

3.2.S.4.1. SPECIFICATION – [OMICRON (XBB.1.5) VARIANT]

The specification for Omicron (XBB.1.5) drug substance is presented in Table 3.2.S.4.1-1.

Table 3.2.S.4.1-1. Specification for Omicron (XBB.1.5) Drug Substance

Quality Attribute	Analytical Procedure	Procedure Number(s)	Acceptance Criteria
Composition and Strength			
Clarity	Appearance (Clarity)	Ph. Eur. 2.2.1	4.2 1st ind
Coloration	Appearance (Coloration)	Ph. Eur. 2.2.2	4.2 1st ind
pH	Potentiometry	Ph. Eur. 2.2.3, USP <791>	4.2 1st ind
Content (RNA Concentration)	UV Spectroscopy	LAB-37658 (PGS Andover) SOP-10071 4.2 1st ind SOP-117646 4.2 1st ind	4.2 1st ind
Identity			
Identity of Encoded RNA Sequence	ddPCR for Identity ^a	LAB-44995 (PGS Andover) SOP-123668 4.2 1st ind	Identity confirmed
	RT-PCR for Identity ^{a, b}	TM-072-070 (BNT Mainz)	
Purity			
RNA Integrity	Capillary Gel Electrophoresis	LAB-41057 (PGS Andover) TM-072-039 (BNT Mainz) SOP-116920 4.2 1st ind	4.2 1st ind
5'- Cap	RP-HPLC	LAB-37879 (PGS Andover) PAN-1239-K 4.2 1st ind PAN-1253-K 4.2 1st ind SOP-117195 4.2 1st ind	4.2 1st ind
Poly(A) Tail	ddPCR	LAB-38823 (PGS Andover) PAN-1280-K 4.2 1st ind SOP-117642 4.2 1st ind	4.2 1st ind
Poly(A) Tail Length	IP-RP-HPLC ^a	LAB-41956 (PGS Andover) SOP-501868 4.2 1st ind SOP-117248 4.2 1st ind	Poly(A) Tail Length Confirmed
Process Related Impurities			
Residual DNA Template	qPCR ^a	LAB-42528 (PGS Andover) TM-072-028 (BNT Mainz) SOP-118245 4.2 1st ind	4.2 1st ind
Product Related Impurities			
dsRNA	Immunoblot ^a	LAB-42529 (PGS Andover) PAN-1356-K 4.2 1st ind SOP-117356 4.2 1st ind	4.2 1st ind

Table 3.2.S.4.1-1. Specification for Omicron (XBB.1.5) Drug Substance

Quality Attribute	Analytical Procedure	Procedure Number(s)	Acceptance Criteria
Safety			
Bacterial Endotoxins	Endotoxin (LAL)	Ph. Eur. 2.6.14, USP <85>, JP 4.01	4.2 1st ind
Bioburden	Bioburden	Ph. Eur. 2.6.12, USP <61>, JP 4.05	4.2 1st ind

a. Assay not performed on stability

b. Analytical Procedure used only at BioNTech Mainz

Abbreviations: NTU = nephelometric turbidity units; B = brown; RT-PCR = reverse transcription polymerase chain reaction; RP-HPLC = reverse phase high performance liquid chromatography; ddPCR = droplet digital PCR; IP-RP-HPLC = Ion-pair-reversed-phase high performance liquid chromatography; qPCR = quantitative PCR; dsRNA = double stranded RNA; LAL = limulus amoebocyte lysate; EU = endotoxin unit; CFU = colony forming unit; PGS = Pfizer Global Supply; BNT = BioNTech