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Original Article

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Effective Management of Polycythemia Vera With Ropeginterferon Alfa-2b Treatment

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Abstract

Background: Polycythemia vera (PV) is a myeloproliferative neoplasm. Ropeginterferon alfa-2b is a new-generation polyethylene glycol-conjugated proline-interferon. It is approved for the treatment of PV at a starting dose of 100 µg (50 µg for patients receiving hydroxyurea (HU)) and dose titrations up to 500 µg by 50 µg increments. The study was aimed at assessing its efficacy and safety at a higher starting dose and simpler intra-patient dose escalation.

Methods: Forty-nine patients with PV having HU intolerance from major hospitals in China were treated biweekly with an initial dose of 250 µg, followed by 350 µg and 500 µg thereafter if tolerated. Complete hematological response (CHR) was assessed every 12 weeks based on the European LeukemiaNet criteria. The primary endpoint was the CHR rate at week 24. The secondary endpoints included CHR rates at weeks 12, 36 and 52, changes of *JAK2*^{V617F} allelic burden, time to first CHR, and safety assessments.

Results: The CHR rates were 61.2%, 69.4% and 71.4% at weeks 24, 36, and 52, respectively. Mean allele burden of the driver mutation *JAK2*^{V617F} declined from 58.5% at baseline to 30.1% at 52 weeks. Both CHR and *JAK2*^{V617F} allele burden reduction showed consistent increases over the 52 weeks of the treatment. Twenty-nine patients (63.0%) achieved partial molecular response (PMR) and two achieved complete molecular response (CMR). The time to CHR was rapid and median time was 5.6 months according to central lab results. The CHRs were durable and median CHR duration time was not reached at week 52. Mean spleen index reduced from 55.6 cm^2 at baseline to 50.2 cm^2 at week 52. Adverse events (AEs) were mostly mild or moderate. Most common AEs were reversible alanine aminotransferase and aspartate aminotransferase increases, which were not associated with significant elevations in bilirubin levels or jaundice. There were no grade 4 or 5 AEs. Grade 3 AEs were reversible and manageable. Only one AE led to discontinuation. No incidence of thromboembolic events was observed.

Conclusion: The 250-350-500 µg dosing regimen was well tolerated and effectively induced CHR and MR and managed spleen size increase. Our findings demonstrate that ropeginterferon alfa-2b at this dosing regimen can provide an effective management of PV and support using this dosing regimen as a treatment option.

Keywords: Ropeginterferon alfa-2b; Polycythemia vera; Complete hematological response; Molecular response; *JAK2*^{V617F} allele burden; Hydroxyurea resistance or intolerance

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Introduction

Polycythemia vera (PV) is a burdensome, myeloproliferative neoplasm (MPN). Most patients suffering from PV carry the driver Janus kinase 2 gene (*JAK*) mutation *JAK2*^{V617F}. PV is associated with an over-production of blood cells, increased incidence of thromboembolic and hemorrhagic complications, and long-term risk of transformation to myelofibrosis (MF) or acute myeloid leukemia (AML) [1-3]. Patients suffering from PV have lower rates of survival and the incidence rate of thrombotic, fibrotic, or leukemic events, which are the main causes of morbidity and mortality, at 20 years is 26%, 16%, and 4%, respectively [4, 5]. Age and history of thrombotic events (TEs) were the factors included in the conventional risk model to determine the risk of thrombosis. In addition, elevated hematocrit (HCT) levels have been demonstrated to have a close relationship with TEs. Several recent studies have also shown the relevance of increased TE risk to white blood cell (WBC) and platelet (PLT) counts, and *JAK2* variant allelic frequency (VAF) [6, 7]. Therefore, the clinical management of PV includes maintaining HCT levels < 45%, achieving complete hematological response (CHR), and reducing the driver *JAK2* mutation VAF to reduce the risk of TEs, and mitigating disease progression to MF and AML.

Ropeginterferon alfa-2b (BESREMi®) is a novel, site-selective polyethylene glycol-conjugated (PEGylated) recombinant proline-interferon-alpha with a favorable pharmacokinetic (PK) profile [8-11]. Its clinical efficacy and safety in the treatment of PV, regardless of race or ethnic background, has been demonstrated in several clinical studies [12-18]. It has been approved for the treatment of adult PV patients in the USA, Europe, and other countries or regions [19, 20]. It is the only interferon (IFN)-based therapy that has been approved by the US Food and Drug Administration (FDA) for treating PV and can be used regardless of the risk category or previous treatment history [19]. Ropeginterferon alfa-2b is used biweekly with an initiating dose of 100 µg (or 50 µg for patients receiving hydroxyurea (HU)), which is followed by 50 µg increments up to the maximum dose of 500 µg. The slow-titration schema with a low starting dose followed by 50 µg intra-patient dose increments could take up to approximately 6 months or more to attain the plateau dose level. During this period of slow dose titration, the hematological parameters and *JAK2*^{V617F} might not be optimally controlled, and patients might potentially face a thrombotic or hemorrhagic risk resulting from inadequate drug exposure [21]. We designed a phase II study to primarily evaluate whether initiating ropeginterferon alfa-2b at a higher dose with a simpler and faster intra-patient dose titration, i.e., the 250-350-500 µg dosing regimen, could result in an efficient CHR rate and molecular response (MR) in PV patients at 6 months based on the existing safety data in other indications for ropeginterferon alfa-2b, including viral hepatitis [21-25]. The results demonstrated that a CHR rate of 61.7% was achieved by this dose regimen in 6 months, which was notably higher than previously reported with the slow-titration schema [26]. However, the longer-term efficacy including CHR and MR, safety, and patient discontinuation rate needed to be investigated. We hereby present the 1-year data for ropeginterferon alfa-2b treatment at the 250-350-500 µg dosing regimen, which indicates the feasibility and effectiveness of this regimen as a treatment option for patients with PV.

Materials and Methods

The phase II, multicenter, single-arm study was conducted at 15 major hospitals in China. Jin et al have previously described the details of the methods and clinical trial information [25]. The phase II study planned to enroll 49 patients with PV who were resistant or intolerant to HU treatment according to the European LeukemiaNet (ELN) criteria [27]. The patients were subcutaneously administered with ropeginterferon alfa-2b once every two weeks. The treatment protocol included a starting dose of 250 µg at week 0, followed by an intra-patient dose escalation to 350 µg at week 2, and then a dose of 500 µg at week 4 thereafter. Dose could be adjusted according to tolerability. The maintenance dose was 500 µg for up to 52 weeks if tolerated. The primary endpoint was CHR rate at week 24. CHR is defined as: HCT < 45% without phlebotomy or erythrocyte apheresis in the preceding 3 months, PLT count $\leq 400 \times 10^9$/L, and leukocyte count $< 10 \times 10^9$/L. The secondary endpoints included CHR rates at weeks 12, 36 and 52; changes of *JAK2*^{V617F} allelic burden from baseline; time to first CHR; and safety assessments including the incidence, causality and severity of adverse events (AEs) assessed according to the Common Terminology Criteria for Adverse Events 5.0 (CTCAE 5.0); the incidence, causality, and severity of AEs of special interest; and the events leading to dose reduction or permanent termination of treatment. Each patient was planned to be in the study for approximately 14 months, encompassing a 28-day screening phase, a 52-week treatment phase, and a 28-day safety follow-up period. Quantitative determination of *JAK2*^{V617F} allele burden and hematological parameters for CHR assessment were carried out by a central laboratory. The 95% confidence interval (CI) of the CHR rate was estimated using the Clopper-Pearson method. The median and 95% CI of the time-event endpoints were estimated using the Kaplan-Meier method. The trial is registered at ClinicalTrials.gov (identifier: NCT05485948) and in China (China National Medical Products Administration registration number: CTR20211664). The institutional review board or ethics committee of all participating clinical sites have approved this study. Written informed consent was obtained from all participating patients. This study was conducted in compliance with the ethical standards of the responsible institution on human subjects as well as with the Helsinki Declaration.

Results

Patient demographics, baseline characteristics and disposition

Forty-nine patients with PV were enrolled in this study. The mean age at enrollment was 53.0 years. All patients had previ-

Table 1. Patient Demographics and Baseline Characteristics

Age (years), mean (SD)	53.0 (10.9)
Sex	
Male	31 (63.3%)
Female	18 (36.7%)
ECOG performance status score	
0	42 (85.7%)
1	7 (14.3%)
PV diagnosis (months), mean (SD)	44.43 (59.0)
History of HU treatment	
Intolerance	49 (100%)
Resistance	0
Total duration of HU treatment (days), median (min. - max.)	125.0 (1 - 7,344)
History of prior IFN treatment, N (%)	30 (61.2%)
Total time of prior IFN treatment (days), median (min. - max.)	153.5 (1 - 6,552)
History of phlebotomy or erythrocyte apheresis, N (%)	23 (46.9%)
History of hemorrhage or thrombosis	
Previous hemorrhage	6 (12.2%)
Previous thrombosis	9 (18.4%)
Spleen size (cm^2)[a], mean (SD)	55.6 (18.8)
Patients with splenomegaly determined by ultrasound[a], N (%)	36 (73.5%)
JAK2^{V617F} mutation	49 (100%)
Baseline parameters, mean (SD)	
Hematocrit (%)	46.0 (5.3)[b]
Leukocytes (10^9/L)	11.4 (9.4)
Platelets (10^9/L)	478.5 (238.8)
JAK2^{V617F} allelic burden (%)	58.5 (25.3)

[a]Spleen size = length × thickness, which was measured by ultrasound. Splenomegaly was recorded in ultrasound reports, being judged mainly based on spleen length. [b]Patients with hematocrit > 45% were generally treated with phlebotomy or erythrocyte apheresis. ECOG: Eastern Cooperative Oncology Group; HU: hydroxyurea; IFN: interferon; PV: polycythemia vera; SD: standard deviation.

ously received and had become intolerant to HU treatment according to the ELN criteria [27]. Of the patients, 61.2% had previously received an IFN-based therapy [26]. All patients needed cytoreductive therapy at baseline. Approximately 30.6% of the patients had previously suffered either a thrombosis or a hemorrhage. The driver mutation *JAK2*^{V617F} was present in all patients with a mean baseline allelic burden of 58.5% (Table 1).

Forty-nine patients were enrolled in the intent-to-treat (ITT) and safety populations, among which two patients withdrew from the study due to personal reasons, one before completing the 24-week treatment and one after completing the 24-week treatment. AEs led to discontinuation of one patient and 46 patients completed the 52-week treatment regimen according to the protocol.

CHR rates, durability, and time to CHR

The CHR rates, based on the central laboratory assessments, at 12, 24, 36, and 52 weeks of the treatment were 44.9% [26], 61.2% [26], 69.4%, and 71.4%, respectively. Previously, a CHR rate of 43% following 1 year of treatment with ropeginterferon alfa-2b at the slow-titration schema was reported in the PROUD-PV study [16]. The CHR rates in the ITT population at different assessment visits are depicted in Figure 1a.

The durable CHR in this study was observed in 17 (34.7%) and 20 patients (40.8%), from week 24 to 36, and week 36 to 52, respectively. Durable CHR is defined as CHR lasting for at least 12 weeks according to the central lab measurements. Furthermore, patients need to have the CHR for all the local lab results available within the 12 weeks. The rates of the durable CHR increased over time and were numerically higher than previously observed in the PV studies with ropeginterferon alfa-2b at the slow-titration schema. For example, the rate of durable CHR from week 36 to 52 was observed to be 27.6% in a phase II study with ropeginterferon alfa-2b at the slow-titration schema [18].

Mean HCT, PLT, and WBC levels decreased over time

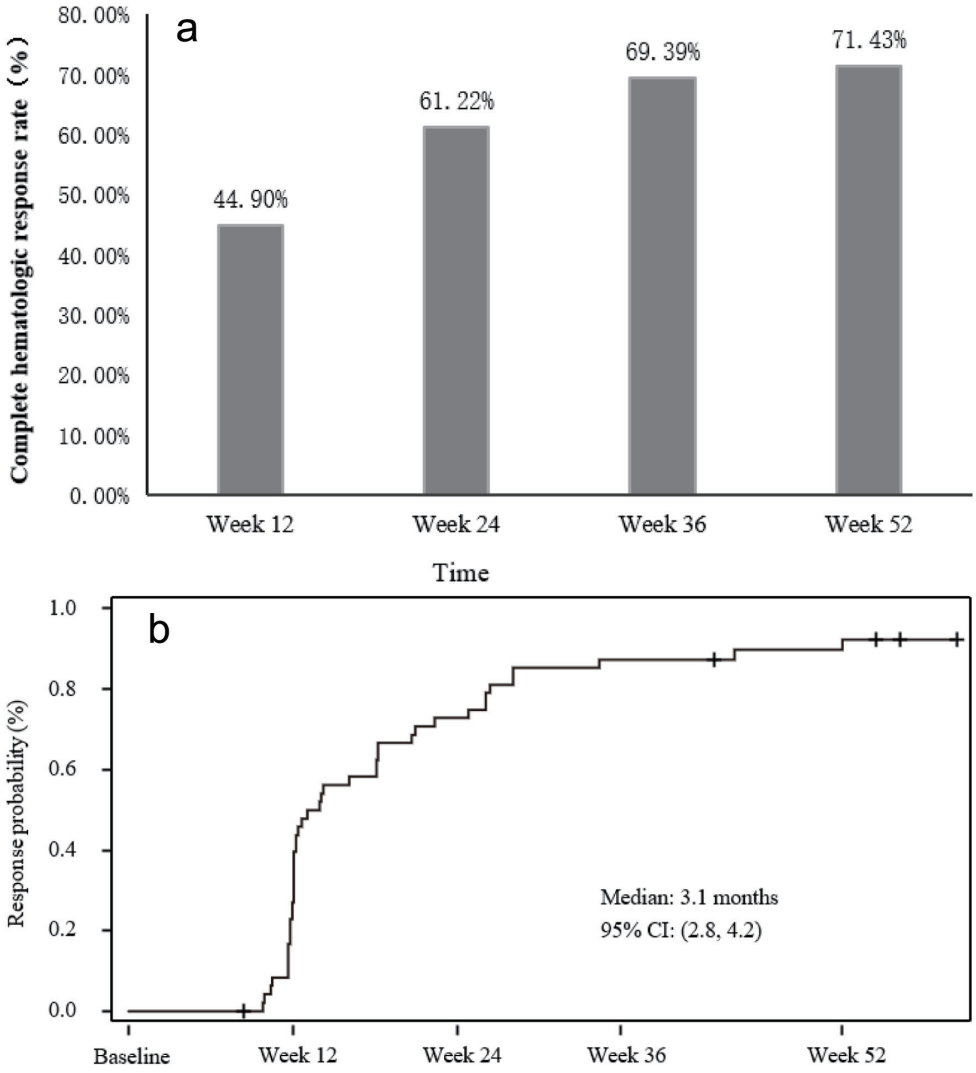

Figure 1. Complete hematological response (CHR) rates and time to CHR. (a) CHR rates at different assessment visits. (b) Graph depicting time to CHR.

from baseline to week 52 in this study: -5.3%, -282.8 × 10^9/L, and -6.5 × 10^9/L, respectively. After 52 weeks of treatment with ropeginterferon alfa-2b, all mean levels of the three peripheral hematological parameters improved to the normal ranges.

The overall CHR of the study was 83.7% with 41 patients achieving a CHR during 52 weeks of the treatment, according to central lab results, which were measured on the assessment visits (once every 12 weeks). The median time to achievement of the first CHR was 5.6 months (95% CI: 2.9 - 5.7) according to the central lab assessments. Based on the local lab measurements which were more frequent, the median time to CHR was 3.1 months (95% CI: 2.8 - 4.2) (Fig. 1b). Previously, the time to CHR with ropeginterferon alfa-2b at the slow-titration schema was observed to be approximately 12 months [18].

Overall CHR duration was further assessed as the number of days between the initiation of CHR and the criteria for CHR being no longer met according to local lab measurements. Forty-four patients (89.8%) achieved at least one CHR during the 52 weeks of the treatment according to local lab assessments. CHR was maintained in 29 patients (59.2%) from the first CHR until the end of the 52-week treatment. As demonstrated in Figure 2, the CHR was very durable, which is evident from the fact that median duration of CHR for patients with at least one response at any visit during the study was not reached at week 52. These results indicate that ropeginterferon alfa-2b at the 250-350-500 µg dosing regimen, which has a higher initiating dose and simpler dose titration, can lead to high levels of CHR, which also occur rapidly and are very durable.

Spleen index

Spleen index was defined as the maximum length on the longitudinal sonogram (cm) times thickness on the transverse sonogram (cm) by ultrasound. The change of spleen index

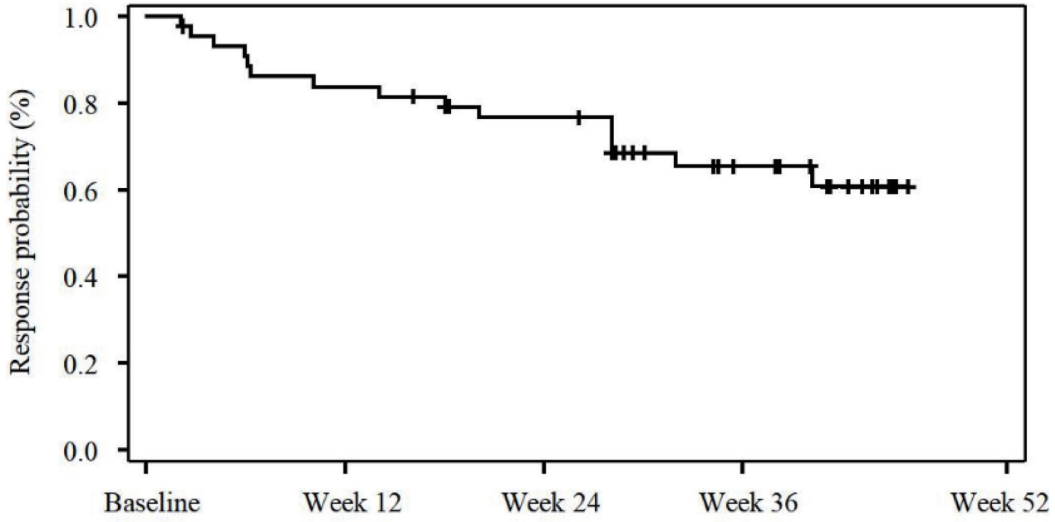

Figure 2. Kaplan-Meier plot demonstrating the complete hematological response (CHR) duration.

(cm^2) was assessed at week 52 and compared with the baseline measurement. Forty-two patients with measurable baseline values were evaluated for spleen response. After the 52-week ropeginterferon alfa-2b treatment, the mean spleen index reduced to 50.2 cm^2 with a mean spleen size reduction of 5.6 cm^2, as shown in Figure 3. This result indicates that ropeginterferon alfa-2b at the 250-350-500 µg dosing regimen can control the spleen size increase in patients with PV.

JAK2^{V617F} MR

After the 52-week treatment with ropeginterferon alfa-2b, the mean *JAK2*^{V617F} allele burden decreased to 30.1% from 58.5%

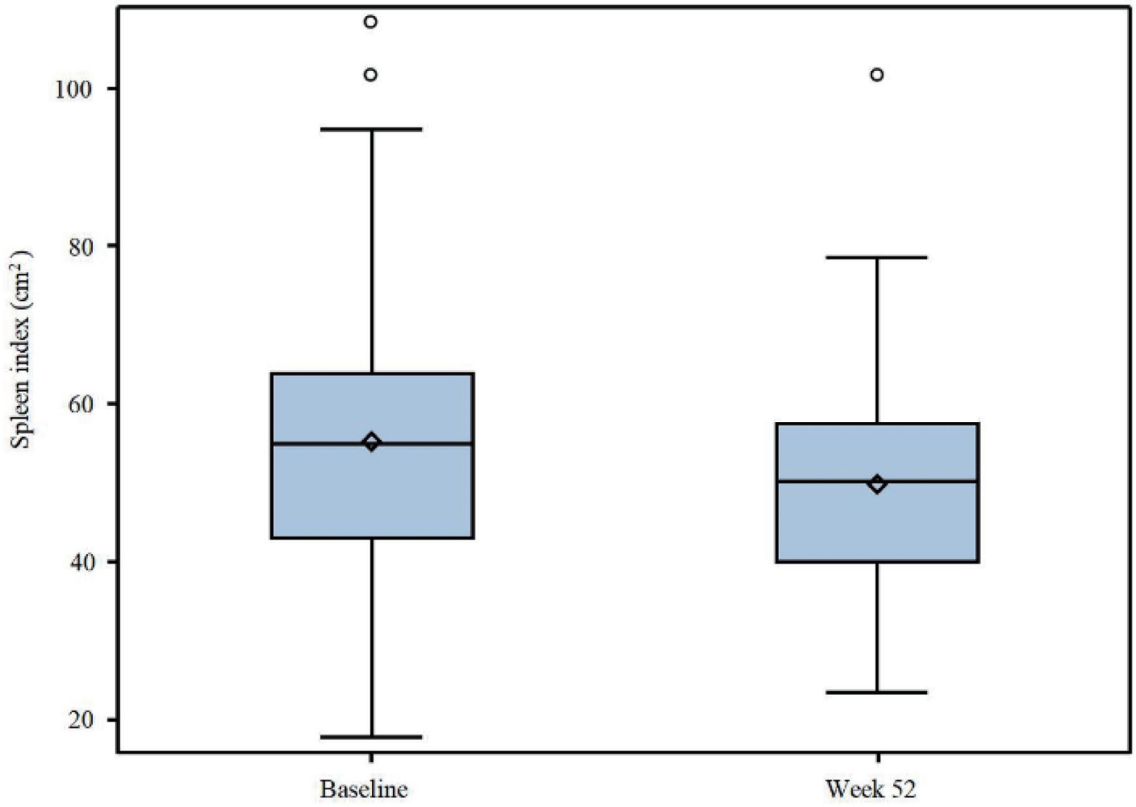

Figure 3. Graph demonstrating the mean spleen index change. The diamond indicates the mean value.

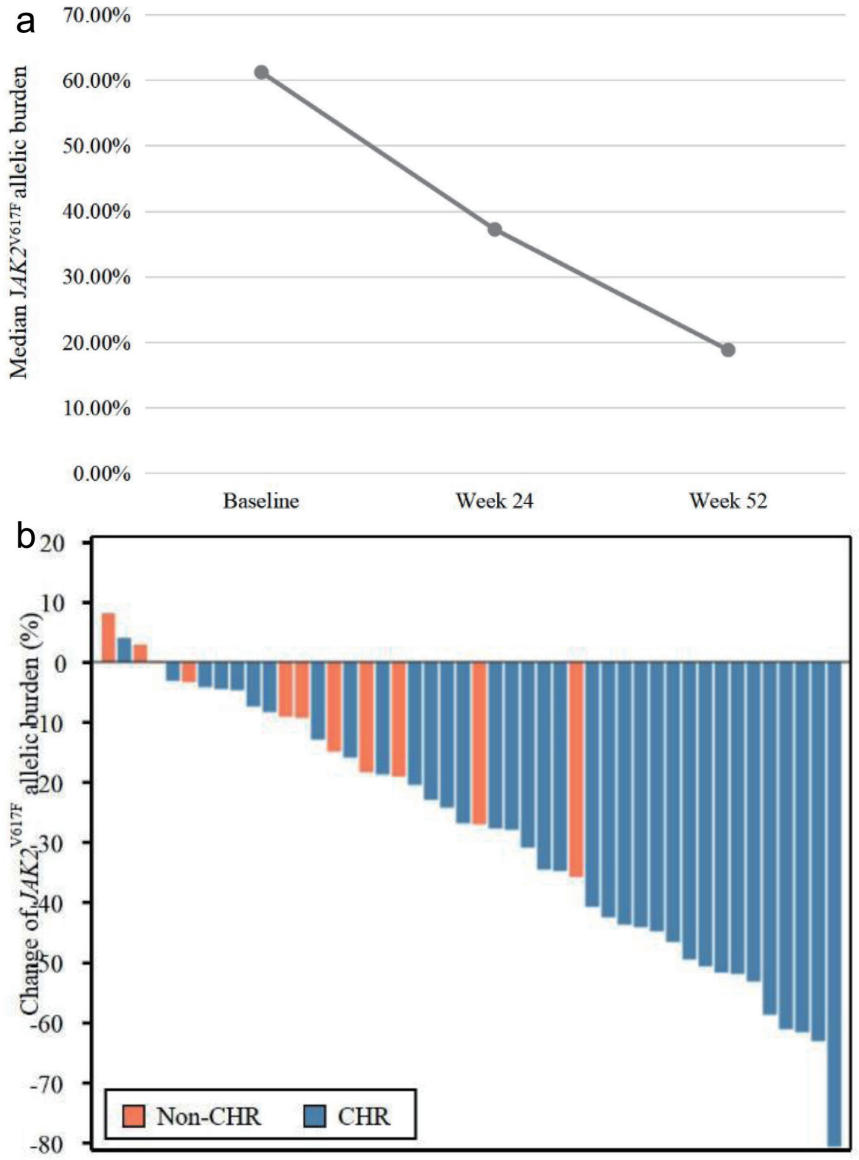

Figure 4. The effect of ropeginterferon alfa-2b on *JAK2*^{V617F} allelic burden. (a) Graph demonstrating the change of median *JAK2*^{V617F} allelic burden (%) during the 52 weeks of treatment. (b) Waterfall plot indicating the change of the *JAK2*^{V617F} allelic burden in individual patients after 52 weeks of treatment. Blue color represents the patients who had complete hematological response (CHR).

at the baseline. The median *JAK2*^{V617F} allelic burden decreased from 61.2% at baseline to 18.8% at week 52 (Fig. 4a). The waterfall plot (Fig. 4b) indicates a reduction in *JAK2*^{V617F} allelic burden in 42/46 patients (91.3%) during the 52 weeks of the treatment. Two patients demonstrated CMR at week 52 as measured by next-generation sequencing technology [28] with a lower limit of quantitation at 1% by the central laboratory. One patient had a baseline *JAK2*^{V617F} allelic burden of 59.2% and the other had 4.7%. The *JAK2*^{V617F} allelic burden was undetectable for both patients after 52 weeks of treatment. Based on the 2009 criteria [29], 29 patients (63.0%) demonstrated PMR, defined as a reduction ≥ 50% in patients with < 50% mutant allele burden, or a reduction ≥ 25% in patients with > 50% mutant allele burden. According to the 2013 ELN criteria [30], 18 patients (39.1%) achieved PMR, defined as ≥ 50% decrease in allele burden in patients with at least 20% mutant allele burden at baseline. The *JAK2*^{V617F} allelic burden of 13 patients (28.3%) decreased to less than 10% following 52 weeks of treatment.

Safety

All 49 patients were included in the safety analysis. All except one patient reached the dose of 500 µg over the 52 weeks of treatment. The median duration of exposure to ropeginterferon alfa-2b was 365 days. Including the first 4 weeks of intra-patient dose

escalations from 250 to 500 µg, the mean dose per patient over 52 weeks was 435.2 ± 59.2 µg once every 2 weeks. The median dose per patient was 462.8 µg once every 2 weeks over the 52 weeks.

Most treatment-emergent AEs were mild or moderate (Table 2). Elevated alanine aminotransferase (ALT) levels (28/49, 57.1%) and aspartate aminotransferase (AST) increase (28/49, 57.1%), WBC count decrease (23/49, 46.9%), hyperuricemia (22/49, 44.9%), gamma-glutamyl transferase (GGT) increase (20/49, 40.8%), neutrophil count decrease (18/49, 36.7%), and hypertriglyceridemia (18/49, 36.7%) were the most common AEs encountered. Most of the AEs were grade 1 or 2 and were reversible. No grades 4 or 5 AEs were observed in the study. Although ALT and AST elevations are common, only one patient had a grade 3 ALT elevation, and the patient did not have elevated bilirubin levels or jaundice. Possible drug-related grade 3 AEs included GGT increase (2/49, 4.1%), lymphocyte count decrease (2/49, 4.1%), neutrophil count decrease (2/49, 4.1%), WBC count decrease (1/49, 2.0%), ALT increase (1/49, 2.0%), pneumonia (1/49, 2.0%), arthralgia and peripheral edema (1/49, 2.0%), and hypertriglyceridemia (1/49, 2.0%). All grade 3 AEs except for one patient who had a grade 3 lymphocyte count decrease, recovered or were resolved. The grade 3 AE of lymphocyte count decrease improved to the grade 2 level. Serious AEs (SAEs) including pneumonia, ALT and AST increase, and myasthenia gravis that were possibly related were observed in three patients. Only one patient discontinued the study due to an AE (grade 2 myasthenia gravis). Five patients showed AEs of special interest (10.2%), including two cases of grade 1 and 2 conjunctival hyperemia (4.1%), respectively, one case of grade 2 diarrhea (2%), one grade 2 hyperthyroidism (2%), and one grade 2 myasthenia gravis (2%). No patient experienced a thromboembolic event and there was no occurrence of transformation to MF or AML.

PK analysis

PK parameters were estimated following administration of the starting dose of 250 µg and intra-patient dose escalation to 500 µg based on the non-compartmental model. Descriptive analyses of concentrations and PK parameters were performed. The summary of PK parameters is given in Table 3. The PK parameters of ropeginterferon alfa-2b following the single dose of 250 µg in this study were observed to be similar to those of a single dose of 270 µg in healthy volunteers [31]. The median time to the maximum serum concentration (T_{max}) in this study and health volunteers was 96.0 vs. 132.0 h. The area under the serum concentration-time curve from time 0 to 336 h ($AUC_{0\text{-}336\ h}$) was 4,082,160 vs. 4,838,047 h × pg/mL and maximum serum concentration (C_{max}) was 18,632 vs. 24,140 pg/mL. The C_{max} and AUC after intra-patient dose titrations (500 µg) in this study were higher than those after the initial dose of 250 µg (Table 3), which is consistent with the intra-patient dose escalation.

Discussion

The results in our study demonstrate that the new dosing regimen of 250-350-500 µg of ropeginterferon alfa-2b is well-tolerated and highly efficacious in patients suffering from PV. This dosing regimen was associated with a rapid achievement of CHR without the need for phlebotomy or erythrocyte apheresis. The CHRs were very durable and the levels of CHR were numerically greater than previously reported for the slow-titration dosing schema. This was also accompanied with a high level of *JAK2*^{V617F} allelic burden reduction. The results suggested that treatment of patients with PV at this dosing regimen of a higher starting dose and simpler intra-patient dose titration can eradicate the mutant cell clones carrying the driver mutation *JAK2*^{V617F} in PV in a more efficient manner. Both hematological parameters including HCT, WBC and PLT counts, as well as *JAK2*^{V617F} are potentially risk factors for patients to develop TEs [32-37]. *JAK2*^{V617F} can also contribute to the neoplastic progression to MF and AML [36-38]. In addition, ropeginterferon alfa-2b treatment appeared to control the spleen size increase with the mean spleen index decreasing from 55.6 cm^2 at baseline to 50.2 cm^2 following 52 weeks of the treatment in this study. Inhibition of disease progression and prolonged progression- or event-free survival in patients with PV could be attributed to PEGylated IFN-based therapies [39, 40]. Thus, the ropeginterferon alfa-2b treatment at the 250-350-500 µg dosing regimen could potentially serve as an effective treatment option for patients with PV.

It is conceivable that at this dosing regimen, ropeginterferon alfa-2b is effective in all PV patients despite the fact that this study enrolled HU-intolerant patients. The Myeloproliferative Disorders Research Consortium studies MPD-RC 111 and 112 with PEGylated IFN alfa-2a indicated that the rate of CHR in PV patients with HU-resistance or -intolerance was comparable to that observed in treatment-naive patients at 12 months (22% and 28%, respectively) [41, 42]. Ropeginterferon alfa-2b has been approved for adult PV patients regardless of prior treatment history or disease risk category. It has demonstrated clinical efficacy in HU-naive as well as -previously treated patients in the phase III PROUD-PV/CONTINUATION-PV studies [16]. Furthermore, ropeginterferon alfa-2b works differently from HU, since it is an IFN-based therapy. The efficacy of ropeginterferon alfa-2b could potentially be attributed to the selective inhibition of the driver mutation *JAK2*^{V617F}-carrying malignant hemopoietic stem or progenitor cells [43], while HU can be non-specific and induce DNA damage. In this study, ropeginterferon alfa-2b treatment at the 250-350-500 µg dosing schema showed a rapid time to achieve the CHR and the levels of CHR were high over the 52 weeks of treatment. Therefore, ropeginterferon alfa-2b at this dosing schema represents an effective treatment regimen for all PV patients, regardless of HU-resistance or intolerance.

In this study, a decrease in *JAK2*^{V617F} allelic burden was observed in almost all patients (91.3%) during the 52 weeks of treatment. The mean reduction from 58.5% at baseline to 30.1% at 52 weeks was observed. Previously, a reduction from 41.9% at baseline to 30.7% at 52 weeks was observed with ropeginterferon alfa-2b at the slow titration dosing schema in the PROUD-PV study. Moreover, CMR was observed in two patients and PMR in 29 patients (63.0%), contributing to an MR rate of 67.3% in this study. It also appeared that most patients with a greater reduction of the *JAK2*^{V617F} allelic burden experienced CHR. This is consistent with the

Table 2. Treatment-Emergent Adverse Events Occurring in ≥ 10% Patients

System organ class preferred term	Patients (N = 49)					
	Grade 1	Grade 2	Grade 3[a]	Grade 4	Grade 5	Total
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Metabolism and nutrition disorder						
Hyperuricemia	22 (44.9%)	0	0	0	0	22 (44.9%)
Hypertriglyceridemia	15 (30.6%)	0	3 (6.1%)	0	0	18 (36.7%)
Decreased appetite	5 (10.2%)	0	0	0	0	5 (10.2%)
Infections and infestations						
Urinary tract infection	6 (12.2%)	6 (12.2%)	0	0	0	12 (24.5%)
COVID-19	5 (10.2%)	1 (2.0%)	0	0	0	6 (12.2%)
Upper respiratory tract infection	3 (6.1%)	3 (6.1%)	0	0	0	6 (12.2%)
General disorders and administration site conditions						
Asthenia	7 (14.3%)	4 (8.2%)	0	0	0	11 (22.4%)
Pyrexia	5 (10.2%)	1 (2.0%)	0	0	0	6 (12.2%)
Skin and subcutaneous tissue disorders						
Alopecia	9 (18.4%)	0	0	0	0	9 (18.4%)
Renal and urinary disorders						
Proteinuria	8 (16.3%)	2 (4.1%)	0	0	0	10 (20.4%)
Musculoskeletal and connective tissue disorders						
Back pain	4 (8.2%)	1 (2.0%)	0	0	0	5 (10.2%)
Hepatobiliary disorders						
Hepatic steatosis	8 (16.3%)	0	0	0	0	8 (16.3%)
Nervous system disorders						
Hypoesthesia	6 (12.2%)	0	0	0	0	6 (12.2%)
Investigations						
Elevated alanine aminotransferase[b]	21 (42.9%)	6 (12.2%)	1 (2.0%)	0	0	28 (57.1%)
Elevated aspartate aminotransferase[c]	21 (42.9%)	7 (14.3%)	0	0	0	28 (57.1%)
Lowered white blood cell count[d]	8 (16.3%)	13 (26.5%)	2 (4.1%)	0	0	23 (46.9%)
Increased gamma-glutamyl transferase[e]	12 (24.5%)	6 (12.2%)	2 (4.1%)	0	0	20 (40.8%)
Decreased neutrophil count	8 (16.3%)	8 (16.3%)	2 (4.1%)	0	0	18 (36.7%)
Decreased lymphocyte count	2 (4.1%)	7 (14.3%)	3 (6.1%)	0	0	12 (24.5%)
Increased beta 2 microglobulin urine	12 (24.5%)	0	0	0	0	12 (24.5%)
Decreased platelet count	9 (18.4%)	1 (2.0%)	0	0	0	10 (20.4%)
Decreased weight	9 (18.4%)	3 (6.1%)	0	0	0	12 (24.5%)
Increased blood bilirubin[f]	5 (10.2%)	1 (2.0%)	0	0	0	6 (12.2%)
Increased blood alkaline phosphatase	4 (8.2%)	2 (4.1%)	0	0	0	6 (12.2%)
White blood cells urine positive	6 (12.2%)	1 (2.0%)	0	0	0	7 (14.3%)
Increased blood lactate dehydrogenase	7 (14.3%)	0	0	0	0	7 (14.3%)
Antinuclear antibody positive	8 (16.3%)	0	0	0	0	8 (16.3%)
Anemia	8 (16.3%)	3 (6.1%)	0	0	0	11 (22.4%)

[a]Fifteen grade 3 AEs were observed in 11 patients. Among them, possible treatment-related TEAEs were observed in eight patients. [b]Four patients (8.2%) had a prior history of grade 1 alanine aminotransferase increase. [c]One patient (2.0%) had a prior history of grade 1 aspartate aminotransferase increase. [d]Three patients received G-CSF treatment for decrease in the white blood count during the study. [e]Three patients (6.1%) had a prior history of grade 1 gamma-glutamyl transferase increase. [f]Two patients (4.1%) had a prior history of grade 1 bilirubin increase. COVID-19: coronavirus disease 2019.

Table 3. Summary of PK Parameters

PK parameters	Starting dose (250 µg)	After intra-patient dose titrations (500 µg)
C_{max} or $C_{max\ ss}$ (pg/mL), mean ± SD	18,632.0 ± 10,288.2	59,534.1 ± 22,498.3
T_{max} (h), median (min. - max.)	96 (48.0 - 168.0)	96 (48.0 - 210.6)
AUC_{0-t} or AUC_{0-tau} (h × pg/mL), mean ± SD	3,960,705.9 ± 2,416,333.4	15,293,612.2 ± 5,625,792.8
$T_{1/2}$ (h), mean ± SD	144.7 ± 100.0	-

T_{max}: median time to the maximum serum ropeginterferon alfa-2b concentration; $T_{1/2}$: terminal-phase half-life; C_{max}: maximum serum concentration; $C_{max\ ss}$: steady state of maximum serum concentration; AUC_{0-t}: area under the serum concentration-time curve from time 0 to the time of the last quantifiable sample within a dosing interval; AUC_{0-tau}: area under the serum concentration-time curve during the dosing interval after repeated dosing; PK: pharmacokinetic; SD: standard deviation.

notion that the potential disease-modifying effect exerted by ropeginterferon alfa-2b is at least in part associated with a decrease in the *JAK2*^{V617F} allelic burden. *JAK2*^{V617F} allelic burden is an important biomarker in PV. Patients with a higher *JAK2*^{V617} VAF had an increased risk of thrombotic complications. *JAK2*^{V617F} MR in patients with PV or essential thrombocythemia was associated with a lower risk of progression to MF [44, 45]. Therefore, further exploration of the relationship between the efficient reduction of the *JAK2*^{V617F} allelic burden by ropeginterferon alfa-2b and improved disease-modifying outcomes such as prolonged progression-free or overall survival for patients is required.

Ropeginterferon alfa-2b was well-tolerated and no new safety issues were identified in this study. Most of the AEs were mild or moderate. Grade 4 or 5 AEs were not observed in any patient. Grade 3 AEs were manageable and reversible. The most common AEs were ALT and AST elevations, with only one case of grade 3 ALT elevation, none of which were associated with the occurrence of jaundice or symptoms. One case of patient discontinuation due to AEs was observed. Previously, initial higher doses of PEGylated IFNs needed to be decreased stepwise based on tolerance [46]. However, our study has demonstrated that ropeginterferon treatment can be initiated with a starting dose of 250 µg, which is close to the highest, target dose of 500 µg. The dose of 500 µg can be achieved within 4 weeks of the first treatment. Moreover, depression which was associated with prior IFN-based therapies was not observed in this study. Furthermore, the PK exposures regarding C_{max} and AUC were found to increase with the dose. Both parameters exhibited a dose-dependent increase. The mean C_{max} at 500 µg in the study was 59.5 ng/mL and the mean AUC was 15,292 ± 5,625.8 h × ng/mL. The higher starting dose regimen appeared to lead to effective plasma exposures associated with good efficacy and tolerability. Thus, the results of this study regarding efficacy, MR, safety, and PK indicate a positive benefit-risk profile with this dosing regimen as a treatment option for patients with PV.

In summary, ropeginterferon alfa-2b at the 250-350-500 µg dosing regimen with a flexibility of dose adjustment according to tolerability appears to show good clinical tolerability and safety, and efficacy with meaningful PK exposures in patients with PV. The results indicate a positive benefit-risk balance in patients treated with ropeginterferon alfa-2b at this dosing regimen, suggesting that the treatment could lead to durable clinical benefits at a greater level with a reasonable safety profile. The participating patients in this study are expected to receive long-term continued treatment to assess progression-free and overall survivals.

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Conflict of Interest

SS Suo, RF Fu, ZH Shao, J Bai, SN Chen, MH Duan, H Zhou, N Xu, SJ Zhang, XL Zuo, X Du, L Wang, P Li, XH Zhang, ZJ Xiao, and J Jin have no conflict of interest to disclose. A Qin, O Zagrijtschuk, and T Sato are employees of PharmaEssentia Corporation. DX Wu, WH Shen, W Wang, JJ Zhang, and YN Li are employees of PharmaEssentia Biotech (Beijing) Ltd.

Informed Consent

Written informed consent was obtained from all participating patients.

Author Contributions

Each author listed contributed to the work and takes responsibility for the content. All authors participated in the writing and review process of the manuscript and approved it for publication. J Jin, ZJ Xiao, A Qin, DX Wu, YN Li, JJ Zhang, W Wang, and O Zagrijtschuk designed the study. DX Wu, A Qin, and J Jin wrote the initial draft of the manuscript. SS Suo, RF Fu, ZH Shao, J Bai, SN Chen, MH Duan, H Zhou, N Xu, SJ Zhang, XL Zuo, X Du, L Wang, P Li, XH Zhang, ZJ Xiao, and J Jin enrolled and treated patients on the trial.

Data Availability

The data supporting the findings of this study can be available from PharmaEssentia upon reasonable request after ropeginterferon alfa-2b has acquired marketing approvals in China.

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