Published in final edited form as:

Clin Lymphoma Myeloma Leuk. 2020 October ; 20(10): e712–e723. doi:10.1016/j.clml.2020.05.018.

Interferon therapy in myelofibrosis - a systematic review and meta-analysis

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Abstract

Background: Myelofibrosis is a Philadelphia chromosome-negative myeloproliferative neoplasm characterized by progressive bone marrow failure, increased risk of progression to AML, and constitutional symptoms. For over three decades various formulations of interferon (IFN) have been used for the treatment of myelofibrosis with variable results and the role of IFN in the treatment of myelofibrosis is evolving.

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Conflicts of Interest:

N.A.P. consulted for and received honoraria from Alexion, Pfizer, Agios Pharmaceuticals, Blueprint Medicines, Incyte, Novartis, Celgene, Bristol-Myers Squibb and CTI biopharma. N.A.P. received research funding (all to the institution) from Boehringer Ingelheim, Astellas Pharma, Daiichi Sankyo, Sunesis Pharmaceuticals, Jazz Pharmaceuticals, Pfizer, Astex Pharmaceuticals, CTI biopharma, Celgene, Genentech, AI Therapeutics, Samus Therapeutics, Arog Pharmaceuticals and Kartos Therapeutics. M.S.T. has received research funding from Abbvie, Cellerant, Orsenix, ADC Therapeutics, and Biosight. R.K.R. has received consulting fees from: Constellation, Incyte, Celgene, Promedior, CTI, Jazz Pharmaceuticals, Blueprint, Stemline, and research funding from Incyte, Constellation, Stemline. M.S.T. has received honoraria for M.S.T. has received research funding from Abbvie, Cellerant, Orsenix, ADC Therapeutics, and Biosight. M.S.T. has received honoraria for advisory board membership from Abbvie, BioLineRx, Daiichi-Sankyo, Orsenix, KAHN, Rigel, Nohla, Delta Fly Pharma, Tetraphase, Oncolyze, and Jazz Pharma. M.S.T. received patents and royalties from UpToDate. A.M.Z. received research funding (institutional) from Celgene, Acceleron, Abbvie, Novartis, Otsuka, Pfizer, Medimmune/AstraZeneca, Boehringer-Ingelheim, Trovogene, Incyte, Takeda, and ADC Therapeutics. A.M.Z. had a consultancy with and received honoraria from AbbVie, Otsuka, Pfizer, Celgene, Jazz, Ariad, Incyte, Agios, Boehringer-Ingelheim, Novartis, Acceleron, Astellas, Daiichi Sankyo, Cardinal Health, Seattle Genetics, BeyondSpring, Trovogene, Ionis, Epizyme, and Takeda. A.M.Z. received travel support for meetings from Pfizer, Novartis, and Trovogene. None of these relationships were related to the development of this manuscript. All other authors report no relevant disclosures/competing interests.

Methods: For this systematic review and meta-analysis MEDLINE and EMBASE via Ovid, Scopus, COCHRANE registry of clinical trials (CENTRAL), and Web of Science were searched from inception through 03/2019 for studies of pegylated IFN (peg-IFN) and non-pegylated IFN (non-peg-IFN) in myelofibrosis patients. The primary outcome of overall response rate (ORR) was defined as a composite of complete response, partial response, complete hematologic response and partial hematologic response. Random-effects models were used to pool ORR and meta-regression analyses were performed to compare peg-IFN and non-peg-IFN formulations.

Results: Among the 10 studies with 141 myelofibrosis patients included, the ORR was 49.9% (95% CI 30.4–69.3%) and there was no statistically significant difference ($p=0.99$) between peg-IFN (50.0%, 95% CI 26.2–73.9%; $I^2=76.9\%$) and non-peg-IFN (49.6%, 95% CI 20.5–79.0%; $I^2=56.7\%$). Treatment discontinuation due to adverse events was common with non-peg-IFN at 35.8% (95% CI 3.5–68.1%) per year and less in the one study on peg-IFN (0.5%/year).

Conclusion: IFN can lead to hematologic improvements in a subset of myelofibrosis patients but study quality is limited and heterogenous. Biomarkers predicting response to IFN and formulations with improved tolerability are warranted.

Microabstract:

Interferon (IFN)- α has been used for several decades for the treatment of myelofibrosis with conflicting results. In this systematic review and meta-analysis of 10 studies with 141 patients we found that IFN led to hematologic improvements in 49% of patients.

Keywords

myelofibrosis; MPN; interferon; meta-analysis; adverse events

Introduction:

Myelofibrosis (MF) belongs to the heterogenous group of Philadelphia chromosome-negative myeloproliferative neoplasms (MPN) and is characterized by bone marrow failure and risk of progression to acute myeloid leukemia (AML).(1, 2) Thanks to advances in molecular testing, driver mutations in the *JAK2*, *CALR*, or *MPL* genes have been identified in about 90% of patients with MF and have led to the development of JAK inhibitors for the treatment of MF.(3, 4) Although the recent approval of the JAK inhibitors ruxolitinib and fedratinib has significantly improved symptom management of MF patients, a substantial proportion of patients becomes refractory to ruxolitinib or is experiencing dose-/treatment-limiting side effects.(5, 6) Additionally, JAK inhibitors are primarily used in the treatment of symptomatic patients with high-risk MF, as defined by the Dynamic International Prognostic Scoring System (DIPSS) or alternative risk stratification tools, and the optimal treatment of patients with low-risk MF and for patients who are intolerant or refractory to JAK inhibitors continues to evolve.(7, 8)

On a molecular basis MF is characterized by the abnormal expression of various pro-inflammatory cytokines leading to the proliferation of aberrant immature megakaryocytes and granulocytes and reactive bone marrow fibrosis.(3, 9, 10) Interferon (IFN)- α has been used in various formulations and dosages for the management of MPNs for over 30 years

and is currently recommended as an alternative to ruxolitinib or hydroxyurea for the treatment of low-risk MF by the National Comprehensive Cancer Network (NCCN).(8) IFN has been shown to have multiple anti-proliferative, pro-apoptotic, and immunomodulatory effects on hematopoietic progenitor and immune cells in the bone marrow and data from patients with essential thrombocytosis (ET) and polycythemia vera (PV) have shown that treatment with IFN can lead to a reduction in the allele burden of driver mutations suggesting a disease-modifying effect.(11–15) However, the role of IFN in the evolving treatment landscape of MF is poorly defined and a more widespread use has been hampered by poor tolerability and the absence of an oral formulation.(15, 16) Therefore, we performed a systematic literature review and meta-analysis to objectively assess efficacy and safety of IFN for the treatment of MF.

Methods:

This systematic review and meta-analysis was conducted according to the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) and Meta-Analysis of Observational Studies in Epidemiology (MOOSE) guidelines.(17) MEDLINE and EMBASE via Ovid, COCHRANE registry of clinical trials (CENTRAL), Scopus and Web of Science electronic databases were searched without language restriction from inception through 3/21/2019, using combinations of free-text terms linked by Boolean operators: ["polycythemia" OR "polycythemia vera" OR "essential thrombocytosis" OR "myelofibrosis" OR "myeloproliferative neoplasm" OR "MPN"] AND ["interferon" OR "IFN" OR "pegylated interferon" OR "peginterferon" or "alpha2b interferon" OR "alpha2 interferon" OR "alpha interferon"]. The study selection process is illustrated in Figure 1. Results for PV and ET are reported separately.

Primary outcome was the ORR defined as a composite of complete response (CR), complete hematologic response (CHR), partial response (PR), and partial hematologic response (PHR) as defined by the individual publications. Safety endpoints were the rate of treatment discontinuation due to adverse events and rate of thromboembolic complications. A Downs and Black checklist was used for quality assessments of individual studies as described previously.(18, 19)

Random-effects models were used to pool ORR. All effect sizes underwent logarithmic transformation prior to pooling using an inverse variance weighting approach. Heterogeneity of studies was determined using Cochran Q and I^2 indices. Planned subgroup analyses and univariate meta regression analyses were performed to statistically compare effect sizes of different studies based on the type of administered interferon (non-pegylated IFN vs. pegylated IFN) and duration of follow up (<24 months vs. ≥ 24 months). All analyses were performed with Comprehensive Meta-Analysis (CMA version 2.2, Biostat).

Results:

Results of literature search:

We identified 6059 citations after duplicate removal. As outlined in Figure 1 studies were excluded based on title and abstract review if they reported results on diseases other than

MPN, or if they were review articles, commentaries without original data, basic research articles, and case series with less than 5 patients. 110 publications were reviewed in full and 10 studies were included in this meta-analysis after applying additional exclusion criteria as illustrated in Figure 1. One additional study was excluded as it used IFN- γ rather than IFN- α , which was used in all other included studies.(20)

Description of included studies:

Ten studies (6 retrospective cohort studies, 1 phase I clinical trial, and 3 phase II clinical trials) with a total of 141 patients with MF were included.(21–29) Thirty out of 66 patients (45.5%) had post-ET/PV myelofibrosis among the 3 studies that reported this information. (23–25) Fourteen out of 18 patients (78%) of patients in the study by Ianotto et al. had secondary MF.(22) Disease-specific risk stratification using standardized tools such as the dynamic international prognostic scoring system (DIPSS) were only used by 3 studies.(23–25) Baseline characteristics of the patients included in the individual studies are shown in Supplemental Table 1.

Patients were treated with various doses and schedules of non-pegylated (non-peg) IFN and pegylated (peg) IFN in 5 (25, 27–30) and 6 (21–26) studies, respectively (Table 1). All studies used a single-arm design which limited study quality (Supplemental Table 2).

Response assessment:

The ORR for IFN treatment among all studies was 49.9% (95% CI 30.4–69.3%) with significant heterogeneity among studies (Cochran's Q statistic: 28.5; I^2 statistic: 68.5%; $p=0.001$) (Figure 2A). Subgroup and meta-regression analyses showed that the ORR was not statistically significantly different ($p=0.998$) between peg-IFN (50.0%, 95% CI 26.2–73.9%; $I^2=76.9\%$) and non-peg-IFN (49.6%, 95% CI 20.5–79.0%; $I^2=56.7\%$) (Figure 2A).

As response rates to IFN can increase over time,(31) we conducted a subgroup and meta-regression analysis of the ORR based on the median duration of follow up with studies stratified by either < 24 months or ≥ 24 months of follow up. Studies which did not report the duration of follow up were excluded from this analysis. In this subgroup analysis, the ORR was not statistically significantly different ($p=0.79$) between studies with a median duration of follow up < 24 months (49.6%, 95% CI 20.5–79%, $I^2=56.7\%$) and studies with a median duration of follow up ≥ 24 months (43.6%, 95% CI 18.8–72.1%, $I^2=60.6\%$) (Supplemental Figure 1).

Due to the heterogeneity of reporting and definitions we were unable to conduct separate meta-analyses on the rate of CHR/CR and PHR/PR. While recent clinical trials of JAK inhibitors in MF have used spleen volume reduction and improvement in symptom burden as primary outcomes, only 6 out of 10 studies explicitly reported effects of IFN on spleen size. When reported, objective spleen size reductions were seen in 20–67% of patients. (20, 22, 25, 27, 28, 30, 32) However, we were unable to perform a meta-analysis of spleen responses given the heterogeneity of assessment (clinical exam vs ultrasound or cross-sectional imaging) and reporting of responses (absolute vs. relative spleen size reduction). A standardized assessment of symptom burden was not reported by any study.

Longitudinal bone marrow assessments were not reported separately in the included studies except for Silver et al., which showed improvements in bone marrow histopathology in 40% of patients.(25) Similarly, only the study by Silver et al. reported molecular response rates and found a minimal reduction in median *JAK2* V617F variant allele frequency (VAF) all patients (median VAF pretreatment: 46.2%; post treatment VAF: 42.4%).(25) However, among the 5 patients who also had an improvement in their bone marrow histopathology, the median reduction in *JAK2* V617F VAF was 23%.(25) Due to the lack of reporting of molecular and histopathologic responses by most studies we were unable to perform a dedicated meta-analysis of this outcome.

Safety assessment and discontinuation rate due to adverse events:

Reporting of adverse events was inconsistent; and 5 studies provided no documentation of adverse events at all. When reported, anemia and thrombocytopenia were the most frequent hematologic adverse events with flu-like symptoms being the most prevalent non-hematologic adverse events (Table 1). The treatment discontinuation rate per patient year was reported by 5 studies (Figure 2B). The annualized treatment discontinuation rate for the one study examining the use of peg-IFN was low at 0.5% per year of follow up.(25) One the contrary, for the studies on non-peg-IFN the treatment discontinuation rate was high at 35.8% (95% CI 3.5–68.1%). This difference was statistically significantly different in meta regression analysis ($p=0.032$).

Discussion:

To our knowledge, this is the first systematic review and meta-analysis on the use of IFN for the treatment of MF. We included 10 studies with a total of 141 patients treated with various formulations and treatment schedules of IFN over a period of more than three decades. Notably, peg-IFN formulations appeared as effective and better tolerated compared to non-peg IFN although this latter effect was largely driven by the study by Silver et al. and comparison of treatment efficacy was limited by variable outcome definitions used by the original studies.(25)

Our analysis revealed significant cross-study heterogeneity for the primary outcome and the annualized treatment discontinuation rate prompting an evaluation of included studies for differences in baseline patient and disease characteristics. The high response rates reported by Barosi et al. might be explained by the higher dose and daily dosing used in this study compared to others using non-peg-IFN as well as the fact that all patients had a hypercellular bone marrow at baseline.(27) The lower response rate in the study by Jabbour et al. could be due to the fact that only patients with primary MF were included in this trial, while studies with higher response rates such as Ianotto et al. had a substantial proportion of patients with secondary MF and responses to IFN might differ between primary and secondary MF.(21, 22) Finally, the study by Lindgren et al. only included patients that had been able to tolerate IFN for 3 months prior to study enrolment, which reduces the negative effect of early treatment discontinuation due to adverse events on ORR.(26) The low annualized treatment discontinuation rate reported by Silver et al. is likely due to the combination of a lower dose of IFN (starting doses 500,000 to 1 million units 3 times weekly for non-peg-IFN; 45 or 90

µg peg-IFNα-2a weekly), the long study duration (median treatment duration 5.6 years), and the inclusion of lower risk patients (73% low and 27% intermediate-1 risk by DIPSS) compared to other studies on MF.(25)

While ruxolitinib is the first-line treatment for intermediate- and high-risk MF and 5-year follow up from the COMFORT trials has shown a 30% relative risk reduction for death compared to best available treatment,(33, 34) NCCN guidelines still recommend IFN as an alternative to ruxolitinib and hydroxyurea for low-risk MF.(8) Several recent studies in PV and ET patients have shown that IFN can lead to molecular remission, suggesting a disease modifying effect.(11, 12, 35, 36) Similar disease-modifying effects have also been reported in studies on MF although complete molecular responses (defined as an undetectable *JAK2* V617F transcript) are rare.(15, 25) Studies showing durable responses even after the discontinuation of IFN treatment in PV patients lend additional support to the disease-modifying potential of IFN.(37) Unfortunately, molecular response data were available only for the studies by Silver et al., which precluded a formal meta-analysis on molecular responses in our study.(25) While molecular responses have also been reported for treatment with ruxolitinib,(32) IFN might be an attractive option for selected patients with low risk MF especially early in the disease course given its safety in pregnancy, the absence of leukemogenic side effects, and potentially less cytopenias compared to ruxolitinib.(15, 25, 38)

While we were unable to assess this question systematically, patients with lower risk MF and patients in the pre-fibrotic/hypercellular disease phase appear to benefit more from IFN treatment compared to patients in the overt disease phase with bone marrow hypocellularity and extensive fibrosis. This assumption is supported by the higher response rates reported in the study by Barosi et al. that enrolled only patients with a hypercellular bone marrow compared to the study by Seewann et al. which included patients with more advanced disease.(30, 39) Additional research to identify molecular biomarkers predicting higher response rates to IFN is needed. In ET and PV patients, *CALR* and *TET2* have been associated with higher and lower response rates to IFN, respectively.(12, 16) However, the effect of other mutations such as *ASXL1*, *EZH2*, or *SRSF2* on response rate and tolerability of IFN treatment in MF is controversial.(15, 25)

Ropeginterferon alfa 2b is a novel IFN formulation that has been shown to be non-inferior to hydroxyurea in terms of hematologic and spleen size responses in the randomized, open label phase III PROUD-PV trial at 12 months and to be superior to hydroxyurea at 36 months for the treatment of patients with PV.(40) Abstract data from patients with MF have also been recently presented. In a phase II study of 25 patients with pre-fibrotic MF (9 patients pretreated with peg-IFN), 50% of patients (6 out of 12 patients) had improvements in anemia and thrombocytosis, 78% (7 out of 9 patients) had improvement in leukocytosis, and no patient had disease progression after 2 years of treatment.(41) However, no improvement in spleen size was documented.(41) In a similar trial among 8 patients (2 early phase, 6 intermediate/high risk patients) treated with ropeginterferon alfa-2b, 2 patients experienced a reduction in spleen size and another patient had symptom improvement by MPN-SAF assessment.(42) Importantly, only 2 out of 25 and 1 out of 8 patients in the 2 studies discontinued treatment due to adverse events, respectively.(41, 42) While those early

results appear promising with comparable efficacy and potentially better tolerability to other formulations, additional data with longer follow up are needed. It will be interesting to see if disease-modifying effects are seen in the long-term follow up which could provide a rationale for active treatment early in the pre-fibrotic phase rather than the currently often practiced active surveillance in this setting.

Recently published data from a phase II study that tested combination treatment with ruxolitinib and peg-IFN in 18 patients with primary or secondary MF (6 patients with low, 9 with intermediate-I, and 3 with intermediate-II risk by DIPSS+ score) showed synergistic effects.(43) Among 18 patients, the combination therapy led to CHR and CR in 75% and 28% of patients, respectively, as well as molecular responses in 50% of patients.(43) However, tolerability of this combination remains a concern with grade 3/4 anemia (33%) and gastrointestinal bleeding (17%) being the most commonly reported adverse events and toxicities led to discontinuation of peg-IFN in 39% of patients.(43) Notably, although 94% of patients had been previously treated with peg-IFN and either been intolerant of or refractory to peg-IFN, 67% of patients were able to complete the trial per protocol, which could be due to the lower dose of peg-IFN used or a modulating effect of ruxolitinib on the IFN-related toxicity.(43) Similar preliminary results have been reported from the RUXOPEG study, a phase I/II trial combining ruxolitinib with peg-IFN in patients with primary or secondary MF ([NCT02742324](#)).(44) Compared with our results of IFN monotherapy, those studies support the synergistic effects of ruxolitinib and IFN and addition of ruxolitinib might be a viable option for both IFN-intolerant or IFN-refractory patients. Other JAK2 inhibitors such as fedratinib, pacritinib, and momelotinib have either been approved (fedratinib) or been tested in phase III clinical trials for the treatment of MF (pacritinib, momelotinib).(6, 45–47) Since adverse event profiles differ between those agents, combining IFN with other JAK2 inhibitors for specifically defined patient subsets could be a feasible strategy to reduce the potential of dose- or treatment-limiting adverse events such as thrombocytopenia or anemia that has been demonstrated with ruxolitinib and peg-IFN.(43, 45–47) However, only ruxolitinib has been combined with IFN in clinical trials thus far and additional research is needed.

Limitations of our study include the heterogeneity of IFN formulations and treatment schedules used, the variable duration of follow up and outcome definitions provided by the original studies. The absence of endpoints such as spleen volume reduction and improvement in symptom burden precluded a formal assessment of IFN effects on those outcomes and comparison with JAK inhibitors. Rates of treatment discontinuation varied widely and data on adverse events were inconsistently reported. Finally, we were unable to distinguish response rates between pre-fibrotic MF, primary MF, and post-PV/ET MF and the pathophysiologic processes in these disease stages might be different.

Conclusion:

In conclusion, this is the first systematic review and meta-analysis of IFN in MF including 141 patients treated in 10 studies. With an ORR of 49.9% and the potential for a durable molecular remission IFN can be an alternative for low-risk MF patients but further studies evaluating its role in the era of JAK inhibitors are needed.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments:

Amer Zeidan is a Leukemia and Lymphoma Society Scholar in Clinical Research and is also supported by a National Cancer Institute (NCI) Cancer Clinical Investigator Team Leadership Award (CCITLA). Research reported in this publication was supported by the NCI of the National Institutes of Health under Award Number P30 CA016359 and Cancer Center Support Grant/Core Grant to Memorial Sloan Kettering Cancer Center (P30 CA008748). The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

List of abbreviations:

AE	adverse events
AML	acute myeloid leukemia
CALR	calreticulin
CI	clinical improvement
CHR	complete hematologic response
CR	complete response
DIPSS	Dynamic International Prognostic Scoring System
ELN	European Leukemia Network
ET	Essential thrombocytosis
IFN	Interferon
IPSS	International Prognostic Scoring System
IWG-MRT	International Working Group for Myeloproliferative Neoplasms Research and Treatment
JAK	Janus kinase
MF	myelofibrosis
MPL	myeloproliferative leukemia virus
MPN	myeloproliferative neoplasm
NCCN	National Comprehensive Cancer Network
ORR	overall response rate
peg	pegylated
PHR	partial hematologic response

PR	partial response
PV	polycythemia vera
VAF	variant allele frequency

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Clinical practice points:

- European and US guidelines recommend interferon as a potential alternative to hydroxyurea and ruxolitinib for the treatment of low-risk myelofibrosis.
- IFN has been used for over 3 decades for the treatment of myelofibrosis with conflicting results prompting this systematic review and meta-analysis. Among the 10 studies with 141 myelofibrosis patients included, 49.9% of patients achieved hematologic improvements including a small proportion of patients with molecular responses suggesting a disease-modifying potential.
- Pegylated and non-pegylated interferon formulations appeared equally efficacious but the adverse event-related treatment discontinuations were more common with the latter one
- Identification of biomarkers predicting response to interferon, combinations of interferon with JAK inhibitors, and attempts to improve tolerability of interferon are needed.

Figure 1: Flow chart showing study selection as per the MOOSE guidelines

Figure 1 illustrates the search strategy and stepwise process of study selection used in this meta-analysis. MEDLINE and EMBASE via Ovid, the COCHRANE registry of clinical trials (CENTRAL), Scopus and the Web of Science electronic databases were searched without language restriction from inception through March 21st, 2019, using the following combination of free-text terms linked by Boolean operators: [“polycythemia” OR “polycythemia vera” OR “essential thrombocythosis” OR “myelofibrosis” OR “myeloproliferative neoplasm” OR “MPN”] AND [“interferon” OR “IFN” OR “pegylated interferon” OR “peginterferon” or “alpha2b interferon” OR “alpha2 interferon” OR “alpha interferon”]. After removal of duplicates, studies were excluded if they were (I) review articles, commentaries or basic research articles, (II) reporting results from diseases other than PV, ET, or MF (e.g. chronic myeloid leukemia), or (III) case series with less than 5

patients. Subsequently, full texts of the potentially eligible studies were reviewed for eligibility. We excluded studies that 1) lacked information on the primary outcomes of overall response rates (ORR), 2) IFN listed only among “other therapies” without separate reporting of outcome data, 3) studies published only in abstract form, 4) duplicate publications from the same patient cohort, 5) IFN given as part of combination therapy, 6) clinical trials without published results, 7) without an available English full text, and 8) studies on ET and PV patients (reported in separate manuscript)

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Figure 2: Meta-analysis
(A) Overall response rate (ORR) to IFN treatment
(B) Discontinuation rate of IFN treatment (per patient year of follow up)

Table 1:

Treatment characteristics, outcomes and adverse effects of studies of myelofibrosis

Author (ref)	Year	Treatment and treatment schedule	N (patients)	Outcome definition	Outcomes	Histopathologic and molecular response	Adverse events (AE)
Barosi et al. (27)	1990	r-IFN α 3–5 $\times$ 10 ⁶ IU daily; maintenance treatment with individualized doses	12	Not reported	ORR: 100% (no definition of CR or PR), 67% with spleen size reduction (median: 3cm, R: 0–3.5cm)	<u>Histopathologic assessment:</u> Baseline: all patients with hypercellular marrow Post treatment: no change in bone marrow fibrosis <u>Molecular response:</u> Baseline: not reported Post treatment: not reported	2 patients discontinued treatment due to AE Hematologic AEs: not reported Non-hematologic AEs: not reported
Gowin et al. (23)	2012	Peg-IFN2 α 30–300 μ g/week	17 (5 patients low, 9 intermediate, and 2 high risk by IPSS; 8 patients with post-ET/PV MF)	2006 IWG-MRT response criteria (48)	ORR: 29% (CR: 0%, PR: 12%, CI: 18%), no spleen outcomes reported	<u>Histopathologic assessment:</u> Baseline: not reported Post treatment: not reported <u>Molecular response:</u> Baseline: 53% JAK2 V617F-positive; no allele burden reported Post treatment: not reported	Not reported separately for MF cohort
Gowin et al. (24)	2017	Peg-IFN2 α 45–270 μ g/week	19 (6 patients low, 3 intermediate-1, 8 intermediate-2, and 2 high risk by DIPSS; 13 patients with post-ET/PV MF)	2013 ELN/IWG-MRT response criteria (49)	ORR: 32% (CR: 0%; PR: 11%); no spleen outcomes reported	<u>Histopathologic assessment:</u> Baseline: not reported Post treatment: not reported <u>Molecular response:</u> Baseline: not reported Post treatment: not reported	Not reported separately for MF cohort
Ianotto et al. (22)	2009	Peg-IFN α –2a dosing and schedule not reported	18 (14 patients with secondary MF)	European Myelofibrosis Network (EUMNET) 2005 response criteria(50)	ORR: 89% (CHR: 33%); 11% with improvement in splenomegaly	<u>Histopathologic assessment:</u> Baseline: not reported Post treatment: not reported <u>Molecular response:</u> Baseline: 78% (14 out of 18) JAK2 positive, no baseline allele burden reported Post treatment: not reported	Hematologic AEs: Not reported Non-hematologic AEs: Not reported
Ibbour et al. (21)	2007	Peg-IFN α –2b 2–3 μ g/kg/week	11 (all primary MF)	CR: symptom resolution with Hgb 11g/dL, platelet count >100 $\times$ 10 ⁹ /L and ANC 1000 ³ /L and normalization of bone marrow fibrosis PR: all criteria for CR except for resolution of bone marrow fibrosis	ORR: 9% (CR: 9%; PR: 0%); no spleen outcomes reported	<u>Histopathologic assessment:</u> Baseline: not reported Post treatment: not reported <u>Molecular response:</u> Baseline: not reported Post treatment: not reported	Not reported separately for MF cohort

Author (ref)	Year	Treatment and treatment schedule	N (patients)	Outcome definition	Outcomes	Histopathologic and molecular response	Adverse events (AE)
Lindgren et al.(26)	2018	Peg-IFN α -2b 90 μ g/week	10	2013 IWG-MRT and ELN criteria(49), no CR or PR given absence of bone marrow assessment	ORR: 90%, no reporting of CR, PR, and spleen responses	<u>Histopathologic assessment:</u> Baseline: not reported <u>Post treatment:</u> not reported <u>Molecular response:</u> Baseline: 50% (4 out of 8) JAK2 V617F, 40% (4 out of 10) CALR-positive; no baseline allele burden reported <u>Post treatment:</u> not reported	Not reported separately for MF patient cohort
List et al.(29)	1992	r-IFN α 0.5 $\times$ 10 ⁶ IU every other day to r-IFN α 3 $\times$ 10 ⁶ IU daily	11	Not reported	ORR: 46% (no reporting of CR, PR, and spleen responses); no symptom improvement	<u>Histopathologic assessment:</u> Baseline: not reported <u>Post treatment:</u> not reported <u>Molecular response:</u> Baseline: not reported <u>Post treatment:</u> not reported	1 transformation to AML; 2 treatment discontinuations due to AE Hematologic AEs: 2 patients each with worsening anemia and thrombocytopenia Non-hematologic AEs: flu-like symptoms, depression and nausea/vomiting as dose-limiting AEs
McCarthy et al.(28)	1991	r-IFN α 3 $\times$ 10 ⁶ IU t.i.w.	8	Not reported	ORR: 45% (no reporting of CR and PR), 3 patients with spleen response	<u>Histopathologic assessment:</u> Baseline: not reported <u>Post treatment:</u> not reported <u>Molecular response:</u> Baseline: not reported <u>Post treatment:</u> not reported	2 treatment discontinuations due to AE (flu-like symptoms, neutropenia) Hematologic AEs: 2 patients with thrombocytopenia Non-hematologic AEs: mild flu-like symptoms in all patients
Seewann et al.(30)	1988	r-IFN α 1-2 $\times$ 10 ⁶ IU daily	5	Not reported	ORR: 20% (no formal criteria), 1 patient with reduction in spleen size	<u>Histopathologic assessment:</u> Baseline: 3 hypocellular, 2 hypercellular; all with increased reticulin fibrosis <u>Post treatment:</u> not reported <u>Molecular response:</u> Baseline: not reported <u>Post treatment:</u> not reported	No treatment discontinuation due to AE Hematologic AEs: 3 patients with neutro- and thrombocytopenia Non-hematologic AEs: None
Silver et al. (25)	2017	rIFN α 2b (500,000-3 million units t.i.w. (18 patients) or 45-90 μ g/week peg-IFN α -2a (8 patients). Five patients began treatment with rIFN α -2b, but changed to peg-IFN α -2a	30 (22 low, 8 intermediate-1 risk by DIPSS; 9 patients with post-ET/PV MF)	2013 ELN/IWG-MRT response criteria(49)	ORR: 50% (CR: 7%; PR: 30%); 45% with spleen size reduction	<u>Histopathologic assessment:</u> Baseline: all patients residual hematopoietic foci >15% <u>Post treatment:</u> improvement in 40% of patients <u>Molecular response:</u> Baseline: JAK2 (66.7%; median VAF 46.2%), CALR 20% (median VAF not reported), MPL (6.7%, median VAF not reported) <u>Post treatment:</u> median JAK2 V617F allele burden 42.4%	109 AE in 30 patients. 2 with grade >3; 0.5% annualized treatment discontinuation rate due to AE Hematologic AEs: anemia 13.2% grade 2, thrombocytopenia 13.2% grade 4 Non-hematologic AEs: 1 treatment discontinuation due to hyperthyroidism, no grade 3

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AE - adverse events; *CI* - clinical improvement; *CHR* - complete hematologic remission; *CR* - complete remission; *ELN* - European Leukemia Network; *ET* - essential thrombocytosis, *IFN* - Interferon; *IWG-MRT* - International Working Group for Myeloproliferative Neoplasms Research and Treatment; *ORR* - overall response rate; *PR* - partial remission; *PV* - polycythemia vera; *Li.W.* - three times per week, *VAF* - variant allele frequency