



European Medicines Agency
Evaluation of Medicines for Human Use

REFUSAL ASSESSMENT REPORT

FOR

Alpheon

International Nonproprietary Name:
recombinant human interferon-alfa-2a

Procedure No. EMEA/H/C/000585

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

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1. BACKGROUND INFORMATION ON THE PROCEDURE

1.1 Submission of the dossier

The applicant BioPartners GmbH submitted on 22 December 2003 an application for a Marketing Authorisation to the European Medicines Agency (EMA) for Alpheon 6 million IU/ml solution for injection. The application was made through the centralised procedure and was validated under the legal basis of Similar Biological Medicinal Product under Article 10(1)(a)(iii) of Directive 2001/83/EC, and with reference to Part II.4 of Annex I of Directive 2001/83/EC, as amended.

Scientific Advice

The applicant received first Scientific Advice from the CHMP (ex CPMP) on 24 March 1999. This Scientific Advice pertained to the clinical aspects of the dossier. The second Scientific Advice (Follow-up request) from the CHMP was given on 1 March 2001.

Licensing status

The product was not licensed in any country at the time of submission of the application.

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

Rapporteur: Harald Enzmann

Co-Rapporteur: Ian Hudson

1.2 Steps taken for the assessment of the product

- The application was received by the EMA on 22 December 2003.
- The procedure started on 21 June 2004.
- The Rapporteur's first Assessment Report was circulated to all CHMP members on 31 August 2004. The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on 31 August 2004.
- During their meeting 11-13 October 2004, the BWP adopted a first report to the CHMP.
- During the meeting 18-21 October 2004, the CHMP agreed on the consolidated List of Questions to be sent to the applicant. The final consolidated List of Questions was sent to the applicant on 21 October 2004.
- The applicant submitted the responses to the CHMP consolidated List of Questions on 16 December 2005.
- The summary report of the inspection carried out at the LG Life Sciences Ltd site between 2 and 10 March 2005 was issued on 16 June 2005. An inspection of the Impfstoffwerk Dessau-Tornau GmbH manufacturing site in Rodleben/ Ortsteil Tornau was carried out on 10-15 November 2005. A summary report was issued on 4 May 2006.
- The Rapporteurs circulated the Joint Assessment Report on the applicant's responses to the List of Questions to all CHMP members on 20 January 2006.
- During their meeting 13-15 February 2006, the BWP discussed the major outstanding quality questions and adopted a second report to the CHMP.

- During the CHMP meeting on 20-23 February 2006, the CHMP agreed on a list of outstanding issues to be addressed in writing and in an oral explanation by the applicant at the 22-24 May 2006 BWP meeting and at the 29 May-1 June 2006 CHMP meeting.
- The applicant submitted the responses to the list of outstanding issues on 26 April 2006.
- The Rapporteurs circulated the Joint Assessment Report on the applicant's responses to the List of Outstanding Issues to all CHMP members on 15 May 2006.
- During the BWP meeting 22-24 May 2006, outstanding issues were presented by the applicant during an oral explanation. The BWP adopted a third report to the CHMP.
- During the CHMP meeting 29 May-1 June 2006, outstanding issues were presented by the applicant during an oral explanation.
- During the meeting on 26-28 June 2006, the CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a negative opinion for granting a Marketing Authorisation to Alpheon on 28 June 2006.
- The CHMP opinions were forwarded, in all official languages of the European Union, to the European Commission, which adopted the corresponding Decisions on 5 September 2006.

2. GENERAL INFORMATION ON THE MARKETING AUTHORISATION APPLICATION

Applicant

BioPartners GmbH
Eisenstrasse 3
D-65428 Rüsselsheim
Germany

Manufacturer(s) of the active substance

LG Life Sciences, Ltd., Iksan Plant 601 Yongje-dong, Iksan-si, Jeonbuk-do 570-350, South Korea.

An inspection of this manufacturing site was carried out by the competent authorities Regierungspraesidium Darmstadt, Landesverwaltungsamt Saxony-Anhalt and an expert from the BfArM. The facility was found to be in compliance with the Community Good Manufacturing Practice requirements (GMP).

Manufacturer(s) of the finished product

Manufacturer of the Drug Product

Impfstoffwerke Dessau-Tornau GmbH, Streetzer Weg 15a, D-06862 Rodleben, Germany.

An inspection of this manufacturing site was carried out by the competent authority Landesverwaltungsamt Saxony-Anhalt and two of the Rapporteur's assessors (BfArM)

Manufacturer responsible for batch release

BioPartners GmbH, Eisenstrasse 3, 65428 Rüsselsheim, Germany.

3 SCIENTIFIC DISCUSSION

3.1 Introduction

BioPartners GmbH applied for a marketing authorisation for Alpheon 6 million IU/ml solution for injection. Alpheon contains recombinant human interferon-alfa-2a as active substance. The application was submitted under the legal basis of Similar Biological Medicinal Product under Article 10(1)(a)(iii) of Directive 2001/83/EC, and with reference to Part II.4 of Annex I of Directive 2001/83/EC, as amended.

The reference medicinal product for this application was Roferon-A Solution for Injection, a recombinant interferon alfa 2a containing product expressed in *E. coli* produced by Roche originally authorised in the EU in 1986. Alpheon was claimed to be similar to this reference medicinal product as approved in the Community.

As required for a Similar Biological Medicinal Product application, the dossier contained a full quality Module 3 and a reduced non-clinical and clinical Modules 4 and 5, with the required elements of the comparability exercise, respectively as required by the CHMP guidelines.

The indication applied for was as follows: Adult patients with histologically proven chronic hepatitis C who are positive for HCV antibodies or HCV RNA and have elevated serum alanine aminotransferase (ALT) without liver decompensation. The efficacy of Interferon-alfa-2a in the treatment of hepatitis C is enhanced when combined with ribavirin. Alpheon should be given alone mainly in case of intolerance or contra-indication to ribavirin.

3.2 Quality aspects

Introduction

The active ingredient of Alpheon was recombinant human interferon alfa-2a expressed in *Saccharomyces cerevisiae*. Human interferon- α 2a protein consists of 165 amino acids with a molecular weight of approx. 19,240 kD and two disulfide bonds.

Alpheon drug product was presented in multidose vials as a ready-to-use solution for injection.

Active Substance

- Manufacture

Cell bank system

The drug substance for Alpheon was produced from a sufficiently well-defined and characterised *S. cerevisiae* cell bank system. Genetic stability was demonstrated during production, at the end of production and in post-production cells, indicating sufficient genetic stability.

Manufacturing Process

The manufacturing process consists of a fermentation process where the secreted product was purified by a protein purification process comprising 5 chromatography and 3 ultrafiltration/diafiltration steps in order to remove process and product-related impurities and reduce product-related substances.

Process validation was performed on small scale as well as commercial scale conditions. All process steps were validated and included in-process and manufacturing controls.

Manufacturing Process Development

The manufacturing process of the drug substance, which was originally developed in 1990, was modified in 1999 in the following ways:

- (1) the replacement of the animal derived bacto-peptone by yeast-derived peptone; and
- (2) the introduction of a second cation exchange chromatography step.

The manufacturing process remained unchanged throughout the Alpheon development programme.

Characterisation

State-of-the-art analytical methods were used to thoroughly investigate recombinant human interferon- α 2a with regard to its structural, physico-chemical, immunological and biological characteristics. The drug substance was considered to have been sufficiently characterised.

The following analytical methods were used:

- Physico-chemical characterisation

Reducing / Non-Reducing SDS-PAGE, Isoelectric Focusing, Extinction Coefficient UV Spectroscopy, Circular Dichroism, Size Exclusion HPLC (SEC-HPLC), Reverse-phase HPLC and Mass Spectrometry

- Structural characterization and conformation

Peptide Mapping, Disulfide Bridges, N-terminal Sequence, C-terminal Sequence, Analysis of Amino Acid Composition, Free Cystein , Carbohydrate Structure

- Biological activities and immunological properties

SDS-PAGE/Western blotting, Epitope Mapping, Enzyme-linked Immunoassay (ELISA), Potency

- Purity

Size Exclusion HPLC (SEC-HPLC), Reverse-phase HPLC, SDS-PAGE, CZE, CM-HPLC.

It was concluded that the drug substance was sufficiently well characterised based on the data provided. However, differences in impurity profile were observed.

Information about the related substances was still insufficient to determine their impact on Alpheon quality and characteristics.

Impurities

Process-related impurities were analysed as well as product-related impurities and product-related substances arising either from the culture/fermentation process or from the purification process (downstream processing), demonstrating sufficient removal of process-related and product-related impurities and reduction of process-related substances.

Control of Drug Substance

The proposed release specifications have been selected taking into account the properties and characteristics of the drug substance. The proposed test items address the physical state, identity, purity and content as well as the presence of a number of related substances and the general safety of the product.

The analytical methods were improved according to state-of-the-art requirements during the procedure and have been validated and harmonised for the drug substance and the drug product, taking into account the large difference in protein concentration (mg/ml) between the drug substance and drug product.

- Stability

Thirty-six month real-time stability data on four batches were presented in the application. Due to the improvement of the analytical methods, new stability data were requested since a retrospective analysis of older drug substance batches was not considered sufficient. A shelf-life could not be assigned at this stage.

Finished Product

Alpheon was presented as a liquid formulation in multidose vials containing 6 individual doses 0.5 mL each. Each 0.5 mL dose represents an activity of 3×10^6 IU interferon alpha-2a corresponding to a label claim of 6×10^6 IU/mL.

- Pharmaceutical Development

Originally, the (lyophilised) product contained human serum albumin (HSA) as a stabilizer. During the development of the liquid formulation, HSA was replaced by an alternative stabilizing agent and preservative system. This formulation was used in all pivotal nonclinical and clinical studies.

- Manufacture

Alpheon finished product is manufactured from drug substance that was compounded with the excipient solutions and aseptically filled into vials.

Process validation

Extensive validation of the manufacturing process was performed; however, unexpected difficulties were encountered during the upgrading of the filling line at the drug product manufacturer IDT, and the process could not be regarded as validated. Thus, the drug product production process was not considered to be fully validated and there was no assurance of control and consistency of the manufacturing process.

Analytical Validation

As a consequence of the improved analytical methods for the drug substance and with the intention to harmonise the methods for drug substance and drug product considering the difference in protein concentration, the methods employed in the testing of the drug product were also revised. Validation of the methods was performed.

Comparability of clinical batches vs. commercial batches

Quantitative differences in the related substance profile of the clinical batches and the recently produced batches of Alpheon were demonstrated. The applicant assumed that the differences were due to ageing of the samples. Conclusions could not be drawn from these experiments with aged samples due to lack of data on the stability profile of Alpheon.

Reliable release data proving comparability of clinical and the recent batches in terms of related substances were not available. Therefore, uncertainties remained regarding the comparability of clinical trial material with the proposed commercial product.

Container Closure System

Alpheon was presented in a 4.0 mL borosilicate Type I glass vial which was closed with a coated butyl rubber stopper and covered with an aluminium overseal and a polypropylene flip-off cap.

Compatibility

Compatibility will be assessed through the ongoing stability studies and in addition on the basis of product-specific migration data on potential leachables and extractables from the coated/laminated stopper material with the drug product.

- Stability of the Product

Sufficient stability data representative of the intended commercial drug product tested with the revised analytical methods were not available and a shelf life could not be assigned.

- Comparability exercise of Alpheon vs. Roferon-A

A number of factors impacted on any study designed to compare Alpheon drug substance and Roferon-A. These included the unavailability of Roferon-A drug substance, formulation effects, and effects of sample preparation. The difficulties of performing a like to like comparison were well recognised.

The applicant designed a number of studies taking into account the above-mentioned factors. The applicant identified that Alpheon drug substance was structurally comparable to the Interferon alfa in Roferon-A. However, while there were similarities between Alpheon drug substance and Roferon-A drug product in terms of the product-related substance profile, the applicant also demonstrated that there were some differences.

Data comparing the two drug products were considered to be incomplete and inconclusive. Alpheon Drug Product manufactured with a validated process representative for the proposed commercial process was not available. As a result of the aforementioned deficiencies and the observed differences in the quantitative and qualitative product-related substance profile, no conclusion regarding the comparability of Alpheon and Roferon-A could be reached.

Issues regarding the samples of Alpheon used (currently no validated drug product) and the impact of sample preparation (concentration procedures and presence of excipients) on the analytical methods were unresolved.

Adventitious Agents Safety Evaluation

No materials of animal origin were used in the manufacture of the drug substance. No human- or animal-derived materials were used in the formulation of the medicinal product.

GMP issues

Compliance with general GMP principles was demonstrated for all manufacturing sites.

Discussion on chemical, pharmaceutical and biological aspects

Active substance

The active ingredient of Alpheon was recombinant human interferon alfa-2a expressed in *Saccharomyces cerevisiae*. Human interferon- α 2a (IFN- α 2a) was a characterized protein consisting of 165 amino acids.

The primary structure of Alpheon was identical to that of a marketed recombinant human interferon alfa 2a product expressed in *E. coli* (Roferon-A). Similarity was claimed to Roferon-A.

The cell banking system for Alpheon was three tiered and included a Master Cell Bank, Intermediate Cell Bank, and Working Cell Banks.

The fermentation process was a two stage process with the main fermentation at 50 L scale. The production organism *Saccharomyces cerevisiae* secreted the drug substance into the fermentation medium. The drug substance was recovered from the fermentation both by a conventional protein purification process comprising 5 chromatography and 3 ultrafiltration/diafiltration steps.

Based on the data presented, it could be concluded that the drug substance was sufficiently well characterised. However, new information on the related substances/ impurities in Alpheon emerged at a very late stage in the assessment process, thus putting into question the previous data provided and the conclusions drawn from it.

Regarding the drug substance stability, the applicant based their response on re-evaluation of old data against the new specifications. This can provide some information but was insufficient if presented without confirmation using the current test methods and specifications.

The proposed shelf-life for the drug substance was not supported by adequate data. The presented data were not regarded as sufficient to support the shelf life of the drug substance at the recommended storage and transportation conditions.

Finished Product

The drug product was presented as a liquid formulation in multidose vials. Target fill volume was 3.60 ml including a 20 % overfill (6 x 0.1 ml) which was stated to be required for the removal of 6 individual doses 0.5 mL each.

The manufacturing process for Alpheon comprised the preparation of the solution, sterile filtration, aseptic filling/closing and secondary packaging.

After the introduction of a new filling line, three new validation batches were manufactured in December 2005. The new validation results demonstrated that the filling process was not sufficiently under control. Major concerns existed regarding the control of the drug product production process. These could only be answered by a full and satisfactory validation of the drug product production process, which takes into account experience during previous failed validations. Thus, the drug product production process was not considered to be fully validated and there was no assurance of adequate control and consistency of the manufacturing process.

It should be highlighted that in the original dossier the applicant had not used a harmonised set of analytical methods for the assessment of related proteins in Alpheon drug substance and drug product. This lack of adequate method transfer led to the significant differences seen in related proteins between the drug substance and the drug product.

ICH compliant stability data were not available for the drug product, since the most recent production batches were not be considered to be validation batches. As a result, a shelf life for Alpheon could not be assigned. An adequate stability study would require validation and finalisation of the manufacturing method and controls. In addition, the impact of the new impurity detected in all batches of the new stability study, needed to be fully evaluated and a specification set prior to the conduct of new stability studies.

Comparability of Alpheon vs. Roferon-A

The data provided for the Alpheon and Roferon-A finished products did not support the key claim of comparability. Assays where product related impurities had been investigated indicated that Alpheon and Roferon-A displayed a different impurity profile. The precise nature, biological properties, and the qualitative and quantitative composition of the different impurities had not been fully studied. Differences in the stability profile also required investigation.

It was therefore concluded that the data comparing the two drug products were incomplete and inconclusive. Issues regarding the samples of Alpheon used (currently no validated manufacturing process) and the impact of sample preparation (concentration procedures and presence of excipients) on the analytical methods were unresolved.

As a consequence of the deficiencies set out above and the quantitative and qualitative differences in the impurity profile, no conclusion regarding the comparability of Alpheon and Roferon-A based upon the available quality data could be reached.

Comparability of clinical batches vs. commercial batches

In addition, the applicant was unable to satisfactorily address the comparability of the clinical trial material with the drug product produced from the intended commercial process. The lack of validation of analytical methods used as well as validation of the production process made this task even more difficult.

The applicant provided retrospective analysis of retained samples, and as much information as possible about the clinical trial material. However, the retrospective analysis of related substances was not considered satisfactory. Quantitative differences in the related substance profile of the clinical batches and the recently produced batches of Alpheon were demonstrated. The applicant considered that these differences were due to ageing of the samples. However, no firm conclusions could be drawn from these experiments with aged samples due to lack of data on the stability profile of Alpheon.

Reliable release data proving comparability of clinical and the recent batches in terms of related substances were not available. Therefore uncertainties remained regarding the comparability of clinical trial material with the proposed commercial product.

3.3 Non-clinical aspects

Introduction

The objectives of the applicant in terms of the non-clinical aspects for Alpheon were to establish its activity as an IFN- α -2a and to demonstrate similarity regarding safety and efficacy to Roferon-A.

In accordance with the *CHMP-Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues (EMEA/CHMP/42832/2005)*, the demonstration of similarity in terms of quality, safety, and efficacy of a medicinal product to another one already authorised in the EU, required an extensive comparability exercise. The required non-clinical studies should be comparative in nature.

For this purpose the applicant submitted an *in vitro* study comparing the activity of Alpheon and Roferon-A in terms of antiviral activity and induction of interferon-sensitive genes together with a 4-week subcutaneous repeated-dose toxicity study.

In addition, the applicant also submitted several non-clinical studies, on pharmacology, pharmacokinetics, and toxicology, which were conducted in the early 1990s with a HSA-stabilised, lyophilised formulation of Alpheon derived from an earlier manufacturing process.

GLP

The 4-week repeated-dose toxicity study in monkeys was conducted in compliance with GLP.

The non-clinical toxicology studies conducted in Korea prior to 1992 were not performed under current GLP regulations, but were in agreement with the regulations for non-clinical studies in force in that country at the time.

Pharmacology

- Primary pharmacodynamics

An *in vitro* study comparing the activity of Alpheon and Roferon-A in terms of antiviral activity against encephalomyelitis virus (EMCV) in two cell lines (A549 cells and HuH7 cells) and gene induction (response to interferon in HuH7 hepatocytes) were performed. A standard interferon α -2a preparation from the National Institute for Biological Standards and Control (NIBSC) was also included as a control.

These preparations were compared in antiviral assays in A549 cells and HuH7 cells using EMCV as the challenge virus and in a gene induction assay in HuH7 cells measuring the induction of the interferon sensitive genes for 2', 5'-oligoadenylate synthetase (2', 5'-OAS) and the human myxovirus resistant protein, MxA, after interferon treatment.

All three interferon alfa-2a preparations exhibited antiviral activity and induced the interferon sensitive genes 2', 5'-OAS and MxA. This was consistent with the known properties of interferon- α -2a. The antiviral activity of the preparations in A549 cells was nearly comparable, but a high variability was observed in the antiviral assay in HuH7 cells for each preparation. Furthermore, considerable differences were observed between the potencies of the different preparations in the gene induction assays, again performed in HuH7 cells.

In A549 cells challenged with EMCV the antiviral activity of the three IFNs was nearly comparable. In contrast, in HuH7 cells challenged with EMCV the antiviral activity of each IFN was extremely variable probably due to difficulties in handling these cells and exclusion of many single outlier measurements. Therefore, a comparison between the different IFNs was not possible in this assay.

In the gene induction assay all three preparations induced the IFN-sensitive genes 2', 5'-OAS and MxA. However, for each interferon a high variability in the magnitude of gene induction was observed and no comparability between the IFNs could be found.

In conclusion, based on this comparative study no definitive statement on the similarity of Alpheon and Roferon-A could be made.

- Secondary pharmacodynamics / safety pharmacology

No specific studies on safety pharmacology of Alpheon have been conducted. Several parameters on safety pharmacology (respiratory rate, heart rate ECG) have been measured during the repeated dose toxicity study in monkeys. No major findings were reported.

Toxicology

- Repeat dose toxicity (with toxicokinetics)

A repeated dose toxicity study in female rhesus monkeys comparing Alpheon and Roferon-A was submitted. This study was a 4-week repeated dose study with an immunological response evaluation in female Rhesus monkeys with a 2-week recovery period. The animals, assigned to 5 groups (3/dose-group), received 6 million IU/kg/dose (Alpheon with preservatives or Roferon-A) or 12 million IU/kg/dose (Alpheon in formulations with and without preservatives) or Alpheon vehicle (control group) every other day.

During this study the following parameters were monitored:

- Toxicological investigations (clinical signs, blood parameters)
- Biological activity (serum neopterin levels)
- Pharmacokinetic parameters: serum IFN- α levels
- Immune response (anti-IFN- α antibody, neutralising capacity)
- Local tolerance at the injection side
- Safety pharmacology (breath rate, heart rate, ECG)

All monkeys survived the treatment and recovery phase without the occurrence of severe adverse effects.

IFN- α was measurable in all animals treated with IFN- α -2a (Alpheon or Roferon-A) and the higher dose resulted in higher titers. Some IFN- α -like effects were observed, such as the appearance of IFN-antibodies and neopterin in serum, slight and transient changes in clinical haematological (decreases in erythrocyte parameters, reticulocyte and platelet counts, increase in APTT) and clinical chemistry (ALT, AST increase) parameters, and a lower food consumption.

Signals of differences between Alpheon and Roferon-A regarding the occurrence of single IFN- α -2a-like adverse effects (food consumption, some clinical haematology and chemistry values) were observed during this study. Low food consumption occurred in all treated groups. Whereas the food consumption in Alpheon treated groups was reduced only on single days, in the Roferon-A group low food consumption was observed on several days during the whole study period in all animals. Changes in clinical haematological and chemistry values, such as a decrease in platelet count and fibrin, increases in APTT, ALT and AST, were observed in animals in the high dose Alpheon group (12 million IU/kg/dose) and in animals receiving 6 million IU/kg/dose of Roferon-A, but not in the dose corresponding Alpheon group. Furthermore, a higher plasma level of IFN- α was observed in the Alpheon group compared to the dose corresponding Roferon-A group.

The low number of animals (3F/group), the use of only female animals, and the high interindividual variability of the data complicated the detection of differences between Alpheon and Roferon-A. Additionally, the time intervals of blood sampling for analysing blood levels of IFN- α , anti-IFN- α -antibodies, and neopterin were quite long and only one dose group was chosen for the comparator product, Roferon-A. This made time-and dose-dependent differences in the occurrence of anti-IFN- α antibodies, the pharmacokinetic profile and the pharmacodynamic effects between Alpheon and Roferon-A difficult to detect.

Therefore, on the basis of this study no conclusion on the similarity of Alpheon and Roferon-A could be drawn.

- Genotoxicity

No genotoxicity studies were required according to the relevant guideline (EMA/CHMP/42832/2005).

- Carcinogenicity

No carcinogenicity studies were performed. These studies were not required according to the *ICH/CPMP-Note for Guidance on preclinical safety evaluation of biotechnology-derived pharmaceuticals* (CPMP/ICH/320/95) and EMA/CHMP/42832/2005.

- Reproduction Toxicity

No reproduction toxicity studies were performed with Alpheon. Such studies were not mentioned as a requirement in the Concept paper on similar medicinal products containing alpha-interferon (CHMP/BWP/7241/2006). Data from published scientific literature indicated that interferons affected male and female fertility and induced abortions in monkeys. Several case reports have described successful pregnancies in women who received interferon-alpha during the first trimester or throughout gestation. In the light of current scientific knowledge it was concluded that there was sufficient information on the reproductive toxicity profile to support a relevant statement in the text of the Summary of Product Characteristics for Alpheon.

- Local tolerance

During the repeated dose toxicity study in monkeys the injection site was examined and was found to have a normal appearance. This indicated that Alpheon was well tolerated at the injection site.

- Other toxicity studies

A battery of standard toxicity studies was conducted with the HSA-stabilised, lyophilised formulation of Alpheon derived from an earlier manufacturing process. These studies included single and repeated dose toxicity studies in mice and rats, genotoxicity studies (Ames test, chromosome aberration assay and micronucleus assay), reproductive toxicity studies (fertility, teratogenicity and peri/postnatal toxicity) in rat and rabbits, acute skin and eye irritation studies in rabbits and antigenicity studies (PCA test and ASA test) in mice, rats, and guinea pigs.

No mutagenic, skin and eye irritant, or antigenic potential of IFN- α -2a was observed during these studies.

Due to the species specificity of IFN- α , the lack of pharmacological response in rodents and the differences in the manufacturing processes used to produce the original HSA-stabilised, lyophilised formulation and the Alpheon product intended for marketing, the relevance of these studies for the safety assessment of Alpheon was limited.

Ecotoxicity/environmental risk assessment

Proteins were unlikely to result in significant exposure to the environment and will be consequently of low environmental risk. Thus, no environmental risk assessment was required for Alpheon.

Discussion on the non-clinical aspects

The applicant submitted an *in vitro* study which compared the pharmacodynamic activity of Alpheon and Roferon-A in assays of antiviral activity and induction of interferon-sensitive genes 2' 5' OAS and MxA.

Insufficient information was submitted on the extent to which the methods used had been previously established and on the analysis of the data.

In addition, the applicant did not provide a comprehensive discussion of the inconsistent results obtained and did not adequately address the differences shown among the three preparations.

In particular whereas the antiviral activity of the preparations in A549 cells was nearly comparable, a high variability in the data was observed in the antiviral assay in HuH7 cells for each preparation.

In the gene induction assay the applicant could demonstrate that both interferons as well as the NIBSC reference standard induced 2', 5'-OAS and MxA genes. However, for each preparation a high variability in the magnitude of the gene induction was observed. In conclusion, based on this comparative study no definitive statement on the similarity of Alpheon and Roferon-A could be made.

The applicant submitted a 4-week repeated dose toxicity study in monkeys. Since the development programme of Alpheon preceded the publication of the *Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues* (EMA/CHMP/42832/2005), the design of this study did not meet the criteria of a comparability study. According to the guideline, non-clinical studies conducted for the investigation of comparability should be used to highlight differences between the product proposed for marketing authorisation and the reference product and should be designed to detect differences in response and not just the response per se. The applicant had justified the development programme through the process.

During the 4-week repeated dose study toxicity in monkeys some signals of differences between Alpheon and Roferon-A regarding the occurrence of single IFN- α -2a-related adverse effects (food consumption, some clinical haematology and chemistry values) were observed. Furthermore, a higher plasma level of IFN- α was observed in the Alpheon group compared to the dose corresponding Roferon-A group.

The study had a number of deficiencies, such as the low number of animals used, the use of only female animals, the long time intervals and number of blood sampling for analysing IFN- α , antibody, and neopterin levels, and the dose levels and use of only one dose group for the comparator product, Roferon-A.

The non-clinical data submitted did not allow a clear conclusion to be drawn regarding the similarity of Alpheon and Roferon-A.

3.4 Clinical aspects

Introduction

The applicant submitted three comparative clinical trials with Alpheon and the reference medicinal product Roferon-A:

- BP-IFN-001 and BP-IFN-005 were conducted to demonstrate a comparable pharmacokinetic profile of Alpheon and Roferon-A. Study BP-IFN-005 was also performed to assess pharmacodynamic comparability.
- Study BP-IFN-002, was an open-label, multicentre, centrally randomised, parallel-group phase III trial to demonstrate the comparable efficacy and safety of Alpheon and Roferon-A. Pharmacokinetic and pharmacodynamic comparability was also assessed in this study.

GCP

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.

A GCP inspection of Study BP-IFN-002 was performed. Although this inspection revealed some potential issues the applicant provided satisfactory assurance that the overall validity of the study data was not compromised.

Pharmacokinetics

No studies on pharmacokinetics (Absorption / Distribution / Elimination; dose proportionality and time dependencies; special populations; pharmacokinetic interaction studies; pharmacokinetics using human biomaterials) were required according the *Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues* (EMA/CPMP/42832/05).

- **Study BP-IFN-001 (comparison of Alpheon and Roferon-A)**

Study BP-IFN-001 was an open study performed with the Roferon-A. The reference product had a different pharmaceutical form and a different strength and had to be refilled at the study site.

The evaluation of data showed supra-bioavailability of Alpheon over the reference product. The evaluation of the reasons for supra-bioavailability revealed a period effect in the Alpheon group. However, there was no carry-over effect because baseline concentrations were always under the limit of quantification and it could also be shown that there was only a slight sequence effect that was not statistically significant. Analytical errors were also excluded by re-analysis of plasma samples.

The Applicant concluded that the ambiguous results of the study were suggestive of problems with the conduct of the study itself.

The results of this study did not support a conclusion of comparable pharmacokinetics between Alpheon and the reference product.

- **Study BP-IFN-005 (comparison of Alpheon and Roferon-A)**

As a consequence of the above, the applicant performed a second comparative pharmacokinetic study. The study design was different from the design of the first study in various aspects: double blind, sampling times were slightly different, the wash-out phase was extended to 12 days (instead of 7), and the same pharmaceutical form and strength of the test product and the reference products was used.

The 90% confidence intervals of the ratios of the geometric means of $AUC_{(0-\infty)}$, $AUC_{(0-36)}$ and C_{max} were contained within the range 80-125%.

Parameter	N	Ratio Alpheon / Roferon-As %	
		Estimate	90% confidence interval
$AUC_{(0-\infty)}$	25	105.78%	(101%,111%)
$AUC_{(0-36)}$	27	107.67%	(103%,112%)
C_{max}	27	114.62%	(110%,119%)
T_{max}	27	0h	(-1.0h,+0.96h)

The statistical analysis of the pharmacokinetic data detected a significant period effect, for both compounds, with higher values for period 2, probably indicative of a carry-over effect.

This study supported a conclusion of comparable pharmacokinetics between Alpheon and the reference product.

- **Study BP-IFN-002 (comparison of Alpheon and Roferon-A)**

Within the clinical study BP-IFN-002, the Applicant planned to perform an analysis of pharmacokinetics for the test and reference formulation. However, this evaluation was not possible, as the serum concentration data exhibited marked variability, not only between, but also within individual patient profiles. In addition, implausible results were obtained in a number of patients (unquantifiable concentrations, very high concentrations at all time-points).

Due to the small sample sizes (21 and 19 patients in the Alpheon and Roferon-A group, respectively) these pharmacokinetic data could not support a conclusion of comparable pharmacokinetics between Alpheon and the reference product.

Pharmacodynamics

- Study BP-IFN-005 (comparison of Alpheon and Roferon-A)**

Study BP-IFN-005 was also performed to assess pharmacodynamic equivalence. The parameters chosen for this assessment were: Beta-2-microglobulin, 2'5'-Oligo-adenylate-synthetase (2'5'-OAS), and neopterin, which were unspecific parameters of the pharmacodynamic effects of interferons. The measured levels for these parameters were corrected for the baseline endogenous levels by subtracting the mean of the measured concentrations in the pre-dose samples. The following tables give an overview of the results of these investigations:

	Parameter	Unit	Alpheon		Roferon	
			Geom. mean	Geom. %CV	Geom. mean	Gom. %CV
Neopterin	AUC (0-t _z)	h*ng/ml	372.65	22.44	371.94	22.18
	C _{max}	ng/ml	4.832	19.76	4.579	19.87
	T _{max} *	h	36.0	28-48	36.0	24-96
β ₂ -micro-globulin	AUC (0-t _z)	h*μg/L	58926	34.28	63080	33.47
	C _{max}	μg/L	939.6	18.19	953.5	19.01
	T _{max} *	h	24.0	20-48	24.0	20-72
2'5' OAS	AUC (0-t _z)	h*pmol/dL	16649	56.84	16790	65.72
	C _{max}	pmol/dL	227.6	55.15	237.1	60.34
	T _{max} *	h	36.0	12-168	36.0	8-72

Summary of the statistical comparisons for pharmacodynamic parameters in study

	Parameter	Ratio Alpheon / Roferon-As %	
		Estimate	90% confidence interval
Neopterin	AUC (0-t _z)	99.87%	(93%, 107%)
	C _{max}	105.44%	(100%, 112%)
β ₂ -microglobulin	AUC (0-t _z)	93.95%	(83%, 106%)
	C _{max}	98.60%	(92%, 105%)
2'5' OAS	AUC (0-t _z)	97.74%	(84%, 114%)
	C _{max}	95.66%	(83%, 110%)

The 90% confidence intervals for the ratios of the AUC and C_{max} values determined were wholly contained within the 80-125% equivalence range. For AUC (0-t) higher values were observed for neopterin and for 2'5'OAS in period 1 compared to period 2 and for β₂-microglobulin in period 2 compared to period 1.

- Study BP-IFN-002 (comparison of Alpheon and Roferon-A)**

Pharmacodynamic equivalence was also assessed in the clinical efficacy and safety study BP-IFN-002. Blood samples were taken from 18 vs. 20 patients. Two selected markers (Beta-2-microglobulin and neopterin) were determined to characterise comparable pharmacodynamics in the target population. The point estimates and 90% confidence intervals for the ratios of the AUC and C_{max} were well within the pre-specified boundaries of bioequivalence (80-125%) for β₂-microglobulin. However, consistently higher values were obtained for neopterin, thus pharmacodynamic equivalence could not be unequivocally concluded for this parameter.

The pharmacodynamic data seem to indicate the similarity of the two compounds tested. An overall conclusion on pharmacodynamic equivalence, however, has to consider the results of the clinical study, which investigated the most valuable pharmacodynamic parameter, viral response.

Clinical efficacy

- Dose response study

No studies were required according to EMEA/CPMP/42832/05.

- Main study

Study BP-IFN-002, was an open-label, multi-centre, centrally randomised, parallel-group phase III trial.

METHODS

Study Participants

Male and female patients, 18-70 years of age, with a positive serum anti-HCV antibody (Ab) screening assay, HCV-RNA serum positive and elevated ALT (>1,5 ULN) were enrolled. Liver biopsy had to have been obtained within 12 months of study start, and histology had to show chronic active viral hepatitis with no evidence of cirrhosis.

Treatments

Patients received Interferon alpha-2a at a dose of 3 million IU three times a week (TIW) s.c. Alpheon was administered with the multi-dose vials (3 ml with 18 million IU corresponding to 6 million IU/ml) intended for marketing. The reference product Roferon-A was administered in the pharmaceutical form of pen-cartridges containing 18 million IU in 0.6 ml (corresponding to 30 million IU/ml).

Objectives

The objectives of the study were to demonstrate the comparable clinical efficacy and safety of Alpheon and Roferon-A monotherapy in patients with compensated chronic hepatitis C.

Outcomes/endpoints

The primary efficacy endpoint was the rate of treatment responders (defined as patients with undetectable HCV-RNA) after 12 weeks of treatment.

Secondary endpoints were: the rate of treatment responders after 24, 48 (end-of-treatment) and 72 weeks (6-months after end of treatment); the rate of sustained virological response (SVR), 6 months after the end of treatment. Biochemical response after 12, 24, 48 and 72 weeks and histological response after 48 weeks were further secondary efficacy endpoints. These secondary analyses were only conducted for those patients with a response after 12 weeks of treatment.

Sample size

The sample size calculation stated in the protocol (or the protocol amendment No. 1) estimated the required size of the study to be 200 patients per treatment arm. However, the study report indicated that the number to be screened was 400 and the number to be randomised was 360.

The sample size of the study and the associated calculations seemed adequate; in particular, the assumed delta of $\pm 15\%$ for the three-month responder rate could be considered appropriate. However, the protocol assumed the calculation of 90% confidence intervals for the difference in responder rates, which did not correspond to the postulated overall significance level of 5%. However, in the final evaluation of the trial appropriate confidence intervals (95%) were presented.

Randomisation

Randomisation was performed centrally with stratification according to genotype (1 and non-1). The randomisation list was generated using the method of random permuted blocks. A separate random scheme was generated for each stratum. Blocks of randomisation numbers within each of the two strata ("Genotype 1" and "Genotype other") were allocated for each site.

Randomisation also led to a balanced allocation with regard to viral load.

Blinding (masking)

Study BP-IFN-002 was an open study. Due to the very different appearances of the two preparations, it was decided that blinding was not possible.

Statistical methods

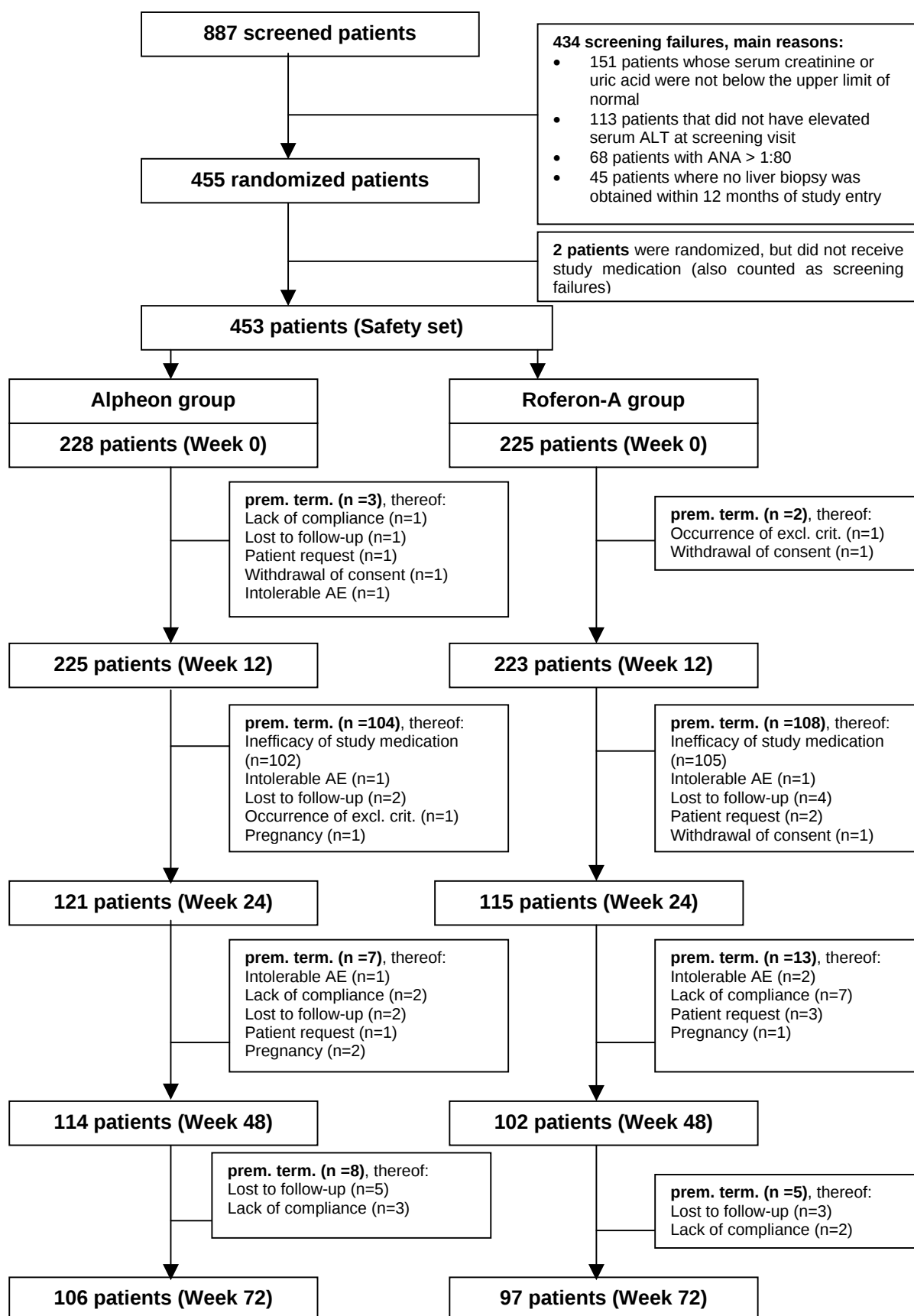
The main analysis of the study was based on the estimation of the 12 weeks rate of HCV-RNA negativity with the calculation of two sided 95% confidence intervals. This interval was expected to lie within a delta of $\pm 15\%$.

Patients with major protocol deviations, incomplete documentation, less than 80% study drug intake or premature termination due to reasons not related to study medication were excluded from the per-protocol (PP) population.

All randomised patients treated with study medication that had at least one post-baseline evaluation of the primary efficacy variable available were included in the intend-to treat (ITT) population.

The safety data set consisted of all patients that took the study medication at least once.

Development of patient numbers in the study course



RESULTS

Recruitment

There was a high rate of screening failures; of the 887 patients screened, 455 were included into the study. The main reasons for exclusion were not meeting exclusion/ inclusion criteria. However, the demographics of included and excluded patients were roughly similar.

There were 22 centres and the number of recruited patients varied from 4 (1.0%) in the smallest centres, up to 35 (8.6%) in the biggest centres.

Conduct of the study

Patients were seen for the screening visit and baseline visit, and then at week 2, 4, 8, 12, 16, 24, 36, 48 and 72. Efficacy assessments were performed at week 12, 24, 48 (end of treatment) and 72 (end of 6-month observation period). Safety evaluations were performed at each visit (antibody formation was determined at week 24, 48, and 72).

Baseline data

The age of the randomised patients ranged from 18 to 63 years, with most patients between 22 and 34 years old. All patients except one in the Alpheon group, who was Asian, were of Caucasian origin. All other baseline demographic data were comparable between the groups, including the distribution of viral genotype.

Numbers analysed

There were 455 patients randomised: 453 patients were included in the safety and ITT populations and 421 in the per-protocol population.

Main efficacy results

Main efficacy results of study BP-IFN-002

Parameter	Per protocol analysis set			Full analysis set		
	Alpheon (n=210)	Roferon-A (n=211)	95 CI for diff.	Alpheon (n=228)	Roferon-A (n=225)	95% CI for diff.
Rate of HCV-RNA negativity at week 12	53.8%	51.7%	-7.39; +11.69	53.1%	51.6%	-7.68; +10.71
Rate of HCV-RNA negativity at week 24	43.8%	39.3%	-4.93; +13.88	42.5%	39.6%	-6.07; +12.04
Rate of HCV-RNA negativity at week 48	46.2%	37.9%	-1.12%; +17.57%	43.9%	37.8%	-2.95%; +15.12%
Rate of HCV-RNA negativity at week 72	31.9%	35.1%	-12.18%; +5.84%	30.7%	34.7	-12.60%; +4.67%
Sustained virological response rate (HCV-RNA negativity at week 48 <u>and</u> 72)	31.4%	32.7%	-10.19%; +7.64%	30.3%	32.4%	-10.72%; +6.36%
Rate of patients with ALT normalisation week 12	69.5%%	73.0%	-12.10%; +5.18	68.4%	72.4%	-12.42 +4.37
Rate of patients with ALT normalisation week 24	46.2%	40.3%	-3.54%; +15.35%	44.7%	40.4%	-4.81; +13.39
Rate of patients with ALT normalisation week 48	48.1%	38.9%	-0.20%; +18.66%	45.6%	38.2%	-1.67%; +16.45%
Rate of patients with ALT normalisation week 72	34.8%	34.1%	-8.44%; +9.72%	33.3%	33.8%	-9.14%; +8.25%

Alpheon and Roferon-A were comparable in terms of the main virological and biochemical endpoints. In addition, the per-protocol and intention-to-treat analyses yielded almost identical results, indicating that the main efficacy analyses were robust.

Liver biopsies were performed at week 48 in 88 (38.6%) patients in the Alpheon and 82 (36.4%) patients in the Roferon-A group (intention to treat analysis). The mean change of stage in comparison to screening biopsy up to week 48 was –0.87 in the Alpheon group, and –0.93 in the Roferon-A group for the PP set and –0.88 and –0.90 for the ITT set. This translated into an “improvement rate” (patients with a decrease of 1 and more stages) of 22.9% and 21.3% in the PP set, and 21.5% and 21.3% in the ITT set for the Alpheon and Roferon-A groups, respectively. An analysis of the grading of the liver biopsies was not performed during the study.

Ancillary analyses

An analysis was performed for the relapse rate between the end of treatment (week 48), and the end of the observation period (week 72).

Relapse rate between the end of treatment and the end of observation period

	Alpheon	Roferon-A	p-value
Number of relapsers (relating to the whole study population)	23/228 10.1%	8/225 3.6%	0.008
Number of relapsers (related to the number of responders at week 48)	23/97 23.7%	8/80 10.0%	0.02

This analysis revealed an overall rate of late relapses of 23 (10.1%) and 8 (3.6%) in the Alpheon and Roferon-A groups, respectively. Since this represented a clinically relevant and a statistically significant difference (p=0.008), the applicant was requested to explore the reasons for this finding.

The following factors were investigated: gender, age, weight, height, and body-mass-index for the demographic factors, antibody status, viral genotype, and baseline viral load.

There were no relevant differences in the patient groups regarding demographic factors at any time point, neither in the whole study population nor in the “patients with late relapse” group.

Anti-interferon antibody status and virological response in safety set and “late relapser” group

Subset	Treatment	No positive results at any visit		Positive result at a single visit		Positive result at more than one visit		Overall	
		n	%	n	%	n	%	n	%
Week 12 responders	Alpheon	64	52.9 %	27	22.3 %	30	24.8 %	121	100
	Roferon	58	50.0 %	26	22.4 %	32	27.6 %	116	100
Week 24 responders	Alpheon	54	55.7 %	19	19.5 %	24	24.7 %	97	100
	Roferon	51	57.3 %	19	21.3 %	19	21.3 %	89	100
Week 48 responders	Alpheon	57	57.0 %	20	20.0 %	23	23.0 %	100	100
	Roferon	44	51.8 %	16	18.8 %	25	29.4 %	85	100
Week 72 responders	Alpheon	39	55.7 %	13	18.6 %	18	25.7 %	70	100
	Roferon	43	55.1 %	14	17.9 %	21	26.9 %	78	100
Late relapses (Week 48 responders relapsed at Week 72)	Alpheon	14	60.9 %	3	13.0 %	6	26.1 %	23	100
	Roferon	4	50.0 %	1	12.5 %	3	37.5 %	8	100

Note: Visits for antibody determination occurred at Weeks 0, 24, 48 and 72

A positive correlation between anti-interferon antibodies and “late relapse” could not be found. Thus antibody formation was highly unlikely to have contributed to the higher relapse rates in the Alpheon group.

The applicant also explored the correlation of genotype and viral load at baseline and “late relapse” subset. The following table gives the results of the subgroup analyses performed according to the genotype at inclusion.

Genotype and viral load at baseline for safety set and “late relapser” group

Subset	Treatment	n	Genotype 1				Non-1			
			> 3 Mio.		≤ 3 Mio.		> 3 Mio.		≤ 3 Mio.	
			n	%	n	%	n	%	n	%
Entire safety set	Alpheon	223	4	1.8 %	77	33.8 %	30	13.2 %	117	51.3 %
	Roferon	225	7	3.1 %	70	31.1 %	32	14.2 %	116	51.5 %
Week 12 responders	Alpheon	121	1	0.8 %	27	22.3 %	20	16.5 %	73	60.3 %
	Roferon	116	3	2.6 %	25	21.6 %	17	14.7 %	71	61.2 %
Week 24 responders	Alpheon	97	1	1.0 %	19	19.6 %	17	17.5 %	60	61.9 %
	Roferon	89	1	1.1 %	19	21.3 %	14	15.7 %	55	61.3 %
Week 48 responders	Alpheon	100	1	1.0 %	17	17.0 %	18	18.0 %	64	64.0 %
	Roferon	85	3	3.5 %	13	15.3 %	12	14.1 %	57	67.1 %
Week 72 responders	Alpheon	70	1	1.4 %	9	12.9 %	11	15.7 %	49	70.0 %
	Roferon	78	3	3.8 %	14	17.9 %	12	15.4 %	49	62.3 %
Late relapsers (Week 48 responders relapsed at Week 72)	Alpheon	23	0	0.0 %	3	34.8 %	7	30.4 %	8	34.3 %
	Roferon	8	0	0.0 %	2	25.0 %	0	0.0 %	6	75.0 %

No significant imbalance at baseline regarding the genotype distribution for the patients with late relapse could be shown.

The following table shows the results for the responder rates in the genotype 1 patients only:

Responders Genotype 1	Alpheon		Roferon-A		Overall	
Evaluable population	n=81	%	n=77	%	n=147	%
Week 12	27	35.1	26	35.6	53	35.3
Week 24	20	26.0	18	24.7	28	25.3
Week 48	18	23.4	15	20.5	28	25.3
Week 72	10	12.3	17	22.1	26	17.3

A 10% difference in SVR for genotype 1 infected patients was observed at Week 72. Moreover, only 30% of the patients included in the clinical study were of genotype 1, compared to ca. 60% in other clinical trials in Hepatitis C and an average 60% infection rate with genotype 1 across Europe (with variable rates in different countries between 40% and over 95%). The low number of genotype 1 patients included raised doubts about the robustness and external validity of the study.

Furthermore, the submitted data could not substantiate the argument that the late relapse rate was associated with a high virus load at baseline, since no significant imbalance regarding the viral load for the patients with late relapse could be shown.

In conclusion, no imbalance regarding baseline factors between the groups leading to the differences in sustained virological response could be identified.

- Discussion on efficacy

The objectives of the efficacy study were to demonstrate comparable clinical efficacy of Alpheon and Roferon-A in patients with compensated chronic hepatitis C.

The primary efficacy analysis was the rate of treatment responders (defined as patients with undetectable HCV-RNA) after 12 weeks of treatment. Secondary endpoints were the virological

response rate at weeks 24, 48 and 72; the biochemical response rate at weeks 12, 24, 48 and 72; and the histological response at week 48.

In all of these secondary parameters comparable efficacy between Alpheon and Roferon-A was demonstrated even though separate non-inferiority margins were not defined in advance. However, the 15% margin as defined for the primary endpoint was met in almost all evaluations regarding virological and biochemical response.

A clinically and statistically significant difference ($p=0.008$) in relapse rate between end of therapy and end of observation period (at 72 weeks) was observed. The applicant explored the reasons for late relapse in these patients. No factors present at baseline contributing to this difference could be identified. The applicant stated that since Alpheon was comparable to Roferon-A in terms of the primary and the secondary endpoints this difference should not be regarded as being of clinical concern and it was implausible that this disparity in relapse rate after week 48, which was not a pre-specified efficacy endpoint, was related to differences in the two interferons.

However, uncertainties remained about the observed difference in relapse rate after week 48. It was therefore concluded that the results presented did not support comparable clinical efficacy between Alpheon and Roferon-A.

The differences in the virological response rates at week 72 in patients with genotype 1 (the “difficult to treat” patients) were also of clinical concern. For this subpopulation, it was stated that the “zero relapse” rate in the Roferon-A group was an extraordinary finding and the virological response at week 72 observed in the Alpheon group was as expected and consistent with the literature. However, with the low numbers in this study - only 30% of the patients included in the clinical trial BP-IFN-002 were of genotype 1, as opposed to ca. 60% in other clinical trials in hepatitis C – no reliable conclusions could be drawn. Furthermore, it had to be acknowledged that within the clinical study there were quite a lot of exceptional results with Roferon-A and with Alpheon due to the fact that the patient population was of a very young age and consisted of a high percentage of patients infected with genotype non-1 virus. With the low rate of included genotype 1 patients – putting the overall validity and robustness of the data into question – a 10% difference in SVR for this subgroup had to be considered clinically relevant and did not allow a clear conclusion to be reached on the comparable clinical efficacy of Alpheon and Roferon-A.

Clinical safety

- Patient exposure

Two single-dose pharmacokinetic/pharmacodynamic studies with the exposure of 56 healthy male volunteers were included in the dossier (Studies BP-IFN-001, and BP-IFN-005).

The clinical study BP-IFN-002 comprised 455 patients with a mean study duration of 336.6 and 337.0 days of treatment in the Alpheon and Roferon-A groups in patients completing the study. The treatment duration for those patients withdrawn prematurely was 135.0 and 132.8 days for Alpheon and Roferon-A, respectively. The figures for the “time in study” were 508.9 and 508.3 days for the patients completing the study, and 297.2, and 293.1 days for those that were prematurely withdrawn. All patients were treated with a dose of 3 million IU three times a week. The number of missed doses and the number of dose reductions was low.

- Adverse events

- Healthy volunteers

The total number of adverse events (AEs) in study BP-IFN-001 was 42 and 34 during treatment with Alpheon and Roferon-A respectively. All of these events were assessed as being possibly or probably related to the treatment. No deaths and no serious AEs were reported during this study.

The total number of AEs reported in study BP-IFN-005 was 256 of which 125 events occurred in 26 subjects in the Alpheon group and 131 events in 27 subjects occurred in the Roferon-A group. 83% of these events were assessed as being probably related to the application of the drug. No deaths and no serious events occurred during the course of the study.

➤ Study BP-IFN-002

A total of 1,444 AEs were reported in 391 patients. Of these, 777 events occurred in 203 (89.0%) patients in the Alpheon group and 667 in 188 (83.6%) patients in the Roferon-A group.

The large majority of adverse events were of mild intensity (512 in the Alpheon group vs. 455 events in the Roferon-A group). Of the remainder, 252 vs. 198 events were of moderate intensity, 7 vs. 10 events were of severe intensity and no intensity was specified in the case of 6 vs. 4 events.

Of all the adverse events, 34 vs. 37 events were unlikely related, 118 vs. 79 events were not related, 132 vs. 105 events were possibly related while 493 vs. 445 events were probably related to treatment. Of the remaining events, 0 vs. 1 event was reported as not assessed.

The following table displays the number of patients with AEs according to body system:

SYSTEM ORGAN CLASS AND PREFERRED TERM	No. of patients with AEs			
	Alpheon n=228		Roferon-A n=225	
	n	(% of total patients)	n	(% of total patients)
General disorders and administration site reactions	190	83.3	175	77.8
Influenza like illness	101	44.3	86	38.2
Pyrexia	86	37.7	81	36.0
Weakness	40	17.5	42	18.7
Weight decreased	18	7.9	7	3.1
Asthenia	5	2.2	6	2.7
Musculo-skeletal and connective tissue disorders	48	21.1	54	24.0
Arthralgia	32	14.0	43	19.1
Myalgia	19	8.3	14	6.2
Investigations	44	19.3	33	14.7
Nervous system disorders	41	18.0	36	16.0
Headache	35	15.4	29	12.5
Rigors	7	3.1	3	1.3
Blood and lymphatic system disorders	36	15.8	32	14.2.
Leucopenia	15	6.6	15	6.7
Thrombocytopenia	10	4.4	12	5.3
Anaemia	7	3.1	8	3.6

White blood cell count decreased	8	3.5	5	2.2
Infections and infestations	22	9.6	25	11.1
Influenza	6	2.6	7	3.1
Skin and subcutaneous tissue disorders	24	10.5	16	7.1
Alopecia	4	1.8	3	1.3
Hypotrichosis	7	3.1	3	1.3
Gastrointestinal Disorders	11	4.8	9	4.0
Metabolic disorders	12	5.3	8	3.6
Appetite decreased	4	1.8	4	1.8
Psychiatric disorders	11	4.8	9	4.0
Depression	3	1.3	2	0.9
Insomnia	4	1.8	3	1.3
Irritability	6	2.6	3	1.3
Endocrine disorders	12	5.3	5	2.2
Thyrotoxicosis	7	3.1	4	1.8
Pregnancies	7	3.1	4	1.8

- Serious adverse event/deaths/other significant events

No deaths were reported during the conduct of the study. Altogether, 10 serious adverse events were reported in 7 vs. 2 patients during the active treatment phase of the study. Three events in 2 vs. 1 patients occurred during the screening phase of the study. All events were considered not related to the study medication.

Laboratory findings

Laboratory changes considered as clinically relevant were reported in 114 patients (63 patients in the Alpheon group and 51 patients in the Roferon group). In these patients 453 events were reported.

Evaluation of laboratory related adverse events judged as clinically relevant:

	Alpheon	Roferon A	p-value
Number of events (counting all reported events)	257	196	N.d.
Number of patients events (counting all events only once per patient)	146/228 64.0 %	104/225 46.2 %	0.02

The main differences were seen in thyroid related and white blood count (WBC) related parameters.

Parameter	Alpheon		Roferon		Overall	
Thyroid related:	28	12.3 %	18	8.0 %	47	10.4 %
TSH	12	5.3 %	10	4.4 %	22	4.9 %
Free T4	8	3.5 %	5	2.2 %	13	2.9 %
T3	7	3.1 %	3	1.3 %	10	2.2 %
Thyroxin	1	0.4 %	0	0 %	1	0.2 %
WBC related:	46	20.2 %	36	16.0 %	82	18.1 %
WBC	27	11.8 %	20	8.9 %	47	10.4 %
Lymphocytes	9	3.9 %	5	2.2 %	14	3.1 %
Neutrophils	8	3.5 %	6	2.7 %	14	3.1 %
Monocytes	1	0.4 %	3	1.3 %	4	0.9 %
Eosinophils	1	0.4 %	2	0.9 %	3	0.7 %

With regard to the other parameters no consistent pattern of laboratory differences could be shown that was to the advantage or the disadvantage of one or the other drug. No relevant changes and no differences between the treatment function screening tests were seen.

- Immunogenicity

The formation of binding antibodies was investigated at baseline, after 24, 48, and 72 weeks of treatment, using a commercial available test kit.

Anti-IFN antibodies (Evaluable patients)		Alpheon		Roferon-A		Overall	
Week 24 (Visit 8)		n = 121	%	n = 115	%	n = 236	%
	missing	0	0.0	5	4.3	5	2.1
	negative	71	58.7	59	51.3	130	55.1
	positive	50	41.3	51	44.3	101	42.8
	Overall	121	100.0	115	100.0	236	100.0
Week 48 (Visit 10)		n = 114	%	n = 102	%	n = 216	%
	missing	3	2.6	3	2.9	6	2.8
	negative	80	70.2	61	59.8	141	65.3
	positive	31	27.2	38	37.3	69	31.9
	Overall	114	100.0	102	100.0	216	100.0
Week 72 (Visit 11)		n = 106	%	n = 97	%	n = 203	%
	missing	2	1.9	1	1.0	3	1.5
	negative	88	83.0	75	77.3	163	80.3
	positive	16	15.1	21	21.6	37	18.2
	Overall	106	100.0	97	100.0	203	100.0

The number of patients that were positive for anti-interferon alfa antibodies was similar in both groups. The absolute rate of antibody formation tended to be higher in the Roferon-A group at all time points. The differences in the rate of antibody formation during treatment between the two groups tended to increase until the end of treatment at week 48, whereas the difference levelled out slightly after cessation of therapy. The applicant provided the one-sided 95% confidence limit for the difference in the proportions of patients with anti-interferon alfa antibodies at week 24 with the lower end given as -3.26% indicating non-inferiority (delta = 15%).

Regarding the validation of the methods used for the detection of antibodies, the applicant had no access to the original validation data and had not performed any pre-validation experiments before starting the relevant studies. Thus comprehensive validation data for the commercial available ELISA (MDS) were not provided. Instead the applicant presented results from a post validation study. The validation experiments included: specificity, precision (repeatability, intermediate precision),

determination of detection limit and quantification limit and short term stability and freeze thaw stability of serum samples. The analytical results were, with the exception for linearity, within the acceptance criteria recommended by the manufacturer. The applicant concluded that the double antigen enzyme immunoassay system was adequate for the determination of anti-Interferon-alpha antibodies by the ELISA applied. However, the applicant failed to exclude that depending on the antibody concentration false negative results might occur, which would appear to be an intrinsic disadvantage of the double antigen enzyme immunoassay system.

In addition, the data provided for the post study validation were not a complete assay validation but more an assay qualification.

An additional analysis was performed for the 70 vs. 78 patients who had anti-IFN alpha-2a antibodies reported at least once and who also had an adverse event. This analysis did not reveal any clinically relevantly different pattern of adverse events in comparison to the rest of the study population

The applicant, contrary to the Scientific Advice received, only reports the occurrence of binding antibodies in the population under investigation.

No other parameters of immunological nature, such as mean titres of antibodies (binding and neutralising, and overall) were reported.

- Discontinuation due to adverse events

Discontinuation due to adverse events was reported in 3 vs. 3 patients in the Alpheon and Roferon-A group.

- Safety related to drug-drug interactions and other interactions

No studies were required according to EMEA/CPMP/42832/05.

- Post marketing experience

N / A

- Discussion on clinical safety

In the clinical study BP-IFN-002 no unexpected events were seen, and the events were regarded as being typical for this kind of medication. However, there was a slightly higher (and almost statistically significant (chi-square test shows a p-value of 0.09)) incidence of adverse events in the Alpheon group compared to the Roferon-A group.

The applicant attributed the differential rate of reporting to the open study design - the open-label study design could have introduced a bias since patients in the Alpheon group knew they were receiving an experimental drug and they may have been more likely to report adverse events.

However, the differences in adverse event rates were mirrored for laboratory related adverse events judged as clinically relevant by the investigators. These events amount to 146 and 104 (when counting the events only once per patient), or 257 and 196 (when counting all reported events) in the Alpheon and Roferon-A groups, respectively. The first of these figures amounts to a highly statistically significant difference when related to the whole safety population ($p < 0.001$; Fisher's exact test).

Although the available immunogenicity data for Alpheon and Roferon A were comparable, the method used was not fully validated and did not sufficiently exclude false negative results. The overall extent of investigations regarding immunogenicity did not fully comply with the *Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues* (EMEA/CPMP/42832/05), as only the binding properties of the antibodies was investigated.

Despite the applicants arguments there was still uncertainty about the safety and immunogenicity data generated in the pivotal clinical study, which did not allow a clear conclusion to be made regarding the similarity of Alpheon and Roferon-A.

3.5 Pharmacovigilance

Detailed description of the Pharmacovigilance system

The CHMP considered that the Pharmacovigilance system as described by the applicant fulfils the legislative requirements.

Risk Management Plan

The CHMP, having considered the data submitted in the application was of the opinion that it was not appropriate to consider risk minimisation activities at this time.

3.6 Overall conclusions, risk/benefit assessment and recommendation

The applicant, BioPartners GmbH, submitted an application for Marketing Authorisation to the European Medicines Agency (EMA) for Alpheon, under the legal basis of Similar Biological Medicinal Product. The reference medicinal product for this application was Roferon-A Solution for Injection, a recombinant interferon alfa 2a containing product expressed in *E. coli* produced by Roche. Alpheon was claimed to be similar to this reference medicinal product as approved in the Community.

Quality

The assays where product related impurities have been investigated indicated that Alpheon and Roferon-A display a different impurity profile. The precise nature, biological properties, and the qualitative and quantitative composition of the different impurities had not been fully studied. Differences in the stability profile also required investigation.

It was therefore concluded that the data comparing the two drug products were incomplete and inconclusive. Issues regarding the samples of Alpheon used (currently no validated manufacturing process) and the impact of sample preparation (concentration procedures and presence of excipients) on the analytical methods were unresolved.

As a consequence of the deficiencies set out above and the quantitative and qualitative differences in the impurity profile, no conclusion regarding the comparability of Alpheon and Roferon-A based upon the quality data currently available could be reached.

The company was unable to fully validate the drug product production process for Alpheon, consequently there was insufficient assurance of control and consistency of the manufacturing process.

No stability data representative for the intended commercial drug product were available and a shelf life could not be assigned. The drug substance stability data were also insufficient.

In addition, the applicant was unable to satisfactorily address the comparability of the clinical trial material with the drug product produced from the intended commercial process. The task was hampered by the lack of validation of analytical methods used as well as validation of the production process.

Furthermore, the applicant provided a retrospective analysis of the clinical stability samples versus consistency lots as part of the comparability exercise. However, no conclusion could be drawn from these experiments with aged samples due to the lack of data on the stability profile of Alpheon.

Reliable release data proving comparability of clinical and the recent batches in terms of related substances were not available. Therefore uncertainties remained regarding the comparability of the clinical trial material with the proposed commercial medicinal product.

Other concerns raised during the assessment process also remained unresolved.

Non-clinical pharmacology and toxicology

The applicant submitted the results of an *in vitro* study, which compared the pharmacodynamic activity of Alpheon and Roferon-A in assays of antiviral activity and induction of interferon-sensitive genes 2' 5' OAS and MxA. For A549 cells the antiviral activity of each preparation was comparable, however, a high variability was observed in the second antiviral assay (using HuH7 cells). In the gene induction assay the applicant could demonstrate that both interferons as well as the reference standard induced 2', 5'-OAS and MxA genes. However, for each preparation a high variability in the

magnitude of the gene induction was observed. On the basis of these data no decision on the similarity of Alpheon and Roferon-A could be drawn.

The applicant submitted a 4-week repeated dose study toxicity in monkeys. Since the development programme for Alpheon preceded the publication of the *Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues* (EMA/CHMP/42832/2005), the design of this study did not meet the current criteria of a comparability study. According to the guideline, non-clinical studies conducted for the investigation of comparability should be used to highlight differences between the product proposed for marketing authorisation and the reference product and should be designed to detect differences in response and not just the response per se. The applicant had justified the development program through the process.

During the 4-week repeated dose toxicity study in monkeys some signals of differences between Alpheon and Roferon-A regarding the occurrence of IFN- α -2a-related adverse effects were observed. Furthermore, a higher plasma level of IFN- α could be observed in the Alpheon group compared to the dose corresponding Roferon-A group.

The non-clinical data submitted did not allow a clear conclusion to be reached regarding the similarity of Alpheon and Roferon-A.

Efficacy

Alpheon and Roferon-A were shown to have comparable pharmacokinetics in one comparative pharmacokinetic study (study BP-IFN-005).

Data from two further pharmacokinetic studies did not support a conclusion of pharmacokinetic comparability between Alpheon and Roferon-A. Data from study BP-IFN-001 were invalid and the pharmacokinetic data obtained from the target population in the clinical study BP-IFN-002 were inconclusive.

A comparable virological response was observed in Hepatitis C patients treated with Alpheon and Roferon-A at Week 12 weeks (primary endpoint), Week 24, Week 48 week (end of treatment) and Week 72 (end of 6-month observation period). Similar results were observed for biochemical response.

However, a clinically and statistically significant difference ($p=0.008$) in virological relapse rate between end of therapy (week 48) and the end of the 6-month observation period (week 72) was observed as well as differences in the SVR for the “difficult-to-treat” genotype 1 patients.

Overall, there was still uncertainty about the comparable clinical efficacy of Alpheon and Roferon-A, which prevented a clear conclusion being made regarding the similarity of the two products.

Safety

There was a slightly higher (and almost statistically significant (chi-square test shows a p-value of 0.09) incidence of adverse events in the Alpheon group compared to the Roferon-A group. The differences in adverse event rates were mirrored for laboratory related adverse events judged as clinically relevant by the investigators. Therefore, from the safety data generated in the pivotal clinical study, clinical comparability of the two compounds from the safety point of view could not be concluded.

As far as immunogenicity was concerned, although the available data did show comparable results for Alpheon and Roferon A, the investigation itself had been incompletely validated and the assay used did not exclude false negative results. Moreover, within the validation data differences between the two compounds were found.

The overall extent of investigations regarding immunogenicity does not fully comply with the *Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance: non-clinical and clinical issues* (EMA/CPMP/42832/05) as only the binding properties of the antibodies had been investigated during the study. The requested post hoc evaluation of neutralising properties of the antibodies could only be performed in a very small number of samples.

Therefore, the investigations on immunogenicity were not fully validated and do not support a conclusion of comparable immunogenicity.

Risk-benefit assessment

Based upon the remaining major quality issues and the resulting clinical uncertainties Alpheon cannot be concluded as being a similar biological medicinal product to Roferon A, the chosen reference medicinal product. Therefore based on the data submitted the benefit/risk balance was not considered to be positive.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the risk-benefit balance of Alpheon in the treatment of Adult patients with histologically proven chronic hepatitis C who are positive for HCV antibodies or HCV RNA and have elevated serum alanine aminotransferase (ALT) without liver decompensation was unfavourable and therefore did not recommend the granting of the marketing authorisation.

GROUNDINGS FOR REFUSAL

Whereas

With regard to Quality a large number of outstanding issues remain. The major objections were as follows:

Comparability of Alpheon versus Roferon-A

-As a consequence of the deficiencies identified and quantitative and qualitative differences in the impurity profile a conclusion on the comparability of Alpheon and Roferon-A based on the quality dossier cannot be reached.

For the drug substance

-Insufficient stability data representative of the drug substance were available and a shelf life cannot be assigned.

For the drug product

-The manufacturing process for Alpheon has not been adequately validated.

-Insufficient stability data representative of the intended commercial drug product were available and a shelf life cannot be assigned.

Clinical comparability of Alpheon versus Roferon-A has not adequately been demonstrated. This relates to:

- Clinically and statistically significant difference in virological relapse rates found between the end of therapy and the end of the observation period.
- Inconclusive data in the response rate for the “difficult-to-treat” genotype 1 patients.
- Different rate of adverse events and the laboratory-related events judged as clinically relevant.
- The inadequate immunogenicity documentation because of the incomplete validation of the assays and methods used (and the consequent insufficient exclusion of false negative results and the factual differences observed in the detection of anti-interferon antibodies).

Due to the remaining major objections regarding quality and differences identified between Alpheon and Roferon-A in the quality and clinical comparability exercise, comparable quality, efficacy and safety of Alpheon and Roferon-A has not been adequately demonstrated.

the CHMP has recommended the refusal of the granting of the Marketing Authorisation for Alpheon.