13 December 2018
EMA/11025/2019
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Besremi

International non-proprietary name: ropeginterferon alfa-2b

Procedure No. EMEA/H/C/004128/0000

Note

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

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List of abbreviations

ADME	absorption, distribution, metabolism, and excretion
ADR	Adverse drug reaction
AE	Adverse event
ALT	Alanine aminotransferase
AS	Active substance
AST	Aspartate aminotransferase
AUC	Area under the curve
CE	European conformity
CHMP	Committee for Medicinal Products for Human Use
CHR	Complete Hematologic Response
CL	Clearance
Cmax	Maximum Serum Concentration
Cmin	Minimum Serum Concentration
CPE	Cytopathic effect
CPP	Critical process parameters
CQA	Critical quality attributes
CSR	Clinical Study Report
CT	Computed Tomography
CTCAE	Common terminology criteria for adverse events
DF	Diafiltration
ECG	Electrocardiogram
eCRF	Electronic case report form
EOT	End-of-treatment (Visit)
EPC	End-of-production cell
EudraCT	Eurpean Union Drug Regulating Authorities Clinical Trials
ELISA	Enzyme-linked immunosorbent assay
EU	European Union
FAS	Full analysis set
FP	Finished product
GCP	Good Clinical Practice
GGT	Gamma-gluamyl-transpeptidase
GMP	Good manufacturing practice
HCP	Host cell proteins
Hct	Haematocrit
Hgb	haemoglobin
HCV	Hepatitis C Virus
HIV	Human Immunodeficiency Virus
HMW	High-molecular weight
HU	Hydroxyurea
ICH	International Conference on Harmonization
IEF	Isoelectric focusing
IFN	Interferon
i.m.	Intramuscular
IPC	In process control
IEX-HPLC	Ion-exchange high performance liquid chromatography
IPTG	Isopropyl β-D-1-thiogalactopyranoside
ISO	International Organization for Standardization
i.v.	Intravenous
JAK2	Janus-kinase 2
L	Litre
LC-MS	Liquid chromatography-mass spectrometry analysis
LDH	Lactate dehydrogenase
LOQ	Limits of quantitation
LVEF	Left Ventricular Ejection Fraction
MedDRA	Medical Dictionary for Regulatory Activities

MCB	Master cell bank
MRI	Magnetic Resonance Imaging
mPEG	Methoxypolyethylene glycol
MPN	Myeloproliferative neoplasm
MTD	Maximum tolerated dose
MUGA	Multigated Acquisition Scan
NIBSC	National Institute for Biological Standards and Control (UK)
PEG	Polyethylene glycol
PD	Pharmacodynamic
Ph. Eur.	European Pharmacopoeia
PK	Pharmacokinetic
PLT	Platelets
PPS	Per protocol set
PTFE	Polytetrafluoroethylene
PTs	Preferred terms
PV	Polycythaemia Vera
QA	Quality attributes
QP	Qualified Person
RBC	Erythrocyte
RP-HPLC	Reverse-phase high performance liquid chromatography
SAE	Serious adverse event
SAP	Statistical Analysis Plan
s.c.	Subcutaneous
SDS-PAGE	Sodium dodecyl sulfate polyacrylamide gel electrophoresis
SE-HPLC	Size-exclusion high performance liquid chromatography
SmPC	Summary of Product Characteristics
SOCs	System organ classes
SPR	Surface plasmon resonance
TEAE	Treatment emergent adverse event
TPOAb	Thyroid peroxidase autoantibody
TSE	Transmissible spongiform encephalopathy
UF	Ultrafiltration
ULN	Upper limit of normal
UV	Ultra-violet
Vd	Volume of distribution
WCB	Working cell bank
WBC	Leukocyte
WHO	World Health Organization

1. Background information on the procedure

1.1. Submission of the dossier

The applicant AOP Orphan Pharmaceuticals AG submitted on 2 February 2017 an application for marketing authorisation to the European Medicines Agency (EMA) for Besremi, through the centralised procedure falling within the Article 3(1) and point 4 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 20 November 2014.

Besremi was designated as an orphan medicinal product EU/3/11/932 on 9 December 2011 in the following condition: Treatment of polycythemia vera.

Following the CHMP positive opinion on this marketing authorisation and at the time of the review of the orphan designation by the Committee for Orphan Medicinal Products (COMP), this product was withdrawn from the Community Register of designated orphan medicinal products on 18 December 2018 on request of the sponsor. The relevant Withdrawal assessment report – Orphan maintenance can be found on Agency's website at ema.europa.eu/en/medicines/human/EPAR/besremi under 'Assessment history' tab.

The applicant applied for the following indication: Besremi is indicated in adults for the treatment of polycythaemia vera without symptomatic splenomegaly.

The legal basis for this application refers to:

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

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

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision(s) P/0178/2013 on the granting of a product-specific waiver.

Information relating to orphan market exclusivity

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.

Applicant's request for consideration

New active Substance status

The applicant requested the active substance ropeginterferon alfa-2b contained in the above medicinal product to be considered as a new active substance, as the applicant claims that it is not a constituent of a medicinal

product previously authorised within the European Union.

The applicant also requested the active substance ropeginterferon alfa-2b contained in the above medicinal product to be considered as a new active substance in comparison to PEGylated-IFN alpha 2b previously authorised in the European Union, as the applicant claimed that ropeginterferon alfa-2b differs significantly in properties with regard to safety and/or efficacy from the already authorised active substance.

Protocol assistance

The applicant received Protocol assistance from the CHMP:

Scientific advice	**date**	**Area**
EMEA/H/SA/2272/1/2012/PA/III	15 March 2012	quality, non-clinical and clinical
EMEA/H/SA/2272/1/FU/1/2012/PA/III	17 January 2013	quality and clinical
EMEA/H/SA/2272/1/FU/2/2014/PA/III	22 May 2014	quality and clinical
EMEA/H/SA/2272/1/FU/3/2015/PA/II	25 February 2016	clinical

1.2. Steps taken for the assessment of the product

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

Rapporteur: Harald Enzmann Co-Rapporteur: Alexandre Moreau

The application was received by the EMA on	2 February 2017
The procedure started on	23 February 2017
The Rapporteur's first Assessment Report was circulated to all CHMP members on	15 May 2017
The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on	17 May 2017
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	29 May 2017
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	N/A
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	22 June 2017
The applicant submitted the responses to the CHMP consolidated List of Questions on	25 May 2018
The following GMP and GCP inspection(s) were requested by the CHMP and their outcome taken into consideration as part of the	

Quality/Safety/Efficacy assessment of the product: – A GCP inspection at 2 Clinical Investigators in Austria and Poland and the sponsor in Austria between 26 June 2017 and 18 August 17. The outcome of the inspection carried out was issued on – A GMP inspection at two sites located in Taiwan involved in the manufacture of the active substance between 18-20/09/2017 and on 22/09/2017. – A GMP inspection at one site located in Taiwan involved in the testing of the active substance on 21/09/2017 and between 23-24/07/18.	Inspection report: 2 October 17 15 January 2018 and 14 June 2018 03 October 2018
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Questions to all CHMP members on	10 July 2018
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	12 July 2018
The CHMP agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	26 July 2018
The applicant submitted the responses to the CHMP List of Outstanding Issues on	15 October 2018
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP members on	5 November 2018
SAG experts were convened to address questions raised by the CHMP on The CHMP considered the views of the SAG as presented in the minutes of this meeting.	07 November 2018 15 November 2018
The CHMP agreed on a 2nd list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	15 November 2018
The applicant submitted the responses to the CHMP List of Outstanding Issues on	28 November 2018
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP members on	5 December 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 Besremi on	13 December 2018

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

Polycythaemia vera (PV) is a myeloproliferative neoplasm (MPN) exhibiting terminal myeloid cell expansion in the peripheral blood, resulting in various combinations of erythrocytosis, leukocytosis, thrombocytosis, bone marrow hypercellularity/fibrosis, and splenomegaly. PV is an acquired form of primary erythrocytosis which is characterised by an excess production of erythrocytes, leukocytes and platelets. Erythrocytosis is defined as an increase in haematocrit or packed red cell volume caused by different pathological conditions. The term "polycythaemia" is often used synonymously with "erythrocytosis", since leukocytes and platelets are present in blood in far smaller proportions than erythrocytes.

2.1.2. Epidemiology

PV is a very rare disease (although being the most common of the MPNs) and typically develops in late adulthood. In a 2006 review of the literature, Johansson and colleagues, as well as the latest Orphanet Report (as of June 2017) reported a prevalence of PV of approximately 3.0 cases per 10,000 population.

2.1.3. Biologic features, Aetiology and pathogenesis

Two key aspects identify the biology of PV: clonality and erythropoietin (Epo) independence (Medscape, 2016). In PV, a single clonal population of erythrocytes, granulocytes, platelets, and variable clonal B cells arises when a haematopoietic stem cell gains a proliferative advantage over other stem cells. The T lymphocytes and natural killer cells remain polyclonal in PV; this is related to their longevity.

Erythropoietin-independence is the ability of erythroid colonies formed from the PV haematopoietic stem cell to grow without erythropoietin. Although the colonies do not require erythropoietin, they remain responsive to it, and the erythropoietin receptor (EpoR) is normal without defects in function or quantity. Experiments using antibodies to neutralise Epo or block the EpoR do not abolish erythropoietin–independent erythroid colony formation.

Significant progress in the elucidation of the underlying molecular mechanism of PV was made with the discovery of a gain of function mutation in the tyrosine kinase Janus-activated kinase-2 (JAK2V617F), which now appears to cause most primary PV cases in adults (McLornan et al.,2006). JAK2V617F is detectable in more than 95% of patients diagnosed with PV (Tefferi et al., 2007; Quintas-Cardama et al., 2009). Several other mutations of JAK2 have since been described (e.g., exon 12, JAK2H538-K539delinsI) (Cario et al., 2003c; Scott et al., 2007b). The JAK2 mutations cause the enzyme to be constitutively active. This leads to an "always on" signal for cell propagation, inducing a massive overproduction of cell lines that express Epo receptors by both cytokine independent proliferation and cytokine hypersensitivity (Tefferi, 2007).

2.1.4. Clinical presentation, diagnosis and stage / prognosis

The key clinical features of PV are highly variable and may include increased red blood cell mass (haematocrit), increased white blood cell (leucocyte) and platelet counts in the peripheral blood, and splenomegaly in advanced disease stages, or any combination of these.

Mild clinical symptoms of PV are unspecific and related to the increased blood cell count resulting in high blood viscosity, e.g. headache, fatigue, dizziness, vision disturbances, vertigo, tinnitus, pruritus, or erythromelalgia.

However, PV is a long-term debilitating and life-threatening condition as it is associated with the risk of thrombosis, haemorrhage and a long term propensity to develop myelofibrosis (MF) and secondary acute myeloid leukaemia (AML) (Griesshammer et al., 2015; Stein et al., 2015).

Since the clinical symptoms are very unspecific, the diagnosis of PV is primarily based on laboratory parameters like increased haemoglobin or haematocrit values and the presence of the two most common JAK2 mutations [about 90-95% of all patients suffering from PV show either a JAK2V617F or JAK2 exon 12 mutation (Tefferi et al, 2007)].

As erythrocytosis may also be a reaction to a natural or artificial increase in the production of erythropoietin, PV has to be distinguished from so-called secondary erythrocytosis.

Table 1: 2016 WHO diagnostic criteria for PV (Arber et al., 2016)

Major criteria	1) Blood count: • Hgb > 16.5 g/dL (men), > 16.0 g/dL (women) or • Hct > 49% (men), >48% (women) or • increased red cell mass (RCM)* 2) BM biopsy showing hypercellularity for age with trilineage growth (panmyelosis) including prominent erythroid, granulocytic, and megakaryocytic proliferation with pleomorphic, mature megakaryocytes (differences in size) 3) Presence of JAK2V617F or JAK2 exon 12 mutation
Minor criterion	Subnormal serum erythropoietin (Epo) level
Diagnosis of PV requires meeting either all 3 major criteria, or the first 2 major criteria and the minor criterion	

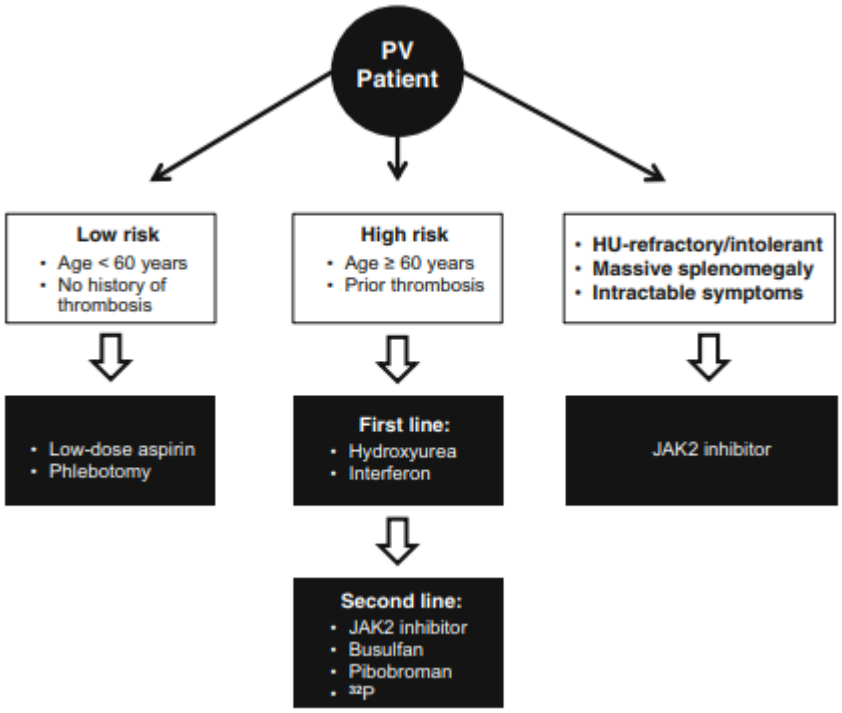
* More than 25% above mean normal predicted value.
†Criterion number 2 (BM biopsy) may not be required in cases with sustained absolute erythrocytosis: hemoglobin levels >18.5 g/dL in men (hematocrit, 55.5%) or >16.5 g/dL in women (hematocrit, 49.5%) if major criterion 3 and the minor criterion are present. However, initial myelofibrosis (present in up to 20% of patients) can only be detected by performing a BM biopsy; this finding may predict a more rapid progression to overt myelofibrosis (post-PV MF).

The median survival of untreated symptomatic patients with PV is 6 to 18 months, whereas survival of treated patients is 10 years or more. Accordingly, all patients with symptomatic PV should be treated.

2.1.5. Management

While the WHO guideline appears to be the most recent, internationally-accepted standard for the diagnosis of PV and other MPNs, there is no international body that has written universally accepted treatment guidelines for PV. The therapeutic goal for PV patients is to alleviate symptoms, reduce the risk of cardiovascular events and decrease and/or minimize the risk of progression to MF, MDS or acute leukaemia. Currently available treatment options based on a proposed algorithm is illustrated in Figure 1 below (Griesshammer et al 2015).

Figure 1: Treatment options based on a proposed algorithm



In the initial phase of the disease, phlebotomy is the cornerstone of treatment with the objective of maintaining haematocrit values below 45%, a cut-off that has been shown to be associated with a lower risk of cardiovascular death and major thrombosis. As PV patients are at a high risk of thrombosis, phlebotomy is often accompanied by low dose aspirin. In a study by the European Collaboration on Low-dose Aspirin in Polycythaemia Vera (ECLAP), the administration of low dose aspirin was shown to significantly reduce the risk of non-fatal myocardial infarction, non-fatal stroke, pulmonary embolism, major venous thrombosis, or death from cardiovascular causes when compared to placebo.

While phlebotomy and low-dose aspirin are accepted as standard of care for the initial therapy of PV patients, cytoreductive therapy is recommended in patients with persistent haematological abnormalities, clinical symptoms, poor compliance with or intolerance of phlebotomy, and those at a high risk of thrombosis. Most PV patients require cytoreductive therapy during the course of their disease.

Hydroxyurea (HU), although not approved in all European countries for the use in the treatment of polycythaemia vera, is often the first-line cytoreductive therapy used in patients with polycythaemia vera. However, hydroxyurea-related toxicities often require either drug reduction or drug discontinuation resulting in inadequate management of the disease. Furthermore due of its mechanism of action HU has the potential to be mutagenic and it is controversially discussed whether HU is associated with an increased risk of leukemic transformation after long-term use in PV patients.

Since 2015 the JAK2-inhibitor ruxolitinib (Jakavi) is approved in the European Union “for the treatment of adult patients with polycythaemia vera who are resistant to or intolerant of hydroxyurea”.

Additional antithrombotic medications are used off-label in the therapy of PV and include non-pegylated and pegylated interferons, anagrelide and alkylating agents (e.g. busulfan).

Interferons (IFNs) are cytokines with a wide range of biologic properties, including antiviral, immune-modulating, anti-proliferative and cellular differentiating effects. Pharmacologically, the binding of IFNα to type I IFN receptors induces a conformational change in the intracellular domain of the receptor molecules resulting in activation of JAK and tyrosine kinase proteins, phosphorylation of signal transducer and activator of transcription (STAT) proteins. Phosphorylated STAT proteins translocate into the nucleus and induce the transcription of IFN-stimulated genes within the interferon stimulated response element, which causes various effects in the immune system such as modulation of immunoglobulin production, inhibition of T-cell cytotoxicity, monocytes / macrophage function and natural killer cell activity (George et al., 2012).

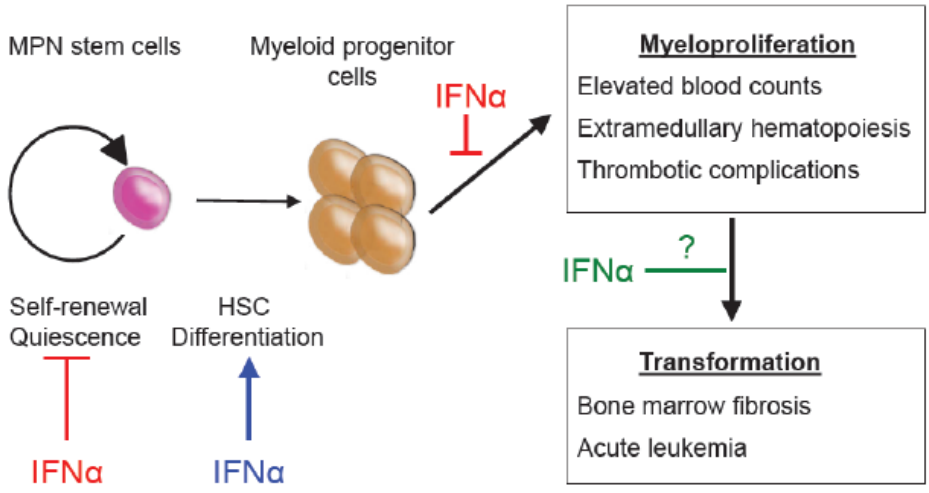
The exact way IFNα works in Polycythaemia Vera (PV) is not fully understood, but it has been shown that it suppresses the proliferation of haematopoietic progenitors, both pluripotent and lineage-committed. It is suggested that IFNα has direct effects on MPN stem cells leading to

- depletion of long-term hematopoietic stem cells,
- exit from quiescence and
- enforced terminal differentiation.

Additionally, IFNα may have direct effects on downstream effector cells leading to reduction in blood counts and extramedullary haematopoiesis. The effects of IFNα on preventing fibrotic and/or leukemic transformation have not yet been defined (Lane and Mullally, 2013). Dose-dependent suppression of both normal (Epo-stimulated) and clonal (neoplastic) haematopoietic progenitors has been demonstrated in vivo and in vitro, and there is evidence of a selective action on clonal haematopoietic progenitor cells (Elliott and Tefferi, 1997).

An overview of known and suspected IFNα mechanism of actions on MPN stem cells, haematopoietic progenitor cells, and the related disease initiation and propagation is illustrated in Figure 2 below.

Figure 2: MPN stem cells are responsible for disease initiation and propagation in vivo



About the product

Besremi contains ropeginterferon alfa-2b as active ingredient, which is a PEGylated, recombinant human interferon alpha-2b, which is modified by an additional proline residue at the N-terminus.

A product specific waiver for ropeginterferon alfa-2b for all subsets of the paediatric population and the condition PV was granted by EMA in 2013 (P/0178/2013).

Type of Application and aspects on development

Orphan Designation (OD) was granted by the European Commission to AOP for ropeginterferon alfa-2b for the treatment of PV on 9 December 2011 (EU/3/11/932) (EMA, 2011).

The OD was based on the significant benefit of the product compared to other medications for PV licensed in EU countries, i.e. the evidence that ropeginterferon alfa-2b may effect haematological remission of the malignant clone in PV, without being associated with an increased leukaemogenic risk.

2.2. Quality aspects

2.2.1. Introduction

The finished product (FP) is presented as a solution for injection containing 250 micrograms/0.5 ml or 500 micrograms/0.5 ml of ropeginterferon alfa-2b (INN) as active substance (AS).

Other ingredients are: sodium chloride, sodium acetate (anhydrous), acetic acid glacial, benzyl alcohol, polysorbate 80 and water for injections.

The product is available as an integrated medicinal product in a pre-filled pen as described in section 6.5 of the SmPC, which may be used up to two times, dependent on the prescribed dose. The pen is not refillable.

2.2.2. Active Substance

General information

The molecular mass of ropeginterferon alfa-2b is approximately 60 kilodaltons (kDa). Ropeginterferon alfa-2b is a covalent conjugate of a recombinant proline-interferon alfa-2b and a two-arm methoxypolyethylene glycol (mPEG) moiety. Pro-IFN alfa-2b, produced in *Escherichia coli,* is an approximately 19 kDa non-glycosylated polypeptide of 166 amino acids; a 40 kDa two-arm PEG moiety attaches to its N-terminal proline.

Ropeginterferon alfa-2b contains two pairs of disulfide bonds (cys-2 pairing with cys-99 and cys-30 pairing with cys-139) as those found in other interferon alpha-2b products. The secondary and tertiary structures elucidated by biophysical characterisation suggest ropeginterferon alfa-2b a monomeric globular protein with a high content of alpha helices.

Manufacture, characterisation and process controls

Active substance manufacturing takes place at PharmaEssentia Corporation, Taichung Plant, Taiwan.

During the procedure, a major objection was raised to request GMP certificates for the AS manufacturing site and a specified AS testing site. Satisfactory certificates were subsequently submitted during the procedure.

Description of manufacturing process and process controls

The process is divided into upstream process, downstream process-1 (proline-interferon alfa-2b AS intermediate purification), synthesis of the 40 kDa PEG intermediate and downstream process-2 (ropeginterferon alfa-2b purification)

The process starts with fermentation in *E. coli*. The product is expressed as an intracellular protein in the form of inclusion bodies. The product is extracted by lysing the cells followed by washing with buffers, and then solubilizing the inclusion bodies that contain the product. Downstream processing steps include protein refolding and chromatography and ultrafiltration/diafiltration purification steps to produce one proline-interferon alfa-2b intermediate batch. This intermediate is stored in a specified Ph.Eur. compliant container (see control of critical steps and intermediates).

Proline-interferon alfa-2b is then PEGylated by attaching a 40 kDa two-arm branched PEG intermediate to the N-terminal proline. It is then further purified via chromatography and UF/DF steps to produce the formulated active substance.

A typical batch size is specified. No reprocessing is claimed.

The description of the active substance manufacturing process is considered acceptable. The routine in-process controls (IPC) along with acceptance criteria, including controls for microbial purity and endotoxin, are described for each step. These are acceptable however, as an additional specified IPC during the early stages of purification is recommended when data from a stipulated number of batches are available (see recommendation).

The active substance solution is stored in a Ph. Eur.-compliant container and the transportation is performed in a temperature-controlled carrier to maintain the quality.

Control of materials

The generation of the cell substrate is well described. The cell substrate for expression of the recombinant protein is based on *E. coli* strain BLR (DE3). Construction of the expression plasmid is documented and the gene encoding Pro-IFN alfa-2b was cloned by polymerase chain reaction (PCR).

A two tiered cell bank system consisting of master cell bank (MCB) and working cell bank (WCB) has been established; characterisation of the cell banks is considered adequate and includes testing for identity, purity, viability and genetic characterisation. Extensive testing demonstrating genetic stability has been performed on end of production cells (EPC) derived from the MCB and WCB. The strategy for monitoring stability of MCB and WCB is considered adequate. A protocol for establishment of future WCBs has been provided.

Sufficient information on raw materials used in the active substance manufacturing process has been submitted. Compendial raw materials are tested in accordance with the corresponding monograph, while specifications (including test methods) for non-compendial raw materials are presented. Information is provided on two materials of animal origin - isopropyl β-D-1-thiogalactopyranoside (IPTG) and stearate (see Adventitious Agents section). No other components of animal and/or human origin are declared.

The 40 kDa mPEG-intermediate, which is critical for the quality of the product, has been re-defined as an intermediate during the procedure as a result of a major objection. The new PEG starting materials have been defined in the dossier accordingly. The mode of preparation and quality controls of the new starting materials were appropriately documented.

Control of critical steps and intermediates

The criticality assessment and the evaluation of CQAs, CPPs, and IPCs are considered acceptable. During the procedure, a major objection was raised on the control strategy for the manufacturing process of the active substance because it had not been unambiguously defined and justified. The criticality assessment was then repeated and the control strategy revised. The applicant has re-evaluated CQAs, CPPs, and IPCs. Consequently more IPC items are now proposed for commercial production. Information on IPC testing at different stages of process development and process validation has been presented. IPC testing for commercial production is now provided in a transparent form and considered satisfactory. Based on the results, critical process steps have been suitably defined. For all process parameters (critical and non-critical), operating ranges were set. The PEGylation reagent is a 40 kDa branched PEG which, as mentioned above, has been re-classified as an intermediate during the procedure.

The route of synthesis as well as the process parameters and in-process controls of the 40 kDa PEG intermediate are suitably described in the dossier. The specification of the 40 kDa PEG intermediate was justified and completed with the requested parameters during the procedure. The fate and purge of impurities generated throughout the production of the intermediate were also satisfactorily addressed. An appropriate QP declaration has been provided confirming appropriate audit of the sites and that the manufacture of the intermediate complies with the principles and guidelines of good manufacturing practice. The shelf-life for the PEG intermediate under the specified storage condition is agreed.

The specification includes appropriate tests for identity, potency/content, purity and physicochemical attributes.

Re-validation of some AS intermediate chromatographic analytical methods has been initiated upon request. While re-validation has been finalised for some of these methods and updated documents have been provided, re-validation reports for other specified chromatographic analytical methods (which may continue to be used for release of marketed product) should be submitted post-authorisation (see recommendations).

Batch data (from process validation and GMP lots) demonstrate that test results met the set acceptance criteria. However, it is recommended to re-evaluate the acceptance criteria for the AS intermediate after re-validation with the new standards (recommendation). In response to a major objection regarding the shelf-life for the intermediate in the specified container, the shelf-life has been acceptably revised.

Process validation

The active substance manufacturing process is considered validated for the process from the seed culture to Pro-IFN alfa-2b and to ropeginterferon alfa-2b. Consistency of the manufacturing process has been shown on a suitable number of batches at commercial production scale. Evaluation of process-parameters and in-process controls demonstrate that the manufacturing process of ropeginterferon alfa-2b is consistent.

To understand the depletion capacity of the manufacturing process for impurities, validation studies for removal of product- and process-related impurities were performed and samples taken from different process steps were monitored. The potential process-related impurities for Pro-IFN alfa-2b AS intermediate may derive from medium components, additives, and host cell components. The medium component and chemical additives have been described. The host cell derived impurities include host cell proteins, residual DNA, and endotoxin. The removal of these process-related impurities is suitably demonstrated. The potential process-related impurities resulted from the PEGylation reaction have also been defined. The removal of these process-related impurities is also suitably demonstrated. In conclusion, the current manufacturing process is capable of effectively and consistently reducing these impurities to low levels. Most of the process-related impurities are below the LOQ.

Sufficient information on holding times of process intermediates has been provided. It has been demonstrated that quality of the commercial AS is representative of that of the materials used in the clinical trials.

The column lifetime was addressed using small-scale and full-scales analyses. The testing program included controls for purity, impurities, recovery and performance. Potential leachables were adequately addressed. The containers used for the storage of AS are generally considered as inert, non-reactive, and non-leachable material for injectable solution. Stability studies have also confirmed the suitability of the storage container.

Manufacturing process development

The process development history is appropriately documented. Comparability between active substances resulting from the different processes over historical development has been demonstrated. The process validation runs were performed using the commercial process. The analytical results at release are consistent between batches. The critical quality attributes (CQAs) of ropeginterferon alfa-2b were identified and risk was assessed using failure mode and effect analysis (FMEA), and then the higher risk process steps and operating parameters were further investigated in process characterisation studies.

Characterisation

Ropeginterferon alfa-2b is a PEGylated recombinant human IFN alpha-2b with addition of an extra amino acid (proline) at the N-terminus. The applicant claims it is a long-acting interferon with only one major isoform in contrast to the 8-14 isomers of other approved, PEGylated interferon products.

Characterisation studies for the AS intermediate as well as for the active substance were performed with batches manufactured using the process considered representative of the commercial manufacturing process.

Interferon alfa-2 is a non-glycosylated polypeptide chain of 165 amino acid residues. Different types of alfa-2 interferon exist, varying in the amino acid residue at position 23. The selected one for this product is interferon alpha-2b, with arginine at position 23. Proline-interferon alfa-2b contains an identical amino acid sequence to interferon alpha-2b plus an extra proline at the N-terminus (166 amino acid residues). Ropeginterferon alfa-2b is a PEGylated proline-interferon alfa-2b synthesized by conjugating a single 40 kDa PEG molecule to the N-terminal proline residue. Similar to Pro-IFN alfa-2b, ropeginterferon alfa-2b was also confirmed to contain two pairs of disulfide bonds, (Cys-2)-(Cys-99) and (Cys-30)-(Cys-139).

The structures of the AS intermediate and the active substance were elucidated using a variety of physico-chemical (e.g. peptide mapping, N- and C-terminal sequencing, electrophoretic and liquid chromatography-mass spectrometry analysis (LC-MS), SE-HPLC, RP-HPLC, IEX-HPLC, etc.), biophysical, and biological (cytopathic effect (CPE)-based potency assay and surface plasmon resonance (SPR) binding assay) techniques to provide a comprehensive understanding of their structure and functional properties and impurity profile.

The primary sequence of the AS was confirmed using peptide map/ RP-HPLC/ LC-MS/MS with 100% sequence coverage. Likewise, molecular mass of the intact protein, extinction coefficient, and presence of disulfide bridges as well as secondary and tertiary structure profiles of AS sample and respective reference batches were confirmed to be superimposable. Only one main PEGylation site was determined; positional isomers, i.e. mono-PEGylated products with different PEGylation sites, were detected at relatively low level using IEX-HPLC.

Active substance samples have been investigated for oxidized forms, deamidated forms, positional isomers and different pegylated forms and levels are suitably controlled. Biological characterisation includes cytopathic effect-based potency assay (antiviral assay) and ligand/receptor binding assay. The antiviral bioassay is proposed for routine use while the ligand/receptor binding assay, intended for characterisation use, monitors

the binding affinity of AS intermediate active substance toward both chains (IFNAR1 and IFNAR2) of the Type I IFN receptor.

Comprehensive data are available for the characterisation of active substance intermediate as well as the active substance. With the D150 and D180 response most of the variants detectable using chromatographic analyses have been identified and quantified.

Specification

The active substance specification includes all the critical quality attributes that affect the manufacturing and quality of the finished product. A summary of testing parameters and methods and the corresponding acceptance criteria have been provided. The specification includes testing of purity (endotoxin-Ph. Eur.; bioburden -Ph. Eur.), product-related impurities, process-related impurities, identity, potency (CPE antiviral assay), protein content (Ph. Eur.), and several general tests.

Most methods used in characterisation studies to determine the purity/impurity profile of ropeginterferon alfa-2b are included in the specification. Based on the additional characterisation data provided with the D150 and D180 response, revised purity/impurity specifications were proposed for the active substance. Furthermore and in accordance with the provided characterisation data SDS-PAGE analysis/ coomassie method has been replaced by SDS-PAGE/ silver staining method and refined impurity acceptance criteria have been implemented. Free Pro-IFN alfa-2b is now determined using SE-HPLC.

The general approach to justify the proposed specification taking into consideration historical process batch data as well as stability data is considered adequate and some limits were requested to be tightened during the procedure.

In addition, it is recommended to re-evaluate the HCP acceptance criteria for the active substance and to tighten the specification once a specified number of commercial batches have been manufactured (see recommendations).

Analytical methods

Many of the analytical procedures applied for control of the active substance are in-house methods for which acceptable descriptions were provided. The basis for calculation of AS purity has been clarified. Representative chromatograms have been provided. In-house, process-specific HCP enzyme-linked immunosorbent assay (ELISA) is used for determining the HCP content. Satisfactory information on analytical validation for non-pharmacopoeial methods has been provided in line with ICH guidelines. Pharmacopoeial methods have been verified.

The applicant proposes to use the antiviral potency assay described in Ph. Eur. monograph for release/stability testing. The bioactivity of active substance is determined using a CPE-based bioassay that measures the ability of the protein sample to protect A549 epithelial lung carcinoma cells from cytotoxicity due to infection with the EMCV. The applicant will include this antiviral potency assay described in the Ph. Eur. monograph for release/stability testing. An alternative assay with improved sensitivity is currently under development and may be introduced as an orthogonal alternative/replacement assay to the antiviral assay (see recommendations).

Batch analysis

Batch information and active substance release data of active substance for a suitable number of process validation lots and several GMP lots - all at commercial-scale- are provided. The results are within the

specifications and confirm consistency of the manufacturing process. Batch results for batches used in clinical studies were provided and support consistency.

Reference materials

In-house reference standards are used for AS intermediate and AS manufacture. Characterisation of the previously and currently used standards is considered adequate. An appropriate qualification protocol for future reference standards was provided. WHO International Standard is used as a primary reference standard in potency assay to calibrate the biological activities of the new in-house primary reference standards. Specific activities of the current AS intermediate primary RS lot as well as AS primary RS lot have been stated. Satisfactory information has been provided on actual potency assignment.

The strategy to establish new standards has been satisfactorily revised.

Stability

The applicant has defined a shelf-life and storage conditions for the AS in specified containers.

The stability claim in line with ICH guidelines is based on real-time data obtained from a suitable number of validation batches produced at PharmaEssentia Corp. Materials used for stability studies were considered as representative of materials derived from the commercial process.

All batches were stored in representative containers.

Accelerated stability and forced degradation studies (thermal, acidic, oxidative, high pH, agitation and light exposure) were also conducted.

The proposed stability protocol containing adequate stability-indicating test parameters is considered appropriate. The parameters chosen for stability monitoring are, in general, the same as those for release testing.

Endotoxins and bioburden are monitored at the beginning and the end of the stability program.

The long-term, real-time, real-condition stability studies show all results remain within the acceptance criteria.

Accelerated stability and forced degradation studies show that the stability-indicating methods are qualified for the intended use.

Based on the results from the photo stability study it is concluded that ropeginterferon alfa-2b active substance is susceptible to photo degradation.

The stability results indicate that the active substance is sufficiently stable and justify the proposed shelf life in the specified container.

2.2.3. Finished Medicinal Product

Description of the product and pharmaceutical development

Ropeginterferon alfa-2b is presented as a colourless to slightly yellowish sterile solution and is intended for subcutaneous administration.

The finished product includes one multi-dose, disposable pen injector, containing one rubber capped glass cartridge containing ropeginterferon alfa-2b solution and two CE-marked safety needles for injection. The material complies with Ph. Eur. and EC requirements.

Two presentations (250 µg and 500 µg, based on protein content, not including the weight of the PEG molecule) are available. The compositions of 1 mL of finished product for 0.5 mg/mL and 1.0 mg/mL concentrations, respectively, include the ropeginterferon alfa-2b active ingredient and the excipients. The excipients include sodium chloride (tonicity agent), sodium acetate, anhydrous (buffer agent), acetic acid, glacial (buffer agent), benzyl alcohol (preservative), polysorbate 80 (stabiliser) and water for injections (vehicle). All excipients are well known and comply with the requirements and specifications of the relevant compendial monographs. There are no novel constituents used in this formulation. The finished product (FP) was developed for subcutaneous administration and its formulation remained unchanged since the start of product development and throughout the clinical trials. The final commercial process was used in phase III clinical trials. An injection pen was chosen for delivery to provide maximum flexibility for dose adjustment while maintaining a high level of dose accuracy, as compared to dose selection by withdrawing solution from a glass vial. The injection pen enables patients to self-administer doses of ropeginterferon alfa-2b at home in between physician / clinic appointments. The suitability of the pen injector has been demonstrated.

The compatibility of the container closure system with the dosage form was assessed with respect to stability testing and extractable/leachable studies. The integrity of the container closure system was verified using the dye ingress test method. The injection pen is intended for multiple uses (twice a month) and benzyl alcohol is used as a preservative in the finished product formulation. The acceptance criteria for the benzyl alcohol concentration are sufficiently justified.

Manufacture of the product and process controls

The FP is released by AOP Orphan Pharmaceuticals, Austria. The GMP status of all the FP manufacturing sites has been confirmed.

The FP batch size has been specified. The FP manufacturing process starts with the compounding of excipient solution, followed by a bioburden reduction filtration. The filtered excipient solution is added to pooled active substance. The process continues through a sequence of operations in a grade A environment (sterile filtration, filling into the stoppered cartridges, sealing and capping) to obtain the filled cartridges. A 100% visual inspection is performed and the filled cartridges are stored at 2-8°C. After that, the pen elements are assembled with the filled cartridges and finally, labelling and packaging is performed. No reprocessing is claimed.

Compounding steps, sterile filtration and filling as well as pen assembly were defined as critical steps. Comprehensive lists of process parameters and in process controls that can be expected to impact the FP CQAs are provided. The listed process parameters and in process controls are considered adequate to control the FP manufacturing process.

The in-process controls performed on the critical steps of the manufacturing process are satisfactory.

Process validation on a suitable number of batches has been conducted and demonstrates that the process is acceptably validated. The defined process conditions and parameters, homogeneity of the bulk and filling of minimum and maximum batch sizes as well as successful pen assembly have been confirmed. Hold times were microbiologically and physico-chemically verified. Filter flush volume for the sterilizing filter was also checked. Release data for all batches support the reproducibility of the manufacturing process.

Product specification

Several potential process-related impurities relevant at active substance level, are not considered relevant for finished product, since all of them are consistently removed by the purification process to acceptable levels and in finished product manufacturing, no further purification step takes place. Levels of product related impurities are controlled by various analytical methods at batch release.

Assessment of elemental impurities was done as defined by ICH Q3D Guideline. Different lots of active substance were tested, demonstrating that levels of these elements are below the defined PDE limits in ICH Q3D Guideline. Equipment and container/closure components were also considered in the risk assessment.

The specification includes appropriate testing of purity, identity, general tests, potency and protein content. The acceptance criteria of the FP release specifications generally correspond to the active substance specification. Justification with respect to clinical qualification/ tightening of some FP limits was requested during the procedure.

Analytical methods

Most of the analytical procedures for testing the finished product are in principle the same as those for the active substance (including for potency). The analytical procedures are sufficiently described and considered adequately validated.

Batch analysis

Batch results were provided for the process validation batches at commercial scale in cartridges (0.5 mg/ mL and 1 mg/ mL strength). The 0.5 mg/mL cartridges were further processed to pre-filled pens. The results on extractable volume are presented. All results comply with the defined acceptance criteria.

Reference materials

See active substance section.

Container Closure System

The primary packaging of the finished product for both presentations consists of a 3 mL, Type I, clear borosilicate glass cartridge which is closed with a grey bromobutyl rubber stopper and an aluminium crimping rubber septum cap. The cartridge and rubber components comply with the requirements of the Ph. Eur. The effectiveness of depyrogenation of the cartridges has been demonstrated.

Medical device

The pen injector for ropeginterferon alfa-2b pre-filled pen 250 µg and 500 µg (functional secondary packaging) is a “dial and push”, multi-dose, disposable needle-based injection system intended exclusively for use in combination with the integrated non-replaceable finished product cartridge. Depending on the individual patient’s dose, one pen may be used for up to two administrations, but has to be discarded at the latest 30 days after the first use. Dose settings visible in the display window of the pen start with 50 µg and can be increased in 50 µg steps. The pens for the two strengths of the finished product are identical except for the printing for dose setting and the push button colour.

The pen injector corresponds to System Designation C of ISO 11608-1 (current version) "Needle-based injection systems for medical use – Requirements and test methods". Product verification and validation demonstrated conformance to ISO 11608-1. Biocompatibility was evaluated in accordance to EN ISO 10993-1. The applicant confirms that the essential requirements of the Medical Device Directive 93/42/EEC, Annex I have been met.

The manufacturing process and quality control of the components and the subassembly of the components are described. Pen injector real-time and accelerated ageing has been studied.

CE-marked injection safety pen needles are provided with the pen injector.

The submitted information on the pen injector for ropeginterferon alfa-2b pre-filled pen 250 µg and 500 µg and the pen needles is considered sufficient.

Stability of the product

A shelf life of 3 years for ropeginterferon alfa-2b 0.5 mg/ml (250 micrograms/0.5 ml) strength and a shelf life of 18 months for ropeginterferon alfa-2b 1.0 mg/ml (500 micrograms/0.5 ml) strength both at +2°C to +8°C, are proposed. Long term stability studies at real time/real temperature conditions as well as accelerated studies were initiated in line with ICH guidelines. All samples were stored in the original containers used for marketing. A suitable number of PV batches have been included in the stability program. For the FP of both strengths stored at 2-8°C, real-time data are available as are accelerated data (25°C/60%RH). In accordance with EU GMP guidelines[1], for ongoing studies, any confirmed out-of-specification result, or significant negative trend, will be reported to the Rapporteur and EMA.

The applicant discusses the stability indicating test parameters and confirms that the test methods used for the stability studies are the same as used for release testing.

The real-time data presented are within specification limits and prove the unimpaired product quality over the current stability testing periods.

After first use, the pre-filled pen may be stored for a maximum of 30 days in the refrigerator (2°C - 8°C) when stored with the pen cap on and kept in the outer carton in order to protect from light. The pre-filled pen may be used up to two times within these 30 days. Any medicine remaining in the pre-filled pen after the second use and/or after 30 days must be discarded. In-use stability has been demonstrated.

Photostability studies demonstrated that ropeginterferon alfa-2b finished product is susceptible to light in the glass cartridges whereas the pen injector is capable of protecting ropeginterferon alfa-2b against light exposure.

A shelf life of 3 years for ropeginterferon alfa-2b 0.5 mg/ml (250 micrograms/0.5 ml) strength and a shelf life of 18 months for ropeginterferon alfa-2b 1.0 mg/ml (500 micrograms/0.5 ml) strength both at +2°C to +8°C are accepted.

Adventitious agents

The applicant provided information on two materials of animal origin - IPTG used for induction of protein expression and stearate, which is used during manufacture of the cation exchange resin matrix. IPTG is manufactured using bovine milk from India; the stearate (from tallow origin) has been manufactured by rigorous processes in accordance with the requirements of the European guideline EMA/410/01 rev.3. The applicant's risk assessment regarding the risk for TSE transmission for both materials is considered adequate.

As the production cell substrate for ropeginterferon alfa-2b is *E. coli*, this represents a major barrier to the transmission of viral adventitious agents. Viruses do not infect or replicate in *E. coli* cells and only a virus-like component, bacteriophage, can infect and replicate there. The strategy for controlling the risk of bacteriophage contamination is inactivation of bacteriophage from the potential source. It is concluded that the risk

[1] 6.32 of Vol. 4 Part I of the Rules Governing Medicinal products in the European Union

assessment on viral safety shows very low risk of transmission of viral adventitious agents. The risk is appropriately minimised with the control strategy.

2.2.4. Discussion on chemical, pharmaceutical and biological aspects

In the initial submission four major objections were identified, which were resolved during the procedure.

The 40kDa PEG intermediate was proposed as a starting material by the applicant. However, as it is critical to the quality of the product, it has been re-classified by the applicant as an intermediate. All steps of its synthesis starting from the newly defined starting materials are performed under GMP. A revised QP declaration has been provided.

A major objection was raised on the control strategy for the manufacturing process of the active substance that was not unambiguously defined and justified. The criticality assessment has been repeated and the control strategy has been revised. The applicant has re-evaluated CQAs, CPPs, and IPCs. Consequently more IPC items are now proposed for commercial production.

IPC testing for commercial production is now clarified and considered satisfactory. The proposed shelf-life for the non-pegylated active substance intermediate has been based on the data submitted further to a major objection at D120 of the procedure.

GMP issues raised as major objections have been resolved and respective certificates have been provided

Besides the MOs, the other issues have also been resolved. Five recommendations are also given. Re-validation of some AS intermediate chromatographic release methods has been initiated, upon request. While re-validation has been finalised for some of these methods and updated documents have been provided, re-validation reports for other specified chromatographic methods (which may continue to be used for release of marketed product) should be submitted post-authorisation (see recommendations).

During review, it was noted that difference exists between Besremi and the standard used, it is therefore recommended to re-evaluate the acceptance criteria for an AS intermediate test after revalidation with the new standards.

For the AS manufacturing process, the routine IPCs are suitably described and found acceptable. However, an additional IPC during the early stages of purification is recommended to assure consistency: establishment of a numerical acceptance criterion for this IPC is recommended when data from a specified number of batches are available.

It has been clarified that with the new in-house ELISA although data exist in order to set an AS specification for HCP, the applicant is recommended to re-evaluate the HCP acceptance criteria for the active substance and to tighten the specification once a specified number of commercial batches have been manufactured.

The bioactivity of active substance is determined using a CPE-based bioassay (antiviral potency assay) that measures the ability of the protein sample to protect A549 epithelial lung carcinoma cells from cytotoxicity due to infection with the encephalomyocarditis virus (EMCV). An alternative assay with improved sensitivity is currently under development. It is therefore recommended to introduce an alternative/replacement method for potency by variation application post-authorisation (see recommendations).

In conclusion, information on development, manufacture and control of the active substance and finished product has been presented in a satisfactory manner. The results of tests carried out indicate consistency and

uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be acceptable when used as defined in the approved SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. Data have been presented to give reassurance on viral/TSE safety. Recommendations for future quality development were agreed by the applicant.

2.2.6. Recommendation(s) for future quality development

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

Area	Number	Description	Classification*
Quality	001	A numerical acceptance criterion for inclusion body recovery should be established when data from further batches are available.	REC
Quality	002	It is recommended to re-evaluate the acceptance criteria for a specified AS intermediate test after re-validation with the new standards.	REC
Quality	003	Re-validation reports for specified chromatographic methods used for the AS intermediate release should be submitted.	REC
Quality	004	It is recommended to re-evaluate the HCP acceptance criteria for the active substance and to tighten the specification once a specified number of commercial batches have been manufactured.	REC
Quality	005	It is recommended to introduce an alternative/replacement method for potency by filing a variation application.	REC

* REC = Recommendation

2.3. Non-clinical aspects

2.3.1. Introduction

The biological activity of ropeginterferon alfa-2b (P1101) was compared to that of peginterferon alfa-2a in Cynomolgus monkeys. PD/PK and toxicokinetic studies were performed in Cynomolgus monkeys; safety pharmacology studies comprised an *in vitro* hERG assay, a cardiovascular study in cynomolgus monkeys and central nervous system and respiration studies in rats.

2.3.2. Pharmacology

Primary pharmacodynamic studies

A Pharmacokinetic and Pharmacodynamic Study of P1101 versus Pegasys in Male Cynomologus Monkeys (Study Number 44101-08-242)

Administration of ropeginterferon alfa-2b or Pegasys induced serum activities of 2',5'-oligoadenylate synthetase (OAS) in a time-dependent manner (see Figure 3 and Figure 4). Both magnitude and kinetic of OAS response induced by ropeginterferon alfa-2b versus Pegasys are comparable till 144 hr postdose. After that time point, OAS activities in the animals receiving Pegasys were gradually subsided, while the OAS activities in the animals receiving ropeginterferon alfa-2b were relatively stable at a high level for about 14 days.

Figure 3: Induction of OAS Activity by Ropeginterferon alfa-2b (P1101) and Pegasys at the dose of 30 mg/kg

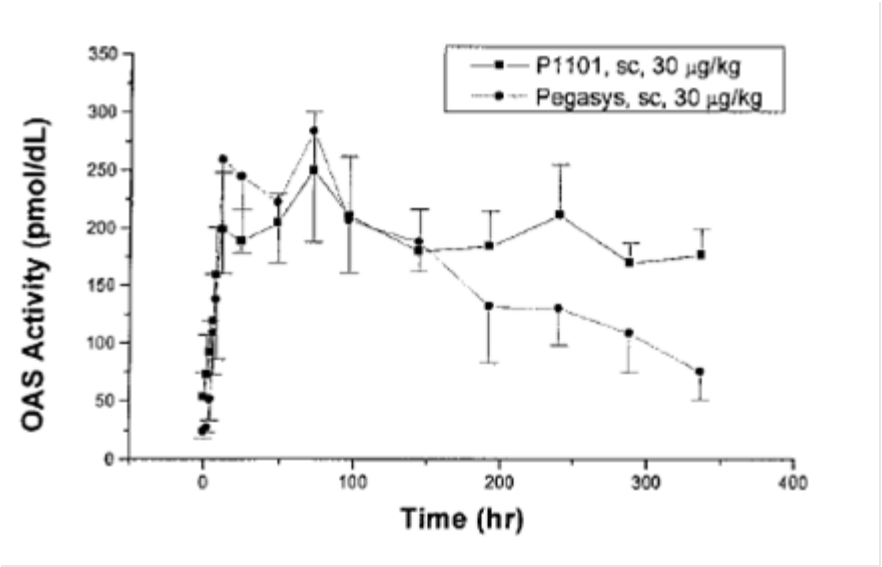
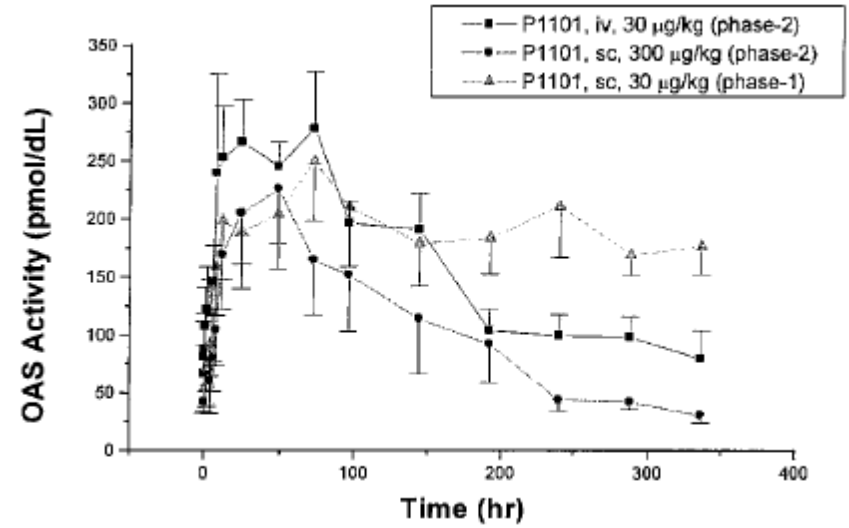


Figure 4: OAS Activity Following s.c. and i.v. Administration of 30 µg/kg Ropeginterferon alfa-2b (P1101)



Pharmacokinetic Analysis

Serum concentration of ropeginterferon alfa-2b is illustrated in Figure **5** and Figure 6. Relative bioavailability of ropeginterferon alfa-2b was 42% for subcutaneous administration of 300 µg/kg in the phase-2 study versus 80% for subcutaneous administration of 30 µg/kg in the phase-I study (see Table 2).

Figure 5:: Serum Concentrations of Ropeginterferon alfa-2b (P1101) and Pegasys -30 mg/kg s.c.

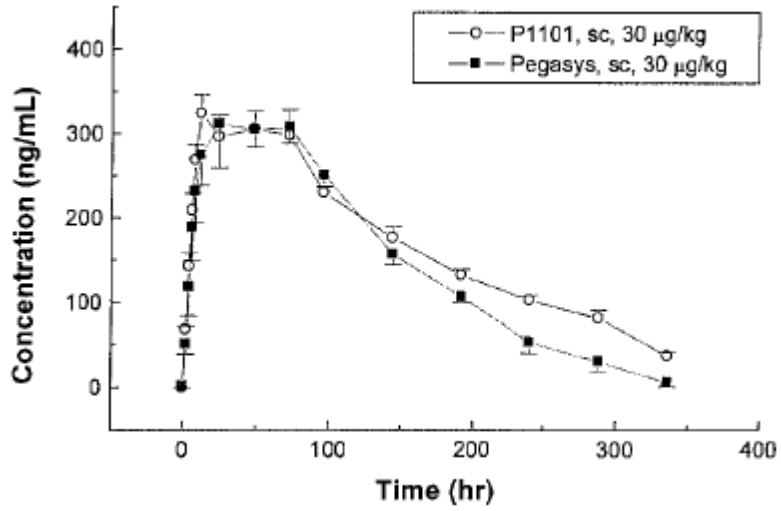


Figure 6: : Serum Concentrations of s.c. and i.v. Administered Ropeginterferon alfa-2b (P1101)

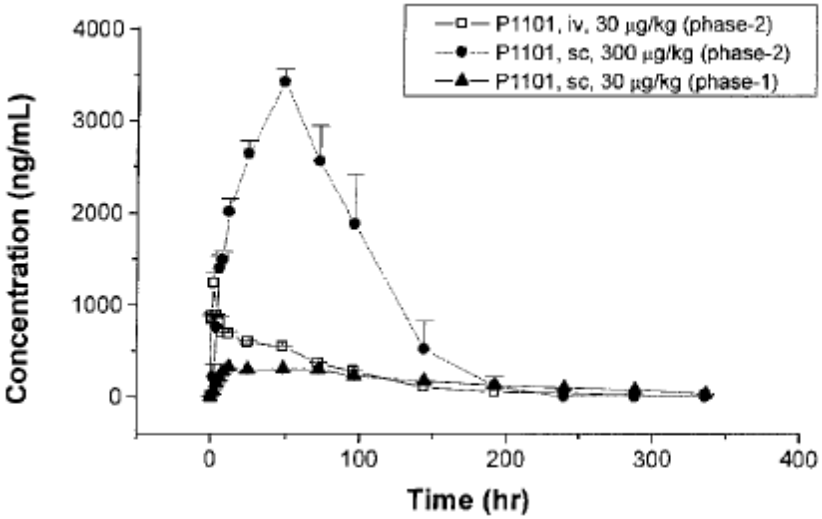


Table 2: PK Parameters of Ropeginterferon alfa-2b (P1101) and Pegasys

Parameter	Phase I			Phase II	
	P1101 (*s.c.*)	Pegasys (*s.c.*)	P1101/Pegasys Ratio	P1101 (*i.v.*)	P1101 (*s.c.*)
Dose (µg/kg)	30	30	1.00	30	300
T_{max} (hours)	33±13.3	66±15.1	0.50	2±0.0	54±6.0
C_{max} (ng/mL)	332.57±22.94	344.05±35.57	0.97	1235.95±109.18	3457.36±106.14
$t_{1/2}$ (hours)	66.06±7.90	61.72±8.44	1.07	67.03±13.55	22.22±7.97
AUC_{last} (hr•ng/mL)	56883.49±3333.57	49546.20±2739.67	1.15	65209.31±9619.71	312071.43±42179.89
AUC_{inf} (hr•ng/mL)	60528.28±3779.34	50882.71±2438.23	1.19	75447.13±6647.68	314370.16±41219.54
MRT_{last} (hours)	120.87±1.15	100.76±7.41	1.20	63.40±14.53	63.21±7.95
Vz_F (mL/kg)	47.36±4.77	53.45±9.38	0.89	NA	29.51±8.57
Cl_F (mL/hr/kg)	0.50±0.034	0.59±0.029	0.85	NA	1.02±0.169
Vz (mL/kg)	NA	NA	NA	37.29±4.54	NA
Cl (mL/hr/kg)	NA	NA	NA	0.41±0.036	NA

Secondary pharmacodynamic studies

No secondary pharmacodynamics studies were performed with ropeginterferon alfa-2b.

Safety pharmacology programme

Effects of Ropeginterferon alfa-2b on Cloned hERG Potassium Channels Expressed in Human Embryonic Kidney Cells

The objective of this study was to examine the *in vitro* effects of ropeginterferon alfa-2b on the hERG (human ether-à-go-go-related gene) channel current (hERG as a surrogate for IKr, the rapidly activating, delayed rectifier cardiac potassium current). Ropeginterferon alfa-2b at 2.5 µM (150 mg/mL; 2.5 µM is approximately the Cmax) was applied to four cells. An additional concentration (0.3 µM (actual 0.25 µM)(15 µg/mL)) was selected and applied to four cells also. Terfenadine (60 nM) was used as positive control.

Ropeginterferon alfa-2b inhibited hERG potassium current by (mean ± SEM) 10.8 ± 0.6% at 0.25 µM (n = 4) and 49.9 ± 2.9% at 2.5 µM (n = 4) versus 0.4 ± 0.2% (n = 3) in control. The IC50 for the inhibitory effect of PEG INF alpha 2b on hERG potassium current was not determined since inhibition was < 50% and the 2.5 µM was

approximately the Cmax. The positive control (60 nM terfenadine) inhibited hERG potassium current by (Mean ± SD; n = 2) 77.1 ± 0.1%. Table 3

Table 3: The Effect of Ropeginterferon alfa-2b on hERG Channel Current

Concentration	% Inhibition (Mean ± SEM)	Measured Cells
0 (Control)	0.4 ± 0.2	3
0.3 µM (0.25 µM, 15 µg/mL actual)	10.8 ± 0.6	4
2.5 µM (150 µg/mL)	49.9 ±2.9	4

Table 3Table 3Table 3Table 3

At concentrations of 2.5 µM (150 µg/mL), a statistically significant inhibition of the tail current amplitude of 49.9% in relation to the normalised control current amplitude was observed. However, the concentrations of 2.5 µM (150 µg/mL) were far in excess of plasma levels achieved clinically (Cmax = 24.84 ± 8.66ng/mL in study P1101-2010; PEGINVERA-PV Study).

Effects on General Activity, Behaviour and Body Temperature in the Rat Following sc Administration

Four groups of male Sprague Dawley rats comprising 6 animals each received a single administration of placebo or test article at doses of 0.2, 2.0 or 20 mg/kg by subcutaneous injection, using a constant dose volume of 4 mL/kg. Irwin observations were performed at 1, 2, 4, 8, 24 and 48 hours post-dose. In addition, the rectal body temperature of each rat was measured pre-dose and immediately following each Irwin observation. The animals were kept for a further 7 days following the day of dosing for general observations.

The formulation analysis of the three doses (0.2, 2.0 and 20 mg/kg) of ropeginterferon alfa-2b showed mean recoveries of 220, 114 and 98%. Subcutaneous administration of ropeginterferon alfa-2b at dose levels of 0.2, 2.0 or 20 mg/kg produced no behavioural, physiological or body temperature changes in rats when compared to placebo-treated animals in this study.

Effects on Respiratory Parameters in the Freely Moving Conscious Rat Using Whole Body Plethysmography

Four groups of male Sprague Dawley rats comprising 6 animals each were acclimatised in plethysmograph boxes (1 rat per box) for one hour prior to dosing. The four groups of rats were administered once *i.v.* with vehicle or ropeginterferon alfa-2b at the doses of 0.1, 1.0 and 10 mg/kg, respectively and returned to the same plethysmograph boxes. The following respiratory parameters were recorded pre-dose and for 6-hour interval after dosing: tidal volume, minute volume and rate of respiration.

Administration of the vehicle as well as the test article (ropeginterferon alfa-2b) at all doses elicited small increases in the tidal volume, rate and minute volume in the first 15 minutes after dose administration. This was observed across all groups and is related to the dosing procedure and the settling period in the plethysmograph environment.

The formulation analysis of the three concentrations (0.1, 1.0 and 10 mg/kg) of ropeginterferon alfa-2b showed mean recoveries of 173, 116 and 95%. It is concluded that under the experimental conditions iv administration of PEG-Pro-IFN alpha-2b at 0.1, 1.0 and 10 mg/kg, did not elicit any statistically significant changes in respiratory parameters in the conscious rat during a 6-hour period, when compared to the vehicle treated group.

***In Vivo* Studies**

Electrocardiographic Investigations on Single-Lead ECGs Using Radio-Telemetry in the Conscious Cynomolgus Monkey Following Intravenous Administration

The objective of this study was to determine the cardiotoxicity of ropeginterferon alfa-2b in the conscious cynomolgus monkey following intravenous administration at a dose volume of 1 ml/kg. Six male non-naïve cynomolgus monkeys were pre-instrumented for single-lead telemetry recordings. Each subject received vehicle or test item administered *i.v.* using an adjusted Latin square design. The telemetric data recording indicated an increase in body temperature in 3 out of 4 animals following the high dose (600 µg/kg) administration. This increase was starting approximately 180 minutes post dose and lasting until approximately 720 or 840 minutes post dose. In 1 out of 4 animals the body temperature was also increased following the low dose (60 µg/kg) administration. In addition, the heart rates were increased with corresponding QT shortening in the same animals and duration (180 minutes to 840 minutes post dose) at 600 µg/kg. In addition, QT intervals were as well shortened in 1 of 4 animals at the mid dose of 60 µg/kg. Furthermore, QTcf was reduced in 2 following intravenous administration of the high dose of 600 µg/kg.

It is concluded that the elevated heart rates with corresponding QT and QTc shortening as well as the elevated body temperatures are a result of the intravenous treatment with the test item ropeginterferon alfa-2b.

The mean arterial blood pressure was only increased in 1 animal following the high dose (600 µg/kg).

Pharmacodynamic drug interactions

Non-clinical studies investigating pharmacodynamic drug interactions with ropeginterferon were not submitted.

2.3.3. Pharmacokinetics

Stand-alone evaluations of PK characteristics of ropeginterferon alfa-2b were not performed. Pharmacokinetic characterization of ropeginterferon alfa-2b was performed as part of the combined pharmacokinetic/pharmacodynamics (PK/PD) study in cynomolgus monkeys (Study No. 44104-08-242; see above). In addition, pharmaco-toxicokinetic evaluations were carried out as part of the GLP toxicity studies in cynomolgus monkeys (see toxicology).

2.3.4. Toxicology

Single dose toxicity

No stand-alone single dose toxicity studies were performed. Single dose toxicity testing was performed as part of the non-GLP dose range finding study in rats (Study No. 7975-100).

Repeat dose toxicity

The program for ropeginterferon alfa-2b was designed taking into account the nature of the product and its intended route of administration. Dose range finding studies were performed in rats (see Table 4) and cynomolgus monkeys (see Table 5). The cynomolgus monkey was selected as a relevant species for the pivotal 4-week repeated dose toxicity study. Two additional bridging toxicity studies in cynomolgus monkeys compared a toxicity lot of ropeginterferon alfa-2b with the clinical lots.

Table 4: dose finding study in rats

Study ID	Species/Sex/ Number/Group	Dose/Route mg/kg/dose	Duration	NOEL (mg/kg/day)	Major findings
7975-100	Crl:CD(SD)rats	0/0.2/2/20 s.c. in 4 mL/kg	single	≥20	No mortality; unremarkable clinical pathology ;
Non-GLP	3/sex/group	0/0.2/2/20 s.c. in 4 mL/kg	multiple on days 1,4,8 and 11	≥20	

Single- and multiple-dose treatment of rats with ropeginterferon alfa-2b did not translate into any pathological or clinical symptoms. Treated rats had normal food consumption and did not show any relevant clinical signs related to treatment. In conclusion, the NOEL following a single or multiple subcutaneous injection of ropeginterferon alfa-2b into rats is at or greater than 20,000 µg/kg/dose (the highest tested dose).

Table 5: dose finding study in cynomolgous

Study ID	Species/Sex/ Number/Group	Dose/Route mg/kg/dose	Duration	NOAEL mg/kg/day	Major findings
7975-102	Cynomolgus Monkey	0.675/2/6.75 s.c. in 2 mL/kg	Dose escalation Daily for 4 non-consecutive days within 2 weeks	<2	Serum ropeginterferon alfa-2b concentrations increased from Day 1 to Day 3 following *s.c.* administration at 2 mg/kg or 6.75 mg/kg. In males and females, serum

Non-GLP	1/sex/group	0/2/6.75 s.c. in 2 mL/kg	Multiple doses up to 14 days	<2

Based on the treatment-related decreases in mean body weights (males) and reticulocyte counts at 2 and 6.75 mg/kg/dose, the NOAEL for Ropeginterferon alfa-2b when given to male and female cynomolgus monkeys *via s.c.* injection once daily for four nonconsecutive days within a 2-week period is <2 mg/kg. A maximum tolerated dose (MTD) was not achieved in this study.

The aim of this GLP study was to assess the toxicity, toxicokinetics and immunogenicity of ropeginterferon alfa-2b in cynomolgus monkeys treated twice weekly over a period of four weeks and to determine reversibility, persistence or delayed occurrence of any treatment-related effect after a 4-week recovery (see Table 6).

Table 6: Study 7975-101

Study ID	**Species/Sex/ Number/Group**	**Dose/Route mg/kg/dose**	**Duration Application on days**	**NOAEL mg/kg/dose**	**Major findings**
7975-101	Cynomolgus Monkey Control group 5/sex Intermediate doses 3/sex Highest dose 5/sex	0/0.675/2/6.75 s.c. in 2mL/kg	1,4,8,11,15,18, 22 and 24 Animals were sacrified on day 29 or after 4 week recovery period	<6.75	No major findings All hematology changes were reversed completely at the end of recovery phase. ketonuria on Day 9; Production of neutralizing antibodies

Toxicokinetics: The toxicokinetic parameters determined in serum are shown in Table 7 below.

Following the first *s.c.* administration (Day 1), the systemic exposure was similar between male and female monkeys. The Cmax and AUC were increased in a linear manner proportionally to the administered dose. Decrease of PK parameters (Cmax, Tmax and AUC) was observed after Dosing Day 24, indicating the development of neutralizing antibodies. In line with this, serum concentrations of ropeginterferon alfa-2b started to decrease (with the exception of one male treated at 0.675 mg/kg/dose and one female that received 6.75 mg/kg/dose) after the last injection (Day 24), further arguing for the development of neutralizing antibodies.

Table 7: Toxicokinetic data study 7975-101

Time Point	PK Parameter	Dose					
		0.675		2		6.75	
		Males	Females	Males	Females	Males	Females
Day 1	AUC_{0-72h} [h•ng/ml]	379403 ± 162352	491057 ± 140308	1687204 ± 674409	1396471 ± 115587	4517087 ± 1068534	4716740 ± 337950
	C_{max} [ng/ml]	6488 ± 2423	8093 ± 2015	28697 ± 13097	23128 ± 2085	82713 ± 18390	81030 ± 4528
	T_{max} [h]	56 ± 14	40 ± 14	40 ± 28	48 ± 24	48 ± 24	34 ± 13
Day 24	AUC_{0-72h} [h•ng/ml]	80138 ± 121850	3433 ± 5535	37300 ± 45454	17558 ± 13183	789175 ± 588682	2549419 ± 3559568
	C_{max} [ng/ml]	1618 ± 1769	105 ± 159	1670 ± 2152	694 ± 513	31296 ± 22840	44561 ± 48106
	T_{max} [h]	13 ± 9	10	7 ± 1	9	12 ± 7	18

Administration of the maximum tested dose of ropeginterferon alfa-2b (6.75 mg/kg/dose) for four weeks did not affect organ weights and did not result in visible changes at necropsy. A decrease in red blood cell counts, hemoglobin, hematocrit, reticulocyte counts, and an increase APTT was observed. Clinical chemistry changes included decreased alkaline phosphatase, total protein, albumin, calcium, inorganic phosphorous, and ketonuria. Most of these treatment-related changes were observed in males and females treated with the highest dose of 6.75 mg/kg and were resolved by the end of the 29-day recovery period. The only treatment-associated microscopic changes were minimal to moderate infiltrates of lymphocytes and macrophages at the injection sites that either resolved or exhibited a reduction in average severity during the recovery phase. Based on non-adverse treatmentrelated effects in the food consumption, body weight loss and mild clinical observations, the NOAEL for ropeginterferon alfa-2b when given to male and female cynomolgus monkeys *via s.c.* injection once daily for 8 non-consecutive days within a 4-week period followed by a 4-week recovery period was defined at 6.75 mg/kg/dose. It is noted, that a decrease of PK parameters after the 8th dose (Day 24) of this study indicated the development of neutralizing antibody. Ropeginterferon alfa-2b concentration decrease in sera was found after the 5th dose indicating that the majority of the monkeys developed neutralizing antibodies by Day 17 on study. The presence of neutralizing antibodies might explain the higher NOAEL in this study compared to the non-GLP 2-week study.

a) 14-Day Repeat-Dose Subcutaneous Toxicity Study with Toxicokinetics and Immunogenicity of P1101 in Cynomolgus Monkeys

The aim of this GLP study (44103 -08-300) was to assess the toxicity and toxicokinetics of ropeginterferon alfa-2b (P1101) in cynomolgus monkeys that were treated at 6.75 mg/kg/dose sc twice weekly over a period of two weeks and to compare the GMP clinical lot with the lot used in the previous toxicity studies.

No mortality or treatment-related clinical signs were observed; A slight reduction in bodyweight could be observed in all ropeginterferon alfa-2b monkeys, which correlated with reduced food consumption; Lower blood pressures (11% -23% lower for toxicity lot, 8% -27% lower for clinical lot) and slightly lower body temperatures in both males and females from both treatment groups were recorded, when compared to the control group. All electrocardiography parameters were within the normal limits. Decreased group mean white blood cell and neutrophil counts were observed and significantly increased group mean basophil levels also noted. In addition, treatment-related prolongation of APTT (21-45%) was noted in groups treated with ropeginterferon alfa-2b,

when compared to the control. ALT, AST, BUN and BUN/C values were increased in test item-treated groups, while levels of total protein, albumin, globulin and triglycerides were decreased. Necropsy of ropeginterferon alfa-2b -treated animals revealed decreased weights of the thymi, microscopically correlating with diffuse atrophies. Animals had mononuclear perivascular immune infiltrates in the brain, while no such infiltrates were found in the control animals, suggesting a treatment related effect. Mixed cell inflammation in the lung was also identified only in the ropeginterferon alfa-2b administered animals.

Immunogenicity: Repeated administration of toxicity and clinical lots of the test item elicited anti-drug antibody responses in most cynomolgus monkeys. However, drug-neutralizing antibodies were not detected using an un-validated cell-based assay.

Toxicokinetics: Following subcutaneous administration to cynomolgus monkeys, the systemic exposure was generally similar between the groups receiving the toxicity lot and clinical lot. Accumulation of test item occurred with repeated doses until Day 13, when most of the animals showed a sharp decrease in ropeginterferon alfa-2b concentrations in serum (see Table 8).

Table 8: Toxicokinetic data study 44103-08-300

Day	Dose (mg/kg)	Lot Number	AUC_{last} [h×µg/ml]		C_{max} [ng/ml]	
			Males	Females	Males	Females
Day 1	6.75	DPL-0006-5	3237.35 ± 881.29	NA	62.68 ± 19.79	NA
		0013-3.375	4090.96 ± 517.05	NA	75.29 ± 16.40	NA
Day 11	6.75	DPL-0006-5	10364.80 ± 3975.96	3635.56 ± 1116.34	161.12 ± 35.52	67.73 ± 18.01
		0013-3.375	8949.93 ± 2420.59	4281.84 ± 523.89	142.20 ± 25.62	66.43 ± 8.24

Twice-weekly repeated *s.c.* administration of ropeginterferon alfa-2b at 6.75 mg/kg/dose with toxicity lot or clinical lot to male and female cynomolgus monkeys over a period of two weeks resulted in lower mean body weights, reduce food consumption, lower blood pressure and body temperature. Treatment also resulted in changes in hematological and clinical pathology parameters, decreased thymus weight, and histopathological findings.

There were no significant differences in treatment-related effects between the two lots of test article.

b) 2-Week Subcutaneous Administration Toxicity, Toxicokinetics and Immunogenicity Bridging Study of Two Lots of P1101 in Cynomolgus Monkeys

The aim of this GLP study (7975-107) was to compare and assess the toxicity, toxicokinetics and immunogenicity of ropeginterferon alfa-2b (P1101) in cynomolgus monkeys that were treated twice weekly at 0. 675 mg/kg/dose, sc, over a period of two weeks with two different lots of ropeginterferon alfa-2b.

No mortality or treatment-related clinical signs were observed. No treatment-related Electrocardiography effects were noted. Moderately reduced reticulocyte counts as well as slightly decreased red blood cell, hemoglobin and hematocrit values were noted. In addition, slightly reduced leukocyte, lymphocyte and eosinophil were noted in both males and females of both test item treated groups, while reduced neutrophils were observed only in ropeginterferon alfa-2b treated males. Changes in clinical chemistry parameters with

unclear association to the test item included slight decreases in levels of total protein, albumin, calcium and inorganic phosphorus could be observed on Day 14 when compared to the control group or pre-dose values.

Immunogenicity: One Group 2 female developed anti-ropeginterferon alfa-2b antibodies after the fourth injection. All other animals were tested negative.

Toxicokinetics: The systemic exposure to ropeginterferon alfa-2b was generally similar following *s.c.* administration of either of the tested lots. Accumulation of ropeginterferon alfa-2b occurred with repeated dosing (see Table 9).

Table 9: Toxicokinetic data study 7975-107

Day	Dose (mg/kg)	Lot Number	AUC_{last} [h×µg/ml]		C_{max} [ng/ml]	
			Males	Females	Males	Females
Day 1	0.675	DPL-0006-5	684.26 ± 198.55	431.30 ± 27.10	13.34 ± 5.20	7.02 ± 0.30
		08-DPL-B001	778.67 ± 94.50	545.58 ± 89.99	15.13 ± 1.59	9.46 ± 1.56
Day 11	0.675	DPL-0006-5	1418.32 ± 223.69	1246.81 ± 595.89	18.06 ± 2.49	20.20 ± 2.59
		08-DPL-B001	1613.93 ± 130.33	1844.03 ± 258.86	25.71 ± 2.81	26.05 ± 0.88

Conclusion: Ropeginterferon alfa-2b-induced changes were observed with both tested lots. Slight differences in the effects of the two lots of ropeginterferon alfa-2b were observed but, however, these differences were not toxicologically relevant and meaningful.

Table 10: Safety Margins between NOAEL in the Toxicity Studies and Maximum Human Dose

Repeat Dose Toxicity Study	Non-GLP (Study No. 7975-102)	GLP (Study No. 7975-101)
NOAEL	2 mg/kg	6.75 mg/kg
HED Conversion[a]	0.645 mg/kg	2.18 mg/kg
Maximum Applied Dose	500 µg, i.e. 7.1 µg/kg (assuming 70 kg body weight)	
Safety Margin (HED/Maximum Human Dose)	90.85	307.04

[a] Human equivalent dose (HED) calculated by applying allometric scaling factor of 3.1 based on body surface area

In conclusion, in the four-week pivotal toxicity study in cynomolgus monkeys treated *s.c.* on eight nonconsecutive days, the NOAEL was determined as 6.75 mg/kg. Exposure to the test item was not affected by the humoral immune response throughout the dosing phase and was similar between the males and females. Reduction in PK parameters such as Cmax and AUC was observed after the administration of the last dose and was mirrored by development of antidrug and neutralizing antibodies. The NOAELs of the both monkey studies (2 and 6.75 mg/kg, respectively) allows for an appropriate safety margin to the maximal applied human dose of 540 µg, i.e. 7.7 µg/kg (based on a 70 kg body weight). (See Table 10).

Genotoxicity

Two in vitro genotoxicity tests were performed with ropeginterferon alfa-2b as summarised in Table 11.

Table 11: **Overview of genotoxicity studies performed with Ropeginterferon alfa-2b**

Type of test / study ID / GLP	**Test system**	**Concentrations / Concentration range / Metabolising system**	**Results Positive / negative / equivocal**
Gene mutations in bacteria / 7975-104 / Yes	Salmonella/E-Coli strains TA98/TA100/TA1535/TA1537/WP2*uvr*A	+/- S9 (TA98/TA100/TA1535/TA1537/ WP2*uvr*A): 33.3, 100, 333, 1000, 3330, 5000 µg/plate Plate incorporation for 52± 4 hours at 37±2°C.	Negative for relevant increase in reverse mutations. No precipitation or signs of cytotoxicity up to 5000 µg/plate.
Chromosome aberration in mammalian cells / 7975-105 / Yes	Chinese hamster ovary (CHO) cells	- S9 20h/20h recovery: 6.25, 12.5, 25, 50 µg/mL + S9 3h/20h recovery: 595, 795, 1190 µg/mL	Negative for chromosomal aberrations –S9, equivocal +S9 at > 795 µg/mL. Dose dependent increase in mitotic indices.

Ropeginterferon alfa-2b was negative in an in vitro Ames test and a chromosome aberration assay without metabolic activation. Equivocal results were obtained at high concentrations in a chromosome aberration assay with metabolic activation.

Carcinogenicity

In accordance with ICH S6 guideline no carcinogenicity studies were performed.

Reproduction Toxicity

Based on the general mechanism of action of IFN alpha, reproductive and developmental studies with ropeginterferon alfa-2b are not considered to add value to understanding of the safety and toxicity profile of ropeginterferon alfa-2b.

Toxicokinetic data

See above section on repeat dose toxicity.

Local Tolerance

The local tolerance evaluation included in the pivotal repeated dose toxicity studies were performed with formulation and route of administration intended for marketing, and thus a separate local tolerance study was not considered necessary.

Local tolerance endpoints confirmed a good tolerability of the treatment with ropeginterferon alfa-2b. A few incidences of slight erythema were related to the dosing procedure, as they occurred also in the control group (Study No. 44103-08-300). Perivascular macrophage and lymphocyte infiltrates without any macroscopic correlates were noted at the injection site at the end of the dosing phase (Study No. 7975- 101). These inflammatory reactions either resolved or were resolving after four-week recovery period.

Other toxicity studies

Analysis of humoral immune response was performed as part of the GLP toxicity studies in cynomolgus monkeys. In a pivotal toxicity study, *s.c.* treatment with ropeginterferon alfa-2b at doses of 0.675, 2 and 6.75 mg/kg consistently induced anti-drug and neutralizing antibodies by the end of the four-week treatment period, demonstrating immunogenicity of the test item.

2.3.5. Ecotoxicity / environmental risk assessment

No ERA studies are submitted, as ropeginterfereon alfa-2b is a protein molecule it is exempted from ERA assessment (see discussion on non-clinical aspects).

2.3.6. Discussion on non-clinical aspects

Interferons are used off-label for the treatment of polycythemia vera since the late 1980s. The applicant did not conduct any nonclinical proof-of-concept study and relies on the existing clinical data to support the use of Besremi (ropeginterferon alfa-2b) in the proposed indication, which was viewed as acceptable from a non-clinical perspective. It is acknowledged that the mechanism of action of ropeginterferon alfa-2b is currently not established; published data suggest that it may involve stimulation of the immune system, direct effect on mutant hematopoietic stem cells to promote their depletion from the stem cell pool, and/or abrogation the proliferative advantage of the mutant hematopoietic stem cells over the normal cells.

PK and magnitude of biological activity of ropeginterferon alfa-2b was similar to that of peginterferon alfa-2a (Pegasys) in terms of inducing an antiviral effect in a study in Cynomolgus monkeys at up to 6 days following a single 30 µg/kg sc dose. From 6 to 14days post-dose, the biological activity of ropeginterferon alfa-2b remained stable whereas that of peginterferon alfa-2a subsided gradually. This sustained response of ropeginterferon alfa-2b vs. peginterferon alfa-2a was attributed to potential differences in the biological activities of the active moieties by Silva et al (Journal of hepatology, 2006) who compared the levels of interferon-induced gene transcripts in 36 patients treated with peginterferon alfa-2b and peginterferon alfa-2a.

Safety pharmacology studies comprised an in vitro hERG assay, a cardiovascular study in cynomolgus monkeys and central nervous system and respiration studies in rats. Although the *in vitro* hERG assay revealed a signal for QT prolongation according to the Redfern et al. (2003) criteria (inhibition of hERG current by 49.9% with 2.5 µM ropeginterferon alfa-2b), this value is lower than the actual IC50 concentration; at the dose of 450 µg in patients a safety margin based on the mean patient serum Cmax of about 1028-fold could be determined compared to the used 2.5 µM in the in vitro assay. Further, in a pivotal 4-week cynomolgus toxicity study the

Cmax values of the animals exceeded the human Cmax values. Therefore, because of the high safety margin, a cardiovascular risk for humans is considered unlikely.

Another study was conducted in rats to study the safety of ropeginterferon alfa-2b in the central nervous system. A single sc administration up to the dose of 20 mg/kg did not elicit any adverse effects on central nervous system or body temperature in rats. Similarly, single iv dose of ropeginterferon alfa-2b at 10 mg/kg to rats had no negative effects on respiration. In conclusion, there was no evidence for significant adverse effects on the central nervous, cardiovascular or respiratory system in the rat. In addition, no clinical signs indicating influence of the test item on central nervous or respiratory system were noted in the toxicity studies in cynomolgus monkeys treated s.c. up to the dose of 6.75 mg/kg. However, as a causal relationship between the use of interferons alfa and beta and the development of pulmonary arterial hypertension, a rare but severe event, cannot be excluded, a relevant warning is included in the SmPC (see section 4.4).

The pharmacokinetics of ropeginterferon alfa-2b were not characterized in stand-alone studies, and it was relied on results obtained in a PD/PK and in toxicokinetic studies performed in Cynomolgus monkeys. In the PD/PK study, the absolute subcutaneous bioavailability could not be accurately determined due to the potential occurrence of anti-IFNα antibodies in animals dosed intravenously. Therefore, the bioavailability of 80% should be viewed as potentially overestimated due to an underestimated value for systemic exposure after intravenous administration. The half-life of ropeginterferon alfa-2b was in the 60-70 hours range, and the volumeof distribution values (37-55 mL/kg) indicate that its distribution is restricted to the intravascular space.

In toxicity studies, the kinetics was shown to be linear over the 0.675-6.75 mg/kg dose range in Cynomolgus monkeys treated once subcutaneously with no significant apparent gender-related difference in systemic exposure. After twice weekly dosing for 2 weeks, Cmax and AUC0-72 levels were 1.4- to 2.9-fold and 2.1- to 3.4-fold higher, respectively, than after the first dose. In the pivotal 4-week toxicity study using the same dosing frequency, systemic exposure levels measured after the last (eighth) dose were much lower than those measured after the first dose, in line with the demonstrated immunogenicity of ropeginterferon alfa-2b.

No stand-alone distribution studies were conducted; reference is made to mass balance, tissue distribution and whole body autoradioluminography studies performed in rats with peginterferon alfa-2a (Pegasys) which is distributed to the liver, kidney and bone marrow in addition to being highly concentrated in the blood. In accordance with the ICH S6 (R1) guidance, studies investigating metabolism of ropeginterferon alfa-2b were not performed as degradation of proteins to short Peptides and amino acids is well understood.

No stand-alone single dose toxicity studies were performed. The non-clinical toxicity studies conducted with ropeginterferon alfa-2b were limited due to species specificity of interferons. Acute and repeated dose toxicity studies have been carried out in rats and cynomolgus monkeys. It is known that there is no pharmacological activity of human interferon in mice and rats and since one study in rats confirmed this assumption, no further repeated dose toxicity studies were conducted in rodents. The duration of the pivotal toxicity study in Cynomolgus monkeys does not cover the chronic use of ropeginterferon alfa-2b in patients with the treatment length exceeding six months and application every two weeks because this study was limited to 4 weeks by the production of neutralizing antibodies. Therefore, the duration of this pivotal toxicity study is justified, as already agreed by CHMP scientific advice EMEA/H/SA/2272/1/2012/PA/III/ March 2012.

The toxicity studies in cynomolgus monkeys, which used clinical route of administration (s.c.) and formulation intended for marketing, did not reveal any serious adverse effects after repeated treatment up to the dose of 6.75 mg/kg. The major findings such as slight weight loss, decreased appetite, reduced red blood cell mass, reticulocytes and platelets, prolonged APTT, transient decrease in mean total protein, albumin and calcium levels in serum and ketonuria were all of mild non-adverse character and reversible after the four-week recovery

phase. Safety margin between NOAEL in the pivotal toxicity study (6.75 mg/kg) and the maximum human dose of 500 µg, i.e. 7.1 µg/kg assuming 70 kg body weight, was calculated to be 307.04. The peak concentrations in sera and exposure were vastly greater in cynomolgus monkeys than in patients, further pointing to a high safety margin of the NOAEL to the maximum applied dose of 540 µg/patient.

Two bridging studies in cynomolgus monkeys showed that clinical lots manufactured with a new producer strain and manufacturing process were comparable in terms of induced effects.

Ropeginterferon alfa-2b was negative in two *in vitro* genotoxicity assays. In accordance with guideline ICH S6 no carcinogenicity studies were performed.

No reproductive and developmental toxicity studies were performed with ropeginterferon alfa-2b. According to the mechanism of action of IFNα effects of ropeginterferon alfa-2b on the reproductive system are expectable and are properly labelled in the SmPC. Women of childbearing potential are advised to use effective contraception during treatment (sections 4.6. and 5.3). Ropeginterferon alfa-2b is not indicated for the use in paediatric patients and thus studies in juvenile animals are not considered necessary.

Analysis of humoral immune response was performed as part of the GLP toxicity studies in cynomolgus monkeys. In a pivotal toxicity study, s.c. treatment with ropeginterferon alfa-2b at doses of 0.675, 2 and 6.75 mg/kg consistently induced anti-drug and neutralizing antibodies by the end of the four-week treatment period, demonstrating immunogenicity of the test item.

The local tolerance evaluation included in the pivotal repeated dose toxicity studies were performed with formulation and route of administration intended for marketing, and thus a separate local tolerance study was not considered necessary. Local tolerance endpoints confirmed a good tolerability of the treatment with ropeginterferon alfa-2b. A few incidences of slight erythema were related to the dosing procedure, as they occurred also in the control group. Perivascular macrophage and lymphocyte infiltrates without any macroscopic correlates were noted at the injection site at the end of the dosing phase. These inflammatory reactions either resolved or were resolving after four-week recovery period.

Ropeginterferon alfa-2b is a mono-PEGylated natural protein and thus belongs to the group of compounds exempted from a Phase I and/or II ERA according to 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 data submitted to address the non-clinical aspects of ropeginterferon alfa-2b are sufficient. All relevant information is reflected in the SmPC.

2.4. Clinical aspects

2.4.1. Introduction

GCP

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

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

- Tabular overview of clinical studies

Clinical Study (Phase)	Study Design	Status
PEGINVERA Ph 1/2	**Stage I** dose finding part • 25 PV patients; HU naïve or pre-treated with HU • 3+3 dose escalation with 3 patients per cohort to determine the MTD (8 dose levels 50 to 540 µg) **Stage II** cohort extension • Additional 26 PV patients; HU naïve or pre-treated with HU • Determination of long term efficacy (up to 1 year and longer) at individually adjusted optimum dose levels. *Treatment schedule:* IMP: ropeginterferon alfa-2b provided in vials at a concentration of 500 µg/mL. One injection (100 to 450 µg) every two weeks for the first 12 months. Thereafter, patients benefitting from treatment were allowed to switch to a once every 4 weeks dosing schedule.	Stage 1: **completed** Stage 2: **completed** 51 patients treated
PROUD-PV Ph 3	• Randomized, controlled study • 257 PV patients; HU naïve or pre-treated with HU • 12 months treatment Test IMP: ropeginterferon alfa-2b s.c., 50 µg to 500 µg every two weeks depending on response and tolerability; provided in vials at a concentration of 0.5 mg/mL Reference IMP: HU per os, 500 mg to 3000 mg daily depending on response and tolerability	**Completed** 257 patients randomized (enrolled) and 254 patients treated (127 patients in the ropeginterferon alfa-2b arm, 127 patients in the HU arm)
CONTINUATION-PV Ph 3b	• Open-label extension study to PROUD-PV Study • 171 PV patients who completed the 12 months treatment of the PROUD-PV Study rolled over into the CONTINUATION-PV Study • Patients who received ropeginterferon alfa-2b in PROUD-PV Study continue to receive ropeginter-feron alfa-2b in the CONTINUATION-PV Study and are treated at individual tolerable dose levels as determined in PROUD-PV. Patients who received HU during the PROUD-PV Study are assigned to the BAT (best available treatment/standard of care) arm. Test IMP: Initially, ropeginterferon alfa-2b was provided in vials at a concentration of 0.5 mg/mL. After availability of results from the PEN-PV Study, patients were allowed to switch to a pre- filled pen (250 µg pen, concentration: 0.5 mg/mL). Patients may be in a 2-, 3- or 4-weeks dosing schedule Comparator: standard first line treatment for treatment of PV disease, as per investigator's discretion	**Ongoing** 171 patients enrolled and treated with ropeginterferon alfa-2b (N=95) respectively best available treatment (N=76) 147 still on study (36-month treatment analysis population; ropeginterferon alfa-2b N=78, BAT N=69)
PEN-PV Ph 3	• Single-arm uncontrolled study • 36 PV patients who completed the ropeginterferon alfa-2b arm of PROUD-PV Study or were enrolled in the ropeginterferon alfa-2b arm of the CONTINUATION-PV Study • Treatment at individual tolerable dose levels determined in PROUD-PV Study • 3 months treatment • Patients performed supervised and unsupervised self-administrations of ropeginterferon alfa-2b with	**Completed** 36 patients enrolled and treated

	the pre-filled pen IMP: ropeginterferon alfa-2b provided as pre- filled pen (250 µg pen, concentration: 0.5 mg/mL).	

2.4.2. Pharmacokinetics

Absorption

Bioavailability

Formal human bioavailablity (BA) studies have not been performed.

- Bioequivalence

No clinical bioequivalence (BE) studies have been performed or *in vitro* dissolution profiles.

A comparative PK study against Pegasys has been performed to evaluate PK parameters of P1101.

Study A09-102-P1101

Design: single centre, double-blind, randomized, active control, single dose escalation study, with P1101 as the test drug and PEGASYS as the active control. The study included dose levels from 0.4 to 4.5 µg/kg (24 to 270 µg, total dose based on a 60 kg subject). These dose levels are comparable to those evaluated in the first clinical study with PEGASYS, an approved PEG-IFN product manufactured with a comparable molecular weight polyethylene glycol.

Subjects: A total of 48 healthy male adult subjects aged 18 to 45 years.

Table 12: Study Treatments (sc administration)

Treatment	Description	Lot number/ expiration date
P1101	PEG-P-IFNα-2b (PEGylated Proline- interferon alpha-2b recombinant) solution for injection, 180 ug/mL, 1.2 mL/vial (test)	08DPL-B002/ 6/24/ 2010
PEGASYS®	(recombinant) for injection, 180 ug/mL, 1.0 mL/vial (Hoffmann-Roche Ltd., Canada) (reference)	B10087 (vial) B1008B017 (box)/ SE2010
Diluent	4.05 mL/vial	08DFL-P001B / 7/23/2010

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Randomisation: Subjects meeting all eligibility requirements were assigned a randomization number in consecutive order Subjects in the same cohort received the same dose of either P1101 or PEGASYS®, with the doses of P1101 ascending in subsequent cohorts. Cohorts comprised 8 subjects where 6 subjects were randomized to receive P1101 and 2 subjects were randomized to receive PEGASYS®. The two sentinel subjects in Cohort 1 were randomized in a 1:1 ratio. The remainder of the subjects, dosed 72 hours later, were randomized in a 5:1 ratio in order to maintain an overall 6:2 (=3:1) ratio in Cohort 1.

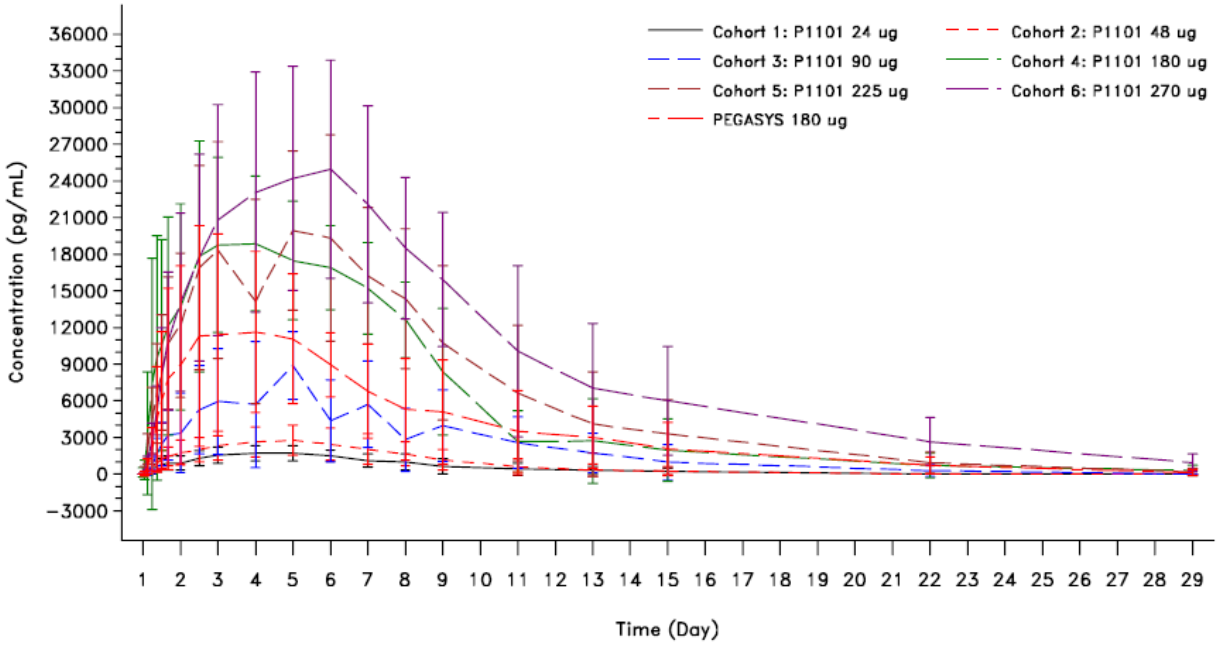
PK blood sampling: Serum samples collected within 1 hour pre-dose, and at 1, 3, 6, 9, 12, 16, 24, 36, 48, 72, 96, 120, 144, 168, 192, 240, 288, 336, 504, and 672 hours post-dose.

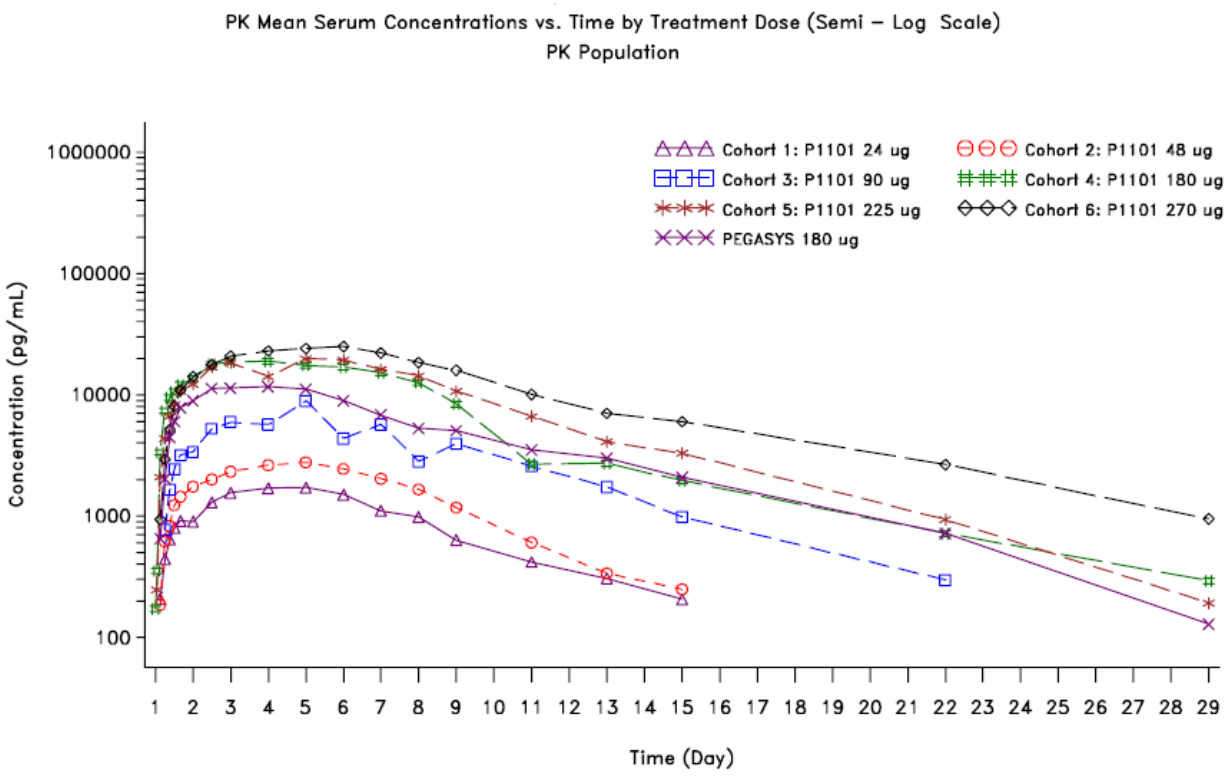
PD blood sampling: samples were collected within 1 hour pre-dose and at 24, 48, 72, 96, 120, 168, 240, 336, 504, and 672 hours post-dose.

Disposition of subjects: Two subjects were lost to follow-up: 1 subject, 001-405, receiving PEGASYS® (Day 18), and 1 subject, 001-501, receiving 225 µg P1101 (Day 13); 1 subject, 001-205, randomized to receive 48 µg withdrew consent (Day 27). All subjects who did not complete the study were dosed and were included in the safety, pharmacokinetic, and pharmacodynamic populations.

Results: Linear and semi-log scale plots of the mean serum concentrations versus time for Cohorts 1 through 6 and PEGASYS® are shown below in the figures below.

PK Mean Serum Concentrations (+/−SD) vs. Time by Treatment Dose (Linear Scale)
PK Population





Mean serum concentration time curves were comparable across treatments. Serum compound concentrations reached a peak at approximately 3 to 5 days after dosing and declined gradually, with elimination half-lives of 60 to 118 hours.

Pharmacokinetic parameters are summarized in the Table 13 below:

Table 13 Study A09-102-P1101 Summary of Mean (SD) PK Parameters by Treatment in HV

Cohort	C_{max} (ng/mL)	T_{max} (hr)	C_t (ng/mL)	λ_z Ke (hr-1)	T½ (hr)	AUC_{0-t} (ng•hr/mL)	AUC (ng•hr/mL)
P1101 24 µg							
N	5	5	5	4	4	5	4
Mean	1.78	86.74	0.61	0.0115	85.21	272.7	372.3
SD	0.48	47.23	0.14	0.0066	62.37	115.1	118.4
P1101 48 µg							
N	5	5	5	4	4	5	4
Mean	2.80	91.35	0.55	0.0171	61.35	466.9	625.2
SD	1.23	20.28	0.23	0.0112	47.99	230.3	115.5
P1101 90 µg							
N	5	5	5	5	5	5	5
Mean	6.64	74.52	0.69	0.0098	78.54	1407.0	1492.4
SD	4.14	42.17	0.33	0.0037	26.64	1056.8	1084.9
P1101 180 µg							
N	5	5	5	5	5	5	5
Mean	20.7	79.22	2.27	0.0162	66.53	3772.1	3945.5
SD	7.66	31.27	3.81	0.0104	48.49	1102.4	1000.8
P1101 225 µg							
N	6	6	6	5	5	6	5
Mean	21.26	88.31	1.35	0.0078	91.68	4290.3	4820.7
SD	8.23	24.77	1.83	0.0016	18.49	1982.4	1912.4
P1101 270 µg							
N	5	5	5	5	5	5	5
Mean	24.84	115.69	1.05	0.0076	118.22	6068.1	6258.0
SD	8.66	31.38	0.54	0.0058	45.74	1465.2	1540.0
PEGASYS® 180 µg							
N	11	11	11	9	9	11	9
Mean	12.95	84.25	1.42	0.0123	89.32	2343.8	2706.0
SD	8.46	26.97	1.30	0.0114	45.13	1104.8	1149.9

Data source: Table 14.2.1.2

The P1101 geometric mean values for Cmax, AUC, and AUC0-t showed an increase of 76%, 66%, and 82%, respectively over PEGASYS at the 180 µg dose. The geometric mean ratios for AUC and AUC0-t indicate no significant statistical differences (p-values of 0.2398 and 0.0687, respectively) at the 180 µg dose level. On the other hand, the Cmax geometric mean ratio is 1.76 (with an increase of 76% over PEGASYS), and this ratio is statistically significant (p-value of 0.0275) at 180 µg dose level. Tmax values were also compared between P1101 (combined and each dose level) and PEGASYS® using Wilcoxon Rank-Sum; however, Tmax values were not log-transformed. These results indicate that the time to maximum P1101 (Tmax) concentration was independent of dose (74.5-115.7 hours) and was similar to PEGASYS (84.3 hours).

Distribution

Plasma protein binding

Interferons (alpha/beta [type I] and gamma [type II] interferons) are blood (glyco)proteins which belong to the protein class of cytokines that are produced by the immune system in response to various stimuli and act as

short distance mediators which are usually present in plasma only during a short period of time. Interferon alpha exhibit their cellular effects by binding to the transmembrane receptor IFNAR which initiates a downstream signalling cascade through the activation of various effector proteins. The specific binding of interferon alpha to IFNAR would suggest that co-binding to another protein would preclude the ligand-receptor interaction. Therefore, interferon alpha is not considered to be bound and transported by other blood proteins. This is further supported by the attempts made to generate fusion proteins with recombinant HSA and recombinant interferon alfa to improve the half-life of interferon alfa (IntronA, recombinant interferon alfa, has a half-life of 2 – 3 hours in humans, in comparison, HSA, has a half-life of 19 days) (Osborn et al., 2002; Bain et al., 2006). Hence, plasma protein binding studies have not been performed with ropeginterferon alfa-2b

Elimination

Elimination studies have not been submitted.

Dose proportionality and time dependencies

Table 14: Analysis of dose proportionality in PK study A09-102-P1101; all cohorts.

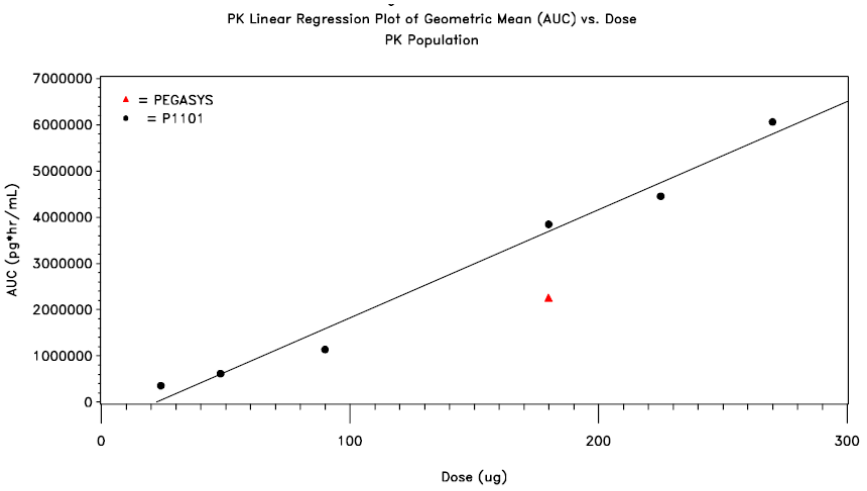
Table 10 Analysis of Dose Proportionality for All the Cohorts

Parameter	Slope	95% Confidence Interval
ln (AUC)	1.2200	1.0047–1.4353
ln (AUC_{0-t})	1.3611	1.1117–1.6105
ln (C_{max})	1.1890	0.9890–1.3890

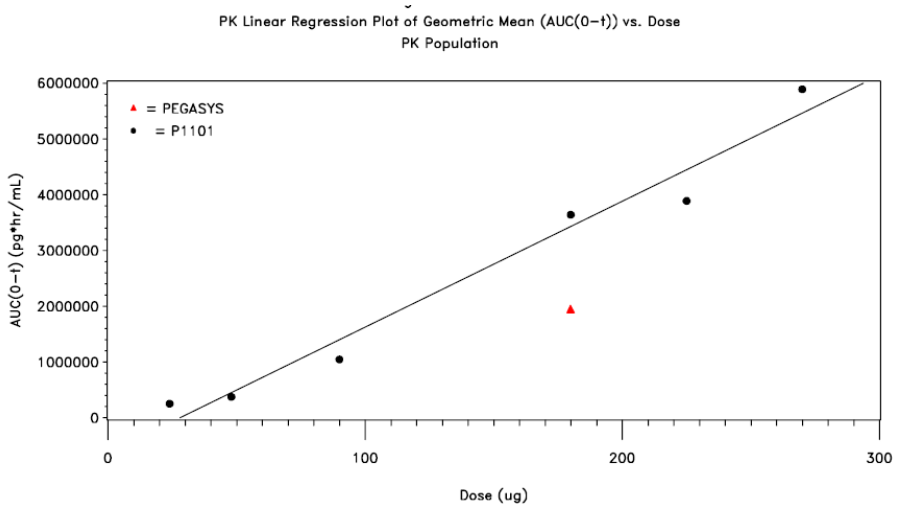
Data source: Table 14.2.1.3

The dose proportionality analysis for P1101 suggests more than proportional kinetics with increasing dose, as the slopes of the power model ranged from 1.19 to 1.36

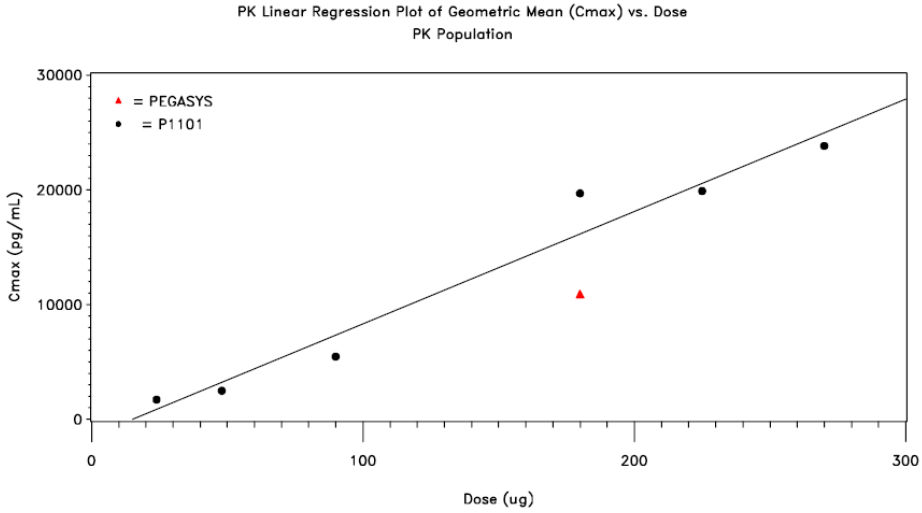
Figure 7: Analysis of dose - proportionality



Regression Equation:
Geometric_Mean = −520010.2 + 23413.88*Dose



Regression Equation:
Geometric_Mean = −634226.1 + 22593.95*Dose



Regression Equation:
Geometric_Mean = −1471.628 + 97.95697*Dose

Intra and inter- subject variability

The pharmacokinetic parameter analysis from study A09-102-P1101 indicated that the inter-subject variability in Cmax, AUC, and AUC0-t for PEGASYS® (mean coefficient variations of 60%, 82%, and 77%, respectively) is higher than that of P1101 (mean coefficient variations of 35%, 25%, and 30%, respectively).

Pharmacokinetics in the target population

Study PEGINVERA: "An open-label, prospective, multicentre, phase I/II dose escalation study to determine the maximum tolerated dose and to assess the safety and efficacy of P1101, PEG-Proline-IFN-α-2b in patients with Polycythaemia Vera".

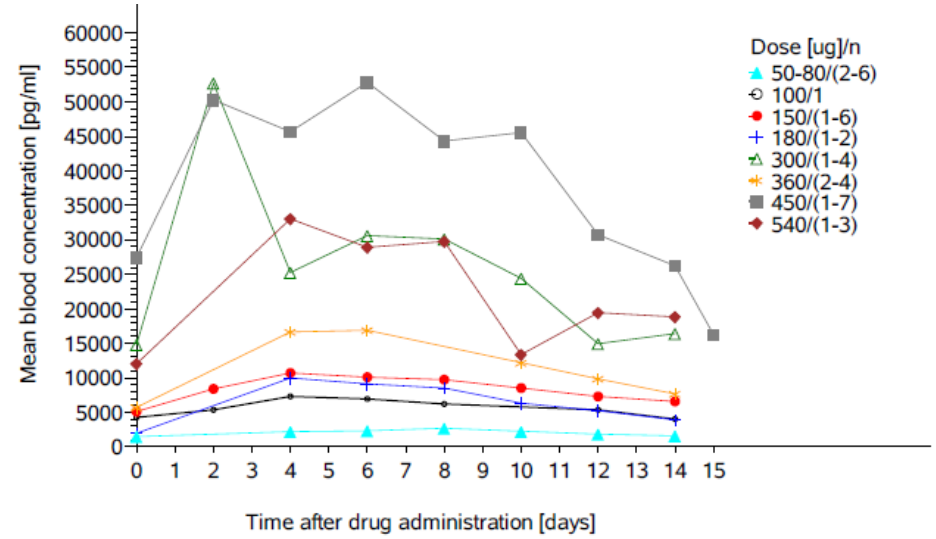
The PEGINVERA Study was conducted as open 3+3 dose escalation multicentre study in Austria with three patients per cohort to determine the maximum tolerable dose (MTD), followed by long-term treatment with an intra-patient dose escalation scheme to the individual tolerable level. Evaluable PK data was obtained from 19 of

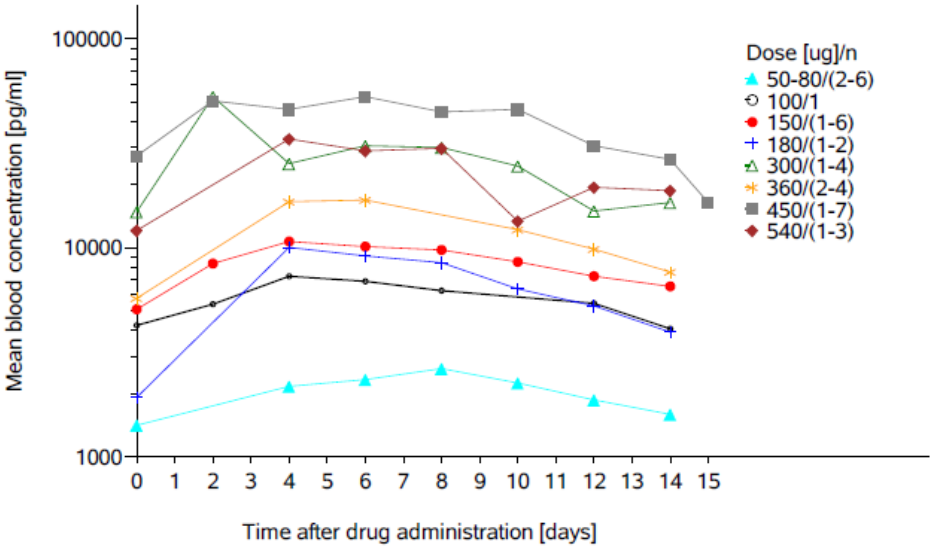
21 patients treated with 50 – 540 µg (administered every two weeks), with the exclusion of data from two patients, in whom concentrations were below the limit of quantification.

Table 15*: Study PEGINVERA: Summary of PK parameters of ropeginterferon alfa-2b in PV patients*

Dose, µg	50–80 (n = 3)	100 (n=1)	150 (n=3-4)	180 (n=1)	300 (n=3)	360 (n=2)	450 (n=5)	540 (n=2)
C_{max} (pg/mL)	2393.3 (712.1–4074.6)	7320 (-)	10957.5 (9403.4–12511.6)	10000 (-)	35866.7 (–74269.2)	17250 (–64801.6)	48640 (20490.8–76789.2)	33750 (–257936)
t_{max} (days)	6 (-)	4 (-)	4.3 (2.2–6.3)	3 (-)	4 (–13.2)	5 (-)	5.6 (4.9–6.3)	4.5 (–38.2)
C_t* (pg/mL)	1596.7 (656.5–2536.8)	4080 (-)	6475 (3065.5–9884.5)	3950 (-)	16466.7 (6002.8–26930.6)	7680 (–20584)	25440 (9730.5–41149.5)	15905 (–180301)
λ_z	0.093 (–0.203)	0.1009 (-)	0.089 (–0.229)	0.1045 (-)	0.092 (0.006–0.179)	0.116 (0.015–0.218)	0.088 (0.067–0.110)	0.09 (–0.501)
$t_{1/2}$ (days)	8.9 (–24.9)	6.9 (-)	10 (–33.4)	6.6 (-)	8.2 (1.0–15.5)	6 (0.8–11.2)	8.2 (5.8–10.5)	8.1 (–44.8)
AUC_{0-t} (pg.h/mL)	28546.7 (7794.1–49299.2)	82235 (-)	118297.5 (101312.1–135282.9)	97935 (-)	369600 (41942.7–697257.3)	181495 (–609389.6)	552570 (224371.3–880768.7)	334375 (–2849366.4)
AUC_{0-t} (pg.h/mL) per day	2074.8 (735.3–3414.3)	5873.9 (-)	8601.7 (7534.4–9669.0)	6995.4 (-)	27026.2 (3959.5–50092.9)	12963.9 (–43527.8)	38713.9 (14831.2–62596.5)	23883.9 (–203526.2)
$AUC_{0-\infty}$ (pg.h/mL)	49930.6 (2355.3–97506.0)	122653.5 (-)	210358.4 (–515123.4)	135744 (-)	569071.2 (191987.4–946155.0)	248254.8 (–902623.2)	844175.5 (343954.0–1344397.1)	537523.2 (–5975587.4)
R^2	0.9 (0.7–1.1)	0.9 (-)	1 (0.9–1.0)	1 (-)	1 (0.9–1.0)	1 (0.8–1.1)	0.9 (0.8–1.0)	1 (1.0–1.0)

Figure 8: Mean blood concentration profiles on linear and semi-logarithmic scale

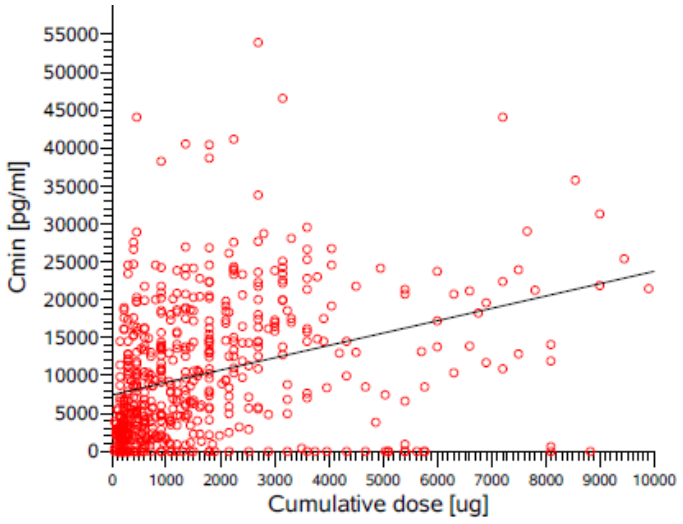




The mean t½ values ranged from 6 - 10 days and did not appear to depend upon the administered dose.

Correlation coefficients between dose administered and concentration level showed a trend to increase with the order of consecutive dosing, that might indicate a cumulative effect over time.

Figure 9: Scatter plots of blood concentrations before next study drug administration (Cmin) vs. cumulative dose (all analyzed data)



Correlation coefficient between cumulative dose and level of concentration r=0.357, Spearman p-value <.001
C_{min} - blood concentration level measured before next study drug administration
Analysis set: All patients with available data, Production date: 22APR2016

Table 16: PK comparison across both studies

Parameter	PEGINVERA Study (AOP-sponsored)	PK Study (non-AOP sponsored)
	(50 µg to 540 µg sc injection, once every two weeks)	(24 µg to 270 µg sc injection, single dose)
C_{max} (ng/ml)	2.39 to 48.64	1.78 to 24.84
t_{max} (d)	3 to 6	3.1 to 4.8
$t_{1/2}$ (d)	6.0 to 10.0	2.5 to 4.9

Special populations

Table 17: Special populations

	Age 65-74 (Older subjects number /total number)	Age 75-84 (Older subjects number /total number)	Age 85+ (Older subjects number /total number)
PK Trial A09-102-P1101	0	0	0
PK evaluation as part of the PEGINVERA study	5	3	0

Pharmacokinetic interaction studies

No PK interaction studies have been submitted.

Pharmacokinetics using human biomaterials

Not applicable.

2.4.3. Pharmacodynamics

Mechanism of action

No studies on mechanism of action have been performed.

Primary and Secondary pharmacology

The proposed biological activity of ropeginterferon alfa-2b in terms of primary pharmacodynamic effects was investigated in PV patients in 3 studies [2010-018768-18 (PEGINVERA), 2012-005259-18 (PROUD-PV), 2012-005259-18 (CONTINUATION-PV)]. The primary pharmacodynamic effect was evaluated by a reduction in mutant JAK2 allelic burden, which is assumed to reflect a reduction of the malignant clone in the bone marrow causing the aberrant haematopoiesis in PV (see above '2.1 Problem statement – Pathophysiology').

The effects of ropeginterferon alfa-2b on JAK2 mutant allelic burden were variable and values fluctuated over the course of the studies. However, taken together the presented results show a sufficient reduction of JAK2 mutant allelic burden by IFN alpha 2-b being acceptable as proof of concept.

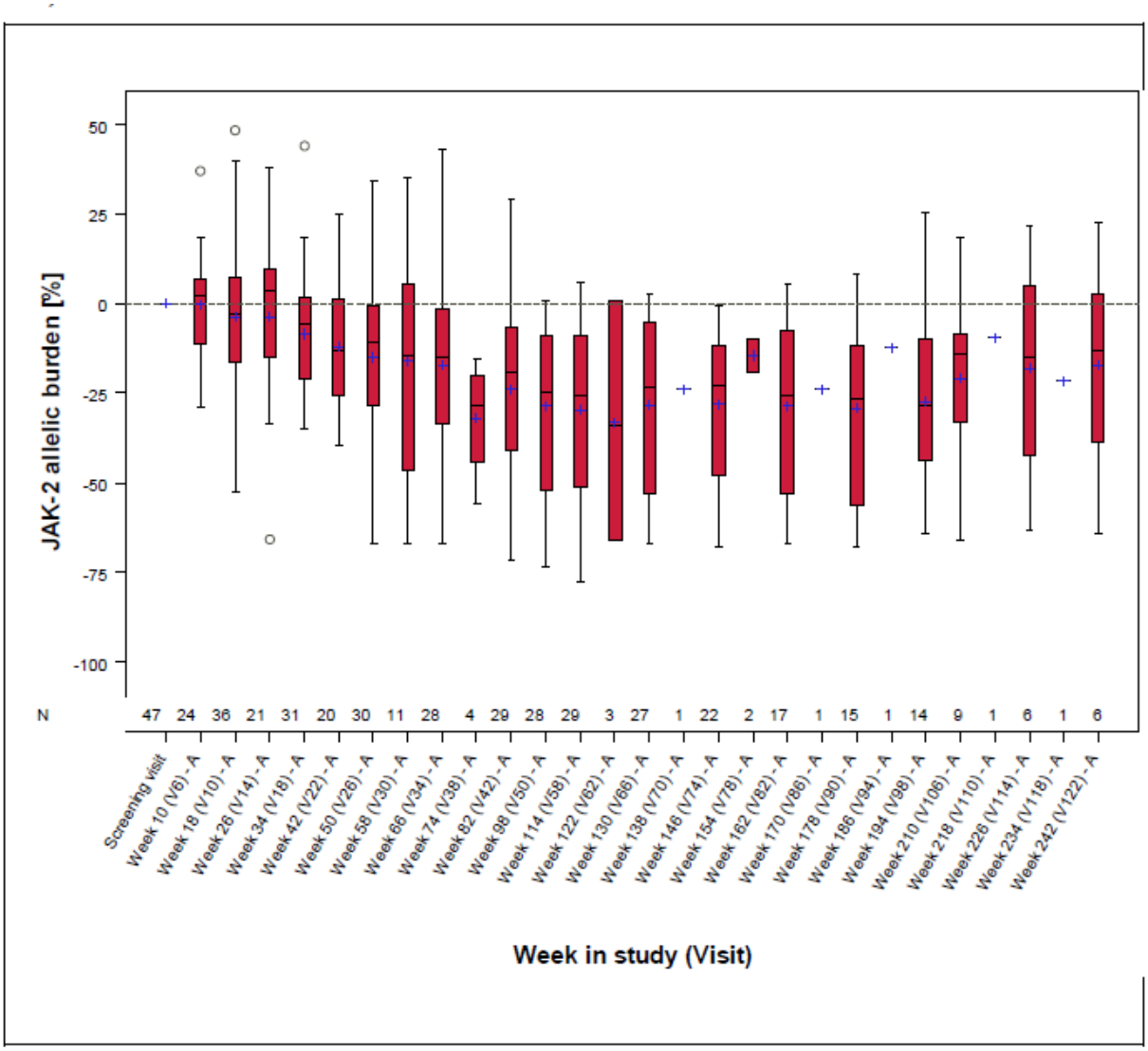
No relevant data regarding the proposed biological activity of ropeginterferon alfa-2b in terms of secondary pharmacology are presented by the applicant.

Pharmacodynamic interactions with other medicinal products have not been investigated. In their responses the applicant discussed that the shared structural similarities with pegIFN alfa-2b in respect of the PEG size and type of PEG linkage should provide evidence that the PD interactions of ropeginterferon alfa-2b and pegIFN alfa-2a are comparable.

With regard to genetic differences the majority of PV patients display the same molecular aberration i.e. the JAK2V617F mutation (>95%) and hence similarity in PD response is expected. Infrequently, alternative targets in the JAK-STAT pathway are affected by somatic mutations in PV. However, JAK2V617F is considered as the genetic determinant for the diagnosis of PV according to the WHO 2008 criteria.

Primary pharmacology data were obtained from the PEGINVERA study

Figure 10: Relative JAK2 allelic burden: boxplot of changes from baseline



Source: PEGINVERVA STAGE 2 CSR Figure 14.2.10.2.1

In study 2012-005259-18 (PROUD-PV) JAK2 allelic burden was measured at baseline and then every six months (i.e. at month 6 and month 12). Since the PROUD-PV study lasted 12 months, the amount of data regarding JAK2 allelic burden during the study is very limited.

A statistically significant (p=0.0045) difference of JAK2 allelic burden was observed at week 26 of treatment (34.6% and 27.4% in the ropeginterferon alfa-2b or HU treatment arm, respectively). However, the difference was no longer statistically significant at EOT with 30.7% and 25.9% for ropeginterferon alfa-2b or HU treated patient.

In study 2012-005259-18 (CONTINUATION-PV) open-label extension study to PROUD-PV study assessing the long-term efficacy and safety of ropeginterferon alfa-2b and best available treatment (BAT) in PV patients who previously participated in the PROUD-PV study; In ropeginterferon alfa-2b arm, the median absolute levels of JAK2 allelic burden ranged from 9.5% (Month 36) to 23.7% (Month 12). JAK2 allelic burden was measured at baseline and then every six months. The median differences from baseline (Month 0, baseline median 37.3%)

ranged from -25.4% (Month 36) to -12.1 (Month 12). In BAT arm, the median absolute levels of JAK2 allelic burden ranged from 18.2% (Month 12) to 42.3% (Month 36). The median differences from baseline (Month 0, baseline median 38.1%) ranged from -18.7% (Month 12) to 0.3% (Month 36).

Comparison of results across studies

During AOP's clinical development program, the stringent requirement for a complete molecular response (i.e. undetectable levels of JAK2 mutant alleles) was only met by a small number of patients, i.e. only seen in the long-term PEGINVERA Study. The less stringent requirement for a partial molecular response (i.e., a relative reduction by 50% of the JAK2 mutant alleles) varied widely, which translated into a highly variable number of partial molecular responders (i.e., 20-60%) over the course of the studies. In general, the observed decrease in JAK2 allelic burden compared to baseline, ranged between 10-30% in all studies. Sustained molecular response is expected as long-term treatment effect (treatment duration >18 months).

2.4.4. Discussion on clinical pharmacology

Overall, the applicant provided minimal own PK data for ropeginterferon alfa-2b in this MAA dossier. It is acknowledged that PK evaluation for Besremi was performed in a rare orphan disease population and therefore limitations are obvious. Moreover, the necessity for dose-titrating has hindered the PK characterisation.

The absorption of ropeginterferon alfa-2b is sustained in patients with peak serum concentrations reached after 3 to 6 days. The absolute bioavailability of subcutaneous administered ropeginterferon alfa-2b was not investigated in humans. Thus, no valid estimation of the absolute bioavailability could be done. Based on data in monkeys, it is approx. 80%, similar to that seen for PEGylated interferon alfa-2a.

Ropeginterferon alfa-2b is found mainly in the bloodstream and extracellular fluid as seen by the volume of distribution at steady-state (V_d) of 6.6 to 17 litres in patients after subcutaneous administration (dose range 50 – 450 micrograms). Mean C_{max} was 2.4 ng/ml (with a dose of 50 – 80 micrograms) to 49 ng/ml (with a dose of 450 micrograms) and AUC_{0-t} ranged from 28.5 ng.h/ml (with a dose of 50 – 80 micrograms) to 552.6 ng.h/ml (with a dose of 450 micrograms) in patients after subcutaneous multiple dose administration. Inter-subject variability was observed with 25-35% for AUC and C_{max}, respectively, in healthy volunteers.

Comparisons of ropeginterferon alfa-2b with marketed PEGylated and un-pegylated interferons are provided which allow further conclusions for Besremi.

The elimination of ropeginterferon alfa-2b is not fully characterised. Studies with a similar interferon alfa medicinal product (pegylated interferon alfa-2a) indicated that the kidney is a major organ for excretion of radiolabelled metabolic products (study in rats) and that the systemic clearance of pegylated interferon alfa-2a in humans is about 100-fold lower compared to the native, un-pegylated interferon alfa-2a.

After subcutaneous multiple dose administration (dose range 50 – 450 micrograms), the terminal half-life of ropeginterferon alfa-2b in patients is approximately 6 to 10 days and the clearance of ropeginterferon alfa-2b is 0.023 to 0.061 L/h.

The involvement of transport proteins in absorption, distribution and elimination of ropeginterferon alfa-2b is not known.

Over a dose range of 24 to 270 micrograms, ropeginterferon alfa-2b C_{max} increased proportionally with dose in a pharmacokinetic study with healthy subjects. A higher than proportional increase in exposure was observed. Inter-subject variability for ropeginterferon alfa-2b was 35% (C_{max}) and 25% (AUC).

Comparable exposure and pharmacokinetic profile was reported for another interferon alfa medicinal product (pegylated interferon alfa-2a) in cirrhotic (Child-Pugh A) and non-cirrhotic patients. Pharmacokinetics were not evaluated in patients with increased severity of hepatic impairment.

The pharmacokinetic profile in patients with moderate or severe renal impairment and in patients with end stage renal disease (ESRD) has been evaluated only for other pegylated interferon alfa medicinal products.

Patients with moderate or severe renal impairment receiving 180 micrograms of pegylated interferon alfa-2a once weekly showed a comparable or 60% higher drug plasma exposure, respectively, compared to subjects with normal renal function.

In 13 patients with ESRD requiring chronic haemodialysis, administration of 135 micrograms pegylated interferon alfa-2a once weekly resulted in a 34% lower drug exposure than in patients with normal renal function.

Patients with renal impairment receiving a single dose of 1.0 micrograms/kg pegylated interferon alfa-2b showed an increased relation of C_{max}, AUC, and half-life to the degree of renal impairment. Following multiple dosing of pegylated interferon alfa-2b (1.0 micrograms/kg subcutaneously administered every week for four weeks), the clearance of pegylated interferon alfa-2b was reduced by a mean of 17% and 44% in patients with moderate or severe renal impairment, respectively, compared to subjects with normal renal function. Based on single dose data, clearance was similar in patients with severe renal impairment not on haemodialysis and in patients who received haemodialysis.

Only limited pharmacokinetic data are available from the use of ropeginterferon alfa-2b in the elderly. Based on the results from the PROUD-PV and CONTINUATION-PV Study on drug exposure, pharmacodynamic response and tolerability, a dose adjustment for ropeginterferon alfa-2b is not considered necessary in the elderly population.

The pharmacokinetic profile of ropeginterferon alfa-2b has not been determined in obese and underweight patients.

Primary pharmacology data regarding reduction in mutant JAK2 allelic burden are acceptable as proof of concept. JAK2V617F mutation is considered as the genetic determinant for the diagnosis of PV according to the WHO 2008 criteria, hence, with regard to genetic differences, for the majority of PV patients (>95%) similarity in PD response is expected.

Enzymes of the protein catabolism are considered to be involved in the metabolism of ropeginterferon alfa-2b. The involvement of transport proteins in absorption, distribution and elimination of ropeginterferon alfa-2b is not known. Interferon alfa has shown to influence the activity of cytochrome P450 (CYP) isozymes CYP1A2 and CYP2D6.

Pharmacokinetic and pharmacodynamic interactions with other medicinal products have not been investigated with ropeginterferon alfa-2b. The applicant discussed differences and similarities between the several un-pegylated and pegylated available interferons with regard to labelled dosing recommendations and other information for patients with hepatic and renal impairment - based on the shared structural similarities with pegIFN alfa-2b the applicant provided evidence that the PD interactions of ropegIFN alfa-2b and pegIFN alfa-2a should be comparable and relevant information was included in the SmPC (see section 4.5).

Pharmacokinetic interaction studies in humans with pegylated interferon alfa-2a indicated a moderate inhibitory effect on substrates metabolised by CYP1A2 and CYP2D6 (see section 4.5).

Co-administration of pegylated interferon alfa-2a with telbivudine in patients with hepatitis B increased the risk of developing peripheral neuropathy. A combination therapy with telbivudine and ropeginterferon alfa-2b is contraindicated (see section 4.3).

Administration of 180 micrograms of pegylated interferon alfa-2a once weekly for 4 weeks in healthy male subjects did not show any effect on mephenytoin, dapsone, debrisoquine and tolbutamide pharmacokinetics profiles, suggesting that pegylated interferon alfa-2a has no effect on *in vivo* metabolic activity of cytochrome P450 (CYP) 3A4, 2C9, 2C19 and 2D6 isozymes. In the same study, a 25% increase in the AUC of theophylline (CYP1A2 substrate) was observed, demonstrating that pegylated interferon alfa-2a is an inhibitor of CYP1A2 activity. Co-administration of pegylated interferon alfa-2b showed no significant interaction with tolbutamide (CYP2C9 substrate), midazolam (CYP3A4 substrate), dapsone (N-acetyltransferase substrate) and modestly increased the exposure of caffeine (CYP1A2 substrate) and desipramine (CYP2D6 substrate). Therefore, care should be taken when ropeginterferon alfa-2b is co-administered with CYP1A2 substrates notably those having a narrow therapeutic margin such as theophylline or methadone. Likewise, caution is recommended with CYP2D6 substrates (e.g. vortioxetine, risperidone) combined with ropeginterferon alfa-2b. Ropeginterferon alfa-2b may inhibit the activity of CYP1A2 and CYP2D6 and thus may increase the blood concentrations of these medicinal products (see SmPC section 4.5).

No dose adaptions for ropeginterferon alfa-2b should be necessary when concomitantly administered with medicinal products metabolised via CYP2C9/19, CYP3A4 or by N-acetyltransferase. Caution must be exercised when administering ropeginterferon alfa-2b in combination with other potentially myelosuppressive/ chemotherapeutic agents. Narcotics, hypnotics or sedatives must be administered with caution when used concomitantly with ropeginterferon alfa-2b. (see SmPC section 4.5).

Similarly, as the applicant did not have own data on patients with renal or hepatic impairment (although renal and hepatic impairment were no special exclusion criteria), recommendations from the interferon 2a PI apply.In patients with compensated cirrhosis (i.e. Child-Pugh A), another pegylated interferon alfa medicinal product (pegylated interferon alfa-2a) has been shown to be safe. No ropeginterferon alfa-2b dose adjustment is required for adult patients with mild liver impairment. The use of interferon alfa has not been evaluated in patients with decompensated cirrhosis (i.e. Child-Pugh B or C) and is contraindicated in these patients (see section 4.3). Increased liver enzyme levels have been observed in patients treated with ropeginterferon alfa-2b. When the increase in liver enzyme levels is progressive and persistent, the dose should be reduced. If the increase in liver enzymes is progressive and clinically significant despite dose reduction, or if there is evidence of hepatic decompensation, therapy should be discontinued (see section 4.4). No dose adjustment for ropeginterferon alfa-2b is required for adult patients with mild (GFR 60-89 mL/min) or moderate (GFR 30-59 mL/min) renal impairment. A reduced starting dose for ropeginterferon alfa-2b of 50 micrograms is recommended for patients with severe (GFR 15-29 mL/min) renal impairment. Ropeginterferon alfa-2b is contraindicated in patients with end stage renal disease (GFR <15 mL/min) (see section 4.3).

Adjustments in the recommended dose for ropeginterferon alfa-2b are not necessary when starting therapy in elderly patients (see section 5.2). The pharmacokinetic profile of ropeginterferon alfa-2b has not been determined in obese and underweighted patients. No recommendation on dose adjustment for ropeginterferon alfa-2b can be given for these patients. The safety and efficacy of Besremi in children and adolescents have not been established. No data are available (see section 4.4).

Furthermore, as no PK data are available with the 1 mg/ml strength, the applicant will provide full PK data with this strength within the ongoing CONTINUATION-PV study. It is planned to collect PK data from both the 500 µg/ml then to 1 mg/ml strengths in order to give reassurance that change in the concentration of the ropeginterferon alfa-2b solution is unlikely to cause significant differences in PK properties after s.c

administration in humans. The proposed PK blood sampling (the same as the one used in in the PEGINVERA Study) would be insufficient to fully characterise the whole PK profile of ropeginterferon alfa-2b, especially with regards to the terminal elimination half-life. Therefore, the applicant committed to extend the proposed PK sampling to reliably estimate the terminal elimination half-life of ropeginterferon alfa-2b, and to include additional intensive PK sampling to estimate the systemic exposure (AUCss, Cmax, Cmin) during the interval between two administrations.

2.4.5. Conclusions on clinical pharmacology

From a clinical pharmacology point of view the data submitted are sufficient.

The CHMP considers the following measures necessary to address the issues related to pharmacology:

- The applicant will further evaluate PK the higher doses in PV patients. Furthermore, as no PK data are available with the 1 mg/ml strength, the applicant commits to provide full PK data with this strength (and the 0.5 mg/ml strength) to give reassurance that change in the concentration of the ropeginterferon alfa-2b solution is unlikely to cause significant differences in PK.

2.5. Clinical efficacy

2.5.1. Dose response study(ies)

Dose finding in study PEGINVERA-PV

Dose finding aspects were part of the PEGINVERA-PV trial which was an open-label, multicentre small-scale phase I study of 51 patients investigated ropeginterferon alfa-2b (named AOP2014 during the clinical development) in long-term treatments of PV (a 2-week regime followed by a 4-week regime).

The study consisted of two stages, corresponding to the primary objective (establishing MTD in Stage 1; N=25) and secondary objectives (PK evaluation during longer treatment in Stage 2; N=51).

In Stage 1, a MTD approach was used in a standard 3+3 dose escalation process and the MTD was defined as the highest dose at which there is at most one patient with a dose limiting toxicity out of 6 patients.

25 evaluable patients entered the stage I dose finding part and the following dose levels have been explored: 50 µg, 100 µg, 150 µg, 225 µg, 300 µg, 360 µg, 450 µg and 540 µg (one treatment cycle, i.e. 2 weeks). From the results the MTD was set to 540 µg, although no dose-limiting toxicity was observed at this dose.

2.5.2. Main study(ies)

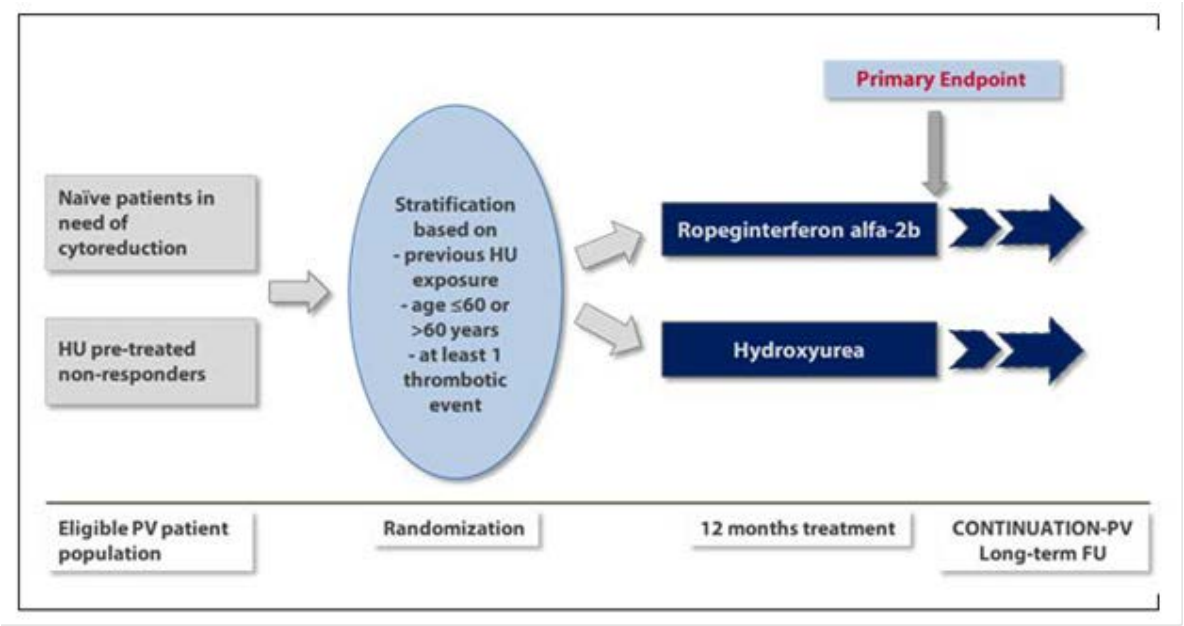
PROUD-PV

This was an open-label, randomized, controlled, parallel-group, non-inferiority study comparing the efficacy and safety of ropeginterferon alfa-2b over hydroxyurea over 12 months.

Methods

Patients at EOT at 12 months who responded on treatment with ropeginterferon alfa-2b had the opportunity to continue treatment in the ongoing extension trial CONTINUATION-PV (supportive).

Figure 11: Study design



Study Participants

Main Inclusion Criteria

- Male or female, 18 years or older.
- Diagnosis of PV according to the World Health Organization (WHO) 2008 criteria [Barbui et al, 2011] with the mandatory presence of JAK2V617F mutation as the major disease criterion.
- For previously cytoreduction untreated patients – documented need for cytoreductive treatment (one or more of the following criteria):

  a. Age >60 years at the planned day of the first drug administration;

  b. At least one previous well documented major cardiovascular PV-related event, except bleeding and PV-related thromboembolic complications in the abdominal area, see excl. criterion 7) in the medical history;

  c. Poor tolerance (defined as a phlebotomy/ procedure-related AE causing significant adverse impact on the patient and limiting ability to apply phlebotomy with the intention to keep Hct <45%) or frequent need for phlebotomy (more than one phlebotomy within last three months prior entering the study, while each of these phlebotomies was performed to reduce Hct level from >45%, or if one phlebotomy was not able to reduce Hct level to <45% for the next three months following phlebotomy);

  d. Progressive splenomegaly (*de novo* appearance of a palpable spleen, or appearance of the symptoms, related to the enlarged spleen, e.g. pain, early satiety etc., with confirmed size increase);

  e. Platelet counts greater than 1000 x 10^9/L (for two measurements within one week);

  f. Leukocytosis (white blood cell [WBC] >10 x 10^9/L for two measurements within one week).

- For patients currently treated or pre-treated with HU, all of the following criteria:

  a. being non-responders (as defined by the response criteria for primary endpoint in this protocol);

  b. Total HU treatment duration shorter than three years;

  c. No documented resistance or intolerance as defined by modified Barosi et al, 2009 criteria.

Main Exclusion Criteria

- Any systemic cytoreduction for PV in the medical history prior to study entry with exception of HU for shorter than 3 years (see respective inclusion criterion).
- Documented autoimmune disease at screening or in the medical history.
- Clinically relevant pulmonary infiltrates, pneumonia, and pneumonitis at screening.
- Known, PV-related thromboembolic complications in the abdominal area (e.g. portal vein thrombosis, Budd-Chiari syndrome) and/or splenectomy in the medical history.
- History or presence of depression requiring treatment with antidepressant or HADS score equal to or above 11 on either or both of the subscales or any risk of suicide at screening or previous suicide attempts.
- Any significant morbidity or abnormality which may interfere with the study participation.
- Pregnancy and breast-feeding females of reproductive potential and males or woman not using effective means of contraception.
- Evidence of severe retinopathy (e.g. cytomegalovirus retinitis, macular degeneration) or clinically relevant ophthalmological disorder (due to diabetes mellitus or hypertension).
- Thyroid dysfunction (clinical symptoms of thyroid hyper- or hypofunction) not adequately controlled.
- Patients tested positively to thyroglobulin (TgAb) autoantibodies and / or thyroid peroxidase (TPOAb) autoantibodies at screening.

Additional exclusion criteria: History of major organ transplantation, history of uncontrolled severe seizure disorder, history of malignant disease, including solid tumours and haematological malignancies (except basal cell and squamous cell carcinomas of the skin and carcinoma in situ of the cervix that have been completely excised and are considered cured) within the last 3 years or history of active substance or alcohol abuse within the last year. Infections with systemic manifestations, e.g., hepatitis B, hepatitis C, or human immunodeficiency virus (HIV) at screening are excluded. Similarly, patients with leukocytopenia or thrombocytopenia at the time of screening or any contraindication to any of the investigational medicinal products (IMPs) (pegylated IFN or HU) or their excipients or pretreatment with pegylated products were also excluded from the trial.

Treatments

- AOP2014 (ropeginterferon alfa-2b; Peg-P-IFN-α-2b), administered, s.c. at the starting dose of 100 µg (50 µg for HU-pretreated patients) every 2 weeks for up to 12 months of treatment, dose titration allows doses up to 500 µg (investigational [main] arm).

- HU, administered orally at the starting dose of 500 mg daily dose, titration allows doses up 3000mg to for up to 12 months of treatment (reference arm).

Low dose aspirin (acetylsalicylic acid) (100 mg/day) was given to patients in both groups, during the 12 months of study treatment, unless contraindicated.

Both drugs (ropeginterferon alfa-2b and HU) were dosed individually, following their pre-specified dosing schemes (10 dose levels each arm) to achieve optimal disease response. The dose was gradually escalated

depending on disease response and tolerability to the dose which delivered the optimal haematological disease response (Hct <45%, platelets <400 x 10^9/L and leukocytes <10 x 10^9/L).

No interferon is approved in PV in the EU; therefore, the CHMP agreed to hydroxyurea (HU) as comparator HU is the most commonly used cytoreductive agent approved in the EU in the target population and recommended as standard first line treatment since decades in scientific treatment guidelines.

Treatment Duration**:** The main study period was expected to be about 13 months per patient, but long-term follow up treatment for ropeginterferon alfa-2b allocated patients was further investigated in the CONTINUATION-PV trial. (After roll-over in the CONTINUATION-PV trial treatment is planned to be continued as long as it is effective and tolerable/safe).

Objectives

Primary objective: To demonstrate non-inferiority of ropeginterferon alfa-2b vs. HU in terms of disease response rate in both HU naïve and currently treated patients, diagnosed with Polycythemia Vera.

Secondary objective: Efficacy in the two treatment arms, safety, QoL and change of JAK2 allelic burden were analysed, using the secondary endpoints outlined, to provide a more comprehensive picture on ropeginterferon alfa-2b compared to HU in patients with PV.

Outcomes/endpoints

Primary efficacy endpoint*:* Disease response rate at 12 months

Composite endpoint (complete haematological response + normal spleen size) was defined as the rate of patients with a complete haematological response (haematocrit < 45% without phlebotomy [at least 3 months since last phlebotomy], platelets < 400 x 10^9/L and leukocytes < 10 x 10^9/L) and a spleen normality (longitudinal spleen length ≤12 cm for females and ≤13 cm for males).

Key secondary endpoint (post-hoc declared as primary endpoint): Disease response rate at 12 months (=complete haematological response only, without normal spleen size). The endpoint 'complete haematological response at Month 12 (excluding spleen normalization)' was not pre-specified in the protocol as primary endpoint. The NI margin (-20%) for this endpoint was not specified in the protocol such that the NI analysis is not considered confirmatory from a methodological, but nevertheless may be considered relevant from a clinical point of view. This endpoint analysis (NI) was defined after data lock date.

Primary endpoint for FDA and Key secondary efficacy endpoints pre-defined:

Durable disease response rate as per the following definition of durable responder:

- A responder at 12 months, i.e., the following criteria were met: Hct <45% without phlebotomy between month 9 visit to month 12 visit, platelets <400 x 10^9/L, leukocytes <10 x 10^9/L, normal spleen size), and
- A responder at 9 months, as measured by blood parameters, i.e. the following criteria were met: Hct <45% without phlebotomy in the past three months (i.e. visits at 6-9 months according to study schedule), platelets <400 x 10^9/L, leukocytes <10 x 10^9/L.

For all parameters mentioned above, central lab blinded assessments and assessment of spleen size at month 12 according to blinded central imaging was used for determination of durable responders.

Other pre-specified secondary endpoints were: Disease response rates at 3 months and 9 months (spleen size measurements and haematological parameters coming from local measurements), time to first disease response, disease response duration, number of phlebotomies performed (per protocol, a phlebotomy was performed any time the patient's Hct was higher than 45%). Hct-, leukocytes-, platelet- and spleen size-change from baseline to last patient visit, disease-related symptoms (microvascular disturbances, pruritus, headache). From the additional efficacy endpoints molecular response as measured with change of JAK2 allelic burden over time and Quality of Life (EQ-5D) are most important since only during interferon therapy long-term remissions regarding complete molecular response (CMR) is described in the literature.

Sample size

Based on expert judgment, the anticipated size of treatment effect in an overall rate of responders at 12 months was considered to be 25% at least. The assumed rate of responders was 15% in patients treated with HU (reference group) and 40% in patients treated with ropeginterferon alfa-2b (tested group). Supposed drop-out was 20%. The dropped-out patients were considered as non-responders according the definition of the primary endpoint which would decrease assumed rate of responders to 12% (reference group) and 32% (tested group). Based on these assumptions 126 patients per group (252 in total) would be needed in order to detect the difference in response rate between treatment groups at 1% (two-sided) significance level with 90% power using standard chi-square test. Taking into account divisibility by 8 (in order to enable equal proportion of all strata subgroups) 128 patients per treatment group (256 in total) were planned to be enrolled.

It is recognized that this was a single pivotal trial. In accordance with EMEA /CHMP/EWP/2330/99 guidance which specifies that "Statistical evidence considerably stronger than $p<0.05$ is usually required, accompanied by precise estimates of treatment effects, i.e. narrow confidence intervals" (CI). The level of significance was set to <5%. For the primary endpoint, 95% and 99% confidence intervals (CI) were calculated.

According to the DMC statement (review of 6-month data) no sample size re-assessment was needed and therefore not performed.

Randomisation

Treatments were assigned to individual patients based on stratified randomization in order to ensure random assignment to treatments and balanced distribution of both treatments within each stratum. The proportion of patients enrolled in each individual stratum was not pre-defined by the protocol. The following stratification factors were defined: [1] previous HU exposure (yes/no), [2] age at screening (≤60 or >60 years) and [3] presence of thromboembolic events in the past (yes/no). Based on these factors 8 stratification groups were established. Block randomization was used. A randomization list was generated by the responsible study statistician using SAS version 9.3 or higher.

Blinding (masking)

This study was open-label.

Statistical methods

The Full Analysis Set (FAS) including all randomized patients according to the treatment to which they were randomized, but excluding patients without intake of study medication and patients not providing baseline data, was the primary set used for efficacy analyses. The Per Protocol Set (PPS) was used for efficacy sensitivity

analysis. It consisted of patients included in the FAS who completed a certain pre-specified minimal exposure to the treatment regimen, had all measurements needed for assessment of the primary endpoint available and did not violate the study protocol in major concerns.

The study was initially designed as a superiority trial, but switched to a non-inferiority trial after completion of the study (after last patient out, prior database lock).

The primary analysis was conducted using a weighted Cochran-Mantel-Haenszel framework for estimation with stratification factors corresponding to those used in the randomization scheme. Corresponding disease response rate difference at 12 months between the two treatment arms (test/reference) and its 95% and 99% CIs were calculated. Non-inferiority was concluded if the lower limit of the 95% two-sided CI of the Mantel-Haenszel common estimate of response rate difference exceeded -0.105 for both the Full Analysis Set (FAS) and the Per Protocol Analysis Set (PPS). All results need to be interpreted in an exploratory sense.

The disease response is a binary (Yes/No) variable defined by four (response) components. Only if all of these four response criteria were met the patient was classified as "responder" (treatment success). Patients withdrawn, for any reason, were classified as treatment failures. A last observation carried forward analysis (LOCF) was provided as sensitivity analysis.

Secondary efficacy analyses were performed for exploratory purposes using descriptive analysis and standard statistical tests. Primary and secondary efficacy endpoint analysis was also done for the individual strata.

Results

Participant flow

A total number of 257 patients was enrolled (127 in the ropeginterferon alfa-2barm, 130 in the HU arm) according the sample size calculation. Sample size re-assessment allowed enrolment for a maximum of 368 patients. Two hundred fifty-four (254) patients received at least one dose and were included in the safety analysis set, and included in the Full Analysis Set (FAS).

Of these, 229 patients (90.2 %) were eligible for Per Protocol Set (PPS).

Table 18: Disposition of study patients and analysis sets

Group of patients	ropeginterferon alfa-2b	HU	All patients
Screened	-	-	
Enrolled	127	130	257
Randomized	127	130	257
Randomized but not treated	0	3	3
Treated (= Safety analysis set)	**127 (100.0%)**	**127 (100.0%)**	**254**
Full Analysis Set (FAS)	127 (100.0%)	127 (100.0%)	254 (100.0%)
Discontinued	21 (16.5%)	16 (12.6%)	37 (14.6%)
Completed the study	106 (83.5%)	111 (87.4%)	217 (85.4%)
Per Protocol Set (PPS)	**115 (90.6%)**	**114 (89.8%)**	**229 (90.2%)**
Per Protocol Set for Sensitivity analysis (PPSS)	119 (93.7%)	118 (92.9%)	237 (93.3%)

The mean age at inclusion was 58.2 years (SD: 11.99) with a median of 60 years (range 21 to 85 years).

- 119/254 (46.9%) were male and 135/254 (53.1%) patients were female.
- 100 % of patients were White, 3/254 (1.2%) had ethnicity Hispanic or Latino.
- The median duration of PV was 1.9 months (range 0-146 months) in the ropeginterferon alfa-2btreatment arm and 3.6 months (range 0-126 months) in the HU treatment arm indicating that patients were diagnosed in an early stage of the disease. Out of 94 HU pre-treated patients, 82 (32.3 %) had records of HU-treatment at study entry, with a median duration of 8.7 months.
- At baseline, the haematological parameters were comparable between the two treatment arms with a mean Hct around 45.5%.
- At baseline, spleen size was comparable between the two treatment arms with a median length of around 13cm, close to normal size. Only 9.4% [12/127] (ropeginterferon alfa-2b) and 11.8% [15/127] (HU) of patients had a spleen size >17cm.
- At baseline, 98.8% [251/254] of the patients were JAK2 V617F positive, with a median JAK2 allelic burden of 37.4% (comparable between the two treatment arms; 37.3% and 37.4%).

Recruitment

The study was conducted between 04 October 2013 and 08. April 2016 in 13 countries (including Austria, Bulgaria, Czech Republic, France, Germany, Hungary, Italy, Poland, Romania, Russia, Slovakia, Spain, and Ukraine) within and outside the European Union. Planned number of centers was 45 to 60; in total 56 sites were activated. Planned: 250 patients; Enrolled: 257 patients; Analysed: 254 patients.

Study initiation date: 04-October-2013

Study completion date: 08-April 2016

Data lock point: 22-August 2016

Conduct of the study

Protocol deviations were rare and occurred in11 patients (5 in the ropeginterferon alfa-2barm, and 6 in the HU arm) and led to exclusion from PPS in 9 patients (4 in ropeginterferon alfa-2b arm and 5 in HU arm).

Table 19: Reasons for exclusion/inclusion from analysis sets

Group of patients		**All patients**
Randomized		**257**
Randomized but not treated		3 (1.2%)
Treated (= Safety set)		**254**
Full Analysis Set (FAS)		254 (100.0%)
Reason for exclusion from PPS	Patient did not enter into maintenance treatment phase	8
	Patient did not enter into maintenance treatment phase + Major PD	1
	Major PD	7
	Missing measurements for assessment of the primary endpoint	8
	Missing measurements for assessment of the primary endpoint + Major PD	1
Per Protocol Set (PPS)		**229**

Reason for inclusion in PPSS	Missing measurements for assessment of the primary endpoint	+8

Protocol Amendments:

There were three amendments to the protocol of PROUD-PV:

Amendment 1: 20 February 2014. Following the discussions with the US FDA, the secondary endpoint "Durable disease response" was added. The amended study protocol states that the endpoint "Durable disease response" was to be analysed as the primary one for study submission in the US; however, for study submission in Europe the primary endpoint "disease response at 12 months" remained unchanged. In order to avoid confusion, a separate US Specific SAP was written for submission in the US. All endpoints as per study protocol were included in both SAPs (for the European and for the US submission). The statistical methods and sample size justification *Amendment 2:* 17 July 2014. Clarification on patients who will have an immunogenicity sample drawn on WK4 and who have already started the study at the time this was implemented.

Amendment 3: 15 June 2016. The primary objective was changed from “To demonstrate superiority of ropeginterferon alfa-2b vs. HU in terms of disease response rate in both HU naïve and currently treated patients, diagnosed with Polycythemia Vera.” to “To demonstrate non-inferiority of ropeginterferon alfa-2b vs. HU in terms of disease response rate in both HU naïve and currently treated patients, diagnosed with Polycythemia Vera.”

Protocol Deviations

A total of 14 major protocol deviations occurred in 11 patients (5 in the ropeginterferon alfa-2b arm, and 6 in the HU arm) and out of those, 11 led to exclusion from PPS in 9 patients (4 in ropeginterferon alfa-2b arm and 5 in HU arm). Major protocol deviations leading to exclusion from PPS were: 6 major deviations due to violation of eligibility criteria (history of malignant disease [in 2 patients], positive TPOAb [in 2 patients], positive HADS without psychiatric assessment [in 1 patients] and a low Hb value [in one patient]) and 5 major protocol deviations due to major efficacy deviations [in 3 patients] (such as a dose reduction or interruption following an AE not related to the study drug [in 4 patients] or due to missed visits [in 1 patients]), led to exclusion from PPS.

Furthermore 3 major safety deviations not leading to the exclusion from PPS were observed in two patients (1 in each treatment arm): included two cases of continuation of the study drug despite a Grade 3 Adverse event (in 1 patient) and an accidental overdose of the ropeginterferon alfa-2b study drug (in 1 patient).

Baseline data

Two hundred fifty-four (254) patients received at least one dose and were included in the safety analysis set, and included in the Full Analysis Set (FAS).

Of these, 229 patients (90.2 %) were eligible for Per Protocol Set (PPS).

Demographics: In the PROUD-PV trial relevant demographic baseline characteristics (mean age: 58.2 y; range 21 -81y) were balanced between both arms. Females were slightly more included (F:53.1% (M:46.9%). Disease specific factors also were balanced regarding median duration of PV, HU pre-treatment, baseline haematological parameters, spleen size and median JAK2 allelic burden (at baseline). At least one previous well documented major cardiovascular PV-related event, except bleeding and PV-related thromboembolic complications in the abdominal area in the medical history was an inclusion criteria; however, cardiovascular risk factors were not systematically reported or used for stratification.

Numbers analysed

Table 20: Disposition of study patients and analysis sets

Group of patients	ropeginterferon alfa-2b	HU	All patients
Screened	-	-	306
Enrolled	127	130	257
Randomized	127	130	257
Randomized but not treated	0	3	3
Treated (= Safety analysis set)	**127 (100.0%)**	**127 (100.0%)**	**254 (100.0%)**
Full Analysis Set (FAS)	127 (100.0%)	127 (100.0%)	254 (100.0%)
Discontinued	21 (16.5%)	16 (12.6%)	37 (14.6%)
Completed the study	106 (83.5%)	111 (87.4%)	217 (85.4%)
Per Protocol Set (PPS)	**115 (90.6%)**	**114 (89.8%)**	**229 (90.2%)**
Per Protocol Set for Sensitivity analysis	119 (93.7%)	118 (92.9%)	237 (93.3%)

Outcomes and estimation

Primary endpoint

The disease response at 12 months of treatment was **21.3%** for ropeginterferon alfa-2b and **27.6%** for HU (FAS), with a difference in responder rates of -6.6 (95% CI: -17.2 to 4.1; p=0.2233), and 22.6% for ropeginterferon alfa-2b and 29.0% for HU (PPS), with a difference in responder rates of -6.3 (95% CI: -17.5 – 5.0; p=0.2353).

Table 21: Primary endpoint outcome: Disease response at month 12 (complete haematological response and spleen normality) FAS and PPS

		ropeginterferon alfa-2b		Hydroxyurea			
Analysis set	**Disease response**	**N (%)**	**Ndisc* (%)**	**N (%)**	**Ndisc* (%)**	**95% CI of difference in responder's percentages**	**p-value*** **
FAS		N/Nmiss = 122/5		N/Nmiss = 123/4			
	Non-responder	96 (78.69%)	21 (17.21%)	89 (72.36%)	16 (13.01%)		
	Responder	**26 (21.31%)**		**34 (27.64%)**		**-6.57 (-17.23 to 4.09)**	**0.2233**
PPS		N/Nmiss = 115/0		N/Nmiss = 114/0			
	Non-responder	89 (77.39%)	16 (13.91%)	81 (71.05%)	10 (8.77%)		
	Responder	**26 (22.61%)**		**33 (28.95%)**		**-6.28 (-17.49 to 4.92)**	**0.2353**

FAS: Full analysis set; PPS: per protocol set; Nmiss means that data has not been recorded at time of data snapshot.
*Ndisc: Number of patients with premature discontinuation. **ropeginterferon alfa-2b-versus HU treated patients.
***Wald test for testing of non-inferiority ropeginterferon alfa-2b versus HU. According to definition of the

disease response discontinued patients are non-responders at visits after discontinuation. Note that the lower boundaries of the 95% CI for both sets crosses the non-inferiority margin of -10.5%,
Source: CSR PROUD-PV Table 14.2.2.1

The disease response at 12 months of treatment was 21.3% for ropeginterferon alfa-2b and 27.6% for HU (FAS), with a difference in responder rates of -6.6 (95% CI: -17.2 to 4.1; p=0.2233), and 22.6% for ropeginterferon alfa-2b and 29.0% for HU (PPS), with a difference in responder rates of -6.3 (95% CI: -17.5 – 5.0; p=0.2353).

A total of 9/217 (FAS) patients had no data for disease response criteria at end of month 12 treatment (EOT) visit available. Four patients in each treatment group had no data for disease response criteria available but other criteria for disease response were fulfilled. For one patient in the ropeginterferon alfa-2b treatment arm, no data for disease response evaluation was available.

Comparison of treatment arms showed that ropeginterferon alfa-2b had 0.69 to 0.75 times lower probability of response, but the results were not statistically significant in either of the statistical models (p ranged from 0.2302 to 0.3909).

Comparison between HU pre-treated and HU naïve patients was statistically significant in all four models (p<0.05) and showed that HU pre-treated patients had a 0.47 to 0.49 times lower probability of response then HU naïve patients, irrespective of treatment. A comparable effect was seen in the two treatment groups, as trend (0.41 to 0.43 probability of response) within the HU treatment group (p ranged from 0.0534 to 0.0657) and non-significant (0.55 to 0.57 probability of response) within the ropeginterferon alfa-2b treatment group (p ranged from 0.2222 to 0.2441).

When adjusted for stratification factors and sex, females were 2.36 times more likely to respond to the treatment (p = 0.0069) and the probability of response was slightly increased in the age group > 60 years (1.15 to 1.18), not showing a significant effect (p ranged from 0.5750 to 0.6484).

Secondary endpoints

Complete haematological response (NI analysis post-hoc defined; without spleen size)

Table 22: Complete haematological response at month 12- FAS and PPS

		ropeginterferon alfa-2b		**Hydroxyurea**			
Analysis set	**Disease response**	**N (%)**	**Ndisc* (%)**	**N (%)**	**Ndisc* (%)**	**95% CI of difference in responder's percentages****	**p-value*****
FAS		N/Nmiss= 122/5		N/Nmiss = 123/4			
	Non-responder	70 (56.91%)	21 (17.07%	68 (54.40%)	16(12.80%)		
	Responder	**53 (43.09%)**		**57 (45.60%)**		**-6.57 (-17.23 to 4.09)**	**0.0028**

PPS		N/Nmiss: 113/2		N/Nmiss = 114/0			
	Non-responder	63 (55.75%)	16 (14.16%	61 (53.51%)	10 (8.77%)		
	Responder	**50(44.25%)**		**53 (46.49%)**		**-6.28 (-17.49 to**	**0.0036**

FAS: Full analysis set; PPS: per protocol set; Nmiss means that data has not been recorded at time of data snapshot.

The disease response at 12 months of treatment was 43.1% for ropeginterferon alfa-2b and 45.6% for HU (FAS), with a difference in responder rates of -3.0 (95% CI: -15.6 to 9.5; p=0.0028), and 44.3% (50/113) for ropeginterferon alfa-2b and 46.0% for HU (PPS), with a difference in responder rates of -2.6 (95% CI: -15.8 to 10.5; p=0.0036). The results did show non-inferiority of ropeginterferon alfa-2b to HU at month 12 in both analysis sets (non-inferiority margin of -20.0%; p<0.01).

Disease response rates over time (i.e. the evaluation of the complete haematological response [haematological parameters measured by central blinded lab] without the disease criterion spleen size at the respective assessment visits from week 12 until week 52) are summarized in

Table 23 (FAS)

Exploratory key secondary endpoint (NI analysis post-hoc defined after data look date):

Complete haematological response (without spleen normalisation) (post-hoc declared as primary endpoint) after the NI margin was widened to -20% (post-hoc declared): Complete hematological response was observed in 43.1% (43/122) of the patients in ropeginterferon alfa-2b arm and in 45.6% (57/123) of the patients in the HU arm after 12 months of treatment (p=0.0028).

Table 23: Disease response rate (excluding spleen size criterion) at assessment visits –FAS

		ropeginterferon alfa 2-b		HU		95% CI of difference in responder's	
Visit	**Disease response**	**N (%)**	**Ndisc* (%)**	**N (%)**	**Ndisc* (%)**	**percentages (AOP-HU)**	**p-value**
Week 12 (V7)-A		N/Nmiss = 116/11		N/Nmiss = 111/16			
	Non-Responder	103 (88.79%)	2(1.72%)	94 (84.68%)	5 (4.50%)		
	Responder	**13 (11.21%)**		**17 (15.32%**		**-3.97 (-12.72/4.78)**	**0.3718**
Week 26 (V14)-A		N/Nmiss =122/5		N/Nmiss =119/8			
	Non-Responder	84 (68.85%)	9 (7.38%))	67(56.30%)	11 (9.24%)		
	Responder	**38 (31.15%)**		**52 (43.70%)**		**-13.28 (25.25/1.31)**	**0.0333**
Week 40 (V21)-A		N/Nmiss =122/5		N/Nmiss =118/9			
	Non-Responder	79 (64.75%)	16 (13.11%)	73 (61.86%)	13 (11.02%)		
	Responder	**43 (35.25%)**		**45 (38.14%)**		**-2.85 (-15.29/9.6)**	**0.6496**
Week 52 (EOT)-A		N/Nmiss =123/4		N/Nmiss =125/2			
	Non-Responder	70 (56.91%)	21 (17.07%)	68 (54.40%)	16 (12.80%)		
	Responder	**53 (43.09%)**		**57 (45.60%)**		**-3.02 (-15.55/9.5)**	**0.6320**

The difference in the response rate, which was most evident at week 26 and decreased with ongoing treatment duration. Ropeginterferon alfa-2b required a longer time to reach the maximal dose plateau (end of titration phase) which may explain the timely delay in disease response.

The following table shows the change in disease response rates (excluding spleen normality) during the PROUD-PV trial:

The following Table 24 summarizes the **outcome to the predefined secondary endpoints**, as far as available.

Table 24: Summary of the outcome for the secondary endpoints reported

Secondary endpoint	**Result**	
	ropeginterferon alfa-2b	**HU**
Disease response rate at 6 months, based on haematological parameters only, defined as Hct <45% without phlebotomy (at least 3 months since last phlebotomy), platelets <400 x 109/L, leukocytes <10 x 109/L (all three lab parameters measured by blinded central lab) for the formal IA (FDA-primary endpoint).	18.6%	31.7%
Time to first disease response (with spleen normality)	50% at362 days	50% at 181 days
Time to first disease response (without spleen normality)	50% at 165 days	50% at112 days
Disease response duration	266 days	167 days
Number of phlebotomies performed (per protocol,	400	276

Secondary endpoint	Result	
	ropeginterferon alfa-2b	HU
a phlebotomy was performed any time the patient's Hct was higher than 45%)	94/127 patients (74.0%)	81/127 patients (63.8%)
Hct change from baseline to last patient visit	45.2%	55.1%
Leukocytes change (< 10 x 10^9/l) from baseline to last patient visit.	78.2%	80.2%
Platelets change from baseline to last patient visit.	74.8%	73.4%
Change in spleen size from baseline to last patient visit	36.1%	48.8%
Disease-related symptoms (microvascular disturbances, pruritus, headache) during the study.	51 disease relat.TEAEs 16/127 patients 12.6%	14 disease relat.TEAEs 11/127 patients 8.7%
Change of JAK-2 allelic burden at EOT	30.7%	25.9%
Quality of Life (EQ-5D)	Change in VAS: 1.3 (±12.56)	Change in VAS: 1.8 (±13.09)
	Total score: 0.2 (±1.08)	Total score: 0.1 (±1.17)

Spleen size change from baseline until EOT

The majority of patients did not have a change in spleen size categorization (normal spleen size/enlarged spleen), 85.1% in the ropeginterferon alfa-2b treatment arm and 84.1% in the HU arm. The absolute decrease in spleen size (change from baseline) was comparable between the treatment arms and ranged from -0.2 cm to -1.4 cm for ropeginterferon alfa-2b and -0.7 cm to -1.9 cm for HU treatment. The absolute increase in spleen size (change from baseline) was also similar for ropeginterferon alfa-2b and HU treatment, with +1.3 cm and +1.0 cm, respectively (Table 25).

Table 25: Absolute change in spleen size (longitudinal diameter) from baseline in the PROUD-PV Study [in cm]

Spleen size normality Screening —› EOT	ropeginterferon alfa-2b		HU	
	n	Mean (±SD)	n	Mean (±SD)
Enlarged spleen —› enlarged spleen	50	-0.3 (±1.63)	44	-1.6 (±1.71)
Enlarged spleen —› normal spleen size	8	-1.4 (±0.74)	14	-1.9 (±0.81)
Normal spleen size —› enlarged spleen	7	1.3 (±0.89)	3	1.0 (±0.50)
Normal spleen size —›normal spleen size	36	-0.2 (±1.02)	46	-0.7 (±0.82)

Summary of main efficacy results

The following tables summarise the efficacy results from the main studies supporting the present application. These summaries should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 26: Summary of efficacy for the pivotal PROUD-PV trial

Title: PROUD-PV			
Study identifier	[2012-005259-18]		
Design	A randomized, open-label, multicenter, controlled, parallel arm, phase III study assessing the efficacy and safety of AOP2014 vs. Hydroxyurea in patients with Polycythemia Vera (PROUD-PV study)		
	Duration of main phase:	04 October 2013 - 08 April 2016	
	Duration of Run-in phase:	not applicable	
	Duration of Extension phase:	Separate trial CONTINUATION-PV (s. below)	
Hypothesis	Superiority planned but switch to **Non-inferiority**		
Treatments groups	Investigational Product Arm (AOP2014, ropeginterferon alfa-2b)	ropeginterferon alfa-2b: administered, subcutaneously (s.c.) at the starting dose of 100 µg (up to 540 µg) every 2 weeks for up to 12 months of treatment, N=127	
	Reference Arm (HU, Hydroxyurea)	HU (reference arm): administered per os at the starting dose of 500 mg daily, according to the local standard of care, for up to 12 months of treatment, N=127	
Endpoints and definitions	**Primary endpoint (Composite)**	Complete haematological response (haematocrit < 45% without phlebotomy [at least 3 months since last phlebotomy], platelets < 400 x 10^9/L and leukocytes < 10 x 10^9/L) **and a spleen normality** (spleen length ≤12 cm for females and ≤13 cm for males) at month 12	
	Key Secondary endpoint (post-hoc*)	Complete haematological response at month 12 **(excluding spleen normalization)**	
	Other Secondary endpoints	Disease response rate at 6 months, based on haematological parameters only, defined as Hct <45% without phlebotomy (at least 3 months since last phlebotomy), platelets <400 x 10^9/L, leukocytes <10 x 10^9/L (all three lab parameters measured by blinded central lab)	
		Time to first disease response (with spleen normality)	
		Time to first disease response (without spleen normality)	
		Disease response duration	
		Number of phlebotomies performed (per protocol, a phlebotomy was performed any time the patient's Hct was higher than 45%)	
		Hct change from baseline to last patient visit	
		Leukocytes change (< 10 x 10^9/l) from baseline to last patient visit.	
		Platelets change from baseline to last patient visit.	
		Spleen size from baseline to last patient visit	
		Disease-related symptoms (microvascular disturbances, pruritus, headache) during the study.	
		Change of JAK-2 allelic burden at EOT	
		Quality of Life (EQ-5D)	

Database lock	Data lock point: 22-August 2016 (final database lock 23-January 2017)

Results and Analysis

Analysis description	**Primary Analysis**
Analysis population and time point description	• FAS (full analysis set; all treated patients without major violation of eligibility criteria=ITT) • during the trial every 2 weeks, EOT 12 months

Treatment group	**Ropeginterferon-alfa-2b**	**Hydroxyurea**
Number of subject	N=127	N=127
Prim. EP Complete haematological response **and spleen normality** at month 12	21.31% (26/122)	27.64% (34/123)
Key sec. EP Complete haematological response at month 12 **(excluding spleen normalization)**	43.1% (53/123)	45.6% (57/125)
Other sec.EP: Time to first disease response (without spleen normality) (median)	165 days	112 days
Other sec.EP: Disease response duration (median)	266 days	167 days
Other sec.EP: Hct change from baseline to last patient visit	45.2%	55.1%
Other sec.EP: Leukocytes change (< 10 x 10^9/l) from baseline to last patient visit	78.2%	80.2%
Other sec. EP: Platelets change from baseline to last patient visit.	74.8%	73.4%
Other sec. EP: Spleen size change from baseline to last patient visit	36.1% from -0.2 cm to -1.4 cm	48.8% From -0.7 cm to -1.9

Effect estimate per comparison	**Prim. EP** Complete haematological response **and spleen normality**	Comparison groups	Ropeginterferon alfa-2b vs. Hydroxyurea
		Difference in responder percentage (using Cochran-Mantel-Haenszel weights)	-6.57
		95% CI	-17.23 to 4.09

		P-value (NI comparison with NI margin -10%)	0.2233
	Complete haematological response at month 12 **(excluding spleen normalization)**	Difference in responder percentage (using Cochran-Mantel-Haenszel weights)	-3.02
		95% CI	-15.55 to 9.52
		P-value (NI comparison with NI margin -20%)	0.0028
Notes	*The endpoint 'complete haematological response at Month 12 (excluding spleen normalization)' was not pre-specified in the protocol. The NI -20% for this endpoint was not pre-specified in the protocol such that the NI analysis is not considered confirmatory. Nevertheless, it is considered as a key secondary endpoint from a clinical point of view as explained in the text above.		
Analysis description	**Including secondary analysis after change in SAP for key sec. EP**		

Clinical studies in special populations

Information regarding efficacy in special populations were initially not reported. However, the applicant has provided a separate analysis regarding gender and several relevant age cohorts which did not demonstrate relevant differences. Limitations of this approach in the small orphan disease population have to be noted. However, the large clinical experience also in special populations with other interferons is considered reassuring.

Analysis performed across trials (pooled analyses AND meta-analysis)

Analysis across the trial regarding differences in efficacy outcome between the four AOP efficacy studies (PEGINVERA; PROUD, CONTINUATION and PEN-PV) shows that the response rate for the haematological parameters Hct, PLT and leukocytes (complete haematological responders) ranged between 43% and 75% in the clinical studies in line with that reported for other IFNs-α in literature.

The following “haematological” criteria have been relatively constant throughout the development of ropeginterferon alfa-2b; comparison was based on: Hct < 45% (without phlebotomy in the previous two (or three) months); Platelet (PLT) count ≤ 400 * 10^9/L and Leukocyte count ≤ 10 * 10^9/L

Table 27: Comparison of individual haematologic responses across studies

	AOP STUDY			
Parameter	**PEGINVERA**	**PROUD-PV**	**CONTI-PV***	**PEN-PV**
Hct median (%)	41.3-44.0	44.0	40.0-42.1	39.7-41.2
Hct responders (<45%, no phleb.)	58-86%	37-55%	66-82%	72-75%
PLT median (*10^9/L)	190-399	220	176-204	176-187
PLT responders (% ≤ 400*10^9/L)	51-97%	60-76%	91-95%	91-94%
leukocytes mean (*10^9/L)	4.6-7.5	5.5	4.7-5.1	5.0-5.3
leukocytes responders (% ≤ 10*10^9/L)	74-97%	78-81%	93-98%	91-94%

In the four AOP efficacy studies, the response rate for the haematological parameters Hct, PLT and leukocytes (complete haematological responders) ranged between 43% and 75%, derived from the AOP study data:

- PROUD-PV Study: 43% after 12 months treatment,
- CONTINUATION-PV Study: 70.5% at treatment month 24 and month 36 (i.e. after 12 and 24 months treatment in the extension study)
- PEN-PV Study: 75% at treatment month 18.8 (range 14.9 to 24.5), after 3 months treatment within the respective study
- PEGINVERA-PV Study: the percentage of complete haematological responders was not evaluated in the study.

Normal spleen size was defined as a longitudinal diameter ≤ 12 cm for females and ≤ 13 cm for males. The spleen size ranges observed per AOP study were the following:

- PROUD-PV Study: 13.1-13.4 cm (mean absolute sizes)
- CONTINUATION-PV Study: 12.4-12.6 cm (median absolute sizes at treatment month 36)
- PEN-PV Study: 13.4-13.5 cm (mean absolute sizes)
- PEGINVERA Study: 12.4-14.0 cm (median absolute sizes)

When the spleen size was included as a criteria, the studies evaluating the disease response (defined as complete haematological response with spleen size normality), reported the following percentages of complete responders:

- PROUD-PV Study: 21% after 12 months treatment
- CONTINUATION-PV Study: 37.4% at treatment month 24 and 42.2 at treatment month 36 (i.e. after 12 and 24 months treatment in the extension study)
- PEN-PV: 48% at treatment month 18.8 (range 14.9 to 24.5), after 3 months treatment within the respective study
- PEGINVERA-PV Study: 48% after (median) 30.5 months treatment

Supportive study: CONTINUATION-PV [2012-005259-18]

CONTINUATION-PV trial is an open-label, multicenter, phase IIIb study assessing the long-term efficacy and safety of ropeginterferon alfa-2b in patients with Polycythemia Vera who previously participated in the PROUD-PV Study. It was planned as a follow on study designed to provide long-term evaluation of ropeginterferon alfa-2b in patients with PV who received the investigational medicinal product (IMP) subcutaneously during the PROUD-PV Study. The integration of the subjects who participated in the PEN-PV Study was also to contribute to this long-term evaluation of the investigational product as well as an alternative mode of administration (pen). Additionally, data from the routine care treatment of patients, who participated in the PROUD-PV Study and were allocated to the HU arm, were to be collected and analysed in the same way as the ropeginterferon alfa-2b arm in this study to enable long term observation of ropeginterferon alfa-2b treatment compared to the real life treatment information. However, the HU/BAT arm was started later following an amendment to the initial study protocol.

Objectives:

Primary:

• To assess the long-term efficacy of ropeginterferon alfa-2b or standard first line treatment (HU or other best available treatmtent [BAT) in terms of disease response rate in patients diagnosed with PV, who were previously treated in the PROUD-PV Study and who completed this study.

• To assess the long-term efficacy including changes in disease burden in patients diagnosed with PV. Disease burden is defined as disease-related signs (clinically significant splenomegaly) and disease-related symptoms (microvascular disturbances, pruritus, headache), assessed by investigator.

Secondary:

• To further assess the long-term efficacy, safety, quality of life (QoL), ease of self-administration of ropeginterferon alfa-2b and change of JAK2 allelic burden in patients diagnosed with PV and previously treated with ropeginterferon alfa-2b or HU in the PROUD-PV Study

Study design / Methodology:

This was a Phase IIIb, open-label continuation of the PROUD-PV Study performed in adults diagnosed with PV. No confirmatory statements or hypotheses were formulated.

Patients who received ropeginterferon alfa-2b or HU in the primary study, PROUD-PV, and completed the end of study visit per protocol, were to be included in this CONTINUATION-PV Study.

Enrolment occurred after eligible patients had signed the informed consent form (ICF). The first day of the CONTINUATION-PV Study coincided with the end of study visit of the primary study, PROUD-PV or the last visit in the PEN-PV Study. Patients who participated in the PEN-PV Study (estimated patient participation was approximately 12 weeks), were allowed to continue their treatment using the pen in the CONTINUATION PV Study.

Subjects were to continue to receive the individualized dosage delivering the optimal disease response (haematocrit [Hct] <45%, platelets [PLTs] <400 x 10^9/L and leukocytes [WBCs] <10 x 10^9/L), as determined in the PROUD-PV or PEN-PV Study, preferably at the level of target blood values.

Treatment was to continue with a periodicity of two weeks, but patients could also be switched to a three or four week administration schedule after PEN-PV Study completion (last subject out) and based on eligibility criteria. Patient visits were to be scheduled every two, three or four weeks.

Target blood values adopted to avoid fluctuations in response assessment due to the variability of laboratory measurements were as follows:

- PLTs and WBCs (approximately 20% - 25% below the upper limit of normal [ULN)]:
    - o ULN for PLTs: 400 x 10^9/L, i.e. target PLTs approximately 300 x 10^9/L.
    - o ULN for WBCs 10 x 10^9/L, i.e. target value for WBCs 8 x 10^9/L.
- Hct level of 40 - 42% without phlebotomies.

Dose adjustment rules, defined in the PROUD-PV and PEN-PV Study, were also to be applied in this continuation protocol. The number of dose decrease and reescalation attempts was not to be limited in this study, since treatment may continue for several years. However, maintaining response or re-gain of response after its loss due to toxicity dose decrease was to be desirable to improve long-term treatment outcomes. Decision on

continuation for each individual patient was to be made by the Investigator upon consideration of benefits and risks.

Disease response assessments were to be made every three months (independently from the administration schedule) by the Investigator to allow continuous data assessment.

A safety follow-up visit was to be planned 28 days after the last IMP administration, including premature discontinuation. Immunogenicity samples were to be drawn at 3 and 6 months following the last ropeginterferon alfa-2b administration.

Also, genetic samples post study (e.g. year 2 and year 3) may be collected to assess disease status following discontinuation of ropeginterferon alfa-2b treatment.

Treatment can be continued as long as it is effective and safe and until ropeginterferon alfa-2b becomes otherwise available.

Safety and efficacy data is planned to be periodically analysed in a descriptive manner.

Diagnosis and main criteria for inclusion:

A patient who met all of the following criteria qualified for entry into the study:

Patients having completed the 12 months PROUD-PV Study and who fulfilled at least one of the following criteria:

a) Normalization of at least two out of three main blood parameters (Hct, PLTs and WBCs) if these parameters were moderately increased (Hct<50%, WBC<20 x 10^9/L, PLTs<600 x 10^9/L) at baseline of the PROUD-PV Study, OR

b) >35% decrease of at least two out of three main blood parameters (Hct, PLTs and WBCs) if these parameters were massively increased (Hct>50%, WBCs>20 x 10^9/L, PLTs>600 x 10^9/L), at baseline of the PROUD-PV Study, OR

c) Normalization of spleen size, if spleen was enlarged at baseline of the PROUD-PV Study, OR

d) Otherwise a clear, medically verified benefit from treatment with ropeginterferon alfa-2b (e.g. normalization of disease-related micro-vasculatory symptoms, substantial decrease of JAK2 allelic burden).

Treatments

The following products were administered to patients during the trial:

IMP: AOP2014 (ropeginterferon alfa-2b, Pegylated Proline-IFN alpha-2b); *Dosage and regimen:* Dosing scheme achieving optimal disease response in the individual patients, as determined in the primary study, PROUD-PV or the PEN-PV Study- administered every two, three or four weeks until the end of the study (sc)

Comparator: The BAT arm received standard first line treatment for treatment of PV disease, as per investigator's discretion.

Low dose aspirin (acetylsalicylic acid) (100 mg/day) was to be given to all patients (ropeginterferon alfa-2b arm) for the duration of study treatment, unless contraindicated.

Study participation was planned to be as long as the treatment was effective and tolerable/safe and until ropeginterferon alfa-2b becomes otherwise available.

Primary efficacy endpoint:

The main efficacy evaluation criterion was to be disease response defined as (1)

- Hct<45% without phlebotomy (at least 3 months since the last phlebotomy),
- PLTs<400 x 10^9/L,
- WBCs<10 x 10^9/L, and
- normal spleen size.

And (2) as

- Hct<45% without phlebotomy,
- PLTs < 400 x 10^9/L,
- WBCs < 10 x 10^9/L,
- resolution and/or clinically improvement of disease-related signs (clinically significant splenomegaly) and disease-related symptoms (microvascular disturbances, pruritus, headache).

Secondary efficacy endpoints:

1. Change in haematological parameters, Hct, WBCs, PLTs and red blood cells (RBCs), from baseline over time up to last patient visit.

2. Change in spleen size from baseline over time up to last patient visit, including change in clinical assessment of asymptomatic to symptomatic /progressive splenomegaly.

3. Maintenance rate of disease response at assessment visits.

4. Duration of response maintenance.

5. Time to disease response.

6. Progression free time.

7. Phlebotomy need.

8. Change of disease related signs and disease-related symptoms (microvascular disturbances, pruritus, headache)

9. Change in QoL (EQ-5D-3L) from baseline over time up to last patient visit.

10. Change in JAK2 allelic burden and other molecular and genetic abnormalities from baseline over time up to last patient visit.

Additional secondary efficacy endpoints defined in Statistical Analysis Plan (SAP) in order to be consistent with endpoints defined for final analysis in PROUD-PV Study:

1. Molecular response

Safety endpoints (safety evaluation):

1. Incidence, causality and intensity of adverse events (AEs) according to common terminology criteria for adverse events (CTCAE 4.0).

2. Events leading to dose reduction or permanent treatment discontinuation.

3. Adverse events of special interest (AESI).

Statistical methods:

Sample size justification

No formal hypothesis is planned to be tested in the CONTINUATION-PV Study. Only patients who completed PROUD-PV.

Statistical analysis

The statistical analysis was to be done in agreement with the ICH Topic E9, Statistical Principles for Clinical Trials and the SAP finalized and approved by the Sponsor and the study statistician before database lock.

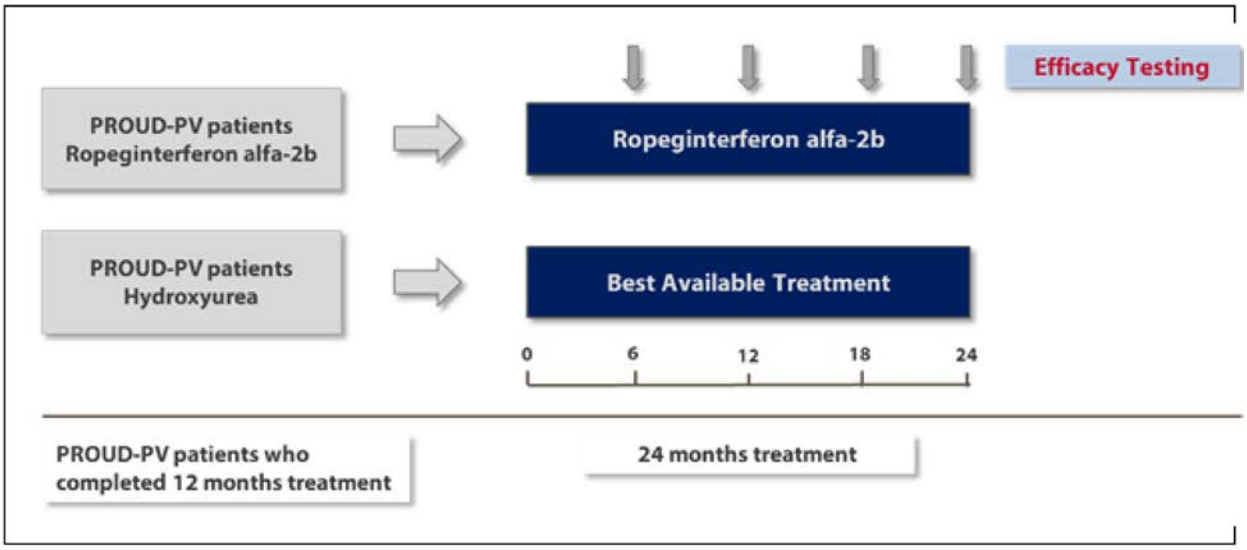
Analyses were to be conducted using Statistical Analysis System (SAS®) software, version 9.3 or higher (SAS Institute, Cary, NC, USA).

Analyses of preliminary data without any impact on the study may have been carried out in order to provide results before study completion.

Efficacy analysis

All statistical analyses were to be performed for exploratory purposes. The standard level of significance, alpha=5%, was set for this study. All confidence intervals and tests were to be two-sided.

Participant flow



Results:

A total of 171 patients were enrolled in this study. All enrolled patients completed PROUD-PV Study, 95 patients previously treated in ropeginterferon alfa-2b arm and 76 patients participated in hydroxyurea (HU) arm. The roll-over rate from PROUD-PV Study into its extension study (CONTINUATION-PV Study) was 78.8% (171/217); 89.6% (95/106) for ropeginterferon alfa-2b-treated and 68.5% (76/111) for (in PROUD-PV Study) HU- treated patients.

Patients entered CONTINUATION-PV Study after a median of 14 days (range 0-122 days) in the ropeginterferon alfa-2b arm, and after a median of 148 days (range 69-481 days) in the BAT arm. The difference is due to the

fact, that the Study Protocol version 4.0, which introduced patients who participated in HU arm of PROUD-PV Study to be included into CONTINUATION-PV Study, is dated 15 Jul 2015, i.e. 9 months after the first patient completed PROUD-PV Study (04 Oct 2014).

Demographics:

All 171 patients were included in the Full Analysis Set (FAS). As all patients enrolled in the CONTINUATION-PV Study previously participated in the PROUD- PV Study, baseline values for demographics and other characteristics of patients continuing in CONTINUATION-PV Study are taken from the screening visit on PROUD-PV Study (i.e. before first study treatment in PROUD-PV Study).

• The mean age at inclusion was 57.5 years (SD: 11.0) with a median of 59.0 years (range 30 to 85 years).

• 83/171 (48.5%) patients were male and 88/171 (51.5%) were female.

• 100 % of patients were Caucasian.

At database (soft) lock (29 May 2018) a total of 24 (14.0%) patients had prematurely discontinued the study, a total of 147 patients is still active in the study.

The mean duration of treatment for all patients included in the 36-month treatment analysis (12 months of treatment within PROUD-PV and 24-month treatment within CONTINUATION-PV Study) was 176.7 weeks (median 181.6, range: 68 to 232).

Efficacy Results:

The roll-over rate from PROUD-PV Study into its extension study CONTINUATION-PV Study shows that more patients were roll-over in the ropeginterferon alfa-2b arm: 89.6% (95/106) for ropeginterferon alfa-2b-treated and 68.5% (76/111) for HU- treated patients in PROUD-PV. Patients included in the HU arm of the PROUD-PV Study were labelled as best available treatment (BAT) arm in CONTINUATION-PV Study. Patients entered CONTINUATION-PV Study after a median of 14 days (range 0-122 days) in the ropeginterferon alfa-2b arm, but significantly later after a median of 148 days (range 69-481 days) in the BAT arm.

All 95 patients enrolled in the ropeginterferon alfa-2b arm received the study drug. The overall number of ropeginterferon alfa-2b doses administered was 1844 doses. The mean dose of ropeginterferon alfa-2b at 12 months (EoT of PROUD-PV Study) for the 36-month population (n=95) was 391 (±137) µg, the median dose was 450 (range: 50 - 500) µg administered by assessment visit. At 24 months the mean dose was 377 (±142) µg, and the median was 450 (range: 50-500) µg. At 36 months, the mean dose of ropeginterferon alfa-2b administered by assessment visit was 363 (±149) µg; the median dose was 425 (range: 35 - 500) µg

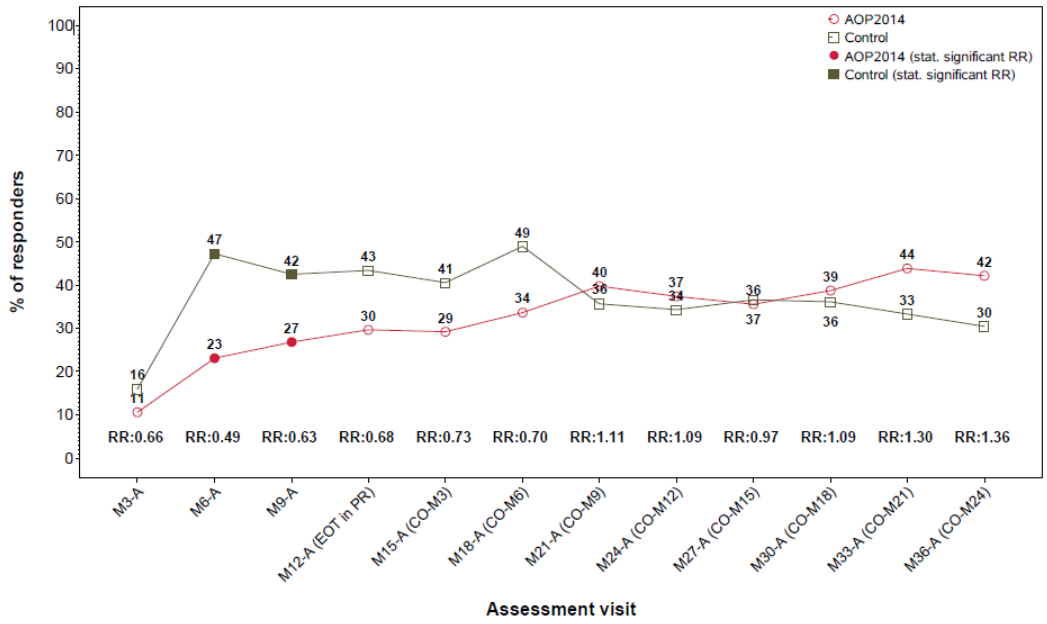
The 76 patients enrolled in the BAT arm received best available treatment for PV disease (at discretion of the investigator). All 76 patients in the BAT arm received HU as the primary treatment for PV at least once during the CONTINUATION-PV study; at month 12 95.8% (46/48), at month 24 98.4% (61/62) and at month 36 97.0% (64/66). The median dose for the control arm remains constant in patients transitioned from the PROUD-PV Study (solely HU-treated) to the CONTINUATION-PV Study (BAT option with almost all patients treated further with HU). The efficacy results indicated that the maximum responder rate was not achieved within PROUD-PV Study (up to Month 12).

Complete haematological response and spleen size normality

Table 28: Disease response (complete haematological response and spleen size normality) at assessment visits (main results) (FAS – 36 month treatment population)

	Disease response			
Visit	**ropeginterferon alfa-2b**		**BAT**	
	N/ Nmiss	N (%)	N/ Nmiss	N (%)
M12	91/4	27 (29.67%)	76/0	33 (43.42%)
M15	89/6	26 (29.21%)	37/39	15 (40.54%)
M18	89/6	30 (33.71%)	47/29	23 (48.94%)
M21	88/7	35 (39.7%)	56/20	20 (35.71%)
M24	91/4	34 (37.36%)	67/9	23 (34.33%)
M27	90/5	32 (35.56%)	71/5	26 (36.62%)
M30	93/2	36 (38.71%)	72/4	26 (36.11%)
M33	89/6	39 (43.82%)	69/7	23 (33.33%)
M36	90/5	38 (42.22%)	69/7	21 (30.43%)

Figure 12: Complete haematologic response and spleen normality at assessment visits (FAS – 36 month treatment population)

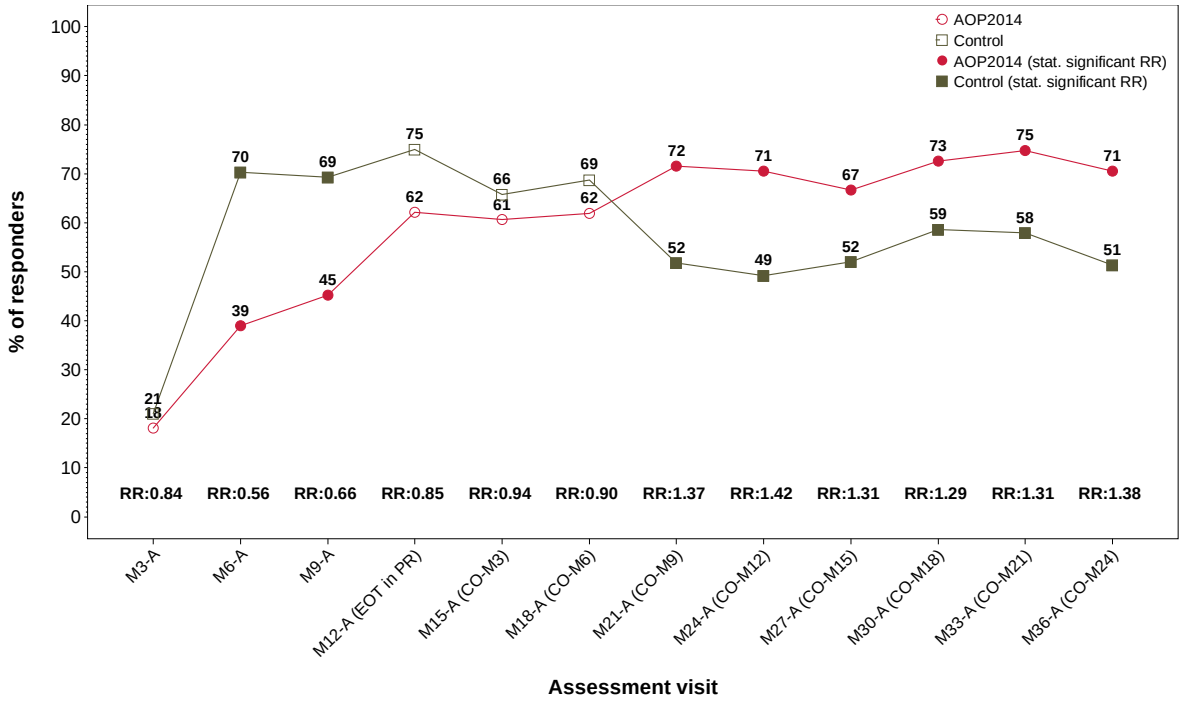


Complete haematological response (without spleen size normality)

Table 29: Complete haematological response at assessment visits (main results) (FAS – 36 month treatment population)

	Disease response			
Visit	**ropeginterferon alfa-2b**		**BAT**	
	N/ Nmiss	N (%)	N/ Nmiss	N (%)
M12	90/5	59 (62.11%)	76/0	57 (75.00%)
M15	94/1	57 (60.64%)	38/38	25 (65.79%)
M18	92/3	57 (61.96%)	48/28	33 (68.75%)
M21	95/0	68 (71.58%)	56/20	29 (51.79%)
M24	95/0	67 (70.53%)	67/9	33 (49.25%)
M27	93/2	62 (66.67%)	75/1	39 (52.00%)
M30	95/0	69 (72.63%)	75/1	44 (58.67%)
M33	95/0	71 (74.74%)	76/0	44 (57.89%)
M36	95/0	67 (70.53%)	74/2	38 (51.35%)

Figure 13: Complete haematologic response and spleen normality at assessment visits (FAS – 36 month treatment population)



Complete haematological response and improvement in disease burden

Table 30: Complete haematological response and improvement in disease burden at assessment visits (main results) (FAS – 36 month treatment population)

	Disease response			
Visit	**ropeginterferon alfa-2b**		**BAT**	
	N/ Nmiss	N (%)	N/ Nmiss	N (%)
M12	95/0	44 (46.3%)	76/0	39 (51.3%)
M15	94/1	46 (48.9%)	52/24	21 (40.4%)
M18	92/3	42 (45.7%)	60/16	28 (46.7%)
M21	95/0	50 (52.6%)	65/11	24 (36.9%)
M24	95/0	47 (49.5%)	71/5	27 (38.0%)
M27	95/0	43 (45.3%)	76/0	32 (42.1%)
M30	95/0	49 (51.6%)	76/0	35 (46.1%)
M33	95/0	53 (55.8%)	76/0	30 (39.4%)
M36	95/0	50 (52.6%)	74/2	28 (37.8%)

Figure 14: Complete haematological response and improvement in disease burden at assessment visits (FAS – 36 months population)

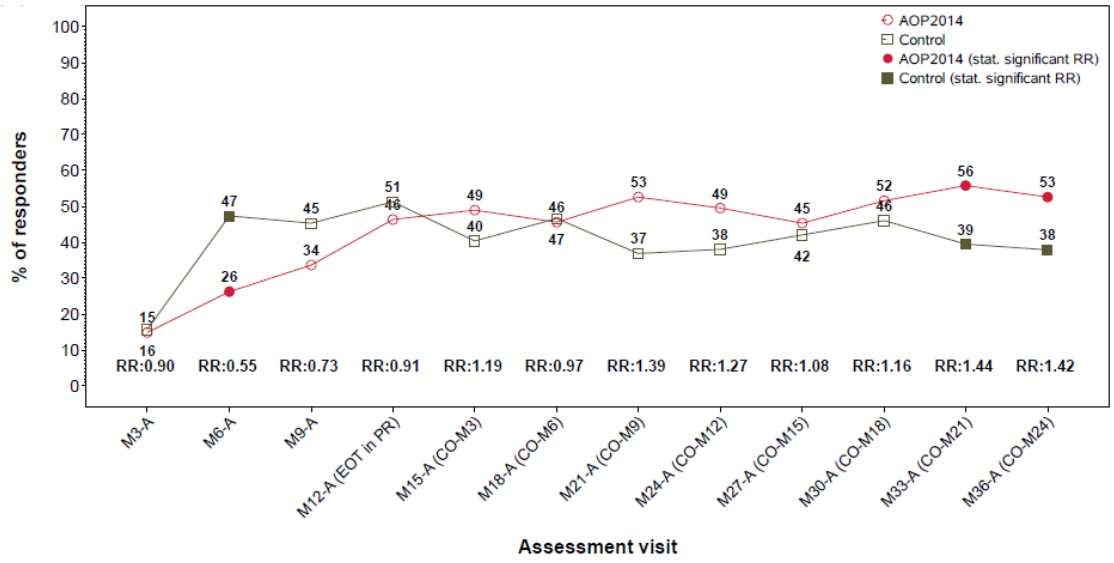


Table 31: Secondary endpoint outcome in CONTINUATION-PV

	Ropeginterferon alfa-2b treatment arm	**BAT treatment arm**
Best observed haematological response	74.7% (71/95) at Month 33 95%CI:NA	75.0% (57/76) at Month 12 95%CI:64.2, 83.4
Hct in target range	Month 12: 66.3% (63/95) Month 24: 80.7% (71/88) Month 36: 80.7% (67/83)	Month 12: 80.3% (61/76) Month 24: 64.1% (41/64) Month 36: 77.9% (53/68)
Leukocytes in target range	Month 12: 92.6% (88/95) Month 24: 96.6% (85/88) Month 36: 98.8% (82/83)	Month 12: 96.1% (73/76) Month 24: 82.8% (53/64) Month 36: 86.8% (59/68)
Platelets in target range	Month 12: 93.7% (89/95) Month 24: 93.2% (82/88) Month 36: 94.0% (78/83)	Month 12: 92.1% (70/76) Month 24: 81.3% (52/64) Month 36: 79.4% (54/68)
Change in spleen size from baseline over time up to last patient visit	Mean differences from baseline (Month 0) ranged from −1.3 cm (Month 36) to 0.1 cm (Month 12)	Mean differences from baseline (Month 0) ranged from -1.24 cm (Month 12) to -0.33 cm (Month 36).
Maintenance of response	Shift in disease response (non-responder to responder) ranged between 9.2% (6/65) at Month 18 and 13.6% (8/59) at Month 21. Complete haematological response was achieved and maintained in 38.9% of patients over 36 months of treatment. In the third year of treatment (Month 24 to 36), 70.5% of patients maintained their response.	Shift in disease response (non-responder to responder) ranged between 8.0% (2/25) at Month 24 to 18.2% (6/33) at Month 18. Complete haematological response was achieved and maintained in 1.45% of patients over 36 months of treatment. In the third year of treatment (Month 24 to 36), 46.1% of patients maintained their response.
Phlebotomy need	Month 12: 28.4% (27/95) Month 24: 14.8% (13/88) Month 36: 14.5% (12/83)	Month12: 10.5% (8/76) Month 24: 18.8% (8/76) Month 36: 10.3% (7/68)
Change in QoL (EQ-5D-3L) from baseline over time up to treatment month 36	Month 12: Mean total score: 6.0 (SD ±1.28) Change from baseline: 0.2 (SD ±1.08) Month 24: Mean total score: 5.8 (SD ±1.19) Change from baseline: 0.0 (SD ±1.11) Month 36: Mean total score: 5.7 (SD ±1.15)	Month 12: Mean total score: 5.9 (SD ±1.26) Change from baseline : 0.1 (SD ±1.17) Month 24: Mean total score: 5.8 (SD ±1.17) Change from baseline: -0.1 (SD ±1.31) Month 36: Mean total score: 6.0 (SD ±1.53)

	Change from baseline: 0.0 (SD ±1.14)	Change from baseline : 0.1 (SD ±1.47)
Mean absolute levels of JAK2 allelic burden	Month 12 23.7% Month 24: 14.3% Month 36: 9.5%	Month 12: 18.2% Month 24: 28.3% Month 36: 42.3%

The best observed disease response rate (defined as complete haematological response and spleen size normality) in the ropeginterferon alfa-2b arm was achieved at Month 33 with 43.8% (39/89), in the BAT arm at Month 18 with 48.9% (23/47). The best observed haematological response in the ropeginterferon alfa-2b arm was again achieved at Month 33 with 74.7% (71/95), whereas the best observed haematological response in BAT arm was already achieved at Month 12 (EOT PROUD-PV Study) with 74.7%.

Up to Month 36, the number of patients with at least one phlebotomy recorded in last 90 days, steadily decreased: 20.5% (35/171) at Month 12 to 12.6% (19/151) at Month 36.

In the ropeginterferon alfa-2b arm the decrease was more prominent: 28.4% (27/95) to 14.5% (12/83), respectively at Month 12 and 36 In the BAT arm, the reverse trend was observed, in that the number of patients with at least one phlebotomy recorded within the last 90 days, increased from 10.5% (8/76) at Month 12 to 18.8% (12/64) at Month 24, and then a decrease to Month 36 to 10.3% (7/68).

No patient reached complete molecular response. (Partial) molecular response was observed for 43.6% (41/94; month 12), 68.1% (64/94; month 24) and 66.0% (62/94; month 36) in patients treated with ropeginterferon alfa-2b (LOCF). In the BAT arm, the (partial) molecular response was observed for 50.7% (38/75; month 12), 33.3% (25/75; month 24) and 27.0% (20/74; month 36).

The median absolute levels of JAK-2 allelic burden ranged in the ropeginterferon alfa-2b treatment arm from 9.5% (Month 36) to 23.7% (Month 12). The median differences from baseline (Month 0) ranged from −24.3% (Month 36) to –12.6%% (Month 12). The median absolute levels of JAK-2 allelic burden ranged in the BAT treatment arm from 18.2% (Month 12) to 42.5% (Month 30). The median differences from baseline (Month 0) ranged from −13.7% (Month 12) to 2.5% (Month 36).

In CONTINUATION-PV Study, the comparator arm was changed to BAT/HU; however, 100% of the patients received at least once HU as the primary treatment for PV. At 36-months (24 months of CONTINUATION-PV Study), 97.0% still received HU as the primary treatment for PV. The roll-over rate from PROUD-PV Study in the extension study CONTINUATION-PV was higher in the ropeginterferon alfa-2b arm with 89.6% (95/106) previously ropeginterferon alfa-2b-treated compared with 68.5% (76/111) previously HU-treated patients in PROUD-PV. Although imbalances increased and more drop-outs in the comparator arm occurred, the results are considered relevant for the efficacy discussion. Additionally, these results will allow assessing duration of efficacy for both drugs at least to some degree.

At this time, only interim analyses of efficacy from this ongoing long term trial are available (i.e. 24-month treatment analysis and 36-month treatment analysis)

Table 32: Maintenance rate of disease response at assessment visits (main results, informative only until M18)

	Disease response			
Visit	**Ropeginterferon alfa-2b**		**BAT**	
	N/ Nmiss	N (%)	N/ Nmiss	N (%)
M12	95/0	44 (46.3%)	76/0	39 (51.3%)
M15	94/1	46 (48.9%)	52/24	21 (40.4%)
M18	92/3	42 (45.7%)	60/16	28 (46.7%)
M21	95/0	50 (52.6%)	65/11	24 (36.9%)
M24	95/0	47 (52.6%)	71/5	27 (38.0%)
M27	95/0	43 (49.5%)	76/0	35 (42.1%)
M30	95/0	49 (51.6%)	76/0	35 (42.1%)
M33	95/0	49 (51.6%)	76/0	30 (39.5%)
M36	95/0	50 (52.6%)	74/2	28 (37.8%)

Within the first 24 months of treatment with ropeginterferon alfa-2b clinical response rates (defined as CHR and improvement of clinical symptoms/signs) consistently increased (46.3% at Month 12, 52.6% at Month 36), whereas the response rates in the control arm (Best available treatment arm, majority of patients still receiving HU) decreased after 12 months of treatment (51.3% at Month 12, 37.8% at Month 36).

Clinical response rates (defined as CHR and improvement of clinical symptoms/signs) remained stable in both treatment arms in the third year of treatment (52.6% and 37.8%, respectively in the ropeginterferon alfa-2b and control arm at Month 36).

Preliminary results from the current interim analysis may indicate that the differences in maintenance of response become similar between ropeginterferon alfa-2b and BAT/HU during long treatment and this seems to be mainly caused by an increase and stabilisation haematological parameters in target range, while impact on spleen size is difficult to interpret and overall less informative in this still early stage population. Interestingly, the need for phlebotomy seemed to be significantly lower after longer treatment periods (e.g. Month 21).

2.5.3. Discussion on clinical efficacy

This application is based on a single pivotal phase 3 trial. The PROUD-PV trial was an open-label, randomized, controlled, parallel-group, non-inferiority study comparing the efficacy and safety of ropeginterferon alfa-2b (AOP2014) over hydroxyurea (HU) over 12 months in 254 patients with Polycythemia Vera. Patients at EOT at 12 months who responded on treatment with ropeginterferon alfa-2b had the opportunity to continue treatment in the ongoing CONTINUATION-PV trial (supportive extension trial).

Two other trials were performed by the applicant: The phase 1/2 trial PEGINVERA-PV aiming to characterise MTD and PK and the PEN-PV trial (phase 3) aiming to assess the self-administration of ropeginterferon alfa-2b using a pre-filled pen. A small PK comparison trial (in healthy adults) with ropeginterferon alfa-2b (P1101) was performed by a different sponsor.

In PROUD-PV Study, randomization and stratification (based on the age: older than 60 years, or younger than 60 years at entry, occurrence of at least one thrombotic event in the past [yes/no], and previous HU exposure [yes/no]) adhered to the general principles laid down in current guidelines (CHMP/ICH/363/96).

It is acknowledged that a double-blind design was not an option due to obvious differences in routes of administration (s.c. vs. oral) and toxicity between study regimens. So the study was open label. Methods were implemented to limit potential bias: relevant blood values at month 12 (primary endpoint) and other endpoints

were measured by a central laboratory, blinded to treatment assignment. Spleen size was assessed by observer-independent imaging (computed tomography/magnetic resonance imaging [CT/MRI]) with blinded radiologic assessment at screening and month 12.

The applicant decided to set the 540 µg dose as MTD although no dose limited toxicity was observed as outcome of both stages of PEGINVERA-PV trial. Thus, it seems questionable whether this dose really represents the MTD. Overall, dose selection was triggered by tolerability reasons mainly and the level of evidence that efficacy at this dose is equivalent to other pegylated interferons was low. No phase II trial was performed. Nevertheless, the dose finding is deemed acceptable considering the limitations clinical development programs in an orphan disease entity.

Due to the absence of an adequate dose finding in a phase II trial, the proposed dose titration in the PROUD-PV trial for ropeginterferon alfa-2b was very conservative. Since dose escalation of ropeginterferon alfa-2b was performed in 50 µg steps up to 500 µg every two weeks, time until full efficacy becomes evident was foreseeable prolonged. A 20 weeks difference to reach the maximum dose plateau was observed between the arms and probably has significantly biased the outcome in favour for HU.

Patients with known risk or contraindication for interferon and hydroxyurea were adequately defined. Additionally, subjects that might have additional intrinsic risk or adding potential difficulties in interpreting the safety outcome are also sufficiently excluded. Overall, the selected population is in accordance with that applied for in this procedure. Although presence of thromboembolic events in the past was used as a stratification factor, differences regarding cardiovascular risks between both arms are not excluded.

HU is the most commonly used approved cytoreductive agent in this disease. This comparator was accepted by the CHMP during a scientific advice procedure since there is no interferon approved for the applied indication. However, due to the known differences in response kinetics between cytotoxic drugs and interferons, time to response is likely to differ significantly. Dose titration and exposure with HU was adequate in PROUD-PV.

The primary endpoint as a composite of clinically relevant outcomes measures of treatment efficacy is acceptable. However, intrinsic difficulties in assessing composite endpoints in small orphan disease population have to be considered. The secondary endpoints allow differentiating the outcome for the different haematological components of the composite endpoint as well as time to response and duration of response. They are all clinically relevant and discriminative. Moreover, the inclusion of change of JAK2 allelic burden over time allows some information about treatment effects on disease related important molecular marker.

The study was initially designed as a superiority trial, but switched to a non-inferiority trial after completion of the study (last patient out, prior database lock). The sample size calculation was based on assumptions for superiority and the change could not influence sample size calculation. The method that was used for blinded sample size re-assessment does in principle not require adjusting the level of significance. The statistical methods are generally considered appropriate, but neither a statistical nor a clinical justification of the -10.5% NI or -20% NI margin was provided. Therefore, from a biostatistical point of view, the actual differences between treatments as well as the results for the additional endpoints need to be interpreted in an exploratory sense only.

Baseline demographic characteristics were balanced between both arms. The same is true with respect to the measurable haematological disease parameters, the spleen size and median JAK2 allelic burden. No relevant imbalances at baseline were noted. Moreover, the drop-out proportion was low and slightly higher for ropeginterferon alfa-2b than for HU. Sensitivity analyses available are considered sufficient to assess impact of drop-outs.

CONTINUATION-PV trial was a follow up study designed to provide long-term evaluation of ropeginterferon alfa-2b in patients with PV who were previously treated in the PROUD-PV Study. Subjects who participated in the PEN-PV Study in order to evaluate an alternative mode of administration (self-administration with PEN) were also integrated in this trial. In a rare disease as PV this overlap of study population is not a major concern.

Additionally, data from the routine care treatment of patients, who participated in the PROUD-PV Study and were allocated to the HU arm, is collected. Currently two interim analyses from this long term trial are available: i.e. 24-month and 36-month treatment analysis (12 months treatment during PROUD-PV study and 12/24 months treatment during CONTINUATION-PV study). The 12/24 months results from the CONTINUATION-PV trial offer the only opportunity for a more valid characterization beyond the 12 months treatment (within PROUD-PV Study) effect with adequately dosed ropeginterferon alfa-2b.

Efficacy data and additional analyses

The pivotal PROUD-PV Study failed to show superiority as well as non-inferiority with respect to the pre-specified primary efficacy endpoint outcome "disease response rate at end of study (12 month)" (i.e. *complete haematological response and spleen size normality*).

Post-hoc the applicant has proposed a redefined primary endpoint "complete haematological response (without spleen normalisation)" in order to demonstrate efficacy. The reason for changing the primary endpoint was that significant splenomegaly was only present in a few patients at baseline and spleen size fluctuations observed during the trial were small in the range of that observed also in a healthy population.

Only for the new post-hoc primary endpoint analysis and post-hoc widened NI margin of – 20% non-inferiority could be demonstrated for ropeginterferon alfa-2b as indicated by a complete haematological response of 43.1% (43/122) in the ropeginterferon alfa-2b arm versus 45.6% (57/123) in the HU arm after 12 months of treatment. The p-value is reported with 0.0028. However, this non-inferiority analysis can hardly be considered confirmatory at least from a methodological point of view.

These weaknesses in reaching clear demonstration of non-inferiority seem to be due to the difference between the arms to reach the maximum dose plateau (on cohort level). In the ropeginterferon alfa-2b arm the maximum median dose plateau level (i.e. end of titration phase) was reached 20 weeks later than in HU arm (week 28 vs week 8). This delay seems to have significantly disadvantaged APO2014 in comparison to HU. At EOT endpoint assessment HU patients were already treated adequately for 10 months, while ropeginterferon alfa-2b patients had only 6 months comparable treatment intensity. At the end source of bias may be reasonably presumed. A comparison of results from the CONTINUATION-PV trial allows to further investigate this issue: Estimating efficacy after 12 and 24 months effective treatment with ropeginterferon alfa-2b and BAT (majority treated with HU) disease response increased to 70.5% (67/95) in the ropeginterferon alfa-2b arm at month 24 and month 36, and 49.3% (33/67) at month 24 and 51.4 (38/74) at month 36 in the BAT arm. Insofar, a similar efficacy after adequate treatment intensity seems likely.

Most importantly, PV is a disease without spontaneous remission. Thus, considering the absolute benefit of a treatment with ropeginterferon alfa-2b based on a comparison to baseline ropeginterferon alfa-2b showed a clear cytoreductive effect. An efficacy comparison of the differences between the four AOP efficacy studies (PEGINVERA; PROUD, CONTINUATION and PEN-PV) shows that the response rate for the haematological parameters Hct, PLT and leukocytes (complete haematological responders) ranged between 43% and 75% in the clinical studies due to different exposure times. It is obvious that response may further increase and can be maintained in up to 70.5% as demonstrated from the interim results after 24 month treatment duration from the CONTINUATION-PV trial (i.e. 36-month overall treatment duration). Insofar, explorative results after 36 months

of treatment indicate a similar efficacy as that reported for IFN- α in the literature. Clinical response rates (defined as CHR and improvement of clinical symptoms/signs) consistently increased within the first 24 months of treatment with ropeginterferon alfa-2b (46.3% at Month 12, 49.5% at Month 24) and remained stable in the third year of treatment (52.6% at Month 36). The prolonged time activity due to the very slow dose titration of ropeginterferon alfa-2b is considered acceptable as in non-responders, phlebotomy is used routinely as rescue in order to normalise blood hyperviscosity. Nethertheless, physicians and patients should be adequately informed about the prolonged time until full efficacy.

The JAK2 allelic burden decreased during treatment in both treatment cohorts (baseline 42.8% and 42.9% in the two treatment arms). The median absolute levels of JAK-2 allelic burden ranged in the AOP204 treatment arm from 9.5% (Month 36) to 23.7% (Month 12); The median absolute levels of JAK-2 allelic burden ranged in the BAT treatment arm from 18.2% (Month 12) to 42.5% (Month 30). At EOT (month 12) the difference was not statistically significant (p=0.0736) between the treatment cohorts, but numerically with 30.7% and 25.9% in favour for AOP2014 versus HU treated patients, respectively. This may be seen as a benefit regarding clinical outcome for ropeginterferon alfa-2b, although the clinical relevance remains unknown at the time being. Nevertheless, it should be considered that from the treatment alternatives in PV only interferon (and probably busulfan) offers at least the chance for complete molecular response in PV patients. Until now, complete molecular response with respect to *JAK2*V617F has been reported in 14% to 24.1% of interferon-treated PV patients, but clearly long-term treatment is needed to achieve this goal. Long-term absence of disease activity in these patients has been reported in the literature (Perricone et al, 2017).

Many clinical trials in the past have characterized the efficacy of interferon-alfa in PV. Thus, external validity for the use of interferons in the applied indication is high, but reflects off-label use only. Since no interferon product is currently approved in the EU for the applied orphan disease target population, hydroxyurea had to be used as comparator in the pivotal trial. In all relevant treatment guidelines interferons are recommended, only the ranking of interferon in the treatment of PV, whether in first or second line, is currently still under discussion. It needs to be considered that the current guidelines (ELN 2018, BSH 2018) recommend the use of pegylated interferon-alfa 2 for first line in patients of all ages.

Additional expert consultation

The SAG Oncology was requested to discuss open issues helping to address the ranking of ropeg-IFN-alfa-2b in the treatment of PV. The CHMP was asked the SAG Oncology the following questions:

1. **The proposed posology of Besremi for the PROUD-PV trial includes a very conservative dose titration which resulted in a delay of 20 weeks to reach the maximum dose plateau between the treatment arms compared with HU. This difference has obviously significantly biased the outcome in favour for the comparator HU and is likely to explain the failure to reach the pre-specified primary endpoint at 12 months in PROUD-PV, while results at 18 months from the extension trial CONTINUATION-PV might indicate non-inferiority in efficacy. Although the dose-titration approach was associated with a favourable safety profile in comparison with HU, it has negatively affected the efficacy outcome at 12 months, since a run-in phase was not included in the trial. Considering the available clinical experience with interferons in PV, the SAG Oncology is requested to comment whether such a conservative dose titration as performed in the PROUD-PV trial is also used in clinical practise with other pegylated interferons in PV patients treated off-label with other pegylated interferons.**

As a general comment, the SAG considered that the pivotal study (PROUD-PV) suffered of critical deficiencies in the design and analysis, making it impossible to formally establish non-inferiority of Besremi compared to HU at

any time point. Furthermore, there are no data to establish that a different titration strategy would have resulted in a better efficacy and acceptable safety of the product. Similarly, any claims about the observed non-inferiority of Besremi at later time points remain to be established given the potential selection bias and lack of pre-specification and handling of multiplicity. Although titration is performed with any type of agent, including off-label pegylated interferons, one cannot extrapolate dosing strategies between different molecules without evidence of similarity between products.

Nevertheless, the SAG agreed that a clinically meaningful effect in terms of response rate has been established and even if formally one cannot exclude a 20% worse response rate compared to HU at 12 months, the loss of efficacy is not considered critical as phlebotomy can be used in the short term to compensate any lack of effect. Availability of a treatment option without the mutagenic and carcinogenic risks associated to HU is certainly of interest, particularly in younger patients more concerned about life-long exposure and especially if in a reproductive age. The toxicity profile observed with Besremi was considered acceptable. Thus, Besremi should only indicated in patients for whom agents with higher activity are not preferred based on the clinical decision that should be shared with physicians experienced in the disease. These types of decisions are normal in clinical practice given the established use of off-label interferon preparations.

2. **Considering the outcome of the pivotal trial PROUD-PV for the post-hoc primary endpoint CHR, demonstration for non-inferiority of Besremi was only reached, if the NI margin was widen to -20%. From a methodological point of view this might indicate that a higher loss of effect (at least at 12 months) than acceptable for a convincing demonstration of non-inferiority cannot be excluded. On the other side phlebotomy, used as rescue treatment, may neutralise potential thromboembolic risks resulting from the very prolonged dose titration in the ropeginterferon-alfa-2b's cytoreductive efficacy in comparison to HU. The SAG Oncology is requested to comment whether the higher loss of effect regarding CHR response under discussion for Besremi in comparison with HU during the first year of treatment is clinically relevant, considering the possibility of additional treatment with phlebotomy.**

A potential loss of 20% or more in CHR is considered clinically significant, especially in the initial treatment where HU is preferred to achieve a rapid response. However, given the availability of phlebotomy, this loss in the short term (<12 months) is considered acceptable even considering a potential for higher risk of thrombotic events (see answer to question No. 1). However, the benefits and risks associated with Besremi need to be clearly communicated patients and physicians. Besremi should be indicated in patients for whom agents with higher activity are not preferred based on an informed clinical decision. It should also be made clear that inferior efficacy compared to HU after 12 months has not been formally established and that patients are likely at higher risk of thrombotic events during the first 12 months (even if the trial did not show clear harms). Thus, before and after 12 months of starting treatment with Besremi, it is important to consider the benefits and risks compared to established active agents like HU and ruxolitinib (where applicable).

3. **The SAG is requested to comment, whether the indication of the applied ropeginterferon-alfa-2b (Besremi) needs to be restricted according the data available for the second line (e.g. "for the treatment of patients with PV without splenomegaly in whom HU is not a suitable treatment [HU intolerant or HU resistant]") or whether it can be approved with the applied broader indication ("treatment of polycythaemia vera (PV) in adults without symptomatic splenomegaly") taking the documented long clinical experience with interferons in this disease into account.**

Based on the available evidence and benefit-risk considerations in the clinical decision, the SAG agreed that the indication should be restricted to patients with PV without splenomegaly for whom treatment with established

active agents like HU or ruxolitinib (where applicable) is not considered acceptable due to the associated risks, and for whom a small increased risk of thrombosis associated with lower activity, especially during the first 12 months, is considered acceptable. Patients should be adequately informed about the benefits, risks, and uncertainties of available options. This is considered feasible in clinical practice.

2.5.4. Conclusions on the clinical efficacy

The efficacy of Besremi in the indication of polycythaemia vera has been demonstrated.

The CHMP recommends the following measures necessary to address issues related to efficacy:

- The final analysis from the CONTINUATION-PV trial.

2.6. Clinical safety

Although the here applied product is currently not marketed, safety assessment needs to consider that similar products containing peginterferon alfa-2b are approved and have been frequently used in other indications (e.g. Hepatitis C, myeloproliferative disorders like CML and also off-label for PV).

At least two types of peginterferon products are currently available on the EU market (Plough's PEG-Intron®; PEGASYS®). As other PEGylated products, both differ in regards to their pharmacokinetics (PK) and recommended dosing in patients. The here applied product was designed to be more stable ("third-generation PEGylated interferon") and allows a twice-monthly (i.e., once every two weeks) s.c. injection for use in PV-population. However, with respect to primary and secondary pharmacology Besremi seemed to be rather similar to the other products.

Thus, the general safety profile of ropeginterferon alfa-2b can be seen as well characterized and safety data provided for Besremi can be compared with that of the already approved products regarding consistency or differences.

INFs in general are used since decades in several indications, insofar the safety profile including long-term safety of interferons is very well characterized, whether they are PEGylated or not.

Specific clinical safety data for Besremi is derived from the PEGINVERA, PEN-PV and the pivotal trials PROUD-PV as well as from the extension trial CONTINUATION-PV(long-term data); for details of these trials please refer to the efficacy section above. Long term safety is available from the phase I PEGINVERA and phase III CONTINUATION-PV trials.

Data from the pivotal phase III trial PROUD-PV (and the extension trial CONTINUATION-PV) allow a safety comparison with Hydroxyurea (or BAT).

For analyses of the relevant safety outcome (TEAEs, ADRs, SAEs and SADRs) the applicant has pooled the data, but differences between the studies are assessable from the presented tables.

Long term experience up to 7 years is available from some patients in the PEGINVERA trial.

There is a lack of information regarding subgroups of patients with higher safety risks (e.g. in general autoimmune and uncontrolled thyroid diseases, psychiatric disorders, severe cardiovascular disease, immunosuppressed transplant recipients), because these patients were excluded from the clinical trials. Treatment of these subgroups is adequately contraindicated in the product information.

Patient exposure

At time of cut-off (29 May 2018) in total 178 patients were exposed and a number of patients participated in consecutive PV studies with the study drug (PROUD-PV Study, CONTINUATION-PV Study and PEN-PV Study). In all clinical studies with ropeginterferon alfa-2b, patients were exposed to the drug for a mean of 38 months; constituting 548.8 patient-years.

Table 33: Overall HU and ropeginterferon alfa-2b exposure in the clinical trials

	Ph III (PROUD-PV)	**Ph III (PROUD-PV)**	**Ph I/II (PEGINVERA)**	**Ph III combined (PROUD-PV + PEN-PV + CONTI-PV)**	**All studies combined (PROUD-PV + PEN-PV + CONTI-PV + PEGINVERA)**
Medicinal product	**Hydroxyurea (HU)**	**Ropeg-interferon alfa-2b**	**Ropeg-interferon alfa-2b**	**Ropeg-interferon alfa-2b**	**Ropeg-interferon alfa-2b**
	N=127	N=127	N=51	N=127	N=178
Exposure to study drug [days] Mean (±SD) Min/Median/Max	336 (±82.95) 14/364/385	336 (±74.97) 28/399/ 336	1379 (±962.2) 14/1816/2604	1024 (±489.3) 43/1201/1568	1125 (±676.7) 14/1237/2604
Exposure to study drug [weeks] Mean (±SD) Min/Median/Max	48 (±11.85) 2/52/55	48. (±10.7) 4/52/57	197 (±137.5) 2/259/372	146 (±69.9) 6/172/224	161 (±96.7) 2/177/372
Exposure to study drug [months] Mean (±SD) Min/Median/Max	12 (±2.9) 0.5/13/13	12 (±2.5) 1/13/14	46 (±32.1) 0/61/87	34 (±16.3) 1/40/52	38 (±22.6) 0/41/87
Patient-Years	N/A	N/A	192.7 years	356.2 years	548.8 years

In the Phase III studies (PROUD-PV, PEN-PV, and CONTINUATION-PV) the mean dose was approximately 360 µg with a median dose of 425 µg. In the PEGINVERA Study (Phase I/II) the median dose was lower with 236.6 µg (median 226.8 µg).

Adverse events

PROUD-PV

Table 34: Overview of adverse events for PROUD-PV Study

		Ropeginterferon alfa-2b		HU		All patients
Characteristics	**Type of event**	**AE**	**n (%)**	**AE**	**n (%)**	**AE n (%)**
All AEs	Any	860	108 (85.0%)	**822**	**113 (89.0%)**	1682 221 (87.0%)
Treatment-emergent	Any	813	104 (81.9%)	**747**	**111 (87.4%)**	1560 215 (84.6%)
Treatment-emergent Adverse Events						
Serious adverse event	No	793	100 (78.7%)	731	111 (87.4%)	1524
	Yes	20	**14 (11.0%)**	16	11 (8.7%)	36
Outcome	Fatal	1	1 (0.8%)			1
	Not Recovered / Ongoing	152	**54 (42.5%)**	84	40 (31.5%)	236
	Recovered	642	94 (74.0%)	640	**110 (86.6%)**	1282
	Recovered with sequelae	16	8 (6.3%)	14	7 (5.5%)	30
	Recovering	2	2 (1.6%)	9	7 (5.5%)	11
Relationship to study treatment	Not Related	444	**92 (72.4%)**	404	82 (64.6%)	848
	Related	369	76 (59.8%)	343	**96 (75.6%)**	712
Relationship to disease under study (PV)	Not Related	716	100 (78.7%)	690	**111 (87.4%)**	1406
	Related	98	38 **(29.9%)**	57	27 (21.3%)	155
Intensity	Grade 1 Mild	556	84 (66.1%)	496	98 **(77.2%)**	1052
	Grade 2 Moderate	223	73 (57.5%)	205	80 **(63.0%)**	428
	Grade 3 Severe	29	21 (16.5%)	45	26 **(20.5%)**	74
	Grade 4 Life-threatening	4	1 (0.8%)	1	1 (0.8%)	5
	Grade 5 Death	1	1 (0.8%)			1

		Ropeginterferon alfa-2b		HU		All patients
Characteristics	**Type of event**	**AE**	**n (%)**	**AE**	**n (%)**	**AE n (%)**
Adverse events of special interest	No	800	104 (81.9%)	740	111 (87.4%)	1540
	Yes	14	11 (8.7%)	7	4 (3.1%)	21

Life-threatening TEAEs in ropeginterferon alfa-2b were Multi-organ failure, Cholecystitis acute, Septic shock and Acute kidney injury – each occurred once. One life-threatening TEAE in the HU arm was Hyponatraemia. One death (Glioblastoma) occurred in the ropeginterferon alfa-2b arm. TEAEs leading to study discontinuation were reported overall in 9/254 patients (3.5%).

Adverse events of special interest were also more frequent in ropeginterferon alfa-2b arm, but overall only observed in a small percentage of patients (ropeginterferon alfa-2b:8,7% vs HU:3.1%).

SAEs after prolonged exposure as available from the pooled PROUD-PV+PEN+CONTI results are only slightly higher than in PROUD-PV (pooled: 12.6 % vs PROUD 11.0%).

In the pooled safety population the percentage of patients reporting at least one ADR was lower for the ropeginterferon alfa- 2b vs. the HU/BAT (66.1% vs. 76.4%, respectively), but the total number of ADRs in the ropeginterferon alfa-2b was slightly higher (590 vs. 431) probably biased also due to the higher number of patients in the ropeginterferon –alfa-2b arm.

In conclusion, the reported key characteristics of safety for ropeginterferon alfa-2b and HU is deemed similar or slightly superior from this analysis; however, limitations from the overall small number have to be considered as well as the overall lower treatment intensity with ropeginterferon alfa-2b in PROUD-PV.

Table 35: Treatment Emergent Adverse Events with Frequency > 5% according SOCs and PTs in ropeginterferon alfa-2b and HU (PROUD-PV Study)

SOC **PTs**	**Ropeginterferon alfa-2b** Number of Events /Percentage of Patients/[n/N]	**HU** Number of Events /Percentage of Patients/[n/N]
Blood and lymphatic system disorders	83 events /28.3% [36/127]	220 events/54.3%/ [69/127]
Anaemia	9 events/6.3%/ [8/127]	50 events/24.4%/ [31/127]
Leukopenia	27 events/8.7%/ [11/127]	53 events/21.3%/ [27/127]
Thrombocytopenia	35 events/15.0%/ [19/127]	99 events/28.3%/ [36/127]
Infections and infestations	56 events/26.0%/ [33/127]	68 events/34.6%/ [44/127]
Influenza	2 events/1.6%/ [2/127]	10 events/7.9%/ [10/127]
Nasopharyngitis	8 events/3.1%/ [4/127]	10 events/6.3%/ [8/127]
Investigations	102 events/37.8%/ [48/127]	48 events/18.9%/ [24/127]
Gamma-glutamyltransferase increased	24 events/14.2%/ [18/127]	1 event/0.8%/ [1/127]
Alanine aminotransferase increased	10 events/7.1%/ [9/127]	1 event/0.8%/ [1/127]
Aspartate aminotransferase increased	9 events/6.3%/ [8/127]	1 event/0.8%/ [1/127]
Platelet count decreased	2 events/1.6%/ [2/127]	14 events/6.3%/ [8/127]
Hepatic enzyme increased	9 events/5.5%/ [7/127]	0 events/0.0%/ [0/127]
Gastrointestinal disorders	52 events/18.1%/ [23/127]	99 events/33.1%/ [42/127]
Nausea	4 events/2.4%/ [3/127]	19 events/11.8%/ [15/127]
Diarrhoea	15 events/6.3%/ [8/127]	20 events/7.9%/ [10/127]

SOC PTs	Ropeginterferon alfa-2b Number of Events /Percentage of Patients/[n/N]	HU Number of Events /Percentage of Patients/[n/N]
Abdominal pain	7 events/5.5%/ [7/127]	11 events/5.5%/ [7/127]
General disorders and administration site conditions	94 events/27.6%/ [35/127]	50 events/20.5%/ [26/127]
Injection site erythema*	0 events/0.0%/ [0/127]	1 event/0.8%/ [1/127]
Injection site pain*	2 events/0.8%/ [1/127]	0 events/0.0%/ [0/127]
Injection site pruritus*	2 events/0.8%/ [1/127]	0 events/0.0%/ [0/127]
Fatigue	43 events/12.6%/ [16/127]	21 events/13.4%/ [17/127]
Asthenia	14 events/5.5%/ [7/127]	7 events/3.9%/ [5/127]
Influenza like illness	7 events/5.5%/ [7/127]	2 events/1.6%/ [2/127]
Pyrexia	11 events/5.5%/ [7/127]	3 events/1.6%/ [2/127]
Nervous system disorders	57 events/21.3%/ [27/127]	44 events/20.5%/ [26/127]
Psychiatric disorders	24 events/12.6%/ [16/127]	16 events/7.9%/ [10/127]
Headache	26 events/9.4%/ [12/127]	17 events/9.4%/ [12/127]
Dizziness	15 events/9.4%/ [12/127]	14 events/7.9%/ [10/127]
Depression*	2 events/1.6%/ [2/127]	1 event/0.8%/ [1/127]
Anxiety*	4 events/3.1%/ [4/127]	3 events/2.4%/ [3/127]
Anxiety disorder*	0 events/0.0%/ [0/127]	1 event/0.8%/ [1/127]
Musculoskeletal and connective tissue disorders	100 events/23.6%/ [30/127]	24 events/15.0%/ [19/127]
Arthralgia	24 events/9.4%/ [12/127]	5 events/3.1%/ [4/127]
Myalgia	13 events/8.7%/ [11/127]	3 events/2.4%/ [3/127]
Back pain	9 events/7.1%/ [9/17]	5 events/3.1%/ [4/127]
Pain in extremity	12 events/7.1%/ [9/127]	3 events/2.4%/ [3/127]
Skin and subcutaneous tissue disorders	78 events/18.1%/ [23/127]	28 events/16.5%/ [21/127]
Pruritus	41 events/7.9%/ [10/127]	9 events/6.3%/ [8/127]
Eye disorders	27 events/15.0%/ [19/127]	16 events/11.0%/ [14/127]
Respiratory, thoracic and mediastinal disorders	20 events/9.4%/ [12/127]	18 events/11.8%/ [15/127]
Cough	7 events/5.5%/ [7/127]	6 events/3.9%/ [5/127]
Vascular disorders	16 events/8.7%/ [11/127]	22 events/11.8%/ [15/127]
Hypertension	6 events/3.9%/ [5/127]	7 events/5.5%/ [7/127]
Cardiac disorders	21 events/9.4%/ [12/127]	31 events/7.9%/ [10/127]
Metabolism and nutrition disorders	19 events/10.2%/ [13/127]	12 events/7.1%/ [9/127]
Renal and urinary disorders	8 events/5.5%/ [7/127]	10 events/7.1%/ [9/127]
Ear and labyrinth disorders	13 events/5.5%/ [7/127]	8 events/6.3%/ [8/127]
Injury, poisoning and procedural complications	9 events/6.3%/ [8/127]	7 events/5.5%/ [7/127]
Hepatobiliary disorders	8 events/5.5%/ [7/127]	5 events/3.1%/ [4/127]

*PT reported with a frequency <5%.

Table 36: comparison of the most common treatment-emergent adverse events (≥10%) with ropeginterferon alfa-2b and HU (PROUD-PV Study)

Ropeginterferon alfa-2b	HU
Thrombocytopenia (15.0%)	Thrombocytopenia (28.3%) and Platelet count decreased (6.3%)
Gamma-glutamyltransferase increased (14.2%)	Leukopenia (21.3%)
Fatigue (12.6%)	Anaemia (24.4%)
	Nausea (11.(%)

Table 37: Treatment Emergent Adverse Events related to study treatment (Adverse Drug Reactions) with Frequency > 5% according SOCs and PTs in ropeginterferon alfa-2b and HU (PROUD-PV Study)

SOC **PTs**	**Ropeginterferon alfa-2b** Number of Events /Percentage of Patients/[n/N]	**HU** Number of Events /Percentage of Patients/[n/N]
Blood and lymphatic system disorders	71 events/23.6%/ [30/127]	207 events/50.4%/ [66/127]
Anaemia	8 events/5.5%/ [7/127]	47 events/22.0%/ [28/127]
Leukopenia	27 events/8.7%/ [11/127]	53 events/21.3%/ [27/127]
Thrombocytopenia	32 events/14.2%/ [18/127]	95 events/26.8%/ [34/127]
Investigations	56 events/22.0%/ [28/127]	29 events/11.8%/ [15/127]
Gamma-glutamyltransferase increased	15 events/9.4%/ [12/127]	0 events/0.0%/ [0/127]
Alanine aminotransferase increased	7 events/5.5%/ [7/127]	0 events/0.0%/ [0/127]
Platelet count decreased	2 events/1.6%/ [2/127]	14 events/6.3%/ [14/127]
Hepatic enzyme increased	9 events/5.5%/ [7/127]	0 events/0.0%/ [0/127]
Gastrointestinal disorders	19 events/7.1%/ [9/127]	50 events/22.0%/ [28/127]
Nausea	1 event/0.8%/ [1/127]	15 events/9.4%/ [12/127]
Diarrhoea	10 events/3.1%/ [4/127]	8 events/5.5%/ [7/127]
General disorders and administration site conditions	69 events/21.3%/ [27/127]	16 events/8.7%/ [11/127]
Fatigue	34 events/7.9%/ [10/127]	10 events/6.3%/ [8/127]
Influenza like illness	7 events/5.5%/ [7/127]	0 events/0.0%/ [0/127]
Nervous system disorders	15 events/7.9%/ [107127]	8 events/4.7%/ [6/127]
Musculoskeletal and connective tissue disorders	41 events/15.0%/ [19/127]	1 event/0.8%/ [1/127]
Arthralgia	11 events/5.5%/ [7/127]	0 events/0.0%/ [0/127]
Myalgia	11 events/7.1%/ [9/127]	0 events/0.0%/ [0/127]
Skin and subcutaneous tissue disorders	64 events/11.8%/ [15/127]	12 events/7.9%/ [10/127]
Pruritus	36 events/5.5%/ [7/127]	4 events/3.1%/ [4/127]

According the PROUD-PV results the most common ADRs (>10%) of both drugs are qualitatively rather similar as the haematotoxicity at the same time also represent the drug's efficacy. Whether the more frequent events for thrombocytopenia, anaemia and leukopenia reflect a beneficial difference or solely indicate the difference in exposure cannot be decided. In conclusion, according the analysis of the adverse events reported at least the acute toxicities between both arms may be comparable, however, different.

Adverse events of special interest

Regarding the major disease-related cardiovascular and thromboembolic events[2] event rates are 8.7% (11/127) of patients in the ropeginterferon alfa-2b arm and 5.5% (7/127) of patients in the HU treatment arm. The most frequently observed major cardiovascular PV-related adverse events in the ropeginterferon alfa-2b arm were atrial fibrillation [(3 events in 2.4% (3/127) of patients)], while in HU patients most events were caused also by atrial fibrillation [2.4% (3/127)] but also due to cardiac failure [3.2% (3/127)].

[2] Sponsor definition included the following PT terms: Atrial fibrillation, brain stem ischaemia, Budd-Chiari Syndrome, cardiac failure, cardiac failure acute, cerebral ischaemia, deep vein thrombosis, embolism, femoral artery occlusion, gastrointestinal haemorrhagic stroke, haematemesis, haemorrhagic transformation stroke, iliofemoral venous thrombosis, intracardiac thrombus, ischaemic stroke, mesenteric infarction, myocardial infarction, myocardial ischaemia pericardial effusion, peripheral arterial occlusive disease, peripheral circulatory failure, peripheral ischaemia, phlebitis, portal thrombosis, pulmonary embolism, pulmonary infarction, retinal artery embolism, splanchnic vein embolism, splenic infarction, stroke, subarachnoid haemorrhage, thrombophlebitis, transient ischaemic attack, truncus coeliacus thrombosis, unstable angina, venous thrombosis limb

Table 38: Thrombo-embolic events occurring during the individual titration phase in PROUD-PV Study.

Thrombo-embolic events (Sponsor's definition)	ropeginterferon alfa-2b (N=127)		HU (N=127)		All patients (N=254)	
	AE	n (%)	AE	n (%)	AE	n (%)
Titration phase (individual per patient)	2	2 (1.6%)	0	0 (0.0%)	2	2 (0.8%)
PT terms						
Haemorrhagic transformation stroke	1	1 (0.8%)	0		1	1 (0.4%)
Ischaemic stroke	1	1 (0.8%)	0		1	1 (0.4%)

Table 39: Thrombo-embolic events occurring during the maintenance phase in PROUD-PV Study.

Thrombo-embolic events	ropeginterferon alfa-2b (N=127)		HU (N=127)		All patients (N=254)	
	AE	n (%)	AE	n (%)	AE	n (%)
Maintenance period (individual per patient)	3	3 (2.4%)	2	2 (1.6%)	5	5 (2.0%)
PT terms						
Splenic infarction	1	1 (0.8%)	.		1	1 (0.4%)
Intracardiac thrombus	1	1 (0.8%)	.		1	1 (0.4%)
Truncus coeliacus thrombosis	1	1 (0.8%).	.		1	1 (0.4%)
Embolism	.		1	1 (0.8%)	1	1 (0.4%)
Femoral artery occlusion	.		1	1 (0.8%)	1	1 (0.4%)

Phlebotomy need during the individual titration phase (i.e. visits with haematocrit > 45%) ranged between 0-27 visits (median 3.0 [Q1-Q3: 1-6]) and 0-6 visits (median 1.0 [Q1-Q3: 0- 2]), respectively in the ropeginterferon alfa-2b and HU treatment arm. Patients with thrombo-embolic events during the individual titration phase (i.e. in ropeginterferon alfa-2b arm only), did not differ significantly with 0 and 2 visits (for the two identified patients).

With respect to the other adverse events of special interest, the results indicate rather similarity in outcome between the arms; particularly for psychiatric, ocular and immunological events. PROUD-PV data concerning the ocular adverse events as well as from the ocular examinations revealed no increased risks from treatment with ropeginterferon alfa-2b. Moreover, there is no doubt that in interferons depression/anxiety events are a risk as well as immunological adverse events. However, the data available indicate no specific risk derived from ropeginterferon alfa-2b in comparison to other interferons.

Serious adverse event / deaths / other significant events

Serious adverse events

In PROUD-PV Study, slightly more SAEs were reported from the ropeginterferon alfa-2b arm [20 events in 14/127 patients; 11.0%] arm than from the HU arm [16 events in 11/127 patients; 8.7%]. 13 SAEs in 5.6% (10/178) of patients were related to ropeginterferon alfa-2b and 3 SAEs in 2.4% (3/127) of patients to HU.

In CONTINUATION-PV 8 additional SAEs were reported in 7/171 (ropeginterferon alfa-2b: 3.2% vs BAT/HU: 2.1%) patients. One patient in the BAT arm died from acute leukaemia, this SAE was the only one to be assessed related to the study drug. After database lock a two HU additional patients developed malignancies (acute leukaemia/ melanoma).

Deaths

In PROUD-PV Study, there was 1 death in the ropeginterferon alfa-2b arm; during CONTINUATION-PV Study, one additional death in the ropeginterferon alfa-2b compared to 2 in the BAT (HU treated patients) occurred.

Two deaths in the ropeginterferon alfa-2b arm were caused by Glioblastoma during the clinical development (PEGINVERA and PROUD-PV Study).

Laboratory findings

There is no signal detected for an increased renal toxicity in both arms. Median changes for creatinine and blood urea from baseline to EOT are comparable between ropeginterferon alfa-2b and HU arms were comparable.

Several clinically important changes in sodium, potassium and calcium (including a few grade 4 events) were observed similarly in both arms (ropeginterferon alfa-2b: 3 vs HU: 5) during PROUD-PV. Whether these events represent a drug related, a disease related or concomitant medication reason cannot be decided. However, one life-threatening event of hyponatremia occurred in the HU arm.

With respect to the safety assessment of ropeginterferon alfa-2b, an increased hepatotoxicity was observed consistently in PROUD-PV and CONTINUATION-PV trial. While no hepatotoxicity was observed in the HU arm, clinically significantly elevated liver enzymes [gamma-glutamyltransferase (GGT), aspartate aminotransferase (AST), and alanine aminotransferase (ALT)] reported for ropeginterferon alfa-2b. This finding indicates rather a mild hepatotoxicity, but two cases of ALT increased grade 3 were reported in the ropeginterferon alfa-2b arm. However, as it was clarified during the procedure hepatotoxicity is a well-known safety class effect of interferons (including cases of DILI).

Significantly abnormal values for the different parameter mentioned occurred in 2 to 10 % of the ropeginterferon alfa-2b-treated target population during the treatment duration (including extension trial). The increase was most pronounced for the GGT, but overall balanced also for AST and ALT. No “Hy’s Law” case was identified up to now during the clinical development of ropeg-IFN-alfa-2b.

Additionally, the impact of concomitant medication, particularly with NSAIDs on hepatotoxicity was sufficiently clarified. Intake of paracetamol was recommended as standard prior administration of ropeg-IFN-alfa-2b as standard for prevention of typical flu-like adverse events in interferons. Thus this concomitant medication may have also contributed to the differences in ALT/AST and y-GT outcome. (Particularly, as blood samples for laboratory analyses were taken < 36 hours after intake of paracetamol). The impact of other factors on hepatotoxicity (e.g. accumulation of PEG) was considered, but could be sufficiently rejected during the procedure since the total PEG concentration is significantly below the levels probably indication safety risk according the recommendations of the safety working party.

Although hepatotoxicity was not very pronounced in the trial, the treatment is a long term one and from the reports of DILI in the literature for all interferons, hepatotoxicity is considered as the most clinical relevant toxicity of interferons, relevant including also ropeg-IFN-alfa-2b.

Safety in special populations

Information regarding the use in special populations was now provided; however, due to the orphan disease character of PV the value of the events observed in the few patients included is not informative. However, it needs to be considered that safety of interferons in general is well characterised also special population due to the long experience with these products.

Table 40. Subgroup analysis of specified TEAEs by Age group

MedDRA Terms	Age <65 number (percentage)	Age 65-74 number (percentage)	Age 75-84 number (percentage)	Age 85+ number (percentage)
	N = 90	N = 29	N = 7	N = 1
Total TEAEs	77 (85.6%)	22 (75.9%)	4 (57.1%)	1 (100.0%)
Related TEAEs – Total	9 (10.0%)	4 (13.8%)	1 (14.3%)	0 (0.0%)
Intensity ≥ Grade 3	15 (16.7%)	7 (24.1%)	1 (14.3%)	0 (0.0%)
Serious TEAEs – Total	9 (10.0%)	4 (13.8%)	1 (14.3%)	0 (0.0%)
- Fatal	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
- Hospitalization/prolong existing hospitalization	8 (8.9%)	4 (13.8%)	1 (14.3%)	0 (0.0%)
- Life-threatening	0 (0.0%)	0 (0.0%)	1 (14.3%)	0 (0.0%)
- Disability/incapacity	1 (1.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
- Other (medically significant)	1 (1.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
TEAE leading to drop-out	5 (5.6%)	5 (17.2%)	1 (14.3%)	0 (0.0%)
Psychiatric disorders *	13 (14.4%)	2 (6.9%)	1 (14.3%)	0 (0.0%)
Nervous system disorders *	14 (15.6%)	2 (6.9%)	2 (28.6%)	0 (0.0%)

Accidents and injuries #	5 (5.6%)	1 (3.4%)	0 (0.0%)	0 (0.0%)
Cardiac disorders #	5 (5.6%)	3 (10.3%)	1 (14.3%)	0 (0.0%)
Vascular disorders #	5 (5.6%)	3 (10.3%)	1 (14.3%)	1 (100.0%)
Cerebrovascular disorders #	2 (2.2%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Infections and infestations *	24 (26.7%)	6 (20.7%)	3 (42.9%)	0 (0.0%)
Anticholinergic syndrome #	13 (14.4%)	3 (10.3%)	1 (14.3%)	0 (0.0%)
Quality of life decreased §	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Sum of postural hypotension, falls, black outs, syncope, dizziness, ataxia, fractures	9 (10.0%)	3 (10.3%)	0 (0.0%)	0 (0.0%)

* as per MedDRA SOC

as per MedDRA SMQ

§ as per MedDRA PT

Immunological events

Repeated administration of recombinant IFNα, can result in a break in immune tolerance to self-antigens in some patients resulting in the production of anti-IFN antibodies. These antibodies may bind to the IFN molecule in such a manner that they are largely without effect, or they may alter IFN pharmacokinetics, or in certain cases neutralize the activity of the IFN by binding to an epitope that prevents IFN from binding to its cell-surface receptor on target cells. Incidence of anti-IFN antibody production are reported as highly variable (7 to 60%) in the literature, but seemed to be lower in pegylated products.

Interferons are known to increase autoantibody titer in general as a class effect. In particular, antibodies against antinuclear antigens (ANAs), erythrocyte and thyroid antigens have been identified as clinically relevant. Therefore, patients with known autoimmune disease and thyroid dysfunction or thyroid autoantibodies were excluded from the PROUD-PV trial, which is adequately reflected in the contraindications.

The presence of immunologic reactions, including development of anti-thyroid antibodies with clinical symptoms as well as IFN-induced hypersensitivity (anti-IFN antibodies) was tested in the clinical studies; the validity of the methods used was clarified during the response.

In the PROUD-PV trial two clinically significant abnormal values of ANA were observed in the ropeginterferon alfa-2b arm at two visits (week 22 and month 9). Starting with study week 12 inclusively (end of titration phase) one clinically significant abnormality was reported in TgAb in the ropeginterferon alfa-2b arm at each study visit excluding follow-up. One (week 20 and month 12) to two (screening and FU) clinically significant abnormalities were recorded for TPOAb in HU arm.

During longer exposure in the extension CONTINUATION-PV trial anti-thyroglobulin antibody titers were significantly increased in six cases during treatment longer than 18 months. Immunogenicity antibodies are

analysed using frequencies of positive results. Positive results were observed in HU arm only (one case per each study visit excluding visit at week 4).

Safety related to drug-drug interactions and other interactions

No product specific interaction studies have been performed with Besremi.

Discontinuation due to adverse events

Patients discontinued the ropeginterferon-alfa-2b were 21/127 versus 16/127 in the HU arm in PROUD-PV. While withdrawal of consent was balanced between the arm, adverse event associated reasons were more frequent in the ropeginterferon-alfal-2b arm (B: 11/21 versus HU: 3/16). Lack of efficacy leading to discontinuation was only described in 2 patients in the HU arm.

Post marketing experience

Not applicable

2.6.1. Discussion on clinical safety

Due to the rarity of the disease (0.8 -2.6/100 000) a number of patients participated in consecutive PV studies with the study drug (PROUD-PV Study, CONTI-PV Study and PEN-PV Study). The total of 178 of PV patients exposed is considered adequate in the context of the disease and a similar size as required for the approval of Jakavi in PV (Besremi: n=127 / Jakavi =110) and is larger than the PV population initially presented for HU for demonstration of efficacy and safety (N=56).

The comparator was adequately dosed in the trial population (~ 125 mg/kgBW /week) taking a literature recommended dose range of 105 -140 mg/kg/week into account. Although times of exposure with ropeginterferon alfa-2b and the comparator HU in the PROUD-PV trial were balanced, the mean dose of approximately 380 µg (with a median dose of 450 µg) indicates that dose intensity of treatment was relatively low for ropeginterferon alfa-2b and reflects the conservative dose escalation already mentioned. The lower dose intensity during the first 6 months may have resulted in lower drug related adverse event rates in the PROUD-PV trial during this time, as indicated by a decreased need for dose reductions due to drug-related adverse events (ropeginterferon alfa-2b: 25.2% versus HU:51.2%). Overall in the clinical development program, patients were exposed to the drug ropeginterferon alfa-2b for a mean of 38 months, constituting 548.8 patient-years. This indicates that long-term experience is sufficient to allow a valid safety evaluation for this application.

Mode of pegylation of ropeginterferon-alfa-2b- is different compared with other PEG interferons. During the procedure it was discussed at which degree PEG may accumulate in the body and potentially may cause organ damage after long-term administration. The applicant has presented meanwhile sufficient comparative data with other interferons regarding PEG-accumulation in the body and valid calculations to preclude this risk. Since this calculations regarding potential accumulation of ropeg-IFN-alfa-2b in humans were satisfying, non-clinical data are deemed not necessary.

Frequencies of TEAEs during the PROUD-PV trial were slightly higher in the HU arm (TEAE: ropeginterferon alfa-2b: 81.9% vs HU:87.4% /ADR: ropeginterferon alfa-2b: 59.8% vs HU:75.6%) and also the differences with respect to the intensity of TEAE as reflected by the grading differences may indicate a slightly superiority for ropeginterferon alfa-2b. Whether these differences are clinically relevant may be challenged as grade 3 and 4 events were rare and existing imbalances are difficult to interpret due to the small numbers of events.

In the PROUD-PV study the most common ADRs (>10%) of both drugs are qualitatively rather similar as haematotoxicity of both drug at the same time also represent the drug's efficacy. Whether more frequent events of thrombocytopenia, anemia and leukopenia observed in the HU arm reflect superiority in favour for ropeginterferon alfa-2b or solely indicate the difference in exposure cannot be decided. According to the analysis of the adverse events reported at least the acute toxicities between both arms may be comparable, however, differences regarding typical interferon adverse events as fatigue, flu-like illness, myalgia, arthralgia, fever are not surprising and intrinsic characteristics. Hepatotoxicity is also known as a known safety risk for interferons.

Frequency and adverse events reported from the PROUD-PV trial for ropeginterferon alfa-2b and HU are in accordance with that well known for interferons and HU.

With respect to the other adverse events of special interest, the results indicate rather similarity in outcome between the arms; particularly for psychiatric, ocular and immunological events. PROUD-PV data concerning the ocular adverse events as well as from the ocular examinations revealed no increased risks from treatment with ropeginterferon alfa-2b. Moreover, there is no doubt that in interferons depression/anxiety events are risks as well as immunological adverse events.

The proposed contraindications are in line with the exclusion criteria in the trials and with those of already licensed interferons and are considered appropriate. After changes performed as requested after the initial indication, the product information in general seems now adequately to reflect remaining uncertainties regarding ropeginterferon-alfa 2b and restrictions for the use of interferons in general.

SAEs in PROUD-PV were reported slightly more frequent in the ropeginterferon alfa-2b arm (ropeginterferon alfa-2b:11.0% vs HU:8.7%). During longer treatment, as reflected by the CONTINUATION-PV results, rates may become lower and more similar (ropeginterferon alfa-2b: 3.2% vs BAT/HU: 2,1%). However, the differences in safety assessment between both trials suggest that PROUD-PV results are more valid due to the more frequent safety monitoring.

During PROUD-PV and CONTINUATION-PV Study, there were mortality was comparable between the two treatment arms (ropeginterferon alfa-2b:2 vs HU: 2); it is important to consider that the only drug-related death was observed in the HU arm from acute leukaemia. An increase in leukaemia rate is well documented in the literature for Hydroxyurea, while the absence of genotoxicity and carcinogenicity is a well-documented benefit for interferons. In this context it should be noted that after database lock two more HU patients developed malignancies (acute leukaemia/ melanoma).

The risk for TE events is significantly increased in the PV population compared with the general population. That is the reason for including the parameter of previous TE event as a risk factor and an indicator to start cytoreductive therapy in PV disease. This is reflected in significant different TE rates between PV patients receiving cytoreductive therapy (< 10%) and not receiving cytoreductive therapy (32.8% - 44.8%). As in the first 6 months treatment intensity in the ropeginterferon alfa-2b arm was significantly lower, these events may indicate an insufficient treatment effect during this period. Although phlebotomy as rescue treatment normalised a potential blood hyperviscosity increased leucocytes may also contribute to the thromboembolic risk.

In the ropeginterferon alfa-2b arm, the risk ratio of thromboembolic events occurring during the individual titration phase vs the non-titration phase was 1.00 (95% CI: 0.14 to 6.99, p>0.999). The statistical analysis did not show that there is a risk of thromboembolic events associated with the titration phase in the ropeginterferon alfa-2b arm. Taking the even longer titration phase definition (time until maximum dosing plateau is achieved, i.e. 28 weeks for ropeginterferon alfa-2b), again, no increased risk of thromboembolic events during the titration

phase compared to the non-titration phase was identified in the ropeginterferon alfa-2b treatment arm (p>0.999).

Two cases of stroke were observed in the ropeginterferon alfa-2b arm during the titration phase compared to none in the HU arm. It is acknowledged that TE events –although significantly more frequent in PV patients than in healthy population and a major risk of this disease- are overall rare events. However, it raises concerns that both events occurred during the titration phase and thus, may probably indicate a higher risk from the longer dose titration phase for ropeginterferon-alfa-2b. As these events are rare, but can become life-threatening, they are clinically relevant and very important. The observed imbalance is adequately reflected in the SmPC in order to information the target population that a potential higher risk for thromboembolic complications during the first 6 months cannot be excluded at present (see SmPC section 4.4.).

The observation of two cases of Glioblastoma leading to death occurred in the ropeginterferon alfa-2b arm was considered as a chance finding as no similar events were documented in the literature or assessed from post marketing sources as potential signals.

With respect to the laboratory assessment of ropeginterferon alfa-2b an increased hepatotoxicity in terms of mild liver enzyme elevations (GGT, AST, ALT), but not with respect to clinical symptoms, was identified during the initial assessment. The applicant clarified in the response that this potential signal can be seen as a well-known class safety effect of all interferons is classified also in most of the product information of approved products. Hepatotoxicity for ropeg-IFN –alfa-2b observed can be classified as mild, but two cases of ALT increased grade 3 were reported and DILI cases are reported for all interferons in the literature. Moreover, the recommended intake of paracetamol or other NSARs shortly before administration of ropeg-IFN-alfa2b is likely to have contributed also to the elevation of liver enzymes. Overall frequency seems to be between 2 to 10 % in the ropeginterferon alfa-2b treated population (including CONTINUATION-PV results). No case fulfilling Hy's law" criteria were reported.

There is no signal detected for an increased renal toxicity in both arms. Median changes for creatinine and blood urea from baseline to EOT were comparable between ropeginterferon alfa-2b and HU arms. Electrolyte disturbances were balanced, although one life-threatening event of hyponatremia occurred in the HU arm.

Increased autoantibody formation is a well-known interferon class effect, but the frequency (up 6 events in CONTINUATION-PV) and grade of increase observed in the limited target population have not given rise to an additional relevant safety signal in the ropeginterferon alfa-2b treated patients. In comparison to other interferons the frequency appears to be lower. Patients at increased risk for immunological reactions are adequately contraindicated. Development of antibodies against interferon was not reported with ropeginterferon-alfa-2b in the limited target population.

No product specific interaction studies have been performed with Besremi to evaluate drug-drug interactions or other interactions. Information provided in section 4.5 of the SmPC is transferred in accordance with that known for other comparable pegylated interferon alfa 2 products. In general, it is reassuring that safety data submitted was generated in a population in which most patients received concomitant medication relevant in the applied target population without any obvious evidence for interactions.

Data or discussion regarding safety in special populations is very limited due to the small population investigated in this orphan disease. Analyses with respect to the safety outcome in the elderly population, potential safety differences with respect to gender, body weight and patients with increased fragility were provided. Similarly, differences in pharmacokinetics and safety outcome in patients with renal and/or hepatic impairment were discussed. The conclusions were adequately reflected in SmPC due to changes performed as requested.

The impact and relevance of the switching strategy in PROUD-PV for those patients who were pre-treated with HU before inclusion seems to be minor also with respect to the outcome on safety events. The guarded dose escalation strategy in the ropeginterferon-alfa-2b arm seems successful in increasing the treatment compliance. Discontinuation results showed an only slightly lower tolerability of ropeginterferon-alfa-2b interferon in comparison to hydroxyurea.

However the dose escalation strategy may have resulted in a higher risk for thromboembolic complications, remains not clear. Considering the data in literature lowering of Hct below 45% in PV patients is essential for the reduction of the disease-associated thromboembolic risk. But, although the 2 thromboembolic events observed during the titration phase may be a chance finding and in general in clinical trials no difference in rates of thromboembolic complications between interferons and HU was observed, a higher risk for thromboembolic events during the first months of treatment cannot be excluded at the end. This is reflected adequately in the product information and in the RMP with appropriate risk minimisation strategy. It is reassuring that during long-term treatment with ropeg-IFN-alfa-2b the cardiovascular and thromboembolic risks seemed to be adequately reduced, - in line also with literature data.

Comparing the clinical safety data assessed for ropeginterferon alfa-2b with those reported for other pegylated interferons. Hepatotoxicity including cases if DILI was identified to be a class safety effects of interferons known for all interferons. As no other interferon is approved for the target population, a head-to head comparison of safety is not possible. Nevertheless, the prolongation of the administration interval for subcutaneous injections up to 2 weeks in comparison to once weekly may be seen as a safety advantage over other interferons. As predictable occurrence of most drug related adverse events (e.g. flu-like illness, arthralgia, myalgia and fever) is promptly related to the interferon injection, a less frequent need for injections may have a positive impact on safety.

The most important undisputed safety benefit of ropeginterferon alfa-2b compared with hydroxyurea is the well-known complete absence of genotoxicity and carcinogenicity compared to hydroxyurea. Taking into account that two cases of acute leukaemia and one melanoma developed during 18 month treatment with hydroxyurea in a small population of 127 patients, a clinical relevant and very important benefit for ropeginterferon alfa-2b was already demonstrated during the clinical development of ropeginterferon alfa-2b.

Additional expert consultations

See SAG Oncology answers under Clinical efficacy discussion

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

The safety profile of Besremi including long-term safety is very well characterized as of the submitted studies and as this is a third generation of interferon and knowledge can be extrapolated from authorised products. The safety results allow concluding that safety profile of ropeginterferon alfa-2b is different to HU, whereas Besremi seems potentially slightly better regarding the frequency of haematological TEAEs (Anaemia, Leukocytopenia and Thrombocytopenia) and the absence of carcinogenicity. Further results from the CONTINUATION-PV trial will be submitted following a recommendation from the CHMP.

2.7. Risk Management Plan

Safety concerns

Summary of safety concerns	
Important identified risks	• Hepatotoxicity • Thyroid dysfunction • Neuropsychiatric adverse effects • Ocular disorders, including decreased visual acuity, loss of vision, blindness, and retinal detachment • Cardiac events including cardiomyopathy, myocardial infarction, myocardial ischaemia • Pulmonary disorders including pulmonary fibrosis, lung infiltration, pneumonitis and pneumonia • Diabetes mellitus
Important potential risks	• Pulmonary arterial hypertension • Thrombotic microangiopathy • Neoplasms, benign and malignant • Reproductive toxicity/ spontaneous abortions • Demyelating disorders
Missing information	None

Pharmacovigilance plan

Study Status	**Summary of objectives**	**Safety concerns addressed**	**Milestones**	**Due dates**
Category 3 - Required additional pharmacovigilance activities				
PASS ropeginterferon alfa-2b: Safety observational study Planned	To further investigate the safety and tolerability of ropeginterferon alfa-2b with a special focus on hepatotoxicity to evaluate the effectiveness of risk minimisation measures. Secondary objective: evaluation of cardiovascular events during titration phase	Hepatotoxicity	*Study protocol* *Patient enrolment* First patient in Last patient in *Study end* Last patient out *Study report* Interim report Final report	Q2 2019 Q2/Q3 2019 Q3 2021 Q1 2023 Q1 2022 Q3 2023

Risk minimisation measures

Safety concern	Risk minimisation measures	Pharmacovigilance activities
Hepatotoxicity	Routine risk communication: SmPC section 4.2, 4.3, 4.4, 4.8, 5.2 PL section 2, 4 Legal status: Prescription only medicine (POM)	Routine pharmacovigilance activities and additional pharmacovigilance activities (PASS ropeginterferon alfa-2b)
Thyroid dysfunction	Routine risk communication: SmPC section 4.3, 4.4, 4.8 PL section 2, 4 Legal status: POM	Routine pharmacovigilance activities
Neuropsychiatric adverse effects	Routine risk communication: SmPC section 4.3, 4.4, 4.7, 4.8 PL section 2, 4 Legal status: POM	Routine pharmacovigilance activities
Ocular disorders, including decreased visual acuity, loss of vision, blindness, and retinal detachment	Routine risk communication: SmPC section 4.4, 4.8 PL section 2, 4 Legal status: POM	Routine pharmacovigilance activities
Cardiac events including cardiomyopathy, myocardial infarction, myocardial ischaemia	Routine risk communication: SmPC section 4.3, 4.4, 4.8 PL section 2, 4 Legal status: POM	Routine pharmacovigilance activities
Pulmonary disorders including pulmonary fibrosis, lung infiltration, pneumonitis and pneumonia	Routine risk communication: SmPC section 4.4, 4.8 PL section 2, 4 Legal status: POM	Routine pharmacovigilance activities
Diabetes mellitus	Routine risk communication: SmPC section 4.4, 4.8 PL section 2, 4	Routine pharmacovigilance activities

Safety concern	Risk minimisation measures	Pharmacovigilance activities
	Legal status: POM	
Pulmonary arterial hypertension	Routine risk communication: SmPC section 4.4, 4.8 PL section 4 Legal status: POM	Routine pharmacovigilance activities
Thrombotic microangiopathy	Routine risk communication: SmPC section 4.8 PL section 4 Legal status: POM	Routine pharmacovigilance activities
Neoplasms, benign and malignant	Routine risk communication: SmPC section 4.5 PL section 2 Legal status: POM	Routine pharmacovigilance activities
Reproductive toxicity/ spontaneous abortions	Routine risk communication: SmPC section 4.6 PL section 2 Legal status: POM	Routine pharmacovigilance activities
Demyelating disorders	Routine risk communication: SmPC section 4.8 PL section 4 Legal status: POM	Routine pharmacovigilance activities

Conclusion

The CHMP and PRAC considered that the risk management plan version 0.5 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 Annex II, Section C of the CHMP Opinion. The applicant did not request alignment of the PSUR cycle with the international birth date (IBD). The new EURD list entry will therefore use the EBD to determine the forthcoming Data Lock Points.

2.9. New Active Substance

Clinical aspects

The CHMP considered that ropeginterferon alpha 2b as investigated in the submitted studies in terms of pharmacokinetic/ pharmacodynamic properties in PV patients in [2010-018768-18 (PEGINVERA), 2012-005259-18 (PROUD-PV), 2012-005259-18 (CONTINUATION-PV)] has shown :

(i) a prolonged half-life in human serum, allowing the dosing interval to be significantly extended, i.e. dosing once every two–four weeks instead of the traditional once every week; thus leading to:

(ii) improved patient tolerability and compliance.

Due to the prolonged half-life in human serum allowing a prolonged dosing interval patient tolerability and compliance is considered to be improved.

Conclusions on clinical aspects

In conclusion, considering that ropeginterferon alpha 2b exhibited a prolonged half-life which resulted in an extended dosing interval, it can significantly improves patient tolerability, ensuring better compliance therefore having a significant impact on the efficacy and safety of the product.

Therefore, the CHMP concluded that ropeginterferon alfa-2b contained in Besremi can be considered a NAS on the basis of indent 3 of the NTA definition of New Active Substance.

Overall conclusions on New active substance

Problem statement

The applicant requested the active substance, ropeginterferon alfa-2b to be considered a new active substance based on the following claim:

(i) an additional amino acid moiety specifically introduced to the N-terminus of IFN alpha-2b for the purpose of site-specific pegylation;

(ii) the redesign of the activated PEG molecule to a more stable linkage, rather than succinimidyl carbonate PEG used for other authorised interferon products, for conjugation of the PEG molecule with proline-IFN alfa-2b;

Collectively, the two structural innovations described in (i) and (ii) led to ropeginterferon alfa-2b consisting of a largely single N-terminal mono-PEGylated interferon versus other authorised interferon products.

(iii) a 40kDa branching PEG molecule used for ropeginterferon alfa-2b versus the 12kDa linear molecule for the authorised EU-marketed PegIntron.

The branching design described in (iii) led to prolonged half-life without the negative side effects of larger linear molecules.

The improvements in the safety and efficacy of ropeginterferon alfa-2b due to its structural innovations over other authorised interferon products can be summarized as follows:

(i) prolonged half-life in human serum, allowing the dosing interval to be significantly extended, i.e. dosing once every two to four weeks instead of the traditional once every week; leading to:

(ii) improved patient tolerability and compliance.

Scientific evaluation

Quality aspects

Position of the Applicant

The applicant claimed the status of a new active substance for ropeginterferon alfa-2b, as no medicinal products containing the active substance ropeginterferon alfa-2b are currently authorised in the European Union.

Ropeginterferon alfa-2b is a mono-pegylated recombinant analogue of human IFNα-2b. Human IFN alpha 2b in its pegylated form is marketed as PegIntron in the EU.

The non-pegylated drug intermediate of ropeginterferon alfa-2b is a human recombinant interferon alpha-2b with an additional proline residue at its N-terminus that, in total, has 166 amino acids in length.

The drug intermediate is covalently modified with a 40 kilodalton (kDa) branched PEG moiety. The 40 kDa PEG moiety is a branched molecule consisting of two 20 kDa linear mPEG branches that are attached to a specified linker. One 40 kDa PEG moiety is attached to one PEGylation site on the interferon protein (i.e. N-terminal proline residue).

The applicant presented data to show that the pegylation site is predominantly at the N-terminus of the drug intermediate, i.e., ropeginterferon alfa-2b consists of a single predominant mono-PEGylated form with only 2 positional isomers as minor variants. By contrast, PegIntron consists of 14 positional isomers.

The applicant also claimed that intentional pegylation at N-terminal cysteine would be challenging due to the C1 tendency to form internal and external disulfide bonds. Incorrect disulfide bond formation is likely to promote protein aggregation.

Thus, in the applicant's view, Pro-IFN alfa-2b was chosen as the drug candidate because of its N-terminal homogeneity, high refolding yield, and high pegylation yield (Patent US81061607).

Further, the applicant stated that the addition of an amino acid should be considered an extension of the original sequence of IFN alfa-2b that enables targeted N-terminal pegylation of the molecule.

To obtain greater homogeneity, a functional group was also introduced to PEG molecule to act as the active site for selective pegylation. According to the applicant, PegIntron uses a less stable carbamate linkage which results in a fairly short half-life in serum and thus requires a weekly dosing interval for PegIntron. The molecular weight of the PEG molecule in 40kDa branched PEG in Besremi is significantly higher than PegIntron's 12kDa thus impacting on the half-life both *in-vitro* and *in-vivo*.

In summary, the applicant claimed that ropeginterferon alfa-2b is structurally different from other EU-authorised PEGylated-IFN alfa-2b due to the following qualitative aspects:

(i) a novel rationally designed N-terminal site for pegylations which is an additional amino acid chosen for its better structural property so that a single isomer is formed during pegylation;

(ii) a larger, branching 40kDa PEG molecule which reduces side effects while increasing the half-life of ropeginterferon alfa-2b;

(iii) the novel linkage between the PEG molecule and interferon in ropeginterferon alfa-2b which features a functional group responsible for much more reliable conjugation and therefore increased half-life.

Discussion on quality aspects

With regard to the NAS status claimed (i.e. a chemical, biological or radiopharmaceutical substance not previously authorised in a medicinal product for human use in the European Union), the primary molecular structure, which is the basic structural element without added functional structures (e.g., pegylation) needs to be different from the basic structural element contained in a medicinal product authorized in the EU. In the CHMP's view, the primary structure of Besremi (ropeginterferon alfa-2b) is different to Pegasys (peginterferon alfa-2a) due to the differences at aa23 and aa112. IFN alfa 2a and IFN alfa 2b are therefore considered different active substances.

However, when comparing Besremi (ropeginterferon alfa-2b) and peginterferon alfa-2b containing products (such as PegIntron) and considering the basic structural element as the primary structure of interferon alpha-2b, CHMP concluded that Besremi and PegIntron are not different when considering structural aspects alone. Indeed both Besremi and PegIntron contain the same primary structural element interferon alpha-2b and the N-terminal proline in Besremi is an addition to this basic structural element. Consequently based on indent 1 of the NTA definition of New Active Substance, Besremi and PegIntron should be considered known active substances unless it can be shown that these differences have a clinical impact in terms of safety and efficacy (indent 3 of Annex I of Chapter 1 - Volume 2A of the Notice to Applicants).

Conclusions on quality aspects

In conclusion, considering that interferon alpha 2b is the same basic structural element of both Besremi and PegIntron and that the addition of an amino acid at the end of the molecule is not considered to have changed the basic structural element of the active substance, the CHMP concluded that ropeginterferon alfa-2b contained in Besremi cannot be considered a NAS on the basis of indent 1 of the NTA definition of New Active Substance.

Clinical aspects

The CHMP considered that ropeginterferon alfa-2b as investigated in the submitted studies in terms of pharmacokinetic/ pharmacodynamic properties in PV patients in [2010-018768-18 (PEGINVERA), 2012-005259-18 (PROUD-PV), 2012-005259-18 (CONTINUATION-PV)] has shown:

(i) a prolonged half-life in human serum, allowing the dosing interval to be significantly extended versus PegIntron, i.e. dosing once every two to four weeks instead of the traditional once every week; thus leading to:

(ii) improved patient tolerability and compliance.

Due to the prolonged half-life in human serum allowing a prolonged dosing interval patient tolerability and compliance is considered to be improved.

Conclusions on clinical aspects

In conclusion, considering that ropeginterferon alfa-2b exhibited a prolonged half-life which resulted in an extended dosing interval, it can significantly improve patient tolerability, ensuring better compliance therefore having a significant impact on the efficacy and safety of the product.

Therefore, the CHMP concluded that ropeginterferon alfa-2b contained in Besremi can be considered a NAS on the basis of indent 3 of the NTA definition of New Active Substance.

Overall conclusions on New active substance

The CHMP considers, based on the available quality and clinical data, that ropeginterferon alfa-2b contained in Besremi is considered to be a new active substance as it differs significantly in properties with regard to safety and efficacy from interferon alpha-2b contained in medicinal product(s) previously authorised within the European Union due to differences in molecular structure, nature of the source material or manufacturing process.

2.10. Product information

2.10.1. 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.*

2.10.2. Additional monitoring

Pursuant to Article 23(1) of Regulation No (EU) 726/2004, Besremi (ropeginterferon alfa-2b) is included in the additional monitoring list as new active substance which, on 1 January 2011, was not contained in any medicinal product authorised in the EU.

Therefore the summary of product characteristics and the package leaflet includes a statement that this medicinal product is subject to additional monitoring and that this will allow quick identification of new safety information. The statement is preceded by an inverted equilateral black triangle.

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

Besremi (Ropeginterferon alfa-2b) is intended as monotherapy for the treatment of polycythaemia vera in adults without symptomatic splenomegaly.

Polycythaemia vera (PV) -a very rare disease -is the most common myeloproliferative neoplasia, an acquired form of primary erythrocytosis which is characterised by an excess production of erythrocytes, leukocytes and platelets which develops when a mutated haematopoietic stem cell gains a proliferative advantage over other stem cells. and typically develops in late adulthood. Although the underlying molecular mechanism is still controversial, current theories argue that the MPN disease forms are actually linked to a similar set of acquired mutations (JAK2, CLR or MPL), the most important being JAK2V617F that can ultimately result in a transition from a “pre-leukaemic” MPN state to leukaemia. PV is a long-term debilitating and life-threatening condition as it is associated with the risk of thrombosis (including cerebrovascular, myocardial and peripheral atrial thrombosis, deep vein thrombosis, transient ischemic attack, stroke, and pulmonary embolism), haemorrhage, and a long term propensity to develop myelofibrosis (MF) and secondary acute myeloid leukaemia (AML).

The median survival of untreated symptomatic patients with PV is 6 to 18 months, whereas survival of treated patients is 10 years or more. Accordingly, all patients with symptomatic PV should be treated in order to increase overall survival by reducing the risk for cardiovascular events and progression to myelofibrosis or acute leukemia.

3.1.2. Available therapies and unmet medical need

In the initial phase of the disease, phlebotomy is the cornerstone of treatment with the objective of maintaining haematocrit below 45%, a cut-off that has been shown to be associated with a lower risk of cardiovascular death and major thrombosis. As PV patients are at a high risk of thrombosis, phlebotomy is often accompanied by low dose aspirin. Cytoreductive therapy is recommended in patients with persistent haematological abnormalities, clinical symptoms, poor compliance with or intolerance of phlebotomy, and those at a high risk of thrombosis ("high-risk PV-patients"). Most PV patients require cytoreductive therapy during the course of their disease.

Hydroxyurea, although not approved in all EU countries for the use in the treatment of polycythaemia vera, is often the first-line cytoreductive therapy used in patients with polycythaemia vera. However, hydroxyurea-related toxicities often require either drug reduction or drug discontinuation resulting in inadequate management of the disease. Furthermore, due to its mechanism of action HU has the potential to be mutagenic and it is controversially discussed whether HU is associated with an increased risk of leukemic transformation after long-term use particularly in younger PV patients.

IFN-α, known for potential efficacy in MPNs and PV particularly since end of 1980s, is still considered an experimental therapy (off label) in Europe due to pending approval in this indication. However, there is sufficient evidence from clinical trials that IFN- α is able to induce major haematological response and occasionally even complete molecular remissions in patients with PV accompanied by a reduction in the risk of thrombosis and bleeding - the major determinants of morbidity in this indication. Moreover, alfa interferons are nowadays even recommended as first line therapy in high-risk PV patients of all ages in the relevant ELN guideline. Previously, it was mainly recommended in younger or HU resistant/intolerant-patients with high-risk PV.

Since 2015 the JAK2-inhibitor ruxolitinib (Jakavi) has been approved in the European Union "for the treatment of adult patients with polycythaemia vera who are resistant to or intolerant of hydroxyurea".

3.1.3. Main clinical studies

This application is based on a single pivotal phase 3 trial (PROUD-PV) and the supportive follow-up trial, CONTINUATION-PV. The PROUD-PV trial was an open-label, randomized (1:1), controlled, parallel-group, non-inferiority study comparing the efficacy and safety of ropeginterferon over hydroxyurea over 12 months in 257 randomised PV patients. While HU was given orally in doses from 500 mg to 3000 mg, ropeginterferon alfa-2b (Besremi) was administered subcutaneously s.c., 50 µg to 500 µg every two weeks, both drugs were given depending on response and tolerability. No relevant imbalances at baseline were observed between treatment arms in the included population of patients with early PV. CONTINUATION-PV trial was an open-label extension study to PROUD-PV designed to provide long-term evaluation of safety as well as efficacy of ropeginterferon alfa-2b in patients with PV who were previously treated in the PROUD-PV Study.

3.2. Favourable effects

Non-inferiority of ropeginterferon alfa-2b was shown for the post-hoc defined NI analysis for endpoint "complete haematological response" and the post-hoc defined NI margin of -20% only. 43.1% (43/122) versus 45.6%

(57/123) of the patients in the ropeginterferon-alfa-2b versus HU arm, respectively, reached this endpoint after 12 months of treatment. However, considering the absolute benefit of a treatment with ropeginterferon alfa-2b based on a comparison to baseline ropeginterferon alfa-2b showed a clear cytoreductive effect. An efficacy comparison of the differences between the four AOP efficacy studies (PEGINVERA; PROUD, CONTINUATION and PEN-PV) shows that the response rate for the haematological parameters Hct, PLT and leukocytes (complete haematological responders) ranged between 43% and 75% in the clinical studies due to different exposure times. Response may further increase and can be maintained in up to 70.5 % as demonstrated from the interim results after 36 months of treatment (i.e. after 24 months treatment duration from the CONTINUATION-PV trial).

Clinical response rates (defined as CHR and improvement of clinical symptoms/signs) consistently increased within the first 24 months of treatment with ropeginterferon alfa-2b (46.3% at Month 12, 49.5% at Month 24) and remained stable in the third year of treatment (52.6% at Month 36).

The accepted PV biomarker JAK2 allelic burden decreased during treatment in both treatment cohorts. At EOT in PROUD-PV Study (month 12) the difference was not statistically significant ($p=0.0736$) between the treatment cohorts with 30.7% and 25.9% for ropeginterferon alfa-2b or HU treated patients, respectively; after 36 months of treatment the difference was statistically significant ($p<0.0001$), with 16.6% and 42.5% in the ropeginterferon alfa-2b and BAT arm, respectively. In detail, the JAK2 allelic burden decreased during treatment in both treatment cohorts (baseline 42.8% and 42.9% in the two treatment arms). The median absolute levels of JAK-2 allelic burden ranged in the AOP204 treatment arm from 9.5% (Month 36) to 23.7% (Month 12). The median absolute levels of JAK-2 allelic burden ranged in the BAT treatment arm from 18.2% (Month 12) to 42.5% (Month 30). The differences between the treatment cohorts were in favour for ropeginterferon alfa-2b versus HU treated patients, respectively.

3.3. Uncertainties and limitations about favourable effects

A very conservative dose escalation strategy in the ropeginterferon alfa-2b arm was associated with a 20 weeks difference in time needed to reach the maximum plateau dose between the arms in favour for the comparator HU. Therefore, it seems that during the 12 months trial duration patients in the ropeginterferon alfa-2b arm were treated with a lower dose intensity which likely had a significant impact on the trial outcome. It appears that the duration of PROUD-PV was too short and assessment of efficacy was planned too early to be able to show the intended superiority or at least clear non-inferiority of ropeginterferon-alfa2b.

However, the prolonged time activity due to the very slow dose titration of ropeginterferon alfa-2b is considered acceptable as in non-responders phlebotomy is used routinely as rescue in order to normalise blood hyperviscosity. Nethertheless, physicians and patients should be adequately informed about the prolonged time until full efficacy.

Information regarding outcome and consistency of favourable effects in relevant subgroups, as defined by age, sex, ethnicity, organ function, disease severity, or genetic polymorphism remains of limited value due to the small orphan disease population investigated.

The clinical relevance of the more pronounced reduction in JAK2 burden in the ropeginterferon alfa-2b arm is not fully understood.

3.4. Unfavourable effects

Frequencies of overall TEAEs as well as ADRs during the PROUD-PV trial were slightly lower in the ropeginterferon alfa-2b arm (TEAE: ropeginterferon alfa-2b: 81.9% vs HU: 87.4% /ADR: ropeginterferon alfa-2b: 59.8% vs HU: 75.6%) and also the differences with respect to the intensity of TEAE as reflected by the grading differences may indicate a slight superiority for ropeginterferon alfa-2b. SAEs in PROUD-PV were reported slightly more frequent in the ropeginterferon alfa-2b arm (ropeginterferon alfa-2b: 11.0% vs HU: 8.7%). During longer treatment, as reflected by the CONTINUATION-PV results, rates may become lower and more similar (ropeginterferon alfa-2b: 3.2% vs BAT/HU: 2.1%). The only death deemed drug-related was observed in the HU arm from acute leukaemia.

The most frequent adverse events noted for ropeginterferon alfa-2b in the target population are thrombocytopenia (16.3%), leukopenia (15.7%) arthralgia (12.4%), fatigue (11.8%), influenza like illness (10.1%) and myalgia (10.1%). The risk for the haematological adverse events described above was higher in the comparator arm [thrombocytopenia 26.8%, leukopenia 21.3%), while Flu-like symptoms including arthralgia and myalgia were more frequently observed in ropeginterferon alfa-2b.

The risk for increased psychiatric disorder in general for the AESI “depression” was higher in the ropeginterferon alfa-2b arm [ropeginterferon alfa-2b: 3.2% (3/127) vs HU: 0.8% (1/127)]. An increased hepatotoxicity in terms of liver enzyme elevations (GGT, AST, ALT) grade 1 or 2 were observed, and two cases of ALT increased grade 3 were reported. The frequency in the ropeginterferon alfa-2b arm was between 2 to 10 % (including CONTINUATION-PV results) and an increase during longer exposure may be discussed. Hepatotoxicity (including cases of DILI) is a well-known class safety effect reported for all interferons. Besremi is contraindicated in patients with severe hepatic impairment due to available knowledge on the interferons.

Antibodies against antinuclear antigens (ANAs), erythrocyte and thyroid antigens have been identified as clinically relevant. In the PROUD-PV trial, two clinically significant abnormal values of ANA were observed in the ropeginterferon alfa-2b arm at two visits (week 22 and month 9). Starting with study week 12 inclusively (end of titration phase), one clinically significant abnormality was reported in TgAb in the ropeginterferon alfa-2b arm at each study visit excluding follow-up. One (week 20 and month 12) to two (screening and FU) clinically significant abnormalities were recorded for TPOAb in the HU arm.

3.5. Uncertainties and limitations about unfavourable effects

Two cases of stroke were observed in the ropeginterferon alfa-2b arm during the (individual) titration phase compared to none in the HU arm. This raises concerns that both events occurred during the titration phase and thus, may probably indicate a higher risk from the longer dose titration phase for ropeginterferon-alfa-2b. The higher risks in the ropeginterferon –alfa 2b arm during the longer dose-titration due to thromboembolic/major cardiovascular events can be sufficiently controlled by phlebotomies (see RMP).

3.6. Effects Table

Table BR 1: Effects Table for Besremi in the treatment of PV without splenomegaly

Effect	Short Description	Unit	Treatment	Control	Uncertainties/ Strength of evidence
Non-inferiority of ropeginterferon alfa-2b against comparator was shown	CHR after 12 months of treatment (without spleen normalisation)	% (n/N)	43.1% (43/122)	HU: 45.6% (57/123)	p-value is reported with 0.0028 /NI:-20%; post-hoc defined; 95% CI:-15.55 to 9.52
Long-term efficacy increased	CHR after 24month CHR after 36 months Rate reported in literature		70.5% (67/95) 70.5% (67/95) 70-80%	HU: 49.3% (33/76) HU: 51.4% (38/76) HU: 80.0%	Data from follow-up trial are subject to selection bias but still reliable
Longer disease response	response at any visit during the study was longer and superior in the ropeginterferon alfa-2b arm	Median (days)	266 days	167 days	Strong evidence (prespecified as sec. EP)
Longer treatment intervals possible	Lower frequency for injections needed	Weeks	ropeginterferon alfa-2b: Once every two weeks	Other peg-IFN-α: Once weekly	Undertreatment is not excluded / Probably strong
External validity	Interferons are subject of well-established use in PV since decades (off-label)	N/A	Complete haematological response (CHR) reported in literature for IFNs: 70-80%	CHR reported in literature for HU: ~80 %	Low/strong
Absence of risk of leukemia	All other cytoreductive drugs are genotoxic		No AML or other leukemia	HU: 2 AML-ADRs	No non-clinical or clinical signal is known for interferon, while HU is genotoxic/carcinogen
Comparable general safety (excluding AML risk)		TEAEs, ADRs, SAEs, drug related deaths	81.9% 59.8% 11.0% 0	HU: 87.4% 75.6% 8.7% 1	Dose intensity in HU was higher, this may lower the TEAEs in ropeginterferon alfa-2b arm, but overall valid, but less discriminative

Effect	Short Description	Unit	Treatment	Control	Uncertainties / Strength of evidence
Increased liver enzymes	Mostly grade 1 to 2 GGT, AST, ALT elevation, but 2 cases grade 3 ALT increase	%	2 – 10%; increasing during longer treatment	N/A	causality currently uncertain
Haematotoxicity	Thrombocytopenia, Leukopenia.	%	16.3% 15.7%	26.8% 21.3%	
Flu-like illness	Arthralgia Myalgia Fatigue	%	12.4% 10.1% 11.8%		
Psychiatric disorders increases	An increased rate for depression (and other psychiatric disorders) is a class effect for INFs.	% (n/N)	3.2% (3/127)	0.8% (1/127)	Risk is small and may indicate the Contraindications/ Exclusion criteria are sufficient
Immunological adverse events	Antithyreoglobulin ABs, ANAs, neutralizing ABs	n	2-6	1	Rates in literature are reported to be higher, absence of neutralizing ABs due to missing method validation ?
Disease-related CV and Thrombo-embolic events	Atrial fibrillation, cardiac failure, PAOD	% (n/N)	8.7% (11/127)	5.5% (7/127)	Risk may be comparable, but 2 thromboembolic events were observed during the dose titration ropeginterferon alfa-2b arm

Abbreviations: CHR= complete haematological response; defined as Hct <45% without phlebotomy (at least 3 months since last phlebotomy), platelets <400 x 109/L, leukocytes <10 x 109/L

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

Besremi showed a clear cytoreductive effect in terms of CHR as absolute benefit based on a comparison to baseline. Significant increase of efficacy after more adequate exposure (12 months with plateau dose) was reported from CONTINUNATION-PV trial. The haematological response rates and clinical response rates increased with treatment duration and remain stable beyond 24 months in line with results reported for other IFNs. It is anticipated that response may further increase and can be maintained in up to 70.5 % as demonstrated from the interim results after 36 months of treatment (i.e. after 24 months treatment duration from the CONTINUATION-PV trial). Thus, a clinically relevant efficacy of Besremi in the applied target population is demonstrated.

The toxicity profile observed with Besremi was considered acceptable. The general safety outcome in terms of TEAEs, ADRs, drug related SAEs, drug related deaths also is favourable to Besremi. The risk of cardiovascular and thromboembolic events during the initial treatment period, i.e. first 6 months, needs to be seen in the context of the treatment needs of the patient. Patients in the ropeginterferon alfa-2b arm were not treated with the maximum plateau dose until month 6. A potential higher risk for thromboembolic complications during the first 6 months cannot be excluded but adequate risk minimisation measures are in place (see RMP). In contrast to all other cytoreductive therapies used in PV, interferons are not genotoxic or carcinogenic.

The absence of genotoxicity, carcinogenicity and leukogenic transformation potential is a benefit very relevant to patients considering the long duration of treatment needed, particularly in younger patients more concerned about life-long exposure and especially if in a reproductive age.

3.7.2. Balance of benefits and risks

The important favourable effects identified and discussed above in principle outweigh the unfavourable effects. A clinically meaningful effect in terms of response rate has been established and even if formally one cannot exclude a 20% worse response rate compared to HU at 12 months, the loss of efficacy is not considered critical as phlebotomy can be used in the short term to compensate any lack of effect. The applicant has proposed adequate risk minimisation measures with respect to the impact of the slow dose titration needed and the thromboembolic complications in PROUD-PV.

3.8. Conclusions

The CHMP concluded that the overall B/R of Besremi in the treatment of polycythaemia vera without symptomatic splenomegaly is positive.

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 Besremi is favourable in the following indication:

Besremi is indicated as monotherapy in adults for the treatment of polycythaemia vera without symptomatic splenomegaly.

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.

The marketing authorisation holder shall submit the first periodic safety update report for this product within 6 months following authorisation.

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

Risk Management Plan (RMP)

The 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.

New Active Substance Status

Based on the review of the available data, the CHMP considers that ropeginterferon alfa-2b is a new active substance as it differs significantly in properties with regard to safety and efficacy from interferon alpha-2b contained in medicinal product(s) previously authorised within the European Union due to differences in molecular structure, nature of the source material or manufacturing process.