20 September 2018
EMA/703393/2018
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Pelmeg

International non-proprietary name: pegfilgrastim

Procedure No. EMEA/H/C/004700/0000

Note

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

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Table of contents

List of abbreviations

ADA	Anti-drug antibody
AE	Adverse event
AEMPS	Agencia Española de Medicamentos y Productos Sanitarios (Spanish Regulatory Authority)
AGES	Österreichische Agentur für Gesundheit und Ernährungssicherheit (Austrian Regulatory Authority)
ALT	Alanine aminotransferase
ANC	Absolute neutrophil count
AS	Active substance
AST	Aspartate aminotransferase
AUC	Area under the curve
AUC	analytical ultracentrifugation
AUC%	Extrapolation of AUC from the last observation to infinity, expressed as a percentage of the total AUC
AUC0-inf	Area under the concentration-time curve (from time zero extrapolated to infinity)
AUC0-last	Area under the concentration-time curve (from time zero to last measurable concentration)
AUEC0-last	Area under the effect time curve
BfArM	Bundesinstitut für Arzneimittel und Medizinprodukte (German Regulatory Authority)
BMI	Body mass index
CD	circular dichroism
CEX	cation exchange chromatography
CI	Confidence interval
cIEF	Capillary isoelectric focusing
CGE	capillary gel electrophoresis
Cmax	Maximum concentration
CQA	critical quality attribute
CPPs	critical process parameters
CSR	Clinical Study Report
CV	Coefficient of variation
Da	Dalton
DSC	Differential scanning calorimetry
ECG	Electrocardiogram
ECL	Electrochemiluminescence
E. coli	*Escherichia coli*
EMA	European Medicines Agency
Emax	Maximum effect
EU	European Union
FIMEA	Finnish Medicines Agency (Finnish Regulatory Authority)
FMEA	Failure Mode and Effect analysis
FN	Febrile neutropenia
FP	finished product
GCP	Good Clinical Practice
GGT	Gamma glutamyltransferase
G-CSF	granulocyte colony-stimulating factor
GLM	General linear model
GMP	good manufacturing practice
HCP	host cell proteins
HIC	hydrophobic interaction chromatography
IMP	Investigational Medicinal Product
INN	international non-proprietary name
IEX-HPLC	ion exchange-high performance liquid chromatography
IPC	in-process controls
LB	Lower boundary
λz	Terminal elimination rate constant
MAA	Marketing Authorisation Application
Max	Maximum
MCB	master cell bank

MedDRA	Medical Dictionary for Regulatory Activities
Mono-PEG	Mono-pegylated product
NA	Not applicable
NOR	normal operating range
NSD	needle-safety device
MTT	tetrazolium compound
OOS	out of specification
PAR	proven acceptable ranges
PD	Pharmacodynamic
PDE	permitted daily exposure
PEG	Polyethylene glycol
Ph. Eur	European Pharmacopoeia
pI	isoelectric points
PK	Pharmacokinetic
PLA	parallel line assay
PPQ	Process Performance Qualification
PT	Preferred term
PVR	process validation run
QA	quality attribute
R	Reference (Neulasta)
rHU-met-G-CSF	recombinant methionyl human granulocycte colony-stimulating factor
RMP	reference medicinal product
RP-HPLC	reversed phase high performance liquid chromatography
SAE	Serious adverse event
SAP	Statistical Analysis Plan
s.c.	Subcutaneous
SE-HPLC	size exclusion high performance liquid chromatography
SmPC	Summary of Product Characteristics
SOC	System Organ Class
SPR	surface plasmon resonance
T	Test (Pelmeg)
t1/2	Half-life
TEAE	Treatment-emergent adverse event
tmax	Time to Cmax
tmax E	Time to Emax
UB	Upper boundary
US	United States
USP	United States Pharmacopoeia
UV	ultraviolet
PFS	pre-filled syringe
WCB	working cell bank

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Cinfa Biotech S.L. submitted on 8 September 2017 an application for marketing authorisation to the European Medicines Agency (EMA) for Pelmeg, through the centralised procedure falling within the Article 3(1) and point 1 Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 10 November 2016.

The applicant applied for the following indication: reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes).

The legal basis for this application refers to:

Article 10(4) of Directive 2001/83/EC – relating to applications for biosimilar medicinal products

The application submitted is composed of administrative information, complete quality data, appropriate non-clinical and clinical data for a similar biological medicinal product.

The chosen reference product is: Neulasta

Medicinal product which is or has been authorised in accordance with Union provisions in force for not less than 10 years in the EEA:

- Product name, strength, pharmaceutical form: Neulasta, 6 mg, solution for injection
- Marketing authorisation holder: Amgen Europe B.V.
- Date of authorisation: 22/08/2002
- Marketing authorisation granted by:
  - Union
- Marketing authorisation number: EU/1/02/227/001-004

Medicinal product authorised in the Union/Member State where the application is made or European reference medicinal product:

- Product name, strength, pharmaceutical form: Neulasta, 6 mg, solution for injection
- Marketing authorisation holder: Amgen Europe B.V.
- Date of authorisation: 22/08/200
- Marketing authorisation granted by:
  - Union
- Marketing authorisation number: EU/1/02/227/001-004

Medicinal product which is or has been authorised in accordance with Union provisions in force and to which bioequivalence has been demonstrated by appropriate bioavailability studies:

- Product name, strength, pharmaceutical form: Neulasta, 6 mg, solution for injection
- Marketing authorisation holder: Amgen Europe B.V.
- Date of authorisation: 22/08/2002
- Marketing authorisation granted by:
  - Union
- Marketing authorisation number: EU/1/02/227/001-004

Information on Paediatric requirements

Not applicable

Information relating to orphan market exclusivity

Not applicable

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.

Scientific advice

The applicant received Scientific Advice from the CHMP on 25 June 2015. The Scientific advice pertained to clinical aspects of the dossier.

1.2. Steps taken for the assessment of the product

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

Rapporteur: Koenraad Norga Co-Rapporteur: Andrea Laslop

The application was received by the EMA on	8 September 2017
The procedure started on	28 September 2017
The Rapporteur's first Assessment Report was circulated to all CHMP members on	19 December 2017
The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on	18 December 2017
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	3 January 2018
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	25 January 2018
The applicant submitted the responses to the CHMP consolidated List of Questions on	26 April 2018
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Questions to all CHMP members on	4 June 2018
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	14 June 2018
The Rapporteurs circulated the updated Joint Assessment Report on the responses to the List of Questions to all CHMP members on	22 June 2018
The CHMP agreed on a list of outstanding issues in writing and/or in an	28 June 2018

oral explanation to be sent to the applicant on	
The applicant submitted the responses to the CHMP List of Outstanding Issues on	14 August 2018
The Rapporteurs circulated the Joint Assessment Report on the Applicant's responses to the List of Outstanding Issues to all CHMP members on	5 September 2018 and 13 September 2018
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 Pelmeg on	20 September 2018

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

The intended indication for Pelmeg (also referred to as B12019 in this report) is "Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes)", ATC code L03AA13. One 6 mg dose (provided as a single prefilled syringe) of pegfilgrastim is recommended for each chemotherapy cycle, to be administered at least 24 hours following cytotoxic chemotherapy.

The Applicant claims the authorisation for Pelmeg as a similar product to Neulasta (EU) which was granted a marketing authorisation in the EU on 22 of August 2002. The proposed indication for Pelmeg is the same as for the reference product Neulasta (EU).

2.1.2. Epidemiology

Chemotherapy-induced neutropenia and its subsequent infectious complications represent the most common dose-limiting toxicity of cancer therapy. Febrile neutropenia, FN, develops in 25% to 40% of treatment-naïve patients during common chemotherapy regimens depending on the patient population; the dosage, timing and type of chemotherapy used[1]. The severity of febrile neutropenia depends on the dose intensity of the chemotherapy regimen, the patient's prior history of either radiation therapy or use of cytotoxic treatment, and comorbidities.

[1] Dinan MA, Hirsch BR, Lyman GH. Management of chemotherapy-induced neutropenia: measuring quality, cost, and value. J Natl Compr Canc Netw. 2015 Jan;13(1):e1-7.

2.1.3. Biologic features

Neutrophils contribute to both, the initiation and the maintenance of inflammation at sites of infection[2].

Chemotherapy regimens are associated with bone marrow suppression, resulting in reduced production of neutrophils (and also other blood cells like erythrocytes and thrombocytes). In clinical practice, neutropenia is the main limiting factor for the applicability of chemotherapy[3]. Both the duration of Grade 4 neutropenia (defined as absolute neutrophil count [ANC] of <0.5 x 109/L) and the depth of the nadir after chemotherapy are correlated with the development of infectious complications[4]. The risk of developing febrile neutropenia (FN) is thereby driven by the chemotherapy dose and schedule, and patient-related factors[5].

The principal regulator of physiological granulopoiesis human G-CSF is a glycoprotein that has been shown to regulate the production and release of neutrophils from the bone marrow, mediated via a single affinity extracellular receptor. By binding and signalling through granulocyte colony-stimulating factor receptor (G-CSFR), G-CSF has multiple effects on circulating neutrophils and on neutrophil precursors in bone marrow[6].

Stimulation of precursor cell proliferation in the bone marrow leads to an increase in the total mass of G-CSFR-expressing cells, which serves as a negative regulator of G-CSF levels through accelerated clearance of G-CSF[7].

2.1.4. Clinical presentation, diagnosis

Chemotherapy-induced neutropenia is a significant dose-limiting toxicity in cancer treatment and a major risk factor for infection-related morbidity and mortality. Febrile neutropenia, FN, develops in 25% to 40% of treatment-naïve patients during common chemotherapy regimens depending on the patient population; the dosage, timing and type of chemotherapy used[1]. In the clinical context, FN is defined as an oral temperature of >38.3°C or two consecutive readings of >38.0°C for 2 hours and an ANC of <0.5 x 109/L, or expected to fall below <0.5 x 109/L [Klastersky, de Naurois et al. 2016]. The occurrence of febrile neutropenia often necessitates chemotherapy delays or dose reductions. It may also lengthen hospital stay; increase monitoring, diagnostic, and treatment costs; and reduce patient quality of life.

[2] Panopoulos AD, Watowich SS. Granulocyte colony-stimulating factor: molecular mechanisms of action during steady state and 'emergency' hematopoiesis. Cytokine. 2008 Jun;42(3):277-88
[3] Khan S, Dhadda A, Fyfe D, Sundar S. Impact of neutropenia on delivering planned chemotherapy for solid tumours. Eur J Cancer Care (Engl). 2008 Jan;17(1):19-25
[4] Green MD, Koelbl H, Baselga J, Galid A, Guillem V, Gascon P, Siena S, Lalisang RI, Samonigg H, Clemens MR, Zani V, Liang BC, Renwick J, Piccart MJ; International Pegfilgrastim 749 Study Group. A randomized double-blind multicenter phase III study of fixed-dose single-administration pegfilgrastim versus daily filgrastim in patients receiving myelosuppressive chemotherapy. Ann Oncol. 2003 Jan;14(1):29-35
[5] Vogel CL, Wojtukiewicz MZ, Carroll RR, Tjulandin SA, Barajas-Figueroa LJ, Wiens BL, Neumann TA, Schwartzberg LS. First and subsequent cycle use of pegfilgrastim prevents febrile neutropenia in patients with breast cancer: a multicenter, double-blind, placebo-controlled phase III study. J Clin Oncol. 2005 Feb 20;23(6):1178-84
[6] Roberts AW. G-CSF: a key regulator of neutrophil production, but that's not all! Growth Factors. 2005 Mar;23(1):33-41
[7] Anderlini P, Champlin RE. Biologic and molecular effects of granulocyte colony-stimulating factor in healthy individuals: recent findings and current challenges. Blood. 2008 Feb 15;111(4):1767-72

2.1.5. Management

Primary prophylaxis with colony-stimulating factors, CSFs, reduces the frequency of chemotherapy induced neutropenia, all-cause mortality during chemotherapy, and need for hospital care e.g. in breast cancer[8]. The administration of G-CSF can accelerate the development of neutrophils from committed progenitors, thereby reducing the incidence, duration, and severity of neutropenia[9]. Forms of G-CSF such as filgrastim and lenograstim including biosimilars, are administered by a course of daily injections, whereas pegfilgrastim allows once-per-cycle administration and may avoid suboptimal daily dosing.

EORTC 2010 guidelines cover use of granulocyte-colony stimulating factor, G-CSF, to reduce the incidence of chemotherapy-induced febrile neutropenia in adult patients with lymphoproliferative disorders and solid tumours. Prophylaxis with a CSF is recommended for:

- Specified chemotherapy regimens with >20% risk of FN
- Specified chemotherapy regimens with 10% to 20% risk of FN, subject to patient specific risk factors such as elderly age (≥65 years) and neutrophil count
- Patients with a previous episode of FN

Pegfilgrastim and filgrastim can accelerate neutrophil recovery, leading to a reduced duration of the neutropenic phase in patients receiving cytotoxic chemotherapy. Filgrastim was initially approved for the prevention of infection as manifested by febrile neutropenia in patients with nonmyeloid malignancies receiving myelosuppressive chemotherapy. The pivotal study in patients with small cell lung carcinoma receiving cyclophosphamide, etoposide, and doxorubicin chemotherapy demonstrated an approximately 50% reduction in the incidence of febrile neutropenia and duration of Grade 4 neutropenia, as well as statistically significant reductions in the incidence of hospitalizations and IV antibiotic usage[10]. Subsequent indications for filgrastim included engraftment following bone marrow transplantation, mobilization of peripheral blood progenitor cells and engraftment following transplantation, induction or consolidation chemotherapy for acute myeloid leukemia, and severe chronic neutropenia. Because of its relatively short half-life of 3.5 hours, filgrastim is administered once daily by SC administration no less than 24 hours after chemotherapy and continuing until absolute neutrophil count (ANC) recovery within each cycle of treatment. Shortcomings of filgrastim include the requirement for either daily visits to the clinic or home injections by the patient during the period of administration, frequent ANC monitoring, the possibility of missed doses, and suboptimal duration of treatment (either too short or too long). Efforts to overcome these limitations led to the PEGylation of the G-CSF protein. The subsequent PEGylation of the G-CSF protein filgrastim altered the pharmacokinetic (PK) profile, resulting in slower clearance and a prolonged half-life (between 15 and 80 hours), thus permitting a single injection per cycle of chemotherapy[11]. Pegylation of filgrastim increases the size of filgrastim so that it becomes too large for renal clearance. Due to its high molecular weight, pegfilgrastim exhibits limited transport into the blood capillaries after SC administration and enters the systemic circulation via an indirect route, through the lymphatics.

[8] Renner P, Milazzo S, Liu JP, Zwahlen M, Birkmann J, Horneber M. Primary prophylactic colony-stimulating factors for the prevention of chemotherapy-induced febrile neutropenia in breast cancer patients. Cochrane Database Syst Rev. 2012 Oct 17;10

[9] Dale DC. Colony-stimulating factors for the management of neutropenia in cancer patients. Drugs. 2002;62 Suppl 1:1-15

[10] Crawford J, Ozer H, Stoller R, Johnson D, Lyman G, Tabbara I, Kris M, Grous J, Picozzi V, Rausch G, et al. Reduction by granulocyte colony-stimulating factor of fever and neutropenia induced by chemotherapy in patients with small-cell lung cancer. N Engl J Med. 1991 Jul 18;325(3):164-70

[11] Foley C, Mackey MC. Mathematical model for G-CSF administration after chemotherapy. J Theor Biol. 2009 Mar 7;257(1):27-44

With a long half-life and target-mediated clearance, pegfilgrastim remains in the circulation until the bone marrow neutrophil precursors start to come back after chemotherapy. Pegfilgrastim (Neulasta) was first authorized for marketing in the EU and US in 2002.

2.1.6. About the product

Filgrastim is a human granulocyte colony-stimulating factor (G-CSF) manufactured via recombinant technology, resulting in recombinant human N-methionyl granulocyte colony-stimulating factor (rHU-Met-G-CSF) of 175 amino acids with a molecular weight of 18,800 Da.

rHU-Met-G-CSF is produced by *Escherichia coli* (E. coli) bacteria and subsequently PEGylated by the conjugation of a polyethylene glycol (PEG) moiety to the primary amino group at the N-terminus of filgrastim to increase the exposure duration and therapeutic activity of the protein due to decreased renal clearance.

Pelmeg is developed as a similar biological medicinal product with Neulasta (pegfilgrastim) as the Reference Medicinal Product. Pegfilgrastim is a PEGylated (polyethylene glycol polymer chain), long-acting form of human recombinant granulocyte colony-stimulating factor (G-CSF; filgrastim). Filgrastim, the protein moiety of Pelmeg, is expressed in Escherichia coli (E. coli) and has an identical amino acid sequence as natural human G-CSF, except for an additional N-terminal methionine residue for the expression in E. coli. Pelmeg is identical to Neulasta with regard to its amino acid sequence. It is delivered in the same formulation (with the exception of pH which is 4.2) and presentation as Neulasta, in prefilled syringes containing 6 mg of the active substance in 0.6 mL solution for subcutaneous (s.c.) injection.

The proposed indication was as follows:

“Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes).”

The final agreed wording was as follows:

“Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes).”

Pelmeg therapy should be initiated and supervised by physicians experienced in oncology and/or haematology.

The posology is as follows: one 6 mg dose (a single pre-filled syringe) of Pelmeg is recommended for each chemotherapy cycle, given at least 24 hours after cytotoxic chemotherapy.

Type of Application and aspects on development

The clinical development programme for Pelmeg was designed and conducted using a sensitive comparability approach, waiving clinical efficacy studies in patients. This approach was chosen because a validated surrogate marker is available for G-CSF. Absolute Neutrophil Count (ANC) was used within the clinical development programme of Pelmeg as a surrogate for efficacy. The clinical approach is furthermore supported by the prerequisite similarity on the physicochemical, biofunctional, and preclinical PK/PD level for Pelmeg.

Two clinical studies were conducted for Pelmeg development:

- The pivotal study (study code: B12019-101) was a single-dose, randomised, double-blind, two-stage, two-way cross-over PK and PD study at a dose of 6 mg. This study enrolled 172 healthy subjects and assessed PK and PD as co-primary endpoints. A comprehensive immunogenicity assay programme, based on actual science for immunogenicity testing was implemented in the study.
- The supportive study (study code: B12019-102) was a multiple-dose, randomised, double-blind, three-period, two-sequence crossover study to assess the immunogenicity and PD comparability of Pelmeg and Neulasta at a dose of 3 mg. This study enrolled 96 healthy subjects and assessed PD and immunogenicity as co-primary endpoints. It comprised a multiple-dose parallel-group part in order to most sensitively detect differences in immunogenicity, using a sensitive set of assays for immunogenicity testing.

The development programme for Pelmeg, including clinical development, is in line with current CHMP guidelines and guidance:

- Guideline on similar biological medicinal products (CHMP/437/04 Rev 1).
- Guideline on similar biological medicinal products containing biotechnologyderived proteins as active substance: non-clinical and clinical issues (EMEA/CHMP/BMWP/42832/2005 Rev 1).
- Guidance on similar biological medicinal products containing recombinant GCSF (EMEA/CHMP/BMWP/31329/2005). This guideline is currently revised, see Concept paper on the revision of the guideline on non-clinical and clinical development of similar biological medicinal products containing recombinant granulocyte-colony stimulating factor (EMA/CHMP/BMWP/214262/2015).

Moreover, the clinical development programme was based on the EMA scientific advice the Applicant received for Pelmeg development in 2015:

1. The CHMP endorsed the assessment of clinical comparability with two clinical trials conducted in healthy volunteers. This study population is most sensitive with regard to detecting any potential differences between Pelmeg and Neulasta. Factors with potential impact on PK, PD, safety and immunogenicity, like drug-drug interactions, co-morbidities or immunosuppression are less or not present in healthy volunteers as compared to cancer patients.

2. The CHMP agreed to the use of ANC as a validated surrogate marker for demonstration of comparable efficacy, particularly if investigated in healthy volunteers.

3. The CHMP endorsed to investigate PK and PD comparability in a crossover study design as proposed for study B12019-101.

4. The CHMP confirmed the approach to evaluate immunogenicity in a clinical trial designed as a parallel-group study (study B12019-102) with repeated dosing of the same IMP. As immunogenicity is known to be low for filgrastim and pegfilgrastim, the investigation of immunogenicity in a parallel-arm design with healthy volunteers uses the most sensitive model for that purpose.

5. The CHMP requested additional PD evaluation, and suggested a dose in the range of 2-4 mg. The 3 mg dose was selected for study B12019-102 as this is within the ascending part of the dose-effect curve of pegfilgrastim[12], thus further enhancing the sensitivity of the chosen clinical study design. Also, the 3 mg dose was chosen based on operational reasons, to ensure precise dosing.

[12] Roskos LK, Lum P, Lockbaum P, Schwab G, Yang BB. Pharmacokinetic/pharmacodynamic modeling of pegfilgrastim in healthy subjects. J Clin Pharmacol. 2006 Jul;46(7):747-57

Based on the above, no confirmatory clinical studies in patients were conducted to demonstrate comparability of Pelmeg with Neulasta.

GCP

The non-clinical PK/PD study was performed according to GLP.

Both submitted clinical trials are claimed to be conducted in accordance with Good Clinical Practice (GCP) as referenced in ICH guidelines and in accordance with the ethical principles stated in the Declaration of Helsinki.

2.2. Quality aspects

2.2.1. Introduction

Pelmeg is a pegylated (polyethylene glycol polymer chain), long-acting form of human recombinant granulocyte colony stimulating factor (G-CSF; filgrastim) and is developed as a similar biological medicinal product to Neulasta (pegfilgrastim), as the reference medicinal product (RMP) for Pelmeg.

The finished product (FP) is presented as a solution for injection containing 6 mg of pegfilgrastim (INN) as active substance. The product is available in a 0.6 ml pre-filled syringe. Other ingredients are: sodium acetate; sorbitol (E420); polysorbate 20 and water for injections.

It is delivered in the same formulation (with the exception of pH (which is 4.2) and presentation, as Neulasta.

2.2.2. Active Substance

General information

The INN for Pelmeg active substance (AS) is pegfilgrastim. It is a clear and colourless liquid. Human G-CSF is a glycoprotein, which regulates the production and release of neutrophils from the bone marrow.

Filgrastim, the protein moiety of Pelmeg AS, is expressed in *Escherichia coli (E. coli)* and has an identical amino acid sequence as natural human G-CSF, except for an additional N-terminal methionine residue for the expression in *E. coli* (recombinant methionyl human granulocycte colony-stimulating factor (rHU-met-G-CSF)). Pelmeg AS is identical to Neulasta with regard to its amino acid sequence.

Recombinant filgrastim is pegylated by the addition of a single 21 kDa polyethylene glycol (PEG) moiety to the amino terminus of the protein. The protein is not glycosylated and polyethylene glycol (PEG) residues are added. Filgrastim contains five cysteine residues. Four of these are involved in disulphide bonds (i.e. cysteines 37 and 43, cysteines 65 and 75) while the cysteine at position 18 remains free.

Manufacture, characterisation and process controls

The manufacture of the active substance takes place at 3P Biopharmaceuticals in Spain. Appropriate GMP certificates have been provided.

Description of manufacturing process and process controls

The manufacturing process of Pelmeg consists of fermentation, cell lysis, inclusion body recovery, inclusion body solubilisation and refolding prior to purification via a series of chromatography and filtration steps to produce the filgrastim critical intermediate. Filgrastim is then purified prior to

pegylation and further chromatographic and filtration purification steps follow. Finally, the active substance is sterile-filtered at room temperature into a container-closure system (of specified composition) and stored as the final AS. No reprocessing has been claimed during AS manufacture. The containers comply with Ph. Eur. and USP requirements for container closure systems and plastic immediate packaging materials. Suitability of the container closure system has been investigated in a leachable study. Relevant process controls are in place to ensure control and consistency of the process and of the active substance. In general, the manufacturing process has been sufficiently described and in-process controls are adequately set to control the process.

Control of materials

All raw materials used in the AS manufacturing process are described and are either compendial grade or are tested according to in-house standards. The composition of the different media and buffers is sufficiently detailed. The starting material of the PEG moiety has been defined; activated PEG is re-classified as an intermediate further to the major objection raised during the procedure. No human or animal derived materials are used in the active substance manufacturing process, nor used in the manufacture of the MCB.

Filgrastim, the protein moiety of Pelmeg AS, is expressed in *E. coli* cells. The complete nucleotide sequence for the expression of filgrastim is included in the marketing authorisation application, along with information on the synthesis of the vector and its transformation into cells.

A two-tiered cell banking system is used and information is provided regarding testing of MCB and WCB and release of future WCBs. Cell banks have been tested for viable cells, purity, identity and genetic stability with characterisation also of end of production cells (EOPCs). Cell bank storage is described and is acceptable. The cell bank system (MCB and WCB) has been described, including information on the cell bank stability program and further testing on the EOPCs. Generation, testing and release of future WCBs will follow the same protocol as used for the initial WCB.

Control of critical steps and intermediates

A comprehensive overview of the process parameters and in-process controls (IPCs) in place throughout the manufacturing process of the AS is given. An iterative approach was utilised to identify critical process parameters (CPPs) for the critical process steps. The potential CPPs identified using a risk assessment failure mode effect analysis (FMEA) approach were further investigated in process characterisation studies and using knowledge gained from commercial scale manufacturing experience. This allowed identification of proven acceptable ranges (PARs) for scale-independent critical process parameters (CPP), which could be used to support assessment of impact on AS quality in the event of deviation from the normal operating range (NOR). The NORs have been set based on the data and experience gained from a suitable number of validation batches.

Intermediate storage times were established by respective stability studies and hold times during manufacture are described and supported by data. Column resin cleaning, regeneration and re-use have been described as has membrane re-use, where applicable. The manufacturing process incorporates several mechanisms to ensure the active substance meets predetermined quality criteria for microbiological control.

Filgrastim is considered as a critical intermediate. Currently, the proposed combination of tests is sufficiently indicative of potency. Batch analysis and stability data of filgrastim are acceptable. The proposed storage condition and time for this intermediate in specified bags is accepted.

The starting material has been defined for the PEG moiety. The manufacturing process has been elaborated in sufficient detail. Data on specifications of the activated mPEG and the starting material,

batch release and stability data of starting material and intermediate and storage conditions have been provided.

Process validation

Following process development and scale-up to the commercial batch scale, the Process Performance Qualification (PPQ) campaign was executed to demonstrate that the process consistently delivers a product that meets pre-defined acceptance criteria for quality. Process performance consistency was demonstrated through the control of CPPs and via introduction of process performance parameters (i.e. IPCs), which were maintained within the pre-determined acceptance criteria.

In general, all the CPPs and IPCs were within the pre-determined acceptance ranges. Any deviations were appropriately investigated and none challenged the conclusions of the process validation. The validation of the Pelmeg DS manufacturing process provides sufficient evidence that the process, when operated within established parameters, performs effectively and reproducibly. The hold time steps used during AS manufacture were properly validated. Also impurity clearance was validated, showing that the process is capable of removing process-related impurities in a consistent way to sufficiently low levels. Host cell proteins (HCPs), host cell DNA and other specified residuals were identified as likely impurities in Pelmeg AS. Acceptable levels of impurities were determined based on safety assessment that determined the acceptable daily exposure and historical data from commercial manufacturing batches.

Clearance of relevant impurities to acceptable levels was demonstrated during the PPQ campaign and will be monitored during commercial manufacturing. In line with guideline ICH Q3D, elemental impurities were assessed. A risk assessment of organic solvents in Pelmeg AS was also conducted. Stated impurities have been present in batches studied in non-clinical and clinical studies.

For the chromatographic purification steps, studies were performed to define and validate the column resin lifetimes.

Manufacturing process development

Manufacturing process development was performed in three stages:

- Early stage development of the upstream and downstream processes at batch scale
- Scale up
- Late stage development to confirm normal operating ranges (NORs) assigned to each CPP and to create proven acceptable ranges (PARs) as well as scale down model qualification.

The biosimilarity testing, non-clinical, and clinical studies were performed with the final commercial scale material. All process changes were introduced prior to non-clinical studies. The non-clinical study, as well as the clinical studies, were conducted using post-change product material of the final commercial scale process.

Overall, the history of the manufacturing process development is well-described. Changes to the process during development are described and justified sufficiently.

Characterisation

To ensure appropriate safety, efficacy, and pharmacokinetics, an extensive analytical program was executed to demonstrate that Pelmeg AS has the expected structure and function based on known information about pegfilgrastim. Characterisation studies were designed to interrogate all aspects of structure and biological function and potency. Physicochemical properties investigated included: intact mass determination; polydispersity analysis; structural characterisation, including analysis of amino acid sequence, pegylation site, and disulphide bridging; higher order structure; post-translational

modifications and biological activity. Pelmeg AS mass distribution was in the expected range. The complete amino acid sequence was verified. The filgrastim protein is pegylated at the same site as described for the reference product (the N-terminal methionine). The disulphide bond structure of pegfilgrastim and filgrastim was shown to be consistent with the expected structure.

Post-translational modifications, other product-related variants and relevant degradation pathways within the protein sequence of pegfilgrastim were identified and quantified for both active substance and finished product. Stressed stability studies showed that some oxidation and deamidation may occur during shelf life. However, these modifications did not impact potency.

The potency of Pelmeg AS was analysed using a cell proliferation assay, which measures the biological activity of pegfilgrastim based on its binding to, and induction of the proliferation of specified cells. The method is closely aligned with the Ph.Eur. method for potency assay of filgrastim. The receptor binding properties of Pelmeg AS were also studied.

Specification

The AS specifications include tests for: characteristics (e.g. colour and clarity of solution), identity, purity/impurities, content, potency and excipients. In general, the specifications proposed are acceptable and properly justified. The applicant is recommended to continue exploring the implementation of an alternative, more accurate analytical method to determine polysorbate 20 levels in the active substance specification (see recommendation). In the meantime, to ensure adequate control of polysorbate 20, the Applicant has introduced appropriate in-process controls on polysorbate 20 solutions used for formulation.

Analytical methods

Method descriptions for all non-compendial analytical procedures used for release and stability testing are provided. For the compendial methods, no method descriptions are submitted which is acceptable, as they are performed according to the respective Ph. Eur. monographs. The descriptions are of sufficient detail. An in vitro proliferation assay is used for potency determination. It determines potency of human modified Granulocyte-Colony Stimulating Factor (i.e. pegylated G-CSF) using a specified cell line, which proliferates only in the presence of G-CSF. The method is closely aligned to the Ph.Eur potency assay for filgrastim.

Batch analysis

Batch data were provided (several commercial-scale and process batches including non-clinical, clinical and process validation batches) showing that all batches were consistent and complied with the specifications.

Reference materials

The applicant has described the qualification of the different reference standards that were used during development of Pelmeg, as well as the primary and secondary reference standard that will be used for commercial production. In addition, an analytical testing programme for future secondary reference standards was provided. It was confirmed and included in the MAA file that any future secondary RS will be tested according to the test panel described in the dossier. In addition to the qualification against the primary reference standard, the future secondary reference standards will be calibrated against the International Pegfilgrastim Reference Standard. This information has been integrated into the dossier.

Stability

The proposed shelf life for the Pelmeg AS in the specified container closure system has been described.

Stability studies under long-term, accelerated and stress conditions have been performed on Pelmeg AS. Some studies are still on-going. Stability studies are conducted in line with ICH guidance.

The proposed shelf is based on the current, available long-term stability data of AS batches.

The primary stability studies included clinical batches and a suitable number of process validation batches. These studies have now been completed. Updated stability data have been provided including additional data from process validation batches. Accelerated and stress condition studies have been completed.

Supportive stability data from a development batch used in the pre-clinical programme (non-GMP batch) are also available.

The initial stability data demonstrated that most of the AS quality parameters are stable and do not show any trends. Updated stability data revealed that most of the AS quality parameters are stable and do not show any trends. However, updated stability data revealed that results for some tests were above the limits for some batches (at several time points) due to storage in small scale containers. No OOS results have been observed at FP level for stability, indicating that full scale AS is being stored properly. Stability data thus far for the commitment batches show that all parameters were well within the acceptance criteria. It was therefore demonstrated that the issues are related to storage in the small containers, and not to the active substance itself. The study is planned to continue. The Applicant is requested to submit the long term stability data of the additional active substance batches when the study has been completed (see recommendation). Based on the data provided, the proposed shelf life for the active substance can be accepted.

2.2.3. Finished Medicinal Product

Description of the product and pharmaceutical development

The final formulation of Pelmeg is a sterile, clear, colourless solution for injection in a disposable, single-use, 0.6 ml pre-filled syringe (PFS) with automatic needle safety guard.

Pelmeg is identical to Neulasta in terms of pharmaceutical strength, composition, route of administration, as well as presentation. Moreover, the formulation of Pelmeg is also identical to Neulasta (with a slightly higher pH for Pelmeg of 4.2). The intended Pelmeg commercial formulation is that used in clinical studies.

The following excipients, all complying with Ph.Eur. are added at the final stage of the manufacturing process of the active substance: sodium acetate (buffer), sorbitol (E420 (solvent)), polysorbate 20 (surfactant) and water for injection; if necessary, pH is adjusted with sodium hydroxide or hydrochloric acid to pH 4.2. Unlike for Neulasta, nitrogen is used during filling to decrease the potential oxidation of the product. This is acceptable. There are no overages. There is an overfill to allow withdrawal of 0.6 ml.

Pelmeg is presented in a pre-filled glass syringe (Type I glass) with a rubber stopper and a stainless steel needle with an automatic needle guard. The needle cap is latex-free. The container closure was selected to mimic Neulasta and complies with EU requirements. The integrity of the container closure system has been studied during development. Leachables and extractables have been studied. The Pelmeg prefilled syringe (PFS) is assembled with a specific needle-safety device (NSD). The same medical device is used in the reference product Neulasta. This was assessed for suitability during the review.

Formulation studies were performed with a focus on the optimisation of the formulation buffer. The choice of the buffer system and pH were discussed and the difference in the pH value compared to the reference product in the formulation sufficiently justified by data. This case is acceptable and does not exclude biosimilar comparability to the reference product.

Critical aspects in the manufacturing process development were discussed by the applicant. Microbiological attributes have been sufficiently discussed, and do not raise any questions or concerns.

Manufacture of the product and process controls

The finished product is released by PharmaKorell GmbH in Germany.

Description of manufacturing process and process controls

The commercial manufacturing scale (fermentation) for the FP has been defined.

The FP manufacturing process therefore comprises formulated bulk sterile filtration of Pelmeg AS (post-filtration integrity testing is conducted), syringe filling, stoppering and final packaging assembly. No further formulation is performed on Pelmeg AS after it is introduced into specified containers. All steps of the Pelmeg FP manufacturing process are considered critical and are thus strictly controlled during the manufacturing process. The processes for filling and packaging of Pelmeg follow established standard operations for the manufacturing of parenteral products in PFS. Critical process parameters and in-process tests are sufficiently described.

Process validation studies using an appropriate number of batches were performed to demonstrate that the manufacturing process operated within the established parameters and was capable of effectively and reproducibly producing product that meets pre-defined acceptance criteria. The following validation studies related to the manufacturing of Pelmeg have been performed: cleaning validation; sterility validation; aseptic filling process validation; packaging and shipping validation.

The validation data show that the process is robust and well controlled. Since manufacturing and evaluation of additional batches is not possible within the EMA review procedure, the Applicant is recommended to provide validation data on filling for additional FP batches (using the optimised nitrogen supply and filling speed), as soon as those data are available.

Product specification

The Pelmeg FP specifications include general tests (e.g., colour and clarity of solution, extractable volume, container appearance) as well as tests for identity, purity, impurities, content and potency. The applicant provided acceptable justifications for the specifications. Analytical methods were sufficiently described and non-compendial methods were validated.

As there are no additional formulation steps prior to Pelmeg filling, the product impurities present in Pelmeg are the same as those identified and controlled in Pelmeg AS.

In general, the product-related impurities of Pelmeg are at low levels. Aggregate levels, which could be a potential safety concern, are at non-critical levels. Charged variants are very low and the total pre- and post-peaks in RP-HPLC testing are also low.

The Applicant is recommended to revise the CEX-HPLC specification limit when data from further batches are available (see recommendation).

For elemental impurities, a risk assessment has been performed for Pelmeg AS. Extractable studies have been performed and no potential leachable compounds were found. For the control of

microbiological attributes, sterility and endotoxin testing as well as container closure integrity are performed as release tests. In conclusion, the specifications are acceptable.

Analytical methods

The FP analytical methods are similar as those used for the AS release testing.

Batch analysis

Batch data from development, clinical and a suitable number of validation lots are presented. Batch data indicate consistent quality of batches which all comply with the specifications.

Reference materials

Pelmeg FP is released against the same reference standards and control materials described for AS.

Stability of the product

A shelf-life of 24 months is proposed for Pelmeg, when stored at 2°C - 8°C and kept in the in the outer carton in order to protect from light.

Stability studies include commercial scale, development and process validation run (PVR) batches. The stability studies involve formal studies at long-term, accelerated and stress conditions, which are performed at 5°C ± 3°C, 30°C ± 2°C/65% ± 5% relative humidity (RH) and 40°C ± 2°C/75% ± 5% RH, respectively in accordance with ICH guidelines.

The proposed shelf life for Pelmeg is 24 months when stored at 5 ± 3°C. No critical changes were observed at long-term conditions. The available stability data show that the FP is stable and support the proposed shelf life of 24 months for the FP. No critical changes were observed at long-term conditions.

In addition, temperature excursion studies (out-of-fridge, OOF and freeze/thaw, F/T) are performed batches to assess the stability of Pelmeg under normal use conditions. Photostability was also studied.

The available stability show that the FP is stable. Overall, the available data support the proposed shelf life of 24 months for the FP.

The applicant is asked to provide the stability data for deamidation and oxidation when the long term stability studies for specified FP batches 17001 and 17002 have been completed (see recommendation).

The FP shelf life may be extended to 36 months via a variation procedure once 36m stability data are available for the 2 batches produced with the commercial process (lots 17001 and 17002).

The Applicant has provided the OOF stability results, which demonstrate that Pelmeg remains stable at 30 ± 2 °C for a single period of 96 hours after 36 months storage at long-term conditions. The F/T results demonstrate that Pelmeg remains stable for two periods of less than 72 hours.

The following special precautions for storage are supported by the stability data and are approved in the SmPC: *Pelmeg may be exposed to room temperature (not above 30°C) for a maximum single period of up to 96 hours. Pelmeg left at room temperature for more than 96 hours should be discarded. Do not freeze. Accidental exposure to freezing temperatures for two periods of less than 72 hours does not adversely affect the stability of Pelmeg.*

A shelf-life of 24 months is agreed for Pelmeg, when stored at 2°C - 8°C.

Adventitious agents

Microbial adventitious agents are controlled throughout the manufacture of Pelmeg AS and Pelmeg FP. Pelmeg AS is checked for bioburden as IPC prior to sterile filtration and filling. The FP is sterile filtered and Pelmeg is tested for sterility and container closure integrity at release. Only raw materials and excipients certified as of non-animal origin are used for manufacture of Pelmeg AS and FP.

The host cell used for the manufacture of Pelmeg AS is an *E. coli* derivative. Based on the microbial nature of the manufacturing process and the materials used (none of animal origin), the risk of viral contamination of Pelmeg can be excluded.

Biosimilarity analysis

Biosimilarity of Pelmeg to the RMP was assessed during development using physicochemical and biofunctional data from side-by-side studies. The side-by-side physicochemical and biofunctional similarity assessments have been supplemented by side-by-side stability testing to demonstrate that the degradation behaviour of Pelmeg is similar to that of the RMP.

First, multiple batches of the RMP were sourced in order to establish the Quality Target Product Profile (QTPP). An initial similarity assessment was performed at pilot scale using several Pelmeg AS and several EU-authorised RMP batches in order to steer the development process. The AS manufacturing process was then scaled-up to the final commercial scale, whereby the manufacturing process was adjusted and modified to fit to the increased scale. This scale-up was guided by the QTPP and the critical quality attribute (CQA) assessment. Next, a side-by-side study at commercial scale, compared Pelmeg FP batches of commercial scale against RMP batches. Finally, a second side-by-side study at commercial scale compared several Pelmeg batches and several RMP batches. The non-clinical and clinical studies were performed with batches from the final manufacturing process at commercial scale. Moreover, non-clinical and clinical batches were included in the aforementioned side-by-side studies.

Pelmeg is delivered in an identical formulation and the same presentation as the RMP, with the exception of pH. Pelmeg's pH (4.2) is slightly higher than that of the RMP and was chosen to provide optimal conditions for a stable buffer system for Pelmeg. This slightly increased pH in comparison to the RMP has no impact on the structure and biological function of Pelmeg as demonstrated by the physicochemical and biofunctional similarity data. Analytical data obtained fora suitable number of EU-authorised RMP batches over all stages of product development formed the basis of the similarity range for each quality attribute (QA) to which Pelmeg results were compared. The analytical similarity exercise was performed on several commercial scale Pelmeg lots versus several RMP batches, depending on the method and parameter tested. To analyse whether similarity to the RMP was met, the results were evaluated against the similarity range. More detailed information including a justification on the batches used (age/number) for specific tests was provided.

State-of-the-art, orthogonal analytical methods were employed to characterise structural and functional similarities and differences to demonstrate analytical similarity between Pelmeg and the RMP. Also comparative stability and degradation profiles were assessed.

Primary Structure

The complete amino acid sequence of Pelmeg was verified and is identical to the amino acid sequence of the RMP. The correct formation of the disulphide bridges (Cys37-43, Cys65-75) was confirmed. No peptides containing disulphide bonded cysteine 18 or mispaired disulphides were found.

Intact mass and PEGylation

MALDI-TOF/TOF and capillary gel electrophoresis (CGE) were used to compare the molecular weight of Pelmeg and RMP. MS analysis showed that Pelmeg and the RMP have similar mass. Mono-PEGylation at the N-terminal methionine was confirmed for Pelmeg. There are 4 lysines (K17, K24, K35 and K41) which are considered putative sites for positional pegylation. No signals for the pegylated peptides containing lysines K17, K24, K35 and K41 were found.

The PEG average molecular weight and polydispersity index were compared for Pelmeg and RMP side-by-side using ESI-MS and found to be similar. The size profiles and PEG moieties of Pelmeg and the RMP were also compared.

Higher order structure

Pelmeg CD data and near-UV CD were compared with the RMP and revealed a high level of similarity. Far-UV CD data suggested α-helices to be the predominant secondary structure (at 25°C between 75% and 78%) with the 3/10 helix being the second most dominant structure. A very small percentage of β-sheets were determined.

Differential scanning calorimetry (DSC) was performed and showed that the melting temperatures of Pelmeg and RMP (to assess the similarity of higher order structure) were comparable between the two products. Fluorescence spectroscopy was used to compare the conformation of Pelmeg and RMP giving similar results.

Purity and impurities

Cation exchange chromatography (CEX) was used to evaluate similarity of Pelmeg and RMP with respect to charge variant profiles. Overall, the profile of main acidic and basic variants of Pelmeg closely matched that of the RMP, suggesting comparable isoform profiles between the two products. The main variant level of Pelmeg showed higher amounts than observed in the RMP, demonstrating a higher charge-related purity of Pelmeg compared to the RMP. However, this difference is not clinically-meaningful given that the material analysed via CEX was also used in a clinical study that showed a high degree of comparability between Pelmeg and the RMP. Thus, it can be concluded that the charged variant profile of Pelmeg is similar to the RMP.

Capillary isoelectric focusing (cIEF) was performed to compare the isoelectric points (pI) of Pelmeg and RMP. The pI of Pelmeg and RMP by cIEF was comparable, suggesting a comparable structure. One batch of Pelmeg showed a slightly higher pI. This difference is covered by the variability of the method. Moreover, this particular batch was also used in a clinical study that showed a high degree of comparability between Pelmeg and the RMP.

Reversed phase chromatography (RP-HPLC), which separates product variants, including oxidised and deamidated species on the basis of hydrophobicity, was performed and showed comparability between Pelmeg and RMP purity profiles. The sums of pre-peaks and post-peaks were also similar between Pelmeg and the RMP, suggesting comparable hydrophobicity-related impurity profiles between the two products.

The level of methionine 122 oxidation was found to be similar between Pelmeg and the RMP. The methionine 127 or 138 oxidation data showed variability, and results for two Pelmeg batches were slightly outside the pre-specified similarity limits. However, this is not considered critical as these values indicate Pelmeg contains less oxidation than the RMP. Furthermore, no impact was observed in the clinical studies performed with these lots.

Post-translational modifications were investigated. For both products, the impurity profile was found to be similar. The overall level of modifications is also similar, but slightly higher in the RMP. However,

the latter may be due to the age differences between Pelmeg and the RMP batches that were analysed. The most prominent post-translational modifications detected were the following: Single oxidation at tryptophan (W) 119 or methionine (M) 122; Single oxidation at methionine (M) 127 or methionine (M) 138; Single deamidation at glutamine (Q) 108, 120 or 121.

The identity measured via immunoreactivity (western blot analysis) for Pelmeg and the RMP was similar. In addition, size exclusion chromatography (SEC-HPLC) was performed to compare the monomer and aggregate content of Pelmeg and the RMP.

Analytical ultracentrifugation (AUC) was performed to provide an orthogonal comparison of monomer, oligomer and higher oligomer proportions of Pelmeg and the RMP. Overall, sedimentation patterns were similar for Pelmeg and the RMP, suggesting comparable amounts of monomers, and low and higher oligomers for the two products.

Free PEG

The levels of residual PEG for Pelmeg and the RMP were compared by RP-HPLC-ELSD and revealed a high level of similarity.

General properties

Extinction coefficients of Pelmeg closely matched that of the RMP, suggesting comparable amino acid content. Pelmeg protein content for all batches was within the range of the RMP, which reveals a high level of similarity for protein content between Pelmeg and the RMP. Osmolality and extractable volumes were also measured for Pelmeg and RMP and closely matched.

Potency comparison by cell proliferation assay

The relative potency of Pelmeg and RMP was compared by proliferative bioassay (cell proliferation assay measures the biological activity of pegfilgrastim) and in general, revealed a high level of similarity. This assay is closely aligned to the Ph.Eur potency assay for filgrastim.

Receptor binding comparison by SPR

The functional similarity of Pelmeg and RMP was also found comparable by measuring receptor binding to the recombinant human G-CSF receptor (rhG-CSFR) using surface plasmon resonance (SPR).

Results of comparative stability

Comparative stability testing of Pelmeg and RMP was performed using a variety of stability-indicating methods. The results of the comparative stability testing showed a high level of similarity between Pelmeg and the RMP for all attributes tested. Purity and impurity profiles as measured by RP-HPLC showed similar degradation rates. Potency also showed similar trends for both Pelmeg and RMP.

The outcome of the physicochemical and biological comparability exercise between Pelmeg and Neulasta is summarised in the tables below.

Table 1: Tabular summary of physicochemical similarity results between Pelmeg and EU Neulasta

Quality attribute	Parameter	Analytical method for control and characterisation	Conclusion
Primary Structure	Primary sequence	LC-MS and Edman	Identical
	Disulphide bonding pattern comparison	LC-MS	Similar
Intact Mass and Pegylation	Pelmeg intact mass	LC-MS	Similar
	Molecular weight	Capillary SDS-PAGE (CGE)	Similar
	Positional Pegylation	LC-MS/Edman	Identical
	PEG polydispersity	ESI-MS	Similar
	PEG identity comparison	SDS-PAGE titrisol	Similar
Higher order structure	Secondary and tertiary structure comparison	Circular Dichroism (CD)	Similar
	Tertiary structure comparison	Differential scanning calorimetry (DSC)	Similar
		Intrinsic fluorescence spectrometry	Similar
Purity and impurities – product related variants	Charge variants	CEX-HPLC	Similar
		Isoelectric Focussing (cIEF)	Similar
	Purity and impurities	RP-HPLC	Similar
	Methionine Oxidation	UPLC-UV-MS	Slight differences observed for Pelmeg, but not considered clinically meaningful nor critical for scientific evaluation of similarity
	Low- (LMW) and high molecular weight (HMW) species	Western blot	Similar
		SEC-HPLC	Similar
		Analytical ultracentrifugation (AUC)	Similar
Purity and impurities – other impurities	Residual free PEG	RP-HPLC-ELSD	Similar
General properties	Extinction coefficient	RP-HPLC followed by fluorescence	Similar
	Protein content comparison	UV/VIS	Similar
	Osmolality comparison	Ph. Eur. 2.2.35	Similar
	Extractable volume	Ph. Eur. 2.9.17	Similar

Table 2: Tabular summary of biofunctional similarity testing results between Pelmeg and EU Neulasta

Quality attribute	Parameter	Analytical method for control and characterisation	Conclusion
Biofunctional testing	Potency	Cell-proliferation assay	Similar
	Receptor binding	Surface Plasmon Resonance	Similar

Table 3: Accelerated (30 °C ± 2 °C, 65% ± 5% relative humidity) and stress (40 °C ± 2 °C, 75% ± 5 % relative humidity) results for comparability stability testing between Pelmeg and EU Neulasta

Quality Attribute	Analytical method	Conclusion
Identity	SDS-PAGE Coomassie	Similar profile
	SDS-PAGE Silver	Similar profile
Higher order structure	CD	Similar profile
Charge variants	Gel IEF	Similar profile
Purity and impurities	RP-HPLC	Similar profile
Immunological ID	Western Blot	Similar profile
Aggregates	SEC-HPLC	Similar profile
Protein content	UV/VIS	Similar profile
Osmolality	Ph. Eur.	Similar profile
Activity	Cell proliferation assay	Similar profile
Clarity/Colour	Visual inspection	Similar profile
pH	Potentiometry	Similar profile
Visible particles	Ph. Eur	Similar profile
Sub-visible particles	Ph. Eur	Similar profile

Table 4: Long-term (5°C ± 3° C) results for comparability stability testing between Pelmeg and EU Neulasta

Quality attribute	Analytical method	Conclusion
Clarity/Colour	Visual inspection	Similar profile
Identity	SDS-PAGE Coomassie	Similar profile
	SDS-PAGE Silver	Similar profile
Higher order structure	CD	Similar profile
Tertiary structure	Intrinsic fluorescence	Similar profile
Aggregates	SEC-HPLC	Similar profile
Purity and impurities	RP-HPLC	Similar profile
Isoelectric point	Gel IEF	Similar profile
	cIEF pI main peak	Similar profile
Activity	Cell proliferation assay	Similar profile
Protein content	UV/VIS	Similar profile
pH	Potentiometry	Similar profile
Osmolality	Ph. Eur.	Similar profile
Visible particles	Ph. Eur.	Similar profile
Sub-visible particles	Ph. Eur.	Similar profile
Extractable volume	Extractable volume	Similar profile

Conclusion on biosimilarity (quality level)

The primary structure of Pelmeg was found to be identical to that of the RMP, with identical site of pegylation for both products. Molecular weight and polydispersity indicated similar PEG moieties between Pelmeg and the RMP. Moreover, the higher order structure, product-related variants, and the impurity and aggregation profiles, were also shown to be similar between Pelmeg and the RMP. Furthermore, relative potency and recombinant human G-CSF receptor binding kinetics were similar for Pelmeg and the RMP. Comparative stability testing demonstrated that Pelmeg and the RMP degrade in a comparable manner. In conclusion, the data of the physicochemical, biofunctional, and stability tests indicate that the proposed biosimilar biological product Pelmeg is similar to the RMP.

2.2.4. Discussion on chemical, pharmaceutical and biological aspects

The AS and FP manufacturing process and process controls are described, as are raw materials. A cell bank system was established and tested and qualified. Critical process parameters were identified and the process was appropriately validated. As regards the activation of PEG into activated PEG, the applicant has provided detailed information on the process and controls.

As regards the AS stability, the Applicant is requested to submit the long term stability data of the additional active substance batches (in a specified container-closure system) post-approval, when the study has been completed (see recommendations).

As regards the FP manufacturing process, validation data on filling for additional FP batches (using specified optimised parameters) are expected post-approval (see recommendations).The AS and FP specifications proposed by the applicant are deemed suitable to control the quality of AS and FP. Analytical methods were described in detail. AS and FP specifications are properly justified. As regards CEX-HPLC, due to the limited data available for this quality parameter, the applicant has proposed a more flexible limit. This is deemed acceptable, however the applicant has been asked to revise the CEX-HPLC acceptance limit when more data are available (see recommendations). The applicant will also submit a variation post-approval to set definitive specification limits for polysorbate 20 based on a higher number of batches (see recommendations).

For Pelmeg AS, an appropriate shelf life and storage conditions have been defined. This shelf life is based on the current, available long-term stability data.

The proposed shelf life for Pelmeg FP is 24 months when stored at 5 ± 3°C. The available stability data support this. The applicant should however provide, post-approval, the results for deamidation and oxidation when the long term stability studies for specified FP batches have been completed (see recommendations).

The OOF study suggests that the FP remains stable when exposed to 30°C for 96 hrs.

Biosimilarity analysis

For the biosimilarity analysis, the applicant has performed an extensive comparability exercise between EU Neulasta and Pelmeg FP (including both process validation and clinical batches). In general, all quality attributes analysed proved to be highly similar between Pelmeg and EU Neulasta. For a few parameters slight differences were observed. However, these differences were properly justified and shown to have no impact on safety or efficacy of the product. Importantly, functional testing showed high similarity between both products. From a quality point of view Pelmeg can be considered as biosimilar to EU Neulasta.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The manufacturing processes for Pelmeg AS and FP are adequately described, sufficiently controlled and properly validated. AS and FP are sufficiently controlled. Container closure systems of AS and FP were qualified. The currently available stability data for AS and FP do not indicate any decrease or trends for potency or purity. The data of the physicochemical, biofunctional, and stability tests confirmed that from a quality point of view Pelmeg can be considered as biosimilar to EU Neulasta.

In conclusion, the quality section of the Pelmeg MAA file is approvable and several recommendations for future quality development are made.

2.2.6. Recommendations 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 the following points for investigation:

- The applicant is recommended to continue exploring the implementation of an alternative, more accurate analytical method to determine polysorbate 20 levels in the active substance. Upon method validation, this alternative method may be implemented via a variation procedure post-approval. When this variation will be submitted post-approval, it is expected that the specification be revised.
- The applicant is recommended to submit the long term stability data of the additional active substance batches in the specified container-closure system when the study has been finalised.
- The applicant is recommended to provide additional validation data on filling of FP using specified optimised conditions as soon as these data are available.
- The applicant is recommended to revise the limit for the CEX-HPLC specification of the finished product (including an appropriate justification) if possible, when additional data are available.
- The applicant is recommended to provide the stability data for deamidation and oxidation when the long term stability studies for finished product batches have been completed.

2.3. Non-clinical aspects

2.3.1. Introduction

In vitro and *in vivo* studies were conducted to assess the biosimilarity of Pegfilgrastim (Pelmeg) compared to Neulasta, using a variety of batches of both Pelmeg and Neulasta. Biosimilarity studies included *in vitro* cell-based models (M-NFS-60 cells, a murine myeloblastic cell line) and receptor binding assays by SPR; and *in vivo* PK/PD studies in normal and neutropenic rats.

The nonclinical programme of pegfilgrasim included a series of *in vitro* comparative studies including a cell proliferation assay on a specified cell line (method closely aligned to the Ph.Eur method for potency assay of filgrastim) and a comparison of binding to granulocyte colony-stimulating factor receptor (G-CSFR) by Surface Plasmon Resonance (SPR).

A GLP-compliant non-clinical comparative study in healthy and neutropenic rats, which evaluated pharmacokinetics (PK) and pharmacodynamics (PD) after a single dose of 15 or 100 µg/kg was also conducted to further support the similarity demonstration between Pelmeg and Neulasta.

2.3.2. Pharmacology

Primary pharmacodynamic studies

1. Cell proliferation assay:

The results of the cell proliferation assay are provided in the figure below.

One measurement showed a value slightly above the specified similarity range, but was within the set specifications for the proliferation assay of 80% to 125%.

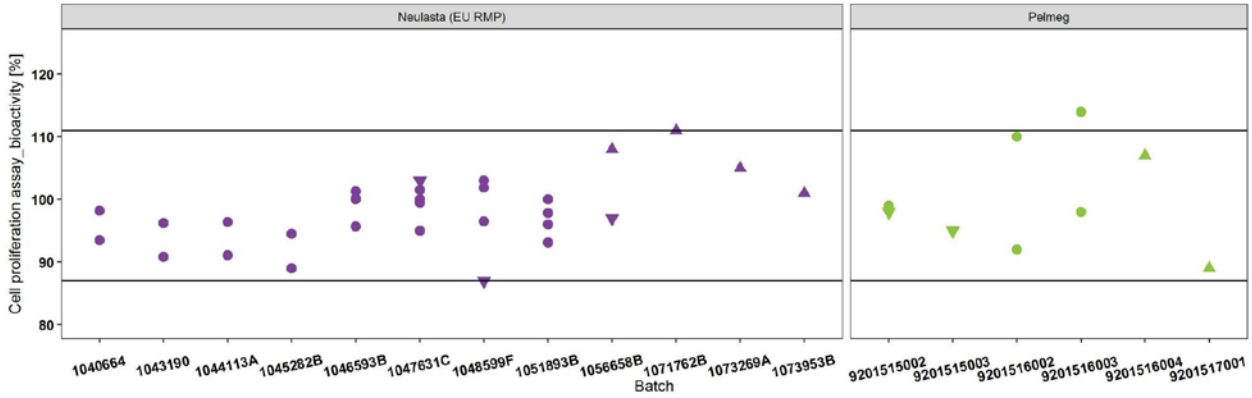
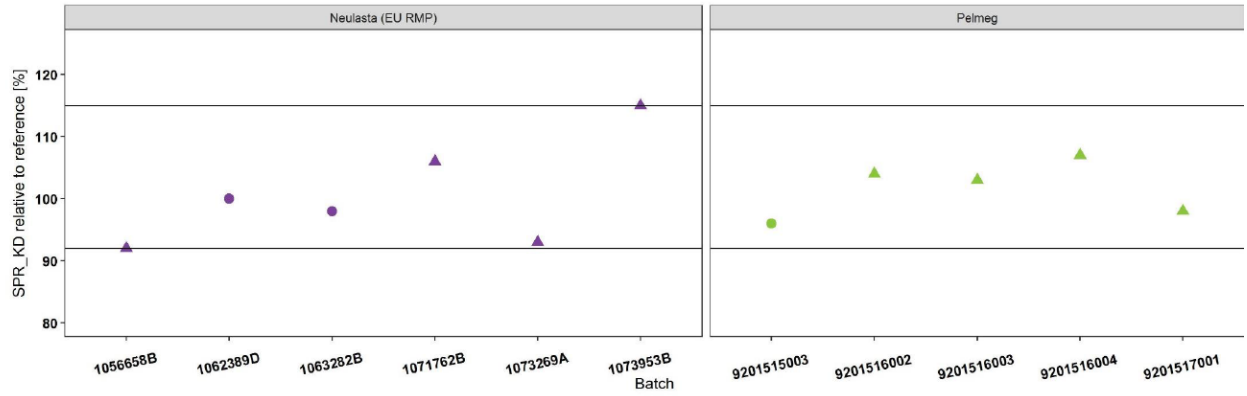


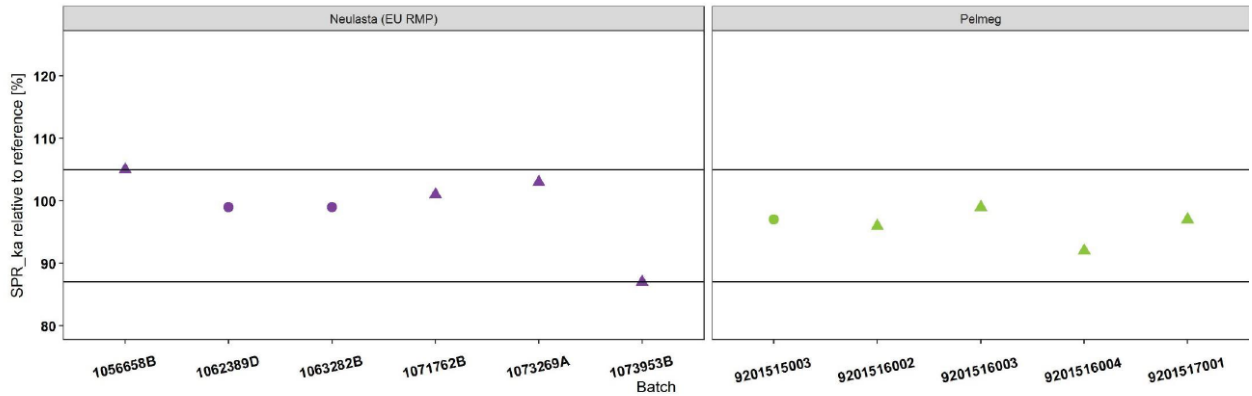
Figure 1: Cell proliferation assay results

2. Receptor binding by SPR:



RMP batches are shown in purple, and Pelmeg batches are shown in green colour. Batches tested in H2H_17 are represented with triangles (▲); Batches tested in H2H_15 are represented with inverted triangles (▼).

Figure 2: Comparison of SPR KD values (relative to reference) for Pelmeg vs reference medicinal product



RMP batches are shown in purple, and Pelmeg batches are shown in green colour. Batches tested in H2H_17 are represented with triangles (▲); Batches tested in H2H_15 are represented with inverted triangles (▼).

Figure 3: Comparison of SPR ka vales (relative to reference) for Pelmeg and ref. medicinal product batches

3. *In vivo* study in rats

Neutropenia was chemotherapy-induced in animals from groups 6 to 10 with a single intraperitoneal dose of 50 mg cyclophosphamide (CPA)/kg b.w. one day before actual dosing with the test, reference or control item (24-hour gap between CPA injection and pegylated GCSF/vehicle injection). The results

of the pharmacodynamic analysis of the neutrophilic granulocytes responses between Pelmeg and Neulasta are given in Table 5 below.

Table 5: Pharmacodynamic analysis of neutrophils in normal and neutropenic rats following Pelmeg or Neulasta injections

Group	CPA dose on test day -1	Dose [μg/kg]	E_{max} [cells x 10^9/ L blood]	t_{max} [h]	$AUEC_{0\text{-}t\ last}$ [h*cells x 10^9/ L blood]	$AUEC_{eff\ 0\text{-}t\ last}$ [h*cells x 10^9/ L blood]#
1	None	0 (Vehicle)	3.10	72	512.62	N/A
2	None	15 (B12019)	15.34	24	1360.81	848.19
3	None	100 (B12019)	23.71	48	2119.72	1607.10
4	None	15 (Neulasta®)	14.94	24	1199.28	686.66
5	None	100 (Neulasta®)	22.09	24	1958.25	1445.63
6	CPA	0 (Vehicle)	2.70	216	226.15	N/A
7	CPA	15 (B12019)	3.60	16	426.99	200.84
8	CPA	100 (B12019)	4.32	12	489.51	263.36
9	CPA	15 (Neulasta®)	4.15	16	464.99	238.84
10	CPA	100 (Neulasta®)	4.30	16	547.49	321.34

#: effective AUEC of the dose level groups 2 to 5: $AUEC_{Gr.\ 2\ to\ 5} - AUEC_{Gr.1}$
effective AUEC of the dose level groups 7 to 10: $AUEC_{Gr.\ 7\ to\ 10} - AUEC_{Gr.6}$
N/A:not applicable

Table 6: Ratios of the pharmacodynamics parameters in healthy and neutropenic rats

Dose (μg/kg)	Healthy rats Pelmeg/Neulasta		Neutropenic rats Neulasta/Pelmeg	
	$AUECeff_{(0\text{-}Tlast)}$	Emax	$AUECeff_{(0\text{-}Tlast)}$	Emax
15	1.24	1.03	1.19	1.15
100	1.11	1.07	1.22	1.00

Secondary pharmacodynamic studies

The applicant did not submit secondary PD studies (see non-clinical discussion). A summary of information from the literature is provided that G-CSF could affect the growth and activity of certain types of non-haematopoietic cells *in vitro*.

Safety pharmacology programme

The applicant did not submit safety pharmacology studies (see non-clinical discussion).

Pharmacodynamic drug interactions

The applicant did not submit pharmacodynamic drug interaction studies (see non-clinical discussion).

2.3.3. Pharmacokinetics

The GLP PK/PD study performed in normal/neutropenic rats consisted of twelve animals/group sampled for G-CSF dosage at the following time points: 0, 4, 8, 12, 16, 24, 36, 48, 72, 96, 120, 144, 168, 192, 216 and 240 hrs after a single SC injection of 0 (vehicle control), 15 or 100 µg/kg Pelmeg or Neulasta.

Table 5: Pharmacokinetic results in healthy and neutropenic rats

Dosage [µg/kg]	Non-compartment analysis of Pegfilgrastim							
	C_{max}#1 [pg/m]	t_{max}#1 [h]	$t_{1/2}$ [h]	K_{el} [1/h]	$AUC_{0\text{-}t\ last}$ [ng·h/m]	$AUC_{0\text{-}inf}$ [ng·h/m]	$AUC_{0\text{-}inf}$/ dose [h·kg·ng/ mL/µg]	DPF
Healthy animals								
15 µg Pelmeg/kg	5104	16	11.53	0.0601	112.3	113.5	7.6	N/A
100 µg Pelmeg/kg	104851	12	11.74	0.0590	2435.9	2437.5	24.4	3.3
15 µg Neulasta/kg	5593	12	11.50	0.0603	102.0	103.4	6.9	N/A
100 µg Neulasta/kg	135130	8	12.30	0.0564	2856.3	2857.9	28.6	4.2
Neutropenic animals								
15 µg Pelmeg/kg	6326	16	10.50	0.0660	177.9	178.7	11.9	N/A
100 µg Pelmeg/kg	178313	24	7.00	0.099	6959.8	6960.3	69.6	5.9
15 µg Neulasta/kg	5609	12	14.41	0.0481	161.4	169.0	11.3	N/A
100 µg Neulasta/kg	164956	16	8.62	0.0804	6423.2	6423.0	64.2	6.0

#1: Values obtained from serum analysis of pegfilgrastim, all other values calculated by pharmacokinetic analysis
DPF: Dose proportion factor
[AUC0-t last (high dose)/AUC0-t last (low dose)]/[(high dose)/ (low dose)]

Table 6: Comparison of exposures between rats given 100 µg/kg and healthy volunteers following a single administration of 6 mg in study B12019-101

Exposure in normal rats (Pelmeg)		Exposure in Neutropenic rats (Pelmeg)		Exposure in HV (Study B012019-101) Pelmeg	
$AUC_{0\text{-last}}$ (h*ng/mL)	2435.9	$AUC_{0\text{-last}}$ (h*ng/mL)	6959.8	$AUC_{0\text{-last}}$ (h*ng/mL)	5030.3
C_{max} (ng/mL)	104.851	C_{max} (ng/mL)	178.313	C_{max} (ng/mL)	137.01
$T_{1/2}$ (h)	11.74	$T_{1/2}$ (h)	7.00	$T_{1/2}$ (h)	39.09
Exposure in Normal rats (Neulasta)		**Exposure in Neutropenic rats (Neulasta)**		**Exposure in HV (Study B012019-101) Neulasta**	
$AUC_{0\text{-last}}$ (h*ng/mL)	2856.3	$AUC_{0\text{-last}}$ (h*ng/mL)	6423.2	$AUC_{0\text{-last}}$ (h*ng/mL)	5435.1
C_{max} (ng/mL)	135.130	C_{max} (ng/mL)	164.956	C_{max} (ng/mL)	152.05
$T_{1/2}$ (h)	12.30	$T_{1/2}$ (h)	8.62	$T_{1/2}$ (h)	40.21

2.3.4. Toxicology

Single dose toxicity

The applicant did not submit single dose toxicity studies (see non-clinical discussion).

Repeat dose toxicity

The applicant did not submit repeat drug toxicity studies (see non-clinical discussion).

Genotoxicity

The applicant did not submit genotoxicity studies (see non-clinical discussion).

Carcinogenicity

The applicant did not submit carcinogencity studies (see non-clinical discussion).

Reproduction Toxicity

The applicant did not submit reproduction toxicity studies (see non-clinical discussion).

Toxicokinetic data

The applicant did not submit toxicokinetic studies (see non-clinical discussion).

Local Tolerance

The applicant did not submit pharmacodynamic drug interaction studies (see non-clinical discussion).

Other toxicity studies

The applicant did not submit other toxicity studies (see non-clinical discussion).

2.3.5. Ecotoxicity/environmental risk assessment

Based on the CHMP Guideline on the environmental risk assessment of medicinal products for human use (CHMP/SWP/4447/00 corr. 2) which states that proteins are exempted from the need to submit studies because they are unlikely to result in a significant risk to the environment due to their nature, the applicant submitted a justification for not submitting an environmental risk assessment.

Pegfilgrastim is already used in existing marketed products and no significant increase in environmental exposure is anticipated. The PEG moiety of Pelmeg is unlikely to result in a significant risk to the environment because of metabolic breakdown before excretion in patients[13,14] and a rapid biodegradation in the environment[15, 16].

2.3.6. Discussion on non-clinical aspects

The potency of Pelmeg was measured on 6 Pelmeg batches *versus* 12 batches of Neulasta by *in vitro* cell-based models (M-NFS-60 cells, a murine myeloblastic cell line) and the results show that their potency is similar.

The binding affinity to the G-CSF receptor of either Pelmeg and Neulasta was also investigated using a validated Surface Plasmon Resonance (SPR). The binding affinities (Kd values) for Pelmeg were found similar to those measured for the reference Neulasta.

Comparability on the non-clinical level was adequately addressed according to the current regulatory requirements. Thus, the focus of the demonstration of biosimilarity was on *in vitro* biofunctional assays which revealed biosimilarity between Pelmeg and the reference medicinal product Neulasta.

A GLP-compliant *in vivo* PK/PD study was additionally performed in normal and neutropenic male CD/Crl:CD(SD) rats with the aim to compare Pelmeg versus reference. Both Pelmeg and Neulasta were compared in terms of absolute neutrophil counts (ANC). Results clearly demonstrate the intended pharmacodynamic effect. However, due to limitations of the neutropenic animal model and the fact that the study was considered descriptive only, a conclusion on statistically-confirmed in vivo biosimilarity could not be drawn. Quality and clinical data are considered to be more relevant in this respect. Therefore, the in vivo data is acceptable and no further discussion is needed.

Safety pharmacology, genotoxicity, carcinogenicity, single/repeat-dose toxicity studies, reproductive and developmental toxicity studies were not submitted and are not required as per the latest European biosimilar guidelines (i.e. Guidelines CHMP/437/04 Rev. 1, EMEA/CHMP/BWP/247713/2012, EMEA/CHMP/BMWP/42832/2005 Rev. 1 and the annex to Guideline on similar biological medicinal products containing biotechnology-derived proteins as active substance, non-clinical and clinical issues; guidance on similar medicinal products containing recombinant granulocyte-colony stimulating factor, EMEA/CHMP/BMWP/31329/2005).

The justification for not submitting environmental risk assessment studies is acceptable. It is unlikely that residues of pegfilgrastim would persist in the environment or cause inadvertent environmental effects. The approval of Pelmeg is not expected to cause increases in environmental exposure above existing levels for this active substance or result in any additional hazards to the environment during

[13] Fruijtier-Pölloth C. Safety assessment on polyethylene glycols (PEGs) and their derivatives as used in cosmetic products. Toxicology. 2005 Oct 15;214(1-2):1-38
[14] Webster R, Didier E, Harris P, Siegel N, Stadler J, Tilbury L, Smith D. PEGylated proteins: evaluation of their safety in the absence of definitive metabolism studies. Drug Metab Dispos. 2007 Jan;35(1):9-16
[15] Bernhard M, Eubeler JP, Zok S, Knepper TP. Aerobic biodegradation of polyethylene glycols of different molecular weights in wastewater and seawater. Water Res. 2008 Dec;42(19):4791-801
[16] Huang M, Wu W, Qian J, Wan DJ, Wei XL, Zhu JH. Body distribution and in situ evading of phagocytic uptake by macrophages of long-circulating poly (ethylene glycol) cyanoacrylate-co-n-hexadecyl cyanoacrylate nanoparticles. Acta Pharmacol Sin. 2005 Dec;26(12):1512-8

storage, distribution, use and disposal. Considering the expected exposure and the nature of the product, the absence of formal environmental risk assessment studies for Pelmeg is considered justified. This is in accordance with the CHMP Guideline on the environmental risk assessment of medicinal products for human use (EMEA/CHMP/SWP/4447/00 corr 2).

2.3.7. Conclusion on the non-clinical aspects

The non-clinical aspects of pharmacology, pharmacokinetic and toxicology for Pelmeg have been well characterised and are considered acceptable. There were no further changes to the SmPC and the product information is aligned with the reference product Neulasta.

2.4. Clinical aspects

2.4.1. Introduction

Pelmeg (B12019) was developed as a similar biological medicinal product to Neulasta (pegfilgrastim). The proposed indication for Pelmeg is identical to the approved indication for Neulasta. The recommended dose is 6 mg pegfilgrastim to be administered subcutaneously (SC).

The clinical programme of Pelmeg was comprised of two studies in healthy volunteers. Clinical comparability of Pelmeg and Neulasta was evaluated in the pivotal PK/PD study B12019-101 using the approved 6 mg dose. The supportive study B12019-102 mainly monitored immunogenicity, but also assessed PD comparability of Pelmeg and Neulasta, using a non-therapeutic, lower dose of 3 mg. The formulation used in the clinical pharmacology studies is the same as the proposed commercial formulation.

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 in the Community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

- Tabular overview of clinical studies

Study	Design	Study population	Dose and Regimen	Objectives/ Endpoints
B12019-101 PK/PD study (pivotal)	Single-dose, randomised, double-blind, two-stage, two-way cross-over study	172 healthy subjects	Single-dose *s.c.* Pelmeg and Neulasta (6 mg) Sequence 1: Pelmeg-Neulasta Sequence 2: Neulasta-Pelmeg Interval between each injection: 6 weeks	Co-primary: PK comparability based on $AUC_{0\text{-last}}$ and C_{max}, PD comparability based on $AUEC_{0\text{-last}}$ for ANC Secondary: Additional PK and PD endpoints, immunogenicity, safety
B12019-102 PD and	Multiple-dose, randomised, double-blind,	96 healthy subjects	Multiple-dose *s.c.* Pelmeg and Neulasta (3 mg)	Co-primary: PD comparability based on $AUEC_{0\text{-last}}$

immuno-genicity/ safety study (supportive)	three periods, two sequences, cross-over study		Sequence 1: Pelmeg-Pelmeg-Neulasta Sequence 2: Neulasta-Neulasta-Pelmeg Interval between each injection: 6 weeks.	for ANC, and immunogenicity/ safety <u>Secondary</u>: PK, additional PD and immunogenicity endpoints

2.4.2. Pharmacokinetics

Absorption

Pegfilgrastim concentrations in serum were determined using a quantitative enzyme linked immunosorbent assay (ELISA) technique. The Human G-CSF DuoSet ELISA kit (R&D Systems) was modified to determine Pegfilgrastim in human serum and was validated for this purpose over a calibration range of 0.20 – 8.00 ng/mL at Celerion Switzerland AG.

<u>Study B12019-101</u>

This was a single-dose, randomised, double-blind, two-stage, two-way cross-over, PK/PD study of Pelmeg versus Neulasta in healthy volunteers. Study drugs were administered as s.c. injections into the abdomen, at a dose of 6 mg. As recommended in different scientific advices, a wash-out phase of 6 weeks between treatments was applied to avoid any potential for carry-over effects.

In order to account for the expected high variability of the relevant PK parameters (AUC and Cmax), the study methodology was based on a two-stage design, planning for a sample size re-calculation after completion of stage 1 and potential sample size adjustment for stage 2. The assumptions used to calculate a sample size of 172 subjects for stage 1 are considered appropriate (i.e. power of 90%, intra-subject CV of maximum 50% for PK parameters, drop out of 10%). According to the predefined decision rules, no stage 2 was performed.

Blood samples for the determination of pegfilgrastim concentrations were collected during the in-patient phase, pre-dose and up to 96 h post-dose (day 5), and during the ambulatory visits in each period (days 6 to 43). The primary PK variables were $AUC_{0\text{-last}}$ and C_{max}, which was agreed upon in scientific advice EMA/CHMP/SAWP/380237/2015. $AUC_{0\text{-inf}}$ and t_{max} were evaluated as secondary PK variables. PK parameters were calculated for all subjects who received a dose of study medication (= PK set).

To demonstrate PK comparability of Pelmeg to the reference product Neulasta, the primary PK parameters $AUC_{0\text{-last}}$ and C_{max} were evaluated using an α1-level of 0.0284. For these parameters the 94.32% (=(1 - 2α)%) CI for the test/reference ratio were to be contained within the standard bioequivalence acceptance interval of 80.00-125.00%. The primary statistical PK analysis was performed on the model-based PK set, defined as all subjects with reliable PK data for both study periods.

Subjects were randomised 1:1 to sequentially receive a 6 mg dose of Pelmeg and Neulasta or vice versa. Of the 172 healthy subjects enrolled and randomised, 171 subjects received study medication (=PK set) and 163 subjects completed both study periods. Two subjects were excluded from the model-based PK set (N= 161) due to missing several consecutive study visits in one period. No major

protocol deviations were reported. All subjects were healthy white males. Mean age of the 171 subjects included in the PK set was 40.9 years (range 19 to 55 years) and mean weight was 81.2 kg (range 61.3 to 99.3 kg).

Results

PK parameters $AUC_{0\text{-last}}$, $_{A}UC_{0\text{-inf}}$ and C_{max} were presented by treatment and showed all a very high inter-subject variability (around 100%). Geometric mean $t_{1/2}$ was about 40 hours and median tmax occurred at 16 hours after both treatments. No positive pre-dose samples were measured, except for one subject in Period 2.

Table 7: Summary of PK parameters of pegfilgrastim (PK set, N= 169)

Parameter	Neulasta® n=164	B12019 n=166
$AUC_{0\text{-last}}$ [h*ng/mL]	5435.1/104.2	5030.3/109.0
$AUC_{0\text{-inf}}$ [h*ng/mL]	5698.3/104.7[a]	5275.5/101.1[b]
C_{max} [ng/mL]	152.05/96.0	137.01/99.9
$t_{1/2}$ [h]	40.208/31.4[a]	39.094/30.8[b]
λ_z [1/h]	0.017239/31.4[a]	0.017730/30.8[b]
t_{max} [h]	16.0 (4.0-36.1)	16.0 (8.0-36.1)

Geometric mean/CV(%) are presented for all values with exception of t_{max}, for which median and range is presented.
[a]n=146, [b] n=148, due to not reliable λ_z in 18 subjects after each treatment

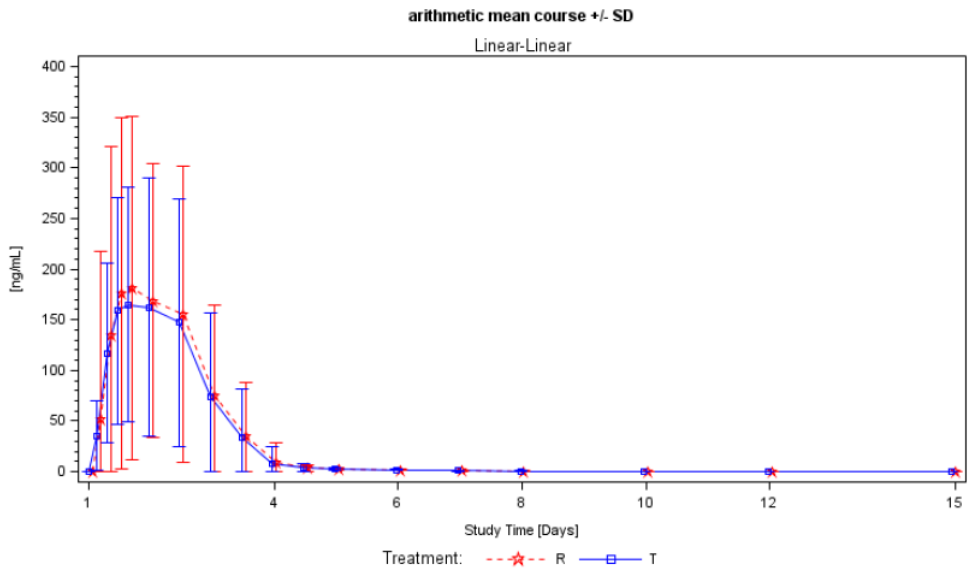


Figure 4: Mean serum concentration time profiles of pegfilgrastim (ng/ml)- until day 15 (Model-based PK set, N=161)

Biosimilarity was claimed as the 94.32% CIs for the ratio of the test and reference product geometric means for the primary PK parameters $AUC_{0\text{-last}}$ and C_{max} were fully contained within the predefined acceptance interval of 80.00-125.00%. Although no statistical analysis of the secondary PK parameter $AUC_{0\text{-inf}}$ was planned per protocol, a post-hoc statistical analysis showed BE criteria were also met for this parameter.

Table 8: Statistical Analysis of Primary PK Parameters in Study B12019-101 (Model-Based PK Set, N=161)

Parameter	Pelmeg/Neulasta		
	Ratio (%)	94.32% CI	Intra-subject CV(%)*
$AUC_{0\text{-last}}$	95.23	86.60;104.73	46.71
C_{max}	92.84	84.36;102.18	47.14

*Intra-individual CV(%) estimated from the residual mean squares.
$AUC_{0\text{-last}}$=area under the concentration time curve from time zero to last measurable concentration, CI=confidence interval, C_{max}=maximum concentration, CSR=Clinical Study Report, CV=coefficient of variation, N=number of subjects, PK=pharmacokinetic.

Table 9: Statistical Analysis of $AUC_{0\text{-inf}}$ in Study B12019-101 (Model-Based PK Set, N=161)**

Pelmeg/Neulasta		
Ratio (%)	94.32% CI	Intra-subject CV(%)*
92.07	82.94; 102.21	45.41

*Intra-individual CV(%) estimated from the residual mean squares.
$AUC_{0\text{-inf}}$=area under the concentration time curve from time zero to infinity, CI=confidence interval, CSR=Clinical Study Report, CV=coefficient of variation, N=number of subjects, PK=pharmacokinetic.
Only subjects with data available were included in the statistical analysis.

**Only subjects of the model-based PK set with data available were included in the statistical analysis (N=146)

A re-evaluation of the protein content data of the test and reference batches used in this study was performed based on an optimized UV method. As a result, a difference in protein content of more than 5% was determined between both batches with the Pelmeg batch having a lower concentration than the Neulasta batch (9.57 mg/ml versus 10.20 mg/ml). The test/reference ratio for both AUC0-last and Cmax (95.23% and 92.84%, respectively) was below 100% and is therefore in line with a lower concentration of the Pelmeg batch. In addition, the 94.32% CIs of the test/reference ratio for both parameters (not corrected for protein content) were fully contained within the acceptance interval of 80.00-125.00%. A post-hoc sensitivity analyses was performed correcting for the difference in protein content.

Other sensitivity analyses that were conducted and included analysis in the subgroup of ADA-negative subjects and an analysis that excluded two outlier subjects. Results for these supportive sensitivity analyses were similar to the results of the overall analyses (see table below).

Table 10: Sensitivity Analyses in Study B12019-101 (Model-Based PK Set)

		Pelmeg/Neulasta		
	Parameter	**Ratio (%)**	**94.32% CI**	**Intra-subject CV(%)***
Protein content corrected (N=161)	$AUC_{0\text{-last}}$ C_{max}	101.47 98.92	92.27;111.58 89.88; 108.87	46.71 47.14
ADA-negative subjects only (N=128)	$AUC_{0\text{-last}}$ C_{max}	95.39 94.62	85.44;106.49 84.85;105.52	48.31 47.76
Excluding subjects 118 and 138 (N=159)	$AUC_{0\text{-last}}$ C_{max}	98.67 96.26	90.92;107.09 88.64;104.54	39.45 39.80
Excluding Subjects 118 and 138 and protein content corrected (N=159)	$AUC_{0\text{-last}}$ C_{max}	105.13 102.56	96.87;114.10 94.44;111.39	39.45 39.80

*Intra-individual CV(%) estimated from the residual mean squares.
ADA=anti-drug antibody, $AUC_{0\text{-last}}$=area under the curve from time zero to last measurable concentration, CI=confidence interval, C_{max}=maximum concentration, CSR=Clinical Study Report, CV=coefficient of variation, N=number of subjects, PK=pharmacokinetic.

Of note, results for the PK parameters AUC0-t and Cmax showed a statistically significant period effect.

Study B12019-102

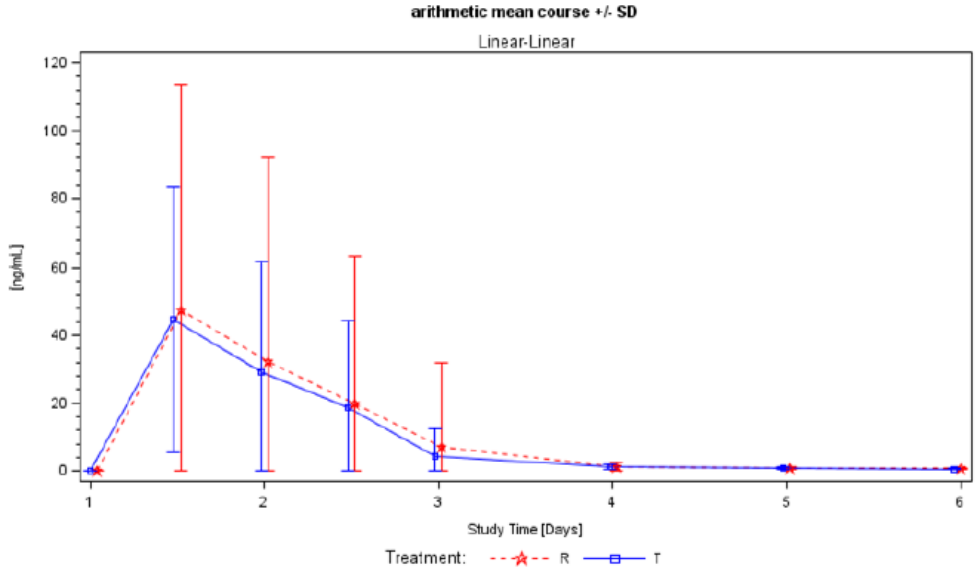
This was a randomised, multiple dose, three-period, two-sequence, cross-over study to mainly assess the immunogenicity and PD comparability of 3 mg Pelmeg (T) and Neulasta (R) in healthy male volunteers. Subjects were randomised (1:1) to the treatment sequence T-T-R or R-R-T. Both treatments were administered as s.c. injection. Dosing was separated by a wash-out period of 6 weeks as was also done for study B12019-101.

Blood sampling was sparse (7 PK samples in each period) since no statistical comparison of PK parameters was aimed for. PK concentrations and PK parameters ($AUC_{0\text{-}120h}$, C_{max} and t_{max}) were evaluated purely descriptively.

Overall, 96 healthy male subjects were randomised and received at least one dose of study medication. Therefore, all subjects were included in the PK set. Mean age of the 96 subjects was 41.4 years (range 21 to 55 years) and mean weight was 81.8 kg (range 62.4 to 99.5 kg). 1 subject was Asian, 5 subjects were black, and the majority was white.

Results

Based on sparse PK sampling, serum concentration time profiles were similar for Pelmeg and Neulasta, with serum concentrations peaking at 12 hours post-dose in most subjects. Variability for the PK parameters was very high with geometric CVs above 100% for AUC and Cmax.



R = reference treatment (Neulasta®), T = test treatment (B12019)

Figure 5: Mean serum concentrations of pegfilgrastim until Day 6 (PK set, N=96)

Table 11: Summary of the PK parameters of pegfilgrastim (PK set, N=96)

Parameter		Neulasta®	B12019
$AUC_{0\text{-}120h}$ [h*ng/mL]	n=133	821.78 / 124.3	847.20 / 116.1
C_{max} [ng/mL]	n=137	29.498 / 143.2	29.880 / 133.3
t_{max} [h]	n=137	12.0000 (11.983-48.050)	12.0000 (11.967-36.050)

Geometric mean/CV (%) are presented for all values with exception of t_{max}, for which median and range is presented.
n = number of subjects' periods contributing to the calculation of the respective descriptive statistics
$AUC_{0\text{-}120h}$ was replaced by AUC_{all} in case the sample was taken earlier than 120h (refer to Section 10.2).

Differences between periods were high, mean peak values ranged between 34.6 and 60.8 ng/mL after Neulasta and between 34.2 and 59.4 ng/mL after Pelmeg. Geometric mean $AUC_{0\text{-}120h}$ ranged between 698 and 934 h*ng/ml after Neulasta and between 687 and 1107 h*ng/mL after Pelmeg.

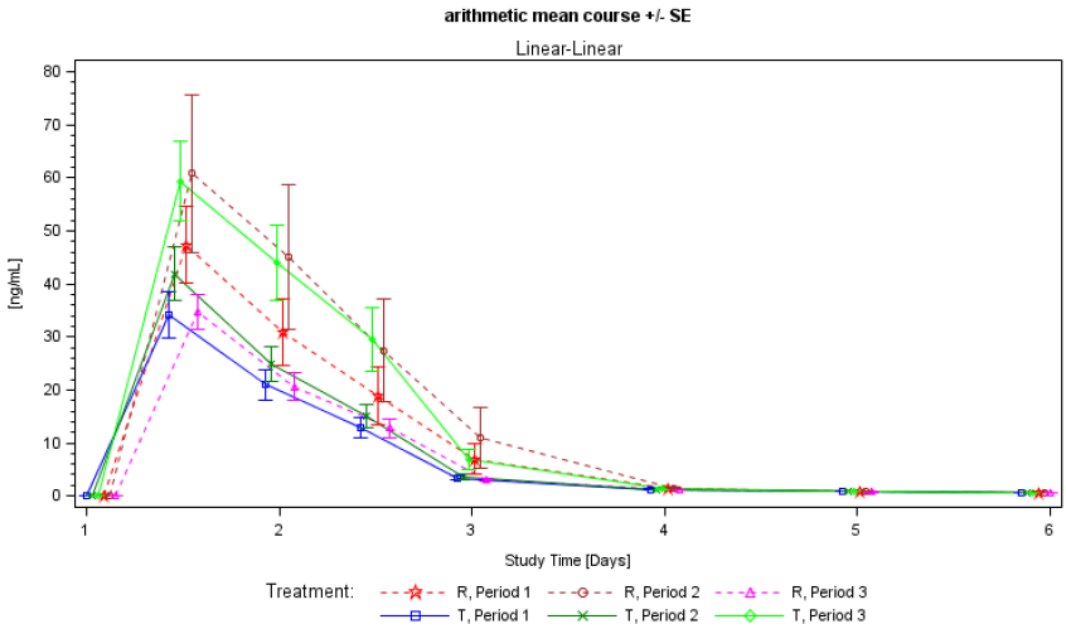


Figure 6: Mean serum concentration time profiles of pegfilgrastim (ng/ml) by treatment and period (PK set, N=96)

As a result of a systematic handling error when the syringes for the first group of 28 subjects (period 1) were prepared, the doses administered to these subjects were between 2 and 2.8 mg pegfilgrastim instead of the per-protocol dose of 3 mg. Therefore, a sensitivity analysis was conducted excluding cohort 1, and period 1 data from cohort 1. This resulted in a slightly higher exposure than in the overall analysis, which is in line with the exclusion of data from underdosed subjects in Period 1.

PK results from pooled analyses

When comparing the PK results of the two studies (one conducted with 6mg and one with 3mg), it is apparent that a dose effect can be seen. Cmax values increased with increasing pegfilgrastim dose. Increases appear to be greater than dose-proportional, which is in line with literature data on the PK of pegfilgrastim[17].

Distribution

The applicant did not submit studies on distribution (see pharmacology discussion).

Elimination

The applicant did not submit studies on elimination (see pharmacology discussion).

Dose proportionality and time dependencies

The applicant did not submit studies on dose proportionality and time dependencies (see pharmacology discussion).

Special populations

The applicant did not submit studies on special populations (see pharmacology discussion).

[17] Roskos LK, Lum P, Lockbaum P, Schwab G, Yang BB. Pharmacokinetic/pharmacodynamic modeling of pegfilgrastim in healthy subjects. J Clin Pharmacol. 2006 Jul;46(7):747-57

Pharmacokinetic interaction studies

The applicant did not submit pharmacokinetic interaction studies (see pharmacology discussion).

Pharmacokinetics using human biomaterials

The applicant did not submit studies on pharmacokinetics using human biomaterials (see pharmacology discussion).

2.4.3. Pharmacodynamics

Mechanism of action

The applicant did not submit studies on the mechanism of action (see pharmacology discussion).

Primary and Secondary pharmacology

Study B12019-101

The following PD parameters were derived from the ANCs:

Primary parameter

$AUEC_{0\text{-}last}$ area under the ANC vs. time curve from dosing to last scheduled sample, by linear trapezoidal rule.

Secondary parameters

- Emax maximum effect level (ANC)
- t_{max}, E time of the maximum effect level
- CD34+ cell counts

The primary statistical analysis is based on the model-based PD set (n=161, subjects who completed both treatment periods without important protocol deviation), PD set included 167 subjects.

Results

The secondary ANC parameters and CD34+ cell counts were evaluated descriptively between treatments.

Table 12: Summary of the PD Parameters of ANC (PD Set, N=167)

Parameter	Neulasta® n=162	B12019 n=166
$AUEC_{0\text{-}last}$ [h*G/L]	7110.5/21.5	7128.1/21.9
E_{max} [G/L]	35.238/24.3	34.594/23.9
t_{max} [h]	60.0 (24-96)	60.0 (24-96)

Geometric mean AUEC0-last and Emax were similar after administration of Neulasta and B12019 (AUEC0-last of 7110 and 7128 h*G/L and Emax of 35.2 and 34.6 G/L, respectively). Variability was moderate with geometric CVs of less than 25%. The geometric mean ratio of AUEC0-last was about 100% and the corresponding 95% confidence interval was very close to 100% (98.67%; 101.75%).

Table 13: Statistical Analysis of Primary PD Parameter $AUEC_{0\text{-}last}$ of ANC in Study B12019-101 (Model-Based PD Set, N=161)

Pelmeg / Neulasta		
Ratio (%)	**95% CI**	**Intra-subject CV(%)***
100.20	98.67;101.75	6.99

*Intra-individual CV(%) estimated from the residual mean squares.

ANC=absolute neutrophil count, $AUEC_{0\text{-last}}$=area under the effect time curve from time zero to last measurable concentration, CI=confidence interval, CV=coefficient of variation, CSR=Clinical Study Report, N=number of subjects, PD=pharmacodynamic.

When evaluating ANC time curves, after similar pre-dose values (3.315 and 3.214 G/L) a similar increase in mean ANC was observed after dosing of Neulasta and B12019: at 36 hours after dosing mean ANC were 32.435 G/L and 31.971 G/L, respectively. ANC values remained on this level until 84 hours after dosing (30.484 G/L and 29.551 G/L, respectively) and decreased thereafter. Pre-dose level was reached again on Day 18 (3.413 G/L and 3.549 G/L, respectively).

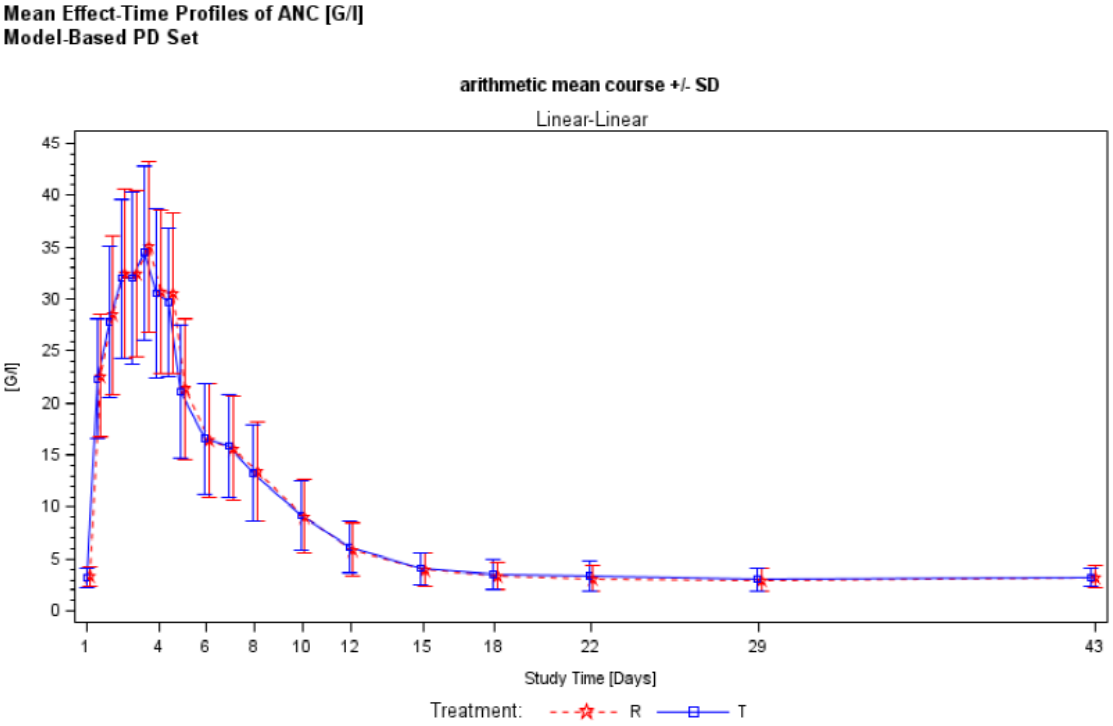


Figure 7: Mean ANC values in Study B12019-101 (Model-based PD set)

In addition, CD34+ counts were analysed as secondary PD endpoint in this study. Similar increases in CD34+ cells were seen after administration of Pelmeg and Neulasta. Values peaked at around 5 days post-dose, and reached pre-dose levels on Day 12 post-dose. Overall, CD34+ profiles were very similar for Pelmeg and Neulasta. In addition, a post-hoc analysis of $AUEC_{0\text{-last}}$ of CD34+ was conducted, using the same model as for the primary PD analysis. In this analysis, the geometric mean ratio (Test/Reference) was 98.46, and the corresponding CI was 93.99; 103.14.

Study B12019-102

A total of 96 healthy subjects were enrolled and treated with study medication. The primary analysis of PD was based on 82 subjects (model-based PD set). The model-based statistical comparisons between treatments only made use of those subjects who completed all treatment periods without any important protocol deviation.

Results

All subjects in this study were male, and the majority (95%) was white. In the primary PD analysis set, median age was 44 years, median weight was 80.3 kg and median BMI was 25.3 kg/m^2. The majority of subjects (84%) were non-smokers.

Table 14: Summary of the PD parameters of ANC (Model based PD Set, N=82)

Parameter	Neulasta	B12019
	n=82	n=82
AUEC0-last [h*G/L]	6170.8 / 21.9	6264.3 / 23.2
Emax [G/L]	28407 / 25.9	28597 / 23.6
tmax [h]	36	36

To demonstrate PD comparability of B12019 versus Neulasta, the primary PD parameter $AUEC_{0\text{-last}}$ was calculated. B12019 and Neulasta were assumed to be comparable with regard to PD if the 95% confidence interval of the test/reference ratio lies within the acceptance range of 80.00-125.00%.

Table 15: Statistical Analysis of Primary PD Parameter $AUEC_{0\text{-last}}$ of ANC in Study B12019-102 (Model-Based PD Set, N=82)

Pelmeg/Neulasta		
Ratio (%)	95% CI	Intra-subject CV(%)*
101.59	99.58; 103.63	7. 49

*Intra-individual CV(%) estimated from the residual mean squares.

ANC=absolute neutrophil count, $AUEC_{0\text{-last}}$=area under the effect time curve from time zero to last measurable concentration, CI=confidence interval, CV=coefficient of variation, CSR=Clinical Study Report, N=number of subjects, PD=pharmacodynamic. Source: CSR B12019-102, Table 14.2.5.1.

The geometric mean ratio of AUEC0-last was 101,59% with the corresponding 95% confidence interval of 99,58% and 103,63%. Emax was 28,4 G/L and median tmax of ANC was 36 hours after both treatments (PD set). Intra-subject CV was low, with 7.49%.

The impact of underdosing Cohort 1 in Period 1 was evaluated in sensitivity analyses, excluding Cohort 1, Period 1, and Period 1 data from Cohort 1, respectively. In all analyses, the geometric mean ratio was around 100% in all sensitivity analyses, and the corresponding 95% CIs were not only fully contained in the 80.00-125.00% interval, but also in the tighter interval of 90.00-111.00%.

When evaluating ANC time curves, similar profiles were seen for Pelmeg and Neulasta. Starting from similar pre-dose levels (around 3 G/L), comparable increases in mean ANC were observed after administration of Pelmeg and Neulasta. Peak levels were reached at around 36 hours post-dose, and decreased thereafter. Pre-dose level was reached again on Day 22.

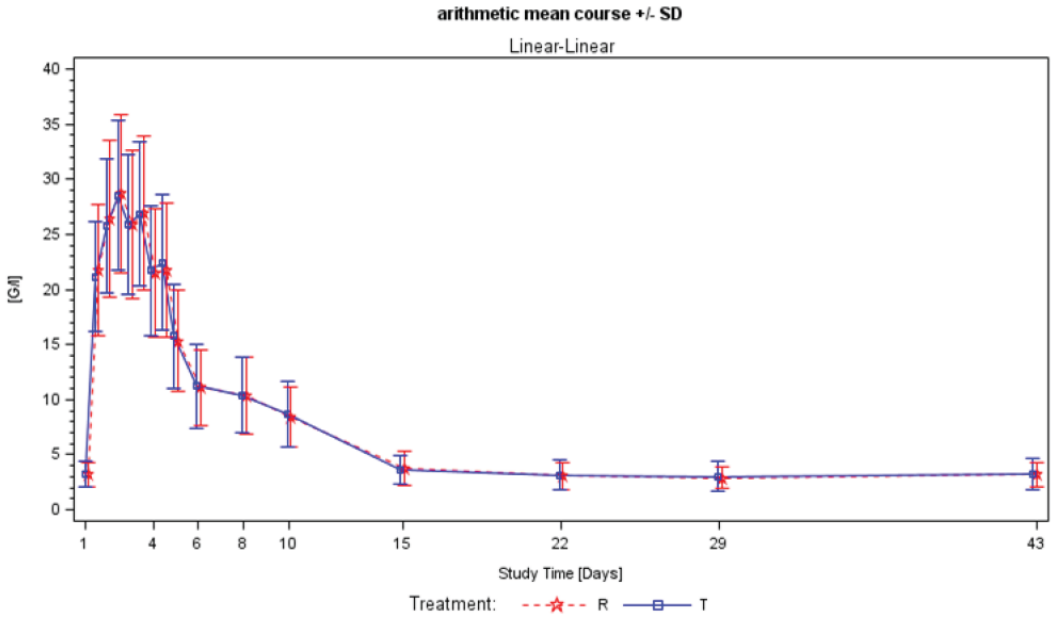


Figure 8: Mean ANC values in Study B12019-102 (PD set, N=93)

R = reference treatment (Neulasta), T = test treatment (B12019) Source: Figure 14.2.3.1.1

Results of ANC parameter per treatment and period:

Table 16: Summary data on pharmacodynamic parameters of ANC values by treatment and period

Model-Based PD Set

Treatment T: B12019
Treatment R: Neulasta®

		Treatment					
		R			T		
Parameter [Unit]	Statistics	Period 1 (N=41)	Period 2 (N=41)	Period 3 (N=41)	Period 1 (N=41)	Period 2 (N=41)	Period 3 (N=41)
AUEC0-last [h*G/L]	n	41	41	41	41	41	41
	Mean	6126.4	6362.0	6456.6	6216.2	6441.9	6619.3
	SD	1248.46	1401.42	1455.59	1384.43	1385.23	1543.08
	CV%	20.4	22.0	22.5	22.3	21.5	23.3
	Minimum	3625	3956	3817	3621	3696	3959
	Median	6148.5	6391.7	6008.2	6184.1	6207.8	6644.0
	Maximum	9240	9868	9745	8790	9591	10455
	Geom. n	41	41	41	41	41	41
	Geom. Mean	6001.6	6215.0	6299.7	6059.2	6296.4	6443.4
	Geom. CV%	20.9	22.2	22.8	23.6	22.0	24.0
Emax [G/L]	n	41	41	41	41	41	41
	Mean	27.115	29.237	31.624	27.076	30.453	30.518
	SD	7.13875	7.80194	6.80801	5.84148	6.45049	6.95990
	CV%	26.3	26.7	21.5	21.6	21.2	22.8
	Minimum	16.14	17.40	18.95	14.13	17.45	16.26
	Median	25.010	28.700	31.470	27.230	29.530	29.430
	Maximum	43.99	53.43	48.88	37.66	47.24	47.79
	Geom. n	41	41	41	41	41	41
	Geom. Mean	26.235	28.265	30.913	26.402	29.784	29.740
	Geom. CV%	26.4	26.8	22.0	23.8	21.8	23.6

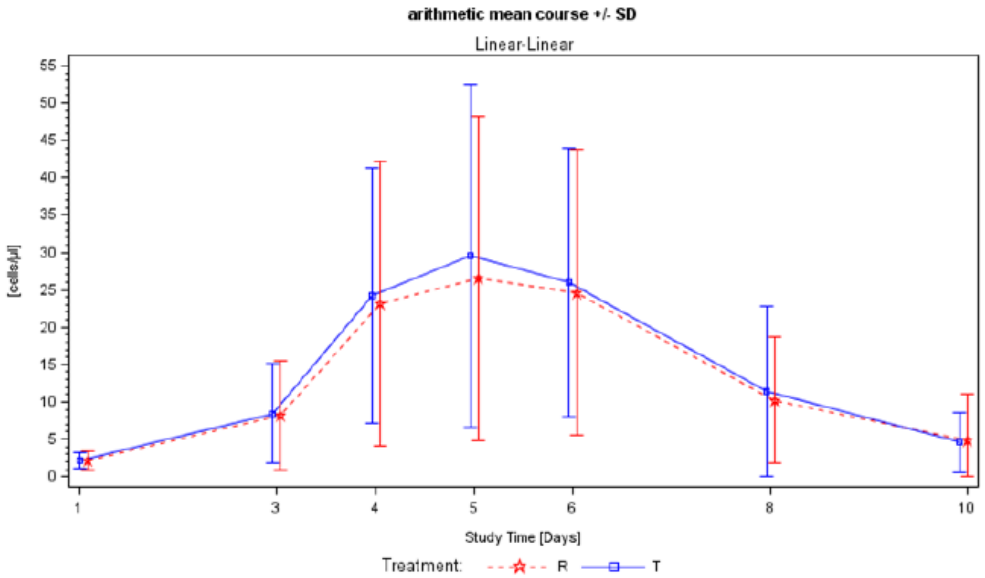
Table 17: Summary data on pharmacodynamic parameters of ANC values by treatment and period - excluding cohort 1 (subjects 1-28)

Treatment T: B12019
Treatment R: Neulasta®

Parameter [Unit]	Statistics	Treatment R Period 1 (N=28)	R Period 2 (N=28)	R Period 3 (N=28)	T Period 1 (N=28)	T Period 2 (N=28)	T Period 3 (N=28)
AUEC0-last [h*G/L]	n	28	28	28	28	28	28
	Mean	6258.0	6337.9	6474.1	6544.7	6596.5	6485.6
	SD	1169.96	1421.41	1263.42	1249.62	1150.37	1395.83
	CV%	18.7	22.4	19.5	19.1	17.4	21.5
	Minimum	4441	3956	4326	3935	4599	3959
	Median	6192.5	6389.7	6134.0	6502.4	6294.4	6611.2
	Maximum	9240	9868	8646	8790	8967	9330
	Geom. n	28	28	28	28	28	28
	Geom. Mean	6156.2	6190.8	6356.7	6426.9	6500.3	6335.3
	Geom. CV%	18.5	22.3	19.7	19.8	17.6	22.7
Emax [G/L]	n	28	28	28	28	28	28
	Mean	27.015	27.456	32.291	28.863	31.545	28.520
	SD	7.20333	6.59734	6.50184	4.81572	5.90231	5.74901
	CV%	26.7	24.0	20.1	16.7	18.7	20.2
	Minimum	16.83	17.40	21.40	19.71	19.96	16.26
	Median	24.990	27.745	32.435	28.685	30.245	28.105
	Maximum	43.99	44.86	48.88	37.66	47.24	40.97
	Geom. n	28	28	28	28	28	28
	Geom. Mean	26.142	26.694	31.685	28.470	31.024	27.935
	Geom. CV%	26.4	24.7	19.9	17.1	18.8	21.4

A slight period effect was observed with both Test and Reference product in the model-based PD set. ANC $AUEC_{0\text{-last}}$ increased 6.3% with Test and 4.97% with Reference when comparing period 3 with period 1. The applicant presented a table of ANC values by treatment and period excluding cohort 1 from all periods: only a slight increase in GMR of ANC $AUEC_{0\text{-t}}$ was observed from period 1 to period 3 with the reference product, whereas with the test product, no increase in ANC was seen (-1.4%).

In addition, CD34+ counts were analysed as secondary PD endpoint in this study.



R = reference treatment (Neulasta®), T = test treatment (B12019)

Figure 9: Mean CD34+ values in Study B12019-102 (PD set, N=93)

Similar increases in CD34+ cells were seen after administration of Pelmeg and Neulasta. Values peaked at around 5 days post-dose, and nearly reached pre-dose levels at the last sampling point on Day 10 post-dose. Overall, CD34+ profiles were very similar for Pelmeg and Neulasta. In addition to the pre-specified analysis, Emax of ANC and AUEC of CD34+ were analysed as secondary PD endpoints, using the Analysis of Variance model as for the primary PD analysis. In both analyses, geometric mean ratios (Test [T]/Reference [R]) were close to 100%, and the corresponding CIs were both fully contained within the acceptance interval of 80.00 - 125.00%.

Pooled ANC and CD34+ analyses across doses:

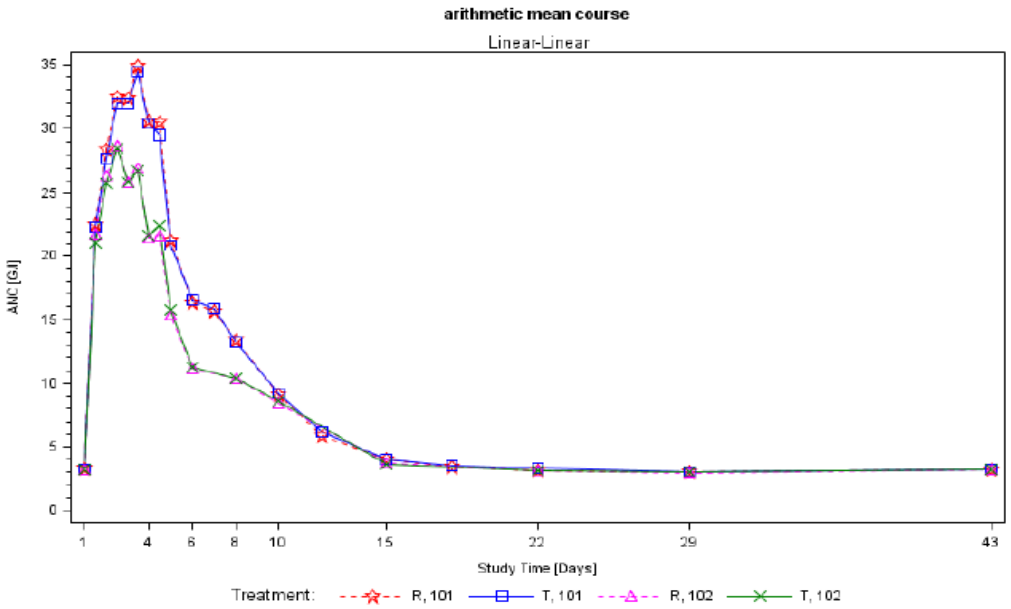


Figure 10: Mean effect-time profiles of ANC [G/l] by treatment study - PD set 101 and 102

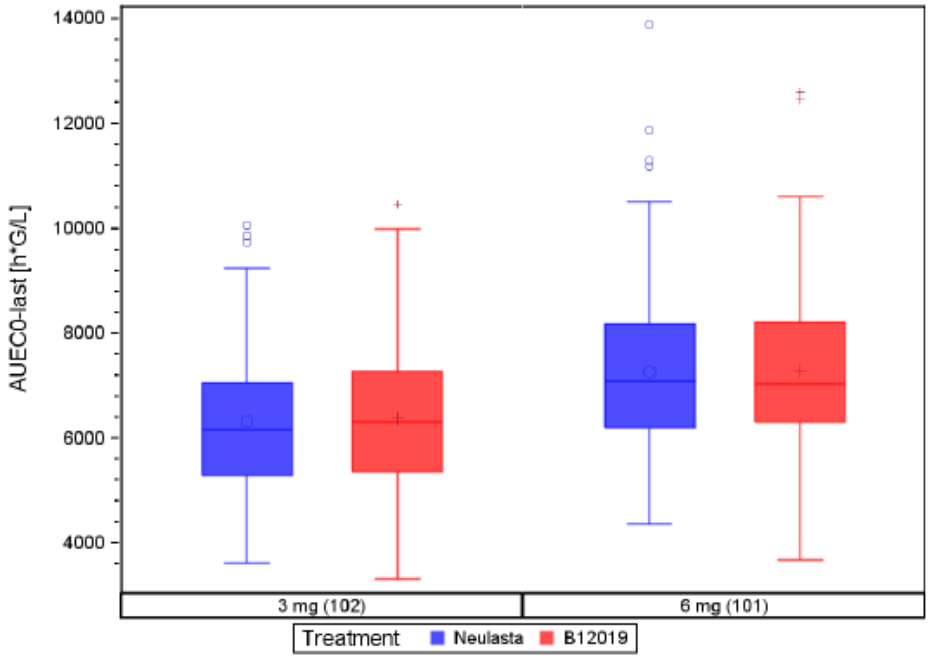


Figure 11: PD parameters AUEC0-last for ANC dose

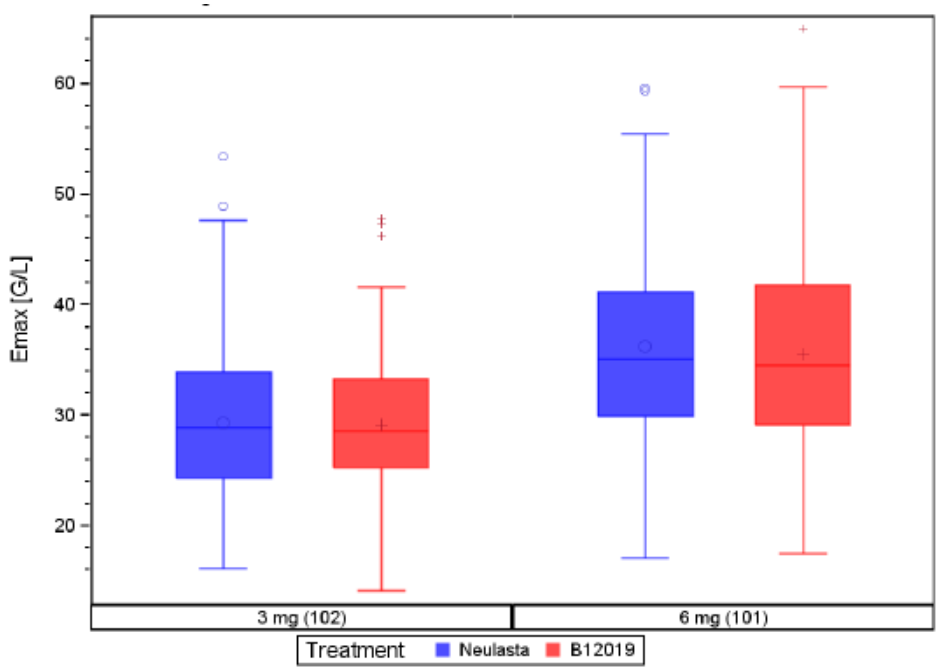


Figure 12: PD parameters Emax for ANC dose

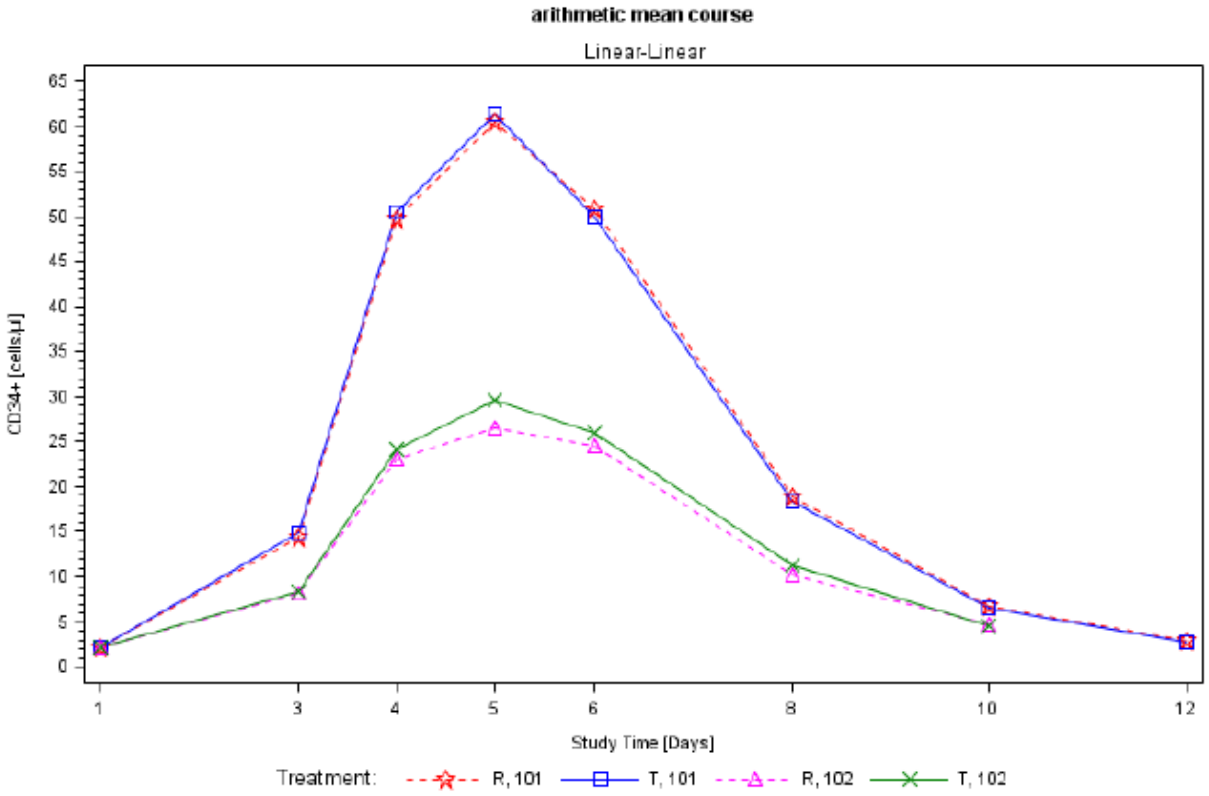


Figure 13: Mean course of CD34+ cell counts [cells/ul] by treatment and study - PD set 101 and 102

2.4.4. Discussion on clinical pharmacology

Two studies investigated the clinical pharmacology of Pelmeg (B12019-101 and B12019-102). The reference medicinal product in both clinical studies was EU-approved Neulasta. While the focus of study B12019-101 was on the PK/PD comparability of Pelmeg and Neulasta, study B12019-102 focused on the PD and immunogenicity comparability of the two products. The main difference between the two studies was the dose used, 6 mg in study B12019-101 and 3 mg in study B12019-102. Further the latter study contained 3 periods to evaluate ADA formation after repeated doses in a partial parallel design. The treatment was switched after two periods for PD analysis with the advantage of a cross-over design. The applicant did not submit studies on distribution, elimination, dose-proportionality and time dependencies, special populations, pharmacokinetics interaction studies, pharmacokinetics using biomaterials and mechanism of action. This is acceptable as according to the guideline EMEA/CHMP/BMWP/31329/2005, these studies are not required.

In general, the Applicant´s development program to demonstrate pharmacokinetic (PK) and pharmacodynamic (PD) similarity between Pelmeg and Neulasta is considered adequate and was performed in line with the guidance on similar biological products and in line with scientific advice received:

- Both clinical studies were conducted in healthy subjects. Compared to cancer patients receiving chemotherapy, healthy subjects lack co-morbidities and co-medications and are not immunosuppressed. Therefore, it was agreed that they represent the most sensitive study population for conducting the PK and PD comparison.
- In the pivotal study, B12019-101, a fixed subcutaneous dose of 6 mg pegfilgrastim was used. It was agreed in the EMA scientific advice that this dose has sufficient PK assay sensitivity since it is in the ascending part of the dose-response profile for AUC and Cmax. In the supportive study, PD comparability was investigated at a lower dose than 6 mg as recommended in the same scientific advice. The underlying rationale for this was that the 6 mg dose alone may not be sufficiently discriminative and sensitive for the comparative investigation of PD between Pelmeg and Neulasta.
- In both studies a wash-out period of 6 weeks was applied between treatments to avoid bias in the PK analysis due to increased clearance mediated by the neutrophils not returned to the baseline level.
- Given the expected high inter- and intra-individual variability for pegfilgrastim, a cross-over design was chosen to evaluate PK and PD comparability in pivotal study B12019-101. In addition, this study was planned as a two-stage design with sample size re-estimation after stage 1. A conservative assumption of an intra-subject CV of 50% was used for sample size estimation for stage 1.
- In study B12019-102, ADA formation was evaluated after the first two periods (TT or RR) prior to switching to the comparative product in period 3. To assess comparative ADA formation after two applications corresponding to a parallel design is acceptable. PD response (ANC, CD34+) was compared, based on the data obtained from all three periods, making use of the advantages of a cross-over design (T/R ratios based upon sequences T-T-R and R-R-T).

Pharmacokinetics

Pegfilgrastim concentrations in serum were determined using an ELISA technique. The Applicant provided additional validation data that showed acceptable precision and accuracy, dilutional linearity and sample long-term stability. However selectivity, without applying any correction for endogenous G-CSF, could only be shown if the acceptance criteria were widened to 30% and not for not for 20% (25% for LLOQ) as defined in the guideline. Since the pivotal PK study B12019-101 had a cross-over design, this bias associated with the presence of low levels of endogenous G-CSF should have influenced the sample measurement of both drugs in a similar way and is thus considered of minor relevance for the similarity assessment. In the pivotal study B12019-101, PK equivalence is claimed as the 94.32% CIs for the ratio of the test and reference product geometric means for the primary and secondary PK parameters $AUC_{0\text{-last}}$, $AUC_{0\text{-inf}}$ and Cmax were fully contained within the standard bioequivalence acceptance interval of 80.00-125.00%.

A difference in protein content of more than 5% between the test and reference batches was determined post-study using an optimized UV method. Although this %difference is not in line with the guideline on the investigation of bioequivalence, the impact on the PK comparability conclusions is considered to be minor. The test/reference ratio for both AUC0-last and Cmax (95.23% and 92.84%, respectively) was below 100% and is therefore in line with a lower concentration of the Pelmeg batch compared to the Neulasta batch used in this study. Furthermore, bioequivalence was demonstrated without applying a correction for protein content, which is considered the worst-case scenario.The results for the PK parameters $AUC_{0\text{-last}}$ and C_{max} showed a statistically significant period effect. Looking at results of LS means by period, it appears that $AUC_{0\text{-last}}$ and C_{max} are lower on average in Period 2. This is in line with what was to be expected. It is known that pegfilgrastim is mainly eliminated by neutrophils/precursors (Yang et al., Clin. Pharmacokinet., 2011) and the period effect observed in study B12019-101 was most likely attributed to target-mediated clearance of pegfilgrastim by the increasing number of neutrophils. The decrease in pegfilgrastim plasma levels could theoretically also be attributed to appearance of ADAs decreasing free pegfilgrastim concentrations. Following an analysis of the relationship between positive signals for PEG-reactive ADAs in period 1 and the PK response profile to administration of 6 mg pegfilgrastim (B12019 or Neulasta) in period 2, a trend for a slight reduction in the PK response in Period 2 for the positive PEG-reactive ADA sub-population was observed, regardless of the identity of the treatment sequence (see Immunogenicity in the clinical safety section). Moreover, subjects classified as ADA-positive showed no meaningful differences in PK, PD or other clinical parameters as compared to ADA-negative subjects. Overall, a modest impact of positive PEG-reactive antibody following the first administration of 6 mg pegfilgrastim (Pelmeg or Neulasta) on PK (AUC) following administration of a second 6 mg dose of pegfilgrastim could not be excluded. The marginal reduction in PK (AUC) was not associated with any reduction in the PD (AUEC for ANC). This observation does not have an effect on biosimilarity.

Study B12019-102 was not intended to evaluate PK equivalence between Pelmeg and Neulasta. However, based on sparse PK sampling, similar PK profiles were observed and overall, the PK data of study B12019-102 support the results on PK comparability between Neulasta and Pelmeg from study B12019-101.

Pharmacodynamics

In line with CHMP scientific advice, PD data in healthy volunteers (absolute neutrophil and CD34+ counts) were evaluated as surrogate marker of clinical efficacy and support of biosimilarity of both products. For the assessment of the PD comparability between Pelmeg and Neulasta, the primary PD parameter $AUEC_{0\text{-last}}$ of ANC was calculated, which had to be contained within the standard acceptance range of 80% - 125% with 95%CI, also for other pharmacodynamic parameters. Lower dose of pegfilgrastim, that is considered more sensitive to detect differences between products, has been tested in repeat dose study B12019-102. In this study, the primary endpoint for PD comparability was based on the data obtained from all three periods, making use of the advantages of a cross-over design (T/R ratios based upon sequences T-T-R and RR-T will be more robust and precise than estimates obtained from a parallel design). As secondary pharmacology the CD34+ cell count were also studied. The time course of mean CD34+ cells was similar when comparing the test and the reference products.

The primary PD endpoints have been met in both studies. At the time of scientific advice the Applicant has justified the use of the 80-125 boundaries in analogy to the PK evaluation and with reference to the bioequivalence guideline. However, a justification has been asked whether the chosen boundaries are sufficiently tight based on clinical argumentation, and why it is not necessary to use e.g. 85-118, or 90-111 as equivalence margins. Following scientific advice, equivalence margins of 90.00-111.00% were retrospectively applied for the PD evaluation in both studies with Pelmeg. The provided PD results support comparability of PD parameters with tighter equivalence margins.

In the Study B12019-101, the PD response was similar between test and reference products. The GMR of the ANC $AUEC_{0\text{-last}}$ was almost 100%; the 95% CI with 99.16; 102.65 was narrow and well within the acceptance interval of 80.00-125.00%. The secondary endpoints E_{max}, t_{max} of ANC and CD34+ count were initially presented only descriptively, their mean values were comparable.

A period effect was evident in ANC response, i.e. AUEC being higher with test and reference in period 2. Higher neutrophil counts are likely to increase pegfilgrastim clearance, resulting in a slightly lower exposure in period 2. This observation does not have an effect on biosimilarity.

In the Study B12019-102, PD comparability between Test and Reference was shown. In the primary analysis the GMR of AUEC 0-last (PEP) was near 100%. The corresponding 95% confidence interval was very close to 100% (99.58%; 103.63%) easily fulfilling the predefined similarity criteria of 80-125%. The intra-subject CV of 7.49% was low. Also the descriptively presented secondary PD endpoints ANC Emax, tmax and CD34+ cell count were very similar. Further sensitivity analyses with exclusion of cohort 1, period 1, period 1 data of cohort 1) also supported similarity for PD endpoints.

For Studies B12019-101 and B12019-102, the Applicant provided a statistical analysis complementing the mean CD34+ $AUEC_{0\text{-last}}$, E_{max} (post-hoc analysis) and median tmax values. The results presented for CD34+ parameters support the comparability.

In the **pooled analyses** of both PK/PD studies, mean responses of 6 mg pegfilgrastim were about 22% higher with Emax and 15% higher with $AUEC_{0\text{-last}}$ compared to 3 mg. Tmax was observed at 60h post dose with 6 mg and 36h post-dose with 3 mg. Dose difference was somewhat more perceivable regarding CD34+. This observation does not have an effect on biosimilarity.

2.4.5. Conclusions on clinical pharmacology

From a PK perspective, the claim of bioequivalence is acceptable since the 94.32% CIs of the test/reference ratio for both the primary and secondary PK parameters were fully contained within the acceptance interval of 80.00-125.00% in the pivotal study B12019-101.

Therefore, overall PK/PD data from the two studies show that similarity between Pelmeg and the reference product Neulasta could be demonstrated.

2.5. Clinical efficacy

No dedicated efficacy studies were performed in patients. For a biosimilar candidate to a pegfilgrastim, pivotal evidence for similar efficacy can be derived from the similarity in physicochemical, functional, pharmacokinetic and pharmacodynamic comparisons. Therefore, a dedicated comparative efficacy trial was not considered necessary.

2.6. Clinical safety

The clinical programme of Pelmeg comprises two studies in healthy volunteers that contributed to safety and immunogenicity analyses. The approved 6 mg dose was used in the pivotal pharmacokinetic (PK)/ pharmacodynamic (PD) study B12019-101. The supportive study B12019-102 mainly monitored immunogenicity, but also assessed PD comparability of Pelmeg and Neulasta, using a non-therapeutic, lower dose of 3 mg which was administered two times in a parallel group part of the study. After two administrations of Pelmeg, subjects received one administration of Neulasta, and in the other arm subjects received Pelmeg after two administrations of Neulasta.

Safety analyses are based on the safety population, defined as all subjects who have received at least one dose of study medication. Safety results are presented for the pooled studies, as well as for the individual studies. For pooling of safety data, the first period of both studies was regarded, as well as all periods from both studies. The rationale for presenting also individual study data is based on the two different doses and dosing regimens in the two studies (two doses of 6 mg in study B12019-101 versus three doses of 3 mg in study B12019-102).

Safety was evaluated from:

- Clinical safety assessments (reported AEs, physical examinations including vital signs and digital standard 12-lead ECGs, local tolerability),
- Laboratory safety assessments (standard haematology, coagulation and biochemistry analyses, urinalysis).

Patient exposure

A total of 268 subjects were randomised, 172 and 96 subjects in Studies B12019-101 and B12019-102, respectively. One subject in Study B12019-101 was randomised but not treated (due to the non treatment-emergent AE tachycardia in the pre-dose ECG). Thus, this subject was not included in the safety population. Therefore, the safety population in Studies B12019-101 and B12019-102 consisted of 171 and 96 subjects, respectively. The pooled safety population comprised 267 subjects.

Table 18: Extent of Exposure (Safety Population)

	Number of subjects						
	Neulasta		**Pelmeg**		**Pelmeg or Neulasta**		
Dose	1 dosing	2 dosings	1 dosing	2 dosings	1 dosing	2 dosings	3 dosings
3 mg	47	45	45	46	5	4	87
6 mg	168	NA	168	NA	6	165	NA
Total	**215**	**45**	**213**	**46**	**11**	**169**	**87**

NA=not applicable. Source: Pooled analyses, Table 2 in Module 5.3.5.3.

Subjects in Study B12019-101 were exposed to two 6 mg doses of study drug (one dose each of Pelmeg and Neulasta). Subjects in Study B12019-102 were exposed to three 3 mg doses of study drug. Depending on the sequence, subjects received either two doses of Pelmeg and one dose of Neulasta (Sequence T-T-R), or two doses of Neulasta and one dose of Pelmeg (Sequence R-R-T).

A total of 259 subjects received at least one dose of Pelmeg (3 mg or 6 mg); 46 of these received two doses. A total of 260 subjects received at least one dose of Neulasta (3 mg or 6 mg); 45 of these received two doses.

In Study B12019-101, all 163 subjects who completed the study, and the two subjects who discontinued after dosing in Period 2, i.e. 165 subjects, received two dosings of 6 mg study drug. Six subjects were dosed in Period 1 only (three with Pelmeg and three with Neulasta).

In Study B12019-102, all 84 subjects who completed the study, and the three subjects who discontinued after dosing in Period 3, i.e. 87 subjects, received one injection of 3 mg study drug per period. Five subjects were dosed in Period 1 only, and four subjects were dosed in Period 1 and 2. One subject randomised to sequence RRT was treated in an incorrect treatment sequence (TRR instead of RRT).

Demographic and Other Characteristics of the Study Population

Table 19: Demographics and Baseline Characteristics (Safety Population)

	Study B12019-101 N=171	Study B12019-102 N=96	Combined B12019-101 and B12019-102 N=267
Age (years)			
Median	42	45	43
Min, max	19, 55	21, 55	19, 55
Weight (kg)			
Median	81.6	80.6	81.3
Min, max	61.3, 99.3	62.4, 99.5	61.3, 99.5
Height (cm)			
Median	179	180	179
Min, max	165, 197	161, 194	161, 197
BMI (kg/m^2)			
Median	25.8	25.3	25.6
Min max	20.0, 30.0	20.0-30.0	20.0, 30.0
Race n(%)			
White	171 (100.0)	90 (93.8)	261 (97.8)
Black	0 (0)	5 (5.2)	5 (1.9)
Asian	0 (0)	1 (1.0)	1 (0.4)
Other	0 (0)	0 (0.0)	0 (0.0)
Smoking status n (%)			
No	139 (81.3)	78 (81.3)	217 (81.3)
Yes	32 (18.7)	18 (18.8)	50 (18.7)
ADA status n (%)			
Negative	137 (80.1)	94 (97.9)	231 (86.5)
Positive	34 (19.9)	2 (2.1)	36 (13.5)

BMI=body mass index, Max=maximum, Min=minimum, N=number of subjects, n=number of subjects in group. All subjects in the studies were male. Thus, subject distribution by sex is not shown.

Source: Pooled analyses, Table 1.2.1 and Table 1.3.1 in Module 5.3.5.3.

All subjects in clinical studies with Pelmeg were male. The majority of subjects overall were white (97.8%) and non-smokers (81.3%). Median age was 43 years, median weight was 81.3 kg and median BMI was 25.6 kg/m^2.

Adverse events

Due to the special design of study B12019-102 (three-periods, two-sequences cross-over), the frequencies of TEAEs were presented by treatment across all periods, but also by treatment utilizing data from Period 1 and 2 only (referring to the onset of the AEs, comparing incidences of AEs which occurred during the first two periods in which the study followed a parallel design).

An overview of TEAEs in clinical studies with Pelmeg is presented for the individual and the pooled studies in Table 22 below.

Table 20: Overview of Adverse Events (Safety Population)

	Number of subjects (%)								
	Study B12019-101 N=171			Study B12019-102 N=96			Combined B12019-101 and B12019-102 N=267		
	R	T	Total	R	T	Total	R	T	Total
Subjects with TEAE	139 (81.3)	147 (86.0)	155 (90.6)	80 (83.3)	76 (79.2)	92 (95.8)	219 (82.0)	223 (83.5)	247 (92.5)
Subjects with drug-related TEAE	136 (79.5)	141 (82.5)	151 (88.3)	73 (76.0)	71 (74.0)	89 (92.7)	209 (78.3)	212 (79.4)	240 (89.9)
Subjects with SAE	0 (0)	1 (0.6)	1 (0.6)	2 (2.1)	1 (1.0)	3 (3.1)	2 (0.7)	2 (0.7)	4 (1.5)
Subjects with TEAE leading to discontinuation	0 (0)	0 (0)	0 (0.0)	2 (2.1)	2 (2.1)	4 (4.2)	2 (0.7)	2 (0.7)	4 (1.5)
Deaths	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Subjects with mild TEAEs	108 (63.2)	108 (63.2)	140 (81.9)	66 (68.8)	67 (69.8)	86 (89.6)	174 (65.2)	175 (65.5)	226 (84.6)
Subjects with moderate TEAEs	111 (64.9)	119 (69.6)	136 (79.5)	52 (54.2)	60 (62.5)	76 (79.2)	163 (61.0)	179 (67.0)	212 (79.4)
Subjects with severe TEAEs	2 (1.2)	2 (1.2)	4 (2.3)	2 (2.1)	5 (5.2)	7 (7.3)	4 (1.5)	7 (2.6)	11 (4.1)
Number of TEAEs	494	485	979	262	308	570	756	793	1549
Number of drug-related TEAEs	400	400	800	210	237	447	610	637	1247
Average number of TEAEs per subject	3.6	3.3	6.3	3.3	4.1	6.2	3.5	3.6	6.3

AE=adverse event, R=reference (Neulasta), N=number of subjects, SAE=serious adverse event, T=test (Pelmeg), TEAE=treatment-emergent adverse event. Percentages are based on N.
Source: Pooled analyses, Table 4.1.1, 4.1.2 and 4.1.4 in Module 5.3.5.3.

The vast majority of subjects in both studies experienced at least one TEAE. Most TEAEs were related to study drug administration. Four subjects overall (1.5%) experienced SAEs, and four subjects overall (1.5%) discontinued due to TEAEs.

Table 21: Treatment-Emergent Adverse Events Reported in ≥2% of Subjects in any Group, by SOC and PT, Across All Periods (Safety Population)

System Organ Class Preferred Term	Number of subjects (%)								
	Study B12019-101			Study B12019-102			Combined B12019-101 and B12019-102		
	R N=171	T N=171	Total N=171	R N=96	T N=96	Total N=96	R N=267	T N=267	Total N=267
Any TEAE	**139 (81.3)**	**147 (86.0)**	**155 (90.6)**	**80 (83.3)**	**76 (79.2)**	**92 (95.8)**	**219 (82.0)**	**223 (83.5)**	**247 (92.5)**
Musculoskeletal and connective tissue disorders	**120 (70.2)**	**122 (71.3)**	**139 (81.3)**	**58 (60.4)**	**55 (57.3)**	**77 (80.2)**	**178 (66.7)**	**177 (66.3)**	**216 (80.9)**
Back pain	109 (63.7)	114 (66.7)	134 (78.4)	57 (59.4)	50 (52.1)	75 (78.1)	166 (62.2)	164 (61.4)	209 (78.3)
Pain in extremity	29 (17.0)	18 (10.5)	41 (24.0)	10 (10.4)	9 (9.4)	16 (16.7)	39 (14.6)	27 (10.1)	57 (21.3)

System Organ Class Preferred Term	**Number of subjects (%)**								
	Study B12019-101			**Study B12019-102**			**Combined B12019-101 and B12019-102**		
	R N=171	**T N=171**	**Total N=171**	**R N=96**	**T N=96**	**Total N=96**	**R N=267**	**T N=267**	**Total N=267**
Neck pain	14 (8.2)	8 (4.7)	21 (12.3)	1 (1.0)	1 (1.0)	2 (2.1)	15 (5.6)	9 (3.4)	23 (8.6)
Arthralgia	5 (2.9)	6 (3.5)	9 (5.3)	4 (4.2)	7 (7.3)	11 (11.5)	9 (3.4)	13 (4.9)	20 (7.5)
Myalgia	9 (5.3)	6 (3.5)	15 (8.8)	3 (3.1)	3 (3.1)	4 (4.2)	12 (4.5)	9 (3.4)	19 (7.1)
Musculoskeletal pain	8 (4.7)	5 (2.9)	13 (7.6)	2 (2.1)	3 (3.1)	5 (5.2)	10 (3.7)	8 (3.0)	18 (6.7)
Musculoskeletal chest pain	4 (2.3)	6 (3.5)	8 (4.7)	1 (1.0)	1 (1.0)	1 (1.0)	5 (1.9)	7 (2.6)	9 (3.4)
Bone pain	5 (2.9)	1 (0.6)	6 (3.5)	0 (0.0)	0 (0.0)	0 (0.0)	5 (1.9)	1 (0.4)	6 (2.2)
Groin pain	2 (1.2)	3 (1.8)	5 (2.9)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.7)	3 (1.1)	5 (1.9)
Nervous system disorders	**58 (33.9)**	**56 (32.7)**	**80 (46.8)**	**25 (26.0)**	**30 (31.3)**	**41 (42.7)**	**83 (31.1)**	**86 (32.2)**	**121 (45.3)**
Headache	52 (30.4)	54 (31.6)	76 (44.4)	22 (22.9)	29 (30.2)	40 (41.7)	74 (27.7)	83 (31.1)	116 (43.4)
Dizziness	4 (2.3)	2 (1.2)	6 (3.5)	1 (1.0)	1 (1.0)	2 (2.1)	5 (1.9)	3 (1.1)	8 (3.0)
Paraesthesia	1 (0.6)	2 (1.2)	3 (1.8)	2 (2.1)	0 (0.0)	2 (2.1)	3 (1.1)	2 (0.7)	5 (1.9)
Infections and infestations	**36 (21.1)**	**28 (16.4)**	**56 (32.7)**	**24 (25.0)**	**25 (26.0)**	**38 (39.6)**	**60 (22.5)**	**53 (19.9)**	**94 (35.2)**
Nasopharyngitis	28 (16.4)	27 (15.8)	50 (29.2)	19 (19.8)	23 (24.0)	36 (37.5)	47 (17.6)	50 (18.7)	86 (32.2)
Oral herpes	3 (1.8)	1 (0.6)	4 (2.3)	1 (1.0)	0 (0.0)	1 (1.0)	4 (1.5)	1 (0.4)	5 (1.9)
Gastroenteritis	1 (0.6)	0 (0.0)	1 (0.6)	2 (2.1)	1 (1.0)	3 (3.1)	3 (1.1)	1 (0.4)	4 (1.5)
Influenza	2 (1.2)	0 (0.0)	2 (1.2)	1 (1.0)	1 (1.0)	2 (2.1)	3 (1.1)	1 (0.4)	4 (1.5)
Metabolism and nutrition disorders	**38 (22.2)**	**34 (19.9)**	**50 (29.2)**	**21 (21.9)**	**23 (24.0)**	**32 (33.3)**	**59 (22.1)**	**57 (21.3)**	**82 (30.7)**
Hypoglycaemia	37 (21.6)	33 (19.3)	49 (28.7)	20 (20.8)	21 (21.9)	29 (30.2)	57 (21.3)	54 (20.2)	78 (29.2)
Hyperkalaemia	0 (0.0)	1 (0.6)	1 (0.6)	1 (1.0)	1 (1.0)	2 (2.1)	1 (0.4)	2 (0.7)	3 (1.1)
Investigations	**25 (14.6)**	**32 (18.7)**	**44 (25.7)**	**22 (22.9)**	**19 (19.8)**	**33 (34.4)**	**47 (17.6)**	**51 (19.1)**	**77 (28.8)**
Blood pressure systolic increased	4 (2.3)	10 (5.8)	11 (6.4)	9 (9.4)	9 (9.4)	14 (14.6)	13 (4.9)	19 (7.1)	25 (9.4)
ALT increased	8 (4.7)	8 (4.7)	12 (7.0)	3 (3.1)	1 (1.0)	4 (4.2)	11 (4.1)	9 (3.4)	16 (6.0)
Blood creatine phosphokinase increased	1 (0.6)	5 (2.9)	6 (3.5)	1 (1.0)	5 (5.2)	6 (6.3)	2 (0.7)	10 (3.7)	12 (4.5)
C-reactive protein increased	3 (1.8)	3 (1.8)	5 (2.9)	4 (4.2)	2 (2.1)	5 (5.2)	7 (2.6)	5 (1.9)	10 (3.7)
GGT increased	4 (2.3)	5 (2.9)	6 (3.5)	1 (1.0)	3 (3.1)	3 (3.1)	5 (1.9)	8 (3.0)	9 (3.4)
AST increased	0 (0.0)	3 (1.8)	3 (1.8)	1 (1.0)	2 (2.1)	3 (3.1)	1 (0.4)	5 (1.9)	6 (2.2)
Blood bilirubin increased	2 (1.2)	1 (0.6)	3 (1.8)	2 (2.1)	0 (0.0)	2 (2.1)	4 (1.5)	1 (0.4)	5 (1.9)
Blood pressure increased	0 (0.0)	0 (0.0)	0 (0.0)	4 (4.2)	4 (4.2)	5 (5.2)	4 (1.5)	4 (1.5)	5 (1.9)
General disorders and administration site conditions	**21 (12.3)**	**27 (15.8)**	**41 (24.0)**	**8 (8.3)**	**13 (13.5)**	**20 (20.8)**	**29 (10.9)**	**40 (15.0)**	**61 (22.8)**
Fatigue	4 (2.3)	7 (4.1)	11 (6.4)	0 (0.0)	1 (1.0)	1 (1.0)	4 (1.5)	8 (3.0)	12 (4.5)
Chest pain	3 (1.8)	3 (1.8)	6 (3.5)	2 (2.1)	3 (3.1)	5 (5.2)	5 (1.9)	6 (2.2)	11 (4.1)

System Organ Class Preferred Term	Number of subjects (%)								
	Study B12019-101			Study B12019-102			Combined B12019-101 and B12019-102		
	R N=171	T N=171	Total N=171	R N=96	T N=96	Total N=96	R N=267	T N=267	Total N=267
Feeling hot	4 (2.3)	5 (2.9)	9 (5.3)	0 (0.0)	1 (1.0)	1 (1.0)	4 (1.5)	6 (2.2)	10 (3.7)
Chest discomfort	3 (1.8)	3 (1.8)	6 (3.5)	1 (1.0)	1 (1.0)	2 (2.1)	4 (1.5)	4 (1.5)	8 (3.0)
Pyrexia	1 (0.6)	3 (1.8)	4 (2.3)	0 (0.0)	2 (2.1)	2 (2.1)	1 (0.4)	5 (1.9)	6 (2.2)
Chills	3 (1.8)	2 (1.2)	5 (2.9)	0 (0.0)	0 (0.0)	0 (0.0)	3 (1.1)	2 (0.7)	5 (1.9)
Discomfort	0 (0.0)	1 (0.6)	1 (0.6)	1 (1.0)	1 (1.0)	2 (2.1)	1 (0.4)	2 (0.7)	3 (1.1)
Feeling cold	0 (0.0)	1 (0.6)	1 (0.6)	0 (0.0)	2 (2.1)	2 (2.1)	0 (0.0)	3 (1.1)	3 (1.1)
Puncture site pain	0 (0.0)	1 (0.6)	1 (0.6)	2 (2.1)	0 (0.0)	2 (2.1)	2 (0.7)	1 (0.4)	3 (1.1)
Gastrointestinal disorders	**20 (11.7)**	**22 (12.9)**	**36 (21.1)**	**4 (4.2)**	**16 (16.7)**	**19 (19.8)**	**24 (9.0)**	**38 (14.2)**	**55 (20.6)**
Nausea	6 (3.5)	6 (3.5)	10 (5.8)	1 (1.0)	8 (8.3)	9 (9.4)	7 (2.6)	14 (5.2)	19 (7.1)
Diarrhoea	3 (1.8)	5 (2.9)	8 (4.7)	1 (1.0)	2 (2.1)	3 (3.1)	4 (1.5)	7 (2.6)	11 (4.1)
Toothache	4 (2.3)	1 (0.6)	5 (2.9)	1 (1.0)	4 (4.2)	4 (4.2)	5 (1.9)	5 (1.9)	9 (3.4)
Abdominal pain	2 (1.2)	3 (1.8)	5 (2.9)	0 (0.0)	3 (3.1)	3 (3.1)	2 (0.7)	6 (2.2)	8 (3.0)
Vomiting	2 (1.2)	1 (0.6)	3 (1.8)	0 (0.0)	4 (4.2)	4 (4.2)	2 (0.7)	5 (1.9)	7 (2.6)
Dry mouth	3 (1.8)	3 (1.8)	6 (3.5)	0 (0.0)	0 (0.0)	0 (0.0)	3 (1.1)	3 (1.1)	6 (2.2)
Dyspepsia	1 (0.6)	3 (1.8)	4 (2.3)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)	3 (1.1)	4 (1.5)
Respiratory, thoracic and mediastinal disorders	**18 (10.5)**	**15 (8.8)**	**30 (17.5)**	**4 (4.2)**	**10 (10.4)**	**14 (14.6)**	**22 (8.2)**	**25 (9.4)**	**44 (16.5)**
Oropharyngeal pain	12 (7.0)	7 (4.1)	18 (10.5)	1 (1.0)	5 (5.2)	6 (6.3)	13 (4.9)	12 (4.5)	24 (9.0)
Cough	4 (2.3)	6 (3.5)	9 (5.3)	1 (1.0)	6 (6.3)	7 (7.3)	5 (1.9)	12 (4.5)	16 (6.0)
Skin and subcutaneous tissue disorders	**10 (5.8)**	**8 (4.7)**	**15 (8.8)**	**1 (1.0)**	**2 (2.1)**	**3 (3.1)**	**11 (4.1)**	**10 (3.7)**	**18 (6.7)**
Hyperhidrosis	1 (0.6)	4 (2.3)	5 (2.9)	0 (0.0)	1 (1.0)	1 (1.0)	1 (0.4)	5 (1.9)	6 (2.2)
Erythema	3 (1.8)	2 (1.2)	5 (2.9)	0 (0.0)	0 (0.0)	0 (0.0)	3 (1.1)	2 (0.7)	5 (1.9)
Cardiac disorders	**7 (4.1)**	**4 (2.3)**	**11 (6.4)**	**0 (0.0)**	**1 (1.0)**	**1 (1.0)**	**7 (2.6)**	**5 (1.9)**	**12 (4.5)**
Palpitations	5 (2.9)	2 (1.2)	7 (4.1)	0 (0.0)	0 (0.0)	0 (0.0)	5 (1.9)	2 (0.7)	7 (2.6)
Injury, poisoning and procedural complications	**3 (1.8)**	**3 (1.8)**	**6 (3.5)**	**3 (3.1)**	**2 (2.1)**	**4 (4.2)**	**6 (2.2)**	**5 (1.9)**	**10 (3.7)**
Arthropod sting	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.0)	1 (1.0)	2 (2.1)	1 (0.4)	1 (0.4)	2 (0.7)
Renal and urinary disorders	**4 (2.3)**	**3 (1.8)**	**6 (3.5)**	**1 (1.0)**	**3 (3.1)**	**3 (3.1)**	**5 (1.9)**	**6 (2.2)**	**9 (3.4)**
Haematuria	1 (0.6)	1 (0.6)	2 (1.2)	1 (1.0)	2 (2.1)	3 (3.1)	2 (0.7)	3 (1.1)	5 (1.9)
Eye disorders	**4 (2.3)**	**2 (1.2)**	**6 (3.5)**	**1 (1.0)**	**1 (1.0)**	**2 (2.1)**	**5 (1.9)**	**3 (1.1)**	**8 (3.0)**
Ocular hyperaemia	2 (1.2)	0 (0.0)	2 (1.2)	1 (1.0)	1 (1.0)	2 (2.1)	3 (1.1)	1 (0.4)	4 (1.5)
Vascular disorders	**3 (1.8)**	**2 (1.2)**	**5 (2.9)**	**1 (1.0)**	**2 (2.1)**	**3 (3.1)**	**4 (1.5)**	**4 (1.5)**	**8 (3.0)**
Hot flush	2 (1.2)	2 (1.2)	4 (2.3)	0 (0.0)	1 (1.0)	1 (1.0)	2 (0.7)	3 (1.1)	5 (1.9)
Haematoma	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.0)	1 (1.0)	2 (2.1)	1 (0.4)	1 (0.4)	2 (0.7)

AE=adverse event, ALT=alanine aminotransferase, AST=aspartate aminotransferase, GGT=gamma glutamyltransferase, MedDRA=Medical Dictionary for Regulatory Activities, N=number of subjects, PT=preferred term, R=reference (Neulasta), SOC=System Organ Class, T=test (Pelmeg), TEAE=treatment-emergent adverse

event. AEs were coded using MedDRA Version 18.1 (study B12019-101) and 19.1 (study B12019-102). Percentages are based on N. Adverse events are sorted by descending frequency in SOC (pooled studies). The sum of subjects with events at the PT level may be greater than the number of subjects with an event at the SOC level. One subject may have had more than one PT event, but was only counted once at the SOC level.

The most frequent TEAEs by SOC were musculoskeletal and connective tissue disorders (80.9% of subjects overall), followed by nervous system disorders (45.3% overall), infections and infestations (35.2% overall), and metabolism and nutrition disorders (30.7% overall). Most frequently reported PTs were back pain (78.3% overall), headache (43.4% overall), nasopharyngitis (32.2% overall), hypoglycaemia (29.2% overall) and pain in extremity (21.3% overall).

In the Study B12019-101, the number of subjects with TEAEs was similar: 81.3% of the subjects reported 494 TEAEs after Neulasta and 86.0% reported 485 TEAEs after B12019. The most frequently reported TEAE was back pain, experienced by 63.7% of the subjects after Neulasta and 66.7% after B12019, followed by headache, experienced by 30.4% of the subjects after Neulasta and 31.6% after B12019.

Notably higher TEAE frequencies after Pelmeg than after Neulasta were seen in Study B12019-102 for the SOCs “gastrointestinal disorders” (16.7% vs. 4.2%) and “respiratory, thoracic and mediastinal disorders” (10.4% vs 4.2%). The imbalance in these SOCs was mainly driven by subjective measurable symptoms and signs such as e.g. nausea (R: 1.0% vs T: 8.3%) or cough (R: 1% vs T: 6.3%). A root cause analysis has been provided to investigate potential possibilities.

Most frequent in both studies and with both treatments were TEAEs in the SOC musculoskeletal and connective tissue disorders. In Study B12019-101, these were reported for 70.2% and 71.3% of subjects after Neulasta and Pelmeg, respectively (first period: 81.4% and 70.6%). In Study B12019-102, a slightly lower percentage of subjects experienced such AEs, 60.4% and 57.3% after Neulasta and Pelmeg, respectively (first period: 63.8% and 69.4%). These slight differences in AE frequencies do not point to a pronounced dose effect. When looking at AEs occurring in the first treatment period only, numbers were similar, which indicates that musculoskeletal AEs occur with similar frequencies at the beginning of treatment and after re-exposure.

Within this SOC, back pain and pain in extremity accounted for the majority of TEAEs, while neck pain, arthralgia, myalgia, musculoskeletal (chest) pain, bone pain and groin pain were each reported in less than 10% of subjects.

In both studies, around 20% of subjects experienced drug-related hypoglycaemia after dosing with Pelmeg or Neulasta. Hypoglycaemia had to be reported as AE if glucose levels decreased to 3.8 mmol/L or below. For further details on events of hypoglycaemia and glucose concentrations, see Section laboratory findings below.

Injection site reactions

Local tolerability at the injection site was assessed pre-dose and several times post-dose in each period. Evaluation made use of the injection site reaction score, ranging from 0 (none) to 3 (severe).The incidence of injection site reactions was higher in study B12019-101, using the 6 mg dose and overall the incidence was higher with Pelmeg relative to Neulasta (7 vs. 4). In particular injection site haematoma were more frequent with Pelmeg treatment (R: 0.0% vs T: 1.8% in study B12019-101). The 267 subjects participating in the two Pelmeg studies received around 600 injections of study drug. Considering data from both studies, only 11 of 267 subjects (4.1%) had injection site reactions, 7 (2.6%) after Pelmeg and 4 (1.5%) after Neulasta. In all subjects, injection site reactions occurred only once, even though subjects received 2 injections in Study B12019-101 and 3 injections in Study B12019-102. For the individual PTs, some imbalances were seen, e.g. puncture site pain occurred only under Neulasta, while injection site haematoma/haematoma was more frequent after Pelmeg.

Serious adverse event / deaths / other significant events

No death occurred during the two studies in healthy volunteers. Serious adverse events were reported for one subject in study B12019-101 (suffering from multiple injuries due to a car accident) and three subjects in study B12019-102 (two suffering from influenza and one suffering from local swelling due to a facelift). The reported serious events can be considered as unrelated to the study drug.

Laboratory findings

Laboratory AEs

Blood samples for haematology and biochemistry parameters and urine samples for urinalysis were collected at screening, on Day -1, and several times post-dose. In both clinical studies with Pelmeg, the investigator decided whether a laboratory abnormality was considered as an AE. In addition, for certain laboratory parameters of interest (i.e. LFTs, creatinine, electrolytes, haematology, muscle and coagulation) specific ranges were defined, outside of which laboratory values were considered clinically relevant, and had to be reported as laboratory AE. For Study B12019-101, this reporting process (including the setup of threshold values for laboratory AEs) was implemented during the study, while for Study B12019-102 the process was pre-specified. Laboratory AEs were reported for both treatments in both studies.

All laboratory abnormalities reported as AEs in B12019 studies are presented below.

Table 22: Laboratory Adverse Events (Safety Population)

	Number of subjects (%)					
Preferred term	**Study B12019-101**		**Study B12019-102**		**Combined B12019-101 and B12019-102**	
	R N=171	**T N=171**	**R N=96**	**T N=96**	**R N=267**	**T N=267**
Hypoglycaemia	37 (21.6)	33 (19.3)	20 (20.8)	21 (21.9)	57 (21.3)	54 (20.2)
ALT increased	8 (4.7)	8 (4.7)	3 (3.1)	1 (1.0)	11 (4.1)	9 (3.4)
Blood creatine phosphokinase increased	1 (0.6)	5 (2.9)	1 (1.0)	5 (5.2)	2 (0.7)	10 (3.7)
CRP increased	3 (1.8)	3 (1.8)	4 (4.2)	2 (2.1)	7 (2.6)	5 (1.9)
GGT increased	4 (2.3)	5 (2.9)	1 (1.0)	3 (3.1)	5 (1.9)	8 (3.0)
AST increased	0 (0.0)	3 (1.8)	1 (1.0)	2 (2.1)	1 (0.4)	5 (1.9)
Blood bilirubin increased	2 (1.2)	1 (0.6)	2 (2.1)	0 (0.0)	4 (1.5)	1 (0.4)
Haematuria	1 (0.6)	1 (0.6)	1 (1.0)	2 (2.1)	2 (0.7)	3 (1.1)
Blood alkaline phosphatase increased	1 (0.6)	2 (1.2)	0 (0.0)	1 (1.0)	1 (0.4)	3 (1.1)
Hyperkalaemia	0 (0.0)	1 (0.6)	1 (1.0)	1 (1.0)	1 (0.4)	2 (0.7)
aPTT prolonged	2 (1.2)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.7)	0 (0.0)
Blood potassium increased	1 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)	0 (0.0)
Hypertransaminasemia	0 (0.0)	1 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)
Leukocytosis	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.0)	0 (0.0)	1 (0.4)
Leukocyturia	0 (0.0)	0 (0.0)	1 (1.0)	1 (1.0)	1 (0.4)	1 (0.4)
Prothrombin time shortened	1 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)	0 (0.0)
WBCs urine	1 (0.6)	1 (0.6)	0 (0.0)	0 (0.0)	1 (0.4)	1 (0.4)

ALT=alanine aminotransferase, aPTT=activated partial thromboplastin time, AST=aspartate aminotransferase, CRP=C reactive protein, GGT= gamma-glutamyltransferase, N=number of subjects, WBC=white blood cell. Source: Pooled analyses, Table 4.2.1 in Module 5.3.5.3.

By far the most frequent laboratory AE was hypoglycaemia, reported in around 20% of subjects in Studies B12019-101 and B12019-102. Decreases in serum glucose after administration of study drug were seen in both clinical studies, and were comparable between treatments.

In Study B12019-101, mean glucose decreased from 5.13 mmol/L on Day -1 to a minimum of 3.88 mmol/L at Day 3 (48 hours) after dosing with Neulasta, and from 5.11 mmol/L on Day -1 to a minimum of 3.93 mmol/L at Day 3 (48 hours) after dosing with Pelmeg. Thereafter blood glucose increased again and reached pre-dose level on Day 15.

In Study B12019-102, mean glucose decreased from 5.13 mmol/L on Day -1 to a minimum of 3.93 mmol/L at Day 3 (48 hours) after dosing with Neulasta, and from 5.13 mmol/L on Day -1 to a minimum of 3.94 mmol/L at 48 hours after dosing with Pelmeg. Thereafter blood glucose increased again and reached pre-dose level on Day 15.

Comparable mean decreases in serum glucose of around 1 mmol/L were observed in both studies and after both treatments, reaching a maximum at 48 hours after dosing. The similarity of results from the two studies suggests that the effect is not dose dependent at the two doses studied. None of the events of hypoglycaemia was associated with clinical signs or symptoms, or required intervention.

In both studies, an inverse relationship was observed for the time pattern of glucose and ANC concentrations. Mean ANC increased until 36 hours after dosing, remained on this level until 84 hours after dosing and decreased thereafter. Pre-dose level of ANC was reached again on Day 18. Mean glucose decreased until 48 hours after dosing, increased thereafter and reached pre-dose level again on Day 15.

Other laboratory AEs reported by more than 2% of subjects in any group were increases in: ALT, creatine phosphokinase, C-reactive protein, GGT, AST, and blood bilirubin, as well as haematuria.

In Study B12019-102, two subjects discontinued due to increased ALT. In all other subjects, AEs of ALT, AST and GGT were mild to moderate, reversible, and did not require intervention.

Vital signs and physical examination

Blood pressure, pulse rate, respiratory rate and tympanic temperature were measured at screening, pre-dose on Day 1, several times post-dose in each period and at the follow-up, after at least 5 minutes rest in supine position. Physical examination was performed during screening, at admission and during the follow-up examination.

In both studies, mean values for systolic and diastolic blood pressure, pulse rate, respiratory rate and body temperature showed no clinically relevant changes after dosing with study drug or differences between treatments.

ECG

In study B12019-101, electrocardiograms were recorded at screening, before and after dosing and at the follow-up. In study B12019-102 Electrocardiograms were recorded at screening, on Day -1 and prior to discharge on Days 5 of each period and on Day 43 of period 3. All 12-lead ECGs were normal or abnormal without clinical significance in both studies, except for one subject, who suffered from tachycardia before study drug administration in study B12019-101. Thus, this subject was not treated with study drug and not included in the safety population.

Immunological events

Immunogenicity

Analytical methods

A multi-tier test strategy (screening, confirmatory and neutralisation assay) was applied to evaluate relative immunogenicity of Pelmeg versus Neulasta. Identical bioanalytical methods were applied to studies B12019-101 and B12019-102.

Initially, samples were subjected to a screening assay. The bridging assay format used biotinylated Pelmeg and Sulfo-TAG-Pelmeg as the labelled antigens, and electrochemiluminescence (ECL) detection of solid-phase-bound complexes of ADA with bridged labelled antigen on streptavidin-coated MSD microtiter plates. An in-house prepared rabbit, affinity-purified, anti-Pelmeg antibody reagent was selected as the primary positive control, whereas a commercial mouse anti-PEG IgM antibody was selected as a positive control for monitoring PEG reactivity in the ADA assay. Antigenic equivalence of the positive control (anti-Pelmeg) antibody for Pelmeg and Neulasta was demonstrated at LPC and HPC level using a single-assay format (with labelled Pelmeg antigen).

All screened positive samples were tested in a confirmatory assay in the same assay format using four different competitive inhibitors (Pelmeg, Neulasta, filgrastim (G-CSF) and PEG_{6000}). Subsequently, titer values were determined for samples that were positive in at least one of the confirmatory antibody assays.

Lastly, all confirmed positive samples were tested using a neutralising assay format.

Results for ADA formation

Table 23: Summary of ADA Results in Study B12019-101 (Safety Population, N=171) and Study B12019-102 (Safety Population without Subject 020, N=95)

Statistic		B12109-101	B12109-102
No. subjects in safety population		171	95*
Results of ADA screening assay			
Total number of samples tested		1835	1409
Total number of reactive samples		539	47
No. subjects with ≥1 reactive sample		98 (57.3%)	25 (26.3%)
Results of ADA confirmatory assay			
No. (%) subjects with ≥1 confirmed ADA positive sample		34 (19.9%)	2 (2.1%)
No. subjects (%) positive with each competing antigen in confirmatory ADA assay	Neulasta + B12019 + PEG_{6000}	6 (3.5%)	1 (1.1%)
	B12019 + PEG_{6000}	3 (1.8%)	0 (0%)
	Neulasta + PEG_{6000}	1 (0.6%)	0 (0%)
	PEG_{6000} only	24 (14.0%)	0 (0%)
	B12019 only	0 (0%)	0 (0%)
	Neulasta only	0 (0%)	1 (1.1%)#
	Filgrastim only	0 (0%)	0 (0%)
No. of confirmed negative samples		396	45
ADA false positives in ADA screening assay (% of total)		21.6%	3.2%
Results of nAb assay			
No. samples positive in nAb assay (%)		0 (0%)	0 (0%)
No. subjects positive in nAb assay (%)		0 (0%)	0 (0%)

ADA=anti-drug antibody, nAb=neutralising antibody.
# Subject 86 in Study B12019-102 has a reactive sample at Period 1 Day 15 which was inhibited to a similar extent by B12019, Neulasta and PEG6000, although only the result for Neulasta was above the confirmatory cutpoint
* Safety population in study B12019-102 was N=96. Subject 20 was excluded from the Safety population since Period 1 and 3 were mixed up, but the sample from this subject are included in the total of 1409 samples tested
Source: Integrated Summary of immunogenicity, Section 5 "Overall Conclusions"

Study B12019-101

Immunogenicity was evaluated as secondary endpoint. Subjects were dosed once with 6 mg Pelmeg and once with 6 mg Neulasta. Blood samples for assessment of immunogenicity were obtained on Day 1 pre-dose, Days 8, 15, 22, 29 of each period and Day 43 of the last period for detection of ADA formation.

Overall, 34 of 171 (19.9%) subjects in the safety population had confirmed ADA positive reactivity with PEG; 9 of these 34 subjects also had confirmed positive ADA reactivity with Pelmeg; 7 of these 34 also had confirmed positive ADA reactivity with Neulasta. Of note, some of these subjects had only a single positive ADA sample throughout the study. No filgrastim-reactive positive samples were detected in any subject treated in study B12019-101. Thus, the detected signals appear to represent antibodies reactive with PEG, or with the PEG moiety of Pelmeg or Neulasta. No samples with neutralising capacity in the cell-based assay were detected.

Overall 6 subjects were ADA positive (positive confirmatory assay of any of the 4 specificity cut points: B12019, Neulasta, filgrastim or PEG 6000) already prior to dosing in Period 1. Only 1 of these subjects remained ADA positive throughout the study. Until the end of Period 1, 15 subjects who received Neulasta and 18 subjects who received B12019 had at least one ADA positive examination. One further subject was ADA positive until the end of Period 2 (he received Neulasta in Period 1 and B12019 in Period 2), i.e. overall 34 subjects had at least one positive confirmatory assay for one time point.

The highest frequency of ADA positive subjects was on Day 15 of Period 1 (12 subjects [14%] each after Neulasta and B12019). Thereafter the number of ADA positive subjects decreased and on Day 43 of Period 2 overall only 9 subjects were ADA positive (3 subjects with the treatment sequence Neulasta/B12019 and 6 subjects with the treatment sequence B12019/Neulasta, all PEG-positive).

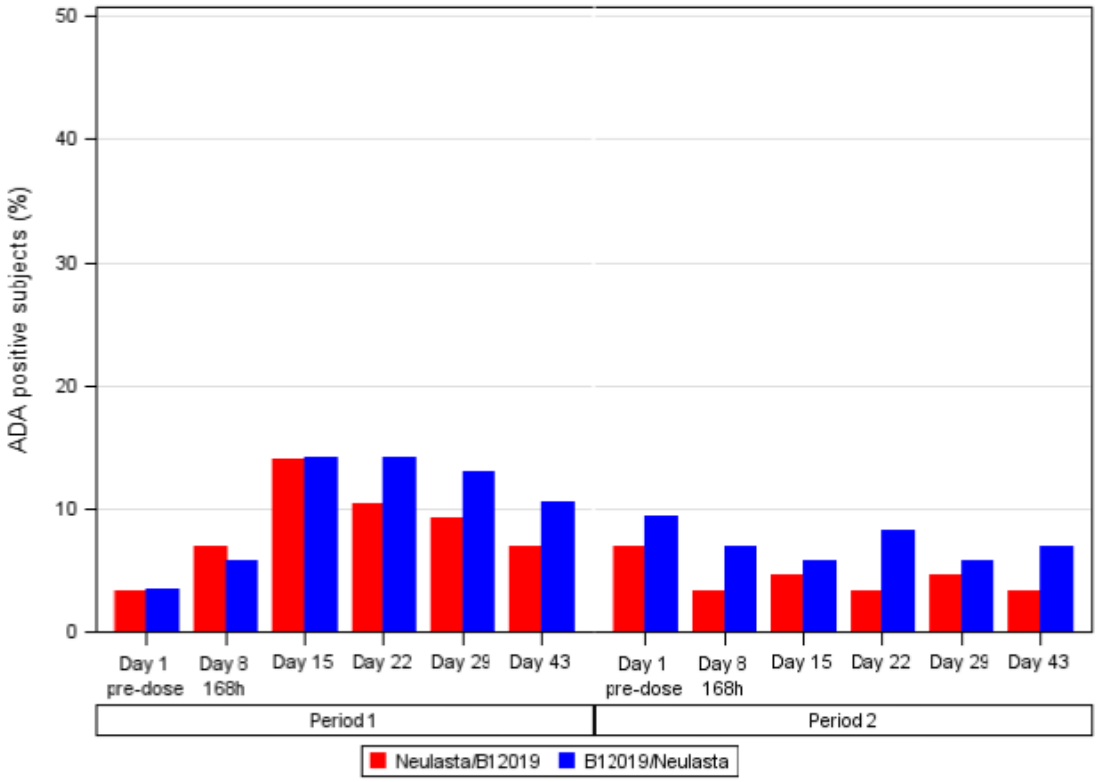


Figure 14: Frequency of ADA-positive Subjects by Treatment Sequence, Period and Timepoint (Safety Set)

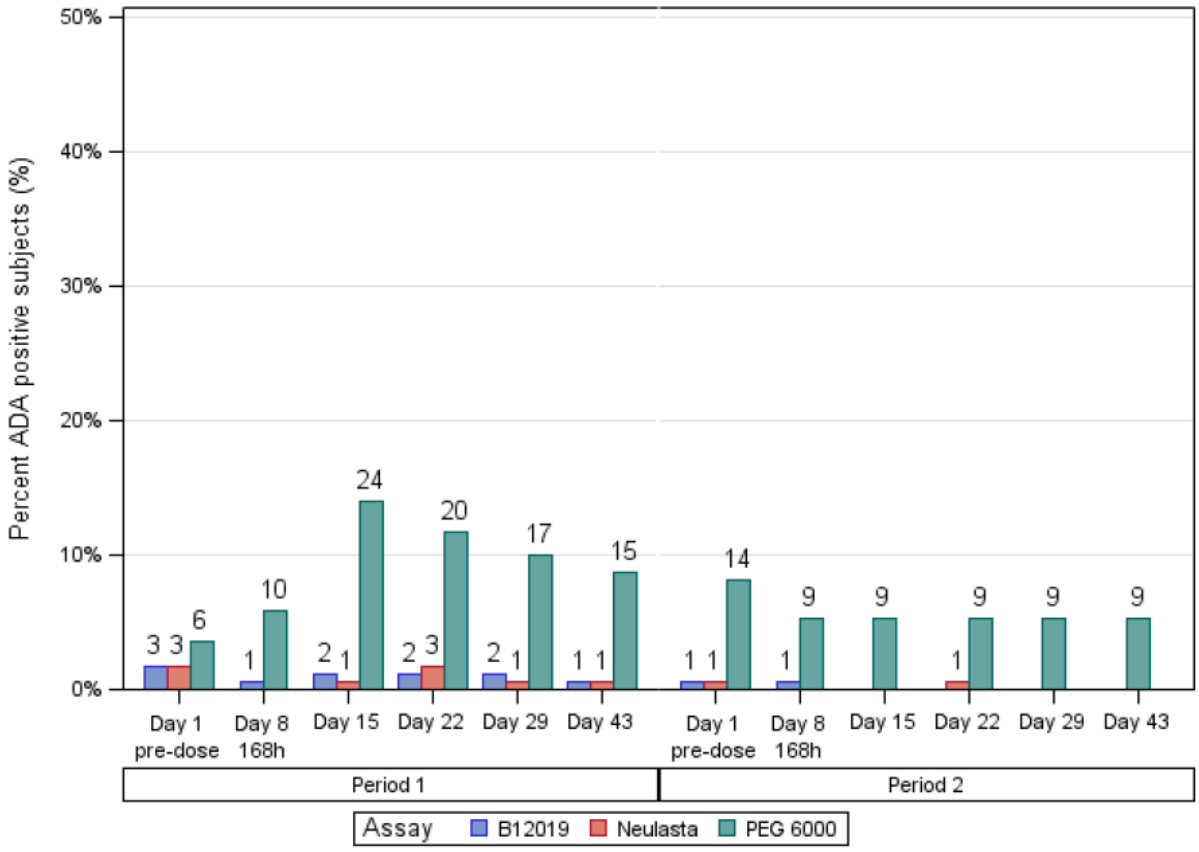


Figure 15: Frequency of ADA-positive Subjects by Assay, Period and Timepoint (Safety Set)

ADA=anti-drug antibody
Source: Integrated summary of immunogenicity. Pooled immunogenicity analysis

A higher incidence of confirmed PEG-reactive antibody signals was detected in Period 1 compared to Period 2 regardless of treatment sequence (Integrated Summary of immunogenicity). ADA titers were relatively low (below 20), consistent with the detection of relatively low affinity and/or low levels of PEG-reactive antibodies in pre- and post-treatment samples in some subjects.

Generally a higher incidence of PEG-reactive antibodies was observed in Period 1 compared to Period 2 regardless of treatment sequence. An analysis of PEG positive subjects shows that there are many subjects who only experienced a single occurrence of PEG positive results (14 subjects). However, there are also many subjects who had PEG positive results for more than three consecutive time points (20 subjects).

Table 24: Treatment-sequence: TR; ADA / mAb results for Period 1 (B12019)

Treatment T: B12019
Treatment R: Neulasta

Statistic *(note 1)*	Pre-dose Period 1	Treatment Day in Period 1				
		8	15	22	29	43 *(note 2)*
No. subjects with valid results	85	85	84	84	84	4
No. subjects with ADA screen positives (%)	12 (14.1%)	34 (40.0%)	42 (50.0%)	34 (40.5%)	36 (42.9%)	1 (25.0%)
No. subjects B12019-reactive (%)	2 (2.4%)	0 (0%)	1 (1.2%)	1 (1.2%)	1 (1.2%)	0 (0%)
No. subjects Neulasta-reactive (%)	2 (2.4%)	0 (0%)	1 (1.2%)	2 (2.4%)	0 (0%)	0 (0%)
No. subjects PEG_{6000}-reactive (%)	3 (3.5%)	5 (5.9%)	12 (14.3%)	11 (13.1%)	10 (11.9%)	1 (25.0%)
No. subjects filgrastim-reactive (%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
No. confirmed negative subjects	9	29	30	22	25	0
No. subjects nAb positive	0	0	0	0	0	0

Note 1: Subjects may have contributed to more than one category due to different signal patterns at different time-points.

Note 2: This visit was performed in Period 1 only in the case that the washout period was longer than 42 days and at discharge from the study. In case of a 42-day washout period this visit corresponds to Day 1 of Period 2. Day 43 was performed in Period 2 for all subjects completing the scheduled treatment.

Table 25: Treatment-sequence: RT; ADA / mAb results for Period 1 (Neulasta)

Treatment T: B12019
Treatment R: Neulasta

Statistic *(note 1)*	Pre-dose Period 1	Treatment Day in Period 1				
		8	15	22	29	43 *(note 2)*
No. subjects with valid results	86	86	86	85	85	4
No. subjects with ADA screen positives (%)	12 (14.0%)	29 (33.7%)	38 (44.2%)	31 (36.5%)	25 (29.4%)	1 (25.0%)
No. subjects B12019-reactive (%)	1 (1.2%)	1 (1.2%)	1 (1.2%)	1 (1.2%)	1 (1.2%)	0 (0%)
No. subjects Neulasta-reactive (%)	1 (1.2%)	0 (0%)	1 (1.2%)	1 (1.2%)	1 (1.2%)	0 (0%)
No. subjects PEG_{6000}-reactive (%)	3 (3.5%)	5 (5.8%)	12 (14.0%)	9 (10.6%)	7 (8.2%)	1 (25.0%)
No. subjects filgrastim-reactive (%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
No. confirmed negative subjects	9	23	26	22	17	0
No. subjects nAb positive	0	0	0	0	0	0

Note 1: Subjects may have contributed to more than one category due to different signal patterns at different time-points.

Note 2: This visit was performed in Period 1 only in the case that the washout period was longer than 42 days and at discharge from the study. In case of a 42-day washout period this visit corresponds to Day 1 of Period 2. Day 43 was performed in Period 2 for all subjects completing the scheduled treatment.

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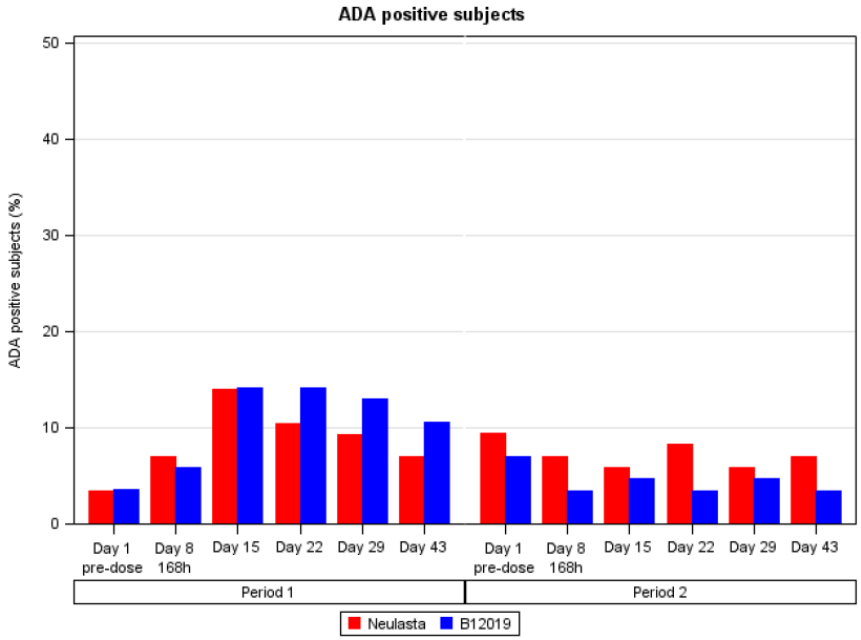

Figure 16: Frequency of ADA-positive subjects by treatment, period and timepoint (Safety set)

The individual profiles over time are presented in Figure 14 to illustrate individuals who had positive results at more than one time point. Figure 14 also lists the individual profiles from subjects that were ADA positive at only one time point. In total, 34 subjects had at least one ADA positive result in at least one of the confirmatory assays.

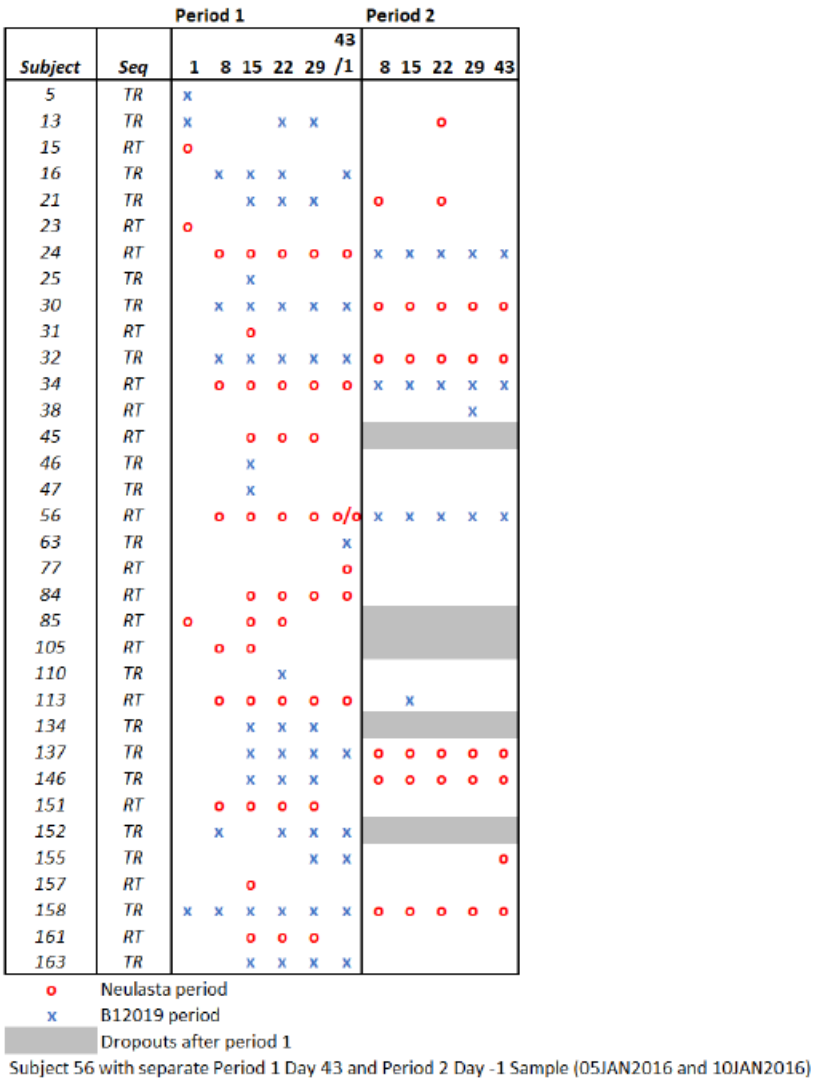

Figure 17: Individual profiles over time for subjects with at least one ADA positive time point

22 out of the 34 subjects were ADA positive at least two consecutive time points; these subject profiles are considered more relevant compared to the profiles of the subjects with only one ADA positive time point. The profiles of the 22 subjects show differing patterns and do not reveal a consistent trend regarding the treatment or treatment sequence. In Period 1, slightly more subjects treated with Pelmeg were ADA positive for at least one time point (Pelmeg: 12 subjects with 46 positive time points; Neulasta: 10 subjects with 39 positive time points). In Period 2, slightly more subjects treated with Neulasta were ADA positive for at least two timepoints (Neulasta: 8 subjects with 29 positive timepoints; Pelmeg: 4 subjects with 16 positive timepoints). Subjects 24, 30, 32, 34, 56, 137, and 158 were consistently positive at all time points from the first ADA positive timepoint until end of study. While subject 158 was already ADA-positive pretreatment, all other subjects where ADA-negative pre-treatment. 6 subjects were consistently ADA-positive during treatment, being balanced across treatment sequences, with 3 subjects in the treatment sequences Neulasta/Pelmeg and 3 subjects in the treatment sequences Pelmeg/Neulasta.

Additionally, a frequency table was generated (Table 28) which shows the number of ADA-positive subjects in Period 1 by the number of consecutive positive time points to assess the immunogenicity after single dosing without crossover.

The number of subjects with at least 4 consecutive ADA-positive time points in Period 1 was balanced between treatments, with 6 subjects after Neulasta and 5 subjects after Pelmeg dosing.

Similar results for the 2 treatments were observed for the number of subjects with at least two consecutive ADA-positive time points in Period 1, with 10 subjects after Neulasta and 12 subjects after Pelmeg dosing.

Table 26: Frequency of ADA-positive subjects in Period 1 by the number of consecutive positive time points

	Period 1, Neulasta (n=86)	Period 1, Pelmeg (n=85)	Period 1, Total (n=171)
Number of subjects with 6 ADA positive consecutive timepoints	1 (1.16%)	1 (1.18%)*	2 (1.17%)
Number of subjects with 5 ADA positive consecutive timepoints	3 (3.49%)	2 (2.35%)	5 (2.92%)
Number of subjects with 4 ADA positive consecutive timepoints	2 (2.33%)	2 (2.35%)	4 (2.34%)
Number of subjects with 3 ADA positive consecutive timepoints	2 (2.33%)	5 (5.88%)	7 (4.09%)
Number of subjects with 2 ADA positive consecutive timepoints	2 (2.33%)	2 (2.35%)	4 (2.34%)

* subject 158 with 6 consecutive ADA positive timepoints was already pre-treatment ADA positive.

Study B12019-102

In this study, the primary immunogenicity endpoint was proportion of ADA positive subjects at the end of Period 2, as detected by a confirmatory assay. The primary immunogenicity analysis was performed on the safety set, defined as all subjects who received at least one dose of study drug. For the primary analysis, proportions of ADA positive subjects at the end of Period 2 were calculated and presented with corresponding 95% CIs per treatment. Furthermore, the difference of proportions of ADA-positive subjects between treatments was calculated and presented with corresponding 95% CIs.

Blood samples for determination of ADAs were collected on Days 1, 8, 15, 22, 29 and 43 of each period.

The analysis was based on 95 subjects, as one subject (020) was excluded from the analysis of ADA frequencies. The respective subject had received an incorrect treatment sequence that was incompatible with the intended study design, i.e. repeated administrations of the same treatment in Period 1 and 2. Two confirmed ADA positive samples were detected in Study B12019-102, both occurring at Day 15 of Period 1. These had a minimal ADA titer, with no filgrastim reactivity and no neutralising capacity.

In Study B12019-102, the 3-mg dose level was chosen to correspond with a dose falling within the linear part of the PD response curve.

Impact of ADAs on pharmacokinetics

The primary PK parameters, $AUC_{0\text{-last}}$ and Cmax, for study B12019-101, were summarized for the ADA-negative and ADA-positive sub-populations (see table below). $AUC_{0\text{-last}}$ and C_{max} were found to be slightly increased (8 to 18%) in the presence of antibodies.

Table 27: Summary of Difference in Primary PK Parameters between ADA Positive vs. ADA Negative Subgroups by Treatment in Study B12019-101 (PK Set Excluding Subjects 118 and 138)

Parameter	Geometric mean					
	Neulasta			Pelmeg		
	ADA negative (N=133)	ADA positive (N=34)	Ratio* ADA pos/neg	ADA negative (N=135)	ADA positive (N=34)	Ratio* ADA pos/neg
	n=129	n=33		n=130	n=34	
$AUC_{0\text{-last}}$	5138.3	5877.6	1.14	4942.4	5588.0	1.13
C_{max}	142.54	168.33	1.18	135.71	146.40	1.08

ADA=anti-drug antibody, $AUC_{0\text{-last}}$=area under the concentration time curve from time zero to last measurable concentration, C_{max}=maximum concentration, N=number of subjects in group, n=number of subjects with data available, PK=pharmacokinetics.
* Ratio was calculated by author.

A summary descriptive statistics for the positive vs. negative PEG-reactive ADA subpopulations for the PK response are shown in the tables below.

For the positive PEG-reactive ADA subjects, the mean Period 2 AUC/Period 1 AUC was 92.5% for the TR (Pelmeg-Neulasta) treatment sequence (n=18 subjects) and 84.2% for the RT (Neulasta-Pelmeg) treatment sequence; the corresponding ratios for the negative PEG-reactive ADA subjects were 104.5% (n=67) and 103.2% (n=70) respectively

Table 28: Descriptive statistics of pegfilgrastim AUC0-last (% Period 2 / Period 1); positive for PEG-reactive ADA

		Sequence		
Variable		TR (N=18)	RT (N=16)	Total (N=34)
Ratio of AUC	n	17	16	33
	Mean	92.5	84.2	88.5
	SD	51.29	52.28	51.13
	CV	55.5	62.1	57.8
	Min	38	19	19
	Median	68.5	75.9	70.5
	Max	213	222	222

Table 29: Descriptive statistics of pegfilgrastim AUC0-last (% Period 2 / Period 1); negative for PEG-reactive ADA

		Sequence		
Variable		TR (N=67)	RT (N=70)	Total (N=137)
Ratio of AUC	n	62	66	128
	Mean	104.5	103.2	103.8
	SD	54.37	74.03	65.00
	CV	52.0	71.7	62.6
	Min	11	2	2
	Median	93.1	92.4	93.1
	Max	275	512	512

In Study B12019-102, only 2 subjects were ADA positive.

Impact of ADAs on pharmacodynamics

In Study B12019-102, only 2 subjects were ADA positive.

In Study B12019-101, antibody formation was primarily directed against the PEG moiety. A slightly lower ANC response is seen in ADA positive subjects (n=34) with both the test and reference product, but overall the mean and geometric mean values for the $AUEC_{0\text{-last}}$ of ANC were comparable for the ADA positive and ADA negative populations in each treatment group, as well as being comparable across treatment groups.

Table 30: Summary Data on Pharmacodynamic Parameters of ANC Values by Treatment and ADA-negative and –positive Subgroups of Study B12019-101 (PD Set)

		Treatment			
		R		T	
Parameter [Unit]	Statistics	Negative (N=133)	Positive (N=34)	Negative (N=133)	Positive (N=34)
AUEC0-last [h*G/L]	n	129	33	132	34
	Mean	7302.3	7158.8	7380.6	6958.4
	SD	1570.33	1618.07	1564.27	1577.17
	CV%	21.5	22.6	21.2	22.7
	Minimum	4473	4376	4323	3682
	Median	7166.2	6951.8	7366.6	6736.9
	Maximum	13887	11187	12606	10609
	Geom. n	129	33	132	34
	Geom. Mean	7142.5	6986.9	7218.8	6786.6
	Geom. CV%	21.3	22.7	21.5	23.2

T: test- Pelmeg, R: reference-Neulasta

Table 31: Summary of Difference in Primary PD Parameter Between ADA Positive vs. ADA Negative Subgroups by Treatment in Study B12019-101 (PD Set)

Parameter	Geometric mean					
	Neulasta			Pelmeg		
	ADA negative N=133	ADA positive N=34	Ratio* ADA pos/neg	ADA negative N=133	ADA positive N=34	Ratio* ADA pos/neg
	n=129	n=33		n=133	n=34	
$AUEC_{0\text{-last}}$	7142.5	6986.9	0.98	7218.8	6786.6	0.94

ADA=anti-drug antibody, ANC=absolute neutrophil count, $AUEC_{0\text{-last}}$=area under the effect time curve from time zero to last measurable concentration, N=number of subjects in group, n=number of subjects with data available, PD=pharmacodynamic. * Ratio was calculated by author. Source: CSR B12019-101, Table 14.2.7.2.

As pre-specified in the study protocol, a sensitivity analysis was conducted in the subgroup of ADA-negative subjects. In this subgroup (N=128), the geometric mean ratio of $AUEC_{0\text{-last}}$ was 100.89% and the corresponding 95% CI was contained in the acceptance interval of 80.00-125.00% (99.16%; 102.65%).

Safety in special populations

All subjects in the conducted studies were male. No differences in AEs due to race between whites and non-whites have been studied. Elderly subjects were not included. Adverse event risk was analysed in the subgroups age, weight, BMI, smoking status, and ADA status. All subgroups were similar with regard to the risk for drug-related TEAEs, indicating that there are no relevant differences in safety between subgroups.

Safety related to drug-drug interactions and other interactions

The applicant did not submit studies relating to drug-drug interactions with Pelmeg (see safety discussion).

Discontinuation due to adverse events

In each study, the majority of subjects completed the study as planned (95.3% and 87.5% in Studies B12019-101 and B12019-102, respectively, based on the safety population). No subject discontinued Study B12019-101 due to an AE. In Study B12019-102, four subjects discontinued due to AEs. Two subjects discontinued due to increased ALT (one each after Pelmeg and Neulasta), one subject discontinued due to lower back pain after the second dose of Pelmeg, and one subject discontinued due to increased blood pressure after the first dose of Neulasta. There was no imbalance between treatments regarding the number of subjects with AEs leading to study discontinuation.

Post marketing experience

There is no post marketing experience with Pelmeg.

2.6.1. Discussion on clinical safety

The safety population (all subjects who had received at least one dose of study drug) in Studies B12019-101 and B12019-102 consisted of 171 and 96 subjects, respectively. Subjects in Study B12019-101 were exposed to two 6 mg doses of study drug (one dose each of Pelmeg and Neulasta). Subjects in Study B12019-102 were exposed to three 3 mg doses of study drug (T-T-R or R-R-T). A total of 259 subjects received at least one dose of Pelmeg (3 mg or 6 mg); 46 of these received two doses. A total of 260 subjects received at least one dose of Neulasta (3 mg or 6 mg); 45 of these received two doses. Although overall safety database is limited, it can be considered sufficient if comparability is demonstrated in terms of PK and PD and if no unexpected AEs are found.

Overall, the most frequent reported TEAEs were back pain (78.3%), headache (43.4%), nasopharyngitis (32.2%), hypoglycaemia (29.2%) and pain in extremity (21.3%).

Generally, the safety profile was in both studies comparable for both treatments apart from a few exceptions discussed below:

Notably higher TEAE frequencies after Pelmeg than after Neulasta were seen in Study B12019-102 for the SOCs “gastrointestinal disorders” (16.7% vs. 4.2%) and “respiratory, thoracic and mediastinal disorders” (10.4% vs 4.2%). The imbalance in these SOCs was mainly driven by subjective measurable symptoms and signs such as e.g. nausea (R: 1.0% vs T: 8.3%) or cough (R: 1% vs T: 6.3%). The imbalances can be considered as chance findings.

Overall, no death occurred during the two studies in healthy volunteers. A total of 4 SAEs were reported in two studies, but were not related to drugs studied. No subject discontinued Study B12019-101 due to an AE. In Study B12019-102, four subjects discontinued due to AEs. There was no imbalance between treatments regarding the number of subjects with AEs leading to study discontinuation.

The majority of subjects in both studies reported at least one TEAE, and in most subjects TEAEs were assessed as related to study drug. The frequencies and pattern of TEAEs were similar between Pelmeg and Neulasta, and in line with the SmPC for Neulasta.

Local tolerability was good in both studies: in the study B12019-101 two subjects (1.2%) had mild, self-limiting, reactions at the injection site after administration of B12019, and in the B12019-102 only 1 subject in each treatment group had mild, self-limiting, reactions at the injection site after study drug administration. Overall, the results do not point to a notably higher frequency of injection site reactions with Pelmeg.
The observed AEs were generally in line with the SmPC for Neulasta, except for the observed AE “hypoglycaemia”. Drug-related hypoglycaemia was reported in around 20% of subjects after administration of Pelmeg and Neulasta. The decrease in serum glucose was comparable between the two treatments. As hypoglycaemia was reversible, not associated with symptoms, and did not require intervention, hypoglycaemia can be considered to be of no clinical relevance.

Therefore, there were no apparent imbalances between Neulasta and Pelmeg treatment regarding the number of subjects with SAEs.
Analyses of glucose results by study period revealed no relevant differences between the two study periods.
There were ADA-positive subjects with AE per treatment and by treatment period in the study B12019-101. Overall, the data are reassuring with similar AE occurring after Pelmeg or after Neulasta in the ADA-positive subjects. No major differences are observed. Moreover, the pattern of AEs in the group of confirmed ADA positive subjects (N=34) was similar to the pattern seen in the overall safety population (N=171). Most common were events in the SOC of musculoskeletal disorders.

There were ADA-positive subjects with AE per treatment and by treatment period in studies B12019-101 and B12019-102. Overall, the data are reassuring with similar AE occurring after Pelmeg or after Neulasta in the ADA-positive subjects, and the AEs seen in ADA positive subjects were representative of AEs seen in the overall population.

Immunogenicity

Overall, a heterogeneous pattern of ADA-positive time points across subjects was observed. In Study B12019-101, about 20% had ADA-positive samples, mainly due to PEG-reactive antibody signals. No antibodies against filgrastim or neutralising antibodies were detected for any of the 2 treatments. In Study B12019-102 two confirmed ADA positive samples were detected. These had no filgrastim reactivity and no neutralising capacity. No consistent trends regarding treatment or treatment sequence were detected. Based on the individual profiles and additional evaluations, no consistent trends towards longer-lasting ADA-positive subjects after Pelmeg treatment could be observed.

Although the immunogenicity is apparently low and no significant ADA emerged whatever the sequence tested in the study (Pelmeg-Pelmeg-Neulasta or Neulasta-Neulasta-Pelmeg), formally long-term immunogenicity could not be assessed as the follow-up period was maximum 18 weeks and the confounding effects of a cross-over design. Therefore, the proposed approach for post-marketing collection of immunogenicity data (collection only for individual requests from physicians and reported via the standard pharmacovigilance reporting lines) is considered acceptable and is agreed by PRAC.

From the safety database all the adverse reactions reported in clinical trials have been included in the Summary of Product Characteristics.

2.6.2. Conclusions on the clinical safety

No unexpected safety signals were observed with Pelmeg and no death occurred during the two studies in healthy volunteers.

The safety profile of Pelmeg in both studies was comparable for both treatments and with the safety of Neulasta. As to immunogenicity, no meaningful differences were observed across treatment sequences. Generally, the observed AEs were in line with the SmPC for Neulasta.

Therefore, the safety data overall support the biosimilarity of Pelmeg and reference product EU-Neulasta.

2.7. Risk Management Plan

Safety concerns

Summary of safety concerns	
Important identified risks	Severe splenomegaly/splenic rupture Cutaneous vasculitis Sweet's syndrome Anaphylactic reactions Capillary leak syndrome Serious pulmonary events including interstitial pneumonia and ARDS Sickle cells crisis in patients with sickle cell disease Musculoskeletal pain-related symptoms Leukocytosis

Summary of safety concerns	
	Thrombocytopenia Glomerulonephritis
Important potential risks	AML/MDS Cytokine release syndrome Medication errors including overdose Drug interaction with lithium Off-label use Immunogenicity (incidence and clinical implications of anti-G-CSF antibodies) Extramedullary haematopoiesis
Missing information	Use in children and adolescents under 18 years of age Use during pregnancy and lactation

Pharmacovigilance plan

There is no planned or ongoing additional study in the pharmacovigilance plan.

Routine pharmacovigilance activities are sufficient to address the safety concerns of this medicinal product.

Risk minimisation measures

Safety concern	Risk minimisation measures	Pharmacovigilance activities
Important identified risks		
Severe splenomegaly/splenic rupture	Routine risk minimisation measures: SmPC sections 4.4 and 4.8 PL section 2 Prescription only medicine	Routine pharmacovigilance
Cutaneous vasculitis	Routine risk minimisation measures: SmPC section 4.8 PL section 4 Prescription only medicine	Routine pharmacovigilance
Sweet's Syndrome	Routine risk minimisation measures: SmPC section 4.8 PL section 4 Prescription only medicine	Routine pharmacovigilance
Anaphylactic reactions	Routine risk minimisation measures: SmPC sections 4.3, 4.4, and 4.8 PL sections 2 and 4 Prescription only medicine	Routine pharmacovigilance
Capillary leak syndrome	Routine risk minimisation measures: SmPC sections 4.4 and 4.8	Routine pharmacovigilance activities beyond adverse reactions reporting and signal

Safety concern	Risk minimisation measures	Pharmacovigilance activities
	PL section 2 Prescription only medicine	detection: Adverse event of special interest follow-up form
Serious pulmonary events including interstitial pneumonia and ARDS	Routine risk minimisation measures: SmPC section 4.4 PL section 2 Prescription only medicine	Routine pharmacovigilance
Sickle cells crisis in patients with sickle cell disease	Routine risk minimisation measures: SmPC sections 4.4 and 4.8 PL sections 2 and 4 Prescription only medicine	Routine pharmacovigilance
Musculoskeletal pain-related symptoms	Routine risk minimisation measures: SmPC section 4.8 PL section 4 Prescription only medicine	Routine pharmacovigilance
Leukocytosis	Routine risk minimisation measures: SmPC sections 4.4 and 4.8 PL sections 2 and 4 Prescription only medicine	Routine pharmacovigilance
Thrombocytopenia	Routine risk minimisation measures: SmPC sections 4.4 and 4.8 PL sections 2 and 4 Prescription only medicine	Routine pharmacovigilance
Glomerulonephritis	Routine risk minimisation measures: SmPC sections 4.4 and 4.8 PL sections 2 and 4 Prescription only medicine	Routine pharmacovigilance
Important potential risks		
AML/MDS	Routine risk minimisation measures: SmPC sections 4.1 and 4.4 PL section 2 Prescription only medicine	Routine pharmacovigilance
Cytokine release syndrome	Routine risk minimisation measures: Prescription only medicine	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Adverse event of special interest follow-up form
Medication errors including overdose	Routine risk minimisation measures:	Routine pharmacovigilance activities beyond adverse

Safety concern	Risk minimisation measures	Pharmacovigilance activities
	SmPC sections 1, 2, 4.2, 4.5, and 4.9 PL section 3 Prescription only medicine	reactions reporting and signal detection: Adverse event of special interest follow-up form
Drug interaction with lithium	Routine risk minimisation measures: SmPC section 4.5 PL Section 2 Prescription only medicine	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Adverse event of special interest follow-up form
Off-label use	Routine risk minimisation measures: SmPC sections 4.1 and 4.4 PL sections 1, 2, and 3 Prescription only medicine	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Adverse event of special interest follow-up form
Immunogenicity (incidence and clinical implications of anti-G-CSF antibodies)	Routine risk minimisation measures: SmPC section 4.4 PL section 2 Prescription only medicine	Routine pharmacovigilance activities beyond adverse reactions reporting and signal detection: Availability of voluntary antibody testing
Extramedullary haematopoiesis	Routine risk minimisation measures: Prescription only medicine	Routine pharmacovigilance
Missing information		
Use in children and adolescents under 18 years of age	Routine risk minimisation measures: SmPC sections 4.2, 4.8, 5.1, and 5.2 PL section 3 Prescription only medicine	Routine pharmacovigilance
Use in pregnancy and/or lactation	Routine risk minimisation measures: SmPC sections 4.6 and 5.3 PL section 2 Prescription only medicine	Routine pharmacovigilance

Routine risk minimisation measures are considered sufficient to minimise the safety concerns of this medicinal product.

Conclusion

The CHMP and PRAC considered that the risk management plan version 0.4 is acceptable.

2.8. Pharmacovigilance

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.

Periodic Safety Update Reports submission requirements

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.

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.*

2.9.2. Quick Response (QR) code

A request to include a QR code in the package leaflet for the purpose of presenting statutory information has been submitted by the applicant and has been found acceptable.

3. Biosimilarity assessment

3.1. Comparability exercise and indications claimed

The claimed indication is identical to the reference product Neulasta: “Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes)”. Clinical studies supporting the application were carried out in healthy volunteers as part of the biosimilarity exercise. The clinical programme of Pelmeg comprised of two studies conducted in male healthy subjects.

Clinical comparability of Pelmeg and Neulasta was established in a pivotal pharmacokinetic (PK)/ pharmacodynamic (PD) study using the approved 6 mg dose. The study B12019-101 (n=172) was a single-dose, randomised, double-blind, two-stage, two-way cross-over study, which assessed PK and PD as co-primary endpoints.

The supportive study B12019-102 (n=96) was a multiple-dose, randomised, double-blind, three-period, two-sequence crossover study to assess the immunogenicity and PD comparability of Pelmeg and Neulasta using a non-therapeutic, lower dose of 3 mg.

3.2. Results supporting biosimilarity

Quality:

For the biosimilarity analysis, the applicant has performed an extensive comparability exercise which included several batches of EU Neulasta and several batches of Pelmeg DP (including both process validation and clinical batches). All quality attributes analysed proved to be highly similar between Pelmeg and EU Neulasta. For a few purity parameters slight differences were observed. However, these differences were very small and are not believed to have any impact on the quality of the product. As such, they do not preclude a conclusion on biosimilarity. The primary structure of Pelmeg was found to be identical to that of the RMP, with identical site of PEGylation for both products. Molecular weight and polydispersity indicated similar PEG moieties between Pelmeg and the RMP. Moreover, the higher order structure, product-related variants, and the impurity and aggregation profiles, were also shown to be similar between Pelmeg and the RMP. Furthermore, relative potency and recombinant human G-CSF receptor binding kinetics were similar for Pelmeg and the RMP. Comparative stability testing demonstrated that Pelmeg and the RMP degrade in a comparable manner. In conclusion, the data of the physicochemical, bio-functional, and stability tests confirm that from a quality point of view Pelmeg could be considered as biosimilar to EU Neulasta.

Non-clinical:

- The non-clinical biosimilarity program comprised comparative assessment of in vitro PD effects as well as an in vivo PD/PK study. Biosimilarity of Pelmeg and Neulasta was demonstrated by comparative assessment of the binding affinity to the G-CSF receptor as well as of the potency to stimulate proliferation of myeloblastic cells. The comparative PD/PK study in naïve and neutropenic rats, although small group sizes, is considered supportive.

Clinical:

- Pharmacokinetics: the primary and secondary PK parameters $AUC_{0\text{-last}}$, $AUC_{0\text{-inf}}$ and Cmax were assessed for bioequivalence between Pelmeg and Neulasta in study B12019 101
    - Biosimilarity in pharmacokinetics of Pelmeg with EU-authorized Neulasta was demonstrated in the pivotal PK/PD study B12019-101 in healthy male volunteers, as the 94.32% CIs for the ratio of the test and reference product geometric means for the primary and secondary PK parameters AUC0-last, AUC0-inf and Cmax were fully contained within the standard BE acceptance interval of 80.00-125.00%.
    - Study B12019-102 was not intended to evaluate PK equivalence between Pelmeg and Neulasta. Although PK sampling was sparse, similar PK profiles were observed that can be considered as supportive for PK similarity between Pelmeg and Neulasta.
- Pharmacodynamics: ANC and CD34+ were investigated in both pharmacological studies in altogether 260 healthy volunteers (PD set) in a cross-over design.
    - In study B12019 101 the GMR of the primary endpoint ANC $AUEC_{0\text{-last}}$ was 100.20%; the 95% CI with 99.16; 102.65 was well within the acceptance interval of 80.00-125.00%. The secondary endpoints ANC E_{max} and t_{max}, as well as CD34+ count which were only presented descriptively, were comparable.
    - In study B12019 102, where a lower dose of 3 mg was used in a 3 period design to assess immunogenicity and PD, comparability between Pelmeg and Neulasta was shown. The GMR of the primary endpoint ANC AUEC0-last was 101.59 with a 95% CI of 99.58%; 103.63%. Tighter similarity limits of 90-111% were defined post-hoc, which

were easily met as well. The descriptively presented secondary PD endpoints Emax and tmax as well as CD34+ cell count support biosimilarity.

- No major safety findings were detected in the clinical part of the biosimilarity program or in terms of immunogenicity:
    - Safety was comparable in both studies for both treatments apart from minor differences. No unexpected safety signals were observed and no death occurred during the two studies in healthy volunteers. The incidence and severity of AEs were comparable between Pelmeg and Neulasta in general in the conducted studies in healthy volunteers. The safety profiles observed were in general in line with the Product Information for Neulasta.
    - Incidence rates of ADAs were equally distributed across treatments and treatment sequences in both studies.
    - Across the two clinical studies, no confirmed filgrastim-reactive ADAs or neutralising antibodies were observed.
    - Subjects classified as ADA-positive showed no meaningful differences in PD or AE as compared to ADA-negative subjects.

3.3. Uncertainties and limitations about biosimilarity

There are no remaining uncertainties and limitations that have an impact on the conclusion of biosimilarity of Pelmeg and Neulasta.

3.4. Discussion on biosimilarity

For a biosimilar, the benefit-risk balance is derived from the reference product provided the totality of evidence collected from the quality, non-clinical and clinical data package that support the comparability of both products.

From a quality perspective, the critical physico-chemical and functional attributes have demonstrated that Pelmeg is highly similar to its reference product EU Neulasta. A few minor differences were observed, but these are negligible and have no impact on the quality of the product or on the conclusion of biosimilarity (at quality level).

From a non-clinical perspective, the requirements for biosimilarity assessment have been met and sufficient evidence for the demonstration of biosimilarity has been provided.

The clinical biosimilarity program consisted of comparative PK, PD, immunogenicity and safety analyses in healthy male volunteers. The studies included only healthy volunteers, which are considered a homogenous and sensitive population to assess the primary objectives of the clinical studies and were in agreement with the scientific advice provided. Biosimilarity between Pelmeg and the reference product Neulasta was demonstrated in the clinical studies, as the primary PK and PD endpoints GMR of $AUC_{0\text{-last}}$, C_{max} and ANC $AUEC_{0\text{-last}}$ including their confidence intervals met the acceptance criteria. The secondary endpoints are also supportive of the biosimilarity exercise.

The safety of Pelmeg was consider similar to Neulasta in both studies, no unexpected safety signals were reported and the incidence rates of ADAs were equally distributed across treatments and treatment sequences in both studies.

Therefore, considering the totality of the evidence on the quality, non-clinical and clinical data, biosimilarity of Pelmeg with the reference product EU Neulasta can be concluded.

3.5. Extrapolation of safety and efficacy

The claimed indication is the only indication currently approved for EU-Neulasta ("Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy [with the exception of chronic myeloid leukaemia and myelodysplastic syndromes"]).

Therefore no extrapolation to other indications is needed for this biosimilar application.

3.6. Additional considerations

Not applicable.

3.7. Conclusions on biosimilarity and benefit risk balance

Based on the review of the submitted data, Pelmeg is considered biosimilar to Neulasta. Therefore, a benefit/risk balance comparable to the reference product can be concluded.

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 Pelmeg is favourable in the following indication:

Reduction in the duration of neutropenia and the incidence of febrile neutropenia in adult patients treated with cytotoxic chemotherapy for malignancy (with the exception of chronic myeloid leukaemia and myelodysplastic syndromes).

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 restricted medical prescription (see Annex I: Summary of Product Characteristics, section 4.2).

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.

Conditions or restrictions with regard to the safe and effective use of the medicinal product

Risk Management Plan (RMP)

The 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.

Conditions or restrictions with regard to the safe and effective use of the medicinal product to be implemented by the Member States

Not applicable