13 November 2025
EMA/CHMP/252486/2025
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

VacPertagen

Common name: Pertussis vaccine (recombinant, acellular, component, adsorbed)

Procedure No. EMEA/H/C/006304/0000

Note
Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted.

Official address Domenico Scarlattilaan 6 ● 1083 HS Amsterdam ● The Netherlands
Address for visits and deliveries Refer to www.ema.europa.eu/how-to-find-us
Send us a question Go to www.ema.europa.eu/contact **Telephone** +31 (0)88 781 6000 An agency of the European Union

Table of contents

List of abbreviations

Abbreviation	Definition
°C	Degree celsius
µg	Microgram
$3aP_{gen}$	Recombinant acellular pertussis vaccine containing PT_{gen}, FHA and PRN
A	Adenine
Å	Angstrom
ACIP	Advisory Committee on Immunization Practices
ACT	Adenylate cyclase toxin
ADP	Adenosine diphosphate
AE	Adverse Event
AEFI	Adverse Event Following Immunisation
Al^{3+}	Aluminium
$AlOH_3$	Aluminium Hydroxide
$AlPO_4$	Aluminium Phosphate
ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
ANZCTR	Australian New Zealand Clinical Trials Registry
aP	Acellular Pertussis vaccine
aP_{gen}	Recombinant acellular pertussis vaccine containing PT_{gen} and FHA
aP_{gen}/ $aP5_{gen}$	Acellular Pertussis vaccine containing genetically detoxified Pertussis Toxin
APQR	Annual Product Quality Review
Assoc.	Associate
ATC	Anatomical Therapeutic Chemical (code)
B.	Bordetella
BCA	Bicinchoninic Acid Assay
BNA	BioNet
BSA	Bovine serum albumin
C	Cytosine
CDC	Centers for Disease Control and Prevention, USA
CFU	Colony forming units
CHO	Chinese Hamster Ovary
CI	Confidence Interval
CM	carboxymethyl (sepharose)
CMI	Cell-mediated immunity
CO_2	Carbon dioxide
CPP	critical process parameter
cPT	Chemically detoxified Pertussis Toxin
CQA	Critical quality attribute
CRL	Charles River Laboratory
CSR	Clinical Study Report
CTD	Common Technical Document
DNA	Deoxyribonucleic acid

DO	Dissolved oxygen
DP	Drug product
DS	Drug substance
DSMB	Data and Safety Monitoring Board
DSMC	Data and Safety Monitoring Committee
DT	Diphtheria toxoid
DTaP	Diphtheria toxoid, Tetanus toxoid, acellular Pertussis vaccine
DTP	Diphtheria toxoid, Tetanus toxoid, Pertussis vaccine
DTwP	Diphtheria toxoid, Tetanus toxoid, whole-cell Pertussis vaccine
E	Glutamic Acid
eCTD	Electronic Common Technical Document
ELISA	Enzyme-linked immunoadsorbent assay
EMA	European Medicines Agency
EMEA	European Agency for the Evaluation of Medicinal Products
EPI	Expanded Program of Immunisation
EU	European Union
F	Female
FBV	Final bulk vaccine
FHA	Filamentous Haemagglutinin
FIM	Fimbriae
FP	Filled product
G	Glycine
G	Guanine
g	Gram
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GMC	Geometric Mean Concentration
GMCR	geometric mean of individual ratio
GMP	Good manufacturing practice
GMT	Geometric Mean Titer
GTP	Guanosine triphosphate
h	hours
HA	Hydroxyapatite
HETP	Height equivalent to theoretical plate
HPLC	High Performance Liquid Chromatography
HRP	Horseradish peroxidase
i.m.	Intramuscular
i.p.	Intraperitoneal
ICH	International Conference on Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
ICH	International Council for Harmonisation
IFN	Interferon
IgG	Immunoglobulin G
IL	Interleukin
INN	International Nonproprietary Names
IU	International Unit

K	Lysine
KCMH	King Chulalongkorn Memorial Hospital
kDa	Kilo Dalton
L	liter
LAL	Limulus Amebocyte Lysate
Lf	Limit of flocculation
LoD	Limit of Detection
LoQ	Limit of Quantification
LPS	Lipopolysaccharide
M	molar
M	Male
MAA	Marketing Authorization Application
MCB	Master cell bank
MedDRA	Medical Dictionary for Regulatory Activities
mg	Milligram
min	Minute
MIT	Mouse Immunogenicity Test
mL	Milliliter
mM	Millimolar
MO	Major objection
MRA	Mutual recognition agreement
MSS	Modified Stainer-Scholte
N, n	Number
N/A, NA	Not applicable
Nab	Neutralizing antibody
NaCl	Sodium Chloride
NaOH	Sodium Hydroxide
NAS	New active substance
nCPP	non-critical process parameter
NCT	National Clinical Trial
NIAID	National Institute of Allergy and Infectious Diseases
NIBSC	National Institute of Biological Standards and Control, UK
NLT	not less than
NLT	Not lower than
nm	Nanometer
NMT	not more than
NRG	Name Review Group
NSS	Normal saline solution
NWP	Normalized Water Permeability
OD	Optical Density
OD_{450}	Optical density at 450 nm
OECD	Organisation for Economic Co-operation and Development
PBMC	Peripheral Blood Mononuclear Cell
PBS	Phosphate buffered saline
pCPP	Potential-Critical process parameter or key process parameter
PCR	Polymerase chain reaction

PerMIT	Pertussis Maternal Immunisation in Thailand
PertADO	Pertagen in Adolescents
pH	potential of hydrogen
Ph. Eur.	European Pharmacopoeia
PP	Per Protocol
PRN	Pertactin
Prof.	Professor
PT	Pertussis Toxin
PT_{chem}	Chemically detoxified
PT_{gen}	Recombinant Pertussis Toxin
PT_{gen} or rPT	Genetically detoxified Pertussis Toxin or Recombinant Pertussis Toxin
PV	Pharmacovigilance
q.s.	Quantum sufficiat, sufficient quantity
QC	Quality control
QP	Qualified person
R	Arginine
r-aP	Recombinant acellular pertussis vaccine
RCT	Randomized Controlled Trial
rPT	Recombinant Pertussis Toxin
S	subunit
SAE	Serious Adverse Event
SAS	Statistical Analysis Software
SD	Standard deviation
SDS-PAGE	Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis
TAMC	Total Aerobic Microbial Count
TCT	Tracheal Cytotoxin
TCTR	Thai Clinical Trials Registry
Td	Tetanus toxoid, Diphtheria toxoid (reduced dose)
$Td3aP_{gen}$	Tetanus toxoid, diphtheria toxoid (reduced dose), recombinant acellular pertussis vaccine containing PT_{gen}, FHA and PRN
TdaP/Tdap	Tetanus toxoid, Diphtheria toxoid (reduced dose), acellular Pertussis vaccine
$Tdap_{chem}$	Tetanus toxoid, Diphtheria toxoid (reduced dose), acellular Pertussis vaccine containing chemically detoxified Pertussis Toxin
$TdaP_{gen}$	Tetanus toxoid, diphtheria toxoid (reduced dose), recombinant acellular pertussis vaccine containing PT_{gen} and FHA
$TdaP_{gen}$ / $TdaP5_{gen}$	Td vaccine combined to VacPertagen aP_{gen} components
TFF	Tangential Flow Filtration
Th1	T helper cell type 1
TMB	3,3',5,5'-Tetramethylbenzidine
TMP	transmembrane pressure
TNF	Tumor necrosis factor
TRS	Technical Report Series
TT	Tetanus toxoid
TYMC	Total Yeast and Mold Count

U	Unit
USA	United States of America
USP/NF	United States Pharmacopeia/National Formulary
VTC	Vaccine Trial Centre
WCB	Working Cell Bank or Working Seed
WFI	Water for injection
WHO	World Health Organization
wP	Whole-cell Pertussis
yoa	Years of age
yrs	Years
µm	Micrometer

1. Background information on the procedure

1.1. Submission of the dossier

The Applicant BioNet Europe submitted on 24 June 2024 an application for marketing authorisation to the European Medicines Agency (EMA) for VacPertagen, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 30 March 2023.

The Applicant applied for the following indication:

VacPertagen is indicated for:

- active booster immunisation against pertussis of individuals 12 years of age and older,
- passive protection against pertussis in early infancy following maternal immunisation during pregnancy.

The use of this vaccine should be in accordance with official recommendations.

1.2. Legal basis and dossier content

The legal basis for this application refers to:

Article 8.3 of Directive 2001/83/EC - complete and independent application

The application submitted is composed of administrative information, complete quality data, non-clinical and clinical data based on Applicants' own tests and studies and/or bibliographic literature substituting/supporting certain test(s) or study(ies).

1.3. Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision(s) P/0406/2023 the granting of a (product-specific) waiver.

1.4. Information relating to orphan market exclusivity

1.4.1. Similarity

Pursuant to Article 8 of Regulation (EC) No. 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the Applicant did not submit a critical report addressing the possible similarity with authorised orphan medicinal products because there is no authorised orphan medicinal product for a condition related to the proposed indication.

1.5. Scientific advice

The Applicant received the following Scientific advice on the development relevant for the indication subject to the present application:

Date	Reference	SAWP co-ordinators
19 May 2022	EMA/SA/0000079788	Jens Reinhardt and Svein Rune

		Andersen
27 June 2024	EMA/SA/0000174337	Jens Reinhardt, Anders Lignell and Vilma Petrikaite

The Scientific advice EMA/SA/0000079788 pertained to the following quality, non-clinical and clinical aspects:

- Specifications of active substance and finished to ensure control of quality and manufacturing consistency and presents a comprehensive data package.
- Adequacy of nonclinical data to support choice of population.
- Adequacy of exposure/safety database, the need for clinical consistency trial to confirm lot-to-lot manufacturing consistency to support MAA.

The Scientific advice EMA/SA/0000174337 pertained to the following quality and clinical aspects:

- Active substance and drug product specifications; need for lot-to-lot clinical consistency
- Sufficiency of the safety database and of the overall clinical programme

1.6. Steps taken for the assessment of the product

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Daniela Philadelphy Co-Rapporteur: Christophe Focke

The application was received by the EMA on	24 June 2024
The procedure started on	18 July 2024
The CHMP Rapporteur's first Assessment Report was circulated to all CHMP and PRAC members on	7 October 2024
The CHMP Co-Rapporteur's first Assessment Report was circulated to all CHMP and PRAC members on	n/a
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC and CHMP members on	21 October 2024
The CHMP agreed on the consolidated List of Questions to be sent to the Applicant during the meeting on	14 November 2024
The Applicant submitted the responses to the CHMP consolidated List of Questions on	26 May 2025
The CHMP Rapporteurs circulated the CHMP and PRAC Rapporteurs Joint Assessment Report on the responses to the List of Questions to all CHMP and PRAC members on	30 June 2025
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	10 July 2025
The CHMP agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the Applicant on	24 July 2025

The following GMP inspection was requested by the CHMP and their outcome taken into consideration as part of the Quality assessment of the product:	
A GMP inspection at one manufacturing site in Thailand between 10 – 14 March 2025. The outcome of the inspection carried out was issued on	8 September 2025.
The Applicant submitted the responses to the CHMP List of Outstanding Issues on	15 September 2025
The CHMP Rapporteurs circulated the CHMP and PRAC Rapporteurs Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP and PRAC members on	01 October 2025
The outstanding issues were addressed by the Applicant during an oral explanation before the CHMP during the meeting on	14 October 2025
The CHMP agreed on a list of 2nd outstanding issues in to be sent to the Applicant on	16 October 2025
The Applicant submitted the responses to the CHMP 2nd List of Outstanding Issues on	21 October 2025
The CHMP Rapporteurs circulated the CHMP and PRAC Rapporteurs Joint Assessment Report on the responses to the 2nd List of Outstanding Issues to all CHMP and PRAC members on	29 October 2025
The CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a positive opinion for granting a marketing authorisation to VacPertagen on	13 November 2025

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

Pertussis (whooping cough) is a bacterial respiratory infection caused by Bordetella pertussis, a gram-negative bacillus, which is transmitted through droplets from infected to susceptible individuals.

Symptoms usually appear 7 to 10 days after infection but may also appear up to 21 days later. Initially, symptoms resemble those of a common cold, including sneezing, runny nose, low-grade fever and a mild cough. Within two weeks, the cough becomes more severe and is characterized by episodes of numerous rapid coughs, followed by a crowing or high-pitched whoop. These episodes frequently end with the expulsion of a thick, clear mucous, often followed by vomiting. They initially occur at night and then become more frequent during the day and may recur for one to two months. In young infants the typical 'whoop' may never develop, and the coughing fits may be followed by brief periods when breathing stops. After this phase, the coughing fits become less frequent and less severe, and the infant gradually gets better although this can take up to three months. Adolescents, adults, or partially immunised children generally have milder or atypical symptoms, so in these groups, in addition to very young infants, pertussis might be more difficult to diagnose.

2.1.2. Epidemiology

Pertussis is a significant cause of infant mortality worldwide and continues to be a public health concern even in countries with high vaccination coverage. Recent estimates from WHO suggest that, in 2008, about 16 million cases of pertussis occurred worldwide and that about 195,000 children died from this disease.

Pertussis is an endemic disease in the EU. Every three to five years, larger epidemics are expected even with high vaccination coverage.

According to ECDC, after a few years of limited circulation in the EU, particularly during the COVID-19 pandemic, more than 25 000 cases of pertussis were reported in 2023, and more than 32 000 between January and March 2024. Similar numbers were observed in 2016 and 2019.

During 2023-24, in 17 EU/EEA countries, infants (those under the age of one year) represented the group with the highest reported incidence, whereas in six countries, the highest incidence is reported in adolescents 10-19 years. The majority of deaths occurred in infants. These surveillance data need to be interpreted with caution due to known differences in Member State surveillance systems, availability of laboratory methods, testing practices, as well as vaccination schedules.

The observed epidemiological picture can be ascribed to a number of factors, which include: expected epidemic peaks, presence of unvaccinated or not up to date vaccinated individuals, waning immunity, decreased contribution of natural boosting in the overall population during the COVID-19 pandemic period.

2.1.3. Pathogenesis

B. pertussis causes a localized infection, rarely disseminating from the respiratory tract. Beyond the paroxysmal cough, however, there are systemic manifestations including lymphocytosis, dysregulated secretion of insulin, alterations in neurologic function and recurrence of paroxysmal cough (days to weeks after the infection has been cleared).

Although the relationship between these additional signs and symptoms and the course of clinical pertussis is unclear, some features appear to be attributable to virulence factors with known activities.

Pertussis toxin (PT) is important for pathogenesis. Its ADP-ribosylation of hetero-trimeric G proteins affects signal transduction (disrupts function) in many cell types. The resulting biological effects include induction of lymphocytosis, alteration in insulin secretion, and enhancement of sensitivity to histamine and other mediators, in humans and/or animals. Each of these effects contributes to pathophysiology; for example, it appears that the elevated numbers of white blood cells are involved in pulmonary hypertension, a significant cause of pertussis morbidity and mortality. Filamentous Hemagglutinin (FHA) can participate in the interaction of *B. pertussis* with host cells (adhesion molecule). More recently, FHA was reported to exert immunomodulatory effects in vivo by unknown mechanisms. Pertactin (PRN) is an adhesin and recent in vivo studies suggest immunomodulator function. Fimbriae (FIM) function as adhesins and is also immunomodulatory.

These 4 surface proteins (particularly PT and FHA) are commonly used in licensed Tdap vaccines due to their attributed roles in pertussis pathogenesis.

2.1.4. Clinical presentation and diagnosis

Pertussis, also known as whooping cough, is a highly infectious bacterial disease that affects the respiratory tract. It is caused by a bacterium found in an infected person's mouth, nose and throat.

Symptoms usually appear 7 to 10 days after infection. Initially, the symptoms are similar to those of a common cold, and include sneezing, runny nose, low-grade fever, mild cough. Pertussis can lead to complications such as pneumonia, ear infection, dehydration, seizures and severe cases can lead to death.

There are three stages in the initial infection of unvaccinated people:

- the catarrhal stage is characterised by cold-like symptoms such as a runny nose and cough, rarely also a slight fever (lasting between 1-2 weeks)
- the convulsive stage, which can last 4-6 weeks, is characterised by coughing attacks. During these barking, paroxysmal and spasmodic coughing attacks, the patient may choke up thick mucus and subsequently vomit
- During the decrementi stage, the coughing fits subside (lasting 6-10 weeks)

In vaccinated persons, pertussis can often manifest itself as a long-lasting cough, but without the classic accompanying symptoms mentioned above. In unvaccinated infants under 6 months of age, pneumonia and even respiratory arrest can occur.

Laboratory confirmation:

- Detection of *Bordetella pertussis* using PCR: the ECDC has specific recommendations for carrying out PCR diagnostics for pertussis (Guidance and protocol Pertussis). A deep nasopharyngeal swab should preferably be taken.
- Detection of *Bordetella pertussis* by culture: it is recommended to culture the pathogen in addition to PCR diagnostics, as this is the only way to carry out molecular biological characterisation and antibiotic resistance testing. A deep nasopharyngeal swab should preferably be taken.
- Serodiagnostics: this is useful between the 2nd and 8th week after the onset of coughing attacks. The recommended method for serological diagnostics is to perform an enzyme-linked immunosorbent assay (ELISA) to detect IgG antibodies against pertussis toxin.

2.1.5. Management

Pertussis is treated with antibiotics. To be effective, treatment must begin as early as possible in the course of the disease. Antibiotic treatment can reduce the bacteria in the nose and throat, which also limits the risk of transmission to other people.

The best prevention against whooping cough is immunisation. Complete immunisation schedules in the childhood should be boosted at school age. After this basic immunisation, the vaccination should also be regularly boosted in adulthood. In order to protect infants in the first months of life, pregnant women in the third trimester in particular are recommended to be vaccinated in several countries.

The first pertussis vaccines developed were whole-cell pertussis vaccines (wP) consisting of suspensions of the entire B. pertussis organism that had been heat or chemically inactivated, using chemicals, usually formaldehyde. Immunization with wP vaccines was found effective and the vaccine was relatively inexpensive. Whole-cell pertussis vaccines, which have been used for more than 50 years, have been shown to provide protection against pertussis and still serve as the foundation of global pertussis control. However, vaccination with wP has been frequently associated with minor adverse reactions such as redness and swelling at the site of injection, along with fever and agitation. Local reactions tend to increase with age and the number of injections; wP vaccines are therefore not recommended for immunization of adolescents and adults.

To address the adverse reactions observed with the wP vaccines, acellular pertussis (aP) vaccines were developed in the early 1980's that contained purified components of B. pertussis such as chemically inactivated pertussis toxin (cPT), usually in combination with other B. pertussis components such as Filamentous Haemagglutinin (FHA), Pertactin (PRN), Fimbriae (Fim) type 2 and 3 in different amount and compositions.

All current aP vaccines contain a pertussis toxoid that is chemically-inactivated by formaldehyde, glutaraldehyde or hydrogen peroxide. Acellular pertussis vaccines have been successfully introduced into many national immunization programmes. All currently marketed pertussis vaccines in Europe (e.g. Adacel, Boostrix) also contain antigens from other pathogens such as tetanus or diphtheria.

Resurgence of pertussis has been recently reported in many countries, especially in aP vaccine using countries. Potential factors of pertussis resurgence have been identified. One of the major concerns is age-related waning immunity so that older children and adults may again become susceptible. The aP vaccines containing cPT induce insufficient T-cell type 1 (Th1) immunity which wanes within 2-4 years. The insufficient immune response is explained by the fact that chemical inactivation of cPT dramatically changes the protein structure resulting in a great reduction (80%) of T-cell binding epitope compared to native PT.

A call for new pertussis vaccine containing genetically detoxified Pertussis Toxin (rPT) and more booster immunizations have been proposed. It is expected to have a better immune response and longer duration than aP vaccine containing cPT.

Genetically-inactivated PT (PT_{gen}) mutants were developed simultaneously in Italy and US at the National Institute of Allergy and Infectious Diseases (NIAID). The PT_{gen} contains two mutations of R9K and E129G in the S1 peptide resulting in a PT devoid of toxicity. Inactivation of PT by chemical treatment is therefore unnecessary. The physicochemical and antigenic properties of PTgen were similar to those of native PT. Studies of PT_{gen} containing $DTaP_{gen}$ vaccine containing diphtheria, tetanus and acellular pertussis showed similar safety profile and efficacy in infants (84%, 95% Confidence Interval; 76-90%) with more Th1 immune response compared to cPT-containing DTaP at 5-time higher PT content. The protective efficacy was sustained for 6 years after primary immunization. These aP vaccines containing PT_{gen} were launched in several countries (including Italy, Korea and Thailand) but were withdrawn in 2000's due to commercial issues.

BioNet has developed a recombinant aP_{gen} pertussis-only vaccine (VacPertagen) and a combined $TdaP_{gen}$ including tetanus and diphtheria toxoids (Boostagen). Both vaccines have been evaluated in clinical trials in children, adolescents and adults including pregnant women.

The two vaccines are licensed for active booster use and for passive immunization to protect infants in Thailand and in Singapore.

Only the aP_{gen} vaccine (VacPertagen) is part of the current MA.

2.2. About the product

Mechanism of action

Pertagen (VacPertagen) contains PT_{gen} and FHA proteins, adjuvanted with Alum for active immunization against pertussis. The exact mechanism of protection has not been determined. VacPertagen elicits binding antibodies to PT and FHA as well as neutralizing antibodies against PT.

VacPertagen is a single-dose acellular Pertussis vaccine (aP) containing FHA and a recombinant PT protein. The PT_{gen} contains two mutations of R9K and E129G in the S1 peptide resulting in a PT devoid of toxicity. Inactivation of PT by chemical treatment is therefore unnecessary.

The Applicant seeks approval for active booster immunisation against pertussis of individuals 12 years of age and older and passive protection against pertussis in early infancy following maternal immunisation during pregnancy.

The proposed posology is intramuscular injection of a single dose (0.5 mL).

2.3. Type of Application and aspects on development

NA

2.4. Quality aspects

2.4.1. Introduction

During the procedure the product name has been revised to VacPertagen (previously Pertagen).

The finished product (FP) is presented as a suspension for injection in prefilled syringe containing 5 µg of recombinant genetically-detoxified Pertussis Toxin (rPT) and 5 µg of Filamentous Haemagglutinin (FHA), adsorbed onto 0.3mg aluminium hydroxide (Al^{3+}) as active substance (AS) per 0.5 ml dose.

Other ingredients are: sodium chloride and water for injection

The product is available in in pack sizes of 1 pre-filled syringe.

2.4.2. Active substance

2.4.2.1. General information

The active substance of the acellular pertussis vaccine consists of two purified Bordetella pertussis antigens. The genetically detoxified, recombinant Pertussis Toxin (rPT, also named PT_{gen}) and Filamentous Haemagglutinin (FHA). The genetically inactivation of the PT is based on the following two amino acid mutations R9K and E129G in the S1 peptide resulting in a PT devoid of toxicity. Therefore, a chemical treatment for detoxification is unnecessary but a low-level formaldehyde is used to stabilize the antigens. The applicant provided information on the structure and properties in section 3.2.S.1 together with a schematic structure representing the nucleotide and amino acid sequence, indicating the mutations and the 2 amino acid substitutions.

All manufacturing steps for this product (including working cell bank (WCB) preparation, storage of master cell bank (MCB) and WCB, manufacturing steps and QC testing) are carried out at a single location in Bang Pa-In Ayutthaya in Thailand.

2.4.2.2. Manufacture, process controls and characterisation

Active Substance Manufacturer - aP antigens

Site: BioNet-Asia Co., Ltd. Hi-Tech Industrial Estate, 81 Moo 1, Baan-Lane, Bang Pa-In Ayutthaya, 13160 Thailand

Activities: Storage of MCB and WCB; Preparation of WCB; Manufacture of Active Substance Intermediate and Active Substance (including Upstream Fermentation, Downstream Purification; Primary Packaging and QC testing.

Satisfactory evidence of GMP compliance has been presented.

Description of manufacturing process and process controls

The manufacturing process consists of an upstream fermentation process with pre-inoculum steps in shake flasks, a pre-fermenter and a production fermenter. At completion of fermentation, the harvested broth is centrifuged followed by filtration and the clarified broth contains the rPT and FHA. The downstream purification process consists of an Ultrafiltration with specific molecular weight cut-off membranes together with conventional column chromatography which finally separates the two antigens. The antigens are concentrated and finally sterile filtrated. The active substances (pre-adsorbed purified rPT and pre-adsorbed purified FHA) are stored at 2 − 8 °C until further processing

For all process steps a re-evaluation was performed and the previously classified pCPPs (potential critical process parameters) were either graded as CPP or nCPPs (Non-critical process parameters), Where applicable acceptance criteria are defined for nCPPs and CPPs. These ranges and/or set points are covered by the process validation. In addition, for some in-process controls acceptance criteria were retrospectively defined in Section 3.2.S.2.4.

The compositions of the various media used in upstream process production are provided. All ingredients of animal origin used in the manufacturing process are listed accordingly in 3.2.A.

A detailed description of the manufacturing process and other necessary information according to ICH M4Q(R1) (e.g. including information on the fermentation process, operating conditions, clarification step, max. antifoam amount and a batch numbering system) were provided. Further information on the different purification steps concerning the removal of the dermonecrotic toxin and the adenylate cyclase toxin is addressed. The management of out of specification (OOS) results is appropriately addressed. For the active substance the proposed batch scale is defined by a lower and an upper limit which is specified based on the results from the process validation and commercial production.

The B. pertussis used to isolate the antigens for the vaccine is derived from B. pertussis Tohama Phase- I through several genetic modifications. The strain was obtained by introducing two mutations into the catalytic subunit S1 of PT (R9K and E129G) of the wildtype gene by homologous recombination and insertion of a second copy of the mutated ptx cluster.. The master seed was produced under GMP. All details of the molecular characterization of the Master Seed Lot are described.

According to ICH Q5B the genetic stability was analysed at end of production at the Fermenter step for the WCB which is representative of the manufacturing steps for the genetic stability analysis and the number of cell doublings. Testing and characterization was performed for the MCB Lot and the WCB A protocol for the establishment of future WCB is provided and therefore no future variation is required if new WCB are established according to the approved protocol. The characterization and testing for the MCB and WCB is acceptable.

The raw materials used in the production mainly comply with Ph. Eur. or USP/NF. Only for three components no compendial reference exists and own acceptance criteria are provided. No direct animal components are used during the production (including establishment of the MCB and WCB). The following three components have been identified as having an indirect animal origin. As regards the TSE compliance, please refer to A.2 Adventitious agents safety evaluation.

During the production of the two antigens rPT and FHA three different chromatography resins are used. Different numbers of runs were carried out for the resins. The claimed shelf-life was supported by retrospective evaluation of the three different resin performances used in commercial production including the column quality attributes but also data from produced active substances passing the

validated impurity clearances The missing runs for the chromatography resins will be carry out and missing data will be submitted as soon as finalised (**REC**).

For all steps during the manufacturing of the active substance in-process controls has been proposed. The IPC were re-categorized and divided into critical and non-critical.

Control of materials

A two tiered cell banking system is used and sufficient information is provided regarding testing of MCB and WCB and release of future WCBs. Genetic stability has been demonstrated for cells at and beyond the limit of cell age.

Raw materials and reagents used in the active substance process comply with Ph. Eur. or USP/NF, except for which no compendial reference exists and for which appropriate specifications have been established.

Appropriate specifications have been established for each chromatographic resin

Control of critical steps and intermediates

A satisfactory overview of critical in-process controls and critical in-process tests performed throughout the active substance manufacturing process is given. Acceptable information has been provided on the control system in place to monitor and control the active substance manufacturing process with regard to critical, as well as non-critical operational parameters and in-process tests. Actions taken if limits are exceeded are specified.

Process validation

Data for three consecutive validation batches are provided. Reprocessing steps are not foreseen. The upstream and downstream manufacturing processes for rPT and FHA active substance have not changed throughout the production of the clinical trial material and the manufacturing of the validation batches. The acceptance criteria for critical process parameters met the established criteria and the purified active Substances (rPT and FHA) met the specifications defined. It is stated that a criticality assessment of all quality attributes and process parameter was performed. Data are also provided for the validation of aseptic processing (media simulation) to validate the aseptic process of sterile filtration adsorption, and formulation at formulation area, DTaP-Hib-HBV vaccine is used to be representative for final formulation, considering most complicated formulation. In addition, challenge study to validate the Filter Media for the sterile Filtration of concentrated FHA and rPT were provided. According to the information provided, TFF systems will be used at different production steps. The validation data provided show that the production is in general consistent, well controlled and robust.

Manufacturing process development

During the product development of VacPertagen only very limited aspects were changed. Small scale batches were used for stability studies and pre-clinical investigation. Commercial scale was used from the beginning of the clinical studies until process validation without introducing any changes to the active substance manufacturing process. Further the production process of the active substance has not changed during finished product upscale. Data to compare small and commercial scale were provided. Some changes in analytical methods were introduced and verified through revalidation.

Characterisation

The Master and Working Cell Bank were characterized by sequencing and restriction map analysis. To confirm the structure-related biological properties of the rPT active substance, epitope binding to a reference antibody (found to be protective in a mouse challenge study) was examined. The characterisation and identification is considered sufficient.

In accordance with the characterization requirements in Ph. Eur. Monograph 1356 studies on adenylate cyclase, tracheal cytotoxin and the absence of residual dermonecrotic toxin were performed on five lots. Once consistency has been demonstrated, these tests do not need to be performed on every batch during routine production.

The specific toxicity test (CHO assay) is not routinely performed. This is acceptable as the toxin is genetically detoxified and genetic stability has been demonstrated. Relevant information is provided under 3.2.S.3.1. From the validation report it can be concluded, that the assay is in line with the compendial requirements (2.6.33).

During the manufacturing of the active substances different process steps are responsible for the removal of product and process related impurities. During validation the depletion capacities were investigated. The removal capacities for the various steps for these components based on the validation data are included in 3.2.S.3.2.

2.4.2.3. Specification

The active substance release and shelf life specification tests for appearance, identity, microbial properties, purity and impurities, concentration and potency.

The active substance release and shelf life specification shown tests for appearance, identity, microbial properties, purity and impurities, concentration and potency.

In general, the proposed specifications and test methods are considered acceptable. The test for residual pertussis toxin can be omitted since it is a genetically detoxified pertussis toxin. The specifications and tests are in line with compendial and ICH Q6B expectations. Identifiers for in-house analytical methods are included in module 3.

Analytical methods

The applicant describes the analytical procedures for all tests previously mentioned in sufficient details. Where appropriate, validity criteria of the tests are included, too. The methods are either compendial or fully validated in accordance with ICH Q2(R2) expectations

The applicant introduced dynamic light scattering as suitable method to control the aggregates. Full validation (**REC**) and a preliminary acceptance criterion for release and stability will be provided post-approval (**REC)**.

The comparability of the in-house residual formaldehyde test with the compendial method has been shown according to Ph. Eur. 5.27.

Some adjustments to the justifications have been introduced, basing them on clinical batches and process validation batches.

For bacterial endotoxins the applicant should adjust the specification as a post-approval measurement once more batch manufacturing data is available (**REC**).

The analytical methods used have been adequately described and (non-compendial methods) appropriately validated in accordance with ICH guidelines.

Primary packaging

The active substances (pre-adsorbed rPT and FHA) are stored according to Ph. Eur. 3.2.1. No compliance to Ph. Eur. 3.1.6 for plastic closures is provided; however, a risk assessment was provided indicating that the quality of the stoppers is deemed acceptable. Certificates of analysis are provided with the submission.

Batch analysis

Batch analysis data on six batches per antigen are provided and meet the acceptance criteria. All batches are within the specifications; these batch data confirm a consistent manufacturing process.

Reference materials

In-house and international (NIBSC) Reference standards have been detailed for rPT and FHA purified antigens.

2.4.2.4. Stability

Stability data for three commercial size batches of each of the two active substances are provided. The studies were carried out in the proposed container closure.

The stability program has been described, including the physical, chemical, biological and microbiological tests to be carried out.

Further characterization of aggregation of rPT and FHA in DS at 25°C and 37°C with dynamic light scattering method are requested post-approval (**REC**).

The provided stability data and results justify the proposed shelf life in the proposed container closure.

2.4.3. Finished medicinal product

2.4.3.1. Description of the product and Pharmaceutical development

The final product of VacPertagen is a suspension for injection. The manufacturing of finished product VacPertagen includes the separate adsorption of the two active substances (rPT, and FHA) to aluminium hydroxide gel, and the formulation of the bulk finished product, with subsequent filling of the bulk finished product into prefilled syringes (1 dose 0.5 mL).

Table 1. Composition of finished product

Name of ingredients	Amount per 1 mL	Amount per dose	Function	Reference to standards
Active substance				
rPT	10 µg	5 µg	Antigen	In-house specification
FHA	10 µg	5 µg	Antigen	In-house specification
Excipients				
Aluminum Hydroxide	0.6 mg (as Al^{3+})	0.3 mg (as Al^{3+})	Adsorbent/adjuvant	Ph. Eur.
Sodium Chloride	8.76 mg	4.38 mg	Isotonic agent	Ph. Eur.
Water for Injection	q.s. to 1.0 mL	q.s. to 0.5 mL	Solvent	Ph. Eur.

The vaccine is supplied in a prefilled syringe as a 0.5-mL single dose presentation.

The syringe consists of Type I borosilicate glass syringe barrel with bromobutyl rubber plunger stopper and a stainless steel needle.

All excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. standards. There are no novel excipients used in the finished product formulation.

2.4.3.2. Manufacture of the product and process controls

The applicant has provided an overview of the development of the finished product. The preclinical and clinical lots used were sufficiently representative of the final commercial product and it was clarified that apart from the early phase clinical lot all other clinical lots (including the pivotal clinical lots) only contained rPT and FHA antigens and no PRN. Batch data were provided for all clinical DP lots.

The manufacturing process of VacPertagen consists of the separate adsorption of the two active substances to aluminium hydroxide gel. Which are intermediates with high antigen concentration. These are formulated with sodium chloride, aluminium hydroxide gel and water to the final bulk vaccine and filled into pre-filled syringes to give the final vaccine. The general process of formulation and filling has not changed from the clinical lots to the current commercial production. Two different manufacturing scale processes were independently developed and individual process validation runs of three consecutive batches for each scale were successful. Comparability between large scale and initial small-scale process could be demonstrated.

Batches and batch results for clinical lots for the active substances up to the filled product are provided in pharmaceutical development section.

The analytical development is described in detail. Reports of the changes are provided. Results demonstrate that the results are comparable after the change.

The development of specifications is described in detail from process validation to commercial production. From process validation to commercial production phase several specifications were established, and recently the description of appearance and readout for identity were simplified.

The vaccine is provided as prefilled syringe presentation. This presentation is described in detail. Studies for extractables and leachables for this presentation were conducted and the results are discussed.

Finished Product Manufacturer

Manufacturing of active product intermediates (adsorbed rPT, and adsorbed FHA), formulation of the final bulk, and filling into PFS is performed at the BioNet-Asia plant in Thailand. Mias Pharma Limited, Ireland is responsible for release.

BioNet-Asia Co., Ltd.

81, Hi-Tech Industrial Estate, Moo 1, Baan-Lane, Bang Pa-In, Ayutthaya, 13160 Thailand

- Formulation, filling, primary and secondary packing.

EU release

MIAS Pharma Limited, Suite 1 First Floor, Stafford House, Strand Road, Portmarnock, D13 WC83, Ireland

Satisfactory GMP certificates for all sites are provided.

Current batch sizes and batch formula for the adsorbed intermediates (rPT adsorbed bulk, FHA adsorbed bulk), and bulk finished product are adequately described.

Manufacture of the FP consists of the adsorption of each antigen bulk concentrate onto aluminium hydroxide, formulation of the Final Bulk aP Vaccine (containing the 2 adsorbed antigens) and filling of Final Vaccine Lot into PFS. Potential critical process parameters (key process parameters), and critical process parameters are defined, and applied for validation batches. Operations and controls for filling are described in detail. Manual visual inspection, and visual inspection by an automatic inspection machine are described for the filled syringes.

Packaging and labelling are described in detail as well, and it is explained at which sites these manufacturing steps are conducted, including the manufacturing operations performed in the EU.

A statistical analysis approach to demonstrate manufacturing process consistency including parameters from small scale, commercial scale and current process parameter results confirmed consistent production.

Current commercial large-scale batch size and batch numbering system are well described. However, the batch numbering system was modified with development of the production process.

The applicant provided a description of the FP manufacturing process and the process controls. Validation data were provided confirming the validated status of the process. A summary for FP transport validation is provided. The start of the shelf life is now clearly mentioned in the dossier and correspond to the date of formulation. All previously declared potential CPPs were re-evaluated and either classified as nCPP or CPP and the nCPPs are continuously monitored based on the identified limits and range. Several Process validation studies with respect to FP manufacture are conducted and consist of

- the evaluation of the single antigen adsorption process.

It was possible to demonstrate that 3 consecutive adsorption processes consistently produce material meeting predetermined specification/acceptance criteria and quality characteristics.

Commercial batch sizes for adsorbed rPT Bulk as well as for the adsorbed FHA Bulk are defined including current commercial experience.

- the evaluation of the vaccine formulation process

It was possible to demonstrate that 3 consecutive vaccine formulation processes consistently produce material meeting predetermined specification/acceptance criteria and quality characteristics.

The commercial batch size for final bulk vaccine is defined the evaluation of filling process

It was possible to demonstrate that 3 consecutive vaccine filling processes consistently produce material meeting predetermined specification/acceptance criteria and quality characteristics.

For the process steps from adsorption to filling process parameters are reported and discussed in validation protocols and reports of these manufacturing steps.

Validation studies of the aseptic process consisting of antigen sterile filtration, adsorption and formulation (media simulation study) and of filling (media fill) were executed for 3 consecutive runs.

Qualification/validation of utilities Cleaning validation has been performed.

Shipment validation

Transport validation within the areas where the product is currently marketed were successful, as well as transport validation to Europe.

Container closure

For storage of adsorbed antigens and bulk vaccine glass bottles with screw cap are used. Materials are described in detail and comply with international standards and regulations. The screw caps are deemed acceptable for the intended use.

The final vaccine is filled into syringes with a needle attached, and a needle closure. The syringe is closed with a plunger and a plunger rod attached. All materials are described in detail and the tests performed on these materials are presented as well. The results of the extractables and leachables studies were provided.

As the pre-filled syringe is a medical device a Notified Body Opinion is provided and is satisfactory.

Further the pre-filled syringe is packed into a blister and put into a carton box.

Control of excipients is discussed. The proposed control strategy is considered acceptable.

The manufacturing process has been validated. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible manner. The in-process controls are adequate.

2.4.3.3. Product specification

The specifications are suitable to confirm the quality of the filled product. Tests are performed for appearance, identity, antigen adsorption, potency, physicochemical and microbiological properties.

In general, the specifications and test methods are considered acceptable.

The validation data for the presented release assays are sufficient and all the validation tests expected according to ICHQ2(R2) were addressed. Batch analysis data for three consecutive process validation batches are presented, with data gained from antigen adsorbed bulk, final bulk vaccine and final vaccine lot. All results are within the pre-defined acceptance criteria.

No new impurities have been identified from the FP manufacturing process. The nitrosamine risk evaluation identified no risk.

The applicant presents justifications for the final vaccine lot specifications. Most of the specifications are justified as identity, physiological or derived from compendial expectations. For bacterial endotoxins the applicant plans to adjust the specification with more batches manufactured which is very much encouraged

For the specifications of adsorbed antigen bulks, final bulk vaccine and filled product, the justification for specifications especially for potency and degree of adsorption have been adjusted based both on clinical batches as well as process validation batches. Also, the FHA potency acceptance criterion was adjusted. The applicant presents reference material for the MIT potency assay and the ELISAs for antigen quantification

An evaluation was carried out in accordance with the conclusions of the EMA BWP Assessment Report EMA/369136/2020 for biological products and the Ph. Eur. general monograph on Substance for Pharmaceutical use (2034, 01/2024). The conclusion of this evaluation is that VacPertagen vaccine is considered at negligible risk of nitrosamine contamination taking into account the relevant process factors.

Appropriate controls for the presence of elemental impurities are applied to process materials and excipients and hence in the finished product.

Analytical methods

The analytical methods used have been adequately described and (non-compendial methods) appropriately validated in accordance with ICH guidelines.

Batch analysis

Batch analysis data for three consecutive process validation batches are presented, with data gained from antigen adsorbed bulk, final bulk vaccine and final vaccine lot. All results are within the pre-defined acceptance criteria.

Reference materials

Reference standards have been provided, with certificates of analysis and procedure for preparation and qualification of new standards. Mouse immunogenicity test (MIT) and ELISA are the main tests used for antigen testing and qualification.

2.4.3.4. Stability of the product

Stability plans, stability data and discussion of stability for adsorbed intermediates, final bulk and final lot batches are provided. Clinical batches are included as well as validation batches from initial small-scale process, and the commercial large scale process.

The following tests are defined as stability indicating tests: pH, appearance, degree of adsorption, integrity test, potency, sterility, bacterial endotoxins, osmolality, and free formaldehyde.

Based on available stability data, the proposed shelf-life of 60 months (5 years) and refrigerated storage conditions at 5 ± 3°C, as stated in the SmPC are acceptable. The product should be stored in the original package in order to protect from light.

A statement has been included in the SmPC that unopened vaccine is stable for a total of 3 days when stored at temperature from 8°C to 25°C (temperature excursion).

2.4.3.5. Adventitious agents

Absence of bacteriophages is controlled by an indirect strategy, i.e. monitoring cell growth during production and testing the cell banks for viability.-

2.4.4. Discussion on chemical, and pharmaceutical aspects

In general, the applicant has described and documented all parts of module 3 of the dossier.

The analytical methods for the active substances are fully validated, the newly introduced dynamic light scattering for purity will be validated post-approval.

The finished product is described, and the manufacturing, and control are addressed in the documents.

The analytical methods for the finished product (adsorbed antigens, final bulk vaccine and filled product) are fully validated.

During the procedure an outstanding GMP Certificate was provided, resolving a Major Objection. A number of Recommendations for future development were also raised.

At the time of the CHMP opinion, there were a number of minor unresolved quality issues having no impact on the benefit-risk balance of the product (see Recommendations).

2.4.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. Data has been presented to give reassurance on viral/TSE safety.

2.4.6. Recommendation(s) for future quality development

In the context of the obligation of the MAHs to take due account of technical and scientific progress, the CHMP recommends five points for investigation.

2.5. Non-clinical aspects

2.5.1. Introduction

2.5.2. Pharmacology

2.5.2.1. Primary pharmacodynamic studies

To demonstrate the proof-of-concept, the Applicant submitted several immunogenicity studies with different vaccine formulations.

In the first study, rats received a single IM dose of aP_{gen}, $3aP_{gen}$,$TdaP_{gen}$ or $Td3aP_{gen}$ at the dose of 0.5 mL per rat. 15 days post-injection, the PT and FHA antibody titers and PT-neutralizing capacity were assessed.

In a second study, mice received a single intraperitoneal dose of aP_{gen}, $3aP_{gen}$, $TdaP_{gen}$ or $Td3aP_{gen}$ at 0.5 mL of different doses. 35 days post-injection, the PT antibody titer and FHA antibody titer were assessed.

In a third study, rats received four IM doses every 2 weeks of $3aP_{gen}$ or the reference vaccine at the dose of 0.5 mL per rat. One to two days after the last dose, PT and FHA antibody titers and PT-neutralizing capacity were assessed. In general, the data generated in both mice and rats demonstrated comparable PT and FHA antibody titers and PT neutralizing capacity between the different formulations used (aP_{gen}, $3aP_{gen}$, $TdaP_{gen}$ and $Td3aP_{gen}$). However, high variation within the treated groups was observed.

In addition, maternal antibody transfer was assessed in pregnant rats. In the first PPND study, female rats were injected with the $TdaP_{gen}$ vaccine at the dose of 0.25 mL per rat, on day 21 prior to their mating, gestational days 6 and 15, and on day 7 of lactation. Blood samples were collected 6-12 days prior to treatment (dams), at termination of gestation (dams), and at termination of the lactation period (dams and pups separately). PT and FHA antibody titers and PT-neutralizing capacity were assessed. In the second PPND study, the $TdaP_{gen}$ vaccine was administered as two IM injections 14 days prior to mating, and on gestation days (GD) 0, 6, and 17 to female rats. Blood samples were collected prior to treatment (dams), at GD21 (dams+ foetuses), LD25 (dams +pups), PND45/70/136 (pups), and anti-PT titers were determined by ELISA. In these studies, maternal anti-PT and anti-FHA antibody transfer to offspring rats was demonstrated.

2.5.2.2. Secondary pharmacodynamic studies

No secondary pharmacodynamics studies were performed. This is acceptable, and line with the WHO guideline on vaccines.

2.5.2.3. Safety pharmacology programme

No separate or dedicated safety pharmacology studies were performed by the Applicant.

Data from the toxicology studies did not suggest that the vaccine regimen may affect physiological functions (e.g. central nervous system, respiratory, cardiovascular, and renal functions) other than those of the immune system. Absence of dedicated safety pharmacology studies is accepted.

2.5.2.4. Pharmacodynamic drug interactions

No pharmacodynamic drug interaction studies were performed by the Applicant. Non-clinical studies evaluating pharmacodynamic drug interactions are generally not considered necessary to support development and licensure of vaccine products for infectious diseases. Absence of pharmacodynamic drug interaction studies is accepted.

2.5.3. Pharmacokinetics

In accordance with WHO guidelines on non-clinical evaluation of vaccines (WHO 2005) and vaccine adjuvants and adjuvanted vaccines (WHO 2014), traditional absorption, distribution, metabolism, and excretion (ADME) evaluations are not generally needed for vaccines. The safety concerns associated with vaccines are generally not related to the pharmacokinetics, but are related to the potential induction of immune response.

2.5.4. Toxicology

2.5.4.1. Single dose toxicity

Two GLP-compliant single dose studies in rats have been provided by the Applicant. In the first, rats (5/sex/group) were IM administered aP_{gen}, $3aP_{gen}$, $TdaP_{gen}$ or $Td3aP_{gen}$ In this study, the dose of aP_{gen} is similar to the intended clinical dose. In the second study, rats (5/sex/group) were IM administered a $Td3aP_{gen}$. Most groups (from both studies) included additional antigens such as PRN, tetanus toxoid and/or diphtheria toxoid. This is not considered to have a major impact on the conclusions of the acute toxicity studies. No systemic toxicity was observed in either of the studies. As may be expected for an aluminum-containing IM-administered vaccine, chronic inflammation was observed at the sites of injection in some of the treated rats.

2.5.4.2. Repeat dose toxicity

In a GLP-compliant repeat-dose toxicity study, rats (10/sex) were administered $3aP_{gen}$ on days 1, 15, 30 and 45. The dose of PT_{gen} and FHA and route of administration are consistent with the intended clinical route. Results were compared with negative control, vehicle (adjuvant) control and a reference product 28-day recovery groups were also included.

As may be expected for an aluminum-containing IM-administered vaccine, $3aP_{gen}$ induced chronic inflammatory change at the sites of injection in some of the treated rats in $3aP_{gen}$ vaccine and the reference vaccine however, all changes were recovered after the 28-Day recovery period. No other (adverse) effects were observed in the animals.

2.5.4.3. Genotoxicity

No genotoxicity studies have been performed in accordance with the WHO Guidelines on Non-clinical Evaluation of Vaccines (2005) and Guidelines on the Non-clinical Evaluation of Vaccine Adjuvants and Adjuvanted Vaccines (2014). The absence of these studies is considered acceptable.

2.5.4.4. Carcinogenicity

No carcinotoxicity studies have been performed in accordance with the WHO Guidelines on Non-clinical Evaluation of Vaccines (2005) and Guidelines on the Non-clinical Evaluation of Vaccine Adjuvants and Adjuvanted Vaccines (2014). The absence of these studies is considered acceptable.

2.5.4.5. Reproductive and developmental toxicity

In a PPND study, female Wistar rats were injected $TdaP_{gen}$ vaccine at the dose of 0.25 mL per rat, on day 21 prior to their mating, gestational days 6 and 15, and on day 7 of lactation. The dose of PTgen and FHA is equivalent to half the human dose. The inclusion of TT and DT is not expected that this has a major impact on the outcome of the toxicity study.

At termination of the gestation or lactation period, there was clear evidence of presence of antibodies against the $TdaP_{gen}$ vaccine in all treated animals. Antibodies were also detected in offspring from treated females. Fertility index, fecundity index and gestation index were slightly lowered in the dams treated with $TdaP_{gen}$ vaccine. Post-implantation losses and early resorptions were slightly higher. None of these differences were statistically significant. A similar incidence of epithelioid granulomas of varying severity was observed in the draining lymph nodes of both the treated and control group, however, the incidence of granulomas of higher severity (showing total replacement of lymphoid cells, exhibiting fibrosis across the node, with central necrosis of minimal severity) was increased in the treated group. This is considered to be related to the pharmacological action of the $TdaP_{gen}$ vaccine along with its adjuvant. No adverse effects on pregnancy, parturition, lactation, embryo-foetal, pre-natal or post-natal development were observed in this study.

In a second PPND study, $TdaP_{gen}$ vaccine was administered as 2 intramuscular injections, at 2 separate injections sites, once at 14 days prior to mating, and on Gestation Days 0, 6, and 17 to female Crl:CD(SD) rats The dose of PT_{gen} and FHA is equivalent to the full human dose.

In treated females, antibodies against rPT were induced, which remained high throughout gestation and lactation. At PND25, antibody levels in serum from offspring from treated females were similar to levels in the treated dams, but dropped following weaning. No adverse effects were noted on F0 survival, maternal pregnancy, parturition, and lactation, F1 growth, viability, development, and reproductive performance, and survival of the F2 embryos.

2.5.4.6. Toxicokinetic data

In all toxicity studies, either a half or a full human dose was administered. This is in line with the recommendations of the WHO guidelines on nonclinical evaluation of vaccines. At this dose, no adverse effects other than chronic inflammatory changes at the sites of injection in some of the treated rats (which may be expected for an aluminium-adjuvanted vaccine) were observed.

2.5.4.7. Tolerance

Local tolerance was studied as a part of the single- and repeat-dose toxicity studies. In the single dose study, chronic inflammatory changes at the sites of injection were observed in some of the treated rats. This is a known effect for intramuscular administered aluminium-adjuvanted vaccines. In the repeated dose study, based on the findings of the gross necropsy and microscopic examination of the sites of injection conducted at termination of the study, it was concluded that the vaccine was well tolerated at the injection sites of.

2.5.4.8. Other toxicity studies

Immunotoxicity of aP_{gen} vaccine was evaluated as part of the repeat-dose toxicity study. No adverse effects on the immune system were observed.

2.5.5. Ecotoxicity/environmental risk assessment

In accordance with the CHMP Guideline on the Environmental Risk Assessment of Medicinal Products for Human Use (EMEA/CHMP/SWP/4447100), due to their nature vaccines are unlikely to result in a significant risk to the environment. Therefore, environmental risk assessment studies are not provided in this application for Marketing Authorisation, which is considered acceptable.

2.5.6. Discussion on non-clinical aspects

Pharmacodynamics

To demonstrate the proof-of-concept, the Applicant submitted several immunogenicity studies with different vaccine formulations. Most studies were not representative of the intended drug product (VacPertagen (aP_{gen}), 5µg rPT and 5µg FHA with 0.3 mg Al) but included additional antigens such as PRN, tetanus toxoid (TT) and diphtheria toxoid (DT). However, it is not expected that the inclusion of these antigens had a major impact on the proof-of-concept studies.

Immunogenicity assessment in the non-clinical studies was limited to PT and FHA antibody titers, and PT-neutralizing capacity after intramuscular administration of different formulations in rats and mice. In addition, maternal antibody transfer was assessed in pregnant rats. There was no characterisation of the immune response, no neutralisation titers for the FHA component, no characterisation of antibody subtypes, no cellular immunity, no duration of immune responses, no data to support the proposed formulation (e.g. no dose response) in particular as to the PT antigen content, and no data to support the proposed booster purposing of the candidate vaccine. Of note, the information in the non-clinical immunogenicity study reports was reported on a high level: only summarizing figures are shown, no raw data was provided, no statistical analysis was performed, and information on the study protocol was missing. However at this stage, the added value of additional animal studies is considered limited. Therefore, the above limitations are not further pursued.

In general, the data generated in both mice and rats demonstrated comparable PT and FHA antibody titers and PT neutralizing capacity between the different formulations used: aP_{gen}, $3aP_{gen}$, $TdaP_{gen}$ and Td3aPgen. However, high variation within the treated groups was observed. In addition, maternal anti-PT and anti-FHA antibody transfer to offspring rats was demonstrated after $TdaP_{gen}$ vaccine administration as part of the prenatal and postnatal development studies.

Toxicology

Intramuscular administration of aP_{gen} was well tolerated in single dose-, repeated dose- and PPND studies in rats, with doses up to the full human dose. Although most studies were not representative of the intended drug product (VacPertagen (aPgen), 5µg rPT and 5µg FHA with 0.3 mg Al) but included additional antigens such as PRN, tetanus toxoid (TT) and diphtheria toxoid (DT), this is not expected to have a major impact on the outcome of the toxicity studies, and the batches used are considered sufficiently representative. No adverse effects other than expected for an aluminum-adjuvanted vaccine were observed.

ERA

In accordance with the CHMP Guideline on the Environmental Risk Assessment of Medicinal Products

for Human Use (EMEA/CHMP/SWP/4447100), due to their nature vaccines are unlikely to result in a significant risk to the environment. Therefore, environmental risk assessment studies are not provided in this application for Marketing Authorisation, which is considered acceptable.

2.5.7. Conclusion on the non-clinical aspects

The non-clinical immunogenicity studies demonstrated an immunogenic response in rats and mice after administration of different aP_{gen} vaccine formulations. However, the supportive value of these non-clinical studies is considered limited, and proof-of-concept and efficacy was demonstrated with clinical data.

Intramuscular administration of aP_{gen} (as such or as $3aP_{gen}$, $TdaP_{gen}$ or $Td3aP_{gen}$ vaccine) in rats was well tolerated and did not result in any adverse effects besides chronic inflammatory changes at the sites of injection, an effect that may be expected for an aluminum-adjuvanted vaccine. In addition, no adverse effects on reproduction or development were observed when a $TdaP_{gen}$ vaccine was administered to pregnant rats.

The non-clinical package is considered acceptable in support of the marketing authorisation.

2.6. Clinical aspects

2.6.1. Introduction

GCP aspects

The Clinical trials were performed in accordance with GCP as claimed by the Applicant

The Applicant has provided a statement to the effect that clinical trials conducted outside the Community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

Tabular overview of clinical studies

Table 2. Tabular overview of clinical studies

Study ID	**Enrolment status** **Start date** **Total enrolment/ enrolment goal**	**Design** **Control type**	**Study & control drugs** **Dose, route of administration and duration** **Regimen**	**Population** **Main inclusion/ exclusion criteria**
Adolescents				
TDA202 (main study)	Complete; FSV 6 July 2015, LSV 18 September 2015 (day 28), 9 August 2016 (1 year); LSV 2-year follow-up study: 4 July 2017, LSV 3-year follow-up study: 31 July 2018, LSV 5-year follow-up study: 02 September	RCT	Single IM dose of BioNet aP (VacPertagen), BioNet Tdap or Adacel (comparator)	Healthy adolescents (12-17 yoa)

	2022; 450/450 enrolled			
PertADO (supportive)	Complete; FSV October 2016, LSV March 2017; 62/60 enrolled	RCT	Single IM dose of VacPertagen (+ Td pur) or Boostrix (comparator)	Healthy adolescents (11-15 yoa)
Adults				
TDA206 (main)	Complete; FSV 10 Feb 2020, LSV 21 Oct 2020; 750/750 enrolled	RCT	Single IM dose of VacPertagen, Boostagen, BioNet Recombinant ap, BioNet Recombinant Tdap or Adacel (comparator)	Healthy adults (18-75 yoa)
Pertaprime-01 (supportive)	Complete; FSV 01 Jul 2020, LSV 26 Sep 2023; 102/102 enrolled	RCT	Single IM dose of VacPertagen or Boostrix (comparator)	Healthy adults (18-30 yoa)
APV301 Only safety (supportive)	Ongoing, Study start: 08 Feb 2025 2100/2300 enrolled	RCT	Single IM dose of VaPertagen or Boostrix (comparator)	healthy adults aged 18 to 75 years
TDA203 (supportive)	Complete; FSV 4 July 2018, LSV 24 January 2019; 250/250 enrolled	RCT	Single IM dose of ap-1,1; Tdap-1,1; Tdap-2,5; Boostagen or Boostrix (comparator)	Healthy female subjects (18-40 yoa)
Healthy pregnant women				
TDA207 (main)	Complete; FSV 18 June 2021, LSV 28 October 2022 (day 28), 02 February 2023 (Mother: Last Subject Last Discharged after Delivery Visit), 01 February 2023 (Infant: Last Subject Last Discharged after Delivery Visit); 240/240 enrolled	RCT	Single IM dose of $ap1_{gen}$, $ap2_{gen}$, $ap5_{gen}$ (VacPertagen), $Tdap2_{gen}$, $Tdap5_{gen}$ or $Tdap_{chem}$ (Adacel; comparator)	Healthy pregnant subjects (18-40 yoa)
TDA204 (supportive)	Complete; FSV 04 January 2019, LSV (day 28) 13 November 2019; LSV Maternal subjects: 14 April 2020 (end-of-study for maternal	RCT	Single IM dose of ap-1,1; Tdap-1,1; Tdap-2,5; Boostagen or Boostrix (comparator)	Healthy pregnant subjects (18-40 yoa)

	subjects) LSV Infant subjects: 23 March 2021 (end-of-study); 400/400 enrolled			
PerMIT (supportive)	Complete; FSV 21 January 2019, LSV 21 May 2020; 584/500 enrolled	Observational	Single IM dose of Recombinant aP (VacPertagen), Recombinant TdaP or licensed Td (comparator)	Healthy pregnant subjects (18-40 yoa)
WoMAN-POWER (supportive)	Complete; 181 enrolled (n=90 received Td-VacPertagen)	RCT Only available as publication (Nakabembe et al 2025)	Either two doses of Td vaccine or the intervention (one dose of Td followed by one dose of Td-VacPertagen) stratified by the mother's HIV status. randomised (1:1:1:1)	Pregnant women with and without HIV (18 years or older, between 16 weeks and 26 weeks of gestation)

2.6.2. Clinical pharmacology

2.6.2.1. Pharmacokinetics

The Applicant did not perform clinical trials to investigate the pharmacokinetic features of VacPertagen. This is in line with the Guideline on clinical evaluation of vaccines (EMEA/CHMP/VWP/164653/05 Rev. 1).

2.6.2.2. Pharmacodynamics

Table 3. Summary of ELISA assays and PT neutralising assay in clinical studies

Study code.	Title of study / Study protocol number or Name / Registry number / Publication	Phase of study	Test performed on serum samples	Immunogenicity test performed by Lab
TDA202	A phase II/III randomized, observer-blind, controlled study to demonstrate non-inferior immunogenicity of a combined Tetanus-diphtheria-acellular Pertussis vaccine as compared to Adacel® vaccine in healthy subjects aged 12-17 years Study protocol number: TDA202 Thai Clinical Trials Registry number: TCTR20150703002	II/III	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell Assay**	VisMederi, Enterprise of Service in Life Science Research, Italy. Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
TDA202 2-year follow-up	Antibody persistence at 2 years after a single dose vaccination of acellular pertussis vaccines among Thai adolescents Study protocol number: TDA202 2-year follow-up ClinicalTrials.gov Identifier: NCT04113655	Follow-up post TDA202 vaccination cohort after 2 years	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell Assay**	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
TDA202 3-year follow-up	Antibody persistence at 3 years after a single dose vaccination of acellular pertussis vaccines containing genetically-detoxified pertussis toxin Study protocol number: TDA202 3-year follow-up ClinicalTrials.gov Identifier: NCT04102137	Follow-up post TDA202 vaccination cohort after 3 years	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell Assay**	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)

Study code.	Title of study / Study protocol number or Name / Registry number / Publication	Phase of study	Test performed on serum samples	Immunogenicity test performed by Lab
TDA202 5-year follow-up	Antibody persistence at 5 years after a single dose vaccination of acellular pertussis vaccines containing genetically-detoxified pertussis toxin Study protocol number: TDA202 5-year follow-up ClinicalTrials.gov Identifier: NCT04529720	Follow-up post TDA202 vaccination cohort after 5 years	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell** Assay	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
TDA203	A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and safety of a single dose of BioNet- Asia's acellular pertussis-only vaccine and its combined tetanus-diphtheria-acellular pertussis vaccine at multiple dose levels or Boostagen® in comparison to Boostrix™, when administered to women of child bearing age Study protocol number: TDA203 Thai Clinical Trials Registry number: TCTR20180321004	II	**ELISA** PT, FHA, DT ,TT	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
TDA204	A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and safety of a single dose of BioNet-Asia's acellular pertussis-only vaccine and its combined tetanus-diphtheria-acellular pertussis vaccine at multiple dose levels or Boostagen® in comparison to Boostrix™, when administered to healthy pregnant women Study protocol number: TDA204 Thai Clinical Trials Registry number: TCTR20180725004	II	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell** Assay	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)

Study code.	Title of study / Study protocol number or Name / Registry number / Publication	Phase of study	Test performed on serum samples	Immunogenicity test performed by Lab
PertADO	A Phase II randomized, observer-blind controlled pilot study to compare the safety and immunogenicity of acellular pertussis vaccines including chemically or genetically-detoxified pertussis toxin in adolescents aged 11-15 years previously immunized with acellular pertussis vaccines Study protocol name: The PertADO Geneva trial ClinicalTrials.gov Identifier: NCT02946190	II	**ELISA** PT, FHA,DT,TT **PT Neutralising CHO cell Assay**	Vaccinology Laboratory, HUG Center for Vaccinology, University of Geneva. Geneva Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
PerMIT	Antibody level in cord sera following immunization with recombinant acellular pertussis vaccines during pregnancy: an observational study Study protocol name: Pertussis Maternal Immunization in Thailand (PerMIT) Thai Clinical Trials Registry number: TCTR20200528006	Observational study	**ELISA** PT, FHA **PT Neutralising CHO cell Assay**	Center of Excellence in Clinical Virology Department of Pediatrics, Faculty of Medicine, Chulalongkorn University Thailand Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
Pertaprime-01	An investigator-driven phase II-III randomised, observer-blind, controlled trial to demonstrate non-inferior immunogenicity of Pertagen® in comparison to Boostrix® in healthy young Australian adults aged 18-30 years Study protocol name: Pertaprime-01 Australian New Zealand Clinical Trials Registry Identifier: ACTRN12619000944134	II/III	**ELISA** PT, FHA **PT Neutralising CHO cell Assay**	Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)

Study code.	Title of study / Study protocol number or Name / Registry number / Publication	Phase of study	Test performed on serum samples	Immunogenicity test performed by Lab
TDA206	A phase III randomized, observer-blind, active-controlled study to compare the safety and immunogenicity of an investigational combined Tetanusdiphtheria-recombinant acellular pertussis vaccine (BioNet Tdap) and licensed recombinant TdaP vaccine (Boostagen®), investigational recombinant monovalent acellular pertussis vaccine (BioNet ap) and licensed recombinant aP vaccine (Pertagen®), and another licensed Tdap vaccine, when administered to healthy adults aged of 18-75 years old Study protocol name: TDA206 (PreBoost Adult) Thai Clinical Trials Registry number: TCTR20190927006	III	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell Assay**	Department of Microbiology, Faculty of medicine, Chulalongkorn University Thailand. (ISO15189: 2012 certified laboratory: accreditation no. 4112/55) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)
TDA207	A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and the safety of BioNet recombinant pertussis vaccines with different doses of genetically detoxified pertussis toxin (PT_{gen}) when administered to healthy pregnant women Study protocol name: TDA207 (PreBoost Pregnant women) Thai Clinical Trials Registry number: TCTR20210128004	II	**ELISA** PT, FHA, DT ,TT **PT Neutralising CHO cell Assay**	Department of Microbiology, Faculty of medicine, Chulalongkorn University Thailand. (ISO15189: 2012 certified laboratory: accreditation no. 4112/55) Human Serology Laboratory at BioNet-Asia Co. Ltd Thailand (ISO 15189:2012 certified laboratory: accreditation no. 4242/63)

2.6.3. Discussion on clinical pharmacology

The Applicant did not perform clinical trials to investigate the pharmacokinetic features of VacPertagen. This is in line with the Guideline on clinical evaluation of vaccines (EMEA/CHMP/VWP/164653/05 Rev. 1) and acceptable, since no new delivery system is employed, and the vaccine does not contain novel adjuvants or excipients.

Mechanism of action

VacPertagen is expected to exert its protective effect by eliciting an adaptive immune response by cell-mediated immunity (CMI) and antibodies against the two pertussis antigens included in the vaccine, namely pertussis toxin (PT) and filamentous haemagglutinin (FHA). These antigens are included in numerous already (centrally and nationally) authorised acellular pertussis (combination) vaccines.

Some vaccines only include PT and FHA to cover pertussis, e.g., Hexyon/Hexacima, which received a positive opinion by EMA in 2013. However, 1 or 2 additional different pertussis antigens (on top of PT and FHA) have been included in several other authorized vaccines, namely pertactin and fimbriae (type 2 and 3). Since support for VacPertagen's vaccine effectiveness in this application is based solely on immunogenicity comparisons, the Applicant was requested to discuss possible effects of missing Pertactin and Fimbriae proteins, which are often contained in licensed Tdap vaccines, on genuine aP vaccine efficacy. The Applicant elaborated on this issue based on the available literature. VacPertagen consistently induces high anti-PT and anti-FHA responses (at least comparable to the vaccine comparators) in the Applicant's different trials. Antibodies against PT and FHA have been demonstrated to wane in parallel with declining effectiveness. In addition, the available nonclinical data also suggest a protective effect of PT and FHA in mouse challenge models. Additional non-clinical and structural data discussed by the Applicant indicate that important epitopes for neutralising antibody responses are retained in PT_{gen} whereas such conformational epitopes might be lost after chemical inactivation as used in the approved comparator vaccines. For passive protection against pertussis in early infancy following maternal immunisation during pregnancy, the effectiveness of the vaccine in infants born to vaccinated mothers is solely via transplacental transfer of anti-pertussis antibodies. There is no CMI involved. Moreover, until now no mono- or two-component Pertussis vaccine is approved to be administered during pregnancy for passive protection. However, antibodies against PT can be considered as a major contributor to protection against pertussis. Indeed, a published real-world effectiveness study conducted in Denmark (Kildegaard et al. 2025) provides evidence that a 1-component vaccine (including only pertussis toxin) may be sufficient to provide protection against laboratory-confirmed pertussis, even if the antibodies were only passively transferred from mothers to their infants.

There is no established correlate of protection for pertussis. However, the limited feasibility of performing efficacy trials due to the low (despite increasing) prevalence of pertussis is acknowledged. Therefore, establishing non-inferiority of immunogenicity of a candidate vaccine vs. authorised vaccines is an acceptable approach to infer efficacy for authorization of new vaccines. The totality of the available immunogenicity data is considered for the assessment.

Dose justification

The submitted dossier includes several clinical trials which investigated different formulations and dose levels. Based on the provided immunogenicity data, the chosen dose for PT and FHA is not objected (see the Clinical Efficacy section for more details) although no clear dose justification was presented. The comparators used in the submitted studies (Adacel and Boostrix) contain 2.5 µg and 8 µg PT, respectively. Therefore, 5 µg PT contained in VacPertagen are somewhat in line with the PT content in frequently used $Tdap_{chem}$ vaccines, although it should be highlighted that all authorised acellular pertussis vaccines include chemically inactivated PT.

The Applicant used fully validated assays for the main immunogenicity analyses (ELISA for anti-PT, anti-FHA IgG, PT neutralization assay), which are deemed suitable for their intended purpose.

Cell mediated immune response (CMI)

The Applicant presented a study report for a cell-mediated immune response assay. There, PBMCs were stimulated with rPT, FHA, a mix of rPT and FHA or media and cytokine responses for IL-2, IL-4, IL-13, IL-17, TNF-alpha, IFN-gamma were measured.

CMI data were only considered as exploratory endpoint of Study TDA202, and these data are only considered supportive. Therefore, it is acceptable that the CMI assay was not fully validated.

2.6.4. Conclusions on clinical pharmacology

The Applicant used fully validated assays for the main immunogenicity analyses (ELISA for anti-PT, anti-FHA IgG, PT neutralization assay), which are deemed suitable for their intended purpose. It is acceptable to base the clinical development programme on immunogenicity studies.

2.6.5. Clinical efficacy

2.6.5.1. Dose response study(ies)

The Applicant chose to include the same antigen concentrations (5 µg each) for pertussis toxin (PT) and filamentous haemagglutinin (FHA) as in Triacelluvax, which obtained an MA in the EU in 1999 (MA voluntarily withdrawn by the Applicant in 2002 due to commercial reasons). Of note, Triacelluvax additionally included a third antigen (Pertactin), which is not contained in VacPertagen. Triacelluvax was indicated for active immunisation of children from 6 weeks up to 7 years of age against diphtheria, tetanus and pertussis.

In addition, different PT and FHA dose levels were investigated in Phase II/III clinical trials (see below).

2.6.5.2. Main study(ies)

From an efficacy perspective, studies TDA202, TDA206 and TDA207 were considered as the main clinical studies. However, study AVP301 was a major contributor to the safety database.

Adolescents

Study TDA202 - A phase II/III randomized, observer-blind, controlled study to demonstrate non-inferior immunogenicity of a combined Tetanus-diphtheria-acellular Pertussis vaccine as compared to Adacel vaccine in healthy subjects aged 12-17 years

Methods

The TDA202 study was a double-center (Thailand), observer-blind, randomized phase II/III study to evaluate immunogenicity, reactogenicity and safety of with VacPertagen (BioNet 2-component aP; PT_{gen} and FHA) and Boostagen (BioNet $TdaP_{gen}$ vaccine) in comparison to the licensed Tdap vaccine Adacel after a single dose in 450 healthy adolescents (12 to 17 yoa) randomized 1:1:1. Primary evaluations were performed approximately 1 month after vaccination at relation to baseline and antibody persistence was investigated 1 year after vaccination.

The TDA202 2-year, 3-year, and 5-year follow-up studies were designed as serological follow-up studies (i.e., no intervention given to the participants) to evaluate long-term persistence of specific antibodies induced by BioNet's recombinant aP and TdaP vaccines and a chemically-detoxified Tdap vaccine (Adacel) in participants who were vaccinated during the TDA202 trial. At every follow up, the principal investigator checked with each of the subjects whether they were willing to participate in long-term follow up studies. Only participants who had been enrolled in the TDA202 study at the Vaccine Trial Centre, Faculty of Tropical Medicine, Mahidol University, Bangkok (Study Site No. 2) were recruited for these long-term follow-up studies due to feasibility issues with Study Site No. 1.

- **Study Participants**

For TDA202 study, a total of 450 healthy adolescent subjects of both gender aged between ≥ 12 to < 18 years were planned to be enrolled and randomized 1:1:1 to the vaccine groups. Only those subjects who fulfilled all of the inclusion criteria and none of the exclusion criteria were randomized.

- **Treatments**

All vaccines were provided as single-dose full liquid formulation in a pre-filled syringe. Vaccines were injected into the non-dominant deltoid following the standard procedure for intramuscular (IM) vaccine administration. One single batch for each vaccine was used for the entire study.

Table 4. Composition of BioNet investigational vaccines

Name of ingredients	Amount *per* 0.5-mL dose	
	BNA aP	BNA TdaP
Active substance		
Tetanus Toxoid (TT)	-	7.5 Lf
Diphtheria Toxoid (DT)	-	2.0 Lf
Recombinant Pertussis Toxin (rPT)	5 µg	5 µg
Filamentous hemagglutinin (FHA)	5 µg	5 µg
Excipients		
Aluminum hydroxide	0.3 mg/dose (as Al^{3+})	0.3 mg/dose (as Al^{3+})
Sodium Chloride	4.38 mg	4.38 mg
Water for Injections	q.s. to 0.5 mL	q.s. to 0.5 mL

Table 5. Composition of Adacel vaccine

Name of ingredients	Amount *per* 0.5-mL dose
	Adacel®
Active substance	
Tetanus Toxoid (TT)	5 Lf
Diphtheria Toxoid (DT)	2 Lf
Pertussis Toxoid (PT)	2.5 µg
Filamentous hemagglutinin (FHA)	5 µg
Pertactin (PRN)	3 µg
Fimbriae type 2/3	5 µg
Excipients	
Aluminum phosphate	1.5 mg/dose (0.33 mg/dose as Al)
Water for Injections	q.s. to 0.5 mL

- **Study assessments**

At Visit 1 (screening) and Visit 3 (Day 28 after vaccination) during the TDA202 study, blood samples of at least 5 mL were taken from each subject in each visit to provide a minimum amount of serum for the immunogenicity tests. At Visit 4 (Day 336±28 after vaccination) during the TDA202 study, a blood sample of 5 mL was taken from each subject in a subset of 150 subjects (50 subjects in each vaccine group) randomly selected for persistence study. These samples were evaluated for ELISA antibodies to PT, FHA, diphtheria and tetanus and for PT neutralizing antibody.

During the TDA202 2-year, 3-year, and 5-year follow-up studies, blood samples of approx. 5 mL were taken from all enrolled participants for antibody persistence analysis at 2, 3, and 5 years after vaccination, respectively. All samples were evaluated for ELISA antibodies to PT, FHA, diphtheria and tetanus using standardized ELISA assay. CHO cell assay to detect PT neutralizing antibody was performed only in the same subset of participants who had been randomly selected for PT neutralizing antibody study since the initial TDA202 study.

- **Objectives**

The primary study objective was to demonstrate non-inferior immunogenicity of one dose of BioNet combined Tetanus, reduced dose of Diphtheria and acellular Pertussis vaccine (BioNet TdaP) as compared to Adacel vaccine.

Statistical Hypothesis

H_0: Seroconversion rates among different vaccine groups are not equivalent.

H_A: Seroconversion rates among different vaccine groups are equivalent.

- **Outcomes/endpoints**

Primary Endpoint

Seroconversion rates as defined by proportion of subjects with ≥ 4-fold increase with respect to baseline of ELISA antibodies to PT and FHA in BioNet TdaP and Adacel vaccine groups.

Secondary Endpoints

1. Seroconversion rates as defined by proportion of subjects with ≥ 4-fold increase with respect to baseline of ELISA antibodies to PT and FHA in BioNet aP vaccine group
2. Seroconversion rates as defined by proportion of subjects with > 0.1 IU/mL ELISA antibodies to Tetanus and Diphtheria in BioNet TdaP and Adacel vaccine groups
3. ELISA Geometric mean antibody concentrations to PT, FHA, Tetanus and Diphtheria in BioNet TdaP and Adacel vaccine groups and to only PT and FHA in BioNet aP vaccine group
4. Seroconversion rates as defined by proportion of subjects with ≥ 4-fold increase with respect to baseline of PT neutralizing antibodies (in a subset of approximately 50 subjects *per* vaccine group)

- **Sample size**

The sample size was calculated based on non-inferiority test with alpha level of 0.05 and 80% power, assuming seroconversion rate in control group was 85%. The sample size required for the study is 150 per arm.

(N.B.: the NI-margin was not presented in the protocol, but a NI-margin of 10% was found as footnote in the SAP.)

- **Randomisation and Blinding (masking)**

Subjects who provided their assent and whose parents/legal guardians providing informed written consent were enrolled into the TDA202 study and randomized according to the randomization list. Subjects who met the study admission criteria were vaccinated.

- Trial TDA202 was an observer-blind study as the appearance of the prefilled syringes was different for BioNet vaccines (aP and TdaP) and Adacel vaccine.

Statistical methods

Analysis sets

The Intention-To-Treat (ITT) set excluded from analysis screened subjects who received no vaccine injection.

The "Per Protocol" (PP) set included the subjects in the ITT set who are compliant with the protocol.

Not in the protocol: at the Day 336±28 days visit (i.e. approximately one year after vaccination), 50 participants per group (i.e. a third) were randomly selected for blood withdrawal to evaluate antibody persistence, and 20 of those were further randomly selected to evaluate CMI (cell-mediated immunity).

Statistical methods (as described in the protocol)

Baseline characteristics of subjects among three vaccine groups were compared, using chi-square test for categorical variables or ANOVA test for continuous variable.

Seroconversion rates, compared with baseline titer, and their 95% CI, were calculated for each antibody titer and each vaccine group. Difference of seroconversion rates among vaccine groups was determined using chi-square test. Geometric mean antibody concentration and its 95% CI, pre- and post-vaccination, were calculated for each vaccine group and then compared among different vaccine groups using ANOVA test. In addition, AE and SAE events were described and compared among different vaccine groups.

Missing data

Missing data were not planned to be imputed.

No subgroup analyses were planned for any immunogenicity, reactogenicity or safety endpoint. In the CSR, additional analyses to assess neutralising anti-PT antibody titer and antibody persistence were defined as subgroups, but they referred to specific endpoints which were only measured in these (randomly drawn) subpopulations.

Seroconversion rates, compared with baseline titer, and their 95% CI, were calculated for each antibody titer and each vaccine group. Difference of seroconversion rates among vaccine groups was determined using chi-square test. GMT and its 95% CI, pre- and post-vaccination, were calculated for each vaccine group and then compared among different vaccine groups using one-way ANOVA test or paired t-test method.

The 95% CI of the proportion of the fourfold responders in each group was calculated using Wald's (Normal Approximation) method. The 95% CI of the difference in proportions of the fourfold responders in the 3 vaccine groups was obtained using Wald's confidence limit.

Non-inferiority test was performed using the SAS version 9.4 programs for seroconversion rates and Geometric Mean Change of ELISA anti-PT and anti-FHA antibodies. The confidence limits were calculated based on one-sided test with alpha at 0.025, using Wald test (Castelloe and Watts, 2015). A pre-defined difference margin of 10% for seroconversion rates and 0.67 for the Geometric Mean Change ratio were used for non-inferiority test based on the study hypotheses and WHO guideline, respectively (WHO TRS No. 979, 2013 and CPMP/EWP/2158/99).

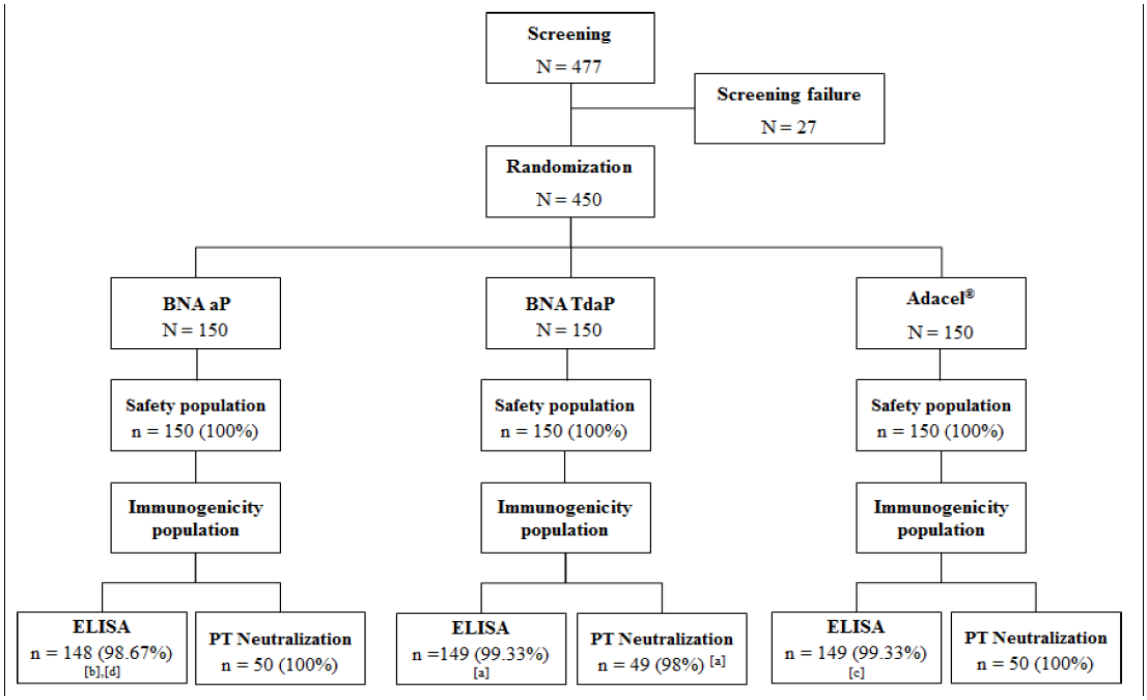
Multiplicity

No multiplicity control within the main study or over the follow-up studies (after 2, 3 and 5 years) is presented in the protocol or the methods section of the CSR, but Bonferroni correction for the three groups within some comparisons is presented.

Results

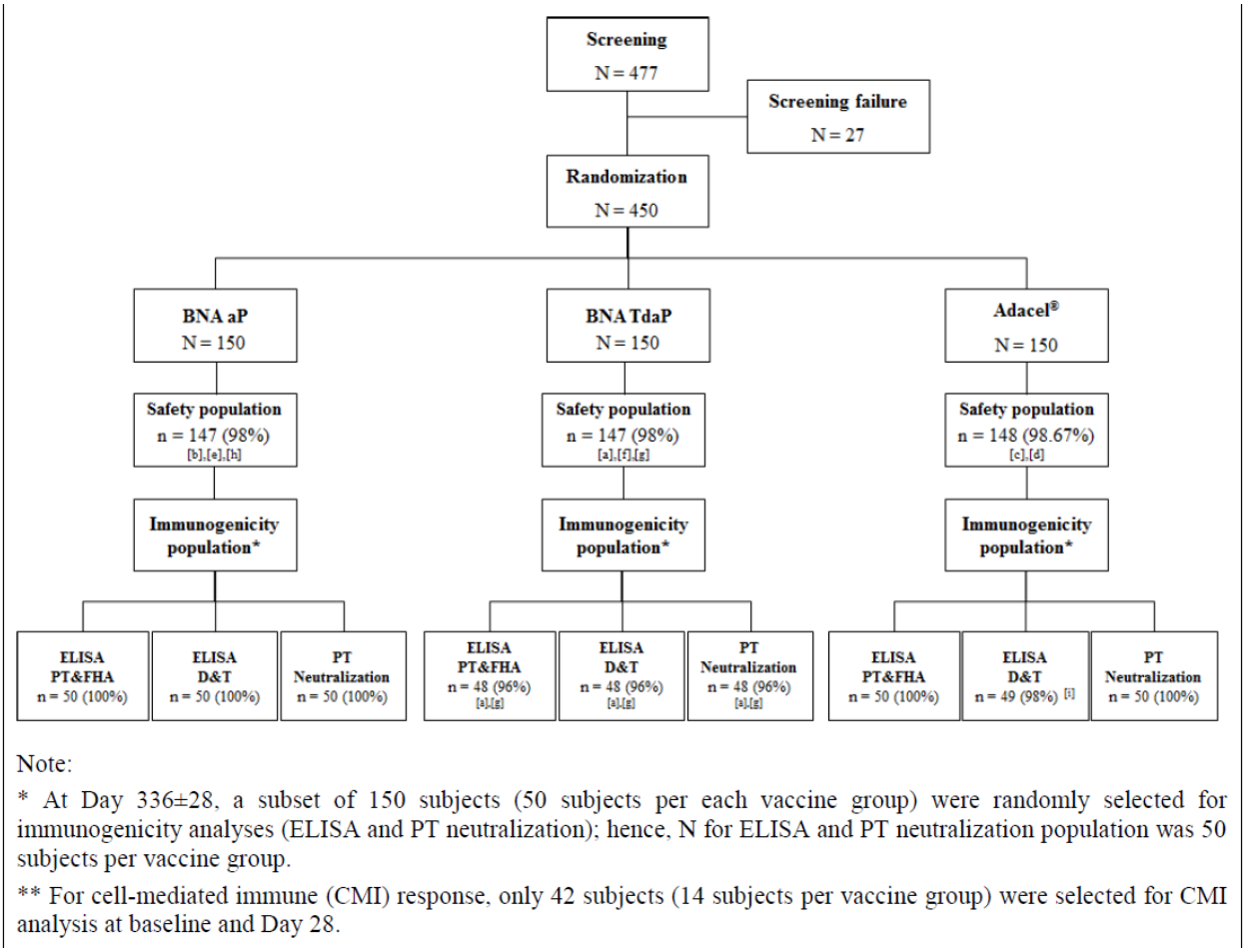
- **Participant flow**

Figure 1. Overall study population at Day 28 after vaccination for TDA202 study



Reason for exclusion lost to follow-up (n=1), Withdrawn by PI for Protocol non-compliance (n=2), Subject refused a blood draw at Day 28 (n=1)

Figure 2. Overall study population at Day 336±28 after vaccination for TDA202 study



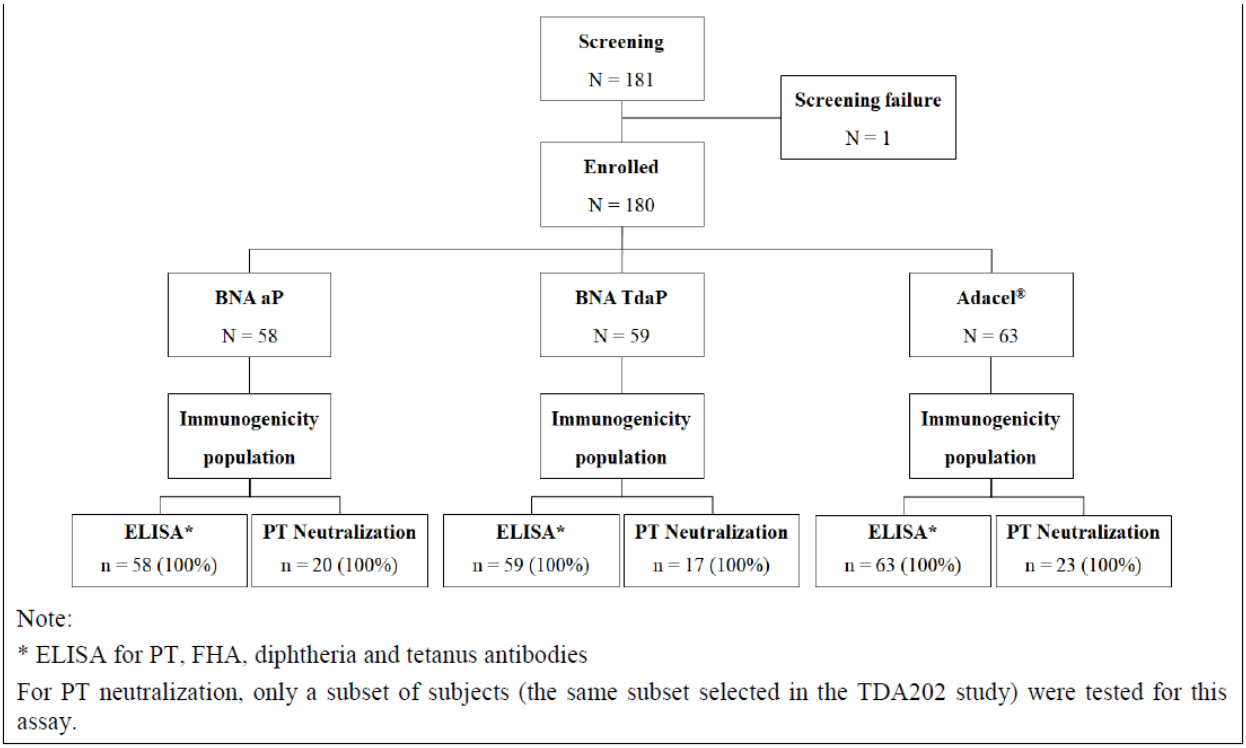
Note:

* At Day 336±28, a subset of 150 subjects (50 subjects per each vaccine group) were randomly selected for immunogenicity analyses (ELISA and PT neutralization); hence, N for ELISA and PT neutralization population was 50 subjects per vaccine group.

** For cell-mediated immune (CMI) response, only 42 subjects (14 subjects per vaccine group) were selected for CMI analysis at baseline and Day 28.

Reason for exclusion: Lost to follow-up (n=1), Withdrawn by PI for Protocol non-compliance (n=2), Subject/Parent/Legal guardian consent withdrawal (n=1), Migrated/moved from the study area (n=3), Lost to follow up (n=1), Excluded from anti-tetanus and anti-diphtheria antibody analysis due to having received Td vaccine 4 days before Day 336 visit(n=1)

Figure 3. Overall study population at 2 years after vaccination for TDA202 2 years follow-up study

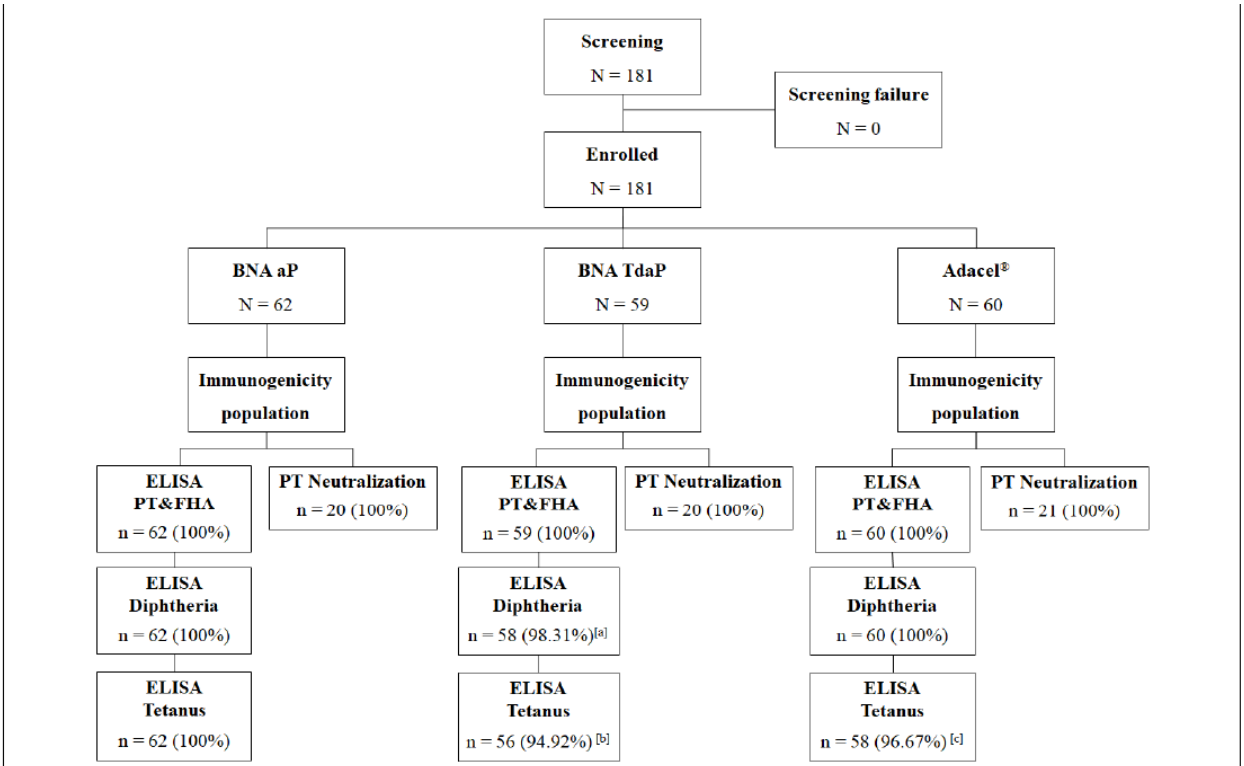


Note:

* ELISA for PT, FHA, diphtheria and tetanus antibodies

For PT neutralization, only a subset of subjects (the same subset selected in the TDA202 study) were tested for this assay.

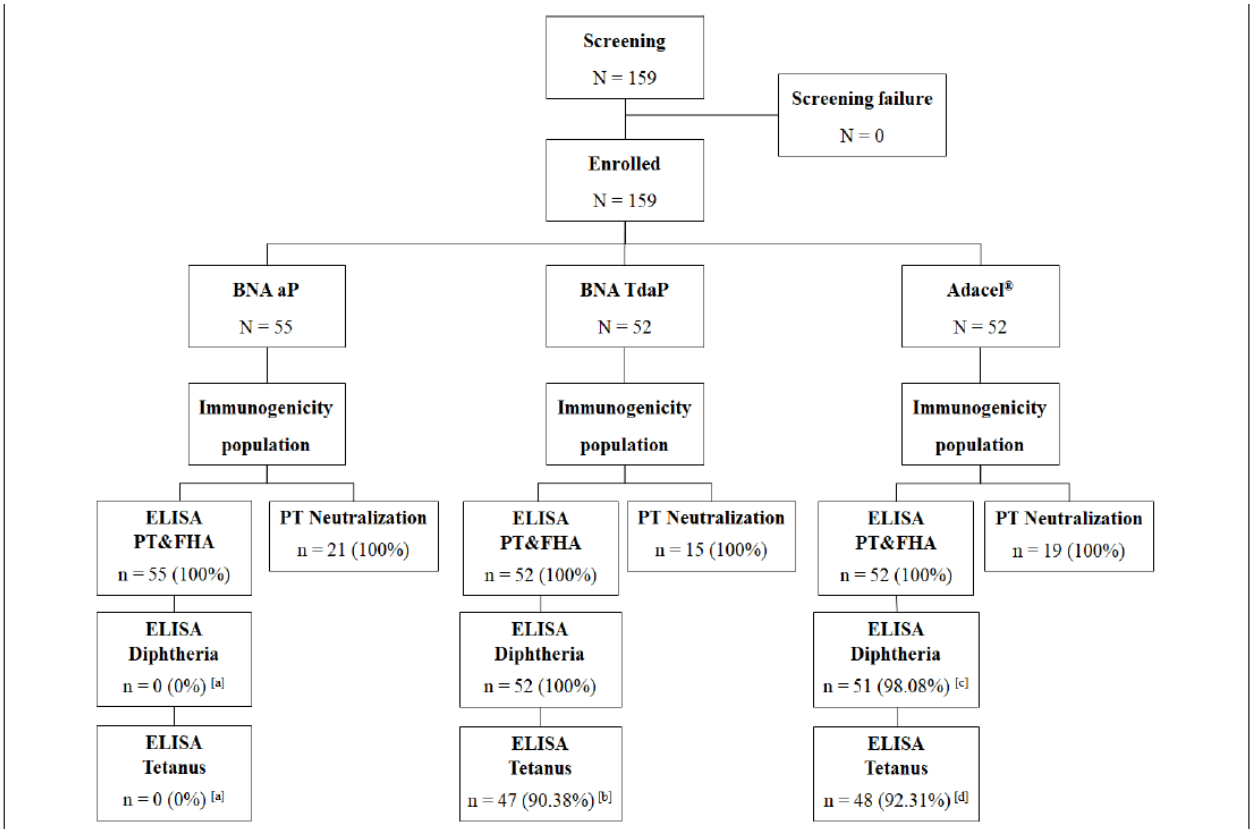
Figure 4. Overall study population at 3 years after vaccination for TDA202 3 years follow-up study



Note: For PT neutralization, only a subset of subjects (the same subset selected in the TDA202 study) were tested for this assay.

Reason for exclusion: Subject received Diphtheria vaccine after the end of the TDA202 2-year follow-up study (n=1), Subject received Tetanus vaccine after the end of the TDA202 2-year follow-up study (n=3), Subject received Tetanus vaccine after the end of the TDA202 2-year follow-up (n=1)

Figure 5. Overall study population at 5 years after vaccination for TDA202 5 years follow-up study



Note: For PT neutralization, only a subset of subjects (the same subset selected in the TDA202 study) were tested for this assay.

Reason for exclusion:

[a] ELISA for anti-tetanus and anti-diphtheria antibodies was not performed for the participants in BNA aP group.

[b] Subject received Tetanus vaccine after the end of the TDA202 2-year follow-up study (n=5),

[c] Subject received Diphtheria vaccine after the end of the TDA202 2-year follow-up study (n=1)

[d] Subject received Tetanus vaccine after the end of theTDA202 2-year follow-up study (n=4)

- **Recruitment**

Date of first subject first visit for TDA202 study: 6 July 2015

Date of last subject for Day 28 visit of TDA202 study: 18 September 2015

Date of last subject for 1 year visit of TDA202 study: 9 August 2016

Date of last subject for 2-year follow-up study: 4 July 2017

Date of last subject for 3-year follow-up study: 31 July 2018

Date of last subject for 5-year follow-up study: 02 September 2022

- **Conduct of the study**

Until 1 year after vaccination (Visit 4), there was no change in the conduct of the study. There was also no change in the conduct of the 2-year, 3-year, and 5-year follow-up studies. Amendments of the CSR primarily concerned the addition of long-term data. There were no changes to relevant analyses throughout TDA202 and the extension studies.

During the conduct of the initial TDA202 study from Visit 1 (screening and vaccination day) to Visit 3 (Day 28 post-immunisation), there were 7 subjects with protocol deviations (mostly refused blood draws at day 28). All subjects were included in the safety analysis for Visit 1 (vaccination) and Visit 2 (Day 7). One subject from BioNet TdaP group was not included in the safety analysis for Visit 3 (Day 28) due to being lost to follow-up. Four subjects were excluded from the immunogenicity analysis.

- **Baseline data**

Table 6. Summary of demographics at baseline for the TDA202 study

Subject status	BNA aP	BNA TdaP	Adacel®	Total	P-value
Demographics at baseline					
(Screening & Vaccination day)					
Gender: n (%)					0.8412 [1]
-N	150	150	150	450	
-Male	61 (40.67)	66 (44.00)	64 (42.67)	191 (42.44)	
-Female	89 (59.33)	84 (56.00)	86 (57.33)	259 (57.56)	
Age (years)					0.5267 [3]
-N	150	150	150	450	
-Mean (SD)	14.31 (1.63)	14.43 (1.70)	14.53 (1.67)	14.42 (1.66)	
-Min/Max	12 - 17	12 - 17	12 - 17	12 - 17	
Height (cm)					0.7704 [2]
-N	150	150	150	450	
-Mean (SD)	158.7 (8.46)	159.4 (8.32)	159.1 (8.57)	159.1 (8.44)	
-Min/Max	136 - 180.8	138 - 184.5	130.7 - 177.7	130.7 - 184.5	
Weight (kg)					0.5494 [3]
-N	150	150	150	450	
-Mean (SD)	53.71 (15.74)	52.16 (13.59)	51.54 (13.45)	52.47 (14.29)	
-Min/Max	28 - 99.1	32 - 106.5	22.5 - 93.6	22.5 - 106.5	

Note:
[1] Overall p-value (2-sided) based on Chi-square test
[2] P-value based on one-way ANOVA
[3] P-value based on Kruskal-Wallis Test
P-value ≤ 0.05 is considered statistically significant.

Table7. Summary of demographics for the TDA202 2-years follow-up study

Subject status	BNA aP	BNA TdaP	Adacel®	Total	P-value
Demographics at baseline					
(Screening & Vaccination day)					
Gender: n (%)					
-N	58	59	63	180	0.0193 [1]*
-Male	22 (37.93)	35 (59.32)	23 (36.51)	80 (44.44)	
-Female	36 (62.07)	24 (40.68)	40 (63.49)	100 (55.56)	
Age					0.1488 [1]
-N	58	59	63	180	
-Mean	16 y 9 m	16 y 8 m	17 y 3 m	16 y 11 m	
-SD	1 y 11 m	1 y 8 m	1 y 8 m	1 y 9 m	
-Min/Max	14 y 0 m - 19 y 11 m	14 y 0 m - 19 y 9 m	13 y 11 m - 19 y 11 m	13 y 11 m - 19 y 11 m	
Height (cm)					0.0419 [2]*
-N	58	59	63	180	
-Mean (SD)	161.5 (7.65)	165.3 (9.29)	162.6 (7.71)	163.1 (8.35)	
-Min/Max	148.5 - 181	146 - 185	147 - 182	146 - 185	
Weight (kg)					0.8221 [1]
-N	58	59	63	180	
-Mean (SD)	56.97 (16.03)	57.81 (15.29)	56.83 (14.80)	57.19 (15.28)	
-Min/Max	34 - 99	34 - 113	33 - 104.5	33 - 113	

y = years, m = months

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on one-way ANOVA

P-value ≤ 0.05 is considered statistically significant.

Table 8. Summary of demographics for the TDA202 3-years follow-up study

Subject status	BNA aP	BNA TdaP	Adacel®	Total	P-value
Demographics at 3 years after vaccination					
Gender: n (%)					0.1581 [1]
-N	62	59	60	181	
-Male	26 (41.94)	33 (55.93)	22 (36.67)	81 (44.75)	
-Female	36 (58.06)	26 (44.07)	38 (63.33)	100 (55.25)	
Age					0.1770 [2]
-N	62	59	60	181	
-Mean	17 y 8 m	17 y 9 m	18 y 2 m	17 y 10 m	
-SD	1 y 11 m	1 y 8 m	1 y 8 m	1 y 9 m	
-Min/Max	15 y 1 m - 20 y 11m	15 y 1 m - 20 y 10 m	15 y 0 m - 21 y 0 m	15 y 0 m - 21 y 0 m	
Height (cm)					0.1427 [3]
-N	62	59	60	181	
-Mean (SD)	162.9 (7.41)	165.7 (9.24)	163.5 (7.85)	164.0 (8.23)	
-Min/Max	149 - 181	146 - 187	147 - 184	146 - 187	
Weight (kg)					0.8471 [2]
-N	62	59	60	181	
-Mean (SD)	60.00 (17.11)	59.09 (14.16)	58.63 (15.36)	59.25 (15.54)	
-Min/Max	38.5 - 110	37 - 96	37.5 - 105	37 - 110	

y = years; m = months

Note:
[1] Overall p-value (2-sided) based on Chi-square test
[2] P-value based on Kruskal-Wallis Test
[3] P-value based on one-way ANOVA

P-value ≤ 0.05 is considered statistically significant.

Table 9. Summary of demographics for the TDA202 5-years follow-up study

Subject status	BNA aP	BNA TdaP	Adacel®	Total	P-value
Demographics at 5 years after vaccination					
Gender: n (%)					0.1078 [1]
-N	55	52	52	159	
-Male	22 (40.00)	29 (55.77)	19 (36.54)	70 (44.03)	
-Female	33 (60.00)	23 (44.23)	33 (63.46)	89 (55.97)	
Age (years)					0.3986 [2]
-N	55	52	52	159	
-Mean (SD)	19.33 (1.85)	19.58 (1.75)	19.79 (1.83)	19.56 (1.81)	
-Min/Max	17-23	17-23	17-23	17-23	
Height (cm)					0.1340 [2]
-N	55	52	52	159	
-Mean (SD)	163.8 (7.55)	167.1 (9.74)	163.9 (8.00)	164.9 (8.55)	
-Min/Max	150-181	147-188	148-186	147-188	
Weight (kg)					0.7699 [2]
-N	55	52	52	159	
-Mean (SD)	63.51 (17.78)	62.77 (14.33)	62.62 (17.79)	62.98 (16.63)	
-Min/Max	40.40 - 102.9	42.20 - 101.9	40.20 - 110.4	40.20 - 110.4	

Note:
[1] Overall p-value (2-sided) based on Chi-square test, [2] P-value based on Kruskal-Wallis Test
P-value ≤ 0.05 is considered statistically significant.

- **Numbers analysed**

For the TDA202 study, a total of 477 volunteers were screened, of whom 450 (225 subjects from each study site) were enrolled and randomized into 3 vaccine groups (150 subjects per vaccine group). The main reason for screening failures was having received tetanus or diphtheria or pertussis vaccine within 1 year prior to study enrollment. Each study site enrolled 225 subjects into the study.

For the primary statistical analysis for data up to Visit 3 (Day 28 after vaccination), all subjects were included in the safety analysis (ITT population). Immunogenicity analysis (PP population; 446 subjects) was performed in 148 subjects in the BioNet aP group and 149 each in the BioNet TdaP and Adacel groups.

The secondary statistical analysis was performed with Visit 4 data, approximately one year after vaccination, and included one year follow up of safety data (SAEs) in all subjects and immunogenicity data, to evaluate antibody persistence in a subset of 50 subjects in each vaccine group.

Details on participant numbers in different analyses are shown in the participant flow figures in the corresponding section above. Participant flow and the numbers analyzed have been presented in sufficient detail. No relevant imbalances between the treatment groups are noted with respect to exclusions from the immunogenicity sets.

- **Outcomes and estimation**

Analysis of Immunogenicity Data at Day 28 Post-vaccination

Seroconversion rates (PT, FHA)

ELISA anti-PT and anti-FHA seroconversion rates were higher in the BioNet aP [anti-PT 96% (95% CI 93-99) and anti-FHA 93% (95% CI 89-97)] than the seroconversion rates in the Adacel group [anti-PT 55% (95% CI 47-63), anti-FHA 54% (95% CI 46-62)].

Table 10. Seroconversion rates as defined by proportion of subjects with ≥4-fold increase in anti-PT and anti-FHA antibody titres at Day 28 post-vaccination compared to baseline as assessed by ELISA in all evaluable subjects by vaccine group

Seroconversion rates	BNA aP (N=148)	BNA TdaP (N=149)	Adacel® (N=149)	P-value
	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	
PT	142 (95.95) (92.77-99.12)	144 (96.64) (93.75-99.54)	82 (55.03) (47.05-63.02)	<0.0001 [1]*
FHA	138 (93.24) (89.20-97.29)	123 (82.55) (76.46-88.64)	81 (54.36) (46.36-62.36)	<0.0001 [1]*

Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

Anti-PT and anti-FHA Geometric Mean Titers (GMTs):

At 28 days after vaccination, anti-PT and anti-FHA GMTs were higher in BioNet aP [562 IU/mL (95% CI 467.79-674.86) for anti-PT antibody; 924 IU/mL (95% CI 809.39-1054.4) for anti-FHA antibody] than those GMTs in Adacel group [63 IU/mL (95% CI 51.05-78.37) for anti-PT antibody; 242 IU/mL (95% CI 208.86-280.05) for anti-FHA antibody].

Table 11. Geometric Mean Titers of ELISA anti-PT and anti-FHA antibodies at baseline (screening) and at Day 28 post-vaccination

Vaccine	PT				FHA			
	Baseline	Day 28 post-vaccination	Geometric Mean Change *from baseline*	P-value [a]	Baseline	Day 28 post-vaccination	Geometric Mean Change *from baseline*	P-value [a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMT Ratio (95% CI)		GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMT Ratio (95% CI)	
BNA aP (N = 148)	13.64 (11.55-16.11)	561.87 (467.79-674.86)	41.20 (34.05-49.86)	<0.0001 [1]*	38.99 (32.54-46.72)	923.80 (809.39-1054.4)	23.69 (19.39-28.96)	<0.0001 [1]*
BNA TdaP (N = 149)	12.91 (11.24-14.83)	365.23 (315.07-423.38)	28.28 (24.04-33.27)	<0.0001 [1]*	41.42 (34.60-49.58)	632.11 (549.85-726.67)	15.26 (12.43-18.74)	<0.0001 [1]*
Adacel® **(N = 149)**	15.57 (13.21-18.36)	63.26 (51.05-78.37)	4.06 (3.47-4.76)	<0.0001 [1]*	45.71 (37.64-55.50)	241.85 (208.86-280.05)	5.29 (4.32-6.48)	<0.0001 [1]*
P-value [b]	0.3631 [2]	<0.0001 [2]*	<0.0001 [2]*		0.5369 [2]	<0.0001 [3]*	<0.0001 [3]*	
Vaccine difference ratio (95%CI)		BNA TdaP - Adacel® 5.77 (4.21-7.92) BNA aP - Adacel® 8.88 (6.47-12.19) BNA aP - BNA TdaP 1.54 (1.12-2.11)	BNA TdaP - Adacel® 6.96 (5.19-9.34) BNA aP - Adacel® 10.14 (7.56-13.62) BNA aP - BNA TdaP 1.46 (1.09-1.96)			BNA TdaP - Adacel® 2.61 (2.06-3.32) BNA aP - Adacel® 3.82 (3.00-4.86) BNA aP - BNA TdaP 1.46 (1.15-1.86)	BNA TdaP - Adacel® 2.88 (2.04-4.09) BNA aP - Adacel® 4.48 (3.16-6.35) BNA aP - BNA TdaP 1.55 (1.10-2.20)	

a : Compared between baseline and Day 28 post-vaccination
b : Compared between vaccine groups
Vaccine difference (95% CI) based on Bonferroni

Note:
[1] P-value based on paired t-test
[2] P-value based on Kruskal-Wallis Test
[3] P-value based on one-way ANOVA
* P-value ≤ 0.05 is considered statistically significant.

Anti-PT neutralizing antibodies

Neutralizing anti-PT antibody was assessed in a subset of 50 subjects in each of the vaccine groups. Neutralizing anti-PT GMTs were similar at baseline across the three vaccine groups but were higher in the BioNet aP group [275.74 IU/mL (95% CI 181.63-418.59)] than in the Adacel group [36.26 IU/mL (95% CI 25.74-51.08)] 28 days after vaccination.

Table 12. Comparison of PT neutralizing GMTs (IU/ml) between baseline and Day 28 post-vaccination as assessed by PT neutralizing assay in CHO cells in a subset of 150 subjects (50 subjects per each vaccine group)

Vaccine	Baseline	Day 28 post-vaccination	Geometric Mean Change *from baseline*	P-value [a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMT Ratio (95% CI)	
BNA aP (N = 50)	9.75 (7.23-13.14)	275.74 (181.63-418.59)	28.28 (20.12-39.77)	<0.0001 [1]*
BNA TdaP (N = 49)	7.93 (6.13-10.27)	215.92 (164.16-284.02)	27.22 (19.99-37.07)	<0.0001 [1]*
Adacel® **(N = 50)**	8.83 (6.50-12.01)	36.26 (25.74-51.08)	4.11 (3.28-5.14)	<0.0001 [1]*
P-value [b]	0.7993 [2]	<0.0001 [2]*	<0.0001 [2]*	
Vaccine difference ratio (95% CI)		BNA TdaP - Adacel® 5.96 (3.27-10.83) BNA aP - Adacel® 7.60 (4.19-13.79)	BNA TdaP - Adacel® 6.63 (4.00-10.98) BNA aP - Adacel® 6.89 (4.17-11.37)	

a: Compared between baseline and Day 28 post-vaccination
b: Compared between vaccine groups
Vaccine difference (95% CI) based on Bonferroni
Note:
[1] P-value based on paired t-test
[2] P-value based on Kruskal-Wallis Test
* P-value ≤ 0.05 is considered statistically significant.

Antibody responses 1 year after vaccination

Seroconversion of ELISA anti-PT and anti-FHA antibodies

ELISA anti-PT and anti-FHA seroconversion rates were higher in the BioNet aP group [Anti-PT 82% (95% CI 71-93), anti-FHA 64% (95% CI 51-77)] than the seroconversion rates in the Adacel group [anti-PT 4% (95% CI 0-9), anti-FHA 28% (95% CI 16-40)] at 1 year after vaccination.

Table 23. Seroconversion rates as defined by proportion of subjects with ≥ 4-fold increase in anti-PT and anti-FDA antibody titers at Day 336±28 post vaccination compared to baseline as assessed by ELISA in all evaluable subjects by vaccine groups

Seroconversion rates	BNA aP (N=50) n (%) (95% CI)	BNA TdaP (N=48) n (%) (95% CI)	Adacel® (N=50) n (%) (95% CI)	P-value
PT	41 (82.00) (71.35-92.65)	36 (75.00) (62.75-87.25)	2 (4.00) (0.00-9.43)	<0.0001 [1]*
FHA	32 (64.00) (50.70-77.30)	27 (56.25) (42.22-70.28)	14 (28.00) (15.55-40.45)	0.0007 [1]*

Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

ELISA anti-PT and anti-FHA GMTs

ELISA anti-PT and anti-FHA GMTs were higher in the BioNet aP group [133 IU/mL (95% CI 92.96-189.77) for anti-PT antibody; 291 IU/mL (95% CI 230.94-367.14) for anti-FHA antibody] than GMTs in Adacel group [22 IU/mL (95% CI 16.05-29.75) for anti-PT antibody; 90 IU/mL (95% CI 64.46-125.39) for anti-FHA antibody] at 1 year after vaccination.

Table 34. Geometric Mean Titers of ELISA anti-PT and anti-FHA antibodies at Day 336±28 post-vaccination in a subset of 150 subjects (50 subjects per group)

Vaccine	PT				FHA			
	Baseline [a] GMT (IU/mL) (95% CI)	Day 336 post-vaccination GMT (IU/mL) (95% CI)	Geometric Mean Change from baseline GMT Ratio (95% CI)	P-value b	Baseline [a] GMT (IU/mL) (95% CI)	Day 336 post-vaccination GMT (IU/mL) (95% CI)	Geometric Mean Change from baseline GMT Ratio (95% CI)	P-value b
BNA aP (N = 50)	15.97 (12.17-20.97)	132.82 (92.96-189.77)	8.32 (6.12-11.30)	<0.0001 [1]*	45.55 (34.00-61.02)	291.18 (230.94-367.14)	6.39 (4.64-8.81)	<0.0001 [1]*
BNA TdaP (N = 48)	13.71 (10.91-17.24)	115.64 (88.09-151.81)	8.43 (6.48-10.97)	<0.0001 [1]*	38.50 (28.66-51.71)	208.57 (156.88-277.28)	5.42 (3.94-7.45)	<0.0001 [1]*
Adacel® (N = 50)	15.64 (11.59-21.11)	21.85 (16.05-29.75)	1.40 (1.16-1.68)	0.0005 [1]*	44.55 (31.37-63.27)	89.91 (64.46-125.39)	2.02 (1.45-2.80)	<0.0001 [1]*
P-value [c]	0.8415 [2]	<0.0001 [3]*	<0.0001 [3]*		0.7150 [3]	<0.0001 [3]*	<0.0001 [3]*	
Vaccine difference ratio (95%CI)		BNA TdaP - Adacel® 5.29 (3.09-9.08) BNA aP - Adacel® 6.08 (3.56-10.37)	BNA TdaP - Adacel® 6.04 (3.90-9.35) BNA aP - Adacel® 5.95 (3.86-9.18)			BNA TdaP - Adacel® 2.32 (1.42-3.78) BNA aP - Adacel® 3.24 (2.00-5.26)	BNA TdaP - Adacel® 2.68 (1.54-4.66) BNA aP - Adacel® 3.17 (1.83-5.47)	

a : Baseline GMTs were calculated from the same subset of subjects tested for immunogenicity at Day 336 post-vaccination.
b : Compared between baseline and Day 336 post-vaccination
c : Compared between vaccine groups
Vaccine difference (95% CI) based on Bonferroni

Note:
[1] P-value based on paired t-test
[2] P-value based on Kruskal-Wallis Test
[3] P-value based on one-way ANOVA
* P-value ≤ 0.05 is considered statistically significant.

Anti-PT neutralizing antibody

Neutralizing anti-PT GMTs were higher in the BioNet aP group [77 IU/mL (95% CI 53.27-111.72)] than the GMTs in the Adacel group [12 IU/mL (95% CI 8.93-16.66)] at 1 year after vaccination.

Table 15. Comparison of PT neutralizing GMTs (IU/ml) between baseline and Day 336±28 post-vaccination as assessed by PT neutralizing assay in CHO cells in a subset of 150 subjects (50 subjects per each vaccine group)

Vaccine	Baseline [a]	Day 336 post-vaccination	Geometric Mean Change *from baseline*	P-value [b]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMT Ratio (95% CI)	
BNA aP (N = 50)	9.75 (7.23-13.14)	77.15 (53.27-111.72)	7.91 (5.86-10.69)	<0.0001 [1]*
BNA TdaP (N = 48)	8.01 (6.16-10.42)	67.48 (50.08-90.91)	8.43 (6.19-11.46)	<0.0001 [1]*
Adacel® (N = 50)	8.83 (6.50-12.01)	12.20 (8.93-16.66)	1.38 (1.14-1.68)	0.0015 [1]*
P-value [c]	0.8198 [2]	<0.0001 [2]*	<0.0001 [2]*	
Vaccine difference ratio (95% CI)		BNA TdaP - Adacel® 5.53 (3.15-9.71) BNA aP - Adacel® 6.32 (3.62-11.04)	BNA TdaP - Adacel® 6.10 (3.84-9.70) BNA aP - Adacel® 5.73 (3.62-9.07)	

a: Baseline GMTs were calculated from the same subset of subjects tested for immunogenicity at Day 336 post-vaccination.
b: Compared between baseline and Day 336 post-vaccination
c: Compared between vaccine groups
Vaccine difference (95% CI) based on Bonferroni

Note:
[1] P-value based on paired t-test
[2] P-value based on Kruskal-Wallis Test
* P-value ≤ 0.05 is considered statistically significant.

Immunogenicity results up to 5 years after immunisation (2-, 3- and 5-year follow-up studies)

During the 2-, 3-, and 5-year follow up studies, antibody responses declined in all groups. The trend of immunogenicity comparisons between BioNet aP and Adacel was similar in all follow up studies. Considering anti-PT and anti-FHA IgG GMCs over the 5-year period after vaccination, BioNet aP can induce considerable pertussis antibody levels (for both PT and FHA) as compared to Adacel.

After 5 years, immune responses against pertussis antigens in terms of seroconversion rates as well as GMCs/GMTs induced by BioNet aP were higher than the responses induced by Adacel. At 5 years after vaccination, seroconversion rates were 33%, 95% CI 20-45, n=55 for BioNet aP and 2%, 95% CI 0-6 for Adacel. Seroconversion rates for ELISA anti-FHA antibody also were higher in the BioNet aP group (45%, 95% CI 32-59, n=55) than in participants vaccinated with Adacel (8%, 95% CI 0- 15).

ELISA anti-PT and anti-FHA GMCs were higher at 5 years after vaccination in the BioNet aP group (33 IU/mL, 95% CI 24.65-43.10 for anti-PT and 70 IU/mL, 95% CI 57.29-86.28 for anti-FHA) than GMCs in Adacel group (GMCs for both PT and FHA were below baseline level: 11 IU/mL, 95% CI 8.78-14.45 for anti-PT and 28 IU/mL, 95% CI 21.16-37.87 for anti-FHA). The neutralizing antibody responses showed a similar pattern over the 5-year period after vaccination.

Figure 6. Geometric Mean Concentrations of ELISA anti-PT antibody over 5 years after vaccination in all evaluable subjects at each timepoint by vaccine groups

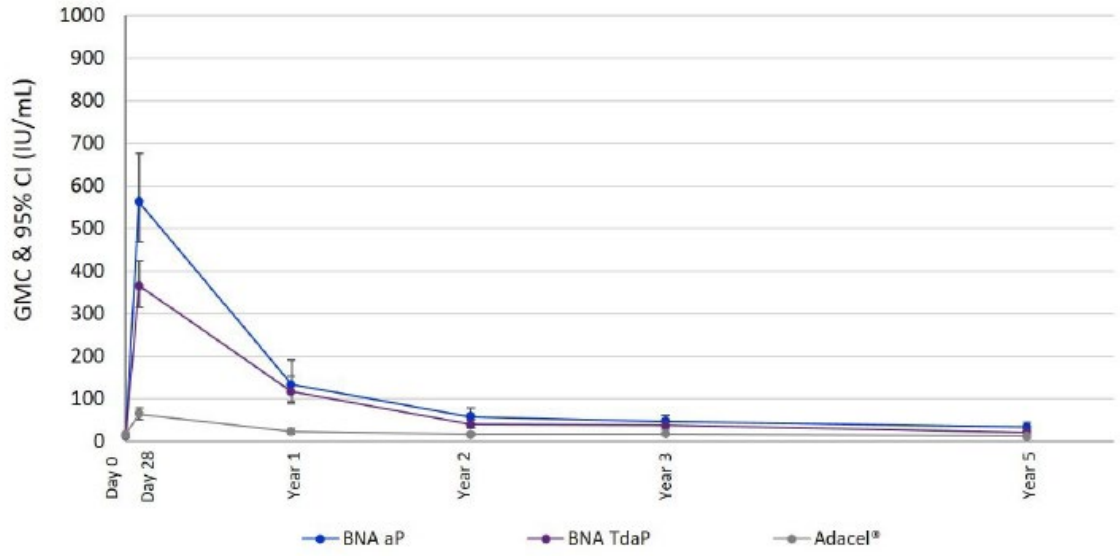


Figure 7. Geometric Mean Concentrations of ELISA anti-FHA antibody over 5 years after vaccination in all evaluable subjects at each timepoint by vaccine groups

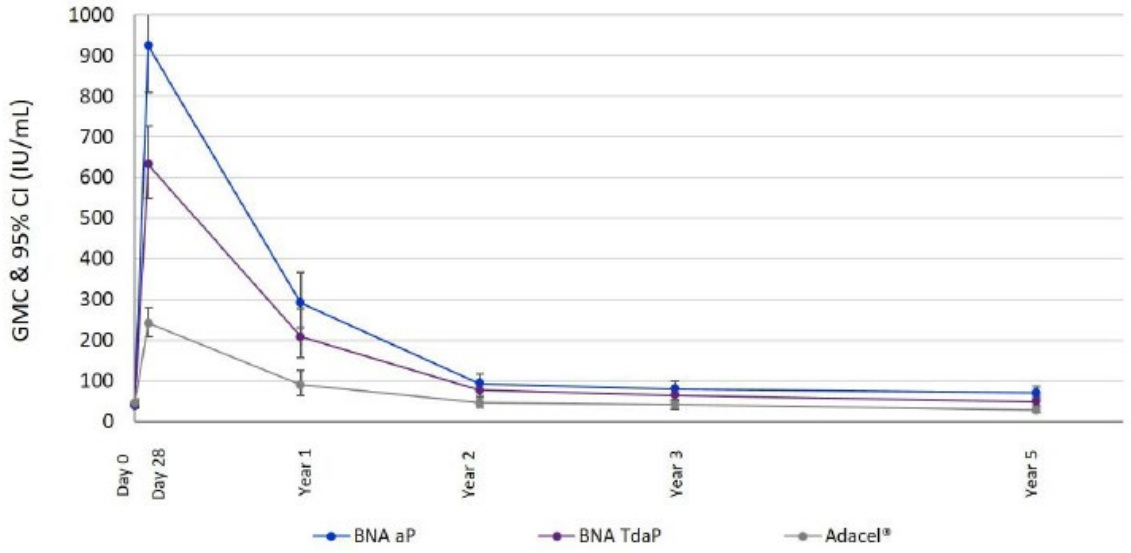
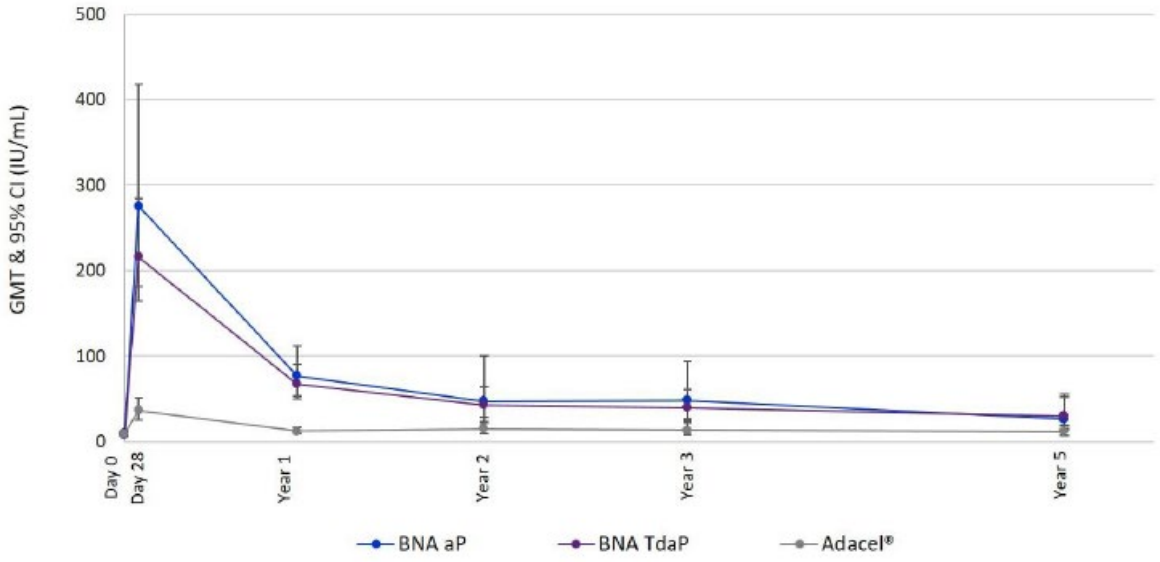


Figure 8. Geometric Mean Titers of PT neutralizing antibody over 5 years after vaccination in all evaluable subjects at each timepoint by vaccine group



Adults

Study TDA206 - A phase III randomized, observer-blind, active-controlled study to compare the safety and immunogenicity of an investigational combined Tetanus-diphtheria-recombinant acellular pertussis vaccine (BioNet Tdap) and licensed recombinant TdaP vaccine (Boostagen), investigational recombinant monovalent acellular pertussis vaccine (BioNet ap) and licensed recombinant aP vaccine (VacPertagen), and another licensed Tdap vaccine, when administered to healthy adults aged of 18-75 years old

Methods

The **TDA206 study** was a phase III, single-site, observer-blind, randomized, active-controlled vaccine trial in 750 Thai healthy adults 18-75 yoa to compare the safety and immunogenicity of a single dose BioNet Tdap and Boostagen, BioNet ap and VacPertagen, and Adacel (randomized in a 1:1:1:1:1 ratio). Primary evaluations were performed approximately 1 month after vaccination at relation to baseline and antibody persistence was investigated 1 year after vaccination. Study TDA206 was conducted at 1 site in Thailand

Table 16. Vaccine groups

Vaccine Groups	*N (18-64 years old)*	*N (65-75 years old)*	*Total in each vaccine group*
BioNet Recombinant ap	120	30	150
BioNet Recombinant Tdap	120	30	150
Pertagen®	120	30	150
Boostagen®	120	30	150
Adacel®	120	30	150
Total	**600**	**150**	**750**

The TDA 206 2- and 3-years follow-up study was conducted to assess the persistence of antibody levels of BioNet recombinant ap and BioNet recombinant Tdap vaccines compared with Adacel at 3 years after vaccination. The study included randomly pre-selected participants who had undergone immunogenicity assessment at Visit 4 in the initial phase III TDA206 study.

- **Study Participants**

A total of 750 participants (600 participants aged 18-64 years and 150 participants aged 65-75 years) were planned to be enrolled. Only those participants who fulfilled all of the inclusion criteria and none of the exclusion criteria were randomized.

- **Treatments**

For each of the 5 study vaccines, a single batch was used for the entire study.

Table 17. Composition of BioNet's investigational vaccines and reference vaccines

Name of ingredients per 0.5-mL dose	Investigational Vaccines		Reference Vaccines		
	Group 1	Group 2	Group 3	Group 4	Group 5
	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®
Active ingredients					
Tetanus Toxoid (TT)	-	7.5 Lf	-	7.5 Lf	5 Lf
Diphtheria Toxoid (DT)	-	2 Lf	-	2 Lf	2 Lf
Pertussis Toxoid (PT)	2 µg[a]	2 µg[a]	5 µg[a]	5 µg[a]	2.5 µg[b]
Filamentous hemagglutinin (FHA)	5 µg	5 µg	5 µg	5 µg	5 µg
Pertactin (PRN)	-	-	-	-	3 µg
Fimbriae type 2/3	-	-	-	-	5 µg
Excipients					
Adjuvant	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Phosphate 1.5 mg/dose (0.33 mg/dose as Al)
NaCl mg/dose	4.38	4.38	4.38	4.38	-
Water for Injection	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL

- **Study assessments**

Three blood draws (approximately 5 mL each of venous blood) were taken for the entire study period from 75 randomly pre-selected participants per each vaccine group: at Day 0 (Visit 1) just before study vaccination and was considered the baseline sample, at Day 28 (Visit 3) after vaccination to evaluate the immune response to study vaccines and were taken at 1 year *(*Visit 4*)* after vaccination to evaluate the antibody persistence.

- **Objectives**

The primary study objective was to assess the safety of BioNet Recombinant ap and BioNet Recombinant Tdap after a single dose vaccination compared to the licensed Tdap comparator (Adacel).

Statistical Hypothesis

H_0: Safety (pain) in BioNet investigational vaccines is worse than that in the licensed Tdap comparator (Adacel) by 12%.

H_A: Safety (pain) in BioNet investigational vaccines is better than or slightly worse not more than 12%

of that in the licensed Tdap comparator (Adacel).

- **Outcomes/endpoints**

Primary Endpoints:

- Percentages of participants with post-immunisation local and systemic reactions during 7 days following vaccination in BioNet Recombinant ap, BioNet Recombinant Tdap, Boostagen, VacPertagen and Adacel vaccine groups
- Percentages of participants with AEs reported during 28 days following vaccination in BioNet Recombinant ap, BioNet Recombinant Tdap, Boostagen, VacPertagen and Adacel vaccine groups
- Percentages of participants with SAEs reported from the day of vaccination until Day 28 following vaccination in BioNet Recombinant ap, BioNet Recombinant Tdap, Boostagen, VacPertagen and Adacel vaccine groups

Secondary Endpoints:

- Seroconversion rates as defined by proportion of participants with ≥ 4-fold increase from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 IU/mL and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL) of ELISA antibodies to PT and FHA in BioNet Recombinant ap, BioNet Recombinant Tdap, VacPertagen, Boostagen and Adacel vaccine groups
- Seroprotection rates as defined by proportion of participants with ≥ 0.1 IU/mL ELISA antibodies to tetanus and diphtheria in BioNet Recombinant Tdap, Boostagen and Adacel vaccine groups
- ELISA geometric mean antibody concentrations to PT, FHA, tetanus and diphtheria in BioNet Recombinant Tdap, Boostagen and Adacel vaccine groups and to only PT and FHA in BioNet Recombinant ap and VacPertagen vaccine groups
- Seroconversion rates as defined by proportion of participants with ≥ 4-fold increase from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 IU/mL and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL) at 28 days after vaccination with respect to baseline of PT neutralizing antibodies
- Geometric mean antibody concentrations to PT neutralizing antibodies in BioNet Recombinant ap, BioNet Recombinant Tdap, VacPertagen, Boostagen and Adacel vaccine groups

- **Sample size**

Sample size was calculated based on a non-inferiority test for safety by using pain as a common safety parameter in all populations with alpha level of 0.05 and 80% power with non-inferior margin of -12%, assuming the percentage of pain in the control group was 78% (Sricharoenchai et al, 2018). The sample size required for the study is 148 per arm.

- **Randomisation and Blinding (masking)**

Participants who provided their informed consent were enrolled into the study and randomized according the randomization list. Two randomization lists were computer generated. One list was for vaccine assignment (to receive one of the five vaccines: BioNet Recombinant ap or BioNet Recombinant Tdap (investigational vaccines) or VacPertagen, Boostagen or Adacel (reference vaccines) in a ratio of 1:1:1:1:1) and the other for immunogenicity testing (to randomly pre-select 75 blood

samples per each vaccine group of each study visit [Day 0: baseline, Day 28 and Day 336 after vaccination]).

The trial was an observer-blind study as the appearance of the pre-filled syringes/vial was different for BioNet Recombinant vaccines (ap and Tdap) and VacPertagen, Boostagen or Adacel vaccines. The study would be carried out in a blinded fashion until the database was locked for primary statistical analysis with data collected until Day 28 (Visit 3) after vaccination.

- **Statistical methods**

Analysis sets

From the protocol:

The Intention-To-Treat (ITT) set will exclude from analysis screened participants who received no vaccine injection.

The "Per Protocol" (PP) set includes the participants in the ITT set who are compliant with the protocol.

In case of the occurrence of premature discontinuation criteria and/or exclusion criteria, all the data from the participant will not be excluded from the PP analyses. For unblinding, the participant will also be excluded from PP analysis only for data collected after the code was broken. Because of the unpredictability of some problems, detailed considerations of the manner of dealing with irregularities will be deferred until the blind review of the data. The blind review will be done at the end of the vaccine period. The precise reasons for excluding participants from a Per Protocol analysis will be documented before unblinding.

The ITT and PP data set will be used for the analysis of short-term safety and also for the analysis of immunogenicity data.

Table templates in the SAP were indeed presented with both ITT and PP populations. In the CSR, mostly tables for the PP population were presented.

Methods for the analysis

From the protocol "Analysis of Immunogenicity in a Subset of 375 Participants (75 Participants per Vaccine Group)" (the same text was repeated in the CSR):

Percentage of participants with seroconversion as defined by a four-fold or higher (≥4-fold) response in ELISA anti-PT and anti-FHA antibody concentrations and PT neutralizing antibody titer measured by CHO cell assay, 28 days and 1 year following immunisation, as compared to baseline, will be computed for each vaccine group.

Percentage of participants with seroprotection as defined by ELISA anti-tetanus and anti-diphtheria antibody concentrations ≥ 0.1 IU/mL at baseline, 28 days and 1 year following immunisation, will be computed with its corresponding exact two-sided 95% CI for each vaccine group, except BioNet Recombinant ap and VacPertagen.

The difference in the percentages of participants between each of Investigational vaccine groups and Reference vaccine groups will be calculated along its two-sided 95% CI.

Geometric mean concentrations (GMCs) of ELISA anti-PT, anti-FHA, anti-tetanus and anti-diphtheria antibody and PT neutralizing antibody titer measured by CHO cell assay at baseline, 28 days and 1 year following immunisation will be calculated for each vaccine groups along with its two-sided 95% CI, by exponentiation the corresponding log-transformed mean and its 95% CI limits.

The ratio of the GMC in each of BioNet Recombinant ap/BioNet Recombinant Tdap groups to that in Adacel/VacPertagen/Boostagen groups will be provided. The log-transformed concentrations/titers will

be used to construct a two-sided 95% CI for the mean difference between the two vaccine groups using ANOVA. The mean difference and corresponding 95% CI limits will be exponentiated to obtain the GMC ratio and its 95% CI.

In addition, a reverse cumulative distribution (RCD) curve for each antigen will be created by vaccine group and visit for all subjects.

Remark: Immunogenicity data will be tabulated by vaccine groups with stratified ages.

Missing data

From the protocol: No information about the handling of missing data in immunogenicity endpoints was found in the protocol.

From the CSR: Missing immunogenicity data was found for a single participant aged 18-64 years in Adacel group. The participant was excluded from immunogenicity analysis since the participant missed Visit 3, hence the blood sample collection was not done.

Multiplicity

None described in the protocol. However, in the footnotes of the SAP template tables for GMC analyses, Bonferroni correction across the groups is mentioned. The same correction was mentioned in the tables in the synopsis and the results sections in the CSR, but it was not described in the methods section of the CSR.

Results

- **Participant flow**

Figure 9. Overall participant disposition at Day 28 after vaccination (Per Protocol population)

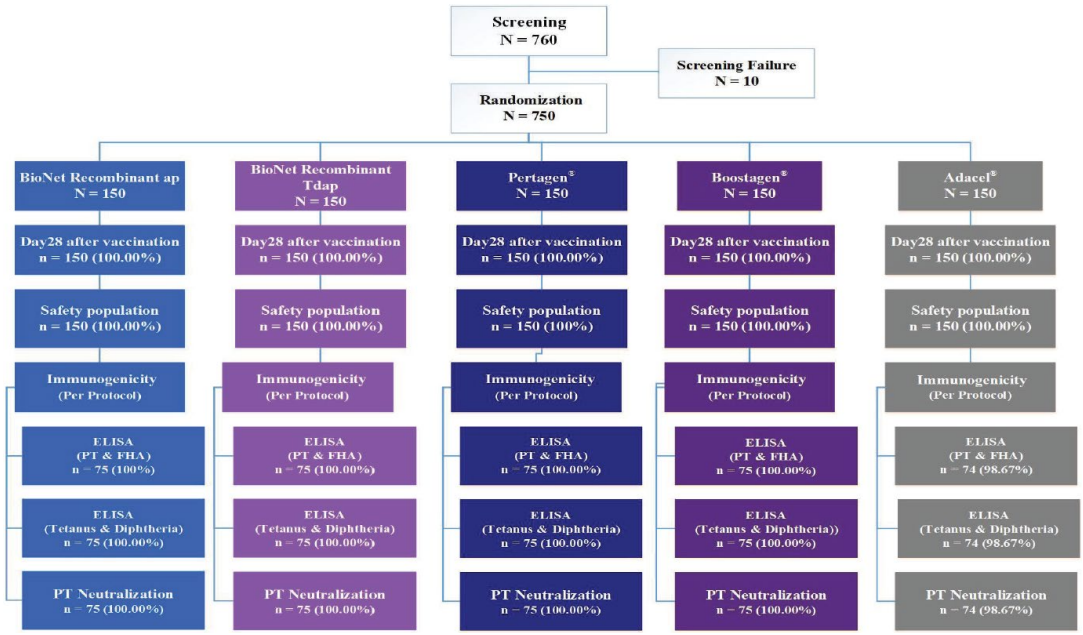


Figure 10. Participant aged 18-64 years old and disposition at Day 28 after vaccination (Per Protocol population)

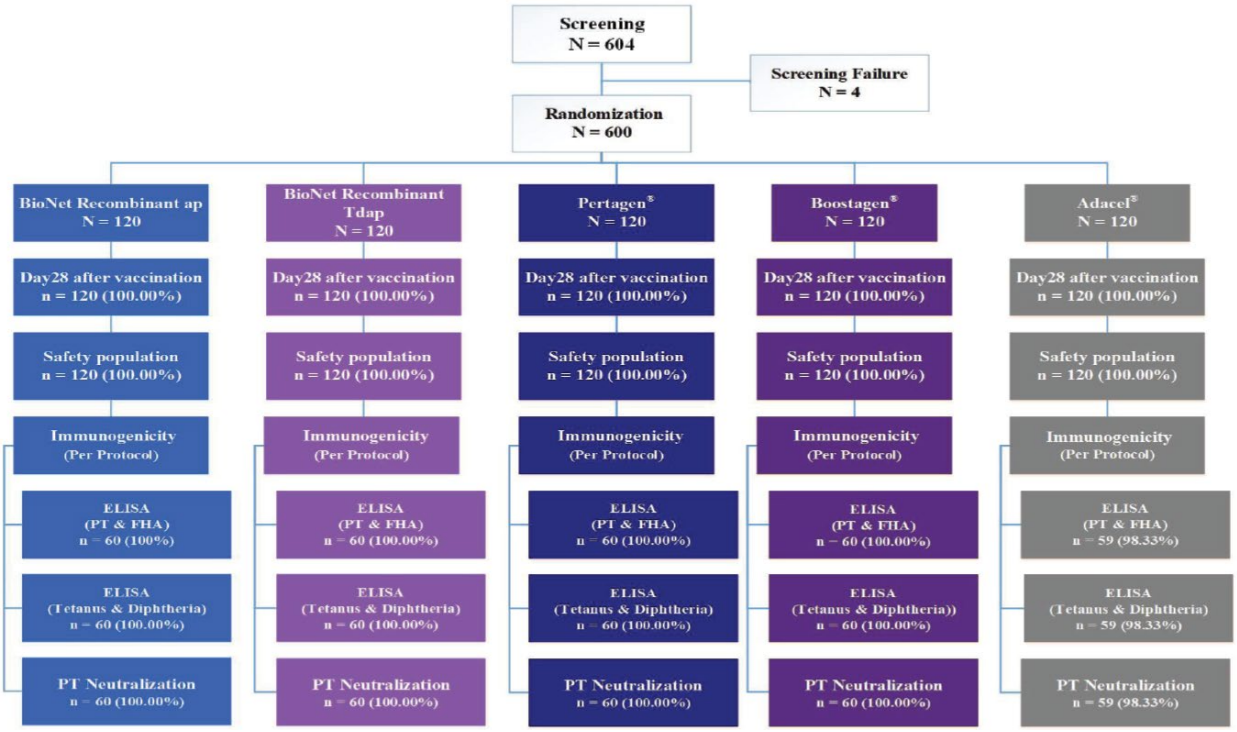
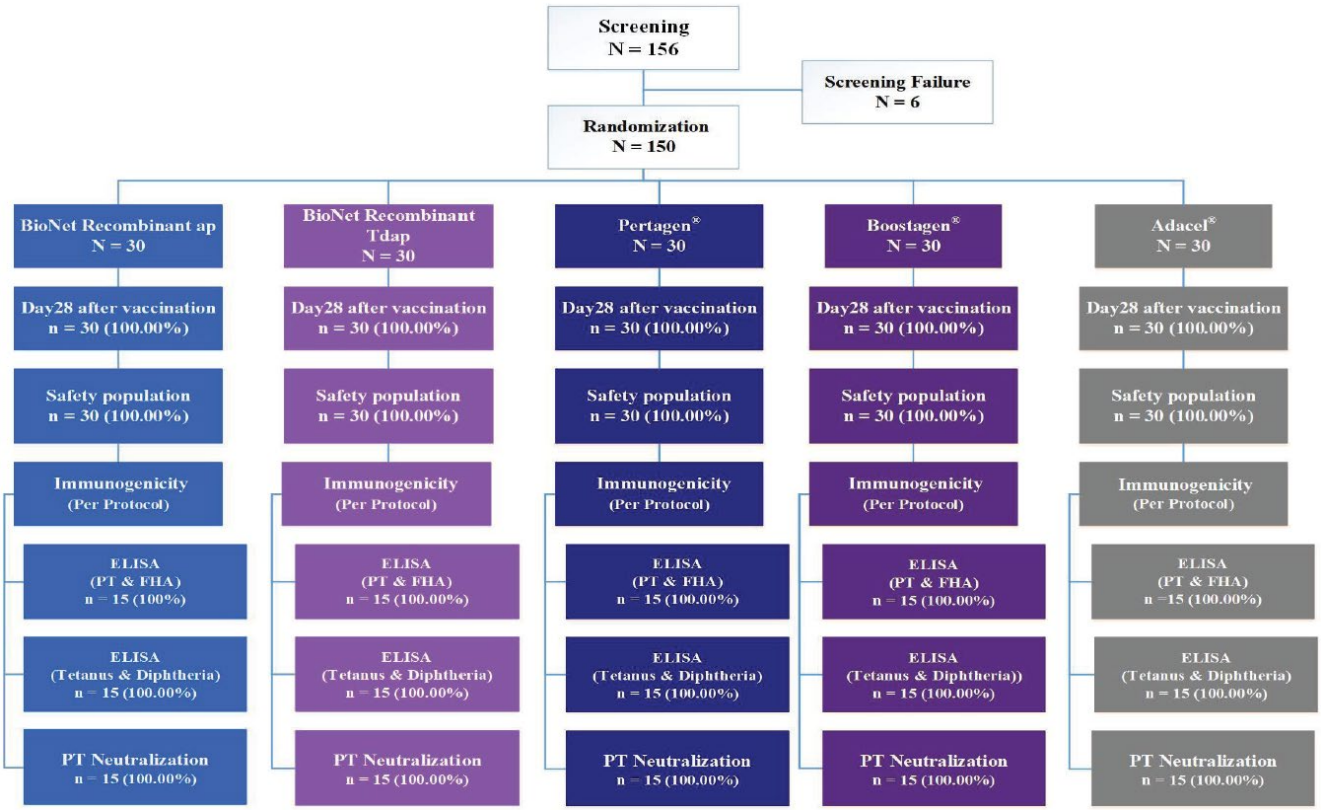


Figure 11. Participant aged 65-75 years old and disposition at Day 28 after vaccination (Per Protocol population)



- **Recruitment**

Date of first subject first visit for TDA202 study: 10 Feb 2020

Date of last subject for Day 28 follow-up (study completion): 21 Oct 2020

- **Conduct of the study**

Until Visit 3 (Day 28) of the study period, there was no change in the conduct of the study. There was no change to the planned analyses for data until visit 3 (Day 28) based on the final version of the SAP part I.

During the conduct of this study from Visit 1 (screening and vaccination day) to Visit 3 (Day 28 post-immunisation), there were 2 participants with major protocol deviations. Both the major protocol deviation did not affect participants safety or led to discontinuation from the study. One participant did not fullfill inclusion number 5 (the urine pregnancy test was not performed) at screening visit, but she was enrolled in the study. The urine pregnancy test was completed at the visit 2 and the result was negative. Another participant missed Visit 3, hence the blood sample collection was not done.

- **Baseline data**

Table 18. Summary of demographics at baseline of all participants

Participants status	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	Total	P-value
Demographics at baseline (Screening)							
Age (years)							
-N	150	150	150	150	150	750	0.8990 [3]
-Mean (SD)	44.22 (15.85)	45.24 (16.08)	43.88 (16.32)	44.77 (15.59)	44.21 (16.07)	44.46 (15.95)	
-Median	41.00	43.00	41.50	41.00	40.00	41.00	
-Min/Max	19 - 75	18 - 75	19 - 74	18 - 75	19 - 75	18 - 75	
Sex: n (%)							0.4277 [2]
-Male	43 (28.67)	44 (29.33)	34 (22.67)	49 (32.67)	43 (28.67)	213 (28.40)	
-Female	107 (71.33)	106 (70.67)	116 (77.33)	101 (67.33)	107 (71.33)	537 (71.60)	
Ethnicity: n (%)							- [1]
-Asian	150 (100.00)	150 (100.00)	150 (100.00)	150 (100.00)	150 (100.00)	750 (100.00)	
-Other	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Height (cm)							0.3511 [3]
-N	150	150	150	150	150	750	
-Mean (SD)	159.5 (8.78)	159.0 (8.17)	158.3 (7.86)	160.4 (8.37)	159.4 (8.03)	159.3 (8.25)	
-Median	158.5	159.0	157.0	160.0	158.0	158.0	
-Min/Max	138 - 183	138 - 185	137 - 179	143 - 190	143 - 185	137 - 190	
Weight (kg)							0.0257 [3]*
-N	150	150	150	150	150	750	
-Mean (SD)	65.65 (14.42)	64.18 (12.59)	61.92 (14.44)	65.69 (14.05)	63.02 (13.06)	64.09 (13.78)	

Table 19. Summary of demographics at baseline of participants aged 65-75 years old

Participants status	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	Total	P-value
Demographics at baseline (Screening)							
Age (years)							
-N	30	30	30	30	30	150	0.7664 [3]
-Mean (SD)	68.83 (3.12)	69.50 (2.89)	69.77 (2.71)	69.70 (3.51)	69.60 (3.30)	69.48 (3.09)	
-Median	69.00	69.00	70.00	69.50	69.00	69.00	
-Min/Max	65 - 75	65 - 75	65 - 74	65 - 75	65 - 75	65 - 75	
Sex: n (%)							0.2979 [2]
-Male	6 (20.00)	10 (33.33)	5 (16.67)	10 (33.33)	5 (16.67)	36 (24.00)	
-Female	24 (80.00)	20 (66.67)	25 (83.33)	20 (66.67)	25 (83.33)	114 (76.00)	
Ethnicity: n (%)							- [1]
-Asian	30 (100.00)	30 (100.00)	30 (100.00)	30 (100.00)	30 (100.00)	150 (100.00)	
-Other	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Height (cm)							0.1695 [4]
-N	30	30	30	30	30	150	
-Mean (SD)	154.9 (7.54)	156.0 (8.52)	152.3 (6.22)	156.6 (9.72)	153.3 (5.77)	154.6 (7.76)	
-Median	154.5	154.5	152.0	155.0	153.0	154.0	
-Min/Max	138 - 172	138 - 173	137 - 166	143 - 190	143 - 164	137 - 190	
Weight (kg)							0.0571 [4]
-N	30	30	30	30	30	150	
-Mean (SD)	57.70 (8.52)	63.60 (12.86)	56.70 (8.59)	61.13 (10.36)	59.83 (7.98)	59.79 (10.00)	

- **Numbers analysed**

A total of 760 volunteers were screened, of whom 750 participants (600 participants aged 18- 64 years and 150 participants aged 65-75 years) were enrolled and randomized into five vaccine groups (150 participants per vaccine group). The main reason for screening failures was having history of significant medical illness such as but not limited to immune deficiency, clinically significant psychiatric, hematologic, pulmonary, cardiovascular, or hepatic, renal, splenic or thymic functional abnormality in participants aged 18- 64 years and having any active clinically significant finding or life-threatening disease that, in the opinion of the investigator, would increase the risk of the individual's having an adverse outcome by participating in the study in participants aged 65-75 years.

All enrolled participants completed the study at Day 28 post-vaccination and were included in the safety analysis. One participant aged 18-64 years in Adacel group was excluded from the immunogenicity analysis for data up to Day 28 since the participant missed Visit 3, hence the blood sample collection was not done. Therefore, 374 participants (75 participants each in the BioNet Recombinant Tdap, BioNet Recombinant ap, VacPertagen and Boostagen vaccine groups and 74 in the Adacel group) were included in the ELISA immunogenicity analysis for anti-PT, anti-FHA and PT neutralization analysis (299 participants aged 18-64 years and 75 participants aged 65-75 years).

- **Outcomes and estimation**

Analysis of Immunogenicity Data at Day 28 Post-vaccination

Seroconversion rates (PT, FHA)

In all participants, seroconversion rate of anti-PT antibody at 28 days after vaccination compared to baseline was higher in VacPertagen [100.00% (95% CI 95.20-100.00)] than the seroconversion rate in Adacel group [74.32% (95% CI 62.84 - 83.78).

In participants aged 18-64 years, seroconversion rate of anti-PT antibody at 28 days after vaccination compared to baseline was higher in the VacPertagen group than in the Adacel group with a difference in seroconversion rate of 22.03% (95%CI 13.33-34.19). Also in participants aged 65-75 years,

seroconversion rate in the VacPertagen group was higher than those in the Adacel group with a difference in seroconversion rate of 40.00% (95%CI 15.24-64.61). The 3 other BioNet vaccines (containing aPgen in same/lower amounts and/or also include Td antigens) also induced higher seroconversion rates 28 days after vaccination compared to Adacel.

In all participants, seroconversion rate of anti-FHA antibody at 28 days after vaccination compared to baseline was similar in all vaccine groups e.g. VacPertagen group [97.33% (95% CI 90.70-99.6)] vs Adacel group [93.24% (95% CI 84.93-97.77). Also when stratified by age, seroconversion rates of anti-FHA antibody at 28 days after vaccination compared to baseline were similar.

Table 20. Seroconversion rates as defined by the proportion of participants in anti-PT antibody concentrations at 28 days after vaccination compared to baseline as assessed by ELISA of all participants by vaccine groups (Per Protocol population)

Seroconversion rates	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	P-value
	(N=75)	(N=75)	(N=75)	(N=75)	(N=74)	
	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	
Anti-PT IgG	71 (94.67)	68 (90.67)	75 (100.00)	74 (98.67)	55 (74.32)	<0.0001 [1]*
	(86.90-98.53)	(81.71-96.16)	(95.20-100.00)	(92.79-99.97)	(62.84-83.78)	

a: Seroconversion as defined by proportions of participants,≥ 4-fold from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL)

95% CI based on Clopper-Pearson method.

Note:
[1] Overall p-value (2-sided) based on Fisher's exact test
* P-value ≤ 0.05 is considered statistically significant.

Table 21. Seroconversion rates as defined by the proportion of participants in anti-PT antibody concentrations at 28 days after vaccination compared to baseline as assessed by ELISA of participants aged 65-75 years old by vaccine groups (Per Protocol population)

Seroconversion rates	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	P-value
	(N=15)	(N=15)	(N=15)	(N=15)	(N=15)	
	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	
Anti-PT IgG	13 (86.67)	13 (86.67)	15 (100.00)	14 (93.33)	9 (60.00)	0.0340 [1]*
	(59.54-98.34)	(59.54-98.34)	(78.20-100.00)	(68.05-99.83)	(32.29-83.66)	

a: Seroconversion as defined by proportions of participants, ≥ 4-fold from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL)
95% CI based on Clopper-Pearson method.

Note:
[1] Overall p-value (2-sided) based on Fisher's exact test
* P-value ≤ 0.05 is considered statistically significant.

Table 42. Seroconversion rates as defined by the proportion of participants in anti-FHA antibody concentrations at 28 days after vaccination compared to baseline as assessed by ELISA of all participants by vaccine groups (Per Protocol population)

Seroconversion rates	BioNet Recombinant ap (N=75) n (%) (95% CI)	BioNet Recombinant Tdap (N=75) n (%) (95% CI)	Pertagen® (N=75) n (%) (95% CI)	Boostagen® (N=75) n (%) (95% CI)	Adacel® (N=74) n (%) (95% CI)	P-value
Anti-FHA IgG	71 (94.67) (86.90-98.53)	67 (89.33) (80.06-95.28)	73 (97.33) (90.70-99.68)	70 (93.33) (85.12-97.80)	69 (93.24) (84.93-97.77)	0.3994 [1]

a: Seroconversion as defined by proportions of participants, ≥ 4-fold from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL)

95% CI based on Clopper-Pearson method.

Note:
[1] Overall p-value (2-sided) based on Fisher's exact test
* P-value ≤ 0.05 is considered statistically significant.

Table 53. Seroconversion rates as defined by the proportion of participants in anti-FHA antibody concentrations at 28 days after vaccination compared to baseline as assessed by ELISA of participants aged 65-75 years old by vaccine groups (Per Protocol population)

Seroconversion rates	BioNet Recombinant ap (N=15) n (%) (95% CI)	BioNet Recombinant Tdap (N=15) n (%) (95% CI)	Pertagen® (N=15) n (%) (95% CI)	Boostagen® (N=15) n (%) (95% CI)	Adacel® (N=15) n (%) (95% CI)	P-value
Anti-FHA IgG	14 (93.33) (68.05-99.83)	14 (93.33) (68.05-99.83)	15 (100.00) (78.20-100.00)	15 (100.00) (78.20-100.00)	14 (93.33) (68.05-99.83)	1.0000 [1]

a: Seroconversion as defined by proportions of participants, ≥ 4-fold from baseline titers of ≥ 5.0 and < 20 IU/mL, ≥ 2-fold increase from baseline titers of ≥ 20 and ≥ 20 IU/mL from seronegative baseline (< 5 IU/mL)

95% CI based on Clopper-Pearson method.

Note:
[1] Overall p-value (2-sided) based on Fisher's exact test

Anti-PT and anti-FHA Geometric Mean Antibody Concentrations (GMCs):

At 28 days after vaccination, GMCs for anti-PT-IgG antibodies were higher in the VacPertagen group [371.83 IU/mL (95% CI 292.76-472.25)] compared to the Adacel group [50.84 IU/mL (95% CI 39.26-65.84)]. Anti-PT GMCs following VacPertagen vaccination were overall comparable in both age groups 28 days after vaccination.

At 28 days after vaccination, GMCs for anti-FHA-IgG antibodies were higher in the VacPertagen group [451.62 IU/mL (95% CI 373.46-546.12)] than in the Adacel group [207.58 IU/mL (95% CI 171.33-251.50)]. In participants aged 65-75 years, GMCs for anti-FHA-IgG antibodies were numerically higher in the in the VacPertagen group [387.88 IU/mL (95% CI 218.85-687.45)] than in the Adacel group [213.11 IU/mL (95% CI 123.84-366.72)].

Table 64. Comparison of anti-PT GMCs (IU/mL) between baseline and Day 28 after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	6.16	147.64	23.96	<0.0001 [2]*
(N=75)	(4.51-8.42)	(116.94-186.40)	(18.23-31.47)	
BioNet Recombinant Tdap	8.47	100.56	11.87	<0.0001 [2]*
(N=75)	(6.24-11.50)	(80.00-126.41)	(9.25-15.22)	
Pertagen®	7.05	371.83	52.72	<0.0001 [2]*
(N=75)	(5.36-9.28)	(292.76-472.25)	(40.78-68.16)	
Boostagen®	9.08	226.20	24.91	<0.0001 [2]*
(N=75)	(6.69-12.32)	(181.17-282.42)	(18.86-32.90)	
Adacel®	5.74	50.84	8.86	<0.0001 [2]*
(N=74)	(4.28-7.70)	(39.26-65.84)	(7.09-11.07)	
P-value[b]	0.1158 [1]	<0.0001 [1]*	<0.0001 [1]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.40 (0.25-0.64)*	BioNet Recombinant ap - Pertagen® 0.45 (0.27-0.76) *	
		BioNet Recombinant ap - Boostagen® 0.65 (0.41-1.05)	BioNet Recombinant ap - Boostagen® 0.96 (0.58-1.61)	
		BioNet Recombinant ap - Adacel® 2.90 (1.81-4.67) *	BioNet Recombinant ap - Adacel® 2.70 (1.61-4.53) *	
		BioNet Recombinant Tdap - Pertagen® 0.27 (0.17-0.43)*	BioNet Recombinant Tdap - Pertagen® 0.23 (0.13-0.38) *	
		BioNet Recombinant Tdap - Boostagen® 0.44 (0.28-0.71) *	BioNet Recombinant Tdap - Boostagen® 0.48 (0.28-0.80)*	
		BioNet Recombinant Tdap - Adacel® 1.98 (1.23-3.18)*	BioNet Recombinant Tdap - Adacel ® 1.34 (0.80-2.24)	
		Pertagen®- Adacel® 7.31 (4.55-11.76)*	Pertagen®- Adacel® 5.95 (3.55-9.97)*	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis.
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 25. Comparison of anti-PT GMCs (IU/mL) between baseline and Day 28 after vaccination as assessed by ELISA in participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	4.56	94.06	20.62	<0.0001 [3]*
(N=15)	(2.67-7.79)	(42.92-206.14)	(10.22-41.62)	
BioNet Recombinant Tdap	5.78	66.83	11.56	<0.0001 [3]*
(N=15)	(2.88-11.60)	(26.33-169.63)	(6.34-21.09)	
Pertagen®	6.77	330.17	48.81	<0.0001 [3]*
(N=15)	(3.27-13.99)	(168.85-645.63)	(24.94-95.52)	
Boostagen®	7.31	162.64	22.24	<0.0001 [3]*
(N=15)	(4.13-12.94)	(84.07-314.63)	(12.19-40.56)	
Adacel®	5.06	34.66	6.85	<0.0001 [3]*
(N=15)	(2.38-10.75)	(15.66-76.67)	(5.38-8.73)	
P-value[b]	0.7044 [1]	0.0005 [2]*	<0.0001 [2]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.28 (0.06-1.25)	BioNet Recombinant ap - Pertagen® 0.42 (0.14-1.30)	
		BioNet Recombinant ap - Boostagen® 0.58 (0.13-2.54)	BioNet Recombinant ap - Boostagen® 0.93 (0.30-2.85)	
		BioNet Recombinant ap - Adacel® 2.71 (0.62-11.93)	BioNet Recombinant ap - Adacel® 3.01 (0.98-9.25)	
		BioNet Recombinant Tdap - Pertagen® 0.20 (0.05-0.89)*	BioNet Recombinant Tdap - Pertagen® 0.24 (0.08-0.73)*	
		BioNet Recombinant Tdap - Boostagen® 0.41 (0.09-1.81)	BioNet Recombinant Tdap - Boostagen® 0.52 (0.17-1.60)	
		BioNet Recombinant Tdap - Adacel® 1.93 (0.44-8.47)	BioNet Recombinant Tdap - Adacel® 1.69 (0.55-5.19)	
		Pertagen®- Adacel® 9.53 (2.17-41.86)*	Pertagen®- Adacel® 7.12 (2.32-21.88)*	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis.
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 26. Comparison of anti-FHA GMCs (IU/mL) between baseline and Day 28 after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	19.63	282.46	14.39	<0.0001 [3]*
(N=75)	(14.66-26.29)	(240.78-331.36)	(10.73-19.29)	
BioNet Recombinant Tdap	23.70	217.75	9.19	<0.0001 [3]*
(N=75)	(17.80-31.55)	(178.50-265.63)	(7.13-11.84)	
Pertagen®	19.34	451.62	23.35	<0.0001 [3]*
(N=75)	(14.82-25.25)	(373.46-546.12)	(17.94-30.39)	
Boostagen®	28.43	367.18	12.92	<0.0001 [3]*
(N=75)	(21.32-37.92)	(307.12-438.98)	(9.54-17.48)	
Adacel®	17.11	207.58	12.13	<0.0001 [3]*
(N=74)	(12.70-23.06)	(171.33-251.50)	(9.36-15.72)	
P-value[b]	0.1066 [2]	<0.0001 [1]*	<0.0001 [2]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.63 (0.43-0.90)*	BioNet Recombinant ap - Pertagen® 0.62 (0.36-1.07)	
		BioNet Recombinant ap - Boostagen® 0.77 (0.53-1.11)	BioNet Recombinant ap - Boostagen® 1.11 (0.64-1.93)	
		BioNet Recombinant ap - Adacel® 1.36 (0.94-1.97)	BioNet Recombinant ap - Adacel® 1.19 (0.68-2.06)	
		BioNet Recombinant Tdap - Pertagen® 0.48 (0.33-0.70) *	BioNet Recombinant Tdap - Pertagen® 0.39 (0.23-0.68) *	
		BioNet Recombinant Tdap - Boostagen® 0.59 (0.41-0.86)*	BioNet Recombinant Tdap - Boostagen® 0.71 (0.41-1.23)	
		BioNet Recombinant Tdap - Adacel® 1.05 (0.72-1.52)	BioNet Recombinant Tdap - Adacel® 0.76 (0.44-1.32)	
		Pertagen® - Adacel® 2.18 (1.50-3.15)*	Pertagen® - Adacel® 1.92 (1.11-3.35)*	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis.
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 27. Comparison of anti-FHA GMCs (IU/mL) between baseline and Day 28 after vaccination as assessed by ELISA in all participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	21.34	206.91	9.69	<0.0001 [3]*
(N=15)	(13.98-32.59)	(139.20-307.55)	(6.25-15.03)	
BioNet Recombinant Tdap	16.14	230.02	14.25	<0.0001 [3]*
(N=15)	(8.72-29.86)	(111.54-474.35)	(7.69-26.43)	
Pertagen®	20.08	387.88	19.32	<0.0001 [3]*
(N=15)	(10.85-37.18)	(218.85-687.45)	(11.81-31.59)	
Boostagen®	23.67	404.38	17.09	<0.0001 [3]*
(N=15)	(13.00-43.07)	(261.46-625.41)	(9.12-32.00)	
Adacel®	13.29	213.11	16.03	<0.0001 [3]*
(N=15)	(6.41-27.56)	(123.84-366.72)	(8.31-30.94)	
P-value[b]	0.6088 [2]	0.0798 [1]	0.4269 [2]	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.53 (0.19-1.52)	BioNet Recombinant ap - Pertagen® 0.50 (0.17-1.50)	
		BioNet Recombinant ap - Boostagen® 0.51 (0.18-1.45)	BioNet Recombinant ap - Boostagen® 0.57 (0.19-1.70)	
		BioNet Recombinant ap - Adacel® 0.97 (0.34-2.76)	BioNet Recombinant ap - Adacel® 0.60 (0.20-1.81)	
		BioNet Recombinant Tdap - Pertagen® 0.59 (0.21-1.69)	BioNet Recombinant Tdap - Pertagen® 0.74 (0.25-2.21)	
		BioNet Recombinant Tdap - Boostagen® 0.57 (0.20-1.62)	BioNet Recombinant Tdap - Boostagen® 0.83 (0.28-2.49)	
		BioNet Recombinant Tdap - Adacel® 1.08 (0.38-3.07)	BioNet Recombinant Tdap - Adacel® 0.89 (0.30-2.66)	
		Pertagen® - Adacel® 1.82 (0.64-5.17)	Pertagen® - Adacel® 1.20 (0.40-3.60)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis.
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Anti-PT neutralizing antibodies

In all participants, seroconversion rate of PT neutralizing antibodies at 28 days after vaccination compared to baseline were higher in the VacPertagen group [96.00% (95% CI 88.75-99.17)] than in the Adacel group [64.86% (95% CI 52.89-75.61). Also when stratified by age, there was a trend for higher seroconversion rates of PT neutralizing antibodies and GMTs at 28 days after vaccination in the VacPertagen group compared to the Adacel group.

Table 28. Comparison of PT neutralising GMTs (IU/ml) between baseline and Day 28 after vaccination as assessed by PT neutralizing assay in CHO cells of all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	4.43	101.10	22.80	<0.0001 [2]*
(N=75)	(3.25-6.05)	(75.85-134.77)	(17.10-30.39)	
BioNet Recombinant Tdap	5.61	75.52	13.47	<0.0001 [2]*
(N=75)	(4.05-7.76)	(59.50-95.87)	(10.13-17.91)	
Pertagen®	5.47	253.34	46.32	<0.0001 [2]*
(N=75)	(4.02-7.45)	(181.35-353.92)	(34.40-62.36)	
Boostagen®	5.93	158.51	26.72	<0.0001 [2]*
(N=75)	(4.31-8.17)	(123.57-203.32)	(20.23-35.31)	
Adacel®	4.41	29.61	6.72	<0.0001 [2]*
(N=74)	(3.31-5.86)	(21.42-40.93)	(5.17-8.73)	
P-value[b]	0.4682 [1]	<0.0001 [1]*	<0.0001 [1]*	
The ratio of GMT or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.40 (0.22-0.71)*	BioNet Recombinant ap - Pertagen® 0.49 (0.28-0.87)*	
		BioNet Recombinant ap - Boostagen® 0.64 (0.36-1.14)	BioNet Recombinant ap - Boostagen® 0.85 (0.48-1.50)	
		BioNet Recombinant ap - Adacel® 3.42 (1.91-6.10)*	BioNet Recombinant ap - Adacel® 3.39 (1.92-5.98)*	
		BioNet Recombinant Tdap - Pertagen® 0.30 (0.17-0.53)*	BioNet Recombinant Tdap - Pertagen® 0.29 (0.17-0.51)*	
		BioNet Recombinant Tdap - Boostagen® 0.48 (0.27-0.85)*	BioNet Recombinant Tdap - Boostagen® 0.50 (0.29-0.89)*	
		BioNet Recombinant Tdap - Adacel® 2.55 (1.43-4.56) *	BioNet Recombinant Tdap - Adacel® 2.00 (1.14-3.53)*	

Geometric mean fold rise (GMFR) is geometric mean of the raio of PT neutralizing antibody titer at Day 28 after vaccination to PT neutralizing antibody titer at baseline.
The ratio of GMT or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 29. Comparison of PT neutralising GMTs (IU/ml) between baseline and Day 28 after vaccination as assessed by PT neutralizing assay in CHO cells of participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	3.26	77.21	23.69	<0.0001 [3]*
(N=15)	(2.03-5.24)	(33.59-177.45)	(13.10-42.85)	
BioNet Recombinant Tdap	4.20	69.43	16.51	<0.0001 [3]*
(N=15)	(2.27-7.79)	(28.26-170.59)	(8.45-32.29)	
Pertagen®	6.07	241.15	39.72	<0.0001 [3]*
(N=15)	(3.06-12.03)	(86.51-672.19)	(17.76-88.83)	
Boostagen®	3.69	122.27	33.17	<0.0001 [3]*
(N=15)	(2.06-6.61)	(64.12-233.14)	(19.21-57.29)	
Adacel®	4.86	25.26	5.20	<0.0001 [3]*
(N=15)	(2.30-10.28)	(10.92-58.45)	(3.12-8.65)	
P-value[b]	0.6817 [1]	0.0035 [1]*	<0.0001 [2]*	
The ratio of GMT or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.32 (0.06-1.65)	BioNet Recombinant ap - Pertagen® 0.60 (0.18-2.00)	
		BioNet Recombinant ap - Boostagen® 0.63 (0.12-3.25)	BioNet Recombinant ap - Boostagen® 0.71 (0.21-2.40)	
		BioNet Recombinant ap - Adacel® 3.06 (0.59-15.72)	BioNet Recombinant ap - Adacel® 4.56 (1.36-15.30) *	
		BioNet Recombinant Tdap - Pertagen® 0.29 (0.06-1.48)	BioNet Recombinant Tdap - Pertagen® 0.42 (0.12-1.40)	
		BioNet Recombinant Tdap - Boostagen® 0.57 (0.11-2.92)	BioNet Recombinant Tdap - Boostagen® 0.50 (0.15-1.67)	
Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMFR (95% CI)	
		BioNet Recombinant Tdap - Adacel® 2.75 (0.53-14.14)	BioNet Recombinant Tdap - Adacel® 3.18 (0.95-10.67)	

Geometric mean fold rise (GMFR) is geometric mean of the raio of PT neutralizing antibody titer at Day 28 after vaccination to PT neutralizing antibody titer at baseline.
The ratio of GMT or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Antibody responses 1 year after vaccination

Table 30. Comparison of anti-PT GMC (IU/ml) between baseline and Day 336 after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 336 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	6.06	31.76	5.24	<0.0001 [2]*
(N=70)	(4.36-8.44)	(23.31-43.26)	(4.02-6.82)	
BioNet Recombinant Tdap	8.57	26.69	3.11	<0.0001 [2]*
(N=72)	(6.27-11.73)	(21.35-33.36)	(2.47-3.92)	
Pertagen®	6.59	70.90	10.76	<0.0001 [2]*

(N=71)	(5.00-8.68)	(52.30-96.11)	(8.19-14.13)	
Boostagen®	8.81	54.32	6.17	<0.0001 [2]*
(N=72)	(6.44-12.06)	(43.43-67.94)	(4.73-8.04)	
Adacel®	5.64	14.12	2.50	<0.0001 [2]*
(N=71)	(4.19-7.58)	(10.98-18.14)	(2.05-3.06)	
P-value[b]	0.1322 [1]	<0.0001 [1]*	<0.0001 [1]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.45 (0.26-0.76)*	BioNet Recombinant ap - Pertagen® 0.49 (0.30-0.80)*	
		BioNet Recombinant ap - Boostagen® 0.58 (0.34-0.99) *	BioNet Recombinant ap - Boostagen® 0.85 (0.52-1.40)	
		BioNet Recombinant ap - Adacel® 2.25 (1.32-3.83)*	BioNet Recombinant ap - Adacel® 2.09 (1.27-3.44)*	
		BioNet Recombinant Tdap - Pertagen® 0.38 (0.22-0.64)*	BioNet Recombinant Tdap - Pertagen® 0.29 (0.18-0.48)*	
		BioNet Recombinant Tdap - Boostagen® 0.49 (0.29-0.83)*	BioNet Recombinant Tdap - Boostagen® 0.50 (0.31-0.83)*	
		BioNet Recombinant Tdap - Adacel® 1.89 (1.11-3.21) *	BioNet Recombinant Tdap - Adacel® 1.24 (0.76-2.04)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 336 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 336 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 31. Comparison of anti-PT GMC (IU/ml) between baseline and Day 336 after vaccination as assessed by ELISA in participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 336 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	4.37	18.35	4.20	0.0002 [3]*
(N=13)	(2.34-8.16)	(8.17-41.20)	(2.25-7.84)	
BioNet Recombinant Tdap	5.39	17.66	3.28	0.0004 [3]*
(N=14)	(2.58-11.26)	(7.59-41.06)	(1.89-5.66)	
Pertagen®	6.90	64.26	9.31	<0.0001 [3]*
(N=14)	(3.15-15.13)	(23.50-175.71)	(3.96-21.91)	
Boostagen®	6.79	34.47	5.07	0.0002 [3]*
(N=13)	(3.55-12.98)	(16.36-72.65)	(2.53-10.19)	
Adacel®	4.85	9.72	2.00	0.0001 [3]*
(N=13)	(2.22-10.61)	(4.34-21.76)	(1.51-2.66)	
P-value[b]	0.7551 [1]	0.0036 [1]*	0.0085 [2]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.23 (0.05-1.11)	BioNet Recombinant ap - Pertagen® 0.45 (0.14-1.51)	
		BioNet Recombinant ap - Boostagen® 0.58 (0.12-2.89)	BioNet Recombinant ap - Boostagen® 0.83 (0.24-2.82)	
		BioNet Recombinant ap - Adacel® 2.22 (0.45-10.98)	BioNet Recombinant ap - Adacel® 2.10 (0.61-7.15)	
		BioNet Recombinant Tdap - Pertagen® 0.18 (0.04-0.82)*	BioNet Recombinant Tdap - Pertagen® 0.35 (0.11-1.15)	
		BioNet Recombinant Tdap - Boostagen® 0.45 (0.09-2.14)	BioNet Recombinant Tdap - Boostagen® 0.65 (0.19-2.15)	
		BioNet Recombinant Tdap - Adacel® 1.69 (0.35-8.13)	BioNet Recombinant Tdap - Adacel® 1.63 (0.49-5.45)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 336 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 336 after vaccination
b : Compared between vaccine groups
Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 32. Comparison of anti-FHA GMCs (IU/ml) between baseline and Day 336 after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 336 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	19.05	100.55	5.28	<0.0001 [3]*
(N=70)	(13.98-25.96)	(81.89-123.46)	(4.09-6.81)	
BioNet Recombinant Tdap	24.45	77.34	3.16	<0.0001 [3]*
(N=72)	(18.34-32.59)	(64.24-93.11)	(2.57-3.89)	
Pertagen®	19.01	142.46	7.50	<0.0001 [3]*
(N=71)	(14.36-25.16)	(114.38-177.44)	(5.74-9.78)	
Boostagen®	28.56	142.56	4.99	<0.0001 [3]*
(N=72)	(21.24-38.40)	(113.45-179.15)	(3.72-6.70)	
Adacel®	17.68	85.26	4.82	<0.0001 [3]*
(N=71)	(13.04-23.98)	(69.77-104.18)	(3.78-6.15)	
P-value[b]	0.1124 [2]	<0.0001 [1]*	0.0001 [2]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.71 (0.46-1.07)	BioNet Recombinant ap - Pertagen® 0.70 (0.42-1.18)	
		BioNet Recombinant ap - Boostagen® 0.71 (0.46-1.07)	BioNet Recombinant ap - Boostagen® 1.06 (0.63-1.76)	
		BioNet Recombinant ap - Adacel® 1.18 (0.78-1.79)	BioNet Recombinant ap - Adacel® 1.09 (0.66-1.83)	
		BioNet Recombinant Tdap - Pertagen® 0.54 (0.36-0.82)*	BioNet Recombinant Tdap - Pertagen® 0.42 (0.25-0.70)*	
		BioNet Recombinant Tdap - Boostagen® 0.54 (0.36-0.82)*	BioNet Recombinant Tdap - Boostagen® 0.63 (0.38-1.05)	
		BioNet Recombinant Tdap - Adacel® 0.91 (0.60-1.38)	BioNet Recombinant Tdap - Adacel® 0.66 (0.39-1.09)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 336 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 336 after vaccination
b : Compared between vaccine groups
Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 33. Comparison of anti-FHA GMCs (IU/ml) between baseline and Day 336 after vaccination as assessed by ELISA in participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	Day 336 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	19.48	75.46	3.87	<0.0001 [3]*
(N=13)	(12.55-30.25)	(45.35-125.55)	(2.71-5.54)	
BioNet Recombinant Tdap	16.98	61.36	3.61	<0.0001 [3]*
(N=14)	(8.82-32.70)	(32.62-115.43)	(2.35-5.57)	
Pertagen®	19.25	129.95	6.75	<0.0001 [3]*
(N=14)	(9.96-37.22)	(67.30-250.90)	(3.87-11.76)	
Boostagen®	25.31	138.45	5.47	<0.0001 [3]*

(N=13)	(13.04-49.12)	(78.15-245.30)	(2.91-10.29)	
Adacel®	15.01	74.03	4.93	<0.0001 [3]*
(N=13)	(6.75-33.35)	(40.08-136.74)	(2.82-8.64)	
P-value[b]	0.8034 [2]	0.1654 [1]	0.3274 [2]	
The ratio of GMC or GMFC between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.58 (0.19-1.82)	BioNet Recombinant ap - Pertagen® 0.57 (0.22-1.52)	
		BioNet Recombinant ap - Boostagen® 0.54 (0.17-1.74)	BioNet Recombinant ap - Boostagen® 0.71 (0.26-1.91)	
		BioNet Recombinant ap - Adacel® 1.02 (0.32-3.25)	BioNet Recombinant ap - Adacel® 0.79 (0.29-2.12)	
		BioNet Recombinant Tdap - Pertagen® 0.47 (0.15-1.45)	BioNet Recombinant Tdap - Pertagen® 0.54 (0.21-1.40)	
		BioNet Recombinant Tdap - Boostagen® 0.44 (0.14-1.39)	BioNet Recombinant Tdap - Boostagen® 0.66 (0.25-1.75)	
		BioNet Recombinant Tdap - Adacel® 0.83 (0.27-2.59)	BioNet Recombinant Tdap - Adacel® 0.73 (0.28-1.94)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 336 after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and Day 336 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Antibody responses 3 year after vaccination

Anti-PT GMCs

Table 34. Comparison of anti-PT GMCs (IU/mL) between baseline and 3 years after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	3 years after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	6.55	22.14	3.38	<0.0001 [2]*
(N=59)	(4.61-9.31)	(16.32-30.04)	(2.52-4.53)	
BioNet Recombinant Tdap	8.60	16.01	1.86	<0.0001 [2]*
(N=62)	(6.07-12.20)	(12.17-21.07)	(1.50-2.32)	
Pertagen®	7.30	43.00	5.89	<0.0001 [2]*
(N=58)	(5.39-9.89)	(31.47-58.75)	(4.54-7.65)	
Boostagen®	9.14	31.60	3.46	<0.0001 [2]*
(N=56)	(6.41-13.03)	(24.76-40.32)	(2.58-4.64)	
Adacel®	5.41	8.75	1.62	<0.0001 [2]*
(N=59)	(3.96-7.39)	(6.55-11.70)	(1.32-1.98)	
P-value[b]	0.2188 [1]	<0.0001 [1]*	<0.0001 [1]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.51 (0.29-0.91)*	BioNet Recombinant ap - Pertagen® 0.57 (0.34-0.96)*	
		BioNet Recombinant ap - Boostagen® 0.70 (0.39-1.25)	BioNet Recombinant ap - Boostagen® 0.98 (0.58-1.64)	
		BioNet Recombinant ap - Adacel® 2.53 (1.43-4.48)*	BioNet Recombinant ap - Adacel® 2.09 (1.26-3.48)*	
		BioNet Recombinant Tdap - Pertagen® 0.37 (0.21-0.66)*	BioNet Recombinant Tdap - Pertagen® 0.32 (0.19-0.52)*	

	BioNet Recombinant Tdap - Boostagen® 0.51 (0.29-0.90)*	BioNet Recombinant Tdap - Boostagen® 0.54 (0.32-0.90)*
	BioNet Recombinant Tdap - Adacel® 1.83 (1.04-3.22)*	BioNet Recombinant Tdap - Adacel® 1.15 (0.70-1.90)

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 3 years after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and 3 years after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 35. Comparison of anti-PT GMCs (IU/mL) between baseline and 3 years after vaccination as assessed by ELISA in participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	3 years after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	4.25	10.47	2.46	0.0191 [3]*
(N=11)	(2.26-8.01)	(4.46-24.63)	(1.20-5.06)	
BioNet Recombinant Tdap	5.42	10.73	1.98	0.0046 [3]*
(N=12)	(2.25-13.04)	(4.41-26.09)	(1.29-3.03)	
Pertagen®	8.88	40.45	4.55	0.0007 [3]*
(N=11)	(3.70-21.33)	(12.31-132.88)	(2.24-9.27)	
Boostagen®	5.98	16.19	2.71	0.0195 [3]*
(N=10)	(2.56-14.00)	(7.63-34.36)	(1.22-5.99)	
Adacel®	5.38	8.25	1.53	0.0209 [3]*
(N=10)	(1.95-14.84)	(3.17-21.44)	(1.08-2.17)	
P-value[b]	0.7352 [2]	0.0319 [1]*	0.1683 [1]	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.26 (0.05-1.48)	BioNet Recombinant ap - Pertagen® 0.54 (0.17-1.70)	
		BioNet Recombinant ap - Boostagen® 0.65 (0.11-3.87)	BioNet Recombinant ap - Boostagen® 0.91 (0.28-2.94)	
		BioNet Recombinant ap - Adacel® 1.27 (0.21-7.60)	BioNet Recombinant ap - Adacel® 1.61 (0.50-5.18)	
		BioNet Recombinant Tdap - Pertagen® 0.27 (0.05-1.47)	BioNet Recombinant Tdap - Pertagen® 0.43 (0.14-1.33)	
		BioNet Recombinant Tdap - Boostagen® 0.66 (0.11-3.83)	BioNet Recombinant Tdap - Boostagen® 0.73 (0.23-2.31)	
		BioNet Recombinant Tdap - Adacel® 1.30 (0.23-7.51)	BioNet Recombinant Tdap - Adacel® 1.29 (0.41-4.07)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 3 years after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and 3 years after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Anti-FHA GMCs

Table 36. Comparison of anti-FHA GMCs (IU/mL) between baseline and 3 years after vaccination as assessed by ELISA in all participants by vaccine groups (Per Protocol population)

Vaccine	Baseline	3 years after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	19.45	68.02	3.50	<0.0001 [3]*
(N=59)	(14.24-26.56)	(55.46-83.42)	(2.66-4.59)	
BioNet Recombinant Tdap	24.77	50.67	2.05	<0.0001 [3]*
(N=62)	(18.38-33.38)	(41.11-62.46)	(1.70-2.47)	
Pertagen®	19.62	92.39	4.71	<0.0001 [3]*
(N=58)	(14.52-26.52)	(71.15-119.98)	(3.67-6.04)	
Boostagen®	29.59	99.33	3.36	<0.0001 [3]*
(N=56)	(21.19-41.33)	(79.31-124.39)	(2.45-4.60)	
Adacel®	15.86	52.36	3.30	<0.0001 [3]*
(N=59)	(11.62-21.64)	(42.05-65.20)	(2.59-4.20)	
P-value[b]	0.0522 [2]	<0.0001 [1]*	0.0001 [2]*	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.74 (0.47-1.15)	BioNet Recombinant ap - Pertagen® 0.74 (0.45-1.24)	
		BioNet Recombinant ap - Boostagen® 0.68 (0.44-1.08)	BioNet Recombinant ap - Boostagen® 1.04 (0.62-1.74)	
		BioNet Recombinant ap - Adacel® 1.30 (0.83-2.03)	BioNet Recombinant ap - Adacel® 1.06 (0.64-1.76)	
		BioNet Recombinant Tdap - Pertagen® 0.55 (0.35-0.85)*	BioNet Recombinant Tdap - Pertagen® 0.43 (0.26-0.72)*	
		BioNet Recombinant Tdap - Boostagen® 0.51 (0.33-0.80)*	BioNet Recombinant Tdap - Boostagen® 0.61 (0.37-1.01)	
		BioNet Recombinant Tdap - Adacel® 0.97 (0.62-1.51)	BioNet Recombinant Tdap - Adacel® 0.62 (0.38-1.02)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 3 years after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and 3 years after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 37. Comparison of anti-FHA GMCs (IU/mL) between baseline and 3 years after vaccination as assessed by ELISA in participants aged 65-75 years old by vaccine groups (Per Protocol population)

Vaccine	Baseline	3 years after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
BioNet Recombinant ap	17.76	49.22	2.77	<0.0001 [3]*
(N=11)	(11.50-27.42)	(30.18-80.27)	(1.96-3.91)	
BioNet Recombinant Tdap	15.56	38.47	2.47	<0.0001 [3]*
(N=12)	(7.47-32.41)	(18.98-77.95)	(1.77-3.46)	
Pertagen®	17.61	84.79	4.81	0.0001 [3]*
(N=11)	(7.83-39.58)	(35.40-203.08)	(2.71-8.55)	

Boostagen®	20.23	82.07	4.06	0.0041 [3]*
(N=10)	(9.20-44.51)	(42.60-158.08)	(1.77-9.32)	
Adacel®	15.87	53.36	3.36	0.0006 [3]*
(N=10)	(6.17-40.84)	(23.21-122.71)	(1.96-5.78)	
P-value[b]	0.9836 [2]	0.3466 [1]	0.3955 [1]	
The ratio of GMC or GMFR between Vaccine Group (95% CI)		BioNet Recombinant ap - Pertagen® 0.58 (0.15-2.20) BioNet Recombinant ap - Boostagen® 0.60 (0.15-2.35) BioNet Recombinant ap - Adacel® 0.92 (0.23-3.62) BioNet Recombinant Tdap - Pertagen® 0.45 (0.12-1.68) BioNet Recombinant Tdap - Boostagen® 0.47 (0.12-1.79) BioNet Recombinant Tdap - Adacel® 0.72 (0.19-2.75)	BioNet Recombinant ap - Pertagen® 0.58 (0.21-1.54) BioNet Recombinant ap - Boostagen® 0.68 (0.25-1.88) BioNet Recombinant ap - Adacel® 0.82 (0.30-2.27) BioNet Recombinant Tdap - Pertagen® 0.51 (0.20-1.35) BioNet Recombinant Tdap - Boostagen® 0.61 (0.23-1.64) BioNet Recombinant Tdap - Adacel® 0.74 (0.27-1.98)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 3 years after vaccination to antibody concentration at baseline.
The ratio of GMC or GMFR between vaccine groups (95%CI) based on Bonferroni post-hoc analysis
a : Compared between baseline and 3 years after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Pregnant women

Study TDA207 – A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and the safety of BioNet recombinant pertussis vaccines with different doses of genetically detoxified pertussis toxin (PT_{gen}) when administered to healthy pregnant women

Methods

This phase II, observer-blind, randomized, active-controlled vaccine trial was aimed to evaluate the immunogenicity and the safety of BioNet recombinant pertussis vaccines with different doses of genetically detoxified pertussis toxin (PT_{gen}) when administered to healthy pregnant women.

The study was conducted at 2 sites in Thailand in 240 (40 participants per vaccine group) healthy pregnant women aged 18-40 years of age with uncomplicated singleton pregnancy. Eligible females were randomized equally (in a 1:1:1:1:1:1 ratio) into one of the following vaccine groups (Table 43):

Table 38. Vaccine groups

Vaccine Groups	Total *N*
BioNet recombinant $ap1_{gen}$	40
BioNet recombinant $ap2_{gen}$	40
BioNet recombinant $aP5_{gen}$	40
BioNet recombinant $Tdap2_{gen}$	40
BioNet recombinant $TdaP5_{gen}$	40
Comparator $Tdap_{chem}$	40
Total	**240**

Maternal participants assigned to receive BioNet recombinant $ap1_{gen}$, $ap2_{gen}$ and $aP5_{gen}$ were unblinded at Day 28 after vaccination to receive one dose of a licensed tetanus toxoid (TT) vaccine after a blood sample collection at this visit.

The results until delivery visit (Visit 3) are presented in the submitted clinical study report. In addition, a very brief CSR addendum is available showing high level immunogenicity data for the 2-month blood draw from infants.

- **Study Participants**

A total of 240 healthy pregnant women aged between 18 and 40 years with singleton uncomplicated pregnancy were enrolled into the study after the screening. Only those subjects who fulfilled all of the inclusion criteria and none of the exclusion criteria were randomized.

- **Treatments**

For each of the test products (groups 1-5), a single batch for each investigational vaccine was used for the entire study. Licensed tetanus toxoid, reduced diphtheria toxoid, and acellular pertussis (Tdap) vaccine ($Tdap_{chem}$, Adacel), which was manufactured by Sanofi Pasteur Limited, was used as comparator vaccine for this study. Two batches of $Tdap_{chem}$ vaccine were used for the entire study. All vaccines were presented in vials, each containing one human dose (0.5 mL). The vaccines were administered by intramuscular injection preferably in the non-dominant deltoid.

Table 39. Composition of vaccines administered to the maternal participants

Name of ingredients per 0.5-ml dose	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
	BioNet Recombinant $ap1_{gen}$	**BioNet Recombinant $ap2_{gen}$**	**BioNet Recombinant $Tdap2_{gen}$**	**BioNet Recombinant $aP5_{gen}$**	**BioNet Recombinant $TdaP5_{gen}$**	**$Tdap_{chem}$**
Active ingredients						
Tetanus Toxoid (TT)	-	-	7.5 Lf	-	7.5 Lf	5 Lf
Diphtheria Toxoid (DT)	-	-	2 Lf	-	2 Lf	2 Lf
Pertussis Toxoid (PT)	1 µg	2 µg	2 µg	5 µg	5 µg	2.5 µg
Filamentous hemagglutinin (FHA)	1 µg	5 µg	5 µg	5 µg	5 µg	5 µg
Pertactin (PRN)	-	-	-	-	-	3 µg
Fimbriae type 2/3	-	-	-	-	-	5 µg
Excipients						
Adjuvant	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Hydroxide 0.3 mg/dose as Al^{3+}	Aluminum Phosphate 1.5 mg/dose (0.33 mg/dose as Al)
NaCl mg/dose	4.38	4.38	4.38	4.38	4.38	-
Water for Injection	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL

- **Study assessments**

Blood samples were taken from all pregnant women at baseline (Day 0) before vaccination, at Day 28, and during delivery. To assess maternal antibody transfer, cord blood samples were collected at delivery and if not possible from the neonate within 72 hours after delivery. To evaluate the response to primary infant immunisation, blood samples were collected from infants at 2 and 7 months of age (Visit 4 and Visit 7, respectively). Only a subset of 20 mother-infant pairs (50%) in each vaccine group (total of 120 pairs) have been tested for PT-neutralizing serum antibody by CHO assay.

- **Objectives**

The primary study objective was to assess the immunogenicity of a single dose vaccination of BioNet recombinant pertussis vaccines; $ap1_{gen}$, $ap2_{gen}$, $aP5_{gen}$, $Tdap2_{gen}$, and TdaP5gen relative to $Tdap_{chem}$ based on level of serum anti-PT antibody measured by ELISA at 28 days following immunisation in healthy pregnant women.

Study hypothesis

To compare each BioNet vaccine formulation with $Tdap_{chem}$ vaccine, the following hypothesis will be tested *with* a significant level of 0.05 for the primary objective,

H_0: GMCS = GMCR

H_A: GMCS ≠ GMCR

Where GMC = geometric mean of pertussis toxin (PT)-specific serum antibodies concentration (GMC) measured 28 days following immunisation in maternal participants, R = reference group (Licensed comparator $Tdap_{chem}$ group) and S = study group (one of BioNet's recombinant $ap1_{gen}$, $ap2_{gen}$, $Tdap2_{gen}$, $aP5_{gen}$ and $TdaP5_{gen}$ group).

- **Outcomes/endpoints**

Primary Endpoint

Geometric mean of pertussis toxin (PT)-specific serum antibodies concentration (GMC) measured 28 days following immunisation in maternal participants, as determined by ELISA.

Selected Secondary Endpoints

In maternal participants:

- Geometric mean of anti-PT antibody concentration measured by ELISA at baseline and at the time of delivery
- Geometric mean of anti-FHA antibodies concentration measured by ELISA at baseline, at 28 days after vaccination, and at delivery
- Percentage of maternal participants with a four-fold or higher response in anti-PT and anti FHA antibodies concentrations measured by ELISA at 28 days after vaccination, and at delivery, as compared to baseline
- Geometric mean of PT-neutralizing antibodies titer (GMT) measured at baseline, at 28 days following immunisation, and at delivery, determined by CHO cell assay
- Percentage of the subset of maternal participants with a four-fold or higher increase in PT neutralizing antibodies titer measured at 28 days following immunisation and delivery, as compared to baseline, determined by CHO cells

In infant participants:

- Geometric mean of anti-PT and anti-FHA antibodies concentrations measured by ELISA at the time of birth (cord blood sample or a neonatal blood sample within 72 hours after birth) and at 2 months of age
- GMT of PT-neutralizing antibodies in infant participants measured at the time of birth (cord blood sample or a neonatal blood sample within 72 hours after birth) and at 2 months of age

- **Sample size**

The sample size was calculated based on the primary objective. According to a previous study, the GMC of anti-PT IgG of VacPertagen and Adacel were 562 IU/mL, (95% CI 468-675) and 63 IU/mL, (95%CI 51-78), respectively, in Thai adolescents (Sricharoenchai et al, 2018). Therefore, with alpha level of 0.05 and 80% power, the sample size required for this study is 6 participants per group. Therefore, 40 participants in each group (total of 240 participants) are sufficient to meet the comparative analysis for the primary objective.

- **Randomisation and Blinding (masking)**

Subjects were vaccinated according to the vaccine group assignment from the randomization list. The first randomization list containing participant numbers and masked vaccine group assignments. The second randomization list containing pre-selected 20 mother-infant pairs per vaccine group whose samples were to be tested for PT-neutralizing antibody by CHO cells

The trial has been carried out in an observer-blind manner for the maternal participants from vaccination until Visit 7 (7 months postpartum) for all vaccine groups, except for BioNet recombinant $ap1_{gen}$, $ap2_{gen}$ and $aP5_{gen}$ groups. Those maternal participants assigned to receive BioNet recombinant $ap1_{gen}$, $ap2_{gen}$ and $aP5_{gen}$ were unblinded at Day 28 (Visit 2) to receive one dose of TT vaccine after blood draw at this Visit 2 and were to receive one dose of Td vaccine soon after delivery.

- **Statistical methods**

Analysis sets

Definition of analysis populations for **maternal participants** to be analysed are:

Enrolled Population includes all screened participants who provide informed consent and received a Participant Number, regardless of the participant's randomization and treatment status in the study.

Full Analysis (FA) Population includes the participants in the enrolled population who were randomized, received a study vaccination, and provide evaluable serum sample at least one time point post-vaccination. The analysis based on this population will serve as supportive results for all secondary immunogenicity objectives pertinent to maternal participants. Participants in the FA population will be analysed "as randomised", i.e. according to the vaccine a participant was designated to receive.

The "Per Protocol" (PP) Population includes the participants in the FA population who correctly received study vaccine per randomization with no major protocol deviations that are determined to potentially interfere with the immunogenicity assessment of the study vaccines. This population will serve as the primary analysis population for all immunogenicity objectives associated with maternal participants.

Definition of analysis populations for **infant** participants are:

Full Analysis (FA) Population includes the infants whose mothers are included in the FA population for maternal participants. The analysis based on this population will serve as supportive results for all secondary immunogenicity objectives pertinent to infant participants.

PP Population includes the infants whose mothers are included in the PP population for maternal participants and who have no major protocol deviations that are determined to potentially interfere with the immunogenicity assessment of the study vaccines. This population will serve as the primary analysis population for all immunogenicity objectives associated with infant participants.

Because of the unpredictability of some irregularities, the criteria for exclusion of mothers/infants from the PP population will be determined based on a blind review of the data before the database is locked. The precise reasons for excluding infants from a PP analysis will be documented before unblinding.

Statistical Method for Primary Objective

The primary endpoint was geometric mean of pertussis toxin (PT)-specific serum antibodies concentration (GMC) measured 28 days following immunisation in maternal participants, as determined by ELISA.

PP population served as the primary analysis population for all immunogenicity objectives associated with maternal and infant participants.

To assess the primary objective, geometric mean of PT-specific serum antibody concentration at 28 days following immunisation and its 95% CI in maternal participants were calculated for each vaccine group by exponentiation the corresponding log-transformed mean and its 95% CI limits. The ratio of the GMC in each of BioNet recombinant $ap1_{gen}$, $ap2_{gen}$, $Tdap2_{gen}$, $aP5_{gen}$ and $TdaP5_{gen}$ to that in Tdapchem group and a two-sided 95% CI of the ratio were provided. The log-transformed concentrations were used to construct a two-sided 95% CI for the mean difference between the two vaccine groups using ANOVA (no covariate adjustment was mentioned in protocol or SAP). The mean difference and corresponding 95% CI limits were exponentiated to obtain the GMC ratio and its 95% CI.

Handling of Dropouts and Missing Values

No missing data imputation techniques were planned for the immunogenicity analysis of this study. Missing immunogenicity data were analysed as if they were missing randomly.

Over the whole study period, the number and percentage of participants who withdrew from the study were provided by treatment group. All withdrawn participants post-randomization were further described regarding their time to dropout, as well as their reasons for withdrawal, and the person responsible for the decision. For participants who withdrew from the study, their data collected before withdrawal were analysed under full analysis (FA) and per protocol (PP) population and safety population when applicable.

Planned subgroup analyses

No subgroup analyses for the primary or secondary endpoints were planned.

A subgroup of participants was selected to assess neutralizing anti-PT antibody titer in a subset of 120 maternal participants (20 participants per each vaccine group).

A subgroup evaluation of serum antibody levels measured by ELISA against PT and PT neutralizing serum antibody measured by CHO cell assay at 28 days following immunisation, at the time of delivery and in cord blood in maternal participants immunized during the second trimester versus maternal participants immunized during the third trimester of pregnancy was performed.

Planned Data Analysis of the Study

The comparisons between BioNet vaccine groups and the $Tdap_{chem}$ group, all statistical tests will be two-sided with a significance level of 0.05 unless indicated otherwise. The 95% CI will be provided for estimates, as appropriate.

The analysis of pregnancy and delivery data and the analysis of data between delivery and 7 months of age were planned to be conducted separately:

- The result of the analysis of pregnancy and delivery data was presented in the CSR. Immunogenicity and safety data were reported on group-level only. Individual listings were reported to be generated without information on the participant's study group. Access to participant-level information about study groups was reported to be restricted.
- The results of the analysis of data between delivery and 7 months of age was presented in a separate CSR addendum. Individual data listings with information on the participant's study group were reported to be generated after full unblinding and provided in the addendum.

From footnotes in the SAP table templates: The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis and asterisk (*) will be added, if the GMC or GMFR between vaccine groups is significantly different.

From CSR synopsis tables (*sic*): The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons.

Results

- **Participant flow**

Figure 12. Overall participant disposition of mothers at Day 28 post-vaccination (Per protocol population)

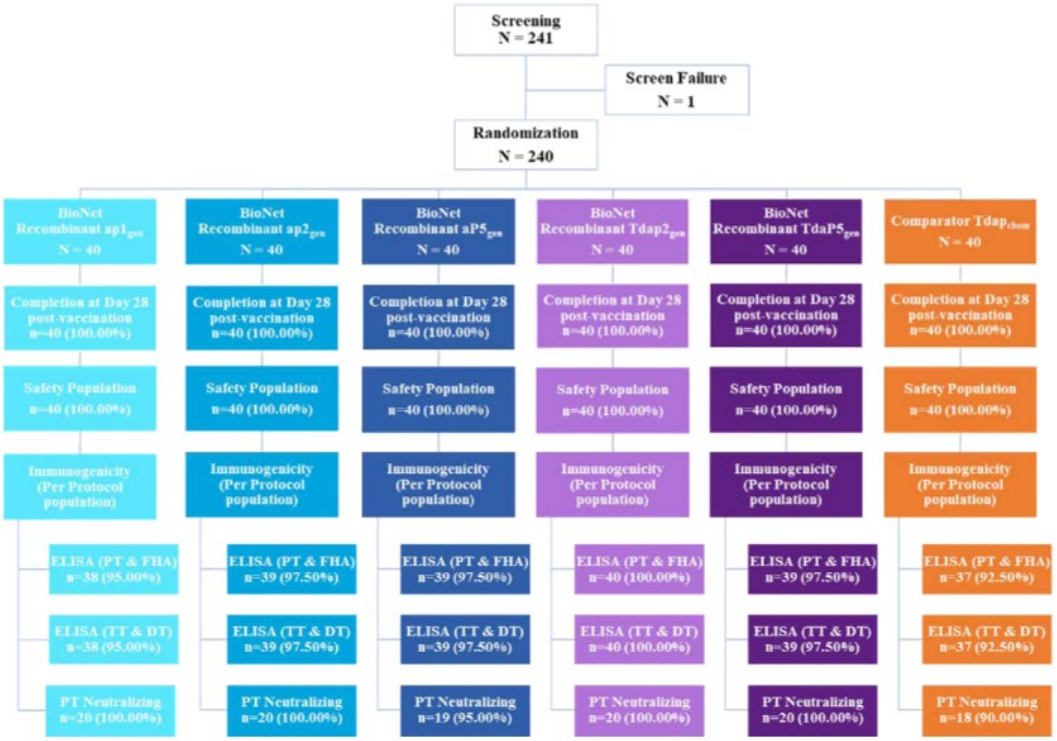
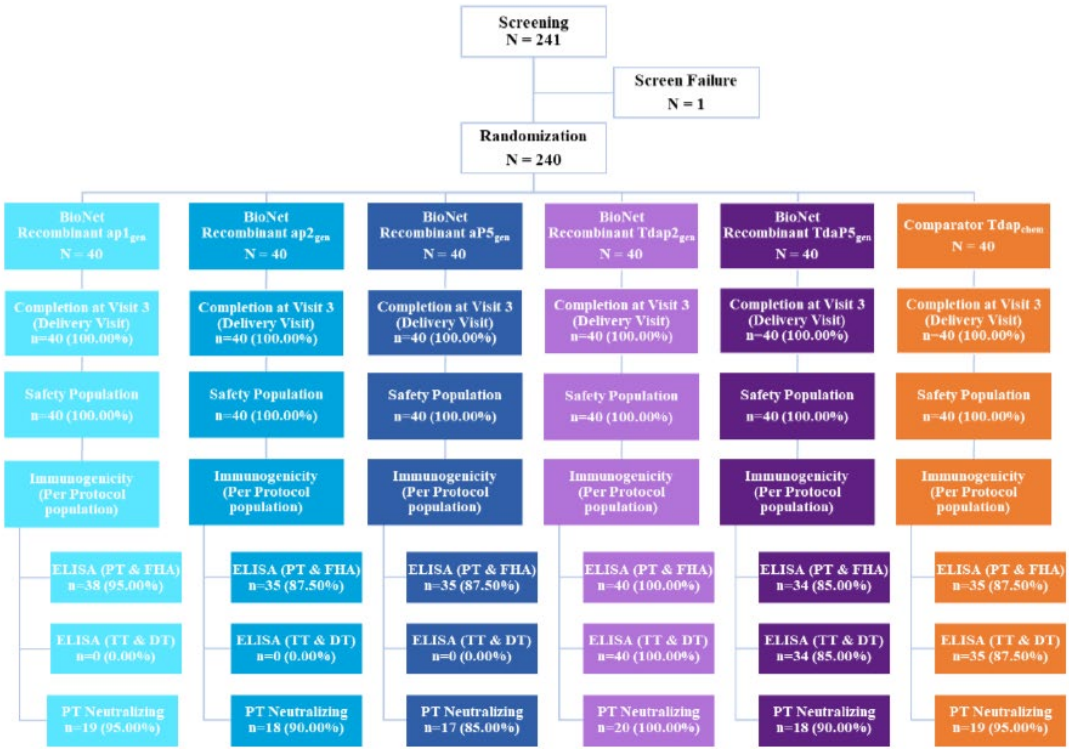


Figure 13. Overall participant disposition of mothers Visit 3 (Delivery visit) (Per protocol population)



- **Recruitment**

Study initiation date: 18 June 2021 (First Subject First Visit, FSFV)

Study completion date: 28 October 2022 (Last Subject Last Day 28 Visit), 02 February 2023 (Mother: Last Subject Last Discharged after Delivery Visit), 01 February 2023 (Infant: Last Subject Last Discharged after Delivery Visit)

- **Conduct of the study**

There was no change in the conduct of the study. All procedures were performed as per the study protocol. No changes had been made to the planned analyses described in the final version of the SAP.

Protocol Deviation

In Maternal Participants: During the conduct of this study from Visit 0 (screening) to Visit 3 (Delivery visit), 64 major and 47 minor protocol deviations were reported. None of the protocol deviations affected participants' safety or led to discontinuation from the study. Most common major protocol deviation was due to no blood/cord blood sample at visit 3 (Delivery) due to the participant delivery at another hospital. Most common minor protocol deviation was due time from blood sample centrifugation to serum storage in deep freezer more than 1 hours at Visit 1 or Visit 2 or Visit 3.

In Infant Participants: For infant participants at Visit 3 (Delivery visit), 9 major and 3 minor protocol deviations were reported. None of the protocol deviations affected participants' safety or led to discontinuation from the study. Most common major protocol deviation was due to infant blood sample not collected within 72 hours after birth. Most common minor protocol deviation was due to delay

(more than 24 hours since site acknowledged the event) in Serious Adverse Event initial report submitted to IRB (Institutional Review Board).

- **Baseline data**

Table 40. Summary of demographics at Visit 0 (screening) of mothers (Full Analysis population)

Participant status	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet recombinant $aP5_{gen}$	BioNet recombinant $Tdap2_{gen}$	BioNet recombinant $TdaP5_{gen}$	Comparator $Tdap_{chem}$	Total	P-value
Demographics at baseline (Screening)								
Age (years)								
-N	40	40	40	40	40	40	240	
-Mean (SD)	29.53 (5.08)	29.38 (4.65)	29.70 (3.59)	30.18 (4.01)	29.40 (5.66)	30.38 (5.95)	29.76 (4.86)	0.9785 [2]
-Median (Q1 - Q3)	30.50 (25.00 - 34.00)	30.00 (25.00 - 32.50)	30.00 (27.50 - 32.00)	29.00 (28.00 - 33.00)	30.00 (24.00 - 34.00)	29.00 (25.00 - 36.50)	30.00 (26.00 - 34.00)	
-Min/Max	20 - 37	22 - 38	23 - 37	22 - 38	19 - 39	21 - 40	19 - 40	
Ethnicity: n (%)								_ [1]
-Asian	40 (100.00)	40 (100.00)	40 (100.00)	40 (100.00)	40 (100.00)	40 (100.00)	240 (100.00)	
-Other	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Height (cm)								
-N	40	40	40	40	40	40	240	
-Mean (SD)	159.5 (5.75)	159.1 (5.58)	158.0 (6.12)	160.9 (4.98)	157.5 (6.39)	159.2 (5.72)	159.0 (5.82)	0.1939 [2]
-Median (Q1 - Q3)	159.0 (155.0 - 165.0)	160.0 (155.5 - 163.0)	159.0 (154.0 - 163.0)	160.5 (158.0 - 165.0)	157.0 (153.0 - 162.0)	158.0 (156.0 - 162.5)	159.0 (155.0 - 163.0)	
-Min/Max	151 - 176	150 - 170	145 - 170	149 - 175	143 - 169	149 - 176	143 - 176	
Weight (kg)								
-N	40	40	40	40	40	40	240	
-Mean (SD)	67.02 (13.95)	66.92 (11.08)	67.69 (14.98)	67.37 (11.34)	62.73 (11.39)	66.77 (13.57)	66.42 (12.78)	0.3530 [2]
-Median (Q1 - Q3)	63.70 (57.25 - 74.50)	66.30 (60.55 - 71.75)	66.10 (56.20 - 71.25)	66.30 (60.00 - 71.95)	59.60 (55.00 - 69.40)	66.85 (55.10 - 77.25)	65.00 (56.95 - 72.50)	
-Min/Max	46 - 105	43.6 - 96	42.9 - 105	48.7 - 96.4	48 - 100	39.8 - 91	39.8 - 105	

Note:
[1] No p-value was computed by SAS
[2] P-value based on Kruskal-Wallis Test

Table 41. Summary of Gestational age at Visit 1: vaccination of mothers

Gestational Age (week)	Visit 1 (Vaccination)						
	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet recombinant $aP5_{gen}$	BioNet recombinant $Tdap2_{gen}$	BioNet recombinant $TdaP5_{gen}$	Comparator $Tdap_{chem}$	P-value
All Participants							
-N	40	40	40	40	40	40	
-Mean(SD)	26.66 (3.68)	26.34 (3.51)	26.21 (3.69)	26.02 (3.49)	25.47 (3.64)	26.26 (3.26)	0.7464 [2]
-Median	25.86	26.43	25.71	25.79	24.64	25.36	
-Min/Max	20.14 - 32.29	20.43 - 32.57	20 - 32.71	20 - 32.71	20.14 - 32.71	21 - 32.14	
2nd Trimester							
-n (%)	21 (52.50)	23 (57.50)	24 (60.00)	23 (57.50)	27 (67.50)	24 (60.00)	0.8490 [1]
3rd Trimester							
-n (%)	19 (47.50)	17 (42.50)	16 (40.00)	17 (42.50)	13 (32.50)	16 (40.00)	

Note:
For this study (TDA207), the gestational age of second trimester of pregnancy is from week 20 to week 26 and gestational age of third trimester of pregnancy is from week 27 up to 33 weeks.
[1] Overall p-value (2-sided) based on Chi-square test
[2] P-value based on Kruskal-Wallis Test

- **Numbers analysed**

The primary analysis was not performed by "intention to treat" but instead in a Per Protocol Population. However, analyses were also provided for the Full Analysis Population "as randomized".

Only one participant failed screening, who met exclusion criterion no. 11 (planning to participate in another clinical trial during the study period). All 240 enrolled participants received a vaccine and completed the study at Day 28 post-vaccination

The number of participants in each group included in the primary and each key secondary analysis is shown in Figure 12 (D28 post-vaccination) and Figure 13 (delivery).

At D28, 8 maternal participants were excluded from ELISA immunogenicity analysis for PP population (1 in the $aP5_{gen}$=VacPertagen and 3 in the $Tdap_{chem}$=Adacel groups) and 3 were excluded from PT neutralization immunogenicity analysis for PP population only. The most common reason for exclusion was caused by lack of an available blood sample due to preterm delivery visit prior to the scheduled Visit 2 at Day 28 in three cases. Two participants were excluded because they received other vaccines (1x COVID-19 vaccine, 1x Influenza vaccine) between study entry and Day 28.

At delivery, 23 maternal participants were excluded from ELISA immunogenicity analysis for PP population (5 in both the VacPertagen and Adacel groups) and 9 were excluded from PT neutralization immunogenicity analysis for PP population. According to Appendix 16.2.3, the most common reason for exclusion of maternal participants from ELISA immunogenicity analysis was caused by no available blood sample at visit 3 (Delivery) due to delivery at another hospital in 11 cases.

The dossier also includes similar tables for infants (as shown above for maternal participants) with numbers of participants for statistical analysis for the delivery visit. These data are not shown in this report, but the most common reason for exclusion was obviously again no available blood sample at visit 3 (Delivery) due to delivery at another hospital.

The participant flow and the numbers analysed have been presented in sufficient detail for Visit 2 (D28 after vaccination) and Visit 3 (delivery). Detailed data from later immunogenicity visits are currently missing. No relevant imbalances between the treatment groups are noted with respect to exclusions from the immunogenicity sets.

- **Outcomes and estimation**

GMCs of anti-PT and anti-FHA antibodies at baseline, 28 days post-vaccination and at delivery

At 28 days after vaccination, GMCs for **anti-PT IgG** antibodies were higher for VacPertagen (=$aP5_{gen}$, 153.98 [95% CI: 107.51 – 220.55]) compared to Adacel (=$Tdap_{chem}$,) 29.53 IU/mL [95% CI: 20.20-43.16]).

At 28 days after vaccination, GMCs for **anti-FHA IgG** antibodies were higher for VacPertagen (214.51 [95% CI: 165.46 – 278.10]) compared to Adacel (83.48 IU/mL [95% CI: 55.60-125.35]).

Table 42. Summary of anti-PT and anti FHA GMCs (IU/ml) as assessed by ELISA at baseline and 28 days after vaccination of mothers by vaccine groups

Vaccine	PT				FHA			
	Baseline	Day 28 after vaccination	GMFR (Day 28 /Baseline)	P-value[a]	Baseline	Day 28 after vaccination	GMFR (Day 28 /Baseline)	P-value[a]
	GMC (95% CI)	GMC (95% CI)	(95% CI)		GMC (95% CI)	GMC (95% CI)	(95% CI)	
BioNet recombinant ap1$_{gen}$ (N=38)	4.98 (3.16-7.87)	73.28 (54.99-97.64)	14.70 (9.80-22.07)	<0.0001 [3]*	12.73 (8.38-19.36)	108.08 (90.38-129.25)	8.49 (5.86-12.29)	<0.0001 [3]*
BioNet recombinant ap2$_{gen}$ (N=39)	4.98 (3.27-7.61)	102.25 (74.10-141.10)	20.51 (13.64-30.85)	<0.0001 [3]*	13.09 (8.85-19.36)	210.83 (160.05-277.72)	16.11 (11.16-23.26)	<0.0001 [3]*
BioNet recombinant aP5$_{gen}$ (N=39)	4.71 (3.17-7.00)	153.98 (107.51-220.55)	32.71 (21.94-48.75)	<0.0001 [3]*	16.05 (10.59-24.33)	214.51 (165.46-278.10)	13.37 (8.89-20.10)	<0.0001 [3]*
BioNet recombinant Tdap2$_{gen}$ (N=40)	3.97 (2.78-5.65)	56.84 (45.33-71.28)	14.33 (10.27-20.00)	<0.0001 [3]*	11.53 (7.57-17.57)	150.30 (120.87-186.90)	13.03 (8.75-19.41)	<0.0001 [3]*
BioNet recombinant TdaP5$_{gen}$ (N=39)	4.80 (3.09-7.45)	137.98 (102.21-186.28)	28.76 (20.26-40.83)	<0.0001 [3]*	11.09 (7.23-17.00)	154.70 (118.99-201.13)	13.95 (9.55-20.39)	<0.0001 [3]*
Tdap$_{chem}$ (N=37)	5.68 (3.62-8.92)	29.53 (20.20-43.16)	5.20 (3.77-7.16)	<0.0001 [3]*	12.35 (7.97-19.14)	83.48 (55.60-125.35)	6.76 (4.58-9.97)	<0.0001 [3]*
P-value[b]	0.9651 [1]	<0.0001 [1]*	<0.0001 [2]*		0.8559 [1]	<0.0001 [1]*	0.0112 [2]*	
Ratio between each of BioNet recombinant vaccines and Tdap$_{chem}$ (95% CI)		ap1$_{gen}$ - Tdap$_{chem}$ 2.48 (1.39-4.43)*	ap1$_{gen}$ - Tdap$_{chem}$ 2.83 (1.43-5.61)*			ap1$_{gen}$ - Tdap$_{chem}$ 1.29 (0.78-2.14)	ap1$_{gen}$ - Tdap$_{chem}$ 1.26 (0.62-2.56)	
		ap2$_{gen}$ - Tdap$_{chem}$ 3.46 (1.95-6.16)*	ap2$_{gen}$ - Tdap$_{chem}$ 3.95 (2.00-7.79)*			ap2$_{gen}$ - Tdap$_{chem}$ 2.53 (1.53-4.17)*	ap2$_{gen}$ - Tdap$_{chem}$ 2.38 (1.18-4.83)*	
		aP5$_{gen}$ - Tdap$_{chem}$ 5.22 (2.93-9.28)*	aP5$_{gen}$ - Tdap$_{chem}$ 6.29 (3.19-12.43)*			aP5$_{gen}$ - Tdap$_{chem}$ 2.57 (1.56-4.24)*	aP5$_{gen}$ - Tdap$_{chem}$ 1.98 (0.98-4.01)	
		Tdap2$_{gen}$ - Tdap$_{chem}$ 1.93 (1.09-3.41)*	Tdap2$_{gen}$ - Tdap$_{chem}$ 2.76 (1.40-5.42)*			Tdap2$_{gen}$ - Tdap$_{chem}$ 1.80 (1.09-2.96)*	Tdap2$_{gen}$ - Tdap$_{chem}$ 1.93 (0.96-3.89)	
		TdaP5$_{gen}$ - Tdap$_{chem}$ 4.67 (2.63-8.32)*	TdaP5$_{gen}$ - Tdap$_{chem}$ 5.53 (2.80-10.93)*			TdaP5$_{gen}$ - Tdap$_{chem}$ 1.85 (1.12-3.06)*	TdaP5$_{gen}$ - Tdap$_{chem}$ 2.06 (1.02-4.19)*	

Geometric mean fold rise (GMFR) geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5pairwise comparisons
a : Compared between baseline and Day 28 after vaccination, b : Compared between vaccine groups
Note: [1] P-value based on Kruskal-Wallis Test, [2] P-value based on One-way ANOVA, [3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

At delivery, the GMCs for **anti-PT IgG** antibodies reduced across all the vaccine groups compared to 28 days after vaccination levels but were higher compared to baseline levels. The **geometric mean fold rise (GMFR)** of **anti-PT IgG** antibodies from baseline were higher in participants from each of BioNet recombinant vaccine groups compared to Tdap$_{chem}$ group.

At delivery, the **GMFR** of **anti-FHA IgG** antibodies from baseline was similar in participants from each of BioNet recombinant vaccine groups compared to Tdap$_{chem}$ group.

Table 43. Summary of anti-PT and anti-FHA GMCs (IU/ml) as assessed by ELISA at baseline and delivery after vaccination of mothers by vaccine

Vaccine	PT				FHA			
	Baseline	Delivery	GMFR (Delivery/Baseline)	P-value[a]	Baseline	Delivery	GMFR (Delivery/Baseline)	P-value[a]
	GMC (95% CI)	GMC (95% CI)	(95% CI)		GMC (95% CI)	GMC (95% CI)	(95% CI)	
BioNet recombinant ap1$_{gen}$ (N=38)	4.99 (3.16-7.88)	54.00 (39.82-73.24)	10.82 (7.27-16.10)	<0.0001 [3]*	11.95 (7.96-17.94)	79.73 (67.54-94.13)	6.67 (4.81-9.24)	<0.0001 [3]*
BioNet recombinant ap2$_{gen}$ (N=35)	4.92 (3.19-7.58)	59.87 (38.42-93.28)	12.18 (7.78-19.05)	<0.0001 [3]*	13.41 (8.75-20.57)	134.10 (99.89-180.04)	10.00 (6.72-14.87)	<0.0001 [3]*
BioNet recombinant aP5$_{gen}$ (N=35)	4.65 (3.10-6.97)	119.75 (81.59-175.74)	25.77 (17.09-38.86)	<0.0001 [3]*	17.07 (11.14-26.18)	155.53 (111.32-217.30)	9.11 (6.02-13.79)	<0.0001 [3]*
BioNet recombinant Tdap2$_{gen}$ (N=40)	3.97 (2.78-5.65)	36.85 (28.93-46.92)	9.29 (6.68-12.92)	<0.0001 [3]*	11.53 (7.57-17.57)	107.10 (88.76-129.24)	9.29 (6.56-13.14)	<0.0001 [3]*
BioNet recombinant TdaP5$_{gen}$ (N=34)	5.49 (3.38-8.90)	102.40 (70.32-149.13)	18.66 (12.44-27.99)	<0.0001 [3]*	11.42 (7.18-18.16)	108.73 (76.78-153.97)	9.52 (6.24-14.52)	<0.0001 [3]*
Tdap$_{chem}$ (N=34)	6.04 (3.73-9.79)	22.23 (15.23-32.45)	3.68 (2.67-5.07)	<0.0001 [3]*	12.31 (7.82-19.38)	65.93 (43.83-99.18)	5.35 (3.64-7.87)	<0.0001 [3]*
P-value[b]	0.9314 [1]	<0.0001 [1]*	<0.0001 [2]*		0.7611 [1]	0.0001 [1]*	0.1300 [2]	
Ratio between each of BioNet recombinant vaccines and Tdap$_{chem}$ (95% CI)		ap1$_{gen}$ - Tdap$_{chem}$ 2.43 (1.28-4.62)*	ap1$_{gen}$ - Tdap$_{chem}$ 2.94 (1.46-5.93)*			ap1$_{gen}$ - Tdap$_{chem}$ 1.21 (0.71-2.06)	ap1$_{gen}$ - Tdap$_{chem}$ 1.25 (0.62-2.48)	
		ap2$_{gen}$ - Tdap$_{chem}$ 2.69 (1.40-5.19)*	ap2$_{gen}$ - Tdap$_{chem}$ 3.31 (1.62-6.77)*			ap2$_{gen}$ - Tdap$_{chem}$ 2.03 (1.18-3.50)*	ap2$_{gen}$ - Tdap$_{chem}$ 1.87 (0.92-3.78)	
		aP5$_{gen}$ - Tdap$_{chem}$ 5.39 (2.80-10.38)*	aP5$_{gen}$ - Tdap$_{chem}$ 7.00 (3.42-14.32)*			aP5$_{gen}$ - Tdap$_{chem}$ 2.36 (1.37-4.06)*	aP5$_{gen}$ - Tdap$_{chem}$ 1.70 (0.84-3.44)	
		Tdap2$_{gen}$ - Tdap$_{chem}$ 1.66 (0.88-3.13)	Tdap2$_{gen}$ - Tdap$_{chem}$ 2.52 (1.26-5.05)*			Tdap2$_{gen}$ - Tdap$_{chem}$ 1.62 (0.96-2.75)	Tdap2$_{gen}$ - Tdap$_{chem}$ 1.73 (0.88-3.43)	
		TdaP5$_{gen}$ - Tdap$_{chem}$ 4.61 (2.38-8.92)*	TdaP5$_{gen}$ - Tdap$_{chem}$ 5.07 (2.47-10.43)*			TdaP5$_{gen}$ - Tdap$_{chem}$ 1.65 (0.95-2.85)	TdaP5$_{gen}$ - Tdap$_{chem}$ 1.78 (0.87-3.61)	

Geometric mean fold rise (GMFR) geometric mean of the ratios of antibody concentration at delivery to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5pairwise comparisons
a : Compared between baseline and Day 28 after vaccination, b : Compared between vaccine groups
Note: [1] P-value based on Kruskal-Wallis Test, [2] P-value based on One-way ANOVA, [3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Maternal participants blood samples at delivery and cord blood or neonatal blood samples

At time of birth (cord blood or neonatal blood within 72 hours after birth), the geometric mean concentration ratio (GMCR) of **anti-PT IgG** antibodies between time of birth (cord blood or neonatal blood) and delivery (maternal blood sample) were similar across all the vaccine groups (Table 44). The same was noted for anti-FHA IgG antibodies.

*Table 44. Summary of anti-PT and anti-FHA GMCs (IU/ml) as assessed by ELISA between delivery (mother) and time of birth (cord blood or neonatal blood within 72 hours after birth) by vaccine groups**

Vaccine	PT				FHA			
	Delivery (mother)	Time of Birth (cord blood or neonatal blood)	GMCR (Delivery (mother)/ Time of Birth (cord blood or neonatal blood))	P-value[a]	Delivery (mother)	Time of Birth (cord blood or neonatal blood)	GMCR (Delivery (mother)/ Time of Birth (cord blood or neonatal blood))	P-value[a]
	GMC (95% CI)	GMC (95% CI)	(95% CI)		GMC (95% CI)	GMC (95% CI)	(95% CI)	
BioNet recombinant $ap1_{gen}$ (N=38)	54.00 (39.82-73.24)	64.80 (45.17-92.97)	1.20 (1.03-1.40)	0.0207 [2]*	79.73 (67.54-94.13)	101.18 (82.14-124.63)	1.27 (1.12-1.44)	0.0005 [2]*
BioNet recombinant $ap2_{gen}$ (N=35)	59.87 (38.42-93.28)	78.90 (52.97-117.51)	1.32 (1.12-1.55)	0.0016 [2]*	134.10 (99.89-180.04)	179.32 (130.44-246.51)	1.34 (1.14-1.57)	0.0008 [2]*
BioNet recombinant $aP5_{gen}$ (N=35)	119.75 (81.59-175.74)	141.40 (94.70-211.12)	1.18 (0.98-1.43)	0.0815 [2]	155.53 (111.32-217.30)	215.54 (157.10-295.74)	1.39 (1.19-1.61)	0.0001 [2]*
BioNet recombinant $Tdap2_{gen}$ (N=40)	36.85 (28.93-46.92)	49.14 (39.04-61.84)	1.33 (1.18-1.50)	<0.0001 [2]*	107.10 (88.76-129.24)	147.49 (117.65-184.90)	1.38 (1.20-1.58)	<0.0001 [2]*
BioNet recombinant $TdaP5_{gen}$ (N=34)	102.40 (70.32-149.13)	122.61 (84.68-177.52)	1.20 (1.05-1.37)	0.0097 [2]*	108.73 (76.78-153.97)	144.61 (104.23-200.62)	1.33 (1.19-1.48)	<0.0001 [2]*
$Tdap_{chem}$ (N=35)	22.09 (15.30-31.89)	27.09 (18.21-40.31)	1.23 (1.11-1.36)	0.0003 [2]*	66.51 (44.74-98.87)	83.85 (56.89-123.60)	1.26 (1.14-1.39)	<0.0001 [2]*
P-value[b]	<0.0001 [1]*	<0.0001 [1]*	0.2664 [1]		0.0001 [1]*	<0.0001 [1]*	0.9141 [1]	
Ratio between each of BioNet recombinant vaccines and $Tdap_{chem}$ (95% CI)	$ap1_{gen}$ - $Tdap_{chem}$ 2.44 (1.29-4.62)*	$ap1_{gen}$ - $Tdap_{chem}$ 2.39 (1.25-4.58)*	$ap1_{gen}$ - $Tdap_{chem}$ 0.98 (0.75-1.27)		$ap1_{gen}$ - $Tdap_{chem}$ 1.20 (0.71-2.03)	$ap1_{gen}$ - $Tdap_{chem}$ 1.21 (0.70-2.07)	$ap1_{gen}$ - $Tdap_{chem}$ 1.01 (0.79-1.28)	
	$ap2_{gen}$ - $Tdap_{chem}$ 2.71 (1.42-5.19)*	$ap2_{gen}$ - $Tdap_{chem}$ 2.91 (1.50-5.64)*	$ap2_{gen}$ - $Tdap_{chem}$ 1.07 (0.82-1.41)		$ap2_{gen}$ - $Tdap_{chem}$ 2.02 (1.18-3.45)*	$ap2_{gen}$ - $Tdap_{chem}$ 2.14 (1.23-3.70) *	$ap2_{gen}$ - $Tdap_{chem}$ 1.06 (0.83-1.35)	
	$aP5_{gen}$ - $Tdap_{chem}$ 5.42 (2.83-10.38)*	$aP5_{gen}$ - $Tdap_{chem}$ 5.22 (2.69-10.12)*	$aP5_{gen}$ - $Tdap_{chem}$ 0.96 (0.74-1.26)		$aP5_{gen}$ - $Tdap_{chem}$ 2.34 (1.37-4.01)*	$aP5_{gen}$ - $Tdap_{chem}$ 2.57 (1.48-4.45)*	$aP5_{gen}$ - $Tdap_{chem}$ 1.10 (0.86-1.40)	
	$Tdap2_{gen}$ - $Tdap_{chem}$ 1.67 (0.89-3.13)	$Tdap2_{gen}$ - $Tdap_{chem}$ 1.81 (0.96-3.44)	$Tdap2_{gen}$ - $Tdap_{chem}$ 1.09 (0.84-1.41)		$Tdap2_{gen}$ - $Tdap_{chem}$ 1.61 (0.96-2.71)	$Tdap2_{gen}$ - $Tdap_{chem}$ 1.76 (1.03-2.99)*	$Tdap2_{gen}$ - $Tdap_{chem}$ 1.09 (0.86-1.38)	
	$TdaP5_{gen}$ - $Tdap_{chem}$ 4.64 (2.41-8.92)*	$TdaP5_{gen}$ - $Tdap_{chem}$ 4.53 (2.32-8.81)*	$TdaP5_{gen}$ - $Tdap_{chem}$ 0.98 (0.74-1.28)		$TdaP5_{gen}$ - $Tdap_{chem}$ 1.63 (0.95-2.81)	$TdaP5_{gen}$ - $Tdap_{chem}$ 1.72 (0.99-3.00)	$TdaP5_{gen}$ - $Tdap_{chem}$ 1.05 (0.82-1.35)	

*Data include only participants with available blood sample at delivery (mother) and birth (cord blood or neonatal blood)
GMC ratio (GMCR) is geometric mean of the ratios of antibody concentration at time of birth (cord blood or neonatal blood) to antibody concentration at delivery
The ratio of GMC or GMCR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between delivery and time of birth (cord blood or neonatal blood)
b : Compared between vaccine groups

Maternal participants vaccinated during the second or the third trimester

Table 50 presents anti-PT antibody GMCs in each subgroup (subgroup 1, vaccine administration during the 2nd trimester of pregnancy; subgroup 2, vaccine administration during the 3rd trimester of pregnancy) for maternal blood samples collected at baseline, on day 28, and at delivery, and in cord blood or neonatal blood samples collected at birth. Regardless of the vaccine and sample used, no marked difference in anti-PT antibody GMCs was found between the 2 subgroups.

Table 45. Summary of anti-PT GMCs (IU/ml) as assessed by ELISA at baseline, Day 28 (Visit 2), Delivery (Visit 3) and time of birth (cord blood or neonatal blood) for mothers vaccinated during the second and third trimester by vaccine groups

Vaccine	GMC (IU/mL) (95% CI)							
	Baseline (maternal blood)		Day 28 (maternal blood)		Delivery (maternal blood)		Birth (cord or neonatal blood)	
	2nd trimester	3rd trimester	2nd trimester	3rd trimester	2nd trimester	3rd trimester	2nd trimester	3rd trimester
BioNet recombinant $ap1_{gen}$	N = 21 5.16 (2.72-9.79)	N = 19 4.65 (2.45-8.83)	N = 20 81.71 (58.68-113.78)	N = 18 64.92 (38.81-108.62)	N = 20 55.99 (34.51-90.85)	N = 18 51.88 (34.50-77.99)	N = 20 66.18 (38.76-112.98)	N = 18 63.31 (37.03-108.24)
BioNet recombinant $ap2_{gen}$	N = 23 4.30 (2.45-7.56)	N = 17 5.61 (2.82-11.13)	N = 23 86.84 (57.35-131.51)	N = 16 129.31 (74.90-223.25)	N = 21 44.77 (25.44-78.78)	N = 14 92.58 (44.13-194.24)	N = 21 59.26 (37.42-93.85)	N = 14 121.21 (58.18-252.56)
BioNet recombinant $aP5_{gen}$	N = 24 3.59 (2.29-5.63)	N = 16 6.50 (3.10-13.63)	N = 24 141.35 (82.57-241.99)	N = 15 176.58 (113.46-274.83)	N = 21 105.91 (62.62-179.13)	N = 14 143.96 (77.48-267.47)	N = 21 132.71 (73.88-238.39)	N = 14 155.51 (87.36-276.84)
BioNet recombinant $Tdap2_{gen}$	N = 23 4.55 (2.89-7.17)	N = 17 3.29 (1.79-6.07)	N = 23 50.81 (36.25-71.21)	N = 17 66.16 (49.01-89.31)	N = 23 31.77 (22.80-44.26)	N = 17 45.04 (31.20-65.01)	N = 23 42.47 (30.77-58.62)	N = 17 59.85 (42.81-83.67)
BioNet recombinant $TdaP5_{gen}$	N = 27 4.07 (2.45-6.74)	N = 13 6.82 (2.83-16.41)	N = 27 121.46 (82.82-178.13)	N = 12 183.85 (111.62-302.82)	N = 22 94.10 (57.10-155.10)	N = 12 119.57 (63.01-226.93)	N = 22 120.82 (74.90-194.89)	N = 12 125.95 (63.65-249.24)
$Tdap_{chem}$	N = 24 3.77 (2.29-6.21)	N = 14 10.29 (4.46-23.72)	N = 23 23.24 (15.20-35.53)	N = 14 43.75 (20.63-92.76)	N = 21 16.86 (11.22-25.34)	N = 14 33.12 (16.49-66.54)	N = 21 21.92 (14.36-33.45)	N = 14 37.24 (16.49-84.11)

Seroconversion rates in anti-PT and anti-FHA antibodies at 28 days post-vaccination and delivery in mother

On Day 28, the percentage of maternal participants with a ≥ 4-fold increase in **anti-PT** antibodies concentrations measured by ELISA at 28 days after vaccination as compared to baseline were higher in each of BioNet recombinant vaccine groups compared to Comparator $Tdap_{chem}$ group.

The percentage of maternal participants with ≥ 4-fold increase in **anti-FHA** antibodies concentrations measured by ELISA at 28 days after vaccination as compared to baseline was similar in each of BioNet recombinant vaccine groups and in Comparator $Tdap_{chem}$ group (Table 46).

Table 46. Summary of difference in percentage of participants with a 4-fold or higher response in anti-PT and anti-FHA antibody concentrations at 28 days after vaccination compared to baseline between each BioNet's vaccine groups and $Tdap_{chem}$ group

of mothers

Comparison groups	PT				FHA			
	BioNet Vaccine	$Tdap_{chem}$	Difference in percentage	P-value	BioNet Vaccine	$Tdap_{chem}$	Difference in percentage	P-value
	n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)		n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)	
BioNet recombinant $ap1_{gen}$ and $Tdap_{chem}$ (N=38 and N=37)	33 (86.84) (71.91-95.59)	23 (62.16) (44.76-77.54)	24.68 (5.05-43.24)	0.0140 [1]*	30 (78.95) (62.68-90.45)	25 (67.57) (50.21-81.99)	11.38 (-8.88-31.10)	0.2651 [1]
BioNet recombinant $ap2_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	35 (89.74) (75.78-97.13)	23 (62.16) (44.76-77.54)	27.58 (8.75-45.62)	0.0047 [1]*	33 (84.62) (69.47-94.14)	25 (67.57) (50.21-81.99)	17.05 (-2.27-35.89)	0.0805 [1]
BioNet recombinant $aP5_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	37 (94.87) (82.68-99.37)	23 (62.16) (44.76-77.54)	32.71 (15.46-49.85)	0.0004 [1]*	32 (82.05) (66.47-92.46)	25 (67.57) (50.21-81.99)	14.48 (-5.22-33.70)	0.1449 [1]
BioNet recombinant $Tdap2_{gen}$ and $Tdap_{chem}$ (N=40 and N=37)	34 (85.00) (70.16-94.29)	23 (62.16) (44.76-77.54)	22.84 (3.19-41.60)	0.0224 [1]*	34 (85.00) (70.16-94.29)	25 (67.57) (50.21-81.99)	17.43 (-1.66-36.17)	0.0709 [1]
BioNet recombinant $TdaP5_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	38 (97.44) (86.52-99.94)	23 (62.16) (44.76-77.54)	35.27 (19.24-51.93)	0.0001 [1]*	32 (82.05) (66.47-92.46)	25 (67.57) (50.21-81.99)	14.48 (-5.22-33.70)	0.1449 [1]

95% CI based on Clopper-Pearson method.
The two-sided 95% CI of the difference in the proportion was obtained based on Miettinen and Nurminen method

Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

At Delivery, the percentage of maternal participants with a ≥ 4-fold increase in **anti-PT** antibodies concentrations measured by ELISA at delivery as compared to baseline were higher in each of BioNet recombinant vaccine groups compared to Comparator $Tdap_{chem}$ group.

The percentage of maternal participants with ≥ 4-fold increase in **anti-FHA** antibodies concentrations measured by ELISA at delivery as compared to baseline was similar in each of BioNet recombinant vaccine groups and in Comparator $Tdap_{chem}$ group (47).

Table 47. Summary of difference in percentage of participants with a 4-fold or higher response in anti-PT and anti-FHA antibody concentrations at delivery compared to baseline between each BioNet's vaccine groups and $Tdap_{chem}$ group of mothers

Comparison groups	PT				FHA			
	BioNet Vaccine	$Tdap_{chem}$	Difference in percentage	P-value	BioNet Vaccine	$Tdap_{chem}$	Difference in percentage	P-value
	n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)		n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)	
BioNet recombinant $ap1_{gen}$ and $Tdap_{chem}$ (N=38 and N=34)	30 (78.95) (62.68-90.45)	15 (44.12) (27.19-62.11)	34.83 (12.47-54.16)	0.0023 [1]*	28 (73.68) (56.90-86.60)	21 (61.76) (43.56-77.83)	11.92 (-9.74-32.97)	0.2788 [1]
BioNet recombinant $ap2_{gen}$ and $Tdap_{chem}$ (N=35 and N=34)	27 (77.14) (59.86-89.58)	15 (44.12) (27.19-62.11)	33.03 (9.99-52.87)	0.0049 [1]*	25 (71.43) (53.70-85.36)	21 (61.76) (43.56-77.83)	9.66 (-12.68-31.25)	0.3945 [1]
BioNet recombinant $aP5_{gen}$ and $Tdap_{chem}$ (N=35 and N=34)	33 (94.29) (80.84-99.30)	15 (44.12) (27.19-62.11)	50.17 (30.38-66.81)	<0.0001 [1]*	26 (74.29) (56.74-87.51)	21 (61.76) (43.56-77.83)	12.52 (-9.64-33.75)	0.2645 [1]
BioNet recombinant $Tdap2_{gen}$ and $Tdap_{chem}$ (N=40 and N=34)	32 (80.00) (64.35-90.95)	15 (44.12) (27.19-62.11)	35.88 (13.93-54.92)	0.0013 [1]*	31 (77.50) (61.55-89.16)	21 (61.76) (43.56-77.83)	15.74 (-5.29-36.16)	0.1399 [1]
BioNet recombinant $TdaP5_{gen}$ and $Tdap_{chem}$ (N=34 and N=34)	32 (94.12) (80.32-99.28)	15 (44.12) (27.19-62.11)	50.00 (30.02-66.70)	<0.0001 [1]*	27 (79.41) (62.10-91.30)	21 (61.76) (43.56-77.83)	17.65 (-4.25-38.23)	0.1102 [1]

95% CI based on Clopper-Pearson method.
The two-sided 95% CI of the difference in the proportion was obtained based on Miettinen and Nurminen method

Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

GMTs of PT-neutralizing antibody titers at baseline,28 days post-vaccination and at delivery

At 28 days after vaccination, PT neutralizing antibody GMTs were higher in BioNet recombinant $ap2_{gen}$, BioNet recombinant $aP5_{gen}$ and BioNet recombinant $TdaP5_{gen}$ compared to $Tdap_{chem}$ group.

Table 48. Summary of PT neutralizing GMT (IU/ml) between baseline and Day 28 after vaccination as assessed by PT neutralization assay in CHO cells in a subset of 120 participants (20 participants per vaccine group) of mothers

Vaccine	Baseline	Day 28 after vaccination	GMFR (Day 28 /Baseline)	P-value[a]
	GMT (95% CI)	GMT (95% CI)	(95% CI)	
BioNet recombinant $ap1_{gen}$ (N=20)	5.37 (3.63-7.94)	78.55 (50.12-123.09)	14.63 (9.22-23.22)	<0.0001 [3]*
BioNet recombinant $ap2_{gen}$ (N=20)	8.31 (4.81-14.38)	174.32 (101.79-298.53)	20.97 (11.15-39.44)	<0.0001 [3]*
BioNet recombinant $aP5_{gen}$ (N=19)	6.63 (4.15-10.59)	206.95 (91.38-468.70)	31.23 (13.91-70.15)	<0.0001 [3]*
BioNet recombinant $Tdap2_{gen}$ (N=20)	5.73 (3.95-8.30)	57.22 (36.75-89.09)	9.99 (6.15-16.23)	<0.0001 [3]*
BioNet recombinant $TdaP5_{gen}$ (N=20)	8.51 (4.50-16.12)	177.19 (119.72-262.24)	20.82 (12.42-34.90)	<0.0001 [3]*
$Tdap_{chem}$ (N=18)	7.67 (4.57-12.87)	32.32 (19.57-53.39)	4.22 (2.88-6.18)	<0.0001 [3]*
P-value[b]	0.6006 [1]	<0.0001 [1]*	<0.0001 [2]*	
Ratio between each of BioNet recombinant vaccines and $Tdap_{chem}$ (95% CI)		$ap1_{gen}$ - $Tdap_{chem}$ 2.43 (0.93-6.37)	$ap1_{gen}$ - $Tdap_{chem}$ 3.47 (1.26-9.57)*	
		$ap2_{gen}$ - $Tdap_{chem}$ 5.39 (2.06-14.13)*	$ap2_{gen}$ - $Tdap_{chem}$ 4.97 (1.80-13.71)*	
		$aP5_{gen}$ - $Tdap_{chem}$ 6.40 (2.41-16.98)*	$aP5_{gen}$ - $Tdap_{chem}$ 7.41 (2.65-20.68)*	
		$Tdap2_{gen}$ - $Tdap_{chem}$ 1.77 (0.68-4.64)	$Tdap2_{gen}$ - $Tdap_{chem}$ 2.37 (0.86-6.53)	
		$TdaP5_{gen}$ - $Tdap_{chem}$ 5.48 (2.09-14.36)*	$TdaP5_{gen}$ - $Tdap_{chem}$ 4.94 (1.79-13.61)*	

Geometric mean fold rise (GMFR) geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline
The ratio of GMT or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups
Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA, [3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

At delivery, PT neutralizing antibody GMTs were higher in BioNet recombinant ap2$_{gen}$, BioNet recombinant aP5$_{gen}$, BioNet recombinant TdaP5$_{gen}$ groups compared to Tdap$_{chem}$ group.

Table 49. Summary of PT neutralizing GMTs (IU/ml) as assessed by PT neutralizing assays in CHO cells delivery (mother) and time of birth (cord blood or neonatal blood within 72 hours after birth) in a subset of 20 mother-infant pairs per vaccine group (Per-protocol population)

Vaccine	Delivery (mother)	Time of birth (cord blood or neonatal blood)
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)
BioNet recombinant ap1$_{gen}$	N = 19	N = 19
	38.74	33.54
	(23.75-63.18)	(18.89-59.54)
BioNet recombinant ap2$_{gen}$	N = 18	N = 18
	92.47	83.72
	(49.57-172.53)	(46.77-149.85)
BioNet recombinant aP5$_{gen}$	N = 17	N = 17
	124.26	137.09
	(49.72-310.51)	(51.57-364.40)
BioNet recombinant Tdap2$_{gen}$	N = 20	N = 19
	28.67	29.85
	(18.09-45.44)	(19.01-46.85)
BioNet recombinant TdaP5$_{gen}$	N = 18	N = 18
	95.04	78.31
	(55.85-161.72)	(50.72-120.91)
Comparator Tdap$_{chem}$	N = 19	N = 17
	15.97	14.04
	(10.04-25.38)	(8.67-22.72)
P-value[a]	<0.0001 [1]*	<0.0001 [1]*
The ratio of GMT or GMFR between Vaccine Group (95% CI)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 2.43 (0.88-6.72) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 5.79 (2.06-16.27)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 7.78 (2.73-22.21)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.80 (0.66-4.91) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.95 (2.12-16.72)*	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 2. 2.39 (0.83-6.90) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 5.96 (2.04-17.47)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 9.77 (3.28-29.04)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.13 (0.74-6.14) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.58 (1.90-16.34)*

Participants who participated in each visit
The ratio of GMT between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between vaccine groups
Note:
[1] P-value based on Kruskal-Wallis Test
* P-value ≤ 0.05 is considered statistically significant

The GMFR of PT-neutralizing antibodies at delivery from baseline were higher in participants from each of BioNet recombinant groups, except BioNet recombinant $Tdap2_{gen}$ compared to $Tdap_{chem}$ group.

*Table 50. Summary of PT neutralizing GMT (IU/ml) between baseline and delivery after vaccination as assessed by PT neutralization assay in CHO cells in a subset of 120 participants (20 participants per vaccine group) of mothers**

Vaccine	Baseline	Delivery	GMFR (Delivery/Baseline)	P-value[a]
	GMT (95% CI)	GMT (95% CI)	(95% CI)	
BioNet recombinant $ap1_{gen}$ (N=19)	5.53 (3.68-8.33)	38.74 (23.75-63.18)	7.00 (4.54-10.79)	<0.0001 [3]*
BioNet recombinant $ap2_{gen}$ (N=18)	9.05 (4.97-16.46)	92.47 (49.57-172.53)	10.22 (4.96-21.07)	<0.0001 [3]*
BioNet recombinant $aP5_{gen}$ (N=17)	6.23 (3.92-9.89)	124.26 (49.72-310.51)	19.95 (8.53-46.68)	<0.0001 [3]*
BioNet recombinant $Tdap2_{gen}$ (N=20)	5.73 (3.95-8.30)	28.67 (18.09-45.44)	5.01 (3.13-8.02)	<0.0001 [3]*
BioNet recombinant $TdaP5_{gen}$ (N=18)	9.56 (4.78-19.10)	95.04 (55.85-161.72)	9.94 (5.87-16.84)	<0.0001 [3]*
$Tdap_{chem}$ (N=18)	7.67 (4.57-12.87)	16.39 (10.05-26.71)	2.14 (1.50-3.04)	0.0002 [3]*
P-value[b]	0.5097 [1]	<0.0001 [1]*	<0.0001 [2]*	
Ratio between each of BioNet recombinant vaccines and $Tdap_{chem}$ (95% CI)		$ap1_{gen}$ - $Tdap_{chem}$ 2.36 (0.84-6.67)	$ap1_{gen}$ - $Tdap_{chem}$ 3.27 (1.20-8.95)*	
		$ap2_{gen}$ - $Tdap_{chem}$ 5.64 (1.97-16.15)*	$ap2_{gen}$ $Tdap_{chem}$ 4.78 (1.73-13.24)*	
		$aP5_{gen}$ - $Tdap_{chem}$ 7.58 (2.61-22.03)*	$aP5_{gen}$ - $Tdap_{chem}$ 9.33 (3.32-26.24)*	
		$Tdap2_{gen}$ - $Tdap_{chem}$ 1.75 (0.63-4.87)	$Tdap2_{gen}$ - $Tdap_{chem}$ 2.34 (0.87-6.32)	
		$TdaP5_{gen}$ - $Tdap_{chem}$ 5.80 (2.03-16.60)*	$TdaP5_{gen}$ - $Tdap_{chem}$ 4.65 (1.68-12.89)*	

Note:
nly for participants who participated in baseline and delivery visit
Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody titer at delivery to antibody titer at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between baseline and delivery
b : Compared between vaccine groups
Note:
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant

Seroconversion rates in PT-neutralizing antibody at 28 days post-vaccination and at delivery

The percentage of maternal participants with a ≥ 4-fold increase in PT neutralizing antibody titers assessed by PT neutralizing assay in CHO cells **at 28 days after vaccination** as compared to baseline were higher in BioNet recombinant $ap1_{gen}$, BioNet recombinant $aP5_{gen}$ and BioNet recombinant $TdaP5_{gen}$ compared to $Tdap_{chem}$ group (Table 51).

Table 51. Summary of difference in percentage of participants with a 4-fold or higher response in PT neutralizing antibody titers at 28 days after vaccination compared to baseline between each of BioNet's vaccine groups and $Tdap_{chem}$ group of mothers

Comparison groups	PT			
	BioNet Vaccine	Comparator $Tdap_{chem}$	Difference in percentage	P-value
	n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)	
BioNet recombinant $ap1_{gen}$ and $Tdap_{chem}$ (N=38 and N=37)	18 (90.00) (68.30-98.77)	10 (55.56) (30.76-78.47)	34.44 (6.40-59.06)	0.0265 [1]*
BioNet recombinant $ap2_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	17 (85.00) (62.11-96.79)	10 (55.56) (30.76-78.47)	29.44 (0.18-55.21)	0.0741 [1]
BioNet recombinant $aP5_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	18 (94.74) (73.97-99.87)	10 (55.56) (30.76-78.47)	39.18 (12.49-62.69)	0.0078 [1]*
BioNet recombinant $Tdap2_{gen}$ and $Tdap_{chem}$ (N=40 and N=37)	17 (85.00) (62.11-96.79)	10 (55.56) (30.76-78.47)	29.44 (0.18-55.21)	0.0741 [1]
BioNet recombinant $TdaP5_{gen}$ and $Tdap_{chem}$ (N=39 and N=37)	18 (90.00) (68.30-98.77)	10 (55.56) (30.76-78.47)	34.44 (6.40-59.06)	0.0265 [1]*

95% CI based on Clopper-Pearson method.
The two-sided 95% CI of the difference in the proportion was obtained based on Miettinen and Nurminen method
Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

The percentage of maternal participants with a ≥ 4-fold increase in PT neutralizing antibody titers assessed by PT neutralizing assay in CHO cells **at delivery** as compared to baseline were higher in all BioNet recombinant vaccine compared to $Tdap_{chem}$ group (Table 52).

Table 52. Summary of difference in percentage of participants with a 4-fold or higher response in PT neutralizing antibody titers at delivery compared to baseline between each of BioNet's vaccine groups and $Tdap_{chem}$ group of mothers

Comparison groups	PT			
	BioNet Vaccine	$Tdap_{chem}$	Difference in percentage	P-value
	n (%) (95% CI)	n (%) (95% CI)	(%) (95% CI)	
BioNet recombinant $ap1_{gen}$ and $Tdap_{chem}$ (N=19 and N=18)	14 (73.68) (48.80-90.85)	3 (16.67) (3.58-41.42)	57.02 (25.65-77.52)	0.0005 [1]*
BioNet recombinant $ap2_{gen}$ and $Tdap_{chem}$ (N=18 and N=18)	14 (77.78) (52.36-93.59)	3 (16.67) (3.58-41.42)	61.11 (29.55-80.67)	0.0002 [1]*
BioNet recombinant $aP5_{gen}$ and $Tdap_{chem}$ (N=17 and N=18)	13 (76.47) (50.10-93.19)	3 (16.67) (3.58-41.42)	59.80 (27.62-79.99)	0.0003 [1]*
BioNet recombinant $Tdap2_{gen}$ and $Tdap_{chem}$ (N=20 and N=18)	13 (65.00) (40.78-84.61)	3 (16.67) (3.58-41.42)	48.33 (17.19-70.65)	0.0025 [1]*
BioNet recombinant $TdaP5_{gen}$ and $Tdap_{chem}$ (N=18 and N=18)	15 (83.33) (58.58-96.42)	3 (16.67) (3.58-41.42)	66.67 (35.72-84.42)	<0.0001 [1]*

95% CI based on Clopper-Pearson method.
The two-sided 95% CI of the difference in the proportion was obtained based on Miettinen and Nurminen method

[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

Infant participants:

GMCs of anti-PT and anti-FHA antibodies at 2 months and 7 months of age

Table 53. Summary of anti-PT and anti-FHA GMCs (IU/ml) in infants as assessed by ELISA at 2 and 7 months of age by vaccine groups

	PT		FHA	
Vaccine	2 Months of age GMC (IU/mL) (95% CI)	7 Months of age GMC (IU/mL) (95% CI)	2 Months of age GMC (IU/mL) (95% CI)	7 Months of age GMC (IU/mL) (95% CI)
BioNet recombinant ap1$_{gen}$	N = 38 22.36 (15.01-33.31)	N = 37 27.09 (18.05-40.64)	N = 38 37.78 (29.89-47.76)	N = 37 13.60 (8.93-20.71)
BioNet recombinant ap2$_{gen}$	N = 37 29.65 (21.36-41.16)	N = 33 24.87 (16.38-37.78)	N = 37 66.27 (51.49-85.30)	N = 33 18.65 (11.53-30.18)
BioNet recombinant aP5$_{gen}$	N = 34 60.46 (38.92-93.92)	N = 35 17.77 (13.06-24.18)	N = 34 83.74 (63.46-110.51)	N = 35 19.01 (12.53-28.84)
BioNet recombinantTdap2$_{gen}$	N = 38 19.28 (15.09-24.63)	N = 38 19.83 (12.23-32.14)	N = 38 59.93 (48.71-73.74)	N = 38 19.87 (11.98-32.94)
BioNet recombinant TdaP5$_{gen}$	N = 31 52.94 (36.25-77.31)	N = 31 20.06 (14.07-28.58)	N = 31 56.87 (40.64-79.58)	N = 31 14.91 (9.20-24.18)
Comparator Tdap$_{chem}$	N = 35 10.74 (7.65-15.07)	N = 33 40.98 (26.59-63.15)	N = 35 33.12 (22.34-49.09)	N = 33 27.26 (15.60-47.63)
P-value[a]	<0.0001 [1]*	0.0333 [1]*	<0.0001 [1]*	0.3858 [1]
The ratio of GMC between Vaccine Group (99% CI)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 2.08 (1.10-3.94)* BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.76 (1.45-5.25)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.63 (2.92-10.85)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.80 (0.95-3.40) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 4.93 (2.52-9.65)*	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 0.66 (0.32-1.38) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.61 (0.28-1.29) BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.43 (0.21-0.91)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.48 (0.23-1.01) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.49 (0.23-1.06)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 1.14 (0.69-1.90) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.00 (1.20-3.34)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 2.53 (1.50-4.27)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.81 (1.09-3.01)* BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 1.72 (1.00-2.94)*	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 0.50 (0.21-1.18) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.68 (0.28-1.66) BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.70 (0.29-1.67) BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.73 (0.31-1.72) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.55 (0.22-1.35)

Note:
The ratio of GMC between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
[a] : Compared between vaccine groups

[1] P-value based on One-way ANOVA
* P-value ≤ 0.05 is considered statistically significant.

Table 54. Summary of anti-PT GMCs (IU/ml) in infants as assessed by ELISA at time of birth (cord blood or neonatal blood within 72 hours after birth) and 2 months of age by vaccine groups

Vaccine	Time of Birth	2 Months of age	GMC Ratio between 2 months of age and time of birth	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMCR (95% CI)	
BioNet recombinant ap1$_{gen}$ (N=37)	62.85 (43.60-90.60)	22.81 (15.17-34.29)	0.36 (0.30-0.43)	<0.0001 [3]*
BioNet recombinant ap2$_{gen}$ (N=34)	78.17 (51.86-117.81)	29.20 (20.46-41.68)	0.37 (0.29-0.49)	<0.0001 [3]*
BioNet recombinant aP5$_{gen}$ (N=34)	141.65 (93.70-214.14)	60.46 (38.92-93.92)	0.43 (0.37-0.49)	<0.0001 [3]*
BioNet recombinant Tdap2$_{gen}$ (N=38)	51.93 (41.34-65.23)	19.28 (15.09-24.63)	0.37 (0.32-0.43)	<0.0001 [3]*
BioNet recombinant TdaP5$_{gen}$ (N=31)	136.40 (92.99-200.07)	52.94 (36.25-77.31)	0.39 (0.33-0.45)	<0.0001 [3]*
Comparator Tdap$_{chem}$ (N=34)	27.26 (18.10-41.05)	10.47 (7.41-14.79)	0.38 (0.32-0.47)	<0.0001 [3]*
P-value[b]	<0.0001 [1]*	<0.0001 [2]*	0.4257 [1]	
The ratio of GMC or GMCR between Vaccine Group (99% CI)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 2.31 (1.19-4.47)* BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.87 (1.46-5.63)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.20 (2.65-10.20)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.90 (0.99-3.67) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.00 (2.51-9.98)*	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 2.18 (1.13-4.18)* BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.79 (1.43-5.43)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.77 (2.97-11.24)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.84 (0.96-3.52) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.06 (2.56-10.00)*	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 0.94 (0.68-1.32) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.97 (0.69-1.36) BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 1.11 (0.79-1.56) BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.97 (0.70-1.34) BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 1.01 (0.71-1.43)	

Note:
Only for participants whose samples available at Time of birth and 2 Months of age
GMC ratio (GMCR) is geometric mean of the ratios of antibody concentration at 2 months of age to antibody concentration at time of birth
The ratio of GMC or GMCR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between Time of birth and 2 Months of age
b : Compared between vaccine groups

[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 55. Summary of anti-PT GMCs (IU/ml) in infants as assessed by ELISA at 2 months of age and 7 months of age by vaccination groups

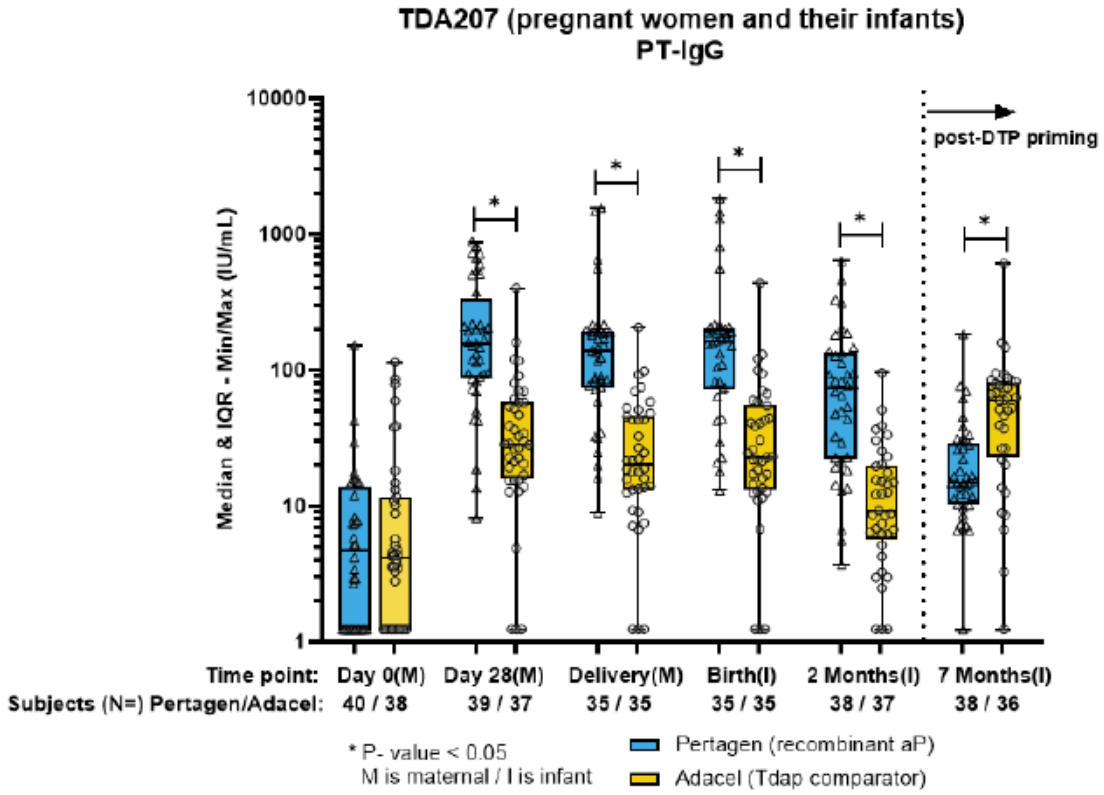
Vaccine	2 Months of age	7 Months of age	GMC Ratio between 7 months of age and 2 months of age	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMCR (95% CI)	
BioNet recombinant ap1$_{gen}$ (N=37)	21.36 (14.34-31.82)	27.09 (18.05-40.64)	1.27 (0.63-2.54)	0.4913 [3]
BioNet recombinant ap2$_{gen}$ (N=32)	31.16 (21.89-44.35)	24.69 (16.04-38.01)	0.79 (0.40-1.56)	0.4866 [3]
BioNet recombinant aP5$_{gen}$ (N=33)	59.64 (37.89-93.87)	17.20 (12.52-23.64)	0.29 (0.17-0.50)	<0.0001 [3]*
BioNet recombinant Tdap2$_{gen}$ (N=38)	19.28 (15.09-24.63)	19.83 (12.23-32.14)	1.03 (0.60-1.77)	0.9175 [3]
BioNet recombinant TdaP5$_{gen}$ (N=31)	52.94 (36.25-77.31)	20.06 (14.07-28.58)	0.38 (0.22-0.65)	0.0009 [3]*
Comparator Tdap$_{chem}$ (N=33)	11.29 (7.95-16.04)	40.98 (26.59-63.15)	3.63 (1.87-7.04)	0.0003 [3]*
P-value[b]	<0.0001 [2]*	0.0308 [1]*	<0.0001 [1]*	
The ratio of GMC or GMCR between Vaccine Group (99% CI)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 1.89 (0.99-3.63) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 2.76 (1.40-5.43)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 5.28 (2.70-10.33)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 1.71 (0.89-3.27)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 0.66 (0.32-1.38) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.60 (0.28-1.30) BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.42 (0.20-0.90)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.48 (0.23-1.01)	BioNet recombinant ap1$_{gen}$ - Comparator Tdap$_{chem}$ 0.35 (0.12-1.05) BioNet recombinant ap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.22 (0.07-0.68)* BioNet recombinant aP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.08 (0.03-0.25)* BioNet recombinant Tdap2$_{gen}$ - Comparator Tdap$_{chem}$ 0.28 (0.09-0.85)*	

	BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 4.69 (2.37-9.27)*	BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.49 (0.23-1.06)	BioNet recombinant TdaP5$_{gen}$ - Comparator Tdap$_{chem}$ 0.10 (0.03-0.33)*

Note:
Only for participants whose samples available at 2 months of age and 7 months of age
GMC ratio (GMCR) is geometric mean of the ratios of antibody concentration at 7 months of age to antibody concentration at 2 months of age
The ratio of GMC or GMCR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between 2 months of age and 7 months of age
b : Compared between vaccine groups

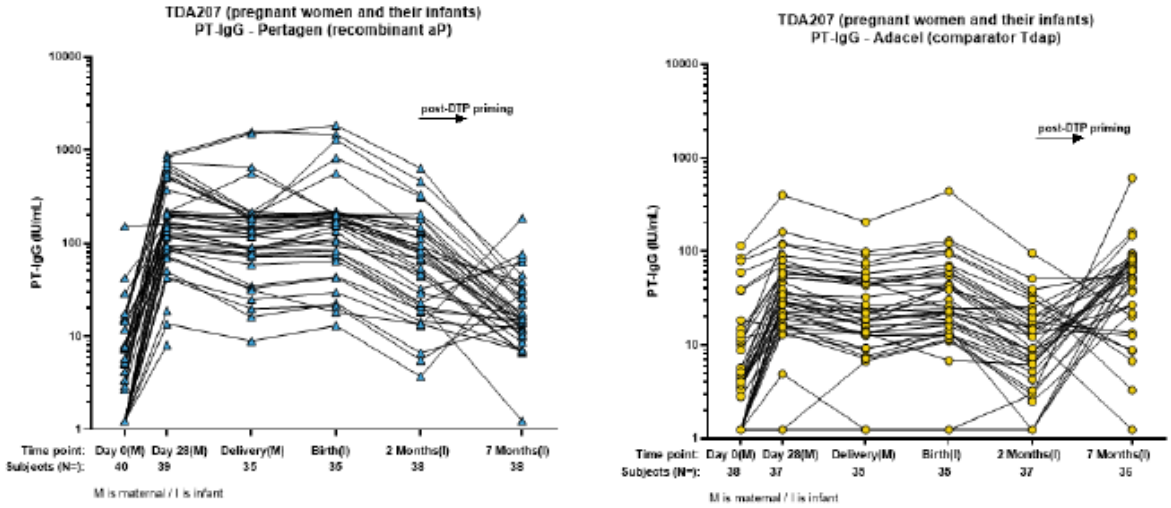
[1] P-value based on Kruskal-Wallis Test
[2] P-value based on One-way ANOVA
[3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Figure 14. Boxplots for PT-IgG levels assessed in pregnant women and their infants in the TDA027 trial



Legend: The figure shows individual data points and boxplots presenting the lowest (minimum) and highest (maximum) value of PT-IgG antibody assessed in pregnant women and their infants participating in the TDA207 trial, before (Day 0), 28 days after and at the time of delivery in mothers (M) after maternal vaccination during pregnancy with VACPERTAGEN or Adacel® (commercialised in Europe as Triaxis), and in infants (I) at the time of birth, at 2 months of age, and at 7 months of age after completing 3-priming doses of diphtheria-tetanus-pertussis (DTP) vaccination at 2, 4 and 6 months of age. Statistical differences between vaccine groups were tested based on Bonferroni post hoc test on pairwise comparisons.

Figure 15. Spaghetti plots for PT-IgG levels assessed in pregnant women and their infants in the TDA027 trial



Legend: The figure shows spaghetti plots presenting PT-IgG concentrations for individual mothers (M) and infants (I) participating in the TDA207 trial, before (Day 0), 28 days after and at the time of delivery in mothers (M) after maternal vaccination during pregnancy with VACPERTAGEN (left hand panel, blue triangular symbols) or Adacel® (right hand panel, yellow circular symbols), and in infants (I) at the time of birth, at 2 months of age, and at 7 months of age after completing 3-priming doses of diphtheria-tetanus-pertussis (DTP) vaccination at 2, 4 and 6 months of age. No statistical testing was performed.

Difference in percentage of infants with a 4-fold or higher response between month 2 (before primary immunisation) and month 7 (= one month after primary immunisation with 3 vaccinations)

Table 56. Summary of difference in percentage of participants with a 4-fold or higher response in anti-PT antibody concentrations at 7 months of age compared to 2 months of age between each of BioNet's vaccine groups and comparator Tdap$_{chem}$ groups of infants (Per Protocol population)

Comparison groups	BioNet vaccine	Comparator Tdap$_{chem}$	Difference in percentage of participants with ≤4-fold response	P-value
	n (%) (95% CI)	n (%) (95% CI)	% (95% CI)	
BioNet recombinant ap1$_{gen}$ and Comparator Tdap$_{chem}$	10 (27.03)	17 (51.52)	-24.49	0.0356 [1]*
(N=37 and N=33)	(13.79-44.12)	(33.54-69.20)	(-45.31-1.48)	
BioNet recombinant ap2$_{gen}$ and Comparator Tdap$_{chem}$	6 (18.75)	17 (51.52)	-32.77	0.0057 [1]*
(N=32 and N=33)	(7.21-36.44)	(33.54-69.20)	(-52.76-9.59)	
BioNet recombinant aP5$_{gen}$ and Comparator Tdap$_{chem}$	3 (9.09)	17 (51.52)	-42.42	0.0001 [1]*
(N=33 and N=33)	(1.92-24.33)	(33.54-69.20)	(-60.52-21.31)	
BioNet recombinant Tdap2$_{gen}$ and Comparator Tdap$_{chem}$	11 (28.95)	17 (51.52)	-22.57	0.0522 [1]
(N=38 and N=33)	(15.42-45.90)	(33.54-69.20)	(-43.54-0.39)	
BioNet recombinant TdaP5$_{gen}$ and Comparator Tdap$_{chem}$	2 (6.45)	17 (51.52)	-45.06	<0.0001 [1]*
(N=31 and N=33)	(0.79-21.42)	(33.54-69.20)	(-62.66-24.40)	

95% CI based on Clopper-Pearson method.
The two-sided 95% CI of the difference in the proportion was obtained based onMiettinen and Nurminen method

Note:
[1] Overall p-value (2-sided) based on Chi-square test
* P-value ≤ 0.05 is considered statistically significant.

GMTs of PT-neutralising antibody titre in infants

Table 57. Summary of PT neutralising GMTs (IU/ml) as assessed by PT neutralising assay in CHO cells between time of birth (cord blood or neonatal blood within 72 hours after birth) and 2 months of age in a subset of 120 infant participants (20 participants each vaccine group) by vaccine groups

Vaccine	Time of Birth	2 Months of age	GMT Ratio between 2 months of age and time of birth	P-value[a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMT Ratio (95% CI)	
BioNet recombinant $ap1_{gen}$	30.66	16.75	0.55	<0.0001 [2]*
(N=18)	(17.24-54.53)	(10.27-27.32)	(0.47-0.64)	
BioNet recombinant $ap2_{gen}$	83.72	39.85	0.48	0.0001 [2]*
(N=18)	(46.77-149.85)	(23.11-68.72)	(0.34-0.66)	
BioNet recombinant $aP5_{gen}$	141.35	62.84	0.44	<0.0001 [2]*
(N=16)	(49.76-401.57)	(25.59-154.35)	(0.35-0.57)	
BioNet recombinant $Tdap2_{gen}$	33.91	17.71	0.52	<0.0001 [2]*
(N=18)	(23.08-49.83)	(11.94-26.26)	(0.42-0.66)	
BioNet recombinant $TdaP5_{gen}$	84.61	39.57	0.47	<0.0001 [2]*
(N=17)	(55.10-129.94)	(27.68-56.57)	(0.38-0.58)	
Comparator $Tdap_{chem}$	13.69	6.83	0.50	<0.0001 [2]*
(N=16)	(8.20-22.84)	(4.35-10.72)	(0.41-0.61)	
P-value[b]	<0.0001 [1]*	<0.0001 [1]*	0.9189 [1]	
The ratio of GMT or GMTR between Vaccine Group (99% CI)	BioNet recombinant $ap1_{gen}$ - Comparator $Tdap_{chem}$ 2.45 (0.94-6.37)	BioNet recombinant $ap1_{gen}$ - Comparator $Tdap_{chem}$ 2.45 (0.94-6.37)	BioNet recombinant $ap1_{gen}$ - Comparator $Tdap_{chem}$ 1.09 (0.72-1.66)	
	BioNet recombinant $ap2_{gen}$ - Comparator $Tdap_{chem}$ 5.83 (2.24-15.16)*	BioNet recombinant $ap2_{gen}$ - Comparator $Tdap_{chem}$ 5.83 (2.24-15.16)*	BioNet recombinant $ap2_{gen}$ - Comparator $Tdap_{chem}$ 0.95 (0.63-1.44)	
	BioNet recombinant $aP5_{gen}$ - Comparator $Tdap_{chem}$ 9.20 (3.44-24.58)*	BioNet recombinant $aP5_{gen}$ - Comparator $Tdap_{chem}$ 9.20 (3.44-24.58)*	BioNet recombinant $aP5_{gen}$ - Comparator $Tdap_{chem}$ 0.89 (0.58-1.36)	
	BioNet recombinant $Tdap2_{gen}$ - Comparator $Tdap_{chem}$ 2.59 (1.00-6.74)	BioNet recombinant $Tdap2_{gen}$ - Comparator $Tdap_{chem}$ 2.59 (1.00-6.74)	BioNet recombinant $Tdap2_{gen}$ - Comparator $Tdap_{chem}$ 1.05 (0.69-1.58)	
	BioNet recombinant $TdaP5_{gen}$ - Comparator $Tdap_{chem}$ 5.79 (2.20-15.26)*	BioNet recombinant $TdaP5_{gen}$ - Comparator $Tdap_{chem}$ 5.79 (2.20-15.26)*	BioNet recombinant $TdaP5_{gen}$ - Comparator $Tdap_{chem}$ 0.94 (0.62-1.43)	

Note:
Only for participants whose samples available at time of birth and 2 months of age
GMT ratio (GMTR) is geometric mean of the ratios of antibody titer at 2 months of age to antibody titer at time of birth
The ratio of GMT or GMTR between vaccine groups (95% CI) based on Bonferroni post-hoc analysis with 5 pairwise comparisons
a : Compared between Time of birth and 2 Months of age
b : Compared between vaccine groups

[1] P-value based on Kruskal-Wallis Test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 58. Summary of PT neutralising GMT (IU/ml) as assessed by PT neutralising assay in CHO cells at 2 months and 7 months of age in a subset of 120 infant participants (20 participants each vaccine group) by vaccine group

Vaccine	2 Months of age	7 Months of age
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)
BioNet recombinant $ap1_{gen}$	N = 18	N = 17
	16.75	10.53
	(10.27-27.32)	(5.39-20.59)
BioNet recombinant $ap2_{gen}$	N = 19	N = 16
	39.96	9.58
	(23.92-66.77)	(5.16-17.78)
BioNet recombinant $aP5_{gen}$	N = 16	N = 18
	62.84	12.57
	(25.59-154.35)	(7.82-20.23)
BioNet recombinant $Tdap2_{gen}$	N = 19	N = 19
	17.87	7.56
	(12.32-25.91)	(5.03-11.37)
BioNet recombinant $TdaP5_{gen}$	N = 17	N = 17
	39.57	6.30
	(27.68-56.57)	(4.28-9.28)
Comparator $Tdap_{chem}$	N = 18	N = 17
	7.43	14.72
	(4.89-11.28)	(7.74-28.02)
P-value[a]	<0.0001 [1]*	0.1501 [1]
The ratio of GMT or GMFR between Vaccine Group (99% CI)	BioNet recombinant $ap1_{gen}$ - Comparator $Tdap_{chem}$ 2.26 (0.91-5.62)	BioNet recombinant $ap1_{gen}$ - Comparator $Tdap_{chem}$ 0.72 (0.28-1.86)
	BioNet recombinant $ap2_{gen}$ - Comparator $Tdap_{chem}$ 5.38 (2.19-13.25)*	BioNet recombinant $ap2_{gen}$ - Comparator $Tdap_{chem}$ 0.72 (0.28-1.86)
	BioNet recombinant $aP5_{gen}$ - Comparator $Tdap_{chem}$ 8.46 (3.30-21.68)*	BioNet recombinant $aP5_{gen}$ - Comparator $Tdap_{chem}$ 0.85 (0.33-2.19)
	BioNet recombinant $Tdap2_{gen}$ - Comparator $Tdap_{chem}$ 2.41 (0.98-5.92)	BioNet recombinant $Tdap2_{gen}$ - Comparator $Tdap_{chem}$ 0.51 (0.20-1.30)
	BioNet recombinant $TdaP5_{gen}$ - Comparator $Tdap_{chem}$ 5.33 (2.11-13.45)*	BioNet recombinant $TdaP5_{gen}$ - Comparator $Tdap_{chem}$ 0.43 (0.16-1.11)*

2.6.5.3. Supportive study(ies)

For the purpose of this application, PertADO, APV301, Pertaprime-01, TDA203, TDA204, PerMIT and WoMAN-POWER are considered supportive clinical studies. APV301 was a safety study only. PerMIT was an observational study which collected antibody level in cord sera.

Adolescents

Study PertADO - A Phase II Randomized, Observer-blind Controlled Pilot Study to Compare the Safety and Immunogenicity of Acellular Pertussis Vaccines Including Chemically or Genetically-detoxified Pertussis Toxin in Adolescents Aged 11-15 Years Previously Immunized With Acellular Pertussis Vaccines

No CSR but a scientific publication (Rohner et al. 2018, Clinical Infectious Diseases, 2019;68(7):1213–22) was provided for this study (Clinical Trials Registration NCT02946190).

Methods

PertADO was an investigator-driven, single-center, phase 2, observer-blinded randomized controlled trial in aP-primed adolescents in Geneva to assess the immunogenicity and reactogenicity of a novel recombinant aP (r-aP) vaccine including recombinant per- tussis toxin (PT) and filamentous hemagglutinin (FHA) co-administered with tetanus-diphtheria toxoids (Td), compared to a licensed tetanus-diphtheria-aP vaccine containing chemically detoxified PT (cd/Tdap).

The primary objective of this RCT was to test whether adolescents primed and boosted with cdPT-containing aP vaccines would respond better to cd/Tdap or to r-aP + Td, based on day 28 geometric mean concentrations (GMCs) of anti-PT neutralizing antibodies. Secondary objectives included seroresponse rates and GMCs of PT-, filamentous hemagglutinin (FHA)–, tetanus toxoid (TT)–, and diphtheria toxoid (DT)–specific total immunoglobulin G (IgG) antibodies, as well as the incidence of adverse events (AEs) and serious adverse events (SAEs) during 7 and 28 days, respectively. The durability of vaccine responses was assessed at day 365 through a study extension.

The sample size was based on practical considerations. Immunogenicity evaluations were descriptive.

Study Population

The study was conducted in Switzerland, between October 2016 and March 2017.

Adolescents 11 to 15 years of age with a documented history of aP immunisation (5 doses) were recruited through private pediatricians and flyers. Vaccination history was retrieved from vaccination records.

Eligible participants were randomized to r-aP (VacPertagen) with contralateral Td (Td-pur; GlaxoSmithKline AG) or the comparator cd/Tdap (Boostrix; GlaxoSmithKline AG) vaccines. Group allocation was performed on a 1:1 basis generated by computer randomization. The study was observer-blinded regarding the administration of 1 or 2 vaccines: The immunizing nurses and participants were aware of the number and type of vaccines administered, whereas the other investigators remained blinded.

There were 2 initial study visits: visit 1 for inclusion, randomization, venous bleed, and vaccination, and visit 2 for safety evaluation, blood draw, and study termination. A third visit was subsequently added on day 365 to assess the durability of vaccine responses.

Study Vaccines

VacPertagen was developed and produced by BioNet. A single dose contains 5 µg of rPT and 5 µg of FHA. Td-pur contains 20 IU of TT and 2 IU of DT. The cd/ Tdap comparator (Boostrix) contains 8 µg of cdPT, 8 µg of FHA, 2.5 µg of pertactin, 20 IU of TT, and 2 IU of DT. All vaccines, adsorbed on aluminium hydroxide, were administered intra- muscularly.

Safety Assessment

Following vaccination, subjects were observed during 30 minutes for immediate reactions. Diary cards recorded solicited local (pain, redness, and swelling) and systemic (fever, head- ache, fatigue, arthralgia, chills, malaise, myalgia, and vomiting) reactions during 7 days after vaccination, and AEs and SAEs for 28 days. Causality of AEs and SAEs to study vaccines was determined by the investigators according to ICH guidelines, as specified in the protocol.

Immunogenicity Assessment

Blood samples were taken at baseline, day 28, and day 365 after vaccination. The primary immunological endpoint was the PT-neutralizing GMCs. Secondary endpoints included seroresponses (defined by ≥4-fold antibody increases) and GMCs of PT-, FHA-, TT-, and DT-specific IgG antibodies.

Statistical Analyses

Statistical analyses were performed by the Center of Excellence for Biomedical and Public Health Informatics, Thailand, using SAS version 9.4 software. The sample size for this proof-of- concept study was based on practical considerations and not on a formal statistical power calculation. Sixty volunteers were planned for randomization, with 30 in each group. The safety analysis included all randomized subjects who had received a dose of study vaccine. The overall percentage of subjects with

at least 1 spontaneously reported AE, with date of onset up to day 28 after vaccination, were tabulated with exact 95% confidence interval (CI), by type of AE, by severity, and by causality. They were displayed by vaccine group as both frequencies and per- centages on the intent-to-treat data set. The seroresponse rates and GMCs were calculated with exact 95% CI. The difference between pre- and post- GMCs or Bmems within a group was assessed using the paired *t* test or Wilcoxon signed-rank test, depending on the distribution of data. The difference between groups was assessed by either $\chi 2$ or Fisher exact test for categorical variables, by the Student *t* test or Mann-Whitney *U* test for continuous variables, and by the Student independent *t* test or Wilcoxon rank-sum test for Bmems. $P \leq .05$ was considered to be statistically significant.

Results

Demographics

In total, 62 aP-primed adolescents (mean age 12.2 years; 92% white) were randomized and vaccinated with r-aP + Td or cd/Tdap. Baseline characteristics including GMCs were similar between the groups.

Immunogenicity evaluation

At 28 days after vaccination, anti-PT GMCs were higher after r-aP + Td (113.74 [95% CI, 88.31–146.50] IU/mL; P = .0006) compared to cd/Tdap (52.43 [95% CI, 36.41–75.50] IU/mL). Also, PT-neutralizing GMCs tended to be higher after r-aP + Td (127.68 [95% CI, 96.73–168.53] IU/ mL; P = .0162) compared to cd/Tdap (73.91 [95% confidence interval {CI}, 49.88–109.52] IU/mL). The day 28 anti-FHA GMCs were similar in both groups. Day 365 anti-PT (but not PT-neutralizing) GMCs were slightly higher in r-aP + Td vaccinees (26.87 [95% CI, 19.51–37.00] versus 15.75 [95% CI, 10.22–24.27]).

Adults

Study Pertaprime - An investigator-driven phase II-III randomised, observer-blind, controlled trial to demonstrate non-inferior immunogenicity of VacPertagen in comparison to Boostrix in healthy young Australian adults aged 18-30 years

Methods

Pertaprime-01 is a phase II-III, observer-blind, randomised, active-controlled vaccine trial including 102 healthy participants (aged 18 to 30 years of age) recruited from a site in Australia who were randomised 2:1 to receive VacPertagen or Boostrix based on a stratification for wP or aP_{chem} vaccination in infancy.

Table 59. Vaccine groups

	wP primed (*51 participants*)	aP primed (*51 participants*)	Total
Pertagen®	34 participants	34 participants	68 participants
Boostrix®	17 participants	17 participants	34 participants

The aim of this study was to demonstrate the safety and non-inferior immunogenicity for pertussis antigens of VacPertagen (BioNet-Asia; not licensed in Australia) compared to the most used pertussis-

containing booster vaccine in Australia (Boostrix GlaxoSmithKline; licensed in Australia for vaccination of individuals aged 4 years and older) in healthy young Australian adults aged 18 to 30 years old.

Study duration: 01 Jul 2020 (First Participant First Visit, FSFV) - 26 Sep 2023 (Last Participant Last Visit). One single study site.

Study Population

A total of 102 eligible 18-40 years of age participants, of whom half (n = 51) had received a whole cell pertussis vaccine as their first pertussis vaccination, and other half (n = 51) had received 3 doses of acellular pertussis vaccines as their first vaccines during infancy were planned to be enrolled. Only those participants who fulfilled all of the inclusion criteria and none of the exclusion criteria were randomized.

Treatments

A single batch for investigational vaccine VacPertagen was used for the entire study. Boostrix vaccine was used as comparator vaccine for this study. Both vaccines were presented in a blinded single-dose of 0.5 mL in a prefilled syringe.

Table 60. Composition of Investigational and reference vaccine administered to the study participants

Name of ingredients	Amount per 0.5-mL dose	
	Pertagen® (Investigational)	Boostrix® (Reference)
Active substance		
Tetanus Toxoid (TT)	-	5 Lf
Diphtheria Toxoid (DT)	-	2.5 Lf
Pertussis Toxoid (PT)	5 µg [a]	8 µg [b]
Filamentous hemagglutinin (FHA)	5 µg	8 µg
Pertactin (PRN)	-	2.5 µg
Excipients		
Aluminum Hydroxide	0.3 mg/dose (as Al^{3+})	0.3 mg/dose (as Al^{3+})
Aluminium Phosphate	-	0.2 mg/dose (as Al^{3+})
Sodium Chloride	4.38 mg	4.5 mg
Water for Injections	q.s. to 0.5 mL	q.s. to 0.5 mL

a) Genetically detoxified; b) chemically detoxified

Randomization and Blinding

Participants were block randomized to receive one dose of the 2 vaccines: VacPertagen (investigational vaccine) or Boostrix (reference vaccine). Eligible participants who provided their written consent were enrolled into the study and randomized according to the randomization list.

The trial was carried out in an observer-blind manner until the end of the study.

Study Assessments

Approximately 10 mL of whole blood was taken from all participants at Visit 1 (day 0) before vaccination (baseline), at Visit 2 (day 28) post-vaccination and at Visit 3 (day 365) post-vaccination to collect serum.

The serum was used to assess immunogenicity and persistence of vaccine antibody responses. PT- IgG and FHA-IgG titres (IU/mL) were assessed using validated ELISA or multiplex immune (MIA) assays. Functional antibodies were assessed using the Chinese Hamster Ovary (CHO) PT neutralization assay. Seroconversion for PT-IgG, FHA-IgG and PT neutralization antibodies were defined as a 4-fold increase of antibody titers at 28 days post-vaccination compared to baseline titers.

Objectives and Endpoints

Primary Objective

- To demonstrate non-inferior immunogenicity of one dose of VacPertagen as compared to Boostrix at 28 days after vaccination based on IgG antibody seroconversion rates for PT and FHA antigens.

Primary Endpoint

- Seroconversion rates as defined by the proportion of participants with ≥ 4-fold increase with respect to baseline of PT-IgG and FHA-IgG antibodies in VacPertagen and Boostrix groups in all participants at day 28 post-vaccination.

Secondary Objectives

- To assess the immunogenicity of one dose of VacPertagen as compared to Boostrix based on antibody geometric mean titer (GMT) day 28 post-vaccination.
- To assess immune persistence 1-year after administration of one dose of VacPertagen as compared to Boostrix based on antibody GMT and seroconversion rates.
- To assess the safety of one dose of VacPertagen as compared to Boostrix based on solicited adverse events (AEs) until day 7 post-vaccination, unsolicited AEs until day 28 post-vaccination, all serious AEs for 6 months post-vaccination, and related serious AEs for 1-year post-vaccination.
- To assess the immunogenicity and immune persistence of pertussis booster vaccination (28 days and 1-year after vaccination with one dose of VacPertagen or one dose of Boostrix, respectively) in young adults whose first dose of pertussis vaccination and optionally subsequent pertussis vaccinations were wP-containing vaccines, or who in the 1st year of life were vaccinated exclusively with 3 doses of chemically inactivated aP_{chem}-containing vaccines

Secondary Endpoints

- Geometric mean titres (GMTs) of PT-IgG, FHA-IgG and PT neutralization antibodies at Day 28 post-vaccination in all participants vaccinated with VacPertagen compared to Boostrix.
- Persistence of GMTs of PT-IgG, FHA-IgG and PT neutralization antibodies 1 year after vaccination in all participants vaccinated with VacPertagen compared to Boostrix.
- Seroconversion rates as defined by proportion of participants with ≥ 4-fold increase with respect to baseline of PT neutralization antibodies in VacPertagen and Boostrix groups at day 28 post- vaccination.

Statistical Analyses

The ITT and PP data set was used for the analysis of safety and immunogenicity.

The primary immunogenicity endpoint (non-inferiority of VacPertagen as compared to Boostrix) was seroconversion rates as defined by percentage of participants with ≥4-fold increase of IgG antibodies to PT and FHA at Day 28 with respect to baseline (Day 0; before vaccination).

The following descriptive statistics were provided for each variable: number of participants, percentages, mean, geometric mean, standard deviation, median, minimum, maximum, and range.

Percentage of participants with a four-fold or higher response, as compared with baseline titer, were computed for each vaccine group along with its corresponding exact two-sided 95% CI: based on Clopper-Pearson method. The difference in the percentages between two vaccine groups were

determined using chi-square test/ Fisher's exact test. The two-sided 95% CI: of the difference in the percentages was obtained based on Miettinen and Nurminen method. Geometric mean antibody concentration and its 95% CI, pre- and post-vaccination, were calculated for each vaccine group. The comparison between pre- and post-vaccination in each vaccine group was determined using paired t-test and then compared between two vaccine groups using Independent t-test / Wilcoxon rank sum test.

Statistical Hypothesis:

H_0: Seroconversion rate among different vaccine groups (VacPertagen minus Boostrix) ≤-0.10.

H_A: Seroconversion rate among different vaccine groups (VacPertagen minus Boostrix) >-0.10

Sample size calculation: the sample size was calculated based on non-inferiority test with alpha level of 0.05, 80% power, 25% drop out and non-inferiority margin of 10%, the sample size required for this study is 102 subjects in total (68 in VacPertagen and 34 in Boostrix groups).

Results

Changes in the planned conduct of the study

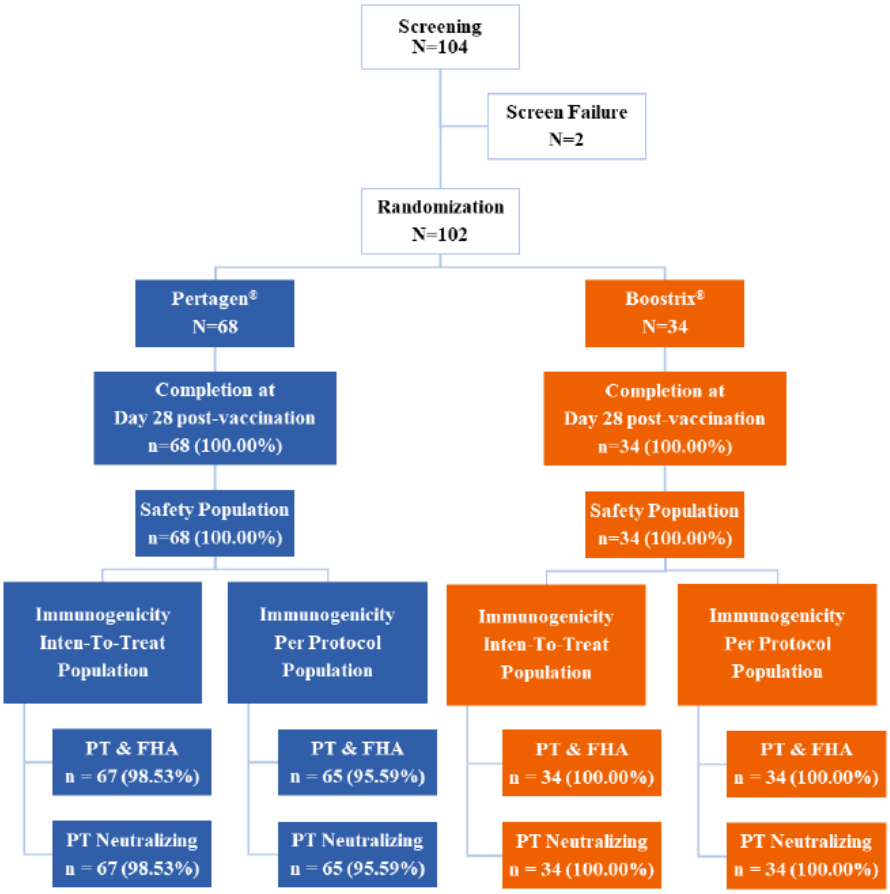
During the conduct of this study from Visit 0 (screening) to Visit 2 (Day 28 post-immunisation), 5 major and 8 minor protocol deviations were reported. None of the protocol deviations affected participants safety or led to discontinuation from the study.

Participant flow

A total of 104 volunteers were screened, of whom 102 participants were enrolled and randomized into two vaccine groups [68 participants in VacPertagen (34 participants each were wP and aP_{chem} primed) and 34 participants (17 participants each were wP and aP_{chem} primed) in Boostrix vaccine group].

At Day 28 post-vaccination, all 102 enrolled participants were included in the safety analysis, 101 (99.02%) participants [except 1 participant in VacPertagen vaccine group due to consent withdrawal] completed the study and were included in immunogenicity analysis for ITT population and 99 participants (97.06%) were included in immunogenicity analysis for PP population. Three (2.94%) participants in VacPertagen vaccine group were excluded from PP population

Figure 16. Overall participant disposition ad Day 28 post-vaccination



Baseline data

Baseline characteristics were balanced between groups. The mean age of subjects was 20.6 years and 84.3% were of Caucasian ethnicity.

Table 61. Summary of demographics at baseline

Participants status	Pertagen®	Boostrix®	Total	P-value
Demographics at baseline (Screening)				
Age (months)				
-N	68	34	102	
-Mean (SD)	20.51 (2.37)	20.82 (2.37)	20.62 (2.36)	0.4873 [4]
-Median	20.00 (18.5-22)	20.50 (19-23)	20.00 (19-22)	
-Min-Max	18-30	18-25	18-30	
Sex				1.0000 [1]
-Male	22 (32.35)	11 (32.35)	33 (32.35)	
-Female	46 (67.65)	23 (67.65)	69 (67.65)	
Ethnicity: n (%)				0.9176 [2]
-Aboriginal and/or Torres Strait Islander	2 (2.94)	0 (0.00)	2 (1.96)	
-Asian	3 (4.41)	2 (5.88)	5 (4.90)	
-Black or African	0 (0.00)	0 (0.00)	0 (0.00)	
-Caucasian	56 (82.35)	30 (88.24)	86 (84.31)	
-Indian-subcontinent	4 (5.88)	1 (2.94)	5 (4.90)	
-Pacific Islander	0 (0.00)	0 (0.00)	0 (0.00)	
-Other	3 (4.41)	1 (2.94)	4 (3.92)	
Anglo/Indian/Burmese	0 (0.00)	1 (2.94)	0 (0.00)	
Asian/Caucasian	2 (2.94)	0 (0.00)	2 (1.96)	
Latino (South American)	1 (1.47)	0 (0.00)	1 (0.98)	
Weight (kg)				
-N	68	34	102	
-Mean (SD)	74.09 (15.73)	75.69 (15.35)	74.63 (15.55)	0.6265 [3]
-Median (Q1-Q3)	72.45 (63.25-80.75)	72.45 (65.5-88.2)	72.45 (64-83.3)	
-Min-Max	41.3-114	44.8-109.4	41.3-114	
Height (cm)				
-N	68	34	102	
-Mean (SD)	172.04 (8.18)	172.65 (10.13)	172.25 (8.83)	0.7468 [3]
-Median (Q1-Q3)	171.50 (167-178)	172.00 (166-179)	172.00 (167-178)	
-Min-Max	152-193	152-194	152-194	
BMI (kg/m2)				
-N	68	34	102	
-Mean (SD)	25.03 (5.24)	25.45 (5.09)	25.17 (5.17)	0.6142 [4]
-Median (Q1-Q3)	24.27 (21.47-27.00)	24.06 (22.49-29.06)	24.27 (21.56-27.14)	
-Min-Max	16.54-40.93	16.86-37.37	16.54-40.93	

Note:
[1] Overall p-value (2-sided) based on Chi-square test
[2] Overall p-value (2-sided) based on Fisher's exact test
[3] P-value based on Independent t-test
[4] P-value based on Wilcoxon rank sum test

Outcomes

At Day 28 post-vaccination, immunogenicity analysis for anti-PT, anti-FHA antibodies measured by ELISA and PT-neutralizing antibody titers measured by CHO cell assay were evaluated in 101 participants (67 participants were from VacPertagen and 34 from Boostrix vaccine group) for ITT population and 99 participants (65 participants were from VacPertagen and 34 participants from Boostrix).

Seroconversion rates for anti-PT and anti-FHA antibodies measured by ELISA

At 28 days after vaccination, seroconversion rates of anti-PT antibodies were higher in VacPertagen group 98.46% (95% CI: 91.72-99.96) than the seroconversion rates in Boostrix group 82.35% (95% CI: 65.47- 93.24). Seroconversion rates of anti-FHA antibody concentrations were higher in VacPertagen group 87.69% (95% CI: 77.18-94.53) than the seroconversion rates in Boostrix group 61.76% (95% CI: 43.56-77.83)].

Table 62. Seroconversion rates as defined by the proportion of participants with ≥4-fold increase with respect to baseline of anti-PT IgG and anti-FHA IgG concentrations at Day 28 after vaccination in all participants by vaccine groups (Per Protocol population)

Seroconversion rate	Pertagen® (N=65) n (%) (95% CI)	Boostrix® (N=34) n (%) (95% CI)	P-value
PT	64 (98.46) (91.72-99.96)	28 (82.35) (65.47-93.24)	0.0062 [2]*
FHA	57 (87.69) (77.18-94.53)	21 (61.76) (43.56-77.83)	0.0027 [1]*

95% CI: using Clopper-Pearson method.
Note:
[1] Overall p-value (2-sided) based on Chi-square test
[2] Overall p-value (2-sided) based on Fisher's exact test
* P-value ≤ 0.05 is considered statistically significant.

Geometric Mean Concentrations (GMCs) for anti-PT antibody and anti-FHA antibody measured by ELISA

At 28 days after vaccination, anti-PT antibody GMC was higher in VacPertagen [132.70 IU/mL (95% CI: 100.21-175.73)] as compared to Boostrix [57.42 IU/mL (95% CI: 42.01-78.48] vaccine group. At 28 days after vaccination, anti-FHA antibody GMC was numerically higher in VacPertagen [290.65 IU/mL (95% CI: 245.16-344.58)] as compared to Boostrix [237.63 IU/mL (95% CI: 185.15-304.98] vaccine group.

Table 63. Comparison of anti-PT IgG (IU/ml) between baseline and Day 28 after vaccination in all participants by vaccine groups

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
Pertagen®	4.87	132.70	27.27	<0.0001 [2]*
(N=65)	(3.89-6.08)	(100.21-175.73)	(21.65-34.35)	
Boostrix®	5.95	57.42	9.64	<0.0001 [2]*
(N=34)	(4.38-8.10)	(42.01-78.48)	(6.96-13.36)	
P-value[b]	0.2860 [1]	0.0001 [1]*	<0.0001 [1]*	
The ratio of GMC or GMFR between vaccine groups (95% CI)	Pertagen®-Boostrix® 0.82 (0.56-1.19)	Pertagen®-Boostrix® 2.31 (1.48-3.61)	Pertagen®-Boostrix® 2.83 (1.91-4.18)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Independent t-test
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Independent t-test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 64. Comparison of anti-FHA IgG (IU/ml) between baseline and Day 28 after vaccination in all participants by vaccine groups (Per protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
Pertagen®	21.18	290.65	13.72	<0.0001 [2]*
(N=65)	(16.21-27.68)	(245.16-344.58)	(10.40-18.11)	
Boostrix®	33.09	237.63	7.18	<0.0001 [2]*
(N=34)	(24.64-44.44)	(185.15-304.98)	(5.09-10.13)	
P-value[b]	0.0264 [1]*	0.1820 [1]	0.0041 [1]*	
The ratio of GMC or GMFR between vaccine groups (95% CI)	Pertagen®- Boostrix® 0.64 (0.42-0.98)	Pertagen®-Boostrix® 1.22 (0.91-1.64)	Pertagen®-Boostrix® 1.91 (1.22-3.00)	

Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at Day 28 after vaccination to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Independent t-test
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Independent t-test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Geometric Mean Titers (GMTs) or PT neutralization antibody

At 28 days after vaccination, PT neutralizing GMT was higher in VacPertagen [146.97 IU/mL (95% CI: 107.95-200.07)] as compared to Boostrix [69.32 IU/mL (95% CI: 50.24-95.66] vaccine group.

Table 65. Comparison of PT neutralizing GMTs (IU/ml) between baseline and Day 28 after vaccination in all participants by vaccine groups (Per protocol population)

Vaccine	Baseline	Day 28 after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMT (IU/mL) (95% CI)	GMT (IU/mL) (95% CI)	GMFR (95% CI)	
Pertagen®	6.69	146.97	21.96	<0.0001 [2]*
(N=65)	(5.34-8.39)	(107.95-200.07)	(16.97-28.42)	
Boostrix®	8.12	69.32	8.54	<0.0001 [2]*
(N=34)	(6.02-10.95)	(50.24-95.66)	(6.11-11.93)	
P-value[b]	0.3008 [1]	0.0010 [1]*	<0.0001 [1]*	
The ratio of GMT or GMFR between vaccine groups (95% CI)	Pertagen®-Boostrix® 0.82 (0.57-1.20)	Pertagen®-Boostrix® 2.12 (1.31-3.43)	Pertagen®-Boostrix® 2.57 (1.68-3.94)	

Geometric mean fold rise (GMFR) is geometric mean of the ratio of antibody titer at Day 28 after vaccination antibody titer at baseline.
The ratio of GMT or GMFR between vaccine groups (95% CI) based on Independent t-test
a : Compared between baseline and Day 28 after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Independent t-test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Analysis of wP-primed and aP-primed subjects

Seroconversion rates for anti-PT antibody measured by ELISA

Table 66. Seroconversion rates as defined by the proportion of participants with ≥4-fold increase with respect to baseline of anti-PT IgG concentrations at Day 28 after vaccination between vaccine group in wP-primed and aP-primed (Per Protocol population)

Seroconversion rate (PT)	Pertagen®	Boostrix®	Difference in percentage with seroconversion [a]	P-value
	n (%) (95% CI)	n (%) (95% CI)	% (95% CI)	
wP-primed	N = 33	N = 17		
	33 (100.00)	13 (76.47)	23.53	0.0103 [1]*
	(89.42-100.00)	(50.10-93.19)	(9.47-47.51)	
aP-primed	N = 32	N = 17		
	31 (96.88)	15 (88.24)	8.64	0.2731 [1]
	(83.78-99.92)	(63.56-98.54)	(-6.44-31.93)	

95% CI: based on Clopper-Pearson method.
a : The two- sided 95% CI: of the difference in the proportions was obtained based on Miettinen and Nurminen method

Note:
[1] Overall p-value (2-sided) based on Fisher's exact test
* P-value ≤ 0.05 is considered statistically significant.

Antibody responses 1 year after vaccination

Table 67. Comparison of anti-PT IgG concentrations (IU/ml) between baseline and 1 year after vaccination in all participants by vaccine groups (Per protocol population)

Vaccine	Baseline	1 year after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
Pertagen®	4.97	27.61	5.56	<0.0001 [2]*
(N=63)	(3.96-6.24)	(18.80-40.53)	(4.18-7.39)	
Boostrix®	5.90	12.51	2.12	<0.0001 [2]*
(N=33)	(4.30-8.10)	(8.39-18.64)	(1.63-2.76)	
P-value[b]	0.3775 [1]	0.0099 [1]*	<0.0001 [1]*	
The ratio of GMC or GMFR between vaccine groups (95% CI)	Pertagen® - Boostrix® 0.84 (0.57-1.24)	Pertagen® - Boostrix® 2.21 (1.21-4.01)	Pertagen® - Boostrix® 2.62 (1.79-3.85)	

* Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 1 year after vaccination to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Independent t-test
a : Compared between baseline and 1 year after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Independent t-test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Table 68. Comparison of anti-FHA IgG concentrations (IU/ml) between baseline and 1 year after vaccination in all participants by vaccine groups (Per protocol population)

Vaccine	Baseline	1 year after vaccination	Geometric Mean Fold Rise from baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMFR (95% CI)	
Pertagen®	21.72	79.77	3.67	<0.0001 [2]*
(N=63)	(16.52-28.56)	(65.57-97.06)	(2.88-4.69)	
Boostrix®	33.29	92.65	2.78	<0.0001 [2]*
(N=33)	(24.57-45.13)	(70.14-122.39)	(2.08-3.72)	
P-value[b]	0.0530 [1]	0.3747 [1]	0.1642 [1]	
The ratio of GMC or GMFR between vaccine groups (95% CI)	Pertagen® - Boostrix® 0.65 (0.42-1.01)	Pertagen® - Boostrix® 0.86 (0.62-1.20)	Pertagen® - Boostrix® 1.32 (0.89-1.95)	

* Geometric mean fold rise (GMFR) is geometric mean of the ratios of antibody concentration at 1 year after vaccination to antibody concentration at baseline
The ratio of GMC or GMFR between vaccine groups (95% CI) based on Independent t-test
a : Compared between baseline and 1 year after vaccination
b : Compared between vaccine groups

Note:
[1] P-value based on Independent t-test
[2] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Study TDA203, A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and safety of a single dose of BioNet-Asia's acellular pertussis-only vaccine and its combined tetanus-diphtheria-acellular pertussis vaccine at multiple dose

levels or Boostagen in comparison to Boostrix, when administered to healthy women of childbearing age

Methods

This is a phase II, dual-site, observer-blind, randomized, active-controlled vaccine trial in which 250 healthy non-pregnant women of childbearing age were recruited from 2 sites in Bangkok, Thailand with 125 women enrolled at each site. Women who were found to be eligible were randomized equally (in a 1:1:1:1:1 ratio) into the following treatment arms:

Table 69. Vaccine groups

Treatment Arm*	*N for Site 1*	*N for Site 2*	*Total N*
Boostagen®	25	25	50
BioNet Tdap-2,5	25	25	50
BioNet Tdap-1,1	25	25	50
Boostrix™	25	25	50
BioNet ap-1,1	25	25	50
Total	**125**	**125**	**250**

*Boostagen®, BioNet Tdap and ap contained 2 pertussis antigens: PTgen and FHA.

Treatments

Each participant received 0.5 mL of a single intramuscular injection of the assigned vaccine, provided in a prefilled syringe. Composition of study vaccines is reported in Table 75 and 76.

Table 70. Composition of BioNet's investigational vaccines

Name of ingredients per 0.5-mL dose	Group 1 **BioNet ap-1,1**	Group 2 **BioNet Tdap-1,1**	Group 3 **BioNet Tdap-2,5**
Active ingredients			
Tetanus Toxoid (TT)	-	7.5 Lf	7.5 Lf
Diphtheria Toxoid (DT)	-	2.0 Lf	2.0 Lf
Pertussis Toxoid (PT)[a]	1 µg	1 µg	2 µg
Filamentous hemagglutinin (FHA)	1 µg	1 µg	5 µg
Pertactin (PRN)	-	-	-
Excipients			
Adjuvant	Aluminum Hydroxide 0.3 mg Al^{3+}	Aluminum Hydroxide 0.3 mg Al^{3+}	Aluminum Hydroxide 0.3 mg Al^{3+}
NaCl mg/dose	4.38	4.38	4.38
Water for Injection	q.s. to 0.5 mL	q.s. to 0.5 mL	q.s. to 0.5 mL

[a] Genetically-detoxified PT (PTgen)

Table 71. Composition of references vaccines

Name of ingredients per 0.5-mL dose	Group 4 **Boostagen®**	Group 5 **Boostrix™**
Active ingredients		
Tetanus Toxoid (TT)	7.5 Lf	5 Lf
Diphtheria Toxoid (DT)	2.0 Lf	2.5 Lf
Pertussis Toxoid (PT)	5 µg[a]	8 µg[b]
Filamentous hemagglutinin (FHA)	5 µg	8 µg
Pertactin (PRN)	-	2.5 µg
Excipients		
Adjuvant	Aluminum Hydroxide 0.3 mg Al^{3+}	Aluminum Phosphate 0.2 mg Al^{3+} Aluminum Hydroxide 0.3 mg Al^{3+}
NaCl mg/dose	4.38	4.50
Water for Injection	q.s. to 0.5 mL	q.s. to 0.5 mL

[a] Genetically-detoxified PT (PTgen)
[b] Chemically-detoxified PT

To evaluate immune responses to the study vaccines, blood samples (approximately 5 mL) were taken from all participants at Day 0 (Visit 1; baseline) just before vaccination and at Day 28 (Visit 2) after vaccination. At Day 28 visit after all study procedures had been performed and blood had been collected, participants in the BioNet ap arm were offered a dose of commercially available Td vaccine.

Objectives/endpoints

Primary Endpoint

Percentage of subjects with seroresponse* in terms of anti-PT ELISA antibody at 28 days after vaccination as compared to baseline

*Seroresponse is defined by the following criteria:

- For seronegative subjects at baseline (<5 IU/mL), post-vaccination anti-PT ELISA antibody concentrations ≥20 IU/mL;
- For seropositive subjects at baseline with pre-vaccination antibody concentrations ≥5 IU/mL and <20 IU/mL, an increase of at least 4 times the pre-vaccination antibody concentration;
- For seropositive subjects with pre-vaccination antibody concentrations ≥20 IU/mL, an increase of at least 2 times the pre-vaccination antibody concentration

Remark: Vaccine formulations that reach a seroresponse rate at 50% or more of the lower limit of the 95% CI for anti-PT antibody at 28 days after vaccination will advance to a subsequent trial in pregnant women.

(Selected) Secondary Endpoints

Percentage of subjects with seroresponse* (as defined above) for anti-FHA ELISA antibody at 28 days after vaccination as compared to baseline

Geometric mean of anti-PT, anti-FHA, anti-tetanus, and anti-diphtheria ELISA antibody concentrations at baseline and at 28 days after vaccination

This study was to be conducted before proceeding to the study on maternal immunisation. Vaccine formulations that achieved a good safety profile and a pre-defined immunogenicity level were to be selected to proceed to the study in pregnant women.

Sample size

This trial is a proof-of-concept phase II study to evaluate the safety and immunogenicity of BioNet's vaccines in healthy nonpregnant women. The sample size of this study was based on clinical and practical considerations.

Randomisation and blinding (masking)

The trial was carried out in an observer-blind manner for all vaccine groups until the end of the study, except for BioNet ap group. The subjects assigned to receive BioNet ap-1,1 vaccine were unblinded at end of Visit 2 (Day 28) and were offered with one dose of commercially available Td vaccine.

A randomization list containing subject numbers and masked vaccine group assignments (the vaccines were masked as WOCBA-1, WOCBA-2, WOCBA-3, WOCBA-4 and WOCBA-5) was provided to the unblind team. The study pharmacist and vaccine administrator were unblinded to treatment assignment; all other investigators and the study participants were blinded to maintain observer-blind status.

Statistical methods

Enrolled Population includes all screened subjects who provided informed consent and received a Subject Number, regardless of the subject's randomization and treatment status in the study.

Full Analysis (FA) Population includes the subjects in the enrolled population who were randomized, received a study vaccination, and provided at least one evaluable serum sample. The analysis based on this population served as supportive results for all immunogenicity objectives.

Subjects in the FA population were analysed "as randomised", i.e. according to the vaccine a subject was designated to receive, which may be different from the vaccine the subject actually received.

The "Per Protocol" (PP) Population includes the subjects in the FA population who correctly received study vaccine per randomization with no major protocol deviations that were determined to potentially interfere with the immunogenicity assessment of the study vaccines. This population served as the primary analysis population for all immunogenicity objectives.

Because of the unpredictability of some irregularities, the criteria for exclusion of subjects from the PP population would be determined based on a blind review of the data before the database was locked. The precise reasons for excluding subjects from a PP analysis would be documented before unblinding.

PP population was used for immunogenicity analyses.

Due to the concept approval nature of this study in terms of determining which formulations of BioNet's vaccines can be further evaluated in pregnant women, all analyses for immunogenicity and safety endpoints were **descriptive** and no multiplicity adjustment was carried out.

The primary immunogenicity endpoint is percentage of subjects with seroresponse based on PT-IgG antibody at 28 days after vaccination as compared to baseline. In order to select the formulations of BioNet's vaccines that can be further evaluated in pregnant women, fifty percent (50%) or more of the lower limit of the 95% confidence interval (CI) of the seroresponse rate was used as a cut-off criterion.

Percentages of subjects with a positive seroresponse for PT-IgG antibody, subjects with a positive seroresponse for FHA-IgG antibody, and percentages of subjects with a seroconversion ($\geq$ 2-fold and $\geq$ 4-fold increase with respect to baseline) for PT-IgG and FHA-IgG antibody concentrations at 28 days following immunisation as compared to baseline were computed for each vaccine group along with its corresponding exact two-sided 95% CI based on Clopper-Pearson method.

Results

Participant flow

No one was excluded from data analysis. FA population was the same as PP analysis population for this study.

Recruitment

The study was initiated on 4 July 2018 and completed on 24 January 2019.

Conduct of the study

During the conduct of this study, no protocol deviation was reported.

Baseline data

Demographic characteristics at baseline were similar across all vaccine groups (Table 77). The mean age of participants at enrollment was 30.47 (±5.51) years. All participants were Asian.

Table 72. Summary of demographic at baseline (Full analysis population)

Subject status	BioNet ap-1,1	BioNet Tdap-1,1	BioNet Tdap-2,5	Boostagen®	Boostrix™	Total	P-value
Demographics at baseline							
Age (years)							0.1943 [2]
- N	50	50	50	50	50	250	
- Mean (SD)	29.74 (5.32)	29.48 (6.32)	30.88 (4.72)	30.28 (5.74)	31.98 (5.18)	30.47 (5.51)	
- Median	30.00	29.00	31.00	31.00	33.00	31.00	
- Min/Max	18 - 39	18 - 39	22 - 39	18 - 39	19 - 39	18 - 39	
Ethnicity: n (%)							_ [1]
- Asian	50 (100.00)	50 (100.00)	50 (100.00)	50 (100.00)	50 (100.00)	250 (100.00)	
- Other	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Height (cm)							0.6096 [3]
- N	50	50	50	50	50	250	
- Mean (SD)	158.5 (5.60)	158.5 (4.49)	157.9 (5.72)	157.7 (5.22)	159.3 (5.51)	158.4 (5.31)	
- Median	158.0	158.5	158.0	158.0	160.0	158.0	
- Min/Max	147 - 173	148 - 170	144 - 174	145 - 167	145 - 175	144 - 175	
Weight (kg)							0.1256 [2]
- N	50	50	50	50	50	250	
- Mean (SD)	57.65 (13.45)	60.10 (13.74)	57.92 (13.11)	62.15 (16.38)	63.76 (14.50)	60.32 (14.37)	
- Median	53.35	57.20	55.60	57.35	60.50	57.20	
- Min/Max	39.3 - 96.1	41.2 - 101.1	38.8 - 95.9	40.8 - 111.6	42.6 - 101.1	38.8 - 111.6	

Note:
[1] No p-value was computed by SAS, [2] P-value based on Kruskal-Wallis Test, [3] P-value based on one-way ANOVA

Numbers analysed

A total of 257 non-pregnant women were screened, of whom 250 (125 from each study site) were enrolled and randomized into 5 vaccine groups (50 participants per each treatment group). The main reason for screening failures was having acute or chronic, clinically significant psychiatric, hematologic, pulmonary, cardiovascular, or hepatic or renal functional abnormality as determined by the Investigator based on medical history and physical examination (Exclusion Criteria No. 1). All enrolled

participants were included in the safety and immunogenicity analyses. No one was excluded from data analysis. FA population was the same as PP analysis population for this study.

Outcomes and estimation

Seroresponse in anti-PT and anti-FHA antibodies

For all tested vaccines, the lower limit of the 95% CI of the PT-IgG seroresponse rate exceeded 50%. Accordingly, all of the tested vaccines had passed the pre-defined cut-off criteria as outlined in the primary objective and can progress to the subsequent trial for maternal immunisation.

Table 73. TDA203 study - Proportion of subjects with a seroresponse for PT-IgG and FHA-IgG antibodies at 28 days after vaccination compared to baseline as assessed by ELISA in all evaluable subjects by vaccine groups

Seroresponse Rate	BioNet ap-1,1 (N=50) n (%) (95% CI)	BioNet Tdap-1,1 (N=50) n (%) (95% CI)	BioNet Tdap-2,5 (N=50) n (%) (95% CI)	Boostagen® (N=50) n (%) (95% CI)	Boostrix™ (N=50) n (%) (95% CI)	P-value
PT	46 (92.00) (80.77-97.78)	44 (88.00) (75.69-95.47)	40 (80.00) (66.28-89.97)	47 (94.00) (83.45-98.75)	39 (78.00) (64.04-88.47)	0.0740 [1]
FHA	45 (90.00) (78.19-96.67)	46 (92.00) (80.77-97.78)	50 (100.00) (92.89-100.00)	48 (96.00) (86.29-99.51)	48 (96.00) (86.29-99.51)	0.1636 [1]

a: Seroresponse

- For seronegative subjects at baseline (< 5 IU/mL), post-vaccination antibody concentrations ≥ 20 IU/mL;
- For seropositive subjects at baseline with pre-vaccination antibody concentrations ≥ 5 IU/mL and < 20 IU/mL, an increase of at least 4 times the pre-vaccination antibody concentrations;
- For seropositive subjects with pre-vaccination antibody concentrations ≥ 20 IU/mL, an increase of at least 2 times the pre-vaccination antibody concentrations.

95% CI based on Clopper-Pearson method
Note:
[1] Overall p-value (2-sided) based on Fisher's exact test

GMCs of anti-PT and anti-FHA antibodies

Table 74. TDA203 study - Geometric mean concentrations (GMCs) of PT-IgG and FHA-IgG antibodies at baseline and 28 days after vaccination as assessed by ELISA in all evaluable subjects by vaccine groups

Vaccine	PT				FHA			
	Baseline	Day 28 after vaccination	Geometric Mean of Fold Change from Baseline	P-value[a]	Baseline	Day 28 after vaccination	Geometric Mean of Fold Change from Baseline	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMC Ratio (95% CI)		GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMC Ratio (95% CI)	
BioNet ap-1,1 (N=50)	9.06 (6.60-12.45)	155.87 (118.38-205.24)	17.20 (11.87-24.90)	<0.0001 [3]*	13.41 (10.31-17.46)	128.55 (107.91-153.13)	9.58 (7.19-12.77)	<0.0001 [3]*
BioNet Tdap-1,1 (N=50)	5.78 (4.42-7.55)	58.26 (43.92-77.28)	10.08 (7.42-13.69)	<0.0001 [3]*	9.80 (7.01-13.69)	74.74 (61.06-91.49)	7.63 (6.01-9.69)	<0.0001 [3]*
BioNet Tdap-2,5 (N=50)	4.56 (3.61-5.74)	60.37 (42.91-84.94)	13.25 (9.77-17.98)	<0.0001 [3]*	7.94 (5.65-11.17)	123.50 (99.91-152.65)	15.55 (11.20-21.58)	<0.0001 [3]*
Boostagen® (N=50)	5.95 (4.31-8.21)	146.92 (106.79-202.12)	24.69 (18.23-33.44)	<0.0001 [3]*	8.95 (6.47-12.38)	198.32 (152.72-257.54)	22.15 (15.66-31.33)	<0.0001 [3]*
Boostrix™ (N=50)	6.51 (4.64-9.15)	64.75 (49.76-84.27)	9.94 (7.38-13.40)	<0.0001 [3]*	11.95 (8.84-16.17)	293.47 (235.41-365.86)	24.55 (16.66-36.18)	<0.0001 [3]*
P-value[b]	0.0321 [1]*	<0.0001 [1]*	0.0001 [2]*		0.1197 [2]	<0.0001 [2]*	<0.0001 [1]*	
Ratio of GMCs between BioNet vaccines and Boostrix™ (95% CI)	BioNet ap-1,1 - Boostrix™ 1.39 (0.77-2.52)	BioNet ap-1,1 - Boostrix™ 2.41 (1.33-4.36)	BioNet ap-1,1 - Boostrix™ 1.73 (0.92-3.26)			BioNet ap-1,1 - Boostrix™ 0.44 (0.28-0.67)	BioNet ap-1,1 - Boostrix™ 0.39 (0.21-0.74)	
	BioNet Tdap-1,1 - Boostrix™ 0.89 (0.49-1.61)	BioNet Tdap-1,1 - Boostrix™ 0.90 (0.50-1.63)	BioNet Tdap-1,1 - Boostrix™ 1.01 (0.54-1.91)			BioNet Tdap-1,1 - Boostrix™ 0.25 (0.17-0.39)	BioNet Tdap-1,1 - Boostrix™ 0.31 (0.16-0.59)	
	BioNet Tdap-2,5 - Boostrix™ 0.70 (0.39-1.27)	BioNet Tdap-2,5 - Boostrix™ 0.93 (0.51-1.69)	BioNet Tdap-2,5 - Boostrix™ 1.33 (0.71-2.51)			BioNet Tdap-2,5 - Boostrix™ 0.42 (0.27-0.65)	BioNet Tdap-2,5 - Boostrix™ 0.63 (0.33-1.20)	
	Boostagen® - Boostrix™ 0.91 (0.50-1.66)	Boostagen® - Boostrix™ 2.27 (1.25-4.11)	Boostagen® - Boostrix™ 2.48 (1.32-4.68)			Boostagen® - Boostrix™ 0.68 (0.44-1.04)	Boostagen® - Boostrix™ 0.90 (0.47-1.71)	

a: Compared between baseline and Day 28 post-vaccination; b: Compared between vaccine groups
Vaccine difference (95% CI) based on Bonferroni post-hoc analysis
[1] P-value based on Kruskal-Wallis Test; [2] P-value based on one-way ANOVA; [3] P-value based on paired t-test
* P-value ≤ 0.05 is considered statistically significant.

Pregnant subjects

Study TDA204, A phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and safety of a single dose of BioNet-Asia's acellular pertussis-only vaccine and its combined tetanus-diphtheria-acellular pertussis vaccine at multiple dose levels or Boostagen in comparison to Boostrix, when administered to healthy pregnant women

Methods

This is a phase II, dual-site, observer-blind, randomized, active-controlled vaccine trial in which healthy women with singleton uncomplicated pregnancy were recruited from 2 sites in Bangkok, Thailand. Participants were randomly and equally (1:1:1:1:1 ratio) allocated into the following study groups:

Table 75. Vaccine groups (N, number of subjects)

Treatment group*	N for Site 1	N for Site 2	Total, N
Boostagen®	40	40	80
BioNet Tdap-2,5	40	40	80
BioNet Tdap-1,1	40	40	80
Boostrix™	40	40	80
BioNet ap-1,1	40	40	80
Total	**200**	**200**	**400**

* BioNet Tdap and BioNet ap contains 2 pertussis antigens: PTgen and FHA (e.g., 2 µg for PT and 5 µg for FHA for Tdap-2,5). Boostagen® contains 5 µg of PT and 5 µg of FHA and Boostrix™ contains 8 µg of PT and 5 µg of FHA.

TDA204 was a study that followed the phase II (TDA203) clinical study. It aimed to primarily evaluate the immunogenicity in terms of anti-PT responses of different dosages of PTgen contained in different formulations of BioNet ap, Tdap, and Boostagen in comparison with Boostrix, in healthy pregnant women. Similarly to TDA203 study, the immunogenicity evaluation is based on immune response of PT-IgG antibody assessed by ELISA in pregnant women. Immune response of FHA-, DT-, and TT-IgG antibodies was evaluated as part of the secondary endpoints. A subset (30%) of maternal and infant subjects are also tested by CHO cell assay for PT-neutralizing serum antibody.

Study Participants

The study population included pregnant woman 18-40 years of age. Only those subjects who fulfilled all of the inclusion criteria and none of the exclusion criteria were randomized.

Treatments

Participants randomized into BioNet vaccine groups received different dose levels of recombinant acellular pertussis vaccine containing varied PT_{gen} and FHA antigen contents (Table 76):

Table 76. Composition of BioNet Investigational vaccines and Boostagen, and comparator Boostrix licensed vaccine

Name of ingredients (per 0.5-mL dose)	Group 1 BioNet ap-1,1	Group 2 BioNet Tdap-1,1	Group 3 BioNet Tdap-2,5	Group 4 Boostagen®	Group 5 Boostrix™ (comparator)
Active ingredients					
Tetanus Toxoid (TT)	-	7.5 Lf	7.5 Lf	7.5 Lf	5 Lf
Diphtheria toxoid (DT)	-	2.0 Lf	2.0 Lf	2.0 Lf	2.5 Lf
Pertussis Toxoid (PT)	1 µg[a]	1 µg[a]	2 µg[a]	5 µg[a]	8 µg[b]
Filamentous hemagglutinin (FHA)	1 µg	1 µg	5 µg	5 µg	8 µg
Pertactin (PRN)	-	-	-	-	2.5 µg
Excipients					
Adjuvant	$Al(OH)_3$: 0.3 mg Al^{3+}				$AlPO_4$: 0.2 mg Al^{3+} $Al(OH)_3$: 0.3 mg Al^{3+}
NaCl mg	4.38 mg				4.50 mg
Water for Injection	q.s. to 0.5 mL				q.s. to 0.5 mL
Batch					
Number	8005A	8006A	8004A	7007A1A	AC37B293CI

µg: microgram; Al^{3+}: Aluminum ion; $Al(OH)_3$: Aluminum hydroxide; $AlPO_4$: aluminum phosphate; aP: acellular pertussis vaccine; ap: acellular pertussis (reduced dose of antigens); d: diphtheria (reduced dose of antigens); Lf: limit of flocculation; mL: milliliter; mg: milligram; NaCl: sodium chloride; q.s.: *quantum satis*; T: tetanus.

[a] genetically detoxified PT (PTgen); [b] chemically-detoxified PT.

Objectives/endpoints

Primary endpoint

In maternal subjects

- Geometric mean of PT-specific serum antibody concentrations (GMCs) measured 28 days after vaccine injection in maternal subjects as well as women of childbearing age*, as determined by ELISA.

* Data from women of childbearing age have been obtained during TDA203 clinical study.

The following non-inferiority hypothesis was tested with a significance level of 0.00625 for the primary objective,

The null hypothesis, H01: $GMC_S/GMC_R \leq 0.5$
The alternative hypothesis, H11: $GMC_S/GMC_R > 0.5$

where R = reference group (Boostrix™ group)
and S = study group (1 of BioNet Tdap/Boostagen® groups or BioNet ap group).

Selected secondary endpoints

In maternal subjects

- Geometric mean of anti-PT ELISA antibody concentrations measured at baseline and at the time of delivery.
- Geometric mean of anti-FHA ELISA antibody concentrations measured at baseline, 28 days after vaccine injection, and at delivery.
- Percentages of maternal subjects with a 4-fold or higher response in anti-PT and anti-FHA ELISA antibody concentrations 28 days after vaccine injection and at the time of delivery, in reference to baseline.
- Geometric mean of PT-neutralizing antibody titer (GMT) measured at baseline, 28 days after vaccine injection, and at delivery, as determined by CHO cell assay.
- Percentage of maternal subjects with a 4-fold or higher response in PT-neutralizing antibody titers measured at 28 days after vaccine injection and at the time of delivery, in reference to baseline, as determined by CHO cell assay.

In infant subjects

- Geometric means of anti-PT and anti-FHA ELISA antibody concentrations measured at the time of birth (cord blood or neonatal blood within 72 hours after birth) and at 2 months of age.
- Geometric mean of PT-neutralizing antibody titers in infant subjects measured at the time of birth (cord blood or neonatal blood within 72 hours after birth) and at 2 months of age.

Sample size

This is a phase II randomized, observer-blind, active-controlled dose ranging study to evaluate the safety and immunogenicity of BioNet vaccine groups relative to Boostrix group in pregnant women. A total of 400 pregnant women were randomized (80 in each group). To assess the primary objective, the immunogenicity data collected in the present study on pregnant women were combined with the data obtained in the TDA203 study on women of childbearing age to determine which formulation of the BioNet vaccines was comparable to Boostrix vaccine.

With 80 enrolled pregnant women per group in this study and 50 women of childbearing age per group in the TDA203 study, there were 130 women per group in the pooled population. The power to show that the ratio of anti-PT GMC in one of the BioNet Tdap groups/Boostagen group/BioNet ap group to Boostrix group at 28 day after vaccination was at least 0.5 was calculated using a 1-sided 2-sample t-test with a significance level of 0.00625, based on assumed different SDs of log10-transformed concentrations of anti-PT antibodies measured by ELISA and number of evaluable subjects per group. Assuming the actual SD of log10 anti-PT ELISA GMC is 0.6, with 117 evaluable subjects per group in the pooled population and a significance level of 0.00625, the study would have a 91% power to detect at least a 0.5-fold anti-PT GMC in BioNet Tdap/ap/Boostagen vs Boostrix group.

Randomisation and blinding (masking)

A randomization list containing subject numbers and masked vaccine group assignments was provided to the unblinded team at the study sites.

Three additional randomization lists were provided to the investigator to select at the study start per vaccine group:

1. 24 mother-infant pairs whose samples were (mother) or will be (infants) tested for PTneutralizing antibody in CHO cell assay.

2. 40 infants per vaccine group whose blood will be taken at 5 months of age (Visit 6), and

3. the other 40 infants per vaccine group whose blood will be taken at 7 months of age (Visit 8).

Then each eligible consented maternal subject received a single intramuscular (IM) injection of the assigned vaccine, which was provided in a monodose prefilled syringe containing 0.5 milliliter (mL) volume.

The study pharmacist and vaccine administration personnel were unblinded to treatment assignment; all other investigators, study staff, and the study maternal subjects were blinded to treatment assignment to maintain observer-blind status. Maternal subjects assigned to receive BioNet ap were unblinded on Day 28 after vaccination, in order to receive one dose of Tetanus-low dose diphtheria (Td) vaccine soon after the second blood collection.

Statistical methods

Analysis populations

Maternal subjects

Enrolled population includes all screened maternal subjects who provided informed consent and received a Subject Number, regardless of the subject's randomization and treatment status in the study.

Full Analysis (FA) population includes the subjects in the enrolled population who were randomized, received a study vaccination, and provide evaluable serum sample at least at one time point postvaccine injection. The analysis based on this population will serve as supportive results for all secondary immunogenicity objectives pertinent to maternal subjects. Subject in the FA population is analysed "as randomised", i.e., according to the vaccine that the subject was designated to receive, which may be different from the vaccine that the subject received.

The "Per Protocol" (PP) population includes the subjects in the FA population who correctly received study vaccine per randomization with no major protocol deviations that could interfere with the immunogenicity assessment of the study vaccines. This population serves as the primary analysis population for all immunogenicity objectives associated with maternal subjects.

For primary objective, FA populations from the present and the TDA203 studies and PP populations from the present and TDA203 studies were pooled together.

Infant subjects

FA population will include the infants whose mothers are included in the FA population for maternal subjects. The analysis based on this population will serve as supportive results for all secondary immunogenicity objectives pertinent to infant subjects.

PP population will include the infants whose mothers are included in the PP population for maternal subjects and who have no major protocol deviations that are determined to potentially interfere with

the immunogenicity assessment of the study vaccines. This population will serve as the primary analysis population for all immunogenicity objectives associated with infant subjects.

Statistical and Analytical Plans

Statistical analyses have been divided into 3 parts. An analysis including all safety and immunogenicity data collected in maternal subjects up to 28 days following immunisation and associated primary and secondary objectives was performed first on cleaned and locked data. Immunogenicity and safety data were reported on group-level only. Individual listings were generated without information on the subject's study group. Access to subject-level information about study groups was restricted.

Two subsequent analyses will be performed as follows:

1. The first subsequent analysis including the rest of safety and immunogenicity data collected in maternal subjects 28 days after vaccine injection to time of delivery and in infants at time of birth and associated secondary and exploratory objectives will be conducted on cleaned and locked data after all maternal subjects will have completed delivery visit (Visit 3).

2. The second subsequent analysis will be conducted on cleaned and locked data including the rest of safety and immunogenicity data collected from infant subjects after birth to 13 months of age, the safety data collected from maternal subjects between the delivery visit and 2 months postpartum visit (Visit 4) and the antibody persistence data collected from a subset of maternal subjects at 1 year after vaccination and associated secondary and exploratory objectives.

The results of these 2 subsequent analyses were presented in an addendum to the CSR. Individual data listings with information on the subject's study group were generated after full unblinding.

The details of the statistical and analytical plan to address the study objectives were given in the SAP produced and finalized before the database lock. All statistical analyses were performed using a statistical analysis software (SAS® software version 9.4). The 95% CI was provided for estimates, as appropriate.

Primary immunogenicity objective

The immunogenicity data collected in the present study on pregnant women were combined with the data obtained in the TDA203 study on women of childbearing age to determine which formulation of the BioNet vaccines was comparable to Boostrix vaccine. No statistical inference was made based on the immunogenicity data obtained in the TDA 203 study on women of childbearing age. The same laboratory that conducted immunogenicity assays for the TDA 203 study performed the immunogenicity testing for the present study, which provided justifiability of combining the immunogenicity data from both studies.

GMC of anti-PT antibodies at 28 days after vaccine injection in the pooled population of maternal subjects and women of childbearing age, as measured by ELISA, was calculated for each vaccine group along with its 2-sided 95% CI, by exponentiating the corresponding log10-transformed mean and its 95% CI limits.

The ratio of the GMC in each of BioNet Tdap/BioNet ap/Boostagen groups to that in Boostrix group and a 2-sided 98.75% CI of the ratio was provided. The log10-transformed concentrations were used to construct a mean difference between the 2 vaccine groups and its 2-sided 98.75% CI using analysis of covariance (ANCOVA). Log10-transformed baseline concentrations and an indicator of study populations were included as covariates. In addition, other possible variables (including age, study site, and gestational age) were evaluated for inclusion in the ANCOVA model as covariates. Interaction terms among selected covariates were evaluated for inclusion as well. The mean difference and corresponding 98.75% CI limits were exponentiated to obtain the GMC ratio and the corresponding

98.75% CI. If the lower confidence limit of the 98.75% CI of the GMC ratio (GMCS/GMCR) was larger than 0.5 which is a non-inferiority margin, the corresponding BioNet vaccine was comparable to Boostrix vaccine. If the 98.75% CI for the ratio the GMC in BioNet vaccine group to that in Boostrix group not only lay entirely above 0.5 but also above 1, there was evidence of superiority in terms of statistical significance at the 1.25% level ($p<0.0125$) according to EMA guidelines (CPMP/EWP/482/99. London, 27 July 2000).

Results

Participant flow

Figure 17. Maternal subjects- Overall disposition at Visit 4 (end of study visit for maternal subjects)

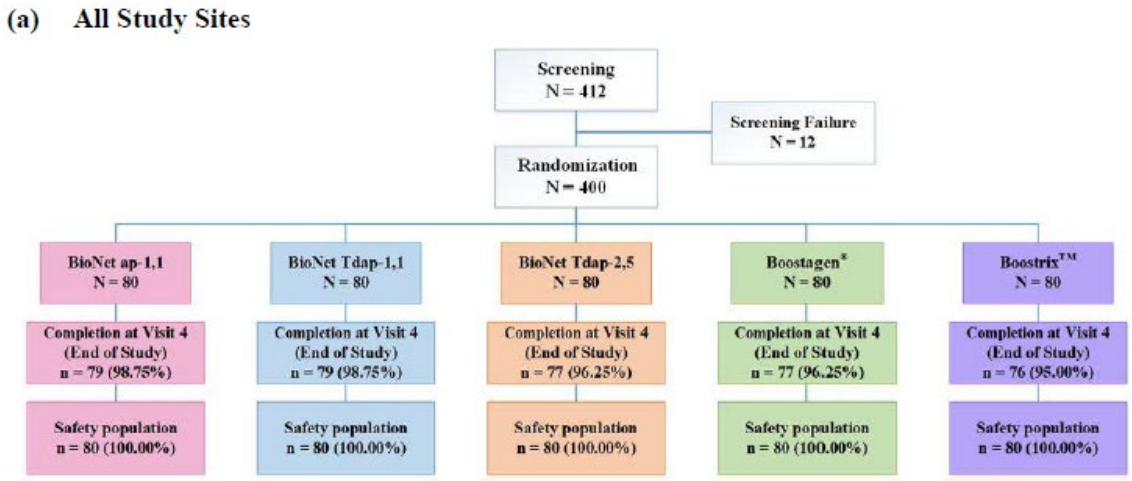
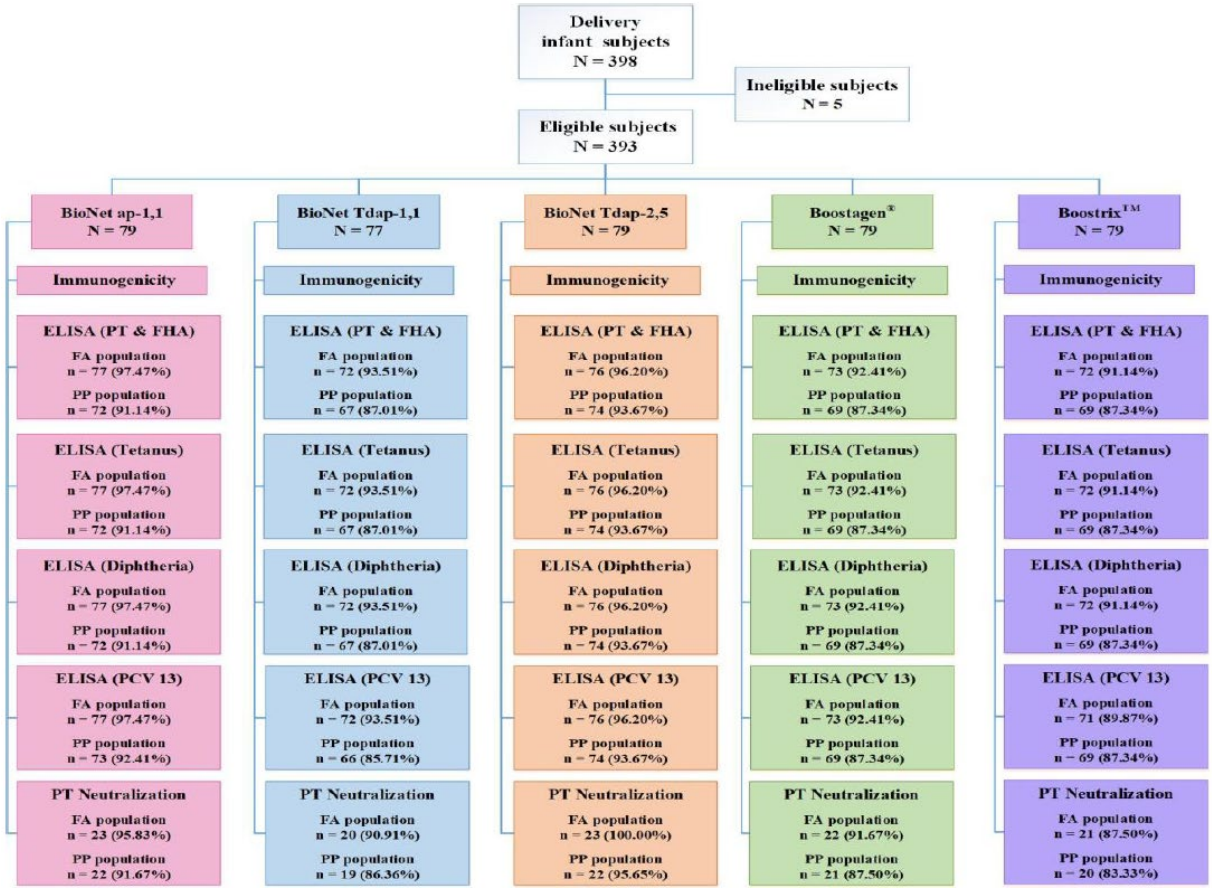


Figure 18. Infant subjects- Overall disposition of subjects at 13 months of age



Recruitment

The first maternal participant visit was in January 2019 and the end of study visit (Visit 10) of the last infant participant was in March 2021.

Conduct of the study

Changes to Planned Analyses

Several statistical analyses, not planned in the SAP, have been carried out. Since this study is only considered supportive for this application, this issue is not pursued.

Baseline data

Maternal Subjects

Maternal subjects were all Asian; their median age at baseline was 30 years (Table 77).

Table 77. Maternal subjects- Summary of demographics at Visit 0 (screening)- Full analysis population

Subject status	BioNet ap-1,1	BioNet Tdap-1,1	BioNet Tdap-2,5	Boostagen®	Boostrix™	Total	P-value
Age (years)							
N	80	80	80	80	80	400	
Mean (SD)	30.55 (4.72)	29.73 (5.12)	29.59 (5.98)	29.14 (5.68)	29.76 (5.15)	29.75 (5.34)	0.6976 [2]
Median	31.00	29.00	30.00	30.00	30.00	30.00	
Min-Max	19-39	18-39	18-39	19-39	19-39	18-39	
Ethnicity: n (%)							
Asian	80 (100.00)	80 (100.00)	80 (100.00)	80 (100.00)	80 (100.00)	400 (100.00)	- [1]
Other	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Height (cm)							
N	80	80	80	80	80	400	
Mean (SD)	158.3 (5.56)	159.5 (5.61)	158.8 (5.27)	159.4 (5.67)	160.0 (5.98)	159.2 (5.63)	0.3335 [2]
Median	158.0	160.0	158.0	159.5	160.0	159.5	
Min-Max	145-170	142-173	150-175	147-172	145/173	142-175	
Weight (kg)							
N	80	80	80	80	80	400	
Mean (SD)	62.13 (10.89)	64.97 (10.68)	64.55 (11.97)	65.13 (12.46)	66.71 (12.26)	64.70 (11.71)	0.1719 [3]
Median	62.10	64.70	62.75	62.80	65.15	63.60	
Min-Max	40.3-90.5	45.4-94.9	41.8-101.4	37.7-101.9	45.5-113.5	37.7-113.5	

cm: centimeters; kg: kilogram; Max: maximum; Min: minimum; SD: standard deviation.
[1] No p-value was computed by S.A.S.
[2] P-value based on Kruskal-Wallis Test.
[3] P-value based on 1-way ANOVA.

Infant Subjects

Approximately half of the infants were male (50.64%). Their birthweight ranged from 2.020 kg and 4.455 kg, with a median of 3.060 kg. Head circumferences (cm) ranged between 30 and 38, with a median of 33.6 cm (Table 78).

Table 78. Infant subjects- Summary of demographic characteristics at Visit 3 (birth)- Safety population

Subject status	BioNet ap-1,1	BioNet Tdap-1,1	BioNet Tdap-2,5	Boostagen®	Boostrix™	Total
Sex: n (%)						
N	79	77	79	79	79	393
Male	40 (50.63)	36 (46.75)	41 (51.90)	41 (51.90)	41 (51.90)	199 (50.64)
Female	39 (49.37)	41 (53.25)	38 (48.10)	38 (48.10)	38 (48.10)	194 (49.36)
Birth weight (gram)						
N	79	77	79	79	79	393
Mean (SD)	3138 (333.4)	3046 (397.4)	3071 (377.9)	3048 (450.6)	3091 (281.0)	3079 (372.1)
Median	3130	3000	3050	3010	3105	3060
Min - Max	2292 - 3870	2140 - 3940	2294 - 4430	2020 - 4455	2370 - 3940	2020 - 4455
Length (cm)						
N	79	77	79	79	79	393
Mean (SD)	49.70 (1.70)	49.15 (2.21)	49.61 (1.67)	49.39 (1.90)	49.92 (1.52)	49.56 (1.83)
Median	50.00	49	49.50	49.50	50.00	49.50
Min - Max	46 - 5305	42 - 54	46 - 54	45 - 53	46 - 54	42 - 54
Head circumference (cm)						
N	79	77	79	79	79	393
Mean (SD)	33.83 (1.15)	33.54 (1.30)	33.71 (1.26)	33.51 (1.41)	33.83 (1.02)	33.69 (1.24)
Median	34.00	33.50	33.50	33.50	34.00	33.60
Min - Max	30.5 - 36	30 – 36.5	31 - 38	30.5 - 37	30 - 36	30 - 38
Gestational size: n (%)						
N	79	77	79	79	79	393
SGA1	1 (1.27)	2 (2.60)	1 (1.27)	3 (3.80)	1 (1.27)	8 (2.04)
AGA2	76 (96.20)	74 (96.10)	74 (93.67)	72 (91.14)	77 (97.47)	373 (94.91)
LGA3	2 (2.53)	1 (1.30)	4 (5.06)	4 (5.06)	1 (1.27)	12 (3.05)

AGA2: appropriate (size) for gestational age; cm: centimeters; kg: kilogram; LGA3: large (size) for gestational age;
Max: maximum; Min: minimum; SD: standard deviation; SGA1: small (size) for gestational age.

Numbers analysed

At Day 28 post-vaccination, immunogenicity analysis (per protocol population) was performed in 394 maternal subjects for PT and FHA immunogenicity analysis measured by ELISA (80 subjects in the BioNet Tdap-1,1 and BioNet Tdap-2,5 groups and 78 subjects in Boostagen (two excluded due to

protocol deviation), the BioNet ap-1,1 (two excluded due to protocol deviation) and Boostrix groups (one excluded due to protocol deviation and one exclusion due to consent withdrawal)).

At delivery, 398 maternal subjects completed this visit. Immunogenicity analysis (per protocol population) was performed in 77 maternal subjects in the BioNet Tdap-1,1 and BioNet ap-1,1, 79 maternal subjects in BioNet Tdap-2,5, 75 maternal subjects in Boostagen and 78 maternal subjects in Boostrix groups.

Infant subjects

At time of birth, there were 398 babies born. Of these, 393 infant subjects enrolled to the study. Immunogenicity analysis (per protocol population) in cord or neonatal blood sample was performed in 76 subjects in the BioNet ap-1,1, BioNet Tdap-1,1 and Boostagen, 79 subjects in BioNet Tdap-2,5 and 78 subjects in Boostrix groups.

At 13 months of age, immunogenicity analysis (PP population) was performed in 69 subjects in Boostagen and Boostrix, 67 subjects in BioNet Tdap-1,1, 72 subjects in the BioNet ap-1,1 and 74 subjects in BioNet Tdap-2,5. Therefore, 351 infants were included for PT and FHA immunogenicity data analysis, and 104 infants were included for PT CHO cell assay.

Outcomes and estimation

GMCs of anti-PT antibody at Day 28 post-vaccination (TDA203 + TDA204 population)

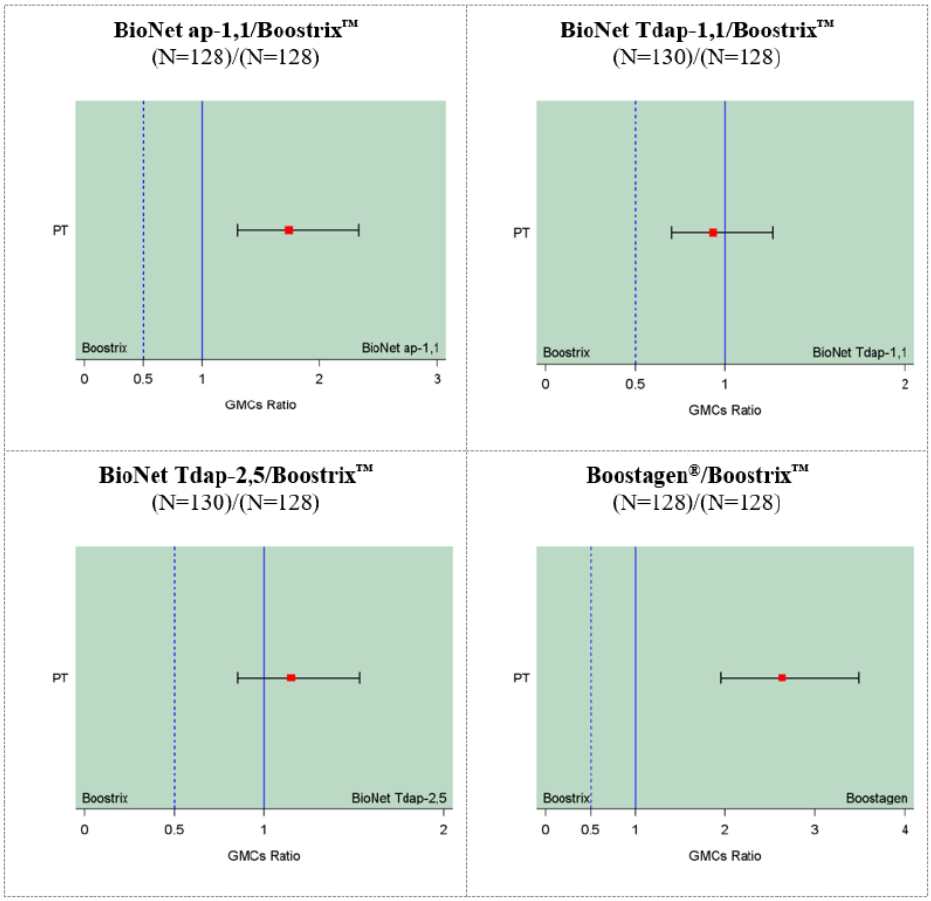
The lower bound of the 98.75% CI of the adjusted GMC ratios was >0.5 for all BioNet vaccines: 1.74 (98.75% CI 1.30-2.33), 0.93 (98.75% CI 0.70-1.26), 1.15 (98.75% CI 0.85-1.53), and 2.63 (98.75% CI 1.95-3.48) for BioNet ap-1,1, Tdap-1,1, Tdap-2,5 vaccines, and Boostagen, respectively. (Table 79 and Figure 19 Non-inferiority test for anti-PT antibody concentrations (ELISA, IU/ml) in BioNet's vaccine vs Boostrix (TDA203 & TDA204))

Table 79. Summary of anti-PT GMCs (IU/ml) as assessed by ELISA at baseline and 28 days after vaccination inn all evaluable subjects from 2 trials (TDA203 & TDA204) by vaccine groups

Vaccine groups	Baseline	Day 28	GMC Ratio	Day 28	Adjusted GMC Ratio**
	GMC (95% CI)	GMC (95% CI)	(Day 28/Baseline) (95% CI)	Adjusted GMC* (95% CI)	(98.75% CI)
BioNet ap-1,1 (N=128)	6.03 (4.97-7.30)	91.74 (73.91-113.87)	15.22 (12.22-18.96)	93.33 (79.63-110.00)	1.74 (1.30-2.33)
BioNet Tdap-1,1 (N=130)	5.55 (4.74-6.50)	49.48 (42.81-57.19)	8.92 (7.55-10.54)	50.12 (43.06-59.35)	0.93 (0.70-1.26)
BioNet Tdap-2,5 (N=130)	4.65 (3.98-5.44)	56.21 (46.54-67.90)	12.08 (10.13-14.42)	61.66 (52.26-72.09)	1.15 (0.85-1.53)
Boostagen® (N=128)	4.99 (4.21-5.91)	133.72 (111.40-160.52)	26.80 (22.47-31.97)	141.25 (118.99-164.25)	2.63 (1.95-3.48)
Boostrix™ (N=128)	6.38 (5.25-7.74)	56.10 (48.16-65.35)	8.80 (7.35-10.53)	53.70 (45.70-63.12)	- -

GMC: geometric mean of concentrations; GMC Ratio: geometric mean of individual ratio of anti-PT antibody concentrations (i.e., Day28/Baseline).
*Adjusted GMC was obtained by ANOVA adjusted for concentrations at baseline, study, and an interaction between vaccine group and study.
** Based on non-inferiority test with the lower confidence limit of the 98.75% CI of the ratio of adjusted GMC of study group of one of BioNet Tdap/Boostagen® groups or BioNet ap group to adjusted GMC of reference group: Boostrix™ group is larger than 0.5.

Figure 19. Non-inferiority test for anti-PT antibody concentrations (ELISA, IU/ml) in BioNet's vaccine vs Boostrix (TDA203 & TDA204)



GMCs of anti-PT and anti-FHA antibodies at baseline, 28 days post-vaccination and delivery in mothers

Table 80. TDA204 study- Summary of anti-PT and anti-FHA antibody GMCs (ELISA, IU/ml) at baseline and on Day 28 by vaccine group of mothers

Vaccine	PT					FHA				
	Baseline GMC (95% CI)	Day 28 GMC (95% CI)	GMCR (95% CI)	Day 28 Adjusted GMC[a] (95% CI)	Adjusted GMC Ratio[b] (98.75% CI)	Baseline GMC (95% CI)	Day 28 GMC (95% CI)	GMCR (95% CI)	Day 28 Adjusted GMC[c] (95% CI)	Adjusted GMC Ratio[d] (95% CI)
BioNet ap-1,1 (N=78)	4.64 (3.69-5.83)	65.31 (49.01-87.03)	14.08 (10.68-18.56)	67.61 (55.86-82.68)	1.48* (1.03-2.09)	7.05 (5.50-9.03)	89.39 (77.91-102.57)	12.69 (10.24-15.72)	93.33 (80.09-109.80)	0.51* (0.41-0.64)
BioNet Tdap-1,1 (N=80)	5.41 (4.43-6.60)	44.67 (38.19-52.26)	8.26 (6.79-10.05)	43.65 (35.70-52.58)	0.93 (0.66-1.33)	9.43 (7.29-12.21)	60.52 (50.81-72.10)	6.42 (5.25-7.85)	57.54 (48.98-66.88)	0.32* (0.25-0.39)
BioNet Tdap-2,5 (N=80)	4.71 (3.81-5.83)	53.76 (42.87-67.42)	11.41 (9.16-14.20)	54.95 (45.76-67.39)	1.20 (0.84-1.71)	7.83 (5.92-10.35)	112.04 (90.48-138.74)	14.31 (10.99-18.63)	112.20 (96.89-132.28)	0.62* (0.50-0.77)
Boostagen® (N=78)	4.46 (3.69-5.39)	125.89 (100.62-157.52)	28.25 (22.70-35.16)	134.90 (109.65-162.37)	2.88* (2.02-4.11)	6.73 (5.23-8.65)	112.44 (94.85-133.30)	16.72 (13.44-20.79)	120.23 (102.42-140.44)	0.66* (0.52-0.82)
Boostrix™ (N=78)	6.29 (4.96-7.98)	51.17 (42.45-61.68)	8.13 (6.47-10.22)	46.77 (38.04-56.40)	- -	9.70 (7.31-12.87)	195.29 (157.57-242.02)	20.14 (15.49-26.19)	181.97 (156.13-214.14)	- -

GMC: geometric mean of concentrations; GMCR: geometric mean of individual ratio of anti-PT and anti-FHA antibody concentrations (i.e., Day28/Baseline).
a Adjusted GMC was obtained by ANOVA adjusted for concentrations at baseline
b The GMC ratio between BioNet vaccine and Boostrix™ with 98.75% CI was calculated based on an ANCOVA model, adjusted for concentrations at baseline
c Adjusted GMC was obtained by ANOVA adjusted for concentrations at baseline and study site.
d The GMC ratio between BioNet vaccine and Boostrix™ with 95% CI was calculated based on an ANCOVA model, adjusted for concentrations at baseline and study site
*The adjusted GMC between BioNet vaccine and Boostrix™ is significantly different.

Table 81. TDA204 study- Summary of anti-PT and anti-FHA antibody GMCs (ELISA, IU/ml) at baseline and delivery by vaccine group in mothers

Vaccine	PT					FHA				
	Baseline GMC (95% CI)	Delivery GMC (95% CI)	GMCR (95% CI)	Delivery Adjusted GMC[a] (95% CI)	Adjusted GMC Ratio[b] (98.75% CI)	Baseline GMC (95% CI)	Delivery GMC (95% CI)	GMCR (95% CI)	Delivery Adjusted GMC[a] (95% CI)	Adjusted GMC Ratio[c] (95% CI)
BioNet ap-1,1 (N=77)	4.68 (3.71-5.89)	42.76 (31.92-57.29)	9.15 (6.90-12.13)	43.99 (35.78 - 54.09)	1.58* (1.09 - 2.30)	7.10 (5.52-9.13)	63.16 (54.45-73.27)	8.90 (7.27-10.89)	65.99 (56.29 - 77.39)	0.54* (0.43 - 0.67)
BioNet Tdap-1,1 (N=77)	5.57 (4.54-6.84)	28.68 (23.83-34.51)	5.14 (4.24-6.24)	27.37 (22.27 - 33.64)	0.98 (0.68 - 1.43)	10.06 (7.78-13.00)	44.09 (36.29-53.56)	4.38 (3.66-5.25)	40.31 (34.38 - 47.26)	0.33* (0.26 - 0.41)
BioNet Tdap-2,5 (N=79)	4.75 (3.84-5.89)	33.57 (26.32-42.81)	7.06 (5.68-8.78)	35.83 (29.21 - 43.95)	1.29 (0.89 - 1.87)	7.97 (6.02-10.54)	77.42 (61.94-96.77)	9.72 (7.56-12.49)	80.33 (68.64 - 93.99)	0.66* (0.52 - 0.82)
Boostagen® (N=75)	4.49 (3.69-5.46)	92.77 (71.41-120.51)	20.66 (15.94-26.76)	98.31 (79.73 - 121.20)	3.53* (2.43 - 5.15)	6.68 (5.15-8.68)	76.83 (63.33-93.19)	11.49 (9.26-14.26)	83.02 (70.66 - 97.57)	0.68* (0.54 - 0.85)
Boostrix™ (N=78)	6.29 (4.96-7.98)	30.86 (25.20-37.80)	4.91 (3.99-6.03)	27.82 (22.65 - 34.18)	- -	9.70 (7.31-12.87)	131.06 (104.05-165.08)	13.52 (10.58-17.27)	122.57 (104.66 - 143.58)	- -

GMC: geometric mean of concentrations; GMCR: geometric mean of individual ratio of anti-PT and anti-FHA antibody concentrations (i.e., Delivery/Baseline).
a Adjusted GMC was obtained by ANOVA adjusted for concentrations at baseline and GA at baseline.
b The adjusted GMC ratio between BioNet vaccine and Boostrix™ with 98.75% CI was calculated based on an ANCOVA model, adjusted for concentrations at baseline and GA at baseline.
c The adjusted GMC ratio between BioNet vaccine and Boostrix™ with 95% CI was calculated based on an ANCOVA model, adjusted for concentrations at baseline and GA at baseline.
*The adjusted GMC between BioNet vaccine and Boostrix™ is significantly different.

Seroconversion rates in anti-PT and anti-FHA antibodies at 28 days post-vaccination and delivery in mothers

Table 82. TDA204 study - Percentages of participants with ⩾4-fold increase in anti-PT and anti-FHA antibody concentrations (ELISA, IU/mL) after vaccine injection by vaccine group in mothers

Comparison groups		PT			FHA		
		BioNet Vaccine n (%) (95% CI)	Boostrix™ n (%) (95% CI)	Difference [a] (%) (95% CI)	BioNet Vaccine n (%) (95% CI)	Boostrix™ n (%) (95% CI)	Difference [a] (%) (95% CI)
BioNet ap-1,1 and Boostrix™	Day 28	66 (84.62) (74.67-91.79)	60 (76.92) (66.00-85.71)	7.69 (-4.83-20.23)	69 (88.46) (79.22-94.59)	69 (88.46) (79.22-94.59)	0.00 (-10.58-10.58)
	Delivery	57 (74.03) (62.77-83.36)	45 (57.69) (45.98-68.81)	16.33 (1.36-30.66)	62 (80.52) (69.91-88.67)	67 (85.90) (76.17-92.74)	-5.38 (-17.49-6.61)
BioNet Tdap-1,1 and Boostrix™	Day 28	63 (78.75) (68.17-87.11)	60 (76.92) (66.00-85.71)	1.83 (-11.25-14.95)	59 (73.75) (62.71-82.96)	69 (88.46) (79.22-94.59)	-14.71 (-26.90-2.53)
	Delivery	49 (63.64) (51.88-74.30)	45 (57.69) (45.98-68.81)	5.94 (-9.44-21.05)	43 (55.84) (44.07-67.16)	67 (85.90) (76.17-92.74)	-30.05 (-43.15--16.12)
BioNet Tdap-2,5 and Boostrix™	Day 28	67 (83.75) (73.82-91.05)	60 (76.92) (66.00-85.71)	6.83 (-5.71-19.42)	68 (85.00) (75.26-92.00)	69 (88.46) (79.22-94.59)	-3.46 (-14.50-7.52)
	Delivery	58 (73.42) (62.28-82.73)	45 (57.69) (45.98-68.81)	15.73 (0.82-30.04)	62 (78.48) (67.80-86.94)	67 (85.90) (76.17-92.74)	-7.42 (-19.61-4.74)
Boostagen® and Boostrix™	Day 28	76 (97.44) (91.04-99.69)	60 (76.92) (66.00-85.71)	20.51 (10.99-31.40)	74 (94.87) (87.39-98.59)	69 (88.46) (79.22-94.59)	6.41 (-2.55-16.13)
	Delivery	69 (92.00) (83.40-97.01)	45 (57.69) (45.98-68.81)	34.31 (21.41-46.62)	65 (86.67) (76.84-93.42)	67 (85.90) (76.17-92.74)	0.77 (-10.63-12.07)

[a] Difference in percentage of participants with ≥4-fold increase in anti-PT and anti-FHA antibody concentrations (IU/mL) as assessed by ELISA.
95% CI based on Clopper-Pearson method. The 2-sided 95% CI of the difference in the proportions was obtained based on Miettinen and Nurminen method

Maternal participants blood samples at delivery and cord blood or neonatal blood samples

Anti-PT antibody GMCs (IU/mL) were always higher in cord blood or neonatal blood samples than in maternal blood sample at delivery.

Table 83. TDA204 study - Summary of anti-PT antibody GMCs (ELISA, IU/ml) at Delivery (maternal blood samples) and at Birth (cord or neonatal blood samples)

Vaccine	GMC (IU/mL) (95% CI)		GMCR (95% CI) (Birth/Delivery)
	Delivery (maternal blood)	Birth (cord or neonatal blood)	
BioNet ap-1,1 (N=76)	42.93 (31.92-57.73)	59.09 (43.36-80.53)	**1.38** (1.27-1.49)
BioNet Tdap-1,1 (N=76)	28.83 (23.91-34.77)	37.20 (30.61-45.20)	**1.29** (1.18-1.41)
BioNet Tdap-2,5 (N=79)	33.57 (26.32-42.81)	49.28 (38.56-62.98)	**1.47** (1.37-1.57)
Boostagen® (N=75)	92.77 (71.41-120.51)	119.77 (94.39-151.97)	**1.29** (1.19-1.40)
Boostrix™ (N=78)	30.86 (25.20-37.80)	46.47 (37.91-56.97)	**1.51** (1.40-1.62)

GMC: geometric mean of concentrations; GMCR: geometric mean of individual ratio of anti-PT antibody concentrations (i.e., Birth/Delivery).

Maternal participants vaccinated during the second or the third trimester

Table 84. TDA204 study - Summary of anti-PT GMCs (ELISA, IU/mL) at Baseline, Day 28 (Visit 2), Delivery (Visit 3) and time of Birth (cord blood or neonatal blood) for mothers vaccinated during the second and third trimester by vaccine groups

Vaccine	GMC (IU/mL) (95% CI)							
	Baseline (maternal blood)		Day 28 (maternal blood)		Delivery (maternal blood)		Birth (cord or neonatal blood)	
	2nd trimester	3rd trimester	2nd trimester	3rd trimester	2nd trimester	3rd trimester	2nd trimester	3rd trimester
BioNet ap-1,1	N = 32 6.23 (4.10-9.48)	N = 46 3.78 (2.94-4.85)	N = 32 88.95 (56.32-140.51)	N = 46 52.68 (36.39-76.27)	N = 32 50.32 (31.88-79.41)	N = 45 38.09 (25.73-56.40)	N = 32 78.73 (47.85-129.55)	N = 44 47.96 (32.20-71.44)
BioNet Tdap -1,1	N = 38 5.98 (4.38-8.16)	N = 42 4.94 (3.79-6.44)	N = 38 42.38 (32.97-54.48)	N = 42 46.85 (38.23-57.41)	N = 36 25.95 (19.60-34.37)	N = 41 31.30 (24.31-40.31)	N = 35 33.03 (24.05-45.38)	N = 41 41.17 (32.15-52.73)
BioNet Tdap-2,5	N = 50 4.45 (3.57-5.55)	N = 30 5.19 (3.32-8.12)	N = 50 55.21 (43.61-69.88)	N = 30 51.44 (31.83-83.11)	N = 49 31.93 (24.16-42.20)	N = 30 36.43 (22.71-58.43)	N = 49 52.78 (39.50-70.54)	N = 30 44.04 (27.81-69.77)
Boostagen®	N = 35 4.68 (3.48-6.29)	N = 43 4.28 (3.31-5.54)	N = 35 124.50 (92.82-166.98)	N = 43 127.04 (90.44-178.47)	N = 35 78.21 (56.11-109.01)	N = 40 107.71 (71.96-161.22)	N = 35 113.36 (82.82-155.17)	N = 41 123.66 (86.56-176.67)
Boostrix™	N = 37 5.65 (4.09-7.81)	N = 41 6.93 (4.85-9.91)	N = 37 56.48 (41.89-76.15)	N = 41 46.81 (36.79-59.55)	N = 37 27.65 (20.32-37.61)	N = 41 34.08 (25.85-44.94)	N = 37 46.41 (33.62-64.06)	N = 41 46.52 (35.52-60.93)
GMC ratio between Boostagen® and Boostrix™ (98.75% CI)			Boostagen® - Boostrix™		Boostagen® - Boostrix™		Boostagen® - Boostrix™	
			2.20 (1.27-3.81)*	2.71 (1.54-4.79)*	2.83 (1.57-5.11)*	3.16 (1.69-5.91)*	2.44 (1.31-4.54)*	2.66 (1.46-4.84)*

GMC ratio between vaccine group with 98.75%CI based on multiplicity adjustment for pairwise comparisons using Bonferroni post-hoc analysis.
* The GMC between BioNet vaccine and Boostrix™ is significantly different.

Infant participants (eligible infants as per protocol)

GMCs of anti-PT and anti-FHA antibodies at birth (cord blood or neonatal blood samples) and at 2 months of age

Table 85. TDA024 Study- Summary of anti-PT and anti-FHA antibody GMCs (IU/ml) at Visit 3 (birth, cord blood or neonatal blood within 72 hours after birth) and Visit 4 (2 months of age) by vaccine group, with ratios between BioNet vaccines and Boostrix

Vaccine	PT			FHA		
	At birth	2 months of age	GMCR (2 months of age /at birth)	At birth	2 months of age	GMCR (2 months of age /at birth)
	GMC (95% CI)	GMC (95% CI)	(95% CI)	GMC (95% CI)	GMC (95% CI)	(95% CI)
BioNet ap-1,1	N = 76	N = 75	N=74	N=76	N=75	N=74
	59.09	16.47	0.30	86.02	24.33	0.29
	(43.36-80.53)	(12.70-21.38)	(0.26-0.34)	(73.93-100.09)	(20.89-28.34)	(0.26-0.31)
BioNet Tdap-1,1	N = 75	N = 73	N=72	N=75	N=73	N=72
	37.77	10.54	0.28	58.13	16.55	0.29
	(31.07-45.90)	(8.59-12.93)	(0.26-0.31)	(48.76-69.32)	(13.80-19.84)	(0.27-0.30)
BioNet Tdap-2,5	N = 79	N = 76	N=76	N=79	N=76	N=76
	49.28	13.88	0.28	111.26	29.82	0.27
	(38.56-62.98)	(11.00-17.51)	(0.25-0.31)	(89.78-137.87)	(23.80-37.37)	(0.25-0.29)
Boostagen®	N = 75	N = 73	N=71	N=75	N=73	N=71
	117.76	32.83	0.28	104.39	29.67	0.29
	(92.82-149.40)	(25.65-42.04)	(0.27-0.30)	(87.58-124.42)	(25.04-35.15)	(0.27-0.30)
Boostrix™	N = 78	N = 74	N=74	N=78	N=74	N=74
	46.47	12.53	0.27	193.27	54.75	0.29
	(37.91-56.97)	(10.14-15.48)	(0.25-0.29)	(154.62-241.58)	(43.23-69.33)	(0.27-0.30)
Ratio between BioNet vaccines and Boostrix™ (98.75% CI)	BioNet ap-1,1 - Boostrix™ 1.27 (0.83-1.95)	BioNet ap-1,1 - Boostrix™ 1.31 (0.87-1.99)	BioNet ap-1,1 – Boostrix™ 1.11 (0.95-1.30)	**BioNet ap-1,1 – Boostrix™ 0.45 (0.32-0.63)***	**BioNet ap-1,1 – Boostrix™ 0.44 (0.31-0.63)***	BioNet ap-1,1 – Boostrix™ 1.01 (0.90-1.13)
	BioNet Tdap-1,1 – Boostrix™ 0.81 (0.53-1.25)	BioNet Tdap-1,1 – Boostrix™ 0.84 (0.55-1.28)	BioNet Tdap-1,1 – Boostrix™ 1.06 (0.90-1.24)	**BioNet Tdap-1,1 – Boostrix™ 0.30 (0.21-0.42)***	**BioNet Tdap-1,1 – Boostrix™ 0.30 (0.21-0.43)***	BioNet Tdap-1,1 – Boostrix™ 1.00 (0.89-1.13)
	BioNet Tdap-2,5 – Boostrix™ 1.06 (0.69-1.62)	BioNet Tdap-2,5 – Boostrix™ 1.11 (0.73-1.67)	BioNet Tdap-2,5 – Boostrix™ 1.04 (0.89-1.22)	**BioNet Tdap-2,5 – Boostrix™ 0.58 (0.41-0.81)***	**BioNet Tdap-2,5 – Boostrix™ 0.54 (0.38-0.77)***	BioNet Tdap-2,5 – Boostrix™ 0.94 (0.84-1.06)
	Boostagen® - Boostrix™ 2.53 (1.65-3.90)*	**Boostagen® - Boostrix™ 2.62 (1.73-3.97)***	Boostagen® – Boostrix™ 1.06 (0.90-1.24)	**Boostagen® – Boostrix™ 0.54 (0.38-0.76)***	**Boostagen® – Boostrix™ 0.54 (0.38-0.77)***	Boostagen® - Boostrix™ 1.00 (0.89-1.12)

GMC and GMCR ratios between vaccine groups with 95% CI based on ANOVA with Bonferroni post-hoc analysis.
GMCRs were evaluated for infant participants who had sample at birth and 2 months of age. * Significant difference between the BioNet vaccine and Boostrix™

Table 86. TDA204 study - Summary of anti-PT and anti-FHA antibody GMCs (IU/ml) at Visit 6 (5 months of age) and Visit 4 (2 months of age) by vaccine group, with ratios between BioNet vaccines and Boostrix

Vaccine	PT			FHA		
	2 months of age	5 months of age	GMCR (5 months of age / 2 months of age)	2 months of age	5 months of age	GMCR (5 months of age / 2 months of age)
	GMC (95% CI)	GMC (95% CI)	(95% CI)	GMC (95% CI)	GMC (95% CI)	(95% CI)
BioNet ap-1,1	N=75	N=35	N=35	N=75	N=35	N=35
	16.47	6.51	0.40	24.33	13.37	0.55
	(12.70-21.38)	(4.73-8.95)	(0.26-0.64)	(20.89-28.34)	(10.92-16.37)	(0.42-0.73)
BioNet Tdap-1,1	N=73	N=34	N=33	N=73	N=34	N=33
	10.54	7.69	0.84	16.55	12.42	0.92
	(8.59-12.93)	(5.15-11.50)	(0.48-1.46)	(13.80-19.84)	(9.48-16.29)	(0.66-1.28)
BioNet Tdap-2,5	N=76	N=37	N=36	N=76	N=37	N=36
	13.88	6.76	0.58	29.82	15.27	0.55
	(11.00-17.51)	(4.40-10.40)	(0.34-1.02)	(23.80-37.37)	(12.15-19.18)	(0.38-0.80)
Boostagen®	N=73	N=35	N=34	N=73	N=35	N=34
	32.83	12.58	0.33	29.67	15.67	0.50
	(25.65-42.04)	(9.23-17.16)	(0.23-0.47)	(25.04-35.15)	(12.90-19.04)	(0.39-0.64)
Boostrix™	N=74	N=33	N=33	N=74	N=33	N=33
	12.53	7.78	0.62	54.75	19.74	0.33
	(10.14-15.48)	(5.16-11.75)	(0.36-1.07)	(43.23-69.33)	(15.71-24.81)	(0.26-0.41)
Ratio between BioNet vaccines and Boostrix™ (98.75% CI)	BioNet ap-1,1 - Boostrix™ 1.31 (0.87-1.99)	BioNet ap-1,1 - Boostrix™ 0.84 (0.43-1.64)	BioNet ap-1,1 – Boostrix™ 0.65 (0.27-1.58)	**BioNet ap-1,1 – Boostrix™ 0.44 (0.31-0.63)***	BioNet ap-1,1 – Boostrix™ 0.68 (0.45-1.01)	BioNet ap-1,1 – Boostrix™ 1.69 (0.99-2.88)
	BioNet Tdap-1,1 – Boostrix™ 0.84 (0.55-1.28)	BioNet Tdap-1,1 – Boostrix™ 0.99 (0.50-1.95)	BioNet Tdap-1,1 – Boostrix™ 1.36 (0.55-3.33)	**BioNet Tdap-1,1 – Boostrix™ 0.30 (0.21-0.43)***	**BioNet Tdap-1,1 – Boostrix™ 0.63 (0.42-0.94)***	**BioNet Tdap-1,1 – Boostrix™ 2.82 (1.64-4.83)***
	BioNet Tdap-2,5 – Boostrix™ 1.11 (0.73-1.67)	BioNet Tdap-2,5 – Boostrix™ 0.87 (0.45-1.69)	BioNet Tdap-2,5 – Boostrix™ 0.94 (0.39-2.27)	**BioNet Tdap-2,5 – Boostrix™ 0.54 (0.38-0.77)***	BioNet Tdap-2,5 – Boostrix™ 0.77 (0.52-1.15)	BioNet Tdap-2,5 - Boostrix™ 1.68 (0.99-2.86)
	Boostagen® - Boostrix™ 2.62 (1.73-3.97)*	Boostagen® - Boostrix™ 1.62 (0.82-3.17)	Boostagen® – Boostrix™ 0.53 (0.22-1.28)	**Boostagen® – Boostrix™ 0.54 (0.38-0.77)***	Boostagen® – Boostrix™ 0.79 (0.53-1.19)	Boostagen® – Boostrix™ 1.54 (0.90-2.62)

GMC and GMCR ratios between vaccine groups with 98.75% CI based on multiplicity adjustment for pairwise comparisons using Bonferroni post-hoc analysis.
GMCRs were evaluated in infant participants who had sample at 2 months of age and 5 months of age. * Significant difference between the BioNet vaccine and Boostrix™

Table 87. TDA204 study - Summary of anti-PT and anti-FHA antibody GMCs (IU/ml) at Visit 8 (7 months of age) and Visit 4 (2 months of age) by vaccine group, with ratios between BioNet vaccines and Boostrix

Vaccine	PT			FHA		
	2 months of age	7 months of age	GMCR (7 months of age / 2 months of age)	2 months of age	7 months of age	GMCR (7 months of age / 2 months of age)
	GMC (95% CI)	GMC (95% CI)	(95% CI)	GMC (95% CI)	GMC (95% CI)	(95% CI)
BioNet ap-1,1	N=75	N=35	N=35	N=75	N=35	N=35
	16.47 (12.70-21.38)	14.88 (9.33-23.74)	0.80 (0.35-1.82)	24.33 (20.89-28.34)	29.16 (22.73-37.42)	1.26 (0.90-1.77)
BioNet Tdap-1,1	N=73	N=36	N=34	N=73	N=36	N=34
	10.54 (8.59-12.93)	20.75 (13.32-32.31)	1.69 (0.89-3.21)	16.55 (13.80-19.84)	29.30 (21.40-40.12)	1.65 (0.98-2.78)
BioNet Tdap-2,5	N=76	N=36	N=36	N=76	N=36	N=36
	13.88 (11.00-17.51)	15.04 (9.38-24.09)	0.82 (0.41-1.65)	29.82 (23.80-37.37)	26.31 (21.10-32.81)	0.80 (0.51-1.26)
Boostagen®	N=73	N=33	N=32	N=73	N=33	N=32
	32.83 (25.65-42.04)	9.83 (6.69-14.43)	0.29 (0.18-0.46)	29.67 (25.04-35.15)	21.51 (15.57-29.72)	0.69 (0.45-1.06)
Boostrix™	N=74	N=32	N=32	N=74	N=32	N=32
	12.53 (10.14-15.48)	21.15 (12.65-35.38)	1.79 (0.86-3.69)	54.75 (43.23-69.33)	25.81 (20.59-32.36)	0.54 (0.32-0.90)
Ratio between BioNet vaccines and Boostrix™ (98.75% CI)	BioNet ap-1,1 - Boostrix™ 1.31 (0.87-1.99)	BioNet ap-1,1 - Boostrix™ 0.70 (0.31-1.59)	BioNet ap-1,1 – Boostrix™ 0.45 (0.13-1.51)	**BioNet ap-1,1 – Boostrix™ 0.44 (0.31-0.63)***	BioNet ap-1,1 – Boostrix™ 1.13 (0.70-1.83)	**BioNet ap-1,1 – Boostrix™ 2.34 (1.05-5.21)***
	BioNet Tdap-1,1 – Boostrix™ 0.84 (0.55-1.28)	BioNet Tdap-1,1 – Boostrix™ 0.98 (0.44-2.21)	BioNet Tdap-1,1 – Boostrix™ 0.95 (0.28-3.21)	**BioNet Tdap-1,1 – Boostrix™ 0.30 (0.21-0.43)***	BioNet Tdap-1,1 – Boostrix™ 1.14 (0.70-1.83)	**BioNet Tdap-1,1 – Boostrix™ 3.06 (1.37-6.84)***
	BioNet Tdap-2,5 – Boostrix™ 1.11 (0.73-1.67)	BioNet Tdap-2,5 – Boostrix™ 0.71 (0.32-1.60)	BioNet Tdap-2,5 – Boostrix™ 0.46 (0.14-1.54)	**BioNet Tdap-2,5 – Boostrix™ 0.54 (0.38-0.77)***	BioNet Tdap-2,5 – Boostrix™ 1.02 (0.63-1.65)	BioNet Tdap-2,5 - Boostrix™ 1.49 (0.67-3.29)
	Boostagen® - Boostrix™ 2.62 (1.73-3.97)*	Boostagen® - Boostrix™ 0.46 (0.20-1.06)	**Boostagen® – Boostrix™ 0.16 (0.05-0.55)***	**Boostagen® – Boostrix™ 0.54 (0.38-0.77)***	Boostagen® – Boostrix™ 0.83 (0.51-1.36)	Boostagen® – Boostrix™ 1.28 (0.57-2.89)

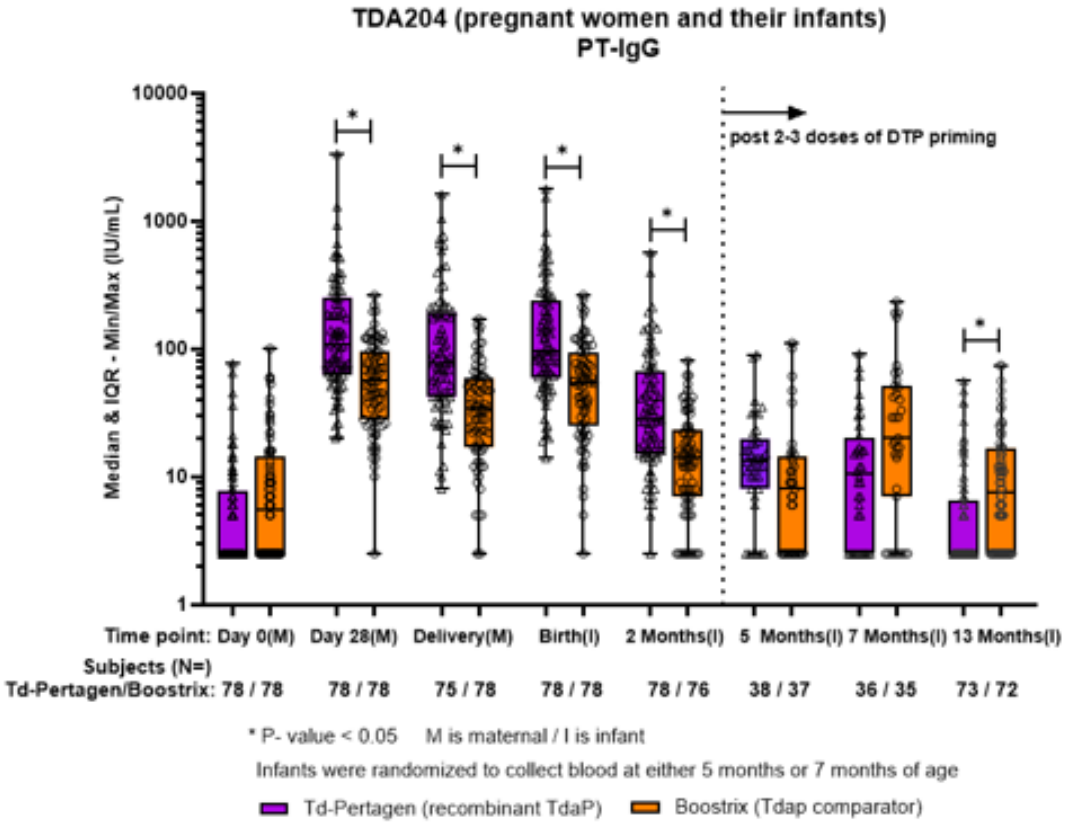
GMC and GMCR ratios between vaccine groups with 98.75% CI based on multiplicity adjustment for pairwise comparisons using Bonferroni post-hoc analysis.
GMCRs were evaluated for infant participants who had sample at 2 months of age and 7 months of age. * Significant difference between the BioNet vaccine and Boostrix™

Table 88. TDA204 study - Summary of anti-PT and anti-FHA antibody GMCs (IU/ml) at Visit 10 (13 months of age) and Visit 4 (2 months of age) by vaccine group, with ratios between BioNet vaccines and Boostrix

Vaccine	PT			FHA		
	2 months of age	13 months of age	GMCR (13 months of age / 2 months of age)	2 months of age	13 months of age	GMCR (13 months of age / 2 months of age)
	GMC (95% CI)	GMC (95% CI)	(95% CI)	GMC (95% CI)	GMC (95% CI)	(95% CI)
BioNet ap-1,1	N=75	N=72	N=72	N=75	N=72	N=72
	16.47 (12.70-21.38)	6.79 (5.15-8.96)	0.41 (0.25-0.67)	24.33 (20.89-28.34)	8.12 (6.52-10.11)	0.34 (0.26-0.46)
BioNet Tdap-1,1	N=73	N=64	N=64	N=73	N=67	N=64
	10.54 (8.59-12.93)	8.65 (6.49-11.52)	0.80 (0.52-1.23)	16.55 (13.80-19.84)	9.61 (7.67-12.06)	0.58 (0.41-0.81)
BioNet Tdap-2,5	N=76	N=73	N=73	N=76	N=74	N=73
	13.88 (11.00-17.51)	6.86 (5.40-8.71)	0.48 (0.32-0.72)	29.82 (23.80-37.37)	7.82 (6.46-9.47)	0.26 (0.18-0.37)
Boostagen®	N=73	N=67	N=67	N=73	N=69	N=67
	32.83 (25.65-42.04)	4.16 (3.38-5.11)	0.12 (0.08-0.17)	29.67 (25.04-35.15)	6.90 (5.44-8.74)	0.22 (0.16-0.30)
Boostrix™	N=74	N=69	N=69	N=74	N=69	N=69
	12.53 (10.14-15.48)	7.72 (6.12-9.74)	0.60 (0.40-0.89)	54.75 (43.23-69.33)	7.49 (6.22-9.01)	0.14 (0.10-0.19)
Ratio between BioNet vaccines and Boostrix™ (98.75% CI)	BioNet ap-1,1 - Boostrix™ 1.31 (0.87-1.99)	BioNet ap-1,1 – Boostrix™ 0.88 (0.57-1.37)	BioNet ap-1,1 – Boostrix™ 0.68 (0.32-1.44)	**BioNet ap-1,1 – Boostrix™ 0.44 (0.31-0.63)***	BioNet ap-1,1 – Boostrix™ 1.08 (0.74-1.58)	**BioNet ap-1,1 – Boostrix™ 2.49 (1.40-4.42)***
	BioNet Tdap-1,1 – Boostrix™ 0.84 (0.55-1.28)	BioNet Tdap-1,1 – Boostrix™ 1.12 (0.72-1.78)	BioNet Tdap-1,1 – Boostrix™ 1.33 (0.62-2.89)	**BioNet Tdap-1,1 – Boostrix™ 0.30 (0.21-0.43)***	BioNet Tdap-1,1 – Boostrix™ 1.28 (0.87-1.89)	**BioNet Tdap-1,1 - Boostrix™ 4.19 (2.32-7.57)***
	BioNet Tdap-2,5 – Boostrix™ 1.11 (0.73-1.67)	BioNet Tdap-2,5 – Boostrix™ 0.89 (0.58-1.40)	BioNet Tdap-2,5 - Boostrix™ 0.80 (0.38-1.70)	**BioNet Tdap-2,5 – Boostrix™ 0.54 (0.38-0.77)***	BioNet Tdap-2,5 – Boostrix™ 1.04 (0.72-1.52)	**BioNet Tdap-2,5 – Boostrix™ 1.89 (1.07-3.36)***
	Boostagen® - Boostrix™ 2.62 (1.73-3.97)*	**Boostagen® - Boostrix™ 0.54 (0.36-0.89)***	**Boostagen® - Boostrix™ 0.19 (0.09-0.41)***	**Boostagen® – Boostrix™ 0.54 (0.38-0.77)***	Boostagen® – Boostrix™ 0.92 (0.63-1.35)	Boostagen® - Boostrix™ 1.59 (0.89-2.85)

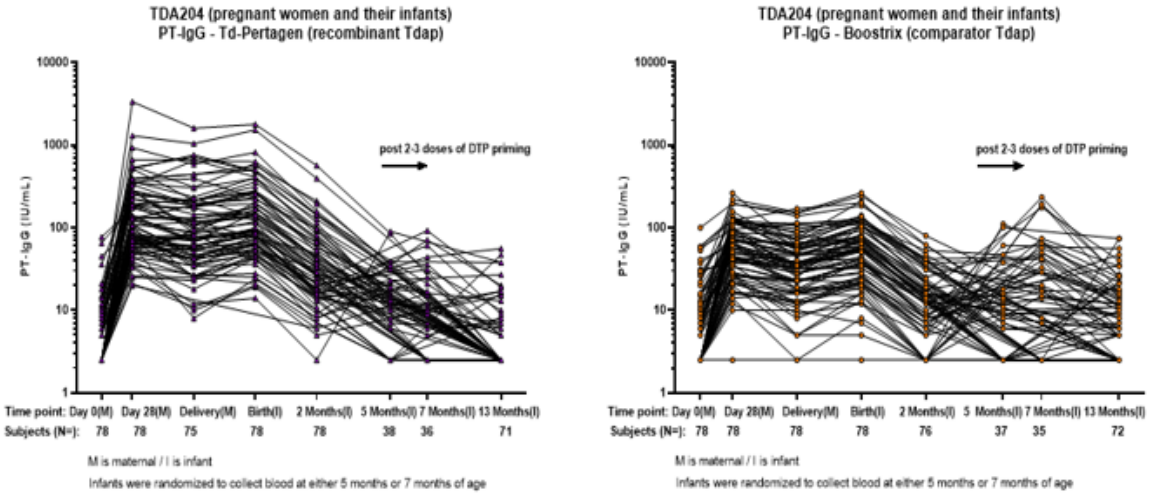
GMC and GMCR ratios between vaccine groups with 98.75% CI based on multiplicity adjustment for pairwise comparisons using Bonferroni post-hoc analysis.
GMCRs were evaluated for infant participants who had sample at 2 months of age and 13 months of age. * Significant difference between the BioNet vaccine and Boostrix™

Figure 20. Boxplots for PT-IgG levels assessed in pregnant women and their infants in the TDA204 trial



Legend: The figure shows individual data points and boxplots presenting the median, interquartile range (IQR) and lowest (minimum) and highest (maximum) value of PT-IgG antibody assessed in pregnant women and their infants participating in the TDA204 trial, before (Day 0), 28 days after and at the time of delivery in mothers (M) after maternal vaccination during pregnancy with Td-VACPERTAGEN or Boostrix™, and in infants (I) at the time of birth, at 2 months of age, and at 7 and 13 months of age after completing 3-priming doses of diphtheria-tetanus-pertussis (DTP) vaccination at 2, 4 and 6 months of age. Blood samples were collected from infants at either 5 or 7 months of age, and not at both time points, based on pre-randomisation. Statistical differences between vaccine groups were tested based on Bonferroni post hoc test on pairwise comparisons.

Figure 21. Spaghetti plots for PT-IgG levels assessed in pregnant women and their infants the TDA204 trial



Legend: The figure shows spaghetti plots presenting PT-IgG concentrations for individual mothers (M) and infants (I) participating in the TDA204 trial, before (Day 0), 28 days after and at the time of delivery in mothers (M) after maternal vaccination during pregnancy with Td-VACPERTAGEN (left hand panel, blue triangular symbols) or Boostrix™ (right hand panel, orange circular symbols), and in infants (I) at the time of birth, at 2 months of age (I), and at 5 months, 7 months (I) and 13 months (I) of age after completing 3-priming doses of diphtheria-tetanus-pertussis (DTP) vaccination at 2, 4 and 6 months of age. Infants were randomised to have a blood sample collected at either 5 months or 7 months of age. No statistical testing was performed.

Functional Antibody Response

GMTs of PT-neutralizing antibody titers at baseline, 28 days post-vaccination and delivery in mother

Table 89. TDA204 study - Summary of PT-neutralizing antibody GMTs (IU/mL) at baseline, on Day 28 and delivery by vaccine group in mother

Vaccine	Day 28					Delivery				
	Baseline GMT (95% CI)	Day 28 GMT (95% CI)	GMTR (95% CI)	Day 28 Adjusted GMT[a] (95% CI)	Adjusted GMT Ratio[b] (95% CI)	Baseline GMT (95% CI)	Delivery GMT (95% CI)	GMTR (95% CI)	Delivery Adjusted GMT[c] (95% CI)	Adjusted GMT Ratio[d] (95% CI)
BioNet ap-1,1	N=23					N=23				
	3.69 (3.16-4.29)	40.19 (20.32-79.48)	10.90 (5.87-20.25)	101.09 (55.34 - 184.71)	2.58* (1.25 - 5.34)	3.69 (3.16-4.29)	28.73 (13.95-59.14)	7.79 (4.01-15.15)	39.43 (25.17 - 61.76)	1.34 (0.70 - 2.54)
BioNet Tdap-1,1	N=24					N=22				
	5.90 (4.17-8.34)	36.50 (26.63-50.02)	6.19 (4.57-8.39)	35.34 (24.13 - 51.77)	0.90 (0.52 - 1.58)	6.27 (4.34-9.07)	29.89 (19.67-45.43)	4.77 (3.34-6.80)	27.75 (17.76 - 43.37)	0.94 (0.51 - 1.75)
BioNet Tdap-2,5	N=24					N=23				
	5.72 (3.79-8.62)	55.12 (36.16-84.02)	9.64 (6.57-14.15)	54.16 (37.01 - 79.27)	1.38 (0.79 - 2.42)	5.88 (3.84-9.00)	46.25 (26.91-79.48)	7.87 (4.77-12.98)	45.03 (29.11 - 69.63)	1.53 (0.83 - 2.83)
Boostagen®	N=24					N=23				
	5.09 (3.95-6.57)	107.04 (66.43-172.49)	21.02 (14.31-30.86)	117.79 (80.19 - 173.06)	3.01* (1.72 - 5.27)	5.21 (4.01-6.77)	91.24 (57.88-143.81)	17.50 (12.06-25.42)	97.05 (62.72 - 150.14)	3.29* (1.77 - 6.12)
Boostrix™	N=24					N=24				
	8.17 (5.27-12.66)	48.18 (32.44-71.56)	5.90 (4.05-8.60)	39.16 (26.06 - 58.84)	- -	8.17 (5.27-12.66)	38.55 (25.74-57.75)	4.72 (3.36-6.64)	29.46 (19.05 - 45.58)	- -

GMT: geometric mean of titers; GMTR: geometric mean of individual ratio of PT-neutralizing antibody titers (i.e., Day28/Baseline and Delivery/Baseine).
a Adjusted GMT was obtained by ANOVA adjusted for titers at baseline and interaction between baseline and vaccine group
b The GMT ratio between BioNet vaccine and Boostrix™ with 98.75% CI was calculated based on an ANCOVA model, adjusted for titers at baseline and interaction between baseline and vaccine group
c Adjusted GMT was obtained by ANOVA adjusted for titers at baseline.
d The GMT ratio between BioNet vaccine and Boostrix™ with 95% CI was calculated based on an ANCOVA model, adjusted for titers at baseline
*The adjusted GMT between BioNet vaccine and Boostrix™ is significantly different.

PerMIT, Antibody level in cord sera following immunisation with recombinant acellular pertussis vaccines during pregnancy: a prospective, observational study

Methods

This was a prospective observational study that was conducted in 18 to 40 years old pregnant women with uncomplicated pregnancies who previously received recombinant acellular pertussis vaccines (exposed cohort) or Td vaccine (unexposed cohort) during pregnancy and delivered in the study center. Informed consent and screening was performed during the routine antenatal care visits until delivery. Pregnant women who received vaccines (recombinant aP, recombinant TdaP or Td vaccine) during pregnancy were offered study information and asked if they were willing to participate in the study. All subjects provided written informed consent prior to participation.

Table 90. Vaccine groups

Vaccine group	Cohort	N
Recombinant aP (Pertagen®)	Exposed	256
Recombinant TdaP (Boostagen®)	Exposed	252
Licensed Td	Unexposed	76
Total	N/A	584

Study Participants

A total of 620 subjects were screened, and 584 subjects were enrolled after determining eligibility. Only those subjects who fulfilled all of the inclusion criteria and none of the exclusion criteria were included in the study. Importantly, subjects with any significant congenital abnormality confirmed by ultrasound, fetal abnormality, stillbirth or neonatal death, and those who have received a pertussis vaccine within 1 year prior to the current pregnancy were excluded from this study.

Study assessments

At delivery, obstetric and birth outcomes were recorded. A sample of umbilical cord blood (6 mL) was collected immediately after birth. Anti-PT and anti-FHA IgG titers were analysed by using commercial ELISA kit according to the manufacturer's instructions. The ELISA kits used were calibrated based on the World Health Organization international standards. The values were expressed in International Units (IU) per milliliter. The lower limit of quantification (LLOQ) of the assay was <5 IU/mL. Anti-PT-neutralizing antibody assay were performed by Human Serology Laboratory of BioNet-Asia using validated CHO cell method.

Objectives

Primary endpoint:

- Geometric mean concentrations of PT-neutralizing, anti-PT and anti-FHA IgG titers in cord sera following maternal immunisation with recombinant aP or TdaP vaccine

Secondary endpoints:

- Seropositive rates of anti-PT GMCs in infants based on antibody levels in cord sera following maternal immunization with recombinant aP or TdaP vaccine

Definition of seropositivity:

Since seroprotective antibody level against pertussis is unknown, the seropositivity in infants at delivery born from mothers who received a pertussis vaccine during pregnancy was estimated to be >30 IU/mL based on anti-PT half-life of 36 days in adults and expected antibody level of > 5 IU/mL in a 3-month old infant (Eberhardt et al, 2016).

Exploratory endpoint:

- Geometric mean concentrations of PT-neutralizing antibody, anti-PT and anti-FHA GMCs in cord sera at delivery following maternal immunisation of recombinant aP or TdaP at different gestational ages (< 27 weeks, 27-36 weeks and > 36 weeks of gestation)

Sample size

Total planned sample size was 500 subjects. The estimated number of subjects enrolled in the exposed cohort was 75-80% (approximately 400 subjects) of total subjects. At the time of this proposal, there was no data of maternal antibody transfer of recombinant acellular pertussis vaccines available.

For the primary objective, sample size was calculated to show the antibody transfer (determining the antibody levels in cord blood) based on the estimated anti-PT antibody levels in mothers who received recombinant acellular pertussis vaccines (VacPertagen or Boostagen) and ratio of cord blood antibody to maternal antibody at delivery.

Statistical methods

Analysis populations

Enrolled Population includes all screened subjects who provided informed consent and received a Subject Number, regardless of the subject's status in the study.

Immunogenicity Analysis Population includes the subjects in the enrolled population and their cord blood sample can be collected at delivery visit with no major protocol deviations that were determined to potentially interfere with the immunogenicity assessment of the vaccine received during current pregnancy. This population served as the primary analysis population for all immunogenicity objectives.

Analysis of immunogenicity endpoints

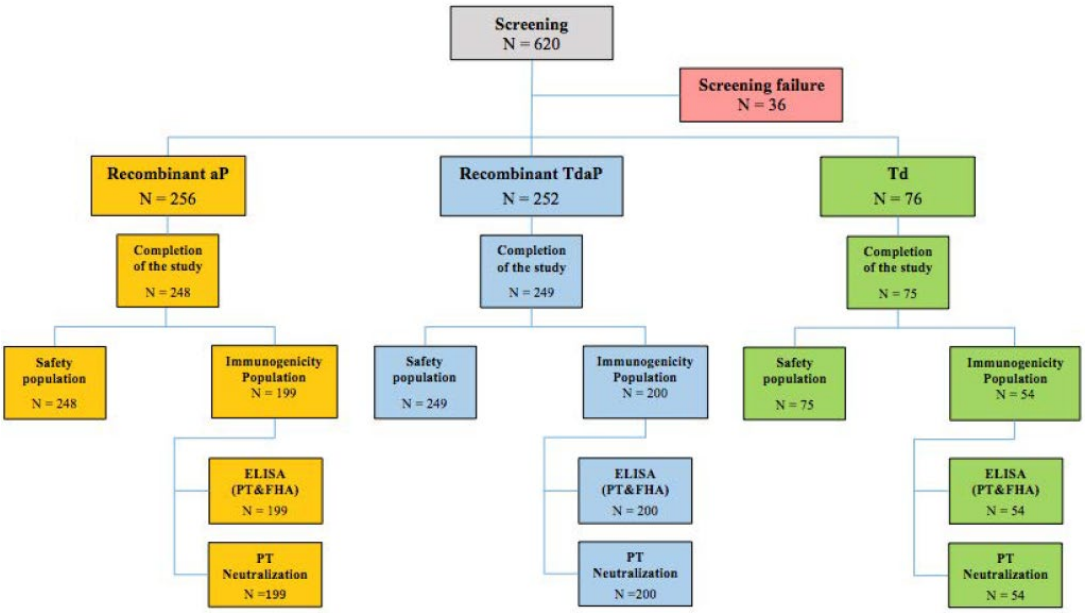
Immunogenicity analysis population was used for immunogenicity analysis. Descriptive statistics were performed for analysis of immunogenicity endpoints for this study. No missing data imputation techniques were used to account for missing, unused or spurious data.

Primary objective: GMCs of anti-PT and anti-FHA antibodies measured by ELISA as well as GMTs of anti-PT-neutralizing antibody titer measured by CHO cell assay in cord sera following maternal immunisation with recombinant aP or TdaP vaccine or licensed Td vaccine were calculated for each vaccine group along with their exact two-sided 95% CI. The ratio of GMCs or GMTs in cord sera following maternal immunisation with recombinant aP or TdaP vaccine to that in licensed Td vaccine and their exact two-sided 95% CI were obtained. The difference between groups for continuous variables were assessed by ANOVA or Kruskal-Wallis test, post-hoc analysis. $P \leq 0.05$ was considered to be statistically significant.

Results

Participant flow

Figure 22. Overall study population



Recruitment

The study was conducted from January 2019 (first subject firs visit) until May 2020 (last subject last visit).

Conduct of the study

During the conduct of this study a total of 155 protocol deviations occurred. The most common protocol deviation occurred when the subject was eligible, but cord blood was not collected (n = 105). In addition, some subjects were not eligible, but their data was collected (n = 18) and some had unusable cord blood that was frozen during storage and could not be used for immunological assays (n = 14). 12 subjects were lost to follow-up due to delivery in another hospital, and 6 subjects were not eligible on delivery day, but their data was collected.

Baseline data

Demographic characteristics at baseline (screening day) were similar across vaccine groups (Table 91). All subjects were Thai.

Table 91. Summary of demographics at baseline

Subject status	Recombinant aP vaccine (Pertagen®) (N = 256)	Recombinant TdaP vaccine (Boostagen®) (N = 252)	Td vaccine (N = 76)	Total (N = 584)	P-value
Demographics (Screening day)					
Age (years)					0.0008 [2]*
- N	256	252	76	584	
- Mean (SD)	29.38 (5.05)	30.59 (4.90)	28.03 (5.74)	29.72 (5.15)	
- Median	30.00	31.00	28.00	30.00	
- Min/Max	18-40	19-40	18-39	18-40	
Ethnicity, n (%)					- [1]
- N	256	252	76	584	
- Thai	256 (100.00%)	252 (100.00%)	76 (100.00%)	584 (100.00%)	
- Other	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
Weight (kg)					0.3136 [2]
- N	256	252	76	584	
- Mean (SD)	68.22 (12.29)	69.37 (11.61)	68.85 (14.09)	68.79 (12.24)	
- Median	67.00	67.75	65.10	67.05	
- Min/Max	47-127.5	48-129	46.1-115.7	46.1-129	
Height (cm)					0.4280 [2]
- N	256	252	76	584	
- Mean (SD)	158.61 (5.91)	158.95 (5.75)	157.84 (5.56)	158.66 (5.80)	
- Median	158.00	159.00	158.00	158.50	
- Min/Max	144-175.5	145-175	140-173	140-175.5	

Note:
[1] No p-value was computed by SAS.
[2] P-value based on Kruskal-Wallis test
* P-value ≤ 0.05 is considered statistically significant.

The majority of subjects across all vaccine groups were experiencing their first pregnancy and had normal ultrasound results. The mean gestational age was 28.54 (±5.14) weeks, and most subjects (73.97%) were vaccinated between 27-36 weeks into current pregnancy (Table 92).

Table 92. Summary of Gestational Age of mothers at vaccination during current pregnancy (screening day)

Subject status	Recombinant aP vaccine (Pertagen®) (N = 256)	Recombinant TdaP vaccine (Boostagen®) (N = 252)	Td vaccine (N = 76)	Total (N = 584)	P-value
Gestational Age (Weeks)					
- N	256	252	76	584	
- Mean (SD)	30.58 (3.50)	29.00 (3.24)	20.12 (6.44)	28.54 (5.14)	<0.0001 [1]*
- Median	31.00	29.00	19.50	29.00	
- Min/Max	21-40	20-36	7-38	7-40	
< 27 weeks					
- n (%)	31 (12.11%)	46 (18.25%)	63 (82.89%)	140 (23.97%)	
27-36 weeks					
- n (%)	214 (83.59%)	206 (81.75%)	12 (15.79%)	432 (73.97%)	
> 36 weeks					
- n (%)	11 (4.30%)	0 (0.00%)	1 (1.32%)	12 (2.05%)	

Note:
[1] P-value based on Kruskal-Wallis test
* P-value ≤ 0.05 is considered statistically significant.

Numbers analysed

A total of 620 subjects were screened, and 584 were enrolled. There were 508 subjects in the exposed cohort where 256 subjects received the recombinant aP vaccine and 252 received the recombinant TdaP. 76 subjects were in the non-exposed cohort and received the Td vaccine.

Table 93. Study subject disposition for safety and immunogenicity analysis

Subject status	Recombinant aP vaccine (Pertagen®)	Recombinant TdaP vaccine (Boostagen®)	Td vaccine	Total
Number of screened subjects, n (%)	N = 620			
Number of enrolled subjects, n (%)	256	252	76	584
Completion of the study	248 (96.88%)	249 (98.81%)	75 (98.68%)	572 (97.95%)
Non-completion of the study	8 (3.13%)	3 (1.19%)	1 (1.32%)	12 (2.05%)
Subjects included in the safety analysis	248 (96.88%)	249 (98.81%)	75 (98.68%)	572 (97.95%)
Subjects included in the ELISA immunogenicity analysis (PT & FHA)	199 (77.73%)	200 (79.37%)	54 (71.05%)	453 (77.57%)
Subjects included in the PT neutralization analysis	199 (77.73%)	200 (79.37%)	54 (71.05%)	453 (77.57%)
Subjects excluded in the safety analysis	8 (3.13%)	3 (1.19%)	1 (1.32%)	12 (2.05%)
Subjects excluded in the ELISA immunogenicity analysis (PT & FHA)	57 (22.27%)	52 (20.63%)	22 (28.95%)	131 (22.43%)
Subjects excluded in the PT neutralization analysis	57 (22.27%)	52 (20.63%)	22 (28.95%)	131 (22.43%)

Outcomes and estimation

ELISA for pertussis antibodies in Cord Blood:

In Table 98, the anti-PT GMCs at delivery were the highest in the VacPertagen group (206.1 IU/mL, 95% CI 164.3-258.6), followed by the Boostagen group (153.1 IU/mL, 95% CI 129.1-181.5).

Comparatively, the anti-FHA GMCs were the highest in the Boostagen group (232.0 IU/mL, 95% CI 199.0-270.6), followed by the VacPertagen group (217.2 IU/mL, 95% CI 184.0-256.4)

In relation to gestational age for maternal immunisation, there was comparable anti-PT IgG titers in pregnant women who received VacPertagen and Boostagen between <27 weeks and 27-36 weeks of gestation (Table 100).

Functional Antibody Response

The PT-neutralizing antibody titers (GMTs) in cord blood were the highest in the VacPertagen group (105.3 IU/mL, 95% CI 81.7-135.8), followed by the Boostagen group (81.5 IU/mL, 95% CI 66.4-100.0).

Table 94. Geometric Mean Concentrations (GMCs) of cord blood PT-IgG, FHA-IgG and PT neutralising antibodies in women vaccinated with VacPertagen, Boostagen or Td vaccine

Antibody	Exposed		Unexposed	P-value*
	Pertagen® GMC (IU/mL) (95% CI)	Boostagen® GMC (IU/mL) (95% CI)	Td-only GMC (IU/mL) (95% CI)	
PT	206.1 (164.3-258.6)	153.1 (129.1-181.5)	6.5 (4.9-8.8)	<0.0001
FHA	217.2 (184.0-256.4)	232.0 (199.0-270.6)	12.2 (8.6-17.4)	<0.0001
PT-Neutralizing	105.3 (81.7-135.8)	81.5 (66.4-100.0)	3.8 (2.8-5.1)	<0.0001

* P-value based on Kruskal-Wallis test

Table 95. Seroresponse rates with the cord blood PT-IgG, FHA-IgG and PT neutralizing titers > 30 IU/ml in women vaccinated with VacPertagen, Boostagen or Td vaccine

Seroresponse Rate	Exposed		Unexposed	P-value*
	Pertagen®	Boostagen®	Td-only	
	N=199	N=200	N=54	
	n (%) (95% CI)	n (%) (95% CI)	n (%) (95% CI)	
PT	175 (87.94) (83.41-92.46)	182 (91.00) (87.03-94.97)	5 (9.26) (1.53-16.99)	<0.0001
FHA	185 (92.96) (89.41-96.52)	195 (97.50) (95.34-99.66)	10 (18.52) (8.16-28.88)	<0.0001
PT-Neutralizing	159 (79.90) (74.33-85.47)	160 (80.00) (74.46-85.54)	3 (5.56) (0.00-11.67)	<0.0001

* Overall P-value (2-sided) based on Chi-square test

Table 96. Cord blood PT-IgG GMCs (IU/ml) in women vaccinated with VacPertagen, Boostagen or Td vaccine at different gestational ages (<27 weeks, 27-36 weeks and >36 weeks of gestation)

Gestational ages at immunization	Recombinant aP vaccine (Pertagen®) (N = 199)	Recombinant TdaP vaccine (Boostagen®) (N = 200)	Td vaccine (N = 54)	P-value[a]
	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	GMC (IU/mL) (95% CI)	
<27 weeks (N = 109)	N = 28	N = 36	N = 45	<0.0001 [2]*
	295.26 (168.68-516.81)	140.65 (96.82-204.32)	6.24 (4.60-8.45)	
27-36 weeks (N = 333)	N = 161	N = 164	N = 8	<0.0001 [2]*
	231.83 (183.46-292.95)	155.99 (128.69-189.07)	9.41 (2.78-31.78)	
>36 weeks (N = 11)	N = 10	N = 0	N = 1	- [1]
	11.36 (4.92-26.27)	0.00 (0.00-0.00)	2.50 (0.00-0.00)	
P-value[b]	<0.0001 [3]*	- [1]	0.5369 [2]	
GMC ratio between two gestational age groups (95% CI)	<27 weeks and 27-36 weeks 1.27 (0.61-2.65)	<27 weeks and 27-36 weeks 0.90 (0.58-1.41)	<27 weeks and 27-36 weeks 0.66 (0.24-1.85)	
	<27 weeks and >36 weeks 25.98 (6.95-97.07)*	<27 weeks and >36 weeks -	<27 weeks and >36 weeks 2.49 (0.17-37.40)	
	27-36 weeks and >36 weeks 20.40 (6.36-65.47)*	27-36 weeks and >36 weeks -	27-36 weeks and >36 weeks 3.76 (0.22-64.43)	

a: Compared between recombinant acellular pertussis-based vaccine and Td vaccine
b: Compared between gestational ages
GMC ratio between two gestational age groups (95% CI) based on Bonferroni post-hoc analysis
Note:
[1] No p-value was computed from SAS.
[2] P-value based on Kruskal-Wallis test
[3] P-value based on one-way ANOVA
* P-value ≤ 0.05 is considered statistically significant.

WoMANPOWER, Safety and immunogenicity of an acellular pertussis vaccine containing genetically detoxified pertussis toxin administered to pregnant women living with and without HIV and their newborns (WoMANPOWER): a randomised controlled trial in Uganda, published in Nakabembe et al. Lancet Glob Health 2025; 13: e81–97

No CSR was provided. A summary of the results was provided.

Methods

WoMANPOWER study was an observer-blind, randomised, phase 2, multicentre, non-inferiority trial evaluating safety and immunogenicity of a vaccine containing genetically detoxified acellular pertussis in pregnant women living with HIV in Uganda. Women aged at least 18 years between 16 weeks and 26 weeks of gestation were randomly assigned to receive either a standard of care (2 doses of tetanus-diphtheria [Td] vaccine) or intervention (one dose of Td followed by one dose of TdaP [Boostagen] vaccine) by intramuscular injection (0.5 mL) with 4-week interval. Stratified block randomisation using blocks of four with a 1:1:1:1 ratio stratified by participant HIV status was used to distribute participants into equal groups (50 participants per group for a total of 200 participants). Participant HIV status was based on confirmatory testing with CD4 count and viral load. All maternal

subjects were followed up for 28 days for AEs. SAEs were followed until 12 months after delivery in mothers and until 12 months of age in infants. Immunogenicity in maternal subjects was assessed at enrolment (baseline) and 4 weeks after the second vaccination and delivery. Antibody concentrations (anti-pertussis toxin and anti-filamentous haemagglutinin IgG concentrations) in infants were measured at birth (cord or neonatal venous blood samples) and 4 weeks following 3-dose of pentavalent vaccine containing whole-cell pertussis at the 18-week visit. Routine infant vaccination was given at 6 weeks, 10 weeks and 14 weeks old.

No details on the immunogenicity assays are provided, including validation status.

Results

Between Oct 28, 2020, and May 21, 2021, 438 pregnant women were screened (2 sites) and 181 were randomly assigned: 90 to Boostagen (40 HIV-positive participants and 50 HIV-negative participants) and 91 to Td vaccine (41 HIV-positive participants and 50 HIV-negative participants). All participants received Td as first vaccination between 16+0 weeks and 25+6 weeks of gestation, and 4 weeks later, 177 received either Td or TdaP (between 20+0 weeks and 29 +6 weeks of gestation). There were 1 woman withdrawing and 3 women being lost to follow-up before the second vaccination, and 1 woman withdrawing after the second vaccination without providing delivery data.

The mean age of enrolled pregnant women was 25 years (range 18–41) and 180/181 participants (99%) were Ugandan. All enrolled pregnant women living with HIV were on combined antiretroviral therapy (cART), with a mean CD4+ T lymphocyte count of 652 cells per µL (range 58–1475); 69 (85·2%) of 81 participants were receiving a dolutegravir-based regimen.

The planned sample size of 40 infants in each group at the 18-week visit after accounting for attrition was not met (TdaP HIV-positive participants [N=36], TdaP HIV-negative participants [N=43], Td HIV-positive participants [N=36], and Td HIV-negative participants [N=44]). Nevertheless, a comparison based on 95% CIs for geometric mean ratios were calculated allowing for *informal* non-inferiority to be assessed.

Table 97. Geometric mean concentrations of anti-PT IgG and anti-FHA IgG by study arm and HIV status in the modified intention-to-treat populations

GMC IU/mL (95% CI)	TdaP vaccine		Td vaccine	
	HIV positive (N=40)	HIV negative (N=50)	HIV positive (N=41)	HIV negative (N=50)
Anti-pertussis toxin IgG				
Baseline	10.7 (7.5, 15.3) [n=39]	10.7 (8, 14.2) [n=48]	12.3 (9.1, 16.8) [n=37]	10.7 (7.8,14.7) [n=48]
Second vaccination + 4 weeks	133.9 (91.4, 196.1) [n=40]	245.1 (167.8, 358) [n=46]	11.8 (8.6, 16.1) [n=38]	9 (6.5, 12.6) [n=46]
Maternal delivery	87.4 (55.1, 138.5) [n=37]	121.7 (79.4, 186.6) [n=42]	10.2 (7, 14.9) [n=36]	6.6 (4.5, 9.6) [n=38]
Infant delivery	114.7 (70.4, 186.8) [n=36]	169.7 (101.2, 284.5) [n=34]	11.9 (8.2, 17.4) [n=33]	7.9 (5.1, 12.2) [n=40]
Infant 18 weeks	16.8 (10.7, 26.1) [n=35]	23.5 (15.6, 35.3) [n=43]	87.6 (43.7, 175.6) [n=35]	141.1 (80.9, 246.1) [n=44]
Delivery to 18-week fold-change	0.14 (0.08, 0.25) [n=32]	0.13 (0.07, 0.24) [n=32]	6.55 (2.34, 18.34) [n=32]	18.82 (7.59, 46.63) [n=37]
Transplacental ratio	1.31 (1.17, 1.46) [n=36]	1.52 (1.2, 1.93) [n=34]	1.31 (1.02, 1.67) [n=33]	1.09 (0.87, 1.37) [n=38]
Anti-filamentous hemagglutinin IgG				
Baseline	14.4 (9.5, 21.8) [n=39]	19.9 (15.4, 25.6) [n=48]	18.6 (13.5, 25.6) [n=37]	20.5 (15.8, 26.6) [n=48]
Second vaccination + 4 weeks	101.7 (65.3, 158.3) [n=40]	313.8 (236.5, 416.4) [n=46]	17.9 (13.3, 24.1) [n=38]	16.2 (12.6, 21) [n=46]
Maternal delivery	66.4 (41.7, 105.7) [n=37]	210.9 (158.1, 281.4) [n=42]	18.8 (13.4, 26.3) [n=36]	16.5 (12.2, 22.2) [n=38]
Infant delivery	74.1 (44.3, 124.2) [n=36]	269.9 (186.5, 390.6) [n=34]	17.5 (11.6, 26.4) [n=33]	17.5 (12.5, 24.7) [n=40]
Infant 18 weeks	20.2 (15.4, 26.5) [n=35]	22.2 (17.1, 28.7) [n=43]	19.8 (14.2, 27.7) [n=35]	17.1 (12.8, 22.7) [n=44]
Delivery to 18-week fold-change	0.25 (0.13, 0.48) [n=32]	0.09 (0.07, 0.11) [n=32]	1.07 (0.59, 1.95) [n=32]	0.96 (0.59, 1.55) [n=37]
Transplacental ratio	1.13 (1, 1.29) [n=36]	1.22 (0.9, 1.66) [n=34]	0.94 (0.78, 1.14) [n=33]	1.02 (0.83, 1.24) [n=38]

Table 98. GMCs of serum bactericidal activity against B.pertussis before and after wP priming (3 doses) in infants (WoMANPOWER)

	Serum Bactericidal Activity (SBA) titers against *B. pertussis*					
Womanpower	**Birth**		**18 weeks Post-DTwP**		**10 months**	
	N	GM (95% CI)	*N*	GM (95% CI)	*N*	GM (95% CI)
Td-VACPERTAGEN	17	13.1 (6.6-26.0)	15	199.3 (132.0-540.3)	13	10.3 (5.3-19.9)
HIV-pos	18	28.8 (12.6-65.8)	19	328.7 (201.1-537.1)	19	22.1 (11.1-44.0)
HIV-neg						
Td	17	12.0 (5.6-25.6)	19	280.1 (152.9-513.1)	16	22.1 (8.8-55.5)
HIV-pos	17	11.9 (5.9-24.1)	20	383.3 (271.9-540.3)	16	27.5 (12.4-61.0)
HIV-neg						

2.6.6. Discussion on clinical efficacy

General aspects of the data package

The submitted dossier includes immunogenicity data from 11 clinical studies. Most of these studies were randomized, observer-blind, controlled studies, except for one prospective observational study in pregnant women (PerMIT). From the 12 submitted studies, 3 were considered as main studies (TDA202, TDA206 and TDA207) investigating adolescents, adults and pregnant subjects, respectively. The remaining studies were considered supportive, either due to their study design or because VacPertagen itself was not investigated but only other comparable formulations were investigated. The Applicant also submitted immunogenicity data from young infants which were obtained with vaccine formulations including PT_{gen} and FHA. Only short summaries or publications were submitted for 2 supportive studies (PertADO and WOMANPOWER). Immunogenicity of VacPertagen was investigated in ~400 subjects in RCTs across the populations (adolescents, adults, elderly, pregnant women) which is overall considered limited yet acceptable to support the full MAA of a novel vaccine although data obtained with Boostagen (Td-VacPertagen) were considered supportive.

Across all studies, most relevant immunogenicity analyses concern the comparison of antibody responses against PT and FHA between VacPertagen or Boostagen and widely used comparators such as Adacel or Boostrix. Other compared formulations are considered of minor relevance for this MAA.

Both Adacel and Boostrix are approved reduced-antigen combined tetanus, diphtheria, and acellular pertussis vaccine (Tdap) vaccines. Adacel, the active comparator of the main studies (TDA202, TDA206 and TDA207), also contains 3 additional pertussis antigens (PRN and FIM Type 2 and 3) as well as antigens from tetanus and diphtheria bacteria, similar to other possible Tdap comparators. Boostrix which was used in the supportive studies, contains the PRN in addition to PT and FHA.

It is considered that GMCs/GMTs (over seroconversion rate) are the most appropriate endpoint to assess responses to pertussis antigens in booster settings. The primary analyses were performed 1 month after vaccination which is acceptable.

The only study for which analyses of the cell-mediated immune responses induced by VacPertagen were provided is study TDA202 (exploratory endpoint). Broader characterisation of humoral responses were planned in the supportive studies Pertaprime-01 and TDA204.

Evaluation of data was mostly to be interpreted descriptively although the Applicant concluded inferential analyses for several studies. Multiplicity control over the multiple study arms and endpoints was seldom implemented. The limited sample sizes influenced the safety assessment, see safety section. Finally, the presentation of relevant statistical details was lacking (NI margins and other details were missing from the protocols and could only be found as footnotes in the SAP after being

referred to from the CSR, summarising plots for the results were only submitted upon request). Overall, the dossier gives the impression of containing well-planned, well-performed proof of concept studies, but none that was specifically planned to generate pivotal evidence for a MAA.

Individual vaccination histories were only available for the Pertaprime-01 and PertADO studies. No individual vaccination histories were available for the pivotal studies provided in Thailand. Instead, the vaccination history was assumed based on national vaccination schedules. According to a publication by Blackwood et al 2013, coverage for 2 or 3 primary doses was low until 1983, increased from 40% to 90% between 1984 and 1990, and has stayed high since (>95%). Direct comparison of wP and aP-primed subjects was only performed in the Pertaprime-01 study where no substantial differences in immunogenicity were observed as described further below. Booster vaccinations after primary immunisation were only routinely performed in Australia and Switzerland (Pertaprime-01 and PertADO studies). The booster doses did not yield increased antibody titres compared to studies in Thailand and Uganda. However, antibody titres cannot be reliably compared between studies and therefore, the relevance of this observation is unclear. Although possible conclusions are limited and individual vaccination history for all studies would have been preferred, the overall picture that VacPertagen induces antibody responses against PT and FHA that are at least comparable to licensed vaccines is consistent and no substantial differences were observed between the different pre-immunisation scenarios and geographical regions.

For the studies conducted in Thailand, it is assumed that subjects received a primary vaccination with whole-cell pertussis combination vaccines (DTwP), which is still the vaccine currently recommended for routine vaccination. This is different from the current situation in Europe for people born after DTaP vaccines were approved and marketed (variable dates pending country), because only a limited number of countries still administer DTwP vaccines in Europe and coverage is unclear. However, also based on data provided by the Applicant, there is still a considerable proportion of women of childbearing potential in Europe who have been primed in infancy with whole cell pertussis vaccines. The majority of subjects in the data package participated in studies conducted in Thailand. Clinical data from outside Thailand are only available from the small PertADO and the Pertaprime-01 studies conducted in Switzerland and Australia, respectively. Upon request, the Applicant provided a discussion on the applicability of the Thai population to Europe in the light of potential differences in genetic (HLA) background and pre-immunisation scenarios. No analyses of possible differences in HLA alleles and their capabilities to present peptides from pertussis proteins or a discussion on possible differences of the elicited T cell responses were provided. Nevertheless, the Applicant sufficiently justified that results of the Thai population can be extrapolated to the European population despite possible genetic differences between populations. The provided literature analysing (pertussis) vaccination in different ethnicities clearly suggests that no substantial differences need to be expected after vaccination of different populations. It is furthermore acknowledged that also the studies conducted in Australia and Switzerland showed at least comparable immune responses after vaccination with VacPertagen when directly compared to licensed vaccines.

Specific aspects of clinical studies

Studies in adolescent subjects

Study TDA202

The TDA202 study was a double-center, observer-blind, randomized phase II/III study conducted in Thailand to evaluate immunogenicity, reactogenicity and safety of with VacPertagen (BioNet 2-component aP; PT_{gen} and FHA) and Boostagen (BioNet $TdaP_{gen}$ vaccine) in comparison to the licensed Tdap vaccine Adacel after a single dose in healthy adolescents randomized 1:1:1. The single dose booster vaccination investigated in this trial is in line with similar commercial Tdap vaccines including

Adacel and the overall study design is acceptable. This trial is considered pivotal in providing evidence for vaccine effectivity after a single dose in adolescents.

A total of **450 adolescent subjects** (≥12 to <18 yoa) were enrolled for the initial TDA202 study. The mean age of adolescent subjects was 14.4 years. Demographic characteristics of study subjects were similar between the 3 vaccine groups. This population is recommended for a booster of Tdap vaccine in several countries of the world. The study population is considered sensitive for the intended immunobridging. Eligibility criteria are acceptable.

The **immunogenicity analyses** (e.g. antibody responses against PT by ELISA and functional antibody titers by PT neutralizing assay on CHO cell) can be considered as established methods in clinical immunogenicity studies for pertussis vaccines and the respective assays were fully validated. The methods for determining immunogenicity are therefore considered fit for purpose.

Primary immunogenicity analyses were performed 28 days after vaccination (primary analyses; approx. 150 subjects per vaccine group) and antibody persistence was investigated 1, 2, 3 and 5 years after vaccination in subsets of 50 to 60 subjects per vaccine group.

The **primary aim** of study TDA202 was to investigate non-inferior immunogenicity of BioNet TdaP vaccine in comparison to Adacel vaccine. The secondary purpose of the study was to demonstrate non-inferior immunogenicity of BioNet aP (VacPertagen) vaccine in comparison to Adacel vaccine and evaluate the safety and reactogenicity of BioNet aP and TdaP vaccines.

Methodologically, some limitations were observed. NI hypotheses were not clearly pre-defined in the protocol (only as footnote in the SAP), the sample size calculation lacked some details in the protocol/SAP and was only clarified upon request, and the analyses were inconsistently adjusted for multiplicity despite the multiple groups and the multiple antigens. Formally, results for secondary endpoints are to be interpreted descriptively only.

At 28 days after vaccination, the three groups showed significantly different seroconversion rates (primary endpoint). The rates in the BioNet aP [anti-PT 96% (95% CI 93-99) and anti-FHA 93% (95% CI 89-97)] group were higher than the seroconversion rates in the Adacel group [anti-PT 55% (95% CI 47-63), anti-FHA 54% (95% CI 46-62)]. Likewise, Anti-PT and anti-FHA GMTs were higher in BioNet aP [562 IU/mL (95% CI 467.79-674.86) for anti-PT antibody; 924 IU/mL (95% CI 809.39-1054.4) for anti-FHA antibody] than those GMTs in Adacel group [63 IU/mL (95% CI 51.05-78.37) for anti-PT antibody; 242 IU/mL (95% CI 208.86-280.05) for anti-FHA antibody]. In addition, seroconversion rates of anti-PT neutralizing antibodies and corresponding GMTs were higher in the BioNet aP group than in the Adacel group. Therefore, consistently higher antibody responses were observed with BioNet aP (VacPertagen) compared to the licensed Adacel approximately 1 month after vaccination. Higher anti-FHA titers following administration of VacPertagen as compared to Adacel are unexpected, considering that same amounts of FHA are formulated in VacPertagen and Adacel.

At 1 year after vaccination, ELISA anti-PT and anti-FHA seroconversion rates were higher in the BioNet aP group [Anti-PT 82% (95% CI 71-93), anti-FHA 64% (95% CI 51-77)] than the seroconversion rates in the Adacel group [anti-PT 4% (95% CI 0-9), anti-FHA 28% (95% CI 16-40)]. Also ELISA anti-PT and anti-FHA GMTs were higher in the BioNet aP and group [133 IU/mL (95% CI 92.96- 189.77) for anti-PT antibody; 291 IU/mL (95% CI 230.94-367.14) for anti-FHA antibody] than GMTs in Adacel group [22 IU/mL (95% CI 16.05-29.75) for anti-PT antibody; 90 IU/mL (95% CI 64.46-125.39) for anti-FHA antibody]. Similarly, neutralizing anti-PT antibody seroconversion rates were higher in both BioNet vaccine groups than the seroconversion rate in Adacel group. Therefore, VacPertagen induced robust antibody persistence against PT and FHA compared to Adacel in adolescents 1 year after vaccination.

During the **2-, 3-, and 5-year follow up studies**, antibody responses declined in all groups as expected. The trend of immunogenicity comparisons between BioNet aP and Adacel was similar in all follow up studies. Considering anti-PT and anti-FHA IgG GMCs over the 5-year period after vaccination, BioNet aP can induce substantial pertussis antibody levels (for both PT and FHA) as compared to Adacel.

After 5 years, immune responses against pertussis antigens in terms of seroconversion rates as well as GMCs/GMTs induced by BioNet aP were higher than the responses induced by Adacel. At 5 years after vaccination, seroconversion rates were 33%, 95% CI 20-45, n=55 for BioNet aP and 2%, 95% CI 0-6, n=52; $P \leq 0.05$ for Adacel. ELISA anti-PT and anti-FHA GMCs were higher at 5 years after vaccination in the BioNet aP group (33 IU/mL, 95% CI 24.65-43.10 for anti-PT and 70 IU/mL, 95% CI 57.29-86.28 for anti-FHA) than GMCs in Adacel group (GMCs for both PT and FHA were below baseline level: 11 IU/mL, 95% CI 8.78-14.45 for anti-PT and 28 IU/mL, 95% CI 21.16-37.87 for anti-FHA). The neutralizing antibody responses showed a similar pattern over the 5-year period after vaccination. The follow-up studies therefore consistently indicated a more robust antibody persistence after vaccination with VacPertagen compared to Adacel.

Taken together, the immunogenicity data show that a single administration of BioNet aP vaccine (VacPertagen) in healthy adolescent subjects elicits a higher immune response compared to Adacel 28 days after vaccination (primary analysis) and antibody persistence was shown up to 5 years after vaccination. Moreover, ELISA GMCs and neutralizing titres against PT were generally higher after BioNet aP throughout TDA202 and the extension studies. Therefore, at least comparable vaccine efficacy against pertussis can be assumed for a single booster dose of BioNet aP in adolescent subjects.

Study PertADO

The PertADO study was an investigator-driven, single-center, phase 2, observer-blinded randomized controlled trial in aP-primed adolescents in Geneva to assess the immunogenicity and reactogenicity of a novel recombinant aP (r-aP) vaccine including recombinant per- tussis toxin (PT) and filamentous hemagglutinin (FHA) co-administered with tetanus-diphtheria toxoids (Td), compared to a licensed tetanus-diphtheria-aP vaccine containing chemically detoxified PT (cd/Tdap, Boostrix).

Therefore, a very specific situation (**co-administration of Td antigens with VacPertagen, r-aP**) was investigated in this study. Furthermore, the sample size was based on practical considerations and not on a formal statistical power calculation. Immunogenicity evaluations were therefore only descriptive which is acknowledged.

In total, only **62 aP-primed adolescents** (mean age 12.2 years) were randomized and vaccinated with r-aP + Td or cd/Tdap. Baseline characteristics including GMCs were similar between the groups.

Anti-PT GMCs were approximately 2-fold higher after r-aP + Td compared to cd/Tdap 28 days after vaccination. Also, PT-neutralizing GMCs tended to be higher after r-aP + compared to cd/Tdap. The day 28 anti-FHA GMCs were similar in both groups. Day 365 anti-PT (but not PT-neutralizing) GMCs were slightly higher in r-aP + Td vaccinees (26.87 [95% CI, 19.51–37.00] versus 15.75 [95% CI, 10.22–24.27]).

Taken together, study PertADO showed higher immune responses against PT 28 days after r-aP (VacPertagen) + Td compared to cd/Tdap (Boostrix) in adolescent subjects. This marked difference in antibody titres was not seen anymore at Day 365 with slightly higher GMCs. Although the investigated vaccination strategy and low sample size limit possible conclusions, the results provide some additional insights into the immunogenicity of VacPertagen. Considering the Caucasian population primarily investigated in study PertADO, the results are considered reassuring for the MA of VacPertagen in the EU.

Studies in adult subjects

Study TDA206

Study TDA206 was a phase III randomized, observer-blind, active-controlled study to compare the safety and immunogenicity of an investigational combined Tetanus-diphtheria-recombinant acellular pertussis vaccine (BioNet Tdap) and licensed recombinant TdaP vaccine (Boostagen), investigational recombinant monovalent acellular pertussis vaccine (BioNet ap) and licensed recombinant aP vaccine (VacPertagen), and another licensed Tdap vaccine, when administered to healthy adults 18-75 yoa randomized 1:1:1:1:1. Adults were stratified by age: 18-64 yoa (adults) and 65-75 yoa (elderly) at approximately 4:1 ratio from one site in Bangkok, Thailand. The overall study design is acceptable. This trial is considered pivotal in providing evidence for vaccine effectiveness after a single dose in adults including elderly.

The primary **purpose** of this study was to investigate whether BioNet recombinant pertussis vaccines with lower recombinant acellular pertussis dosages (BioNet ap and Tdap; 2 µg PT_{gen} instead of 5 µg in VacPertagen) could be comparable to Adacel in terms of safety and immunogenicity. The main objective of this phase III study was to assess the safety of BioNet ap and Tdap vaccines. In particular, the sample size calculation and the only NI-hypothesis of the trial are formulated for safety only; all immunogenicity points were only addressed descriptively as secondary endpoints, although p-values were reported for these analyses as well.

In total, **750 healthy adults** (600 participants aged 18-64 years and 150 participants aged 65-75 years) were enrolled and randomized into the 5 vaccine groups (150 participants *per* vaccine group). Eligibility criteria are acceptable. Demographic characteristics were similar across vaccine groups across age groups. The mean age of participants, aged 18-64 years old and aged 65-75 years was 38.21 years and 69.48 years, respectively.

Overall, 74 subjects were included into the **immunogenicity analyses** of Adacel as compared to 75 subjects in the BioNet vaccine groups (299 participants aged 18-64 years and 75 participants aged 65-75 years were analysed in total). Hence, the number of immunogenicity blood samples taken was similar between vaccine groups in TDA202 but the sample size for the elderly population can be considered as rather low, especially after stratification by vaccine groups (15 subjects examined after vaccination with VacPertagen in the 65-75 yoa group). Primary immunogenicity evaluations were performed approximately 1 month after vaccination. Furthermore, antibody persistence was investigated after 1 year. The immunogenicity analyses rely on validated methods and are overall acceptable.

Immune responses to 4 different aP_{gen} BioNet vaccines and the comparator Adacel were investigated in adults (including the elderly). The comparison of the VacPertagen vs. the Adacel group is of paramount interest since only VacPertagen is part of the current MAA.

In all participants, **seroconversion** rate of **anti-PT antibody at 28 days** after vaccination compared to baseline was higher in VacPertagen [100.00% (95% CI 95.20-100.00)] than the seroconversion rate in Adacel group [74.32% (95% CI 62.84 - 83.78)]. In participants aged 18-64 years, seroconversion rate of anti-PT antibody at 28 days after vaccination compared to baseline was higher in the VacPertagen group than in the Adacel group with a difference in seroconversion rate of 22.03% (95%CI 13.33-34.19). Also in participants aged **65-75 years**, seroconversion rate in the VacPertagen group was higher than those in the Adacel group with a difference in seroconversion rate of 40.00% (95%CI 15.24-64.61). Furthermore, the 3 other BioNet vaccines (containing aP_{gen} in same/lower

amounts and/or also include Td antigens) also induced higher seroconversion rates 28 days after vaccination compared to Adacel which is reassuring considering the low sample size for the elderly, especially after stratification into vaccine groups.

At 28 days after vaccination, **GMCs** for **anti-PT-IgG antibodies** were higher in the VacPertagen group [371.83 IU/mL (95% CI 292.76-472.25)] compared to the Adacel group [50.84 IU/mL (95% CI 39.26-65.84)]. Anti-PT GMCs following VacPertagen vaccination were overall comparable in both age groups 28 days after vaccination. In all participants, seroconversion rate of **PT neutralizing** antibodies at 28 days after vaccination compared to baseline were higher in the VacPertagen group [96.00% (95% CI 88.75-99.17)] than in the Adacel group [64.86% (95% CI 52.89-75.61). Also when stratified by age, there was a trend for higher seroconversion rates of PT neutralizing antibodies and GMTs at 28 days after vaccination in the VacPertagen group compared to the Adacel group. In all participants, **seroconversion** rate of **anti-FHA antibody** at 28 days after was similar in all vaccine groups e.g. VacPertagen group [97.33% (95% CI 90.70-99.6)] vs Adacel group [93.24% (95% CI 84.93-97.77). Also when stratified by age, seroconversion rates of anti-FHA antibody at 28 days after vaccination compared to baseline were similar. At 28 days after vaccination, **GMCs** for **anti-FHA-IgG antibodies** were higher in the VacPertagen group [451.62 IU/mL (95% CI 373.46-546.12)] than in the Adacel group [207.58 IU/mL (95% CI 171.33-251.50)].

After 1 year, GMCs for anti-PT-IgG and for anti-FHA-IgG antibodies were higher in the VacPertagen group compared to Adacel. At 336 days after vaccination, GMCs for anti-PT-IgG antibodies after VacPertagen were 70.90 IU/mL (95% CI 52.30-96.11) and 14.12 IU/mL (95% CI 10.98-18.14) after Adacel. The GMCs for anti-FHA-IgG antibodies were 142.46 IU/mL (95% CI 114.38-177.44) for VacPertagen and 85.26 IU/mL (95% CI 69.77-104.18) for Adacel.

After 3 years, GMCs for anti-PT-IgG and for anti-FHA-IgG antibodies were higher in the VacPertagen group compared to Adacel. The GMCs for anti-PT-IgG antibodies after VacPertagen were 43.00 IU/mL (95% CI 31.47-58.75) and 8.75 IU/mL (95% CI 6.55-11.70) after Adacel. The GMCs for anti-FHA-IgG antibodies were 92.39 IU/mL 95% CI 71.15-119.98) for VacPertagen and 52.36 IU/mL (95% CI 42.05-65.20) for Adacel.

Taken together, results of study TDA206 suggest that a single administration of VacPertagen in healthy adult subjects (including the elderly) elicits a considerable immune response after 28 days in comparison to Adacel although only descriptive immunogenicity analyses were provided. The totality of evidence indicates at least a comparable antibody response following vaccination with VacPertagen compared to Adacel across the investigated age groups. Only 75 elderly subjects (65-75 yoa) were included in the immunogenicity analyses (15 subjects vaccinated with VacPertagen) which limits possible conclusions in this age group. Nevertheless, it is acknowledged that VacPertagen (and other BioNet aP_{gen} vaccines; partly containing only less than half the amount of PT than VacPertagen) generally induced higher antibody responses to PT and FHA also in the 65-75 yoa group.

Study Pertaprime-01

The **aim** of the phase II-III study Pertaprime-01 was to demonstrate the safety and non-inferior immunogenicity for pertussis antigens of VacPertagen compared to the licensed Boostrix (randomized 2:1) in healthy young Australian adults aged 18 to 30 years old, with NI-margin for seroconversion (primary endpoint) being set at 10%. In addition, the study explored whether responses to pertussis booster vaccine in young adulthood may be affected by the nature of pertussis vaccines (wP or chemically inactivated aP (aP_{chem}) vaccines) in early life.

In total, **102 subjects were enrolled**. Baseline characteristics were balanced between groups. The mean age of subjects was 20.6 years and 84.3% were of Caucasian ethnicity. Antibody analyses was

conducted in 99 participants (65 participants were from VacPertagen and 34 from Boostrix vaccine group) for PP population. According to the Applicant's information, 68 participants were required for adequate power of the NI comparison. The sample size was reduced from V1 to V6 of the protocol, but neither the change in the assumption nor the calculation of the sample size could be properly followed. The sample size is overall considered rather small. As in the main studies, no multiplicity correction was implemented in the trial and the results of the secondary endpoints are to be interpreted descriptively.

At 28 days after vaccination, seroconversion rates of **anti-PT antibodies** were not inferior, and even higher, in the VacPertagen group: 98.46% (95% CI: 91.72-99.96), with respect to the Boostrix group: 82.35% (95% CI: 65.47- 93.24). At 28 days after vaccination, anti-PT antibody GMC was higher in VacPertagen [132.70 IU/mL (95% CI: 100.21-175.73)] as compared to Boostrix [57.42 IU/mL (95% CI: 42.01-78.48] vaccine group. Also, PT neutralizing GMT was higher in VacPertagen [146.97 IU/mL (95% CI: 107.95-200.07)] as compared to Boostrix [69.32 IU/mL (95% CI: 50.24-95.66] vaccine group.

Seroconversion rates of **anti-FHA antibody** concentrations were higher in VacPertagen group 87.69% (95% CI: 77.18-94.53) than the seroconversion rates in Boostrix group 61.76% (95% CI: 43.56-77.83)]. At 28 days after vaccination, anti-FHA antibody GMC was numerically higher in VacPertagen [290.65 IU/mL (95% CI: 245.16-344.58)] as compared to Boostrix [237.63 IU/mL (95% CI: 185.15-304.98] vaccine group. In total, 34 subjects in the VacPertagen group had been previously vaccinated with acellular (PT_{chem}-based) vaccines. In this limited dataset, the overall impression of higher immune responses after vaccination with VacPertagen compared to Boostrix was **unchanged when subjects were stratified according to pre-immunisation with aP or wP** vaccines.

After 1 year, GMCs for anti-PT-IgG were higher in the VacPertagen group and GMCs for anti-FHA-IgG antibodies were similar compared to Boostrix. GMCs for anti-PT-IgG antibodies after VacPertagen were 27.61 IU/mL (95% CI: 18.80-40.53) and 12.51 IU/mL (95% CI: 8.39-18.64) after Boostrix. The GMCs for anti-FHA-IgG antibodies were 79.77 IU/mL (95% CI: 65.57-97.06) for VacPertagen and 92.65 IU/mL (95% CI: 70.14-122.39) for Boostrix.

Taken together, study Pertaprime-01 provided additional evidence for VacPertagen vaccine effectiveness in Australian adult subjects, by showing overall higher immune responses to PT and FHA after VacPertagen than after the licensed Boostrix, 28 days after vaccination.

Studies in pregnant subjects

Data supporting an indication for passive protection against pertussis in infancy following maternal immunisation during pregnancy were obtained in 2 trials, namely TDA204 and TDA207, and in one observational study (PerMIT) conducted post-approval in Thailand. The Applicant further cited a recently published study conducted in Uganda (no CSR available, WOMANPOWER study, Nakabembe et al, 2025).

Study TDA207

TDA207 was a phase II, observer-blind, randomized, active-controlled vaccine trial conducted at two sites in Thailand in 240 healthy pregnant women. Eligible females were randomized equally (n=40 per group) into one of the following vaccine groups: $ap1_{gen}$, $ap2_{gen}$, $aP5_{gen}$ (= VacPertagen), $Tdap2_{gen}$, $TdaP5_{gen}$ (= Boostagen), and $Tdap_{chem}$ (=licensed comparator Adacel).

Blood samples were taken from all pregnant women at baseline (Day 0) before vaccination, at Day 28, and during delivery. To assess maternal antibody transfer, cord blood samples were collected at delivery and if not possible (n=2 in the VacPertagen group, n=1 in the Adacel group) from the neonate within 72 hours after delivery. To evaluate the response to primary infant immunisation, blood samples

were collected from infants at 2 and 7 months of age (Visit 4 and Visit 7, respectively). Only a subset of 20 mother-infant pairs (50%) in each vaccine group (total of 120 pairs) have been tested for PT-neutralizing serum antibody by CHO assay.

The primary study objective was to assess the immunogenicity of a single dose vaccination of BioNet recombinant pertussis vaccines relative to $Tdap_{chem}$ based on the geometric mean of pertussis toxin (PT)-specific serum antibodies concentration (GMC) measured 28 days following immunisation in maternal participants, as determined by ELISA. The primary endpoint did not include a non-inferiority hypothesis of VacPertagen ($aP5_{gen}$) vs Adacel. For this application, the comparison of the VacPertagen vs. the $Tdap_{chem}$ group (licensed comparator Adacel) is considered of relevance. Therefore, the discussion of the results will focus on these two treatment groups.

From a methodological perspective, the study presents several shortcomings that make it more similar to a well-planned, well-conducted proof-of-concept study rather than a pivotal trial for the proposed indication. Of note, this is the pivotal study to support the proposed indication of "passive protection against pertussis in early infancy", but newborn/infant endpoints are only secondary endpoints. They are not included in the strategy to address multiplicity.

At D28, 1 and 3 maternal participants were excluded from ELISA immunogenicity analysis for PP population of the VacPertagen and Adacel groups, respectively. At delivery, 5 participants in each the VacPertagen and Adacel groups were excluded from ELISA immunogenicity analysis for PP population. The most common reason for exclusion was caused by no available blood sample at visit 3 due to delivery at another hospital. While 5 exclusions of in total 40 participants per group (VacPertagen and Adacel) is rather substantial, the exclusions were at least balanced between the two groups and it is acknowledged that delivery at another hospital cannot be prevented.

At 28 days after vaccination, the **anti-PT** geometric mean concentration (GMC) was higher in the VacPertagen group compared to the $Tdap_{chem}$ group (153.98 IU/mL [95% CI: 107.51 – 220.55] vs. 29.53 IU/mL [95% CI: 20.20 – 43.16], respectively). The geometric mean fold rise (GMFR) in relation to the respective baseline titres was 32.71 in the VacPertagen group and 5.2 in the $Tdap_{chem}$ group, indicating a markedly stronger response for VacPertagen compared to the licensed comparator. Comparable results were shown for **PT neutralizing GMTs**, with a GMFR between baseline and Day 28 of 31.23 in the VacPertagen group and 4.22 in the $Tdap_{chem}$ group. The anti-PT **seroconversion rate** at Day 28, defined as the percentage of maternal participants with a $\geq$ 4-fold increase in anti-PT antibodies concentrations, was higher in the VacPertagen group with 94.87% [95% CI: 82.68 – 99.37] vs. the $Tdap_{chem}$ group with 62.16% [95% CI: 44.76-77.54].

A stronger response with respect to **anti-FHA** titres was also noted in the VacPertagen group (GMFR 13.37), compared to the $Tdap_{chem}$ group (GMFR 6.76). The seroconversion rate for anti-FHA antibody concentrations was 82.05% [66.47 – 92.46] in the $aP5_{gen}$ group and 67.57% [50.21 – 81.99] in the $Tdap_{chem}$ group.

At delivery, the antibody titres **in maternal blood samples** did slightly decrease, but the overall picture was comparable to the results of D28. The **anti-PT** GMC in the VacPertagen group was 119.75 [95% CI: 81.59 – 175.74] and 22.23 [95% CI: 15.23 – 32.45] in the $Tdap_{chem}$ group. Importantly, comparable results were also observed **in cord blood** (or neonatal blood within 72 hours after birth) at delivery (141.40 IU/mL [95% CI: 94.70 – 211.12] vs. 27.09 IU/mL [95% CI: 18.21 – 40.31], respectively). The GMFRs at delivery (maternal samples)/baseline were 25.77 in the VacPertagen group and 3.68 in the $Tdap_{chem}$ group. In line with the earlier sampling time point, a comparable trend with respect to **anti-PT neutralizing GMTs** was also observed at delivery (GMFR aP5gen: 19.95; GMFR $Tdap_{chem}$: 2.14). The seroconversion rate remained higher in the VacPertagen group, with 94.29% [95% CI: 80.84 – 99.30] in the $aP5_{gen}$ group and 44.12% [95% CI: 27.19 – 62.11].

For **anti-FHA**, the GMFR at delivery remained numerically higher in the VacPertagen group (GMFR: 9.11), compared to the $Tdap_{chem}$ group (5.35). With respect to seroconversion rates of anti-FHA concentrations at delivery, no notable difference was noted, as the confidence intervals were overlapping ($aP5_{gen}$: 74.29% [95% CI: 56.74 – 87.51]; $Tdap_{chem}$: 61.76% [95% CI: 43.56 – 77.83]).

When comparing maternal blood samples at delivery to the **cord blood samples**, numerically higher anti-PT GMCs were noted in cord blood for both relevant treatment groups ($aP5_{gen}$ GMCR: 1.18; $Tdap_{chem}$ GMCR: 1.23), which was also shown for anti-FHA GMCs.

Anti-PT titres were comparable between participants who were vaccinated in the **2nd trimester vs. the 3rd trimester**, for both VacPertagen and for $Tdap_{chem}$. However, for both treatment groups there was a consistent trend for numerically higher GMCs in participants who were vaccinated in the 3rd trimester.

For the **2-month blood samples in infants**, the reported antibody levels (GMC in IU/mL) for anti-PT IgG, anti-FHA IgG and PT neutralising antibodies remained higher in the VacPertagen group (anti-PT IgG: 60.46 [38.92 – 93.92], anti-FHA IgG: 83.74 [63.46 – 110.51], PT-neutralising: 54.04 [25.12 – 116.24]), compared to the $Tdap_{chem}$ group (anti-PT IgG: 10.74 [7.65 – 15.07], anti-FHA IgG: 33.12 [22.34 – 49.09], PT-neutralising: 7.43 [4.89 – 11.28]).

At the **7-month time point**, infants already received their primary immunisation with three doses (at months 2, 4 and 6) of mostly whole cell (but in a few cases also acellular) pertussis antigen containing vaccines. In contrast to the earlier time point, the anti-PT GMCs at 7 months were **lower** in the VacPertagen group (=$aP5_{gen}$, 17.77 IU/ml [95% CI: 13.06-24.18]) vs. the licensed comparator Adacel ($Tdap_{chem}$, 40.98 IU/ml [26.59-63.15]). Between 7 months and 2 months, anti-PT IgG GMCs were **significantly lower** for VacPertagen [GMCR 0.29 (95% CI: 0.17-0.50), p<0.0001] and significantly higher for Adacel [GMCR 3.63 (95% CI: 1.87-7.04), p=0.0003] which indicate a **decrease** in anti-PT IgGs between month 2 (pre-primary timepoint) and month 7 (after three primary vaccinations) for VacPertagen, while infants of mothers who received Adacel during pregnancy showed an increase. The strong blunting effect of VacPertagen was also observed for the percentages of infants with a 4-fold or higher response in anti-PT antibody concentrations measured by ELISA at 7 months of age, in reference to 2 months of age (VacPertagen: 9.1% [95% CI: 1.92-24.33), Adacel: 51.5% [95% CI: 33.54-69.20]).

Overall, the presented data of Study TDA207 show that a single administration of VacPertagen in pregnant women elicits a considerable immune response, as described for the blood samples taken at 28 days after vaccination, at delivery, and by data from infant samples 2 months after delivery. The study did not include formal non-inferiority testing against the licensed comparator vaccine Adacel but the presented descriptive results clearly suggest that VacPertagen can be expected to elicit an immune response which is at least as pronounced as the licensed comparator. However, the results also suggest a very pronounced "blunting" effect, characterised as a reduced response to primary immunisation in infants. See the respective discussion on this issue further below.

Study PerMIT

The Pertussis Maternal Immunisation in Thailand (PerMIT) trial was a post-marketing (both VacPertagen and Boostagen are authorized in Thailand) observational study in healthy pregnant women between 18-40 years of age who previously received VacPertagen (aP_{gen}), Boostagen ($TdaP_{gen}$) or Td vaccine during pregnancy. The purpose of this study was to investigate the maternal antibody transfer to neonates of the recombinant acellular pertussis vaccines (VacPertagen, Boostagen). Therefore, a sample of umbilical cord blood (6 mL) was taken at delivery.

The primary objective was to determine geometric mean concentrations of PT-neutralizing, anti-PT and anti-FHA IgG titers in cord sera.

Immunogenicity analysis was performed in the Immunogenicity Analysis Population, which includes enrolled participants with cord blood sample and no major protocol deviations that were determined to potentially interfere with the immunogenicity assessment. The study results were descriptively analysed and no missing data imputation techniques were used, which is acceptable for the purpose of this study.

The study protocol describes an anticipated enrolment of ~500 eligible subjects, with ~400 subjects expected to be exposed, and a ratio of exposed (VacPertagen or Boostagen) to unexposed (licensed Td vaccine) cohort of 4:1. At the end of the study, a total of 620 subjects were screened and 584 were enrolled, with comparable numbers of subjects exposed to VacPertagen (N=256) or Boostagen (N=252), and fewer subjects exposed to licensed Td vaccine (N=76). Fewer than planned unexposed subjects were enrolled, which is more relevant for the interpretation of the safety data, since no immune response to PT and FHA is expected in this group.

The most frequent protocol deviation was that cord blood was not collected (n=105). Immunogenicity analyses (ELISA, PT-neutralization assay) were performed in 199 participants in the VacPertagen group, 200 participants in the Boostagen group, and 54 participants in the Td vaccine group.

The mean age at enrollment was 29.7 (±5.1) years.

The immunogenicity results show clearly higher GMCs in cord blood in the exposed groups compared to unexposed participants with respect to anti-PT IgG (VacPertagen: 206.1 IU/mL [164.3-258.6], Boostagen: 153.1 IU/mL [129.1-181.5], Td-only: 6.5 IU/mL [4.9-8.8]), anti-FHA IgG (VacPertagen: 217.2 IU/mL [184-256.4], Boostagen: 232 IU/mL [199-270.6], Td-only: 12.2 IU/mL [8.6-17.4]), and PT-neutralization (VacPertagen: 105.3 IU/mL [81.7-135.8], Boostagen: 81.5 IU/mL [66.4-100], Td-only: 3.8 IU/mL [2.8-5.1]).

No notable differences for neither VacPertagen nor Boostagen were seen when anti-PT IgG were compared between participants vaccinated in the 2nd vs. the 3rd trimester.

Overall, the presented results of this observational study investigating immunogenicity in cord sera clearly show maternal transfer of antibodies as determined by anti-PT IgG, anti-FHA IgG and PT-neutralization assays.

Studies TDA203 + TDA204

Additional supportive information is available from 2 randomized controlled observer-blind clinical trials in non-pregnant females at childbearing age (TDA203) and pregnant women (TDA204). Data from study TDA203 are actually more relevant for the adult indication than for the maternal immunisation indication but are described here because the Applicant pooled the study results for the primary analysis of TDA204. A total of 250 participants were recruited for Study TDA203, and 400 participants in Study TDA204 (participants equally randomized into 5 treatment groups in both studies). For interpretation of the data, it should be pointed out that VacPertagen, which includes 5 µg of pertussis toxoid (PT) and 5 µg of filamentous hemagglutinin (FHA), was not investigated. Studies TDA203 and subsequently TDA204 actually investigated a lower dosed version of VacPertagen (1 µg PT antigen, 1 µg FHA antigen) and different dose levels of the Applicant's Tdap vaccine, in comparison to the licensed comparator Boostrix. Of note, one study vaccine (Boostagen, licensed in Thailand) includes the same amount of PT and FHA antigens (5 µg each) as VacPertagen, but additionally tetanus and diphtheria antigens.

The primary analysis of study TDA204 was made by combining results of studies TDA204 and TDA203, while all other secondary endpoints were only measured in the TDA204 population. This strategy was

not fully justified in the protocol. The attempt to formally demonstrate the non-inferiority of Boostagen versus Boostrix by assessing the non-inferiority of the immune response specific to PT induced by Boostagen versus Boostrix in the pooled per-protocol population of pregnant women and women of childbearing age in Thailand (TDA203-TDA204) is not appropriate. In addition, there were various methodological issues and results of study TDA203 were likely already known at time of planning which defies the equipoise assumption in setting up the study hypothesis. Due to the supportive nature of this study, as well as to the consistent results across the trials, these concerns are not further pursued, and the data generated in this trial are only interpreted descriptively.

The detailed data are described in the results section above. In short, the presented data from studies TDA203 and TDA204 show that the investigated BioNet vaccines induced a considerable response with respect to anti-PT antibodies at Day 28 after vaccination and at delivery, which were often higher than the licensed comparator Boostrix, depending on the endpoint and formulation (see results section above). With respect to anti-FHA antibodies, there was a trend for significantly lower responses compared to Boostrix. These data need to be interpreted with caution due to the different antigen formulations/concentrations administered and several methodological concerns.

Data of the study ***WoMANPOWER*** indicate a booster effect of PT- and FHA- specific immune responses following vaccination with Boostagen in both HIV positive and HIV negative pregnant women. Importantly, these data also indicate an interference of the maternal antibodies on the infants' PT-specific immune response expected to be mounted in response to the childhood vaccination (DTwP, 3 doses).

In conclusion, concerning the transfer of maternal antibodies (passive immunisation of newborns), studies TDA207 and TDA204 showed GMCs of anti-PT IgGs in infants of women who received VacPertagen during pregnancy which were at least as high as for the licensed comparators. Passive transfer of maternal antibodies was also confirmed in an observational study (PerMIT). As there is no immune correlate of protection (ICPs) for pertussis, it is not known if the transferred anti-PT and anti-FHA antibodies will be sufficient to protect against (severe) disease. Evidence from an effectiveness study conducted in Denmark (Kildegaard et al 2025) indicate that passively transferred anti-PT antibodies might be sufficient to confer protection against pertussis in newborns. However, uncertainties remain on the magnitude and duration of the assumed protection of newborns. Effectiveness needs to be confirmed post-marketing and the Applicant committed to conduct a real-world effectiveness study in Europe. The respective study protocol should be submitted within 6 months after EC decision.

Importantly, the blunting effect to primary vaccination in infants, as described for study TDA207 further above, was also observed in study TDA204 (7-month and 13-month time points).

Blunting of pertussis response following primary immunisation is known to occur in infants born to women vaccinated with Boostrix and Adacel. This phenomenon has been extensively described in the literature, with difference in the magnitude of interference and in the resolution after the toddler's boost. The quantity and quality of the antibody response after primary vaccination with DTwP or DTaP in infants born to mothers who received a Tdap during pregnancy might be different. Maternal vaccination (transfer of antibodies) might also modulate the cellular immune response induced by the primary vaccination of infants.

Indeed, at least for anti-PT IgGs (less clear for neutralising antibodies based on limited data), studies (TDA207, TDA204) suggest a more pronounced blunting effect with (Td-) VacPertagen compared to the licensed comparators Adacel/Boostrix.

Longitudinal follow-up data presented in the spaghetti plot for Study TDA207 indicate that infants with low anti-PT IgG responses at birth had an increase in levels when comparing anti-PT IgG at month 2

and month 7. While infants with high anti-PT IgG responses at birth had no increase or a decrease in levels when comparing anti-PT IgG at month 2 and month 7. This is consistent with data reported by Knuutila et al. 2023 that investigated the maternal immune response to immunisation during pregnancy (IP) with Boostrix and the effect of IP and pre-existing antibodies on infants' primary vaccine responses in an open-label, non-randomized trial. Reported data show differences in levels of antibody responses to primary vaccination with a DTaPa-HB-IPV-Hib vaccine on the basis of categorization to low and high baseline antibody concentration (cutoff defined by distributing the infants in proportional increases based on antibody concentrations measured pre-vaccination at three months of age). Data indicate a comparatively higher blunting of the responses to primary vaccination in infants with higher circulating levels of anti-PT, anti-FHA and anti-PRN pre-vaccination.

The PIs of vaccines currently approved for passive immunisation suggest that the blunting effect may not be clinically significant. The Applicant provided a recent publication of an effectiveness study conducted in Australia suggesting slightly lower VE point estimates for the third dose of infant pertussis vaccine among maternally vaccinated compared to unvaccinated infants, but the study authors did not observe higher rates of pertussis infection (Regan et al. 2023). Post toddler booster dose immunogenicity data available for vaccines currently approved for passive immunisation indicate effective priming of the immune system. The more pronounced effect with (Td-) VacPertagen of actually decreasing titres between month 2 (=pre-priming) and month 7 (=one month after primary vaccination consisting of 3 DTwP/DTaP injections) may be relevant for decision making by NITAGs. Post-toddler booster dose data are lacking for (Td-) VacPertagen. Available data are shown in section 5.1 of the SmPC and section 4.4 includes a warning that maternal antibodies interfere with induction of PT-specific immune response to primary immunisation with DTwP/DTaP in infants born to women vaccinated with VacPertagen during pregnancy. Furthermore, the Applicant presented plans how to further investigate the impact of maternal immunisation on primary vaccination and boosting in infants.

2.6.7. Conclusions on the clinical efficacy

The available immunogenicity data consistently showed elevated binding antibody concentrations and neutralizing titres against PT (and often-increased FHA binding antibody concentrations) after a single dose of VacPertagen (aP_{gen}) compared to the licensed (chemically inactivated) Tdap comparators Adacel and Boostrix in adolescents and adults (including pregnant women), previously vaccinated with pertussis vaccines. Limited descriptive data indicate longer persistence of anti-PT and anti-FHA responses as compared to Adacel.

There is no established serological correlate of protection for pertussis and the protective potential of the elicited humoral immune response cannot be predicted. Based on a clear trend for higher immunogenicity across studies throughout the available clinical trials, however, a certain level of vaccine efficacy against pertussis can be assumed for VacPertagen for the targeted booster vaccination of adolescents and adults.

The Applicants also sought an indication for *passive protection against pertussis in early infancy following maternal immunisation during pregnancy*. Overall, based on the robust anti-PT (binding and neutralising) and anti-FHA (binding) antibody levels in infants (at birth and at 2 months of age), the CHMP concluded that it is reasonable to assume that VacPertagen will be effective in the setting of passive protection against pertussis in early infancy following maternal immunisation during pregnancy. However, uncertainties remain on the magnitude and duration of the assumed protection of newborns. Effectiveness will be confirmed post-marketing.

2.6.8. Clinical safety

2.6.8.1. Patient exposure

Table 99. Patient exposure in randomised controlled clinical trials for VacPertagen

Study name	Adolescents	Adults	Elderly	Pregnant women
TDA202	150	-	-	-
PertADO	31	-	-	-
Pertaprime-01	-	68	-	-
TDA206	-	120	30	-
APV301	-	1977	123	-
TDA207	-	-	-	40
Subtotal	181	2165	153	40
TOTAL	2539			

Study APV301

A pivotal, multi-site, phase III, observer blind, randomised, active-controlled vaccine trial in which 2400 healthy adults aged 18 to 75 years were recruited from three sites in Bangkok, Thailand. After obtaining the informed consent and confirmation of eligibility, the participants were randomised in a 7:1 ratio into one of the following vaccine groups:

Vaccine groups	N
Pertagen®	2100
Boostrix™	300
Total	2400

Safety was evaluated based on the incidence of predefined local and systemic solicited adverse reactions occurring within 7 days post-vaccination, as well as unsolicited adverse events (both serious and non-serious) reported within 28 days. Immunogenicity was not assessed in this study.

Objectives

Primary objective: To descriptively evaluate the safety profile of VacPertagen and Boostrix after a single booster dose vaccination by assessing the incidence and nature of all adverse drug reactions (ADRs) in healthy adults.

Secondary objective: To assess the nature, severity, and duration of adverse events (AEs), serious adverse events (SAEs), and adverse drug reactions (ADRs) following a single booster dose of recombinant acellular pertussis (aP) vaccine and Boostrix.

Exploratory objectives: To describe the safety of three batches of recombinant acellular pertussis (aP) vaccine after a single booster dose vaccination. To assess the safety of recombinant acellular pertussis (aP) vaccine in pooled populations from all randomised controlled trials with aP vaccine including APV301, TDA206, TDA202, TDA207, Pertaprime-01 and any relevant studies.

Participants

As of 8 May 2025, a total of 2400 were enrolled and vaccinated with either VacPertagen (2100 participants) or Boostrix (300 participants). A summary of the demographic characteristics at baseline of the total study population is presented in the Table below.

Table 100. Summary of demographics at baseline in study APV301

Characteristics	
Age; n (%)	
18 - 65 years old	2289 (95.37)
> 65 years old	111 (4.63)
Missing data	0 (0.00)
Sex; n (%)	
Male	945 (39.37)
Female	1455 (60.63)
Missing data	0 (0.00)
Ethnicity; n (%)	
Asian	2400 (100.00)

Safety data is presented in the sections below as follows:

- Solicited and unsolicited adverse events:

  - pooled data for adolescents and adults (including pregnant women). This includes data from RCTs TDA202, TDA206, TDA207, Pertaprime-01 and APV301 and was the basis for identification of ADR. The frequencies reported in 4.8 were adjusted to the highest reported frequency in any of the studies mentioned above.
  - study TDA207 for pregnant women

- Serious adverse events:

  - study TDA202 for adolescents

Pooled non-pregnant adult safety data. This includes data from RCT TDA206, Pertaprime-01 and APV301 safety data from RCT TDA 207 pregnant women Regarding PertADO study, no CSR was provided but a scientific publication. Thus, no data from this study is presented but is discussed in discussion section.

2.6.8.2. Adverse events

Adolescents and adults (including pregnant women)

Table 101. Pooled safety data for solicited AEs reported within 30 minutes post-vaccination following administration of VacPertagen in adolescents and adults (including pregnant women) from RCTs TDA202, TDA206, TDA207, Pertaprime-01 and APV301

Solicited adverse events within 30 minutes post-vaccination			
	MedDRA System Organ Class (SOC)	**Preferred Term (PT)**	**Adolescents and adults (including pregnant women)[1] (12-75 years) n (%) (95% CI)**
Participants with at least one AE			N=2508 439 (17.50) (16.04-19.05)
Local reactions	General disorders and administration site conditions	Injection site pain	N=2508 307 (12.24) (10.98-13.59)
		Injection site erythema	N=2508 5 (0.20) (0.06-0.46)
		Injection site swelling	N=2358 1 (0.04) (0.00-0.24)
		Injection site induration	N=2508 1 (0.04) (0.00-0.22)
		Injection site pruritus	N=2290 30 (1.31) (0.89-1.86)
Systemic reactions		Malaise	N=2508 17 (0.68) (0.40-1.08)
		Fatigue	N=2508 39 (1.56) (1.09-2.09)
		Chills	N=2508 8 (0.32) (0.14-0.63)
		Pyrexia	N=2508 0 (0.00) (0.00-0.15)
	Musculoskeletal and connective tissue disorders	Myalgia	N=2508 171 (6.82) (5.86-7.88)
		Arthralgia	N=2508 30 (1.20) (0.81-1.70)
	Nervous system disorders	Headache	N=2508 66 (2.63) (2.04-3.34)
	Gastrointestinal disorders	Vomiting	N=2508 2 (0.08) (0.01-0.29)
		Nausea	N=2358 7 (0.30) (0.12-0.61)

Notes:

[1] Data from TDA202 (adolescents), TDA206 (adults), TDA207 (pregnant women), Pertaprime-01 (adults) and APV301 (adults).

- N is the total number of participants in each group
- n is number of participants with at least one adverse event
- Two-sided 95% CIs were computed using the Clopper-Pearson method

Table 102. Pooled safety data for solicited AEs reported within 7 days following administration of VacPertagen in adolescents and adults (including pregnant women) from RCTs TDA202, TDA206, TDA207, Pertaprime-01 and APV301

Solicited adverse events within 7 days after vaccination			
	MedDRA System Organ Class (SOC)	**Preferred Term (PT)**	**Adolescents and adults (including pregnant women)[1] (12-75 years) n (%) (95% CI)**
Participants with at least one AE			N=2507 1523 (60.75) (58.81-62.67)
Local reactions	General disorders and administration site conditions	Injection site pain	N=2507 1256 (50.10) (48.12-52.08)
		Injection site erythema	N=2507 22 (0.88) (0.55-1.33)
		Injection site induration	N=2507 23 (0.92) (0.58-1.37)
		Injection site swelling	N=2357 9 (0.38) (0.17-0.72)
		Injection site pruritus	N=2289 98 (4.28)

			(3.49-5.19)
Systemic reactions		Malaise	N=2507 292 (11.65) (10.42-12.97)
		Fatigue	N=2507 386 (15.40) (14.01-16.87)
		Chills	N=2507 78 (3.11) (2.47-3.87)
		Pyrexia	N=2507 26 (1.04) (0.68-1.52)
	Musculoskeletal and connective tissue disorders	Myalgia	N=2507 775 (30.91) (29.11-32.76)
		Arthralgia	N=2507 239 (9.53) (8.41-10.75)
	Nervous system disorders	Headache	N=2507 425 (16.95) (15.50-18.48)
	Gastrointestinal disorders	Vomiting	N=2507 34 (1.36) (0.94-1.89)
		Nausea	N=2357 55 (2.33) (1.76-3.03)

Notes:

[1] Data from TDA202 (adolescents), TDA206 (adults), TDA207 (pregnant women), Pertaprime-01 (adults) and APV301 (adults).

- N is the total number of participants in each group
- n is number of participants who have at least one adverse event
- Two-sided 95% CIs were computed using the Clopper-Pearson method.

Table 103. Pooled safety data for all unsolicited AEs (related and unrelated) reported within 28 days following administration of VacPertagen in adolescents and adults (including pregnant women) from RCTs TDA202, TDA206, TDA207, Pertaprime-01 and APV301

All unsolicited AEs (related and unrelated) within 28 days after vaccination		
MedDRA System Organ Class (SOC)	Preferred Term (PT)	Adolescents and adults (including pregnant women)[1] (N=2507) n (%) (95% CI)
Participants with at least one AE		315 (12.56) (11.29-13.93)
Blood and lymphatic system disorders	Iron deficiency anaemia	1 (0.04) (0.00-0.22)
	Lymphadenitis	3 (0.12) (0.02-0.35)
	Lymphadenopathy	3 (0.12) (0.02-0.35)
Cardiac disorders	Palpitation	1 (0.04) (0.00-0.22)
Ear and labyrinth disorders	Cerumen impaction	1 (0.04) (0.00-0.22)
	Ear congestion	1 (0.04) (0.00-0.22)
	Tinnitus	1 (0.04) (0.00-0.22)
Endocrine disorders	Thyroiditis	1 (0.04) (0.00-0.22)
Eye disorders	Eye pain	1 (0.04) (0.00-0.22)
	Swelling of eyelid	1 (0.04) (0.00-0.22)
	Vision blurred	2 (0.08) (0.01-0.29)
Gastrointestinal disorders	Abdominal distension	2 (0.08) (0.01-0.29)
	Abdominal pain	2 (0.08) (0.01-0.28)
	Aphthous ulcer	3 (0.12) (0.02-0.35)
	Cheilitis	1 (0.04) (0.00-0.22)
	Colitis	1 (0.04) (0.00-0.22)
	Dental caries	2 (0.08) (0.01-0.29)
	Diarrhoea	14 (0.56) (0.31-0.94)
	Dyspepsia	3 (0.12) (0.02-0.35)

	Enteritis	1 (0.04) (0.01-0.22)
	Food poisoning	1 (0.04) (0.00-0.22)
	Gastrooesophageal reflux disease	2 (0.08) (0.01-0.29)
	Haemorrhoids	1 (0.04) (0.00-0.22)
	Mouth ulceration	1 (0.04) (0.00-0.22)
	Nausea	6 (0.24) (0.09-0.52)
	Toothache	4 (0.16) (0.04-0.41)
	Vomiting	2 (0.08) (0.01-0.29)
General disorders and administration site conditions	Chills	2 (0.08) (0.01-0.29)
	Fatigue	16 (0.64) (0.37-1.03)
	Induration	1 (0.04) (0.00-0.22)
	Influenza like illness	5 (0.20) (0.06-0.46)
	Injection site bruising	1 (0.04) (0.00-0.22)
	Injection site erythema	2 (0.08) (0.01-0.29)
	Injection site haematoma	2 (0.08) (0.01-0.29)
	Injection site induration	8 (0.32) (0.14-0.63)
	Injection site pain	71(2.83) (2.22-3.56)
	Injection site pruritus	5 (0.20) (0.06-0.46)
	Injection site swelling	4 (0.16) (0.04-0.41)
	Malaise	8 (0.32), (0.14-0.63)
	Pyrexia	5 (0.20) (0.06-0.46)
	Vessel puncture site haematoma	1 (0.04) (0.00-0.22)
Immune system disorders	Allergy to arthropod bite	1 (0.04)

		(0.00-0.22)
	Food allergy	1 (0.04) (0.00-0.22)
Infections and infestations	Bronchitis	1 (0.04) (0.00-0.22)
	COVID-19	4 (0.16) (0.04-0.41)
	Candida infection	1 (0.04) (0.00-0.22)
	Conjunctivitis	1 (0.04) (0.00-0.22)
	Cystitis	1 (0.04) (0.00-0.22)
	Fungal infection	1 (0.04) (0.00-0.22)
	Gastroenteritis	2 (0.08) (0.01-0.29)
	Gingivitis	3 (0.12) (0.02-0.35)
	Hordeolum	1 (0.04) (0.00-0.22)
	Influenza	4 (0.16) (0.04-0.41)
	Laryngitis	1 (0.04) (0.00-0.22)
	Lower respiratory tract infection	1 (0.04) (0.00-0.22)
	Nasopharyngitis	20 (0.80) (0.49-1.23)
	Otitis externa	1 (0.04) (0.00-0.22)
	Otitis media	2 (0.08) (0.01-0.29)
	Otitis media acute	1 (0.04) (0.00-0.22)
	Pharyngitis	3 (0.12) (0.02-0.35)
	Pneumonia	1 (0.04) (0.00-0.22)
	Rhinitis	3 (0.12) (0.02-0.35)
	Sexually transmitted disease	1 (0.04) (0.00-0.22)
	Sialoadenitis	1 (0.04) (0.00-0.22)

	Sinusitis	1 (0.04) (0.00-0.22)
	Tonsillitis	1 (0.04) (0.00-0.22)
	Upper respiratory tract infection	15 (0.60) (0.34-0.98)
	Viral infection	4 (0.16) (0.04-0.41)
	Viral upper respiratory tract infection	4 (0.16) (0.04-0.41)
	Vulvovaginal candidiasis	2 (0.08) (0.01-0.29)
Injury, poisoning and procedural complications	Accident	1 (0.04) (0.00-0.22)
	Animal bite	1 (0.04) (0.00-0.22)
	Contusion	2 (0.08) (0.01-0.29)
	Heat exhaustion	1 (0.04) (0.00-0.22)
	Joint dislocation	1 (0.04) (0.00-0.22)
	Ligament sprain	4 (0.16) (0.04-0.41)
	Limb injury	3 (0.12) (0.02-0.35)
	Muscle strain	6 (0.24) (0.09-0.52)
	Occupational exposure to communicable disease	1 (0.04) (0.00-0.22)
	Road traffic accident	2 (0.08) (0.01-0.29)
	Skin abrasion	2 (0.08) (0.01-0.29)
	Stab wound	1 (0.04) (0.00-0.22)
	Thermal burn	2 (0.08) (0.01-0.29)
Musculoskeletal and connective tissue disorders	Arthralgia	17 (0.68) (0.40-1.08)
	Costochondritis	1 (0.04) (0.00-0.22)
	Myalgia	44 (1.76) (1.28-2.35)
	Pain in extremity	5 (0.20)

		(0.06-0.46)
	Patellofemoral pain syndrome	1 (0.04) (0.00-0.22)
Nervous system disorders	Dizziness	4 (0.16) (0.04-0.41)
	Dysgeusia	1 (0.04) (0.00-0.22)
	Headache	45 (1.79) (1.31-2.39)
	Hypoaesthesia	1 (0.04) (0.00-0.22)
	Ischaemic stroke	1 (0.04) (0.00-0.22)
	Migraine	5 (0.20) (0.06-0.46)
	Tension headache	5 (0.20) (0.06-0.46)
Pregnancy, puerperium and perinatal conditions	Gestational diabetes	1 (0.04) (0.00-0.22)
	Threatened labour	1 (0.04) (0.00-0.22)
Psychiatric disorders	Bruxism	1 (0.04) (0.00-0.22)
	Depression	2 (0.08) (0.01-0.29)
	Insomnia	2 (0.08) (0.01-0.29)
Reproductive system and breast disorders	Dysmenorrhoea	16 (0.64) (0.37-1.03)
	Endometriosis	1 (0.04) (0.00-0.22)
	Intermenstrual bleeding	1 (0.04) (0.00-0.22)
	Menstruation irregular	1 (0.04) (0.00-0.22)
Respiratory, thoracic and mediastinal disorders	Cough	5 (0.20) (0.06-0.46)
	Oropharyngeal pain	4 (0.16) (0.04-0.41)
	Rhinitis allergic	4 (0.16) (0.04-0.41)
	Rhinorrhoea	3 (0.12) (0.02-0.35)
Skin and subcutaneous tissue disorders	Chloasma	1 (0.04) (0.00-0.22)

	Dermatitis contact	1 (0.04) (0.00-0.22)
	Diabetic foot	1 (0.04) (0.00-0.22)
	Night sweats	1 (0.04) (0.00-0.22)
	Rash	2 (0.08) (0.01-0.29)
	Urticaria	3 (0.12) (0.02-0.35)
	Urticaria papular	1 (0.04) (0.00-0.22)
Vascular disorders	Hot flush	1 (0.04) (0.00-0.22)
	Hypertension	1 (0.04) (0.00-0.22)

Notes:
[1] Data from TDA202 (adolescents), TDA206 (adults), TDA207 (pregnant women), Pertaprime-01 (adults) and APV301 (adults).
- N is the total number of participants in each group
- n is number of participants who have at least one adverse event
- Two-sided 95% CIs were computed using the Clopper-Pearson method.

Table 104. Pooled safety results for related unsolicited AEs reported within 28 days following administration of VacPertagen in adolescents and adults (including pregnant women) from RCTs TDA202, TDA206, TDA207, Pertaprime-01 and APV301

All unsolicited AEs (related) within 28 days after vaccination		
MedDRA System Organ Class (SOC)	Preferred Term (PT)	Adolescents and adults (including elderly and pregnant women)[1] (N=2507) n (%) (95% CI)
Participants with at least one AE		126 (5.03) (4.20-5.96)
Blood and lymphatic system disorders	Lymphadenitis	2 (0.08) (0.01-0.29)
	Lymphadenopathy	2 (0.08) (0.01-0.29)
Cardiac disorders	Palpitation	1 (0.04) (0.00-0.22)

Eye disorders	Eye pain	1 (0.04) (0.00-0.22)
Gastrointestinal disorders	Diarrhoea	1 (0.04) (0.00-0.22)
	Enteritis	1 (0.04) (0.00-0.22)
	Nausea	3 (0.12) (0.02-0.35)
General disorders and administration site conditions	Chills	1 (0.04) (0.00-0.22)
	Fatigue	8 (0.32) (0.14-0.63)
	Influenza like illness	1 (0.04) (0.00-0.22)
	Induration	1 (0.04) (0.00-0.22)
	Injection site bruising	1 (0.04) (0.00-0.22)
	Injection site erythema	2 (0.08) (0.01-0.29)
	Injection site haematoma	2 (0.08) (0.01-0.29)
	Injection site induration	7 (0.28) (0.11-0.57)
	Injection site pain	71 (2.83) (2.22-3.56)
	Injection site pruritus	5 (0.20) (0.06-0.46)
	Injection site swelling	4 (0.16) (0.04-0.41)
	Malaise	6 (0.24) (0.09-0.52)
	Pyrexia	3 (0.12) (0.02-0.35)
Infections and infestations	Pharyngitis	1 (0.04) (0.00-0.22)
Musculoskeletal and connective tissue disorders	Arthralgia	12 (0.48) (0.25-0.83)
	Myalgia	35 (1.40) (0.97-1.94)
Nervous system disorders	Headache	10 (0.40) (0.19-0.73)
	Ischaemic stroke	1 (0.04) (0.00-0.22)
	Rash	1 (0.04)
Skin and subcutaneous tissue disorders		(0.00-0.22)
	Urticaria	1 (0.04) (0.00-0.22)

Notes:

[1] Data from TDA202 (adolescents), TDA206 (adults), TDA207 (pregnant women), Pertaprime-01 (adults) and APV301 (adults).

- N is the total number of participants in each group
- n is number of participants who have at least one adverse event
- Two-sided 95% CIs were computed using the Clopper-Pearson method.

Pregnant women and infants

TDA 207

Table 105. TDA 207 study: Local and systemic post-immunisation reactions in maternal subjects during 7 days after vaccination by vaccine groups

Local and Systemic Reactions		Visit 1 (within 30 minutes after vaccination)						Day 0 – Day 7					
		BioNet recombinant $ap1_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $ap2_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $aP5_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $Tdap2_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $TdaP5_{gen}$ (N = 40) n (%) (95% CI)	Comparator $Tdap_{Adacel}$ (N = 40) n (%) (95% CI)	BioNet recombinant $ap1_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $ap2_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $aP5_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $Tdap2_{gen}$ (N = 40) n (%) (95% CI)	BioNet recombinant $TdaP5_{gen}$ (N = 40) n (%) (95% CI)	Comparator $Tdap_{Adacel}$ (N = 40) n (%) (95% CI)
Local	Pain	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	31 (77.50) (61.55 - 89.16)	27 (67.50) (50.87 - 81.43)	27 (67.50) (50.87 - 81.43)	36 (90.00) (76.34 - 97.21)	34 (85.00) (70.16 - 94.29)	33 (82.50) (67.22 - 92.66)
	Redness	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	1 (2.50) (0.06 - 13.16)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	1 (2.50) (0.06 - 13.16)	0 (0.00) (0.00 - 8.81)
	Swelling	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)
	Induration	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	1 (2.50) (0.06 - 13.16)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)
	Pruritus	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	3 (7.50) (1.57 - 20.39)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)	1 (2.50) (0.06 - 13.16)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)
Systemic	Headache	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	6 (15.00) (5.71 - 29.84)	6 (15.00) (5.71 - 29.84)	8 (20.00) (9.05 - 35.65)	6 (15.00) (5.71 - 29.84)	9 (22.50) (10.84 - 38.45)	9 (22.50) (10.84 - 38.45)
	Fatigue	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	8 (20.00) (9.05 - 35.65)	6 (15.00) (5.71 - 29.84)	13 (32.50) (18.57 - 49.13)	9 (22.50) (10.84 - 38.45)	11 (27.50) (14.60 - 43.89)	10 (25.00) (12.69 - 41.20)
	Arthralgia	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)	1 (2.50) (0.06 - 13.16)	3 (7.50) (1.57 - 20.39)	5 (12.50) (4.19 - 26.80)
	Chills	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	1 (2.50) (0.06 - 13.16)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	1 (2.50) (0.06 - 13.16)	2 (5.00) (0.61 - 16.92)	0 (0.00) (0.00 - 8.81)
	Malaise	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	3 (7.50) (1.57 - 20.39)	1 (2.50) (0.06 - 13.16)	3 (7.50) (1.57 - 20.39)	6 (15.00) (5.71 - 29.84)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)
	Myalgia	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	11 (27.50) (14.60 - 43.89)	10 (25.00) (12.69 - 41.20)	9 (22.50) (10.84 - 38.45)	11 (27.50) (14.60 - 43.89)	12 (30.00) (16.56 - 46.53)	10 (25.00) (12.69 - 41.20)
	Vomiting	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	1 (2.50) (0.06 - 13.16)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	0 (0.00) (0.00 - 8.81)	1 (2.50) (0.06 - 13.16)
	Nausea	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)	1 (2.50) (0.06 - 13.16)	2 (5.00) (0.61 - 16.92)	2 (5.00) (0.61 - 16.92)	3 (7.50) (1.57 - 20.39)
	Fever (≥37.5°C)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)	0 (0.00) (0.00 - 8.81)

Table 106. TDA 207 study: Maternal subjects with adverse events by severity during Day 0 to Day 28 post-vaccination, by vaccine groups

Study Number Phase	TDA207 Phase II						
Vaccine groups	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet $aP5_{gen}$ (Pertagen®)	BioNet recombinant $Tdap2_{gen}$	BioNet $TdaP5_{gen}$ (Boostagen®)	Comparator $Tdap_{Adacel}$	Total
Number of subjects per group, N	40	40	40	40	40	40	240
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
With one or more adverse events							
By severity							
Mild	6 (15.00)	8 (20.00)	7 (17.50)	9 (22.50)	7 (17.50)	8 (20.00)	45 (18.75)
Moderate	1 (2.50)	1 (2.50)	2 (5.00)	2 (5.00)	2 (5.00)	1 (2.50)	9 (3.75)
Severe	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Potential life-threatening	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
With vaccine-related adverse events							
By severity							
Mild	2 (5.00)	1 (2.50)	1 (2.50)	1 (2.50)	1 (2.50)	1 (2.50)	7 (2.92)
Moderate	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)	1 (0.42)
Severe	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Potential life-threatening	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Discontinued due to an adverse event	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)

Table 107. TDA 207 study: Maternal subjects with adverse events (medically attended adverse events and adverse events leading to withdrawal) by severity from Day 0 - Visit 3 (Delivery visit) post-vaccination by vaccine groups.

Study Number Phase	TDA207 Phase II						
Vaccine groups	BioNet recombinant ap1$_{gen}$	BioNet recombinant ap2$_{gen}$	BioNet aPS$_{gen}$ (Pertagen*)	BioNet recombinant Tdap2$_{gen}$	BioNet TdaPS$_{gen}$ (Boostagen*)	Comparator Tdap$_{chem}$	Total
Number of subjects per group, N	40	40	40	40	40	40	240
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
With medically attended adverse event							
By severity							
Mild	9 (22.50)	5 (12.50)	8 (20.00)	11 (27.50)	9 (22.50)	7 (17.50)	49 (20.42)
Moderate	2 (5.00)	1 (2.50)	3 (7.50)	3 (7.50)	4 (10.00)	2 (5.00)	15 (6.25)
Severe	1 (2.50)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	2 (0.83)
Potential life-threatening	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Discontinued due to an adverse event							
By severity							
Mild	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Moderate	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Severe	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Potential life-threatening	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)

* also referred to as Visit 1 (vaccination)

Table 708. TDA 207 Summary of specific complications during pregnancy and delivery of mother (Safety population)

	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet recombinant $aP5_{gen}$	BioNet recombinant $Tdap2_{gen}$	BioNet recombinant $TdaP5_{gen}$	Comparator $Tdap_{chem}$
	(N=40) n (%)	(N=40) n (%)	(N=40) n (%)	(N=40) n (%)	(N=40) n (%)	(N=40) n (%)
Delivery mode						
Normal	23 (57.50)	24 (60.00)	15 (37.50)	18 (45.00)	21 (52.50)	12 (30.00)
Caesarean section	17 (42.50)	14 (35.00)	25 (62.50)	22 (55.00)	18 (45.00)	28 (70.00)
Emergency Caesarean (C)-section	8 (20.00)	7 (17.50)	18 (45.00)	15 (37.50)	7 (17.50)	12 (30.00)
Elective Caesarean (C)-section	9 (22.50)	7 (17.50)	7 (17.50)	7 (17.50)	10 (25.00)	16 (40.00)
No data	0 (0.00)	2 (5.00)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)
Complication during pregnancy						
Yes	8 (20.00)	5 (12.50)	7 (17.50)	11 (27.50)	10 (25.00)	6 (15.00)
Pregnancy loss or stillbirth	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Preterm delivery (<37 weeks of gestation)	4 (10.00)	3 (7.50)	4 (10.00)	4 (10.00)	6 (15.00)	2 (5.00)
Preterm premature rapture of membranes	2 (5.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)
Pregnancy-induced hypertension	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Pre-eclampsia/eclampsia	2 (5.00)	0 (0.00)	1 (2.50)	3 (7.50)	1 (2.50)	0 (0.00)
Intrauterine growth restriction (IUGR)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Obstetric hemorrhage	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)

Gestational diabetes	0 (0.00)	1 (2.50)	1 (2.50)	3 (7.50)	2 (5.00)	1 (2.50)
Other	2 (5.00)	3 (7.50)	2 (5.00)	3 (7.50)	3 (7.50)	3 (7.50)
Chorioamnionitis	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)
COVID-19 infection	2 (5.00)	2 (5.00)	0 (0.00)	2 (5.00)	0 (0.00)	1 (2.50)
False labour pain	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)
Fetal movements decreased	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)
Oligohydramnios	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)
Otitis media	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)
Preterm labour without delivery	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	1 (2.50)
Threatened premature labour	0 (0.00)	1 (2.50)	1 (2.50)	1 (2.50)	0 (0.00)	0 (0.00)
No	32 (80.00)	35 (87.50)	33 (82.50)	29 (72.50)	30 (75.00)	34 (85.00)
Complication during delivery						
Yes	8 (20.00)	8 (21.05)	17 (42.50)	11 (27.50)	9 (21.05)	8 (20.51)
Postpartum hemorrhage	1 (2.50)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Maternal fever or infection	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Other	8 (20.00)	7 (17.50)	17 (42.50)	11 (27.50)	8 (20.00)	8 (20.00)
Breech Presentation	0 (0.00)	1 (2.50)	1 (2.50)	1 (2.50)	0 (0.00)	0 (0.00)
Cephalopelvic disproportion	1 (2.50)	2 (5.00)	9 (22.50)	3 (7.50)	1 (2.50)	3 (7.50)
Chorioamnionitis	0 (0.00)	0 (0.00)	1 (2.50)	1 (2.50)	0 (0.00)	0 (0.00)
Fetal distress	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)
Fetal growth restriction	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)
Fetal macrosomia	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Footling breech	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)
Forceps Extraction due to Fetal Mal position	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)
Non-reassuring fetal heart rate	1 (2.50)	0 (0.00)	2 (5.00)	0 (0.00)	1 (2.50)	2 (5.00)
Non-reassuring fetal status	0 (0.00)	1 (2.50)	1 (2.50)	1 (2.50)	1 (2.50)	0 (0.00)
Oligohydramnios	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	1 (2.50)
Placenta previa totalis	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)
Preeclampsia	1 (2.50)	0 (0.00)	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)
Severe preeclampsia	1 (2.50)	0 (0.00)	1 (2.50)	1 (2.50)	0 (0.00)	0 (0.00)
Unfavorable cervix	0 (0.00)	1 (2.50)	0 (0.00)	2 (5.00)	0 (0.00)	1 (2.50)
Unprogress of labour	3 (7.50)	1 (2.50)	1 (2.50)	3 (7.50)	4 (10.00)	0 (0.00)
Vacuum-assisted vaginal delivery due to Maternal effort	0 (0.00)	1 (2.50)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
No	32 (80.00)	30 (75.00)	23 (57.50)	29 (72.50)	30 (75.00)	31 (77.50)
No data / Unknown	0 (0.00)	2 (5.00)	0 (0.00)	0 (0.00)	1 (2.50)	1 (2.50)

Table 109. Summary of infants with prematurity, small for gestational age and low birth weight at Visit 3 (Delivery visit) (Full analysis population)

Infant participant status	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet recombinant $aP5_{gen}$	BioNet recombinant $Tdap2_{gen}$	BioNet recombinant $TdaP5_{gen}$	Comparator $Tdap_{chem}$	Total
	(N=40)	(N=40)	(N=40)	(N=40)	(N=40)	(N=40)	(N=240)
Infants with one or more criteria: n (%)	4 (10.00)	3 (7.50)	4 (10.00)	4 (10.00)	7 (17.50)	2 (5.00)	24 (10.00)
1.Prematurity (<37 weeks of gestational age)	4 (10.00)	3 (7.50)	4 (10.00)	4 (10.00)	6 (15.00)	2 (5.00)	23 (9.58)
2.Small for gestational age (SGA)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	2 (5.00)	0 (0.00)	3 (1.25)
3.Low birth weight (< 2000 grams)	0 (0.00)	0 (0.00)	0 (0.00)	1 (2.50)	2 (5.00)	1 (2.50)	4 (1.67)

2.6.8.3. Serious adverse event/deaths/other significant events

Adverse events of special interest were not defined in the clinical study program for VacPertagen.

Serious adverse events

Adolescents

TDA 202 (main study)

During 28 days post-vaccination, only one SAE was reported in the BioNet aP group(open wound after road accident). The SAE was assessed as not related to vaccination. A summary of SAEs from day 29 to 365 is presented below.

Table 110. TDA202: Summary of serious adverse events by MedDRA Term during Day 29 – Day 336 post-vaccination by vaccine groups

MedDRA System Organ class	Preferred Term	BNA aP		BNA TdaP		Adacel[a]		Total	
		Number of events	Number of subjects	Number of events	Number of subjects	Number of events	Number of subjects	Number of events	Number of subjects
		N=5	N=147	N=6	N=147	N=2	N=148	N=13	N=442
		n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Gastrointestinal disorders	Enteritis	1(20.00)	1(0.68)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Gastrointestinal disorders	Gastroenteritis	1(20.00)	1(0.68)	1(16.67)[a]	1(0.68)	0(0.00)	0(0.00)	2(15.38)	2(0.45)
Gastrointestinal disorders	Helicobacter gastritis	0(0.00)	0(0.00)	1(16.67)[a]	1(0.68)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Infections and infestations	Dengue fever	0(0.00)	0(0.00)	2(33.33)	2(1.36)	2(100.00)	2(1.35)	4(30.77)	4(0.90)
Infections and infestations	Herpangina	0(0.00)	0(0.00)	1(16.67)	1(0.68)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Injury, poisoning and procedural complications	Multiple fractures	1(20.00)	1(0.68)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Injury, poisoning and procedural complications	Snake bite	1(20.00)	1(0.68)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Injury, poisoning and procedural complications	Substance-induced psychotic disorder	0(0.00)	0(0.00)	1(16.67)	1(0.68)	0(0.00)	0(0.00)	1(7.69)	1(0.23)
Injury, poisoning and procedural complications	Wound	1(20.00)	1(0.68)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(7.69)	1(0.23)

*. 2 SAEs occurred in the same subject.

Non-pregnant adults

Table 111. Summary of pooled safety results for SAEs reported within 28 days following vaccination with VacPertagen in adult subjects (including the elderly) from RCTs - TDA206, Pertaprime-01 and APV301

SAEs within 28 days after vaccination					
MedDRA System Organ Class (SOC)	Preferred Term (PT)	Adults and the elderly[1] (18-75 years) (N=2317) n (%) (95% CI)	Elderly (65-75 years)[2] (N=123) n (%) (95% CI)	Adults (18-64 years)[3, 7] (N=2096) n (%) (95% CI)	Elderly (65-75 years)[4] (N=153) n (%) (95% CI)
Participants with at least one SAE		3 (0.13) (0.03-0.38)	0 (0.00) (0.00-2.95)	2 (0.10) (0.01-0.34)	0 (0.00) (0.00-2.38)
Nervous system disorders	Ischaemic stroke[5]	1 (0.04) (0.00-0.24)	0 (0.00) (0.00-2.95)	1 (0.05) (0.00-0.27)	0 (0.00) (0.00-2.38)
Psychiatric disorders	Depression[6]	1 (0.04) (0.00-0.24)	0 (0.00) (0.00-2.95)	0 (0.00), 0 (0.00-0.18)	0 (0.00), 0 (0.00-2.38)
Skin and subcutaneous tissue disorders	Diabetic foot[5]	1 (0.04) (0.00-0.24)	0 (0.00) (0.00-2.95)	1 (0.05) (0.00-0.27)	0 (0.00) (0.00-2.38)

[1] Data from **TDA206** and **APV301** (adults including the elderly) and **Pertaprime-01** (adult participants)
[2] Data from **APV301** (65-75 years)
[3] Data from **APV301** (18-64 years) and **TDA206** (18-64 years)
[4] Data from **APV301** (65-75 years) and **TDA206** (65-75 years)

[5] In the **APV301** study, SAEs were actively monitored throughout the entire 28-day study period. Two SAEs were reported, both in recipients of VacPertagen: one case of inpatient hospitalisation due to ischaemic stroke in a >60-year-old participant, and one case of diabetic foot requiring inpatient hospitalisation in a >40-year-old participant. The ischaemic stroke was assessed by the investigator as possibly related to the study vaccine. However, the sponsor considered the event as vaccine-unrelated based on the established safety profile of inactivated pertussis vaccines and the presence of plausible alternative aetiology specifically, longstanding untreated hypertension and neuroimaging findings consistent with chronic small vessel cerebrovascular disease. The case of diabetic foot was assessed as vaccine-unrelated by both the investigator and the sponsor. Both cases had resolved, and the participants completed the study.

[6] In the **Pertaprime-01** study, SAEs were actively monitored up to 6 months post-vaccination, and vaccine-related SAEs were monitored up to 1 year. During the 28-day follow-up period, one vaccine-unrelated SAE was reported in a >20-year-old recipient of VacPertagen: an inpatient hospitalisation due to depression. An additional vaccine-unrelated SAE (biliary colic, considered medically significant) was reported five months after vaccination in a >18-year-old participant who received VacPertagen. Both events had resolved. No SAEs were considered related to VacPertagen at any time during the 1-year follow-up period.

[7] In the **TDA206** study, SAEs were actively monitored throughout the entire 1-year study period. No SAEs following vaccination with VacPertagen were reported within the initial 28 days post vaccination. One vaccine-unrelated SAE was reported five months after vaccination in a >40-year-old participant who received VacPertagen: an inpatient hospitalisation due to breast mass. This event resolved and the participant completed the study. No SAEs were considered related to VacPertagen during the course of the study.
- Two-sided 95% CIs were computed using the Clopper-Pearson method.

Adult pregnant women and their infants

TDA 207

Table 112. TDA 207- Summary of maternal participants with serious adverse events (SAEs) from Day 0 – Visit 3 (Delivery visit) post vaccination among vaccine groups (Safety population)

AE Summary	BioNet recombinant $ap1_{gen}$	BioNet recombinant $ap2_{gen}$	BioNet recombinant $aP5_{gen}$	BioNet recombinant $Tdap2_{gen}$	BioNet recombinant $TdaP5_{gen}$	Comparator $Tdap_{chem}$	Total
	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)
Number of participants (N)	40	40	40	40	40	40	240
Serious adverse events							
Participants with one or more SAEs	9 (22.50), 12 (10.84-38.45)	5 (12.50), 7 (4.19-26.80)	9 (22.50), 11 (10.84-38.45)	11 (27.50), 14 (14.60-43.89)	7 (17.50), 10 (7.34-32.78)	5 (12.50), 5 (4.19-26.80)	46 (19.17), 59 (14.39-24.72)
Participants with vaccine-related SAEs	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)
Participants with AE leading to hospitalization	6 (15.00), 7 (5.71-29.84)	2 (5.00), 3 (0.61-16.92)	5 (12.50), 5 (4.19-26.80)	8 (20.00), 10 (9.05-35.65)	6 (15.00), 8 (5.71-29.84)	5 (12.50), 5 (4.19-26.80)	32 (13.33), 38 (9.30-18.30)
Participants with AE leading to death	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)
Participants with medically significant events	4 (10.00), 5 (2.79-23.66)	4 (10.00), 4 (2.79-23.66)	5 (12.50), 6 (4.19-26.80)	5 (12.50), 6 (4.19-26.80)	2 (5.00), 2 (0.61-16.92)	0 (0.00), 0 (0.00-0.00)	20 (8.33), 23 (5.16-12.58)
Participants with SAE leading to withdrawal	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)	0 (0.00), 0 (0.00-0.00)

Note:
n is number of participants with event and E is numbers of events

Table 113. TDA207- Infant participants with serious adverse events at delivery visit among vaccine groups (safety population)

Adverse Event	BioNet recombinant ap1$_{gen}$	BioNet recombinant ap2$_{gen}$	BioNet recombinant aP5$_{gen}$	BioNet recombinant Tdap2$_{gen}$	BioNet recombinant TdaP5$_{gen}$	Comparator Tdap$_{chem}$	Total
	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)
Number of Participants (N)	40	40	40	40	40	40	240
Participants withone or more serious adverse events	8 (20.00), 11	6 (15.00), 6	14 (35.00), 19	8 (20.00), 11	9 (22.50), 19	5 (12.50), 9	50 (20.83), 75
	(9.05-35.65)	(5.71-29.84)	(20.63-51.68)	(9.05-35.65)	(10.84-38.45)	(4.19-26.80)	(15.88-26.53)
Death	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Life-threatening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Inpatient's hospitalization / prolongation of existing hospital	6 (15.00), 7	2 (5.00), 2	11 (27.50), 14	3 (7.50), 5	6 (15.00), 10	4 (10.00), 5	32 (13.33), 43
	(5.71-29.84)	(0.61-16.92)	(14.60-43.89)	(1.57-20.39)	(5.71-29.84)	(2.79-23.66)	(9.30-18.30)
Persistent or significant disability or incapacity	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	1 (0.42), 1
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.06-13.16)	(0.00-0.00)	(0.01-2.30)
Congenital anomaly or birth defect in the offspring of participant	0 (0.00), 0	0 (0.00), 0	2 (5.00), 2	1 (2.50), 1	1 (2.50), 1	2 (5.00), 4	6 (2.50), 8
	(0.00-0.00)	(0.00-0.00)	(0.61-16.92)	(0.06-13.16)	(0.06-13.16)	(0.61-16.92)	(0.92-5.36)
Medically significant AE	3 (7.50), 4	4 (10.00), 4	3 (7.50), 3	5 (12.50), 5	4 (10.00), 7	0 (0.00), 0	19 (7.92), 23
	(1.57-20.39)	(2.79-23.66)	(1.57-20.39)	(4.19-26.80)	(2.79-23.66)	(0.00-0.00)	(4.83-12.09)
Neonatal blood screening abnormalities	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	1 (0.42), 1
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.06-13.16)	(0.00-0.00)	(0.01-2.30)
Hearing loss detected through neonatal screening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	1 (0.42), 1
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.06-13.16)	(0.00-0.00)	(0.01-2.30)

Table 114. TDA207- Summary of serious adverse events by MedDRA term starting from day 0 – visit 4 (Study End of mothers) among vaccine groups of mothers (safety population)

MedDRA System Organ class	Preferred Term	BioNet recombinant ap1$_{gen}$	BioNet recombinant ap2$_{gen}$	BioNet recombinant aP5$_{gen}$	BioNet recombinant Tdap2$_{gen}$	BioNet recombinant TdaP5$_{gen}$	Comparator Tdap$_{chem}$	Total
		n (%), E	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E
Infections and infestations	COVID-19	3 (7.50), 3	3 (7.50), 3	3 (7.50), 3	5 (12.50), 5	2 (5.00), 2	2 (5.00), 2	18 (7.50), 18
	COVID-19 pneumonia	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	1 (2.50), 1	0 (0.00), 0	3 (1.25), 3
	Endometritis	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.42), 1
Pregnancy, puerperium and perinatal conditions	Amniotic cavity infection	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	2 (0.83), 2
	False labour	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	1 (0.42), 1
	Postpartum haemorrhage	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.42), 1
	Postpartum sepsis	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.42), 1
	Pre-eclampsia	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	2 (5.00), 2	0 (0.00), 0	0 (0.00), 0	3 (1.25), 3
	Premature delivery	3 (7.50), 3	3 (7.50), 3	1 (2.50), 1	2 (5.00), 2	3 (7.50), 3	0 (0.00), 0	12 (5.00), 12
	Premature labour	1 (2.50), 1	0 (0.00), 0	4 (10.00), 4	2 (5.00), 2	4 (10.00), 4	3 (7.50), 3	14 (5.83), 14
	Preterm premature rupture of membranes	2 (5.00), 2	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	3 (1.25), 3
	Threatened labour	0 (0.00), 0	1 (2.50), 1	1 (2.50), 1	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	3 (1.25), 3
Reproductive system and breast disorders	Uterine inflammation	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	1 (0.42), 1

Note:
n is number of participants with event and E is number of events

Table 115. Summary of serious adverse events in infants born to mothers vaccinated during pregnancy by MedDRA term starting during day 3 (delivery visit) until visit 7 (7 months after delivery) among vaccine groups

MedDRA System Organ class	Preferred Term	BioNet recombinant aplgen	BioNet recombinant ap2$_{gen}$	BioNet recombinant aP5$_{gen}$	BioNet recombinant Tdap2$_{gen}$	BioNet recombinant TdaP5$_{gen}$	Comparator Tdap$_{chem}$	Total
		n (%), E	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E
Number of Participants (N)		40	38	39	39	35	37	228
Participants with one or more serious adverse events		11 (27.50), 15 (14.60-43.89)	7 (18.42), 8 (7.74-34.33)	16 (41.03), 25 (25.57-57.90)	11 (28.21), 12 (15.00-44.87)	15 (42.86), 25 (26.32-60.65)	7 (18.92), 11 (7.96-35.16)	67 (29.39), 96 (23.56-35.76)
Blood and lymphatic system disorders	Anaemia	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	1 (0.44), 1
	Jaundice neonatal	0 (0.00), 0	2 (5.26), 2	2 (5.13), 2	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	4 (1.75), 4
	Polycythaemia	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	1 (0.44), 1
Congenital, familial and genetic disorders	Congenital diaphragmatic hernia	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Congenital hydronephrosis	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	2 (0.88), 2
	Congenital megacolon	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Ventricular septal defect	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.70), 1	1 (0.44), 1
Ear and labyrinth disorders	Deafness unilateral	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	1 (0.44), 1
Gastrointestinal disorders	Diarrhoea	0 (0.00), 0	1 (2.63), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Necrotising enterocolitis neonatal	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Pylorospasm	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
General disorders and administration site conditions	Fever neonatal	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
Hepatobiliary disorders	Jaundice neonatal	2 (5.00), 3	1 (2.63), 1	4 (10.26), 5	0 (0.00), 0	6 (17.14), 6	1 (2.70), 1	14 (6.14), 16
Infections and infestations	Bacterial infection	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	1 (0.44), 1
	COVID-19	1 (2.50), 1	1 (2.63), 1	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	2 (5.41), 2	5 (2.19), 5
	COVID-19 pneumonia	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	1 (2.86), 1	0 (0.00), 0	2 (0.88), 2
	Congenital pneumonia	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Croup infectious	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.70), 1	1 (0.44), 1
	Diarrhoea infectious	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	2 (0.88), 2
	Laryngitis	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	1 (0.44), 1
	Nasopharyngitis	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Omphalitis	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	1 (2.70), 1	2 (0.88), 2
	Pharyngitis	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.70), 1	1 (0.44), 1
	Pneumonia	0 (0.00), 0	0 (0.00), 0	2 (5.13), 2	0 (0.00), 0	1 (2.86), 1	0 (0.00), 0	3 (1.32), 3
	Pneumonia aspiration	1 (2.50), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Pneumonia respiratory syncytial viral	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	2 (5.71), 2	1 (2.70), 1	3 (1.32), 3
	Sepsis neonatal	2 (5.00), 2	0 (0.00), 0	4 (10.26), 4	2 (5.13), 2	1 (2.86), 1	1 (2.70), 1	10 (4.39), 10
	Streptococcal sepsis	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
Injury, poisoning and procedural complications	Exposure to SARS-CoV-2	0 (0.00), 0	1 (2.63), 1	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	2 (0.88), 2
Pregnancy, puerperium and perinatal conditions	Low birth weight baby	1 (2.50), 1	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	2 (5.71), 2	0 (0.00), 0	4 (1.75), 4
	Neonatal pneumothorax	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
	Premature baby	4 (10.00), 4	2 (5.26), 2	4 (10.26), 4	3 (7.69), 3	4 (11.43), 4	1 (2.70), 1	18 (7.89), 18
Renal and urinary disorders	Renal aplasia	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1
Respiratory, thoracic and mediastinal disorders	Nasopharyngitis	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (2.70), 1	1 (0.44), 1
	Neonatal respiratory distress syndrome	0 (0.00), 0	0 (0.00), 0	1 (2.56), 1	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	1 (0.44), 1

2.6.8.4. Laboratory findings

Clinical laboratory tests of haematology, biochemistry and urinalysis were not performed in studies TDA 202, TDA 206 or TDA 207. Data on vital signs and physical examination were provided for the study visits at day 7 and day 28 for TDA 202, TDA 206 and TDA 207 (also day 336 for study TDA 202).

2.6.8.5. In vitro biomarker test for patient selection for safety

Not applicable

2.6.8.6. Safety in special populations

Elderly subjects

Safety data in elderly subjects is presented separated by study and pooled (studies TDA206 and APV301)

TDA206

Table 116. TDA 206- Summary of number of participants with adverse events by severity started during Day 0 - Day 28 post-vaccination of participants aged 65-75 years old among vaccine groups in study TDA206

Adverse Events	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	Total
	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E	n (%), E
Number of participants (N)	30	30	30	30	30	150
with one or more adverse events by severity						
Mild	6 (20.00), 6	5 (16.67), 6	8 (26.67), 17	7 (23.33), 9	9 (30.00), 17	35 (23.33), 55
Moderate	2 (6.67), 3	1 (3.33), 1	0 (0.00), 0	0 (0.00), 0	1 (3.33), 1	4 (2.67), 5
Severe	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Potential life-threatening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
with vaccine-related adverse events by severity						
Mild	1 (3.33), 1	0 (0.00), 0	1 (3.33), 1	1 (3.33), 1	1 (3.33), 1	4 (2.67), 4
Moderate	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Severe	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Potential life-threatening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
discontinued due to an adverse event by severity						
Mild	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Moderate	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Severe	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
Potential life-threatening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0

Table 117. Participants with serious adverse events during Day 0 - Day 28 post-vaccination of participants aged 65-75 years old among vaccine groups in study TDA206

Adverse Event	BioNet Recombinant ap	BioNet Recombinant Tdap	Pertagen®	Boostagen®	Adacel®	Total
	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)	n (%), E (95% CI)
Number of Participants (N)	30	30	30	30	30	150
Participants with one or more serious adverse events	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Death	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Life-threatening	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Inpatient's hospitalization / prolongation of existing hospitalization	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Persistent or significant disability or incapacity	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Congenital anomaly or birth defect in the offspring of participants	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)
Medically significant AE	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0	0 (0.00), 0
	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)	(0.00-0.00)

APV301

Table 118. Solicited AEs reported within 7 days following vaccination with VacPertagen in elderly subjects from APV301 study, by System Organ Class and Preferred Term

Solicited AEs within 7 days after vaccination		
MedDRA System Organ Class	Preferred Term	Elderly subjects (aged 65 to 75 years) N=123 n (%) (95% CI)
Participants with at least one reaction		39 (31.71) (23.61-40.71)
General disorders and administration site conditions	Injection site pain	30 (24.39) (17.10-32.95)
	Injection site erythema	1 (0.81) (0.02-4.45)
	Injection site swelling	0 (0.00) (0.00-2.95)
	Injection site induration	0 (0.00) (0.00-2.95)
	Injection site pruritus	8 (6.50) (2.85-12.41)
	Chills	3 (2.44) (0.51-6.96)
	Fatigue	9 (7.32) (3.40-13.44)
	Malaise	9 (7.32) (3.40-13.44)
	Pyrexia	2 (1.63) (0.20-5.75)
Nervous system disorders	Headache	9 (7.32) (3.40-13.44)
Musculoskeletal and connective tissue disorders	Arthralgia	8 (6.50) (2.85-12.41)
	Myalgia	17 (13.82) (8.26-21.20)
Gastrointestinal disorders	Vomiting	3 (2.44) (0.51-6.96)
	Nausea	4 (3.25) (0.89-8.12)

Table 119. Related unsolicited AEs reported within 28 days following vaccination with VacPertagen in elderly subjects from APV301 study, by System Organ Class and Preferred Term

Related unsolicited AEs within 28 days after vaccination		
MedDRA System Organ Class	Preferred Term	Elderly subjects (aged 65 to 75 years) N=123 n (%) (95% CI)
Participants with at least one AE		3 (2.44) (0.51-6.96)
Cardiac disorders	Palpitations	1 (0.81) (0.02-4.45)
General disorders and administration site conditions	Chills	1 (0.81) (0.02-4.45)
	Malaise	1 (0.81) (0.02-4.45)
Musculoskeletal and connective tissue disorders	Myalgia	1 (0.81) (0.02-4.45)

Pooled data TDA206 and APV301

Table 120. Summary of AEs by age group (<65 vs ≥65 years of age) following vaccination with VacPertagen from pooled data of studies TDA206 and APV301

Safety outcomes	Adults aged <65 years (N=2096)[a] n (%) (95 % CI)	Adults aged ≥65 years (N=153)[b] n (%) (95 % CI)
Participants with at least one solicited adverse reaction within 7 days post vaccination	1250 (59.64) (57.50-61.75)	53 (34.64) (27.14-42.75)
Participants with any unsolicited AE (related or unrelated) within 28 days post vaccination	215 (10.26) (8.99-11.64)	12 (7.84) (4.12-13.30)
Participants with at least one SAE within 28 days post vaccination	2 (0.10) (0.01-0.34)	0 (0.00) (0.00-2.38)
Participants with vaccine-related unsolicited AE within 28 days post vaccination	97 (4.63) (3.77-5.62)	4 (2.61) (0.72-6.56)
Participants with vaccine-related SAEs within 28 days post vaccination[c]	1 (0.05) (0.00-0.27)	0 (0.00) (0.00-2.38)
Participants with vaccine-related SAEs within 1 year post vaccination[d]	0 (0.00) (0.00-0.00)	0 (0.00) (0.00-0.00)

[a] Data from APV301 (18-64 years) and TDA206 (18-64 years)
[b] Data from APV301 (65-75 years) and TDA206 (65-75 years)
[c] Data from APV301 study in which SAEs were collected and monitored for 28 days.
[d] Data from TDA206 study in which SAEs were collected and monitored throughout the entire one-year study period.

2.6.8.7. Immunological events

Please refer to the discussion on immunology as surrogate of efficacy of this vaccine in the clinical pharmacology sections.

2.6.8.8. Safety related to drug-drug interactions and other interactions

No dedicated drug-drug interaction studies were provided.

2.6.8.9. Discontinuation due to adverse events

No discontinuation due to adverse events were reported in any of the studies.

2.6.8.10. Post marketing experience

The Applicant has presented post-authorisation data in Thailand and Singapore. In addition, data from 3 observational studies in pregnant women have been submitted.

Post authorisation studies

Table 121. Tabular overview of post-authorisation studies in non-EU countries

Study ID	Enrolment status Start date Total enrolment/ enrolment goal	Design Control type	Study & control drugs Dose, route of administration and duration Regimen	Population Main inclusion/ exclusion criteria
Healthy pregnant women				
PerMIT	Complete; FSV 21 January 2019, LSV 21 May 2020; 584/500 enrolled pregnant women who prior to the study have received VacPertagen or Boostagen or the control Tdvaccine were enrolled.	Observational, prospective, pregnancy and neonatal safety outcomes	Single IM dose of Recombinant aP (VacPertagen), Recombinant TdaP or licensed Td (comparator) – all prior to the study	Healthy pregnant subjects (18-40 yoa)
PERMIS	Completed; Women who received VacPertagen (0,5 ml, single dose) during pregnancy and delivered between January 2021 and April 2024 in Thailand Safety information for mothers and infants was obtained through systematic review of medical records.	Observational, retrospective, pregnancy and neonatal safety outcomes	Single IM dose of recombinant aP (VacPertagen) Women who received VacPertagen (0,5 ml, single dose) during pregnancy and delivered between January 2021 and April 2024 in Thailand	Healthy pregnant women
Pertagen-MOM	Completed, 585 pregnant women In this observational, retrospective study, safety information was obtained from medical records of pregnant women who received VacPertagen (0.5 ml,	observational, retrospective study, pregnancy and neonatal outcomes	Single IM dose of Recombinant aP (VacPertagen),	Pregnant women

	single dose) as part of routine antenatal care between 2021 and 2023,			

PerMIT: Antibody level in cord sera following immunisation with recombinant acellular pertussis vaccines during pregnancy: a prospective, observational study

A prospective, observational study to descriptively evaluate the safety and maternal antibody transfer to infants based on antibody **in cord sera** following maternal immunisation (one dose of recombinant aP or TdaP or Td vaccine) of pregnant women 18-40 years of age at the time of pregnancy.

Please refer to section 2.6.5. for a description of the methods and results on maternal antibodies transfer. This section only describes the pregnancy outcomes.

Table 122. PerMIT- Summary of pregnancy and neonatal outcome (Delivery day)

Subject status	Recombinant aP vaccine (Pertagen®)	Recombinant TdaP vaccine (Boostagen®)	Td vaccine	Total	P-value
	N = 248	N = 249	N = 75	N = 572	
Pregnancy outcome, n (%)					- [1]
- Live birth	248 (100.00%)	249 (100.00%)	75 (100.00%)	572 (100.00%)	
Full term	239 (96.37%)	230 (92.37%)	68 (90.67%)	537 (93.88%)	
Pre-term	9 (3.63%)	19 (7.63%)	7 (9.33%)	35 (6.12%)	
- Stillbirth	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
- Spontaneous abortion	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
- Elective termination	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
- Fetal death	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
Neonatal outcome					
Birthweight (gram)					0.4608 [4]
- N	248	249	75	572	
- Mean (SD)	3127.38 (432.25)	3108.36 (397.22)	3059.39 (417.29)	3110.18 (415.19)	
- Min/Max	1540-4535	1892-4210	2202-4140	1540-4535	
Apgar score at 1 minute					0.9106 [3]
- N	247*	249	75	571	
- Mean (SD)	8.88 (0.43)	8.84 (0.61)	8.77 (0.85)	8.85 (0.58)	
- Median	9.00	9.00	9.00	9.00	
- Min/Max	5-9	3-9	4-9	3-9	
Apgar score at 5 minute					0.5357 [3]
- N	247*	249	75	571	
- Mean (SD)	9.89 (0.41)	9.87 (0.41)	9.88 (0.33)	9.88 (0.40)	
- Median	10.00	10.00	10.00	10.00	
- Min/Max	6-10	7-10	9-10	6-10	
Neonatal outcome, n (%)					0.1595 [2]
- Normal	230 (92.74%)	220 (88.35%)	64 (85.33%)	514 (89.86%)	
- Low birth weight	13 (5.24%)	16 (6.43%)	8 (10.67%)	37 (6.47%)	
- Birth defect/Congenital anomalies	0 (0.00%)	0 (0.00%)	0 (0.00%)	0 (0.00%)	
- Other	5 (2.02%)	13 (5.22%)	3 (4.00%)	21 (3.67%)	

Table 123. Summary of delivery status (Delivery day)

Subject status	Recombinant aP vaccine (Pertagen®)	Recombinant TdaP vaccine (Boostagen®)	Td vaccine	Total	P-value
	N = 248	N = 249	N = 75	N = 572	
Gestational age at delivery (weeks)					
- N	248	249	75	572	0.2282 [2]
- Mean (SD)	38.36 (1.21)	38.32 (1.22)	38.55 (1.39)	38.37 (1.24)	
- Median	38.00	38.00	39.00	38.00	
- Min/Max	33-41	33-41	35-41	33-41	
Delivery mode					
Vaginal, n (%)	150 (60.48%)	129 (51.81%)	47 (62.67%)	326 (56.99%)	0.1396 [1]
- Normal	135 (90.00%)	120 (93.02%)	39 (82.98%)	294 (90.18%)	
- Abnormal	15 (10.00%)	9 (6.98%)	8 (17.02%)	32 (9.82%)	
Caesarean section, n (%)	98 (39.52%)	120 (48.19%)	28 (37.33%)	246 (43.01%)	0.5957 [1]
- Normal	45 (45.92%)	55 (45.83%)	10 (35.71%)	110 (44.72%)	
- Abnormal	53 (54.08%)	65 (54.17%)	18 (64.29%)	136 (55.28%)	
Cord blood collection performed, n (%)					0.2231 [1]
- No	41 (16.53%)	45 (18.07%)	19 (25.33%)	105 (18.36%)	
- Yes	207 (83.47%)	204 (81.93%)	56 (74.67%)	467 (81.64%)	

Note:
[1] Overall p-value (2-sided) based on Chi-square test
[2] P-value based on Kruskal-Wallis Test

PerMIS-01

This completed, observational, retrospective study assessed pregnancy and neonatal safety outcomes in women who received VacPertagen during pregnancy and delivered between January 2021 and April 2024 in 16 clinical sites in Thailand. Safety information for mothers and infants was obtained through systematic review of medical records. In this observational study, solicited AEs were not collected as it was not part of the study design. However, safety data on pregnancy and neonatal outcomes were collected. Safety endpoints were collected and reported as follows:

- Number and proportion of pregnant women vaccinated with VacPertagen either in the second or third trimester of pregnancy who had full term and preterm (or premature) delivery.
- Number and proportion of pregnant women vaccinated with VacPertagen either in the second or third trimester of pregnancy who had experienced complications during delivery.
- Number and proportion of healthy infants born to mothers who received VacPertagen during pregnancy.
- Number and proportion of not healthy infants (e.g., difficulty of breathing, congenital abnormality and others) born to mothers who received VacPertagen during pregnancy.

A total of 1980 pregnant women were included with a mean maternal age of 29.46 years. The median height and weight are not known. Most participants received vaccination in the third trimester (84.82%) and 15.18% in the second trimester. Concomitant vaccination was common (87.51%), primarily influenza (83.87%). The majority of deliveries occurred in 2023 (52.12%).

A total of 1985 infants were delivered from 1980 pregnant women. The total accounts for five additional infants, comprising one extra infant from twin gestations in three women and two additional infants from a triplet gestation. Demographic data i.e., sex was available for 1979 infants included in the analysis and sex data were unavailable for 6 infants. From a total of 1979 infants, 50.23% were male and 49.77% were female.

Delivery outcome data were available for 1968 women: 54.73% delivered vaginally and 45.27% by caesarean section. Most deliveries were at term (94.31%), with a preterm delivery rate of 5.69%.

Among the 1980 pregnant women who received VacPertagen, complications at delivery data were available for 1978 pregnant women.

A total of 241 complications were reported by 198 pregnant women [10.01%] during delivery. The most commonly reported complication was postpartum haemorrhage [1.87%], followed by meconium-stained amniotic fluid [0.86%], abnormal foetal heart rate pattern (Category II) [0.81%], gestational diabetes mellitus (not insulin treated), gestational hypertension [0.71%], uterine atony [0.61%], immediate postpartum haemorrhage, non-reassuring foetal heart rate pattern, pre-eclampsia [0.30%], late latent syphilis, preeclampsia with severe features, premature rupture of membranes [0.25%], cephalopelvic disproportion, foetal distress, preeclampsia without severe features [0.20%]; birth before arrival at hospital or other healthcare facility, cervical laceration, chronic hypertension, foetal heart rate abnormality, gestational diabetes mellitus, insulin treated, laceration of lower uterine segment, oligohydramnios, pregnancy-induced hypertension, preterm premature rupture of membranes [0.15%]; and intrapartum fever, placenta previa, preterm delivery, prolonged second stage of labour, retained placenta, shoulder dystocia, surgical site infection [0.10%]. Other complications were reported in less than 0.10% pregnant women. Foetal death (one twin) was reported in one pregnant woman (twin pregnancy) (0.05%) since gestational age 22+4 weeks before vaccination.

Of the 1984 infants with available infant outcome, 1676 infants [84.48%] were reported as healthy at birth. Among the 308 infants classified as non-healthy at birth [15.52%], a total of 441 clinical events were reported. The most frequently reported conditions included: difficulty of breathing in 100 cases [5.04%], neonatal jaundice in 79 cases [3.98%], congenital abnormality in 30 cases [1.51%], hypoglycaemia in 25 cases [1.26%], transient tachypnoea of the newborn in 15 cases [0.76%], low birth-weight liveborn infant in 12 cases [0.60%], caput succedaneum in 10 cases [0.50%], hydrocele in 9 cases [0.45%, ABO incompatibility and ankyloglossia in 8 cases [0.40%], hypoxemia in 6 cases [0.30%]; and cephalohematoma, congenital syphilis, glucose-6-phosphate dehydrogenase deficiency, preterm birth, small for gestational age in 5 cases each [0.25%]. Other clinical events were reported in less than 5 (0.25%) infants.

Among the 30 reported cases of congenital abnormalities in infants, no consistent pattern or clustering was identified: 4 cases [0.20%] each of bilateral pes planus and unspecified or unknown abnormalities; 3 cases [0.15%] of gastroschisis; and 1 case [0.05%] each of the following: atrial septal defect (ASD), ASD with homozygous chromosomal syndrome, bilateral congenital curly toe deformity, bilateral congenital talipes equinovarus (clubfoot), cleft lip, cleft lip with cleft palate, congenital overlapping toe deformity, congenital tracheomalacia, foetal supraventricular tachycardia, left-sided microtia, micrognathia with glossoptosis, patent ductus arteriosus with ASD, patent foramen oval, posterior urethral valves, severe early-onset foetal growth restriction, suspected Noonan syndrome, transposition of the great arteries with pulmonary atresia with ventricular septal defect (VSD) with ASD, and unilateral cleft lip and cleft palate.

As part of an exploratory endpoint, preterm delivery rates were compared with national data for women who received the licensed Td vaccine between 2019 and 2020. VacPertagen recipients had a preterm delivery rate of 5.69% (112 preterm delivery out of 1968 delivery outcomes) compared with

13.25% among Td vaccine recipients (82603 preterm delivery out of 623472 delivery outcomes), a difference that was statistically significant ($p < 0.001$).

Pertagen-MOM

The final Statistical analysis report (SAR) has been submitted for Pertagen-MOM with 585 pregnant women vaccinated with VacPertagen.

In this observational, retrospective study, safety information was obtained from medical records of pregnant women who received VacPertagen (0.5 ml, single dose) as part of routine antenatal care between 2021 and 2023, as well as from records of their infants. In this observational study, solicited AEs were not collected as it was not part of the study design. However, safety data on pregnancy and neonatal outcomes were collected. Safety endpoints were collected and reported as follows:

- Incidence of preterm delivery in pregnant women who received VacPertagen
- Comparison of obstetric outcomes between pregnant women who delivered during 2007-2018 and pregnant women who received monovalent pertussis vaccine during 2021-2023.

The average age of the 585 pregnant women after being vaccinated with VacPertagen was 29.84 years old, the average weight and height were 71.41 kilograms and 158.51 centimeters, respectively. The average gestational age at vaccination with VacPertagen was 31.65 weeks. Of the 585 pregnant women, 13 (2.24%) women received VacPertagen at 2nd Trimester and 568 (97.76%) women received VacPertagen at 3rd Trimester. The timing of vaccination was not available in 4 pregnant women.

Of the 585 pregnant women, 149 (25.47%) women received Td vaccine, 84 (14.36%) received COVID-19 vaccine and 482 (82.39%) received Influenza vaccine.

There were 50.60% (296/585) pregnant women with normal delivery, whereas 49.40% (289/585) with caesarean section. The caesarean section indications including CS due to previous Caesarean section (128/585, 21.88%), CS due to cephalopelvic disproportion (65/585, 11.11%), CS due to foetal non-reassuring (21/585, 3.59%), CS due to breech presentation (17/585, 2.91%), vacuum extraction (17/585, 2.91%) and other indications (41/585, 7.00%).

A total of 585 babies were born to 585 pregnant women (no twin pregnancy). Majority were male babies, 309 (52.82%) with mean birthweight of 3052.43 grams and mean gestational age of 38.56 weeks. Of the 585 newborn babies, 468 (80.00%) were assessed as healthy whereas 117 (20.00%) babies had abnormalities at birth including 14 hydrocele, 10 hypoglycaemia, 8 caput succedaneum, 3 neonatal cardiac arrhythmia, 3 bradypnea and desaturation, 5 cephalhematoma, 2 gastroesophageal reflux, 2 undescended testes, 2 congenital syphilis, 2 systolic ejection murmur gr. I, 2 foetal pyelectasis, 2 microcephaly, 2 sacral dimple, 2 bradycardia and 58 other abnormalities. The newborn stayed at the hospital for an average of 5 days.

Of the 585 newborn babies, 55 (9.40%) babies were premature or preterm (<37 weeks GA) and 64 (10.94%) babies were low birthweight (< 2500 grams).

Table 124. Comparison of obstetric outcomes between pregnant women during 2007-2018 and pregnant women who received monovalent pertussis vaccine during 2021-2023.

Obstetric outcomes	Monovalent pertussis vaccine (2021-2023) N = 585 n (%)	Control group (2007-2018) N = 106,946 n (%)	P-value
Preterm delivery (< 37 weeks)	55 (9.40)	14641/105471[1] (13.88)	0.002*
Low birth weight (< 2500 grams)	64 (10.94)	12086 (11.3)	0.783
Mode of delivery			
Normal	296 (50.60)	56221/105,471[1] (53.30)	0.018*
Caesarean section	289 (49.40)	45141/105,471[1] (42.80)	
Birth asphyxia (Apgar Score ≤ 7)	46 (7.86)	6600 (6.17)	0.090
Neonatal outcomes			
Disability/other abnormalities	117 (20.00)	54739 (51.18)	<0.001*
Length of hospital stay			
1-3 days	248 (42.39)	31652 (29.60)	<0.001*
4-7 days	284 (48.55)	66672 (62.34)	
8-14 days	34 (5.81)	6225 (5.82)	
15-30 days	11 (1.88)	674 (0.63)	
> 30 days	8 (1.37)	248 (0.23)	

[1] Number of pregnant women during 2007-2018 (N = 105,471).
*p-value < 0.05 was statistically significant.

Post- authorisationdata in Thailand and Singapore

The MAH provided data on spontaneous report in Thailand and Singapore, including data reported in the last PSUR (30 September 2016 - 30 September 2023).

Following licensure of VacPertagen vaccine in 2016, BioNet has conducted an active post marketing surveillance on the enhanced safety of the vaccine in individuals aged 11 years and older.

From 2016 to 2023, in the active post-marketing surveillance in Thailand, collected data from 15,618 vaccinees including 5,414 who received VacPertagen (2,953 non-pregnant adults , 36 elderly 65-75 yoa, and 10,425 pregnant adults). The remaining 10,204 receved Boostagen (Td-VacPertagen) (437 adolescents; 8,268 non-pregnant adults, and 1,499 pregnant women).

In total, 17 AEFIs were reported in 15 individuals vaccinated with VacPertagen. Twelve non-pregnant adults 18-64 yoa experienced injection site pain (mostly females), and 1 non-pregnant adult 18-64 yoa experienced injection site pain and injection site bruise. There were no AEFI reported in elderly. One pregnant woman reported rash at the point of injection, and 1 experienced injection site pain and injection site induration. Incidence rates (IR) were very low: 2.4/1000 for injection site pain and 0.2/1000 for all others (versus frequencies common ≥1/100 to <1/10 or very common ≥1/10 in clinical studies).

Among the 2425 pregnant women who received VacPertagen between 2016 and 2023, pregnancy safety outcome reports were only received for 559 women through active follow-up with healthcare practitioners. Of the 559 pregnancies (women vaccinated with VacPertagen), 22 (3.9%) involved pregnancy-related complications: premature labour (8/22, 36.4%), non-reassuring foetal status (2/22, 9.1%), premature rupture of membranes (2/22, 9.1%), antepartum haemorrhage due to placenta praevia (2/22, 9.1%), gestational hypertension (2/22, 9.1%), cephalopelvic disproportion (2/22,

9.1%), abruptio placenta (1/22, 4.5%), superimposed preeclampsia (1/22, 4.5%) and chorioamnionitis (1/22, 4.5%). In one case (1/22; 4.5%), the specific nature of the complication was not documented. All cases were assessed by the reporting physician as unrelated to vaccination. However, without details, it is not possible to check this assessment. Among the 559 pregnant women, a total of 570 liveborn infants were delivered, consisting of 546 singleton births and 11 twin pregnancies (22 infants). In addition, two stillbirths were reported.

Following licensure in Singapore on 23 July 2021, VacPertagen has been indicated for booster immunisation against pertussis in individuals aged 11 years and older. As of December 2024, a total of 114 individuals had received VacPertagen under routine use in Singapore. All vaccinees were adults aged 18 to 64 years (113 between 18-44 yoa and 1 between 45-64 yoa), including 79 pregnant women (69.3%). Eight questionnaires were collected from healthcare professionals during the interview period in Year 2023 – 2024. No adverse events following immunisation (AEFIs) were reported during this period. No further safety information is available in the provided (very limited) statistical analysis report.

Out of 114 vaccinated persons, no reactogenicity or unsolicited AE was observed. In this post-marketing setting, the usefulness of these data is questioned without control group.

2.6.9. Discussion on clinical safety

Safety Assessment

The Applicant submitted 9 studies in the dossier following a RCT design (TDA202, TDA203, TDA204, TDA206, TDA207, APV301, PerMIT, PertADO, PertaPrime). Of these, 6 studies included VacPertagen as study vaccine, 3 studies (TDA203, TDA204 and PertADO) only included vaccines with a lower concentration of Pertussis antigens and/or additional antigens for tetanus and diphtheria (e.g. Boostagen), but no study arm with only VacPertagen in its intended composition was included in these studies. Notably, non-pregnant adult subjects vaccinated with VacPertagen were followed only in RCTs TDA206, APV301 and Pertaprime-01. Non-pregnant adult subjects (including elderly) vaccinated with Boostagen were followed in RCTs TDA203 and TDA206, adolescent subjects were followed in TDA202, supported by PertADO. Adult pregnant women were followed in RCTs TDA207 and TDA204 (VacPertagen in TDA207, Boostagen in TDA207 and TDA204). A pooled safety database for adolescents and adults (including pregnant women) was provided for studies TDA202, TDA206, TDA207, APV301 and Pertaprime-01, which also serves as basis for the determination of ADRs. Safety data from pregnant women (including pregnancy outcomes) will be discussed separately, as the vaccination during pregnancy constitutes a separate indication. Safety data collection in supportive post-authorisation studies on adult subjects (PerMIT, PerMIS) will be described as required. Safety events in conducted RCTs were assessed at the vaccination day (immediate responses), at day 7 (solicited) and day 28 (AEs) after vaccination. In pregnant women of study TDA 207, possible complications during delivery and the health status of the offspring were evaluated (SAEs). Safety beyond day 28 were provided upon request for studies TDA206 and for TDA207 beyond the delivery visit. The outlined safety assessment appears appropriate to cover unwanted effects of the study vaccine and to assess any possible complication caused by the vaccination during pregnancy. As per protocol of the pivotal studies all safety events that were not categorized as solicited reaction from vaccination to day 7 (and beyond) were recorded as adverse event and are reported in the list of AEs until day 28.

Exposure

The interpretation of resulting safety events from participants that were vaccinated with Boostagen needs to be cautious due to the co-administration of tetanus-diphtheria toxoids included in the vaccine (included in studies: TDA202 with n=150 adolescent subjects, TDA203 with n=50 non-pregnant adult women, TDA204 with n=80 pregnant adult women, TDA206 n=150 adults and elderly, TDA207 n=40

pregnant women). Boostagen contains VacPertagen (i.e. PT and FHA in the intended concentration of 5µg each), but also contains tetanus and diphteria toxoids (7.5Lf and 2Lf, respectively). A clear causal relationship of any given safety event (including reactogenicity) to the Pertussis antigens is not possible from participants that were vaccinated with Boostagen. Still, one would expect a stronger reactogenicity and a potentially wider safety profile from a vaccination with additional antigens. Thus, safety data from participants that were either co-administered with other vaccines besides VacPertagen or have received Boostagen still serve as important supportive safety information. However, these data cannot be used to conclude on ADRs related to the vaccination with VacPertagen, due to confounding antigens not included in the intended product.

As per Guideline on clinical evaluation of vaccines (EMEA/CHMP/VWP/164653/05 Rev. 1), at least uncommon adverse events (occurring in between 1/100 and 1/1000 vaccinated persons) should be possible to be described by the **safety database.** The full safety database collected for **adult and adolescent subjects** in a RCT setting after vaccination with VacPertagen (and without co-administration of Td-antigens) comprises n=2508 subjects (APV301: n=2100, TDA206: n=150, Pertaprime-01: n=68, TDA202: n=150, TDA207: n=40 pregnant women), and is further supported by n=320 non-pregnant adult subjects that were followed after vaccination with Boostagen (TDA203: n=50, TDA206: n=150, TDA204: n=80 pregnant women, TDA207 n=40 pregnant women) and n=181 adolescent subjects that were followed after co-vaccination of VacPertagen with Td-antigens (Boostagen in TDA202 and PertADO). This dataset includes the subgroups of elderly (n=153) that were vaccinated with VacPertagen in studies APV301 (n=123) and TDA206 (n=30; also supported by some elderly vaccinated with Boostagen: n=30). In conclusion, the provided safety database from RCTs seems sufficiently large to describe the safety profile of VacPertagen. Adverse events of **pregnant women** that were vaccinated with VacPertagen and followed in a RCT setting were only reported in study TDA 207 (n=40). Further pregnancy outcomes were also reported in the PerMIT trial (n=256) and a retrospective observational cohort safety study (PerMIS with n=1980), as well as in the scope of a PSUR and a postmarketing-observational studies (e.g. comparison of obstetric outcomes in Pertagen-MOM with n=585). The safety dataset is also supported by additional data from pregnant women vaccinated with Boostagen (n=40 in TDA207, n=80 in TDA204, n=90 in WoMANPOWER and n=252 PerMIT).

All subjects of the main clinical studies (TDA202, TDA206, TDA207 and APV301) were Asian and all these studies were conducted in Thailand. The only two studies outside Thailand and with other included ethnicities are PertaPrime-01 conducted in Australia (n=68) and PertADO conducted in Switzerland (n=31). Limited information is available for study PertADO (single publication), the study population was adolescent and VacPertagen was co-administered with tetanus-diphtheria toxoids. PertADO is the only study conducted geographically in Europe. Thus, only limited information is available from a European population vaccinated with VacPertagen. No explicit safety concern is evident, but there appears to be a tendency for the European population to report more safety related events compared to the Thai population as followed in the clinical studies (TDA202, 206 and 207; see detailed discussion below).

All studies were done on healthy subjects. None were performed in patients with relevant co-morbidities such as clinically significant renal, hepatic, cardiac impairment, or immunodeficiency and there is no data available to assess the effect of administration of VacPertagen on breastfed infants or on milk production/excretion. However, the safety profile in these populations is not expected to differ significantly from the targeted population and thus, routine pharmacovigilance suffices for further characterising it.

Adverse Events pooled from main RCTs in adults (including pregnant women) and adolescents

Solicited and unsolicited AE

Immediate reactions after VacPertagen vaccination were mostly local injection site pain (in 12.24%) and systemic myalgia (in 6.82%). Except for local injection site pruritus (in 1.31%) as well as systemic headache (2.63%) and fatigue (1.56%), all other solicited immediate reaction were reported in <1% of subjects. A similar pattern is also evident for **solicited reactions** within 7 days after vaccination, with injection site pain (in 50.1%) being the most common local reaction and myalgia (in 30.91%) the most common systemic reaction. No concern arises from the pattern and principal frequency of reported solicited reactions after vaccination with VacPertagen. Solicited reactions (severe and general) within 7 days were more common in adolescent subjects compared to adult subjects (for VacPertagen, Boostagen and Adacel in TDA202 as compared to TDA206). However, patient numbers are low, which limits the interpretation of potential age-related differences in event frequencies. Within the largest trial the proportion of participants reporting any solicited AEs within 7 days after vaccination was around 10% lower in the VacPertagen group compared with the comparator Boostrix group.
Unsolicited adverse events within 28 days after vaccination were reported by 12.56% of subjects in the pooled set of safety data. However, most events were observed in <1% of subjects, only injection site pain (in 2.83%), headache (in 1.79%) and myalgia (in 1.76%) were reported by ≥1% of subjects. Reported AEs within 28 days after vaccination reflect an expected response to vaccination. The proportion of subjects with AEs, related AEs and severe AEs within 28 days is only mildly higher in adolescent subjects of study TDA202 compared to the rates reported for adult subjects in study TDA206 . In study the largest trial APV301, the proportion of participants reporting any unsolicited AEs within 28 days post-vaccination was around 5% higher in the VacPertagen group compared to the Boostrix comparator group. Most of them were solicited AEs. No safety follow-up beyond 28 days after vaccination was provided for study APV301. Long-term safety data are available from subjects followed for one year (n=68 in Pertaprime-01, n=150 in TDA206 and n=150 in TDA202) and further supported by data from subjects vaccinate with Boostagen.

Notably, a substantial discrepancy in reported event rates is evident for study Pertaprime-01 compared to other main studies. A clearly higher proportion of subjects has reported solicited and unsolicited reactions in study Pertaprime-01 compared to studies TDA206 and APV301. However, reporting frequencies within Pertaprime were more comparable to the comparator vaccine in the same study (i.e. Boostrix). The discrepancy in reported safety data from the Pertaprime-01 compared to studies TDA206 and APV301 trials might be driven by e.g. a discrepancy in reporting of adverse reactions/events between study sites, a relation of reported AE to ethnic differences in the study population of both trials, or related to the age distribution (Pertaprime-01 mean age: 20.62 vs. TDA206 mean age: 44.46 and APV301 mean age: 40.66) of the studied population. Notably, the proportions of subjects with solicited reactions and AEs in Pertaprime-01 appear more comparable to study PertADO in adolescent subjects. The pivotal studies, including TDA206 and APV301 in adult subjects, were conducted exclusively in Thailand and in an Asian population, whereas Pertaprime-01 was conducted in Australia and PertADO in Switzerland, both with substantial inclusion of Caucasian subjects. Adverse events reported in PertADO might be confounded by the applied coadministration of a tetanus-diphtheria toxoids vaccine, but the discrepancy also holds in comparison to subjects vaccinated with Boostagen in study TDA206. The exact driving factor for the observed discrepancy between studies remains elusive and is most like multifactorial. Still, in summary, it cannot be excluded that higher event rates have to be anticipated for the European population compared to the event rates reported from main clinical evidence in the adolescent and adult population exclusively reported from studies in Thailand, especially since the information provided from a European population is rather limited (n=31 adolescent subjects in study PertADO).

As a consequence of the above-described discrepancy in observed event rates across studies the reporting frequency of ADRs for section 4.8 was determined based in on the highest reported frequency as reported in individual studies and not based on the frequency of the pooled dataset.

Overall, the most frequently reported adverse reactions after vaccination with VacPertagen were injection site pain (77.6%, Pertaprime-01 study), headache (59.7%, Pertaprime-01 study), fatigue (52.2%, Pertaprime-01 study), myalgia (45.3%, TDA202 study), arthralgia (24%, TDA202 study), malaise (22.7%, TDA202 study) and nausea (22.4%, Pertaprime-01). The majority of the reactions were mild in severity and resolved within a few days of onset.

Additionally, diarrhoea, enteritis, injection site induration, injection site haematoma, rash, urticaria and lymphadenopathy were identified as ADRs with higher frequencies reported in study Pertaprime-01 compared to the safety pool and are adequately reflected with higher frequency in section 4.8. Similarly, lymphadenitis as identified ADR was reported with higher frequency in study TDA202 compared to the safety pool and is adequately reflected with higher frequency in section 4.8. Besides the above mentioned and those events already covered as solicited events (i.e. vomiting, injection site pruritus, injection site erythema, injection site swelling, chills, pyrexia), only palpitations was additionally identified as an ADRs that is possibly related to the vaccination with VacPertagen (rare; see SmPC section 4.8).

Serious adverse events (SAEs)

Adolescent

During study TDA202 only one **SAE** was reported within 28 days after vaccination. The adolescent subject was vaccinated with VacPertagen and experienced an open wound after accident 18 days after vaccination. From day 29 until day 336 a few subjects reported SAEs in all treatment groups, with a mildly higher rate in subjects vaccinated with VacPertagen or Boostagen (3.4% both) compared to those vaccinated with Adacel (1.35%). However, the rate of SAEs in adolescent subjects is small and none of the reported SAEs in the study was considered vaccine-related. Narratives of SAEs were provided, which support the assessment as unrelated to study vaccine. No subject discontinued due to a SAE. In conclusion, no concern arises from reported SAEs within 336 days after vaccination in adolescent subjects.

Adults

No serious adverse event was reported from any of the subjects vaccinated with VacPertagen in study TDA206 until day 28 after vaccination. From subjects that were vaccinated with Boostagen, one subject (0.67%) experienced a SAE that was a medically significant event (knee injury). Similarly, for subjects vaccinated with Adacel one subject (0.67%) experienced a SAE that was leading to hospitalization (ruptured appendicitis). No vaccine related SAEs were reported throughout the study. No concern arises from reported SAEs within 28 days after vaccination in adult subjects.

Only one serious adverse event has occurred until day 336 in the study group vaccinated with VacPertagen (PT Breast mass occurred >5 months after vaccination in a >40-year-old female subject, resolved within the reporting year). Relation of this event to the study vaccine is unlikely.

During study APV301 a total of two SAEs were reported in the VacPertagen group. One SAE (type 2 diabetes mellitus with an infected wound on lower extremity) was vaccine-unrelated and the other one (acute ischaemic stroke in a >60-year-old male, 21 days after vaccination) was considered possibly related by the investigator but deemed vaccine-unrelated by the Sponsor due to known safety profile of inactivated pertussis vaccine and plausible alternative cause: untreated hypertension with imaging findings of chronic cerebrovascular lesions. No SAE of stroke was reported in study TDA206 nor in

other clinical studies (RCT TDA207 and supportive RCT Pertaprime-01, or the RCT in adolescent subjects TDA202).

In Pertaprime-01 study, 4 serious adverse events have been reported within the full year in 2 subjects (appendicitis and tooth impacted in one subject as well as biliary colic and depression each in one subject). Causal relation to the study vaccine does not seem evident.

Elderly subjects (TDA206 and APV301)

The provided information on elderly subjects ≥65 years old that were vaccinated with VacPertagen is limited to data from study TDA 206 (n=30) and APV301 (n=123). From these subjects, only 34.64% have reported at least one solicited event and 7.84% have reported an unsolicited adverse event (2.61% with vaccine-related unsolicited event). No serious AE was reported by any subject ≥65 years old. No specific concern arises from reported safety events in elderly. The direct comparison t the safety profiles of subjects <65 years of age (pooled data of studies TDA206 and APV301) even suggest a more favourable safety profile for the elderly population compared to adults aged <65 years (Solicited AEs within 7 days following vaccination: 34.64% vs. 59.64%, Unsolicited AEs within 28 days following vaccination: 7.84% vs. 10.26%, Related unsolicited AEs within 28 days following vaccination: 2.61% vs. 4.63%, respectively). Therefore, no specific risk minimisation measure is needed for the elderly.

PertADO (adolescent)

For PertADO trial insufficient background information was provided. The submission in the dossier is restricted to the publication Blanchard Rohner et al. 2019 ("Boosting Teenagers With Acellular Pertussis Vaccines Containing Recombinant or Chemically Inactivated Pertussis Toxin: A Randomized Clinical Trial"). Participants were co-administered with VacPertagen and a tetanus-diphtheria toxoid vaccine, or have received a tetanus-diphtheria-aP vaccine containing chemically detoxified PT. The study population was adolescent. As per study methods, serious AEs were to be recorded within 28 days after vaccination, but no serious AEs were reported in that study.

Boostagen

Pooled data from adolescent and adult subjects that have received Boostagen (i.e. VacPertagen including Td-antigens) studied in TDA202, TDA206, TDA207, PertADO, Pertaprime-01 and APV301 do not indicate any critical difference in reported safety events compared to the pooled data from subjects vaccinated with Adacel or Boostrix (both also containing Td-antigens; data not presented here).

Overall, no safety concern was identified in adolescents and adults.

Adverse events in pregnant Women

TDA207

No **immediate reactions within 30 minutes** after vaccination were reported by any of the pregnant women vaccinated in study TDA207. Within the first **7 days** after vaccination 67.5% of women vaccinated with VacPertagen have experienced a local reaction and 50% have experienced a systemic reaction. This is an expectable proportion. The rate in local reactions is a bit lower and the rate in systemic reactions slightly higher compared to the other vaccines in the study. Especially fatigue (32.5%), myalgia (22.5%) and headache (20%) were reported systemic reactions and pain was the most frequently reported local reaction (67.5%) after VacPertagen vaccination. Other systemic and local reactions were reported in ≤3 subjects (equivalent to ≤7.5%). Importantly, no fever and no severe solicited event was reported from pregnant women after vaccination with VacPertagen. The rates of solicited events were also comparable after vaccination with Boostagen. No concern arises

from safety data reported at the day of vaccination and form solicited reaction until day 7 after vaccination. Within **28 days** after vaccination 17.5% have reported any AE and in one subject (2.5%) 3 events were considered as vaccine-related (mild injection site pain, mild fatigue, one moderate headache) and in 2 subjects the AE required medical attendance (3 events, mild COVID-19, mild dermatitis and moderate threatened preterm labour). Until delivery day in total 11 subjects (27.5%) reported an AE that required medical attendance after vaccination with VacPertagen, which is a comparable ratio as for the other vaccine groups. No severe adverse event was reported in pregnant women vaccinated with VacPertagen after vaccination until the day of delivery. As per study plan, at the delivery day and beyond only serious AEs were recorded.

Complications during pregnancy within 28 days after vaccination affected 2 subjects (5%), but relationship to the vaccination does not appear plausible for gestational diabetes and is difficult to examine for the threatened preterm labour (without preterm birth) due to the lack of placebo control and the limited number of included subjects. From 28 days after vaccination until delivery, additional 5 subjects reported complication during pregnancy (7 events: 4 preterm delivery (<37 weeks of gestation) 1 preterm premature rupture of membranes, 1 pre-eclampsia/eclampsia and 1 chorioamnionitis). None of the infants was born with low birth weight (<2000g) or considered small for gestational age after the mother was vaccinated with VacPertagen during pregnancy. Two such events were reported for each category after the mother was vaccinated with Boostagen during pregnancy. No pregnancy loss or stillbirth was reported in any of the study groups. Reported complications appear principally in an expected range and appear comparable in the other study groups.

Complications during delivery were frequently reported in all study groups. Around two-fold more subjects reported complications after vaccination with VacPertagen compared to the other study groups (42.5% of subjects and e.g. 20.52% in the $Tdap_{chem}$ comparator group). Of note, also in the group treated with Boostagen the rate was much lower, despite the fact that VacPertagen is part of the Boostagen vaccine combination (21.05% of subjects with complications during delivery). The most common complication during delivery in the VacPertagen group was cephalopelvic disproportion (n=9, 22.5%). Other events were reported in ≤2 subjects in that study group (2 with non-reassuring foetal heart rate and each 1 with breech presentation, chorioamnionitis, footling breech, forceps extraction due to foetal malposition, non-reassuring foetal status, placenta previa totalis, Severe preeclampsia and unprogress of labour). Cephalopelvic disproportion was also reported in the other study groups (2.5.-7.5%), but was exceptionally high in the VacPertagen group. Events reported for the mothers and infants do not suggest any specific event in relation to head/body size that might have cause the high rate of cephalopelvic disproportion in subjects vaccinated with VacPertagen. Of note, cephalopelvic disproportion was also the main cause for emergency C-sections in that study group. In fact, the rate of C-sections (62.5%) and especially emergency C-sections (45% of deliveries in women vaccinated with VacPertagen) was very high in the study group vaccinated with VacPertagen. Causes for the emergency C-section are equivalent to the reported complications during delivery above (mostly cephalopelvic disproportion), but with additional previous C-sections as cause reported. Elective C-sections were all decided based on previous C-section in the VacPertagen study group. Notably, the high rate in emergency C-sections was not observed for Boostagen (n=7, 17.5%), as also only one subject had cephalopelvic disproportion reported in that group. Thus, a direct effect of VacPertagen on observed rates in cephalopelvic disproportion (and in consequence emergency C-sections) cannot be established, as VacPertagen is also part of the combination vaccine Boostagen. Furthermore, no biological plausibility mechanism was identified by which vaccination with VacPertagen could be related to increased cephalopelvic disproportion.

SAEs in infants

As a likely consequence of the emergency C-sections also a high rate of **serious AEs** at delivery were observed for **infants** of those mothers that were vaccinated with VacPertagen (n=14, 35%) (versus

Boostagen 12.5%, Adacel 20.83%). The vast majority of these were "Inpatient's hospitalization / prolongation of existing hospital" (n=11, 27.5%), which is likely a consequence of the emergency C-section that was required for 18 deliveries. With respect to other active treatment groups of this study, the highest rate of SAEs was reported for infants from mothers that were vaccinated with Boostagen (22.5%), which also contains VacPertagen. This rate still appears comparable to other treatment groups of the study that were vaccinated with genetically inactivated antigens (15-20%), but it is noted that the group vaccinated with the chemically inactivated comparator vaccine had the lowest rate of serious events in infants (12.5%, also mostly caused by Inpatient's hospitalization / prolongation of existing hospital in n=4 subjects). Notable are cases of neonatal sepsis (in 12.5%, 2.5% and 2.5% of participants vaccinated with VacPertagen, Boostagen and Adacel, including one case reported as PT Streptococcal sepsis) and neonatal jaundice (in 5%, 0% and 0% of participants vaccinated with VacPertagen, Boostagen and Adacel). None of these SAEs was considered vaccine related by the investigator and a clear causal relationship cannot be established as subject numbers are too low.

Physical examination as well as growth and development were assessed in infants at months 2, 4, 6 and 7 without any critical outcome.

SAEs in pregnant women

During the time from vaccination until delivery 22.5% of **pregnant women** vaccinated with VacPertagen have reported a SAE, mostly PTs COVID-19 (7.5%) and premature labour (10%), whereas amniotic cavity infection, premature delivery, preterm premature rupture of membranes and threatened labour were reported each by a single subject. The rate of subjects with SAEs is a bit higher compared to women vaccinated with Boostagen or Adacel (17.5% and 12.5%, respectively). None of the SAEs reported in study TDA207 was considered vaccine-related. Narratives were assessed, which support the categorization as not-related to study vaccine. No death and no withdrawal due to SAE was reported.

From vaccination until visit 4 (2 months after delivery), in total 22.5% of women have reported a serious AE (3 (7.5%) COVID-19, 1 (2.5%) amniotic cavity infection, premature delivery in 1 (2.5%), premature labour in 4 (10%), preterm rapture of membrane in 1 (2.5%), threatened labour in 1 (2.5%)). For the comparator vaccine (Adacel), 15% have reported a SAE (2 Covid19 (5%), 1 false labour (2.5%) and 3 premature labour (7.5%). From delivery until visit 7 (i.e. 7 months after delivery), those infants that were born to mothers vaccinated with VacPertagen or Boostagen have the clearly highest proportion in serious AEs (in 41.03% and 42.86%, respectively) compared to all other study groups (all with SAEs in <30%) and especially compared to the comparator vaccine (SAEs in 18.92%). The most frequent events were neonatal jaundice, neonatal sepsis, and premature baby (each in 10.26% of infants), all other events were reported in <10% of infants. None of these SAEs were considered vaccine-related by the investigators and none of them led to withdrawal from the study nor death.

In summary, the available data does not allow to conclude in a causal association between VacPertagen and SAEs since subject numbers in this single RCT in pregnant women are too low. It cannot be excluded that the observed differences are chance findings. No further randomised trial was conducted in pregnant women.

Post-authorisation safety studies in pregnant women in non-EU countries

PerMIT

As supportive information on vaccination in pregnant women, the Applicant has conducted PerMIT in non-EU countries as a post-authorisation observational (non-randomised, non-blinded), prospective, study in pregnant women with pregnancy and neonatal outcomes reported (including any complications during pregnancy and delivery), but at the day of delivery only. No solicited events, AEs or SAEs were planned to be recorded in the study. In total 256 women were enrolled that were vaccinated with VacPertagen before, and an additional 252 and 76 pregnant women were enrolled that were vaccinated with Boostagen or a licensed Td vaccine before study enrolment. Notably, the vaccination as such was not part of the study and subjects were not eligible if they had "any significant congenital abnormality confirmed by ultrasound" or "foetal abnormality, stillbirth or neonatal death" at study entry. Thus, subjects were not randomized (which could favour a subject selection bias in favour of the study vaccine) and a preselection of participants based on problematic safety outcomes was performed after vaccination and before study safety evaluation. In the end, only 9 subjects were excluded from the study due to reasons above-mentioned. Nonetheless, the exclusion criteria seem inadequate with respect to safety evaluation of the vaccines. Pregnancy outcomes were followed for n=248, n=249 and n=75 women that have received VacPertagen, Boostagen or a licensed Td vaccine during pregnancy and before the study, respectively. The difference in gestational age between the study groups that had received VacPertagen or the VacPertagen containing vaccine Boostagen (mean and median around week 30) and the group that has received a licensed Td vaccine (mean and median around week 20) appears large, which compromises the direct comparison within the study. Around 40% of subjects vaccinated with VacPartagen had a C-section and in >50% of those cases an abnormal C-section (i.e. unplanned C-sections due to complications during delivery) was recorded. However, the rate was comparable throughout all study groups. Cephalopelvic disproportion (the main cause of emergency C-section in study TDA 207, with an imbalance in subjects vaccinated with VacPertagen compared to other study vaccines in that study) was reported in >10% of subjects, in all study groups.

Reported neonatal outcomes do not indicate any specific event or pattern of concern with respect to the vaccination under evaluation and relation to the study vaccine does not appear evident, considering that vaccination was given around 8 weeks before delivery. No safety concern is evident from this study.

PerMIS-01

The observational, retrospective, cohort, safety study PerMIS-01 gathered information on women that were vaccinated with VacPertagen during pregnancy in Thailand and that gave birth from January 2021 to April 2024. The infant health status was also reported. C-sections were reported in 45.27% of births, complications were reported in 10% of deliveries and preterm deliveries (i.e. before gestational week 37) in 5.69%. The infant health status was compromised in 15.52% of newborns (e.g. difficulty breathing in 5.04%, neonatal jaundice in 3.98%, congenital abnormality of variable kind in 1.51%). Neonatal sepsis was reported in only 0.15% of newborns, (aspiration) pneumonia in 0.05%.

As a comparison, in study TDA207, serious cases of neonatal jaundice were reported in 15.39% of infants born to mothers that have received VacPertagen and 17.14% of infants born to mothers that have received Boostagen. Serious neonatal sepsis was reported in 10.26% (plus streptococcal sepsis in 2.56%) and serious pneumonia in 5.13% of infants born to mothers that have received VacPertagen. The potential risk of chorioamnionitis upon pertussis immunisation during pregnancy (see e.g. Kildegaard 2025) would also require a larger dataset to provide more reassuring evidence. These examples demonstrate the difference between the retrospective reporting compared to the reporting in the scope of RCT. Notably, it is unclear whether the serious cases reported in TDA207 were indeed causally related to the vaccination during pregnancy, as chance findings could also be possible considering the very limited sample size of this RCT.

Overall, no safety concerns were observed in the pregnancy outcomes reported in this study.

Importantly, as no solicited events and no AEs were assessed, study data from the PerMIT and PerMIS-01 trials do not contribute to the estimation of ADRs in pregnant women upon vaccination.

Pertagen-MOM

This was an observational, retrospective study in pregnant women who received VacPertagen. Only pregnancy and neonatal outcomes were collected. A comparison has been done between the obstetric outcomes between an historical group (2017-2018) and pregnant women who received VacPertagen (n=585) between 2021 and 2023.

Results show a statistically significant difference in the incidence rate of preterm delivery (13.88% vs. 9.40%, respectively; p-value=0.002) and in the neonatal outcomes with disability/other abnormalities (51.18% vs. 20%, respectively, p-value<0.001). However, there was no significant difference in terms of low birth weight (<2500 grams) and birth asphyxia (Apgar score ≤ 7). The mode of delivery seems to show a significantly higher rate of C-sections upon vaccination with VacPertagen compared to the control group.

However, the historical group has not been matched for the years, and it is critically noted that obstetric outcomes can evolve. Moreover, as pointed-out for PerMIS-01, this is an observational study without direct comparison to control which severely limit the interpretation of the results. Furthermore, no ADRs for pregnant women can be concluded from the study as safety events after vaccination were not systematically recorded as per protocol.

Additional studies in pregnant women

WOMANPOWER: This was a randomised clinical trial for which a literature reference was provided. Because of the small number of pregnant women in each group and the comparator without pertussis antigen (40 HIV+ Td-VacPertagen, 50 HIV- Td-VacPertagen, 41 HIV+ Td-only comparator, 50 HIV- Td-only comparator), the clinical relevance of these supporting data is very limited.

Further safety support and conclusion on vaccination in pregnant women

Following the reported data from RCTs that is supported by data from observational studies and post-marketing reporting, none of the discussed events regarding pregnancy outcomes can be associated to the vaccination with VacPertagen. The number of pregnant women followed in RCTs is rather low and results from those pregnant women vaccinated with VacPertagen (TDA207) do not fully correspond to those from women vaccinated with Boostagen, which raises uncertainty regarding the relation of events to VacPertagen vaccination. Considering the rather low number of pregnant women followed in study TDA207 (n=40 after VacPertagen), the fact that no critical events were considered vaccine-related and concerning imbalances were not seen to the same extent for subjects vaccinated with Boostagen (n=80 in TDA204 and n=40 in TDA207) and without concerning evidence from observational trials and post-marketing data (see below), it appears likely that reported imbalances in study TD207 constitute chance findings.

In order to further strengthen the evidence on safety for pregnant women and their infants when vaccinated with VacPertagen, the Applicant has committed to submit by Q1 2026 the final study reports for the prospective, observational studies PERTg001 and BOOSTg001 that have evaluated pregnancy outcomes after vaccination with VacPertagen (n=559) and Boostagen (n=816), respectively. Furthermore, a PASS is planned to be conducted in the EU to collect structured safety data from pregnant women vaccinated with VacPertagen, including information on pregnancy outcomes and infant health status, and the study is included in the RMP. The study is planned as observational, non-interventional cohort study utilizing a prospective pregnancy exposure registry with intended >10 000 births/year. Additional safety data from pregnant women and their infants are

planned to be reported during the post-authorisation immunogenicity study APV302, a phase 3 multicenter, randomized, controlled trial in Belgium, Australia, and the United States (n=164 subjects intended to be vaccinated with VacPertagen).

Reported imbalances in events around delivery as observed in study TDA207 remain highly uncertain, due to the very low subject numbers vaccinated with VacPertagen alone and lack of equivalent patterns observed for Boostagen in RCTs. Chance findings appear likely. Also, none of the supportive information from observational studies and post-marking data did identify any specific concern regarding the pregnancy outcomes after vaccination with VacPertagen. Altogether, the safety profile for vaccination during pregnancy is acceptable. Still, additional safety data from ongoing prospective trials (PERTg001 and BOOSTg001) and post-marketing studies (APV302 and PASS) are highly encouraged to be submitted post-authorisation in order to further characterise remaining uncertainties **(REC).**

Complementary Safety Data (all indications)

Post-marketing experience

The Applicant has submitted three reports from post-marketing observations in Thailand - trial PerMIT, a PSUR and a post-marketing observational report (Pertagen-MOM). Respective results are discussed above were considered of relevance. Of note, the PSUR as well as the report on the post-marketing observation lack detail and appear rather vague. However, an active post-marketing surveillance between 2016 and 2023 collected data from 15,618 vaccinees including 5,414 who received VacPertagen (2,989 non-pregnant adults, 2,425 pregnant adults). The remaining 10,204 received Boostagen (Td-VacPertagen) (437 adolescents; 8,268 non-pregnant adults, and 1,499 pregnant women).

Overall, in the active post-marketing surveillance setting with VacPertagen, with very low incidence rates observed for the solicited AEs and no ADR observed, pregnancy safety outcome reports were not received for all vaccinated pregnant women (559 out of 2425), and with the information missing for 2 liveborn infants. A limitation inherent to the nature of the post-marketing reporting is the risk of underreporting of safety events, and the usefulness of these data is questioned without control group. As discussed above, the Applicant intends to conduct two further post-marketing studies in pregnant women Europe (APV302 and a PASS).

Discontinuations, AESIs, Drug-drug interaction, laboratory data and examination vital/physical function

It is acknowledged that no discontinuation due to AE was reported for any of the pivotal studies TDA206 and 207 until day 28 after vaccination, but it is critically noted that no adverse events of special interest were defined to be reported for the pivotal clinical studies. No dedicated drug-drug interaction studies were performed to assess respective safety concerns. Of note, it should be considered that patients receiving immunosuppressive treatment may not elicit an adequate immune response after vaccination, but the Applicant has clarified that no data on immunocompromised patients were generated. No clinical laboratory tests of haematology, biochemistry and urinalysis were submitted for participants after vaccination in the main clinical studies TDA206 or TDA207. This is acceptable in the context of the overall safety evaluation and findings and in line with prior authorisations of vaccines. Data on physical examination and vital signs did not indicate any pattern of concern after vaccination with VacPertagen and also the provided physical examination of infants born to women that were vaccinate with VacPertagen during pregnancy did not reveal any concerning outcome. Physical examination as well as growth and development were assessed in infants at months 2, 4, 6 and 7 without any critical outcome.

. As mentioned above, additional safety data in pregnant women and their infants will be obtained in post-authorisation studies.

2.6.10. Conclusions on the clinical safety

The Applicant submitted 9 studies that were conducted in a RCT design (TDA202, TDA203, TDA204, TDA206, TDA207, APV301, PerMIT, PertADO, Pertaprime-01) and 6 of these studies included VacPertagen as study vaccine. Studies TDA206, TDA207 and APV301 are considered the pivotal evidence from randomised controlled trials, Pertaprime-01 is considered a supportive randomised controlled trial. Data generated from vaccination with Boostagen are considered supportive evidence, as the vaccine contains additional tetanus and diphtheria toxoids (7.5Lf and 2Lf, respectively) besides the two VacPertagen antigens (i.e. PT and FHA in the intended concentration of 5 µg each). Thus, a clear causal relationship of any given safety event (including reactogenicity) to the Pertussis antigens is not possible from participants that were vaccinated with Boostagen. The full safety database collected for adult and adolescent subjects in a RCT setting after vaccination with VacPertagen comprises 2508 subjects (APV301: n=2100, TDA206: n=150, Pertaprime-01: n=68, TDA202: n=250, TDA207: n=40 pregnant women) and is further supported by 320 adult subjects that were followed after vaccination with Boostagen (TDA203: n=50, TDA206: n=150, TDA204: n=80 pregnant women, TDA207 n=40 pregnant women) and 181 adolescent subjects that were followed after co-vaccination of VacPertagen with Td-antigens (Boostagen in TDA202 and PertADO). Based on these data from RCTs, the safety database seems sufficiently large to describe the safety profile of VacPertagen in adult and adolescent subjects.

Data on pregnant women that were followed in a RCT setting for adverse events after vaccination with VacPertagen are sparse (n=40) but are supported by observational data (PerMIT n=256, Permis n=1980, Pertagen-MOM=585), as well as post-authorisation spontaneous data in non-EU-countries. The safety dataset is also supported by additional data from pregnant women vaccinated with Boostagen (n=40 in TDA207, n=80 in TDA204, n=90 in WoMANPOWER and n=252 PerMIT). Further safety data to characterise the safety profile after vaccination in pregnant women are expected from ongoing prospective trials (PERTg001 with n=559 VacPertagen vaccinations and BOOSTg001 with n=816 Boostagen vaccination) and post-marketing studies (APV302 with n=164 planned VacPertagen vaccinations and a PASS utilizing a prospective pregnancy exposure registry with intended >10 000 births/year). However, no safety concerns were identified regarding the vaccination in pregnant women from available information.

In summary, reported safety results on immediate reactions, solicited events and AEs including SAEs do not cause reason of concern for the vaccination of adolescent or adult subjects, including elderly and adult pregnant women. Notably, limited safety information is available for the European population. This will be further characterised with routine pharmacovigilance activities.

In conclusion, the safety profile of VacPertagen in adults and adolescent subjects appears well tolerated, without unexpected safety signal. Therefore, licensure of VacPertagen in adult and adolescent subjects as well as for pregnant women is acceptable from a safety perspective.

2.7. Risk Management Plan

2.7.1. Safety concerns

Table 125. Summary of safety concerns

Important identified risks	None
Important potential risks	None
Missing information	Limited information on use in pregnant women in the European population

2.7.2. Pharmacovigilance plan

Table 126. Summary of ongoing and planned additional pharmacovigilance activities

	Summary of objectives	Safety concerns addressed	Milestones	Due dates
Category 1 - Imposed mandatory additional pharmacovigilance activities which are conditions of the marketing authorisation				
None				
Category 2 – Imposed mandatory additional pharmacovigilance activities which are Specific Obligations in the context of a conditional marketing authorisation or a marketing authorisation under exceptional circumstances				
None				
Category 3 - Required additional pharmacovigilance activities				
Planned	-Evaluation of the safety of VacPertagen in the second and third trimesters of pregnancy in relation to maternal and neonatal health outcomes. -Estimation of the effectiveness of maternal vaccination with VacPertagen in reducing the risk of pertussis in infants born to vaccinated mothers by comparing pertussis incidence in infants whose mothers received VacPertagen during pregnancy with infants of unvaccinated mothers, where appropriate data are available.	Limited information on use in pregnant women in the European population	Feasibility assessment	Within four to six months of EC decision

	Summary of objectives	Safety concerns addressed	Milestones	Due dates
			Protocol submission	Within six months of EC decision (end of procedure)
			Registration in EU PAS	Within two weeks after protocol approval
			Start of data collection	Within nine months of study approval
			End of data collection	After completion of follow up period for last patient in
			Final study report completion and submission	Within 12 months of data lock.

2.7.3. Risk minimisation measures

Table 127. Summary table of pharmacovigilance activities and risk minimisation activities by safety concern

Safety concern	Risk minimisation measures	Pharmacovigilance activities
Missing information: Limited information on use in pregnant women in the European population and its impact on neonates	Routine risk minimisation measures: SmPC section 4.6 PL section 2 Additional risk minimisation measures: No additional risk minimisation measures	Routine pharmacovigilance activities as per current regulatory guidance Additional pharmacovigilance activities: PASS in the EU to collect structured safety data from pregnant women vaccinated with VacPertagen, including information on pregnancy outcomes and infant health status

2.7.4. Conclusion

The CHMP considers that the risk management plan version 1.0 is acceptable.

2.8. Pharmacovigilance

2.8.1. Pharmacovigilance system

The CHMP considered that the pharmacovigilance system summary submitted by the Applicant fulfils the requirements of Article 8(3) of Directive 2001/83/EC.

2.8.2. Periodic Safety Update Reports submission requirements

The requirements for submission of periodic safety update reports for this medicinal product are set out in the Annex II, Section C of the CHMP Opinion. The Applicant did request alignment of the PSUR cycle with the international birth date (IBD). The IBD is 30.09.2016. The new EURD list entry will therefore use the IBD to determine the forthcoming Data Lock Points.

2.9. Product information

2.9.1. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the Applicant show that the package leaflet meets the criteria for readability as set out in the *Guideline on the readability of the label and package leaflet of medicinal products for human use.*

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

The Applicant seeks the following indications for VacPertagen:

- booster immunisation against pertussis of individuals aged 12 years of age and older,
- passive protection against pertussis in early infancy following maternal immunisation during pregnancy.

Whooping cough, also known as pertussis, is a highly infectious bacterial disease involving the lungs and airways. It is caused by bacteria in the mouth, nose and throat of an infected person. Whooping cough is spread via airborne droplets produced when the infected person coughs. Whooping cough can also be spread by an individual who has only a mild form of the disease, or by an infected individual who has no symptoms at all. Frequently, older siblings and parents who may be carrying the bacteria bring the disease home and infect an infant in the household.

Symptoms usually appear 7 to 10 days after infection but may also appear up to 21 days.

The most severe forms of whooping cough are in infants. Whooping cough in unvaccinated or incompletely vaccinated infants or infants whose mother was unvaccinated during pregnancy can be particularly severe. Complications include pneumonia, encephalopathy (a disease of the brain),

seizures and even death. In adults and older children, complications include inability to breathe for short periods, broken ribs, rectal prolapse and hernias.

3.1.2. Available therapies and unmet medical need

The most important way to prevent whooping cough is through complete immunisation. There's no whooping cough only vaccine authorised in EU. The vaccine for whooping cough is usually given in combination with diphtheria and tetanus vaccinations (often in combination also with poliomyelitis, Haemophilus influenzae and hepatitis B vaccination). A primary course of 2-3 doses is usually given between 2 and 12 months of age, in accordance with the national vaccination schedule. A third or fourth dose is recommended at 11-24 months of age, and another dose between 3 and 7 years of age.

Some EU/EEA countries recommend boosters for adolescents, adults, and/or women during pregnancy, which also temporarily protects the baby once it is born. Some countries also recommend a booster to unprotected women soon after they have delivered, to reduce the risk of transmission of the disease to the baby.

Antibiotics can be used to treat whooping cough and prevent further spread of the disease. However, in order to be most effective, treatment must begin early in the course of disease, during the first one to two weeks before the episodes of numerous rapid coughs occur.

There are a plethora of combination vaccines nationally/centrally authorized in EU (in combination with diphtheria and tetanus vaccinations or additionally in combination also with poliomyelitis, Haemophilus influenzae and hepatitis B vaccination). Some of these include 1 to 5 pertussis antigens and are indicated for primary vaccination of infants and for toddler's booster doses (DTaP / DTaP-IPV / DTaP-IPV-Hib / DTaP-IPV-Hib-HBV). Other vaccines, which include 1 to 5 pertussis antigens in comparatively reduced amounts (Tdap vaccines combined or not with inactivated poliomyelitis viruses), are authorised for booster vaccinations at older ages. Two Tdap vaccines (combined or not with inactivated poliomyelitis viruses) are indicated in Europe also for passive protection against pertussis in early infancy following maternal immunisation during pregnancy.

Boostrix (3 pertussis antigens) and Adacel (5 pertussis antigens), were alternatively used as active comparators within the submitted dossier for VacPertagen.

3.1.3. Main clinical studies

TDA202 is a pivotal phase II/III randomized, controlled clinical trial in a total of 450 healthy adolescent volunteers 12-17 years of age who either received Boostagen (BioNet $TdaP_{gen}$ vaccine, or Td-VacPertagen, a vaccine containing VacPertagen components together with tetanus and diphteria antigens), VacPertagen (BioNet 2-component aP; PT_{gen} and FHA) or the active comparator Adacel (n=150 per group). The study objectives were to demonstrate non-inferior immunogenicity of one dose of Boostagen as compared to Adacel vaccine (primary objective), to assess safety of Boostagen and VacPertagen, and non-inferior immunogenicity of VacPertagen as compared to Adacel vaccine (secondary objectives). Antibody persistence at one year after vaccination was also evaluated as part of the TDA202 study and for a cohort of participants at later time points (TDA202 2-year, 3-year, and 5-year follow-up).

TDA206 is a phase III randomized, observer-blind, active-controlled study in healthy adults 18-75 years of age to compare the safety and immunogenicity of different formulations of acellular pertussis vaccines, including VacPertagen and Td-VacPertagen and a licensed $Tdap_{chem}$ vaccine (Adacel). A total of 750 subjects were enrolled and randomized into 150 per group (total 5 groups). Immunogenicity assessment

was evaluated at baseline (before vaccination), at 28 days after vaccination and at approximately 1 year after vaccination in a randomly pre-selected subset of 375 subjects.

TDA207 is a phase II randomized, observer-blind, active-controlled study to evaluate the immunogenicity and the safety of the different doses and formulations of the Applicant's recombinant acellular pertussis vaccines containing genetically-inactivated pertussis toxin (PT_{gen}) compared to licensed $Tdap_{chem}$ vaccine (Adacel) when administered to healthy pregnant women. A total of 240 subjects were enrolled and randomized into 40 per group (6 groups in total). The maternal antibody response was primarily assessed at 28 days after vaccination and monitored at the time of delivery. Immune response in infants was assessed during the following time points: at birth, at 2 months of age prior to the first dose of pertussis primary immunisation, and at 7 months of age.

In addition, the dossier includes data from 9 clinical trials which are considered supportive evidence.

3.2. Favourable effects

Adolescents

At **28 days after vaccination** (study TDA202), ELISA anti-PT and anti-FHA seroconversion rates were higher in the VacPertagen group [anti-PT 96% (95% CI 93-99) and anti-FHA 93% (95% CI 89-97)] than the seroconversion rates in the Adacel group [anti-PT 55% (95% CI 47-63), anti-FHA 54% (95% CI 46-62)]. Also, anti-PT and anti-FHA GMTs were higher in VacPertagen [562 IU/mL (95% CI 467.79-674.86) for anti-PT antibody; 924 IU/mL (95% CI 809.39-1054.4) for anti-FHA antibody] than those GMTs in Adacel group [63 IU/mL (95% CI 51.05-78.37) for anti-PT antibody; 242 IU/mL (95% CI 208.86-280.05) for anti-FHA antibody] at 28 days after vaccination. Similar results were obtained with neutralizing anti-PT GMTs.

At **1 year after vaccination** (study TDA202), ELISA anti-PT and anti-FHA seroconversion rates were higher in the VacPertagen group [Anti-PT 82% (95% CI 71-93), anti-FHA 64% (95% CI 51-77)] than the seroconversion rates in the Adacel group [anti-PT 4% (95% CI 0-9), anti-FHA 28% (95% CI 16-40)] at 1 year after vaccination. Also, ELISA anti-PT and anti-FHA GMTs were higher in the VacPertagen and group [133 IU/mL (95% CI 92.96-189.77) for anti-PT antibody; 291 IU/mL (95% CI 230.94-367.14) for anti-FHA antibody] than GMTs in Adacel group [22 IU/mL (95% CI 16.05-29.75) for anti-PT antibody; 90 IU/mL (95% CI 64.46-125.39) for anti-FHA antibody]. Similar results were obtained with neutralizing anti-PT GMTs.

At **5 years after vaccination** (study TDA202), anti-PT seroconversion rates were 33% (95% CI 20-45) for VacPertagen and 2% (95% CI 0-6) for Adacel. Seroconversion rates for ELISA anti-FHA antibody also were higher in the VacPertagen group (45%, 95% CI 32-59, n=55) than in participants vaccinated with Adacel (8%, 95% CI 0-15). Also, ELISA anti-PT and anti-FHA GMCs were higher in the VacPertagen group (33 IU/mL, 95% CI 24.65-43.10 for anti-PT and 70 IU/mL, 95% CI 57.29-86.28 for anti-FHA) than GMCs in Adacel group (GMCs for both PT and FHA were below baseline level: 11 IU/mL, 95% CI 8.78-14.45 for anti-PT and 28 IU/mL, 95% CI 21.16-37.87 for anti-FHA).

Adults

In study TDA206, the seroconversion rate of anti-PT antibody in adult subjects at **28 days after vaccination** compared to baseline was higher in the VacPertagen group [100.00% (95% CI 95.20-100.00)] than the seroconversion rate in the Adacel group [74.32% (95% CI 62.84 - 83.78). Seroconversion rate of anti-FHA antibody at 28 days after vaccination compared to baseline was similar in all vaccine groups e.g. VacPertagen group [97.33% (95% CI 90.70-99.68)] vs Adacel group [93.24% (95% CI 84.93-97.77)]. Also, GMCs for anti-PT-IgG antibodies were higher in the VacPertagen group [371.83 IU/mL (95% CI 292.76-472.25)] compared to the Adacel group [50.84

IU/mL (95% CI 39.26-65.84)] at 28 days after vaccination. GMCs for anti-FHA-IgG antibodies were higher in the VacPertagen group [451.62 IU/mL (95% CI 373.46-546.12)] than in the Adacel group [207.58 IU/mL (95% CI 171.33-251.50)]. Similar results were obtained with neutralizing anti-PT GMTs.

In participants **aged 65-75 years** (n=15 per group), seroconversion rate in the VacPertagen group was higher than those in the Adacel group with a difference in seroconversion rate of 40.00% (95%CI 15.24-64.61). Furthermore, the 3 other BioNet vaccines (containing VacPertagen in same/lower amounts and/or also include Td antigens) also induced higher seroconversion rates 28 days after vaccination compared to Adacel. Also, GMCs for anti-PT-IgG antibodies were higher for the VacPertagen group [330.17 IU/mL (95% CI 168.85-645.63)] compared to the Adacel group [34.66 IU/mL (95% CI 15.66-76.67)] at 28 days after vaccination. GMCs for anti-FHA-IgG antibodies were numerically higher in the VacPertagen group [387.88 IU/mL (95% CI 218.85-687.45)] than in the Adacel group [213.11 IU/mL (95% CI 123.84-366.72)]. Similar results were obtained with neutralizing anti-PT GMTs.

At **3 years after vaccination**, GMCs for anti-PT-IgG and for anti-FHA-IgG antibodies were higher in the VacPertagen group compared to Adacel. The GMCs for anti-PT-IgG antibodies after VacPertagen were 43.00 IU/mL (95% CI 31.47-58.75) and 8.75 IU/mL (95% CI 6.55-11.70) after Adacel. The GMCs for anti-FHA-IgG antibodies were 92.39 IU/mL 95% CI 71.15-119.98) for VacPertagen and 52.36 IU/mL (95% CI 42.05-65.20) for Adacel.

Pregnant subjects and their infants

In maternal participants of Study TDA207, the **anti-PT GMCs** elicited by VacPertagen were higher compared to Adacel **at Day 28** after vaccination (153.98 IU/mL [95% CI: 107.51 – 220.55] vs. 29.53 IU/mL [95% CI: 20.20 – 43.16], respectively). This was also observed **in cord blood** (or neonatal blood within 72 hours after birth) at delivery (141.40 IU/mL [95% CI: 94.70 – 211.12] vs. 27.09 IU/mL [95% CI: 18.21 – 40.31], respectively) and **in infants 2 months after delivery** (60.46 IU/ml [95% CI: 38.92 – 93.92] vs. 10.74 IU/ml [95% CI: 7.65 – 15.07], respectively). Significant differences at these 3 time points were also reported with respect to seroconversion rates (percentage of maternal participants with a $\geq$ 4-fold increase in anti-PT antibodies concentrations). These findings are supported by PT neutralization data. Numerically higher GMCs and seroconversion rates were also noted in the VacPertagen group for **anti-FHA** antibodies, but confidence intervals were often overlapping. At **7 months after delivery**, infants had already received primary vaccination against pertussis (3 doses at months 2, 4 and 6), mostly with a whole cell vaccine. The anti-PT GMCs at 7 months were **lower** in the VacPertagen group, compared to the Adacel group (17.77 IU/ml [95% CI: 13.06 – 24.18] vs 40.98 IU/ml [95% CI: 26.59 – 63.15], respectively).

Supportive studies performed in adolescent, adult and pregnant subjects corroborated results from the main studies.

3.3. Uncertainties and limitations about favourable effects

While effectiveness has been demonstrated for other pertussis-containing vaccines, no efficacy or effectiveness data are available for VacPertagen.

There is no established immune correlate of protection for pertussis. Therefore, there is uncertainty regarding the relationship between the elicited antibody titres and the potential for vaccine efficacy. The Applicant has committed to conduct effectiveness studies with VacPertagen post-authorisation.

Available data on the impact of the maternal vaccination with VacPertagen on infants' response to primary vaccination suggest a strong "blunting" effect. While this phenomenon is known for licensed

vaccines with an indication of passive protection in infants after maternal immunisation, the effect was more pronounced with VacPertagen, at least with respect to anti-PT IgGs. This was also shown in the supportive study TDA204. The Applicant committed to further evaluate the immune interference with pertussis vaccination in infants after maternal pertussis vaccination in a post-authorisation study **(REC)**.

There are only limited data in adults aged 65-75 years. There are no data in individuals > 75 years of age.

There are no data in immunocompromised individuals for VacPertagen. There's limited data from a randomised clinical trial (WoMANPower) where Boostagen (Td-VacPertagen) was evaluated in a randomised clinical trial in pregnant women living with HIV in Uganda who received antiretroviral therapy. Results from this study indicate a booster effect of PT- and FHA- specific immune responses following vaccination with Boostagen in both HIV positive and HIV negative pregnant women. Because of the small number of pregnant women in each group the clinical relevance of these supporting data with Td-VacPertagen is very limited.

Methodologically, none of the clinical trials had an appropriate multiplicity control and most trials seemed to be rather suited to a well-planned, well-conducted proof-of-concept than to the generation of pivotal data for a novel vaccine.

3.4. Unfavourable effects

Quantitative

Pooled safety data for adolescent and adult subjects (including pregnant women) from RCTs (TDA202, TDA206, TDA207, Pertaprime-01 and APV301)

Immediate reactions within 30 minutes after vaccination were reported by 17.5% of subjects in the RCT pool and the most common events (≥1%) were injection site pain (12.2%), headache (2.6%), fatigue (1.5%), injection site pruritus (1.3%) and arthralgia (1.2%).

Solicited events were reported by 60.8% of subjects in the RCT safety pool. The most commonly reported solicited local event was injection site pain (50.1%) and the most commonly reported solicited systemic events were myalgia (30.9%), headache (17%), fatigue (15.4%) and malaise (11.7%). All other solicited events (injection site erythema, injection site swelling, injection site induration, injection site swelling, injection site pruritus, arthralgia, chills, pyrexia, arthralgia, vomiting and nausea) were reported by <10% of subjects. All solicited local and systemic events are listed in section 4.8 of the SmPC.

Unsolicited events within 28 days after vaccination were reported only by 12.7% of subjects (in 5.3% considered related events) in the RCT safety pool and the most commonly reported (≥1%) were injections site pain (2.8%), headache (1.79%) and myalgia (1.76%). The only event that was considered related and reported by ≥1% of subjects was myalgia (related in 1.4%).

The frequencies for all adverse reactions reported in section 4.8 were adjusted to the highest reported frequency in any of the studies mentioned above. Overall, the most frequently reported adverse reactions after vaccination with VacPertagen were injection site pain (77.6%, Pertaprime-01 study), headache (59.7%, Pertaprime-01 study), fatigue (52.2%, Pertaprime-01 study), myalgia (45.3%, TDA202 study), arthralgia (24%, TDA202 study), malaise (22.7%, TDA202 study) and nausea (22.4%, Pertaprime-01). The majority of the reactions were mild in severity and resolved within a few days of onset.

Additionally, diarrhoea, enteritis, injection site induration, injection site haematoma, rash, urticaria and lymphadenopathy were identified as ADRs with higher frequencies reported in study Pertaprime-01 compared to the safety pool and are adequately reflected with higher frequency in section 4.8. Similarly, lymphadenitis as identified ADR was reported with higher frequency in study TDA202 compared to the safety pool and is adequately reflected with higher frequency in section 4.8. Besides the above mentioned and those events already covered as solicited events (i.e. vomiting, injection site pruritus, injection site erythema, injection site swelling, chills, pyrexia), only palpitations was additionally identified as an ADRs that is possibly related to the vaccination with VacPertagen (rare; see SmPC section 4.8).

No discontinuation due to AE was reported for any of the studies until day 28 after vaccination.

Pregnant Women and offspring (TDA207)

No **immediate reactions within 30 minutes** after vaccination were reported by any of the pregnant women vaccinated with VacPertagen in study TDA207.

Within the first **7 days** after vaccination 67.5% of women vaccinated with VacPertagen have experienced a local reaction and 50% have experienced a systemic reaction. Fatigue (32.5%), myalgia (22.5%) and headache (20%) were reported systemic reactions and pain was the most frequently reported local reaction (67.5%) after VacPertagen vaccination. Other systemic and local reactions were reported in ≤3 subjects (equivalent to ≤7.5%). Importantly, no fever and no severe solicited event was reported from pregnant women after vaccination with VacPertagen.

Within **28 days** after vaccination 17.5% have reported any AE and in one subject (2.5%) 3 events were considered as vaccine-related (mild injection site pain, mild fatigue, one moderate headache) and in 2 subjects the AE required medical attendance (3 events, mild Covid-19, mild dermatitis and moderate threatened preterm labour). Until delivery day in total 11 subjects (27.5%) reported an AE that required medical attendance after vaccination with VacPertagen, but no severe adverse event was reported in pregnant women vaccinated with VacPertagen after vaccination until the day of delivery.

Complications during pregnancy within 28 days after vaccination affected 2 subjects (5%), but relation to the vaccination does not appear plausible for gestational diabetes and is difficult to examine for the threatened preterm labour (without preterm birth).

Preterm delivery (<37 weeks of gestation) was reported in 4 (10%) subjects and additional 5 subjects (in total n=7, 17.5%) have reported complications during pregnancy (1 preterm premature rupture of membranes, 1 pre-eclampsia/eclampsia, 1 gestational diabetes, 1 chorioamnionitis and 1 threatened premature labour). None of the infants was born with low birth weight (<2000g) or considered small for gestational age after the mother was vaccinated with VacPertagen during pregnancy. No pregnancy loss or stillbirth was reported.

Around two-fold more subjects reported complications during delivery after vaccination with VacPertagen compared to the other study groups (VacPertagen: 42.5%, Boostagen: 21.05%, Adacel: 20.52%). The most common complication during delivery in the VacPertagen group was cephalopelvic disproportion (VacPertagen: n=9, 22.5%, Boostagen: 2.5%, Adacel: 7.5%). Other events were reported in ≤2 subjects in that study group. Cephalopelvic disproportion was also the main cause for emergency C-sections in that study group. The rate of emergency C-sections of deliveries in women vaccinated with VacPertagen was rather high in the study group vaccinated with VacPertagen compared to other study groups (VacPertagen: 45%, Boostagen: 17.5%, Adacel: 30%). Events reported for the mothers and infants do not suggest any specific event that might have caused the high rate of cephalopelvic disproportion in subjects treated with VacPertagen. Low patient numbers are noted with respect to possible chance findings.

A high rate of **serious AEs** at delivery were observed for infants of those mothers that were vaccinated with VacPertagen (n=14, 35%) (versus Boostagen 12.5%, Adacel 20.83%). The vast majority of these were "Inpatient's hospitalization / prolongation of existing hospital" (n=11, 27.5%), which is likely a consequence of the emergency C-sections that were required for 18 deliveries. Notable serious events in infants were neonatal sepsis (in 12.5%, 2.5% and 2.5% of participants vaccinated with VacPertagen, Boostagen and Adacel, including one case reported as PT Streptococcal sepsis, and neonatal jaundice (in 5%, 0% and 0% of participants vaccinated with VacPertagen, Boostagen and Adacel). None of these SAEs was considered vaccine related by the investigator and a clear causal relationship cannot be established as subject numbers are too low. Chance findings cannot be excluded.

From vaccination until visit 4 (2 months after delivery) in total 22.5% of women have reported a serious AE (3 (7.5%) Covid-19, 1 (2.5%) amniotic cavity infection, premature delivery in 1 (2.5%), premature labour in 4 (10%), preterm rapture of membrane in 1 (2.5%), threatened labour in 1 (2.5%)). For the comparator vaccine (Adacel), 15% have reported a SAE (2 Covid19 (5%), 1 false labour (2.5%) and 3 premature labour (7.5%)).

From delivery until visit 7 (i.e. 7 months after delivery), those infants that were born to mothers vaccinated with VacPertagen or Boostagen have the clearly highest proportion in serious AEs (in 41.03% and 42.86%, respectively) compared to all other study groups (all with SAEs in <30%) and especially compared to the comparator vaccine (SAEs in 18.92%).The most frequent events were neonatal jaundice, neonatal sepsis, and premature baby (each in 10.26% of infants), all other events were reported in <10% of infants. None of the SAEs reported in infants were considered related to study vaccination by the study investigator.

Data on physical examination and vital signs in vaccinated adult subjects, as well as physical data from newborns in study TDA207 do not indicate any negative effect from vaccination.

Qualitative

Comparative profiles of subjects <65 years of age and subjects ≥65 years of age from pooled data of studies TDA206 and APV301 suggest a more favourable safety profile for the elderly population compared to adults aged <65 years.

3.5. Uncertainties and limitations about unfavourable effects

The only two studies outside Thailand and with other included ethnicities are Pertaprime-01 conducted in Australia and PertADO conducted in Switzerland. PertADO is the only study conducted geographically in Europe, but limited information is available for this study (single publication), the study population was adolescent and VacPertagen was co-administered with tetanus-diphtheria toxoids (n=31), rendering the interpretation of resulting safety events ambiguous. Thus, limited information is available for the European population vaccinated with VacPertagen and based on reported safety outcomes, higher AE rates might have to be anticipated for the European population compared to the population studied in main clinical studies in Thailand. A discrepancy in reported frequencies of solicited reactions and adverse events between adult subjects vaccinated with VacPertagen at the study sites in Thailand (TDA206) and Australia (Pertaprime-01) is noted.

Only 40 pregnant women were followed in a randomised, controlled trial after vaccination with VacPertagen. Adverse events after vaccination with VacPertagen during pregnancy were recorded only from these subjects in a systematic manner. Chance findings reported within this trial cannot be ruled out from this low sample size. This includes complications during delivery (mostly cephalopelvic disproportion) and SAEs in infants (mostly prolonged hospitalisation) as reported after vaccination with

VacPertagen during the only randomised controlled trial that has followed pregnant women (including adverse event reporting). Importantly, post-authorisation safety data in pregnant women is available from non-EU countries and do not raise any safety concern. 'Limited information on use in pregnant women in the European population' is included as missing information in the RMP and an observational post-authorization study in pregnant women in EU is planned as part of the pharmacovigilance plan. Adverse events of special interest were not recorded or defined in the pivotal clinical studies as per respective study protocols.

VacPertagen includes 5µg of PT_{gen}, which is considered as high dosage not comparable to the reduced dosages of chemically detoxified PT included in approved Tdap vaccines. Whether repeated doses with VacPertagen could lead to an increase of reactogenicity in adolescents and adults, including pregnant women, as observed when DTaP were administered as 4th and/or 5th doses is unclear.

All studies were done on healthy subjects. None were performed in patients with relevant co-morbidities such as clinically significant renal, hepatic, cardiac impairment, or immunodeficiency. However, the safety profile in these populations is not expected to differ significantly from the targeted population and thus, routine pharmacovigilance suffices for further characterising it.

3.6. Effects Table

Table 128. Effects Table for VacPertagen

Effect	Short Description	Unit	Treatment	Control	Uncertainties / Strength of evidence	References
Favourable Effects						
Anti-PT and seroconversion rates (ELISA)	Peripheral blood sample from adolescent subjects, 28 days after vaccination	%	VacPertagen (n=148) PT: 96% (95% CI 93-99) FHA: 93% (95% CI 89-97)	Adacel (n=149) PT: 55% (95% CI 47-63) FHA: 54% (95% CI 46-62)	Immunogenicity NI comparison with uncertainties; descriptive analysis unambiguous; PT-ELISA results consistent with neutralizing antibody responses (CHO assay)	TDA202

Effect	Short Description	Unit	Treatment	Control	Uncertainties / Strength of evidence	References
Anti-PT and anti-FHA Geometric Mean Titers (GMTs):	Peripheral blood sample from adolescent subjects, 28 days after vaccination	IU/mL	VacPertagen (n=148) PT: 562 IU/mL (95% CI 467.79-674.86) FHA: 924 IU/mL (95% CI 809.39-1054.4)	Adacel (n=149) PT: 63 IU/mL (95% CI 51.05-78.37) FHA: 242 IU/mL (95% CI 208.86-280.05)	Immunogenicity NI comparison with uncertainties; descriptive analysis unambiguous; PT-ELISA results consistent with neutralizing antibody responses (CHO assay)	TDA202
Anti-PT and seroconversion rates (ELISA)	Peripheral blood sample from adult subjects, 28 days after vaccination	%	VacPertagen (n=75) PT: 100.00% (95% CI 95.20-100.00) FHA: 97.33% (95% CI 90.70-99.68)	Adacel (n=74) PT: 74.32% (95% CI 62.84 - 83.78 FHA: 93.24% (95% CI 84.93-97.77)	Descriptive analysis, low sample size in the elderly but consistent results with younger subjects	TDA206
Anti-PT and anti-FHA Geometric Mean Titers (GMTs):	Peripheral blood sample from adult subjects, 28 days after vaccination	IU/mL	VacPertagen (n=75) PT: 371.83 IU/ml (95% CI 292.76-472.25) FHA: 451.62 IU/mL (95% CI 373.46-546.12)	Adacel (n=74) PT: 50.84 IU/ml (95% CI 39.26-65.84) FHA: 207.58 IU/mL (95% CI 171.33-251.50)	Descriptive analysis, low sample size in the elderly but consistent results with younger subjects	TDA206
Anti-PT IgG GMC (ELISA)	Cord blood sample at delivery (or neonatal blood within 72 hours after birth)	IU/mL	VacPertagen (n=35) 141.40 IU/mL (95% CI: 94.70 – 211.12)	Adacel (n=35) 27.09 IU/mL (95% CI: 18.21 – 40.31)	Descriptive analysis, Per Protocol Population, low sample size but results consistent with D28 post-vaccination sample in mothers and 2-month data in infants	TDA207
Unfavourable Effects						

Effect	Short Description	Unit	Treatment	Control	Uncertainties / Strength of evidence	References
Immediate solicited reactions	Solicited events within 30 minutes after vaccination	% of Subjects with at least one event	VacPertagen (Safety data pool, n=2508) 17.5%	Adolescent TDA202 (Adacel, n=150): 0% Adult TDA206 (Adacel, n=150): 8% with local and 0% with systemic reaction Pregnant Women TDA207 (Adacel, n=40): 0% Adult APV301 (Boostrix, n=300): 27.33% Adult Pertaprime-01 (Boostrix, n=34): 17.65% with local and 20.59 with systemic reaction		Safety data and safety data pool from studies: TDA202, TDA206, TDA207, Pertaprime-01 and APV301
Solicited reactions	Solicited events within 7 days after vaccination	% of Subjects with at least one event	VacPertagen (Safety data pool, n=2507) 60.75%	Adolescent TDA202 (Adacel, n=150): 14.67% Adult TDA206 (Adacel, n=150): 76% with local and 50% with systemic reaction Pregnant Women TDA202 (Adacel, n=40): 82.5% Adult APV301 (Boostrix, n=299): 67.89% Adult Pertaprime-01 (Boostrix n=34): 79.41% with local and 79.41 with systemic reaction		Safety data and safety data pool from studies: TDA202, TDA206, TDA207, Pertaprime-01 and APV301

Effect	Short Description	Unit	Treatment	Control	Uncertainties / Strength of evidence	References
Adverse events	Unsolicited adverse events from vaccination until day 28	% of Participants with at least one event from vaccination until day 28	VacPertagen (Safety data pool, n=2507) 12.56%	Adolescent TDA202 (Adacel, n=150): 32.67% Adult TDA206 (Adacel, n=150): 22% Pregnant Women TDA207 (Adacel, n=40): 22.5% Adult APV301 (Boostrix, n=299): 6.02% Adult Pertaprime-01 (Boostrix n=34): 44.12%		Safety data and safety data pool from studies: TDA202, TDA206, TDA207, Pertaprime-01 and APV301
Serious Adverse Events	Events from vaccination until day 28	% of subjects with at least one event	VacPertagen Adolescent TDA202 (n=150): 0% Adult TDA206 (n=150): 0% Pregnant Women TDA207 (n=40): 5% Adult APV301 (n=2099): 0.1% Pertaprime-01 (n=68): 1.47%	Adolescent TDA202 (Adacel, n=150): 0% Adult TDA206 (Adacel, n=150): 0.67% Pregnant Women TDA207 (Adacel, n=40): 5% Adult APV301 (Boostrix, n=300): 0% Adult Pertaprime-01 (Boostrix, n=34): 0%	Conclusions on (serious) events in infants born to vaccinated mothers are uncertain due to the long time from vaccination and due to the low patient numbers included in study TDA207.	Safety data from studies TDA202, TDA206, TDA207, APV301, Pertaprime-01

Abbreviations: GMC=geometric mean concentration, PT=pertussis toxin, FHA=filamentous haemagglutinin

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

VacPertagen offers advantage over widely used Tdap vaccines by enabling selective booster immunisation against pertussis without administering antigens from other pathogens for which booster immunisation might not be needed notably for maternal immunisation.

The presented immunogenicity data show that a single booster dose with VacPertagen elicits a considerable humoral immune response, as determined by anti-PT/anti-FHA IgG (ELISA) and by neutralizing antibodies (functional CHO based assay) in (non-pregnant) adult (Study TDA206) and

adolescent (Study TDA202) individuals. This was also observed in pregnant women, cord blood samples at delivery, and by 2-month data in infants.

The clear and consistent trend of increased immunogenicity of VacPertagen compared to Adacel and Boostrix across all submitted studies does compensate for the methodological shortcomings and the immunogenicity of VacPertagen is not questioned.

Booster vaccination with VacPertagen in adults and adolescents consistently induces high anti-PT and anti-FHA responses (a least comparable to the vaccine comparators) in the different trials. Antibodies against PT and FHA have been demonstrated to wane in parallel with declining effectiveness. In addition, the available non-clinical data also suggest a protective effect of PT and FHA in mouse challenge models. Additional non-clinical and structural data discussed by the Applicant indicate that important epitopes for neutralising antibody responses are retained in VacPertagen whereas such conformational epitopes might be lost after chemical inactivation as used in the approved comparator vaccines.

The Applicant also seeks an indication for passive protection against pertussis in early infancy following maternal immunisation during pregnancy. Until now, no mono- or two-component pertussis vaccine is approved to be administered during pregnancy for passive protection in Europe. However, antibodies against PT can be considered as a major contributor to protection against pertussis. Furthermore, a recent real-world effectiveness study conducted in Denmark (Kildegaard et al. 2025) investigated a 1-component vaccine (DiTekiBooster) which was used temporarily (off-label) due to a surge of pertussis cases in 2019. This study can be considered as a proof of concept that a 1-component vaccine (including only pertussis toxin) may be sufficient to provide protection against laboratory-confirmed pertussis, even if the antibodies were only passively transferred from mothers to their infants.

The results of study TDA207 in pregnant women are consistent with an observational post marketing study (in Thailand) and with a supportive randomised controlled study investigating Boostagen (Td-VacPertagen) and lower-dose formulations, indicating considerable antibody levels and maternal transfer of antibodies into cord blood after vaccination during pregnancy. However, clinical data from study TDA207 and the supportive studies TDA204 and WoMANPOWER in pregnant mothers and their infants suggests a strong "blunting" effect of maternal vaccination on their infant's response to primary vaccination. This was characterised by lower levels of anti-PT IgGs at month 7 (one month after primary vaccination with three doses of mostly whole cell vaccines), compared to month 2 (pre-priming). While blunting is a known phenomenon for licensed pertussis containing vaccines, it was more pronounced with VacPertagen (vs. the comparator groups), at least with respect to anti-PT IgG responses. The clinical relevance of this observation is not known but infants are most vulnerable to pertussis during the first months of life and infants of women who received VacPertagen during pregnancy indicated high GMCs of anti-PT IgGs within this time period. Overall, based on the robust anti-PT (binding and neutralising) and anti-FHA (binding) antibody levels in infants (at birth and at 2 months of age), the CHMP concluded that it is reasonable to assume that VacPertagen will be effective in the setting of passive protection against pertussis in early infancy following maternal immunisation during pregnancy. However, uncertainties remain on the magnitude and duration of the assumed protection of newborns. Effectiveness will be confirmed post-marketing. To inform NITAGs, the observed interference with induction of PT-specific immune response to primary immunisation with DTwP/DTaP in infants born to women vaccinated with VacPertagen during pregnancy is described in the product information.

For adult and adolescent subjects, the reported safety results on immediate reactions, solicited events and AEs (including SAEs) do not cause immediate reason of concern, as the reported pattern of events appears to cover an expected safety profile, severe and serious AEs were not very common and no discontinuation due to AEs were reported. Safety data pooling across studies was provided and ADRs

were defined based on this pool. No adverse events of special interest were defined or assessed during clinical trials. However, the safety database collected for adult and adolescent subjects in a RCT setting after vaccination with VacPertagen is sufficiently large to describe the safety profile of VacPertagen (n=2508 adult and adolescent subjects followed after vaccination with VacPertagen in RCTs).

Reported imbalances in events around delivery as observed in study TDA207 (pregnant women) remain highly uncertain, due to the very low patient numbers vaccinated with VacPertagen alone, the fact that no critical events were considered vaccine-related and concerning imbalances were not seen to the same extent for subjects vaccinated with Boostagen (n=80 in TDA204 and n=40 in TDA207). Notably, none of the supportive information from observational studies and post-marking data did identify any specific concern regarding the pregnancy outcomes after vaccination with VacPertagen. Chance findings appear likely based on the low number of pregnant women observed in trial TDA207. Altogether, the safety risk profile for vaccination during pregnancy seems acceptable as no immediate safety concerns were identified regarding the vaccination in pregnant women from available information. Still, additional safety data from ongoing perspective trials (PERTg001 and BOOSTg001) and post-marketing studies (APV302 and PASS) are highly encouraged to further resolve remaining uncertainties.

Limited safety information is also available for the European population but based on the sparse data reported (only in study PertADO in adolescent subjects), a higher rate in solicited reactions and adverse events compared to the population in Thailand seems evident. It is unclear whether the geographical or demographical setup might have contributed to the discrepancy, but in consequence higher event rates than those reported in pivotal clinical trials from Thailand cannot be excluded for the European population. The clinical relevance of these findings do not impact the overall benefit-risk balance of VacPertagen.

3.7.2. Balance of benefits and risks

The immunogenicity data consistently showed that a booster dose with VacPertagen elicited increased antibody responses against PT and FHA compared to Adacel or Boostrix in adolescents, adults and pregnant subjects approximately 1 month after vaccination. Although there is no established serological correlate of protection, a reasonable vaccine effectiveness against pertussis in adolescents and adults could be assumed for VacPertagen based on the generally higher antibody responses against PT (the major mediator of pathogenicity) compared to approved vaccines.

Based on the robust anti-PT (binding and neutralising) and anti-FHA (binding) antibody levels in infants (at birth and at 2 months of age), the CHMP concluded that it is reasonable to assume that VacPertagen will be effective in the setting of passive protection against pertussis in early infancy following maternal immunisation during pregnancy. However, uncertainties remain on the magnitude and duration of the assumed protection of newborns. Effectiveness will be confirmed post-marketing.

The clinical safety database that is submitted for the current MA is sufficiently large to support licensure in adult and adolescent subjects, but long-term safety data are not abundant. Solicited and unsolicited events were well described, and no specific safety concern has been identified by the available data in non-pregnant subjects. However, the safety database from RCTs on vaccinations in pregnant women is small and had to be supported by post-authorisation data in non-EU countries where VacPertagen has been authorised since 2016. This includes prospective and retrospective post-authorisation observational studies with both, VacPertagen and Boostagen as well as spontaneous reports. To date, no safety concern has been identified in pregnant women. Nevertheless, a post-authorisation safety study in pregnant women is also planned in Europe, to collect structured data and further characterise the safety profile in this population.

Overall, the benefit-risk balance of VacPertagen is positive for the applied indications.

3.8. Conclusions

The overall benefit/risk balance of VacPertagen is positive, subject to the conditions stated in section 'Recommendations'.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the benefit-risk balance of VacPertagen is favourable in the following indication(s):

VacPertagen is indicated for:

• booster immunisation against pertussis of individuals 12 years of age and older,

• passive protection against pertussis in early infancy following maternal immunisation during pregnancy (see sections 4.4, 4.6 and 5.1).

The use of this vaccine should be in accordance with official recommendations.

The CHMP therefore recommends the granting of the marketing authorisation subject to the following conditions:

Conditions or restrictions regarding supply and use

Medicinal product subject to medical prescription.

Official batch release

In accordance with Article 114 Directive 2001/83/EC, the official batch release will be undertaken by a state laboratory or a laboratory designated for that purpose.

Other conditions and requirements of the marketing authorisation

- **Periodic Safety Update Reports**

The requirements for submission of periodic safety update reports for this medicinal product are set out in the list of Union reference dates (EURD list) provided for under Article 107c(7) of Directive 2001/83/EC and any subsequent updates published on the European medicines web-portal.

The marketing authorisation holder shall submit the first periodic safety update report for this product within 6 months following authorisation.

Conditions or restrictions with regard to the safe and effective use of the medicinal product

- **Risk Management Plan (RMP)**

The marketing authorisation holder (MAH) shall perform the required pharmacovigilance activities and interventions detailed in the agreed RMP presented in Module 1.8.2 of the marketing authorisation and any agreed subsequent updates of the RMP.

An updated RMP should be submitted:

- At the request of the European Medicines Agency;
- Whenever the risk management system is modified, especially as the result of new

information being received that may lead to a significant change to the benefit/risk profile or as the result of an important (pharmacovigilance or risk minimisation) milestone being reached.