CHAPTER 35
Hematopoietic growth factors

David J. Kuter
Division of Hematology, Massachusetts General Hospital and Harvard Medical School, Boston, MA, USA

Introduction
Over the past several decades, the major hematopoietic growth factors have been identified, purified, and therapeutic products made. These hematopoietic growth factors affect the growth and differentiation of stem cells and later progenitor cells of all lineages. Several of these molecules play important roles in patient care and are widely used. After a brief review of general principles of hematopoietic growth factor function, this chapter will focus on the clinically relevant hematopoietic growth factors that affect erythroid, myeloid, and megakaryocyte differentiation, specifically erythropoietin (epoetin alfa and darbepoetin alfa), granulocyte colony-stimulating factor (G-CSF; filgrastim, tbo-filgrastim, and pegfilgrastim), granulocyte macrophage colony-stimulating factor (GM-CSF; sargramostim), and thrombopoietin (TPO; romiplostim and eltrombopag). The general biology and clinical use of each will be described as well as their role in transfusion medicine.

General principles of hematopoietic growth factors
Pluripotent stem cells give rise to all of the final differentiated blood cells (Figure 35.1). The molecular differentiation steps for some of these precursor cells have been described and in general involve a random (“stochastic”) process in which lineage-specific differentiation occurs at multiple stages. For example, a micro-RNA (miR-150) binds to and downregulates c-Myb, which causes the common erythroid–megakaryocyte precursor to undergo megakaryocytic, not erythroid, differentiation.1 But the subsequent survival of cells at each differentiation stage is determined by the presence of specific hematopoietic growth factors; if the stage- or lineage-specific hematopoietic growth factor is absent, the cells undergo programmed cell death. For example, erythroid burst-forming cells (BFU-Es) will continue their divisions as long as erythropoietin is present; if absent, BFU-Es undergo programmed cell death.

In this complicated hierarchy of cell division, differentiation, and apoptosis, the hematopoietic growth factors can be generally grouped into those that are early acting, whose circulating levels are not altered and affect multiple lineages (interleukin-3 [IL3], IL6, IL11, GM-CSF, stem cell factor, and TPO), versus those that are late acting, whose levels are modulated and affect single lineages (erythropoietin, G-CSF, macrophage colony-stimulating factor [M-CSF], and TPO). All of these hematopoietic growth factors are active at very low concentrations and work via specific cell surface receptors, thereby promoting the viability of the target cells. For the clinically relevant hematopoietic growth factors, G-CSF, erythropoietin, M-CSF, and TPO, a number of other general physiological principles should be noted:

- The circulating level of each factor in normal physiology is inversely proportional to the mass of the differentiated cell it regulates; for example, as the hemoglobin, absolute neutrophil count (ANC), monocyte count, and platelet count fall, the respective levels of erythropoietin, G-CSF, M-CSF, and TPO rise.
- The normal physiologic response is usually "log-linear," that is, erythropoietin levels increase exponentially as the hemoglobin declines on a linear scale (Figure 35.2).2
- Circulating levels of each factor are determined by the relative rates of its production and metabolism. For erythropoietin, there is a precise mechanism that regulates its production with a rather fixed rate of renal clearance. For TPO and M-CSF, levels are primarily regulated through clearance by the platelet or macrophage, respectively. These cells have very high-affinity receptors for their factor and bind, internalize, and degrade it. There is little effect on the rate of production. Finally, G-CSF is regulated by both production and clearance. Mature neutrophils bind and clear G-CSF, but many other cells (eg, monocytes, endothelial cells) can increase G-CSF production when stimulated. In normal basal physiology, G-CSF production is relatively constant.
- This normal physiologic response may be altered in pathologic states.
  - In anemia of chronic disease, renal erythropoietin production is decreased.2
  - In acute infection, there is a marked increase in G-CSF production.
  - In liver failure, TPO production is decreased.3
- The clinical effect of pharmacologic administration of a hematopoietic growth factor also operates on an exponential dose–response curve. In a healthy subject, linear increases in any blood cell require an exponentially greater dose of the specific hematopoietic growth factor.4,5

Erythroid growth factors
Ever since the initial studies by Paul Carnot in 1906, it has been known that blood contained a “hémopoïétine” that had the ability to

© 2016 John Wiley & Sons, Ltd. Published 2016 by John Wiley & Sons, Ltd.

418
Figure 35.1 Scheme of normal hematopoiesis. The many different stages of differentiation of the myeloid, erythroid, and megakaryocyte lines are illustrated along with their relevant early- and late-acting hematopoietic growth factors. BFU, blast-forming unit; CFU, colony-forming unit; CLP, common lymphoid progenitor; CMP, common myeloid progenitor; Epo, erythropoietin; G-CSF, granulocyte colony-stimulating factor; GM-CSF, granulocyte–macrophage colony-stimulating factor; GMP, granulocyte–macrophage progenitor; IL, interleukin; LTR-HSC, long-term repopulating hematopoietic stem cell; M-CSF, monocyte/macrophage colony stimulating factor; MEP, megakaryocyte–erythroid progenitor; SCF, stem cell factor; STR-HSC, short-term repopulating hematopoietic stem cell; TNF, tumor necrosis factor; Tpo, thrombopoietin. Source: Sieff CA, Zon LI. Anatomy and physiology of hematopoiesis. In: Nathan and Oski’s Hematology of Infancy and Childhood, 7th ed, Orkin et al. (eds.), Elsevier Philadelphia, 2009. Reproduced with permission.
stimulate red cell production. Decades after identifying a factor in the urine that stimulated red cell production, erythropoietin (EPO) was finally purified and a recombinant form was approved by the US Food and Drug Administration (FDA) in 1989. EPO was the first hematopoietic growth factor available for clinical use and paved the way for the development of many others.

Structure, function, and physiology

Erythropoietin is made as a 193-amino-acid precursor, of which 27 amino acids are cleaved off to produce a 166-amino-acid form that loses a terminal arg166 to give the final 165-amino-acid (molecular weight \( = 30,400 \text{ Da} \)) heavily glycosylated (30% carbohydrate) protein. It is made by a single gene (7q22.1) and produced by interstitial peritubular fibroblasts in the kidney with maybe a small amount being made by the liver. Using an efficient renal sensor of hypoxia (hypoxia inducible factor [HIF]), circulating EPO levels are primarily determined by its rate of transcription. HIF is a dimeric protein made of \( \alpha \) and \( \beta \) subunits that binds to a promoter region of the EPO gene and increases its rate of transcription. However, the HIF\( \alpha \) subunit undergoes proline hydroxylation in the presence of oxygen and is degraded by the proteasome. So with normoxia, HIF\( \alpha \) is rapidly degraded and transcription of the EPO gene reduced; with hypoxia, HIF\( \alpha \) survives and increases transcription. EPO has no storage form and once made is immediately secreted into the circulation and cleared by the kidney.

EPO binds to and activates preformed, inactive, dimeric EPO receptors (Figure 35.3) that are present on pronormoblasts, basophilic normoblasts, and polychromatophilic and orthochromatophilic normoblasts, but not on reticulocytes or mature red cells. When EPO is present, these precursor cells do not undergo apoptosis but survive and produce mature red cells.

The usual log-linear relationship between the EPO level and the hematocrit (Figure 35.2) seen in disorders such as iron deficiency is blunted in conditions historically referred to as anemia of chronic disease (e.g., diabetes, cancer, renal failure, and inflammation).

Clinically available erythroid growth factors

**Epoetin alfa (Epogen, Procrit)**

This 165-amino-acid (MW \( = 30,400 \text{ Da} \)) glycosylated protein (Figure 35.3) is made in Chinese hamster ovary (CHO) cells and is available in vials and prefilled syringes in a wide variety of doses. Epoetin alfa is FDA approved for:

- the treatment of anemia due to chronic kidney disease (CKD), including patients on dialysis and not on dialysis, to decrease the need for red blood cell (RBC) transfusion;
- the treatment of anemia in patients with nonmyeloid malignancies where anemia is due to the effect of concomitant myelosuppressive chemotherapy, and upon initiation, there is a minimum of two additional months of planned chemotherapy;
- the treatment of anemia due to zidovudine administered at \( \leq 4200 \text{ mg/week} \) in HIV-infected patients with endogenous serum erythropoietin levels of \( \leq 500 \text{ IU/mL} \); and
- reducing the need for allogeneic RBC transfusions among patients with perioperative hemoglobin \( > 10 \) to \( \leq 13 \text{ g/dL} \) who are at high risk.

---

**Figure 35.2** Log-linear relationship between EPO level and hematocrit. EPO levels were determined for normal blood donors (triangles) and patients with anemia (squares; those with renal disease, rheumatoid arthritis, or solid tumors were excluded). Dashed line denotes limits of detection for this assay. Note that EPO does not begin to rise until hematocrit falls below \( \sim 36 \), suggesting that other factors (i.e., androgens) account for the higher values. Source: Erslev (1991). Reproduced with permission of Elsevier.

**Figure 35.3** Binding of epoetin alfa and darbepoetin alfa to the EPO receptor. Epoetin alfa has three N-linked carbohydrate chains with a maximum of 14 sialic acids. It has a MW of 30,400 Da and is 40% carbohydrate. Darbepoetin alfa has five N-linked carbohydrate chains with a maximum of 22 sialic acids. It has a MW of 39,000 Da and is 51% carbohydrate. Both bind to and activate the preformed dimeric EPO receptor. Source: D. Kuter. Reproduced with permission.
risk for perioperative blood loss from elective, noncardiac, nonvascular surgery and who are not willing to donate autologous blood preoperatively.

The starting dose of epoetin alfa is usually 50 U/kg tiw for renal failure, 100 U/kg tiw for HIV, 150 U/kg tiw for chemotherapy (40,000 units per week has become a standard for oncology patients11), and either 300 U/kg daily for 10 days for preoperative anemia with surgery in under three weeks or 600 U/kg weekly for preoperative anemia when surgery is greater than three weeks away. Subsequent doses are adjusted every three to four weeks based upon response. This drug is much more effective when given subcutaneously than IV, with levels rising at two hours, peaking at 18 hours, and with a T1/2 of 8.5 hours.

Darbepoetin alfa (Aranesp)
Darbepoetin alfa was designed from understanding how epoetin alfa is cleared from the circulation. Epoetin alfa has many isoforms due to variations in the number of sialic acids present (Figure 35.3).12,13 Isoforms with the maximum 14 sialic acids have a prolonged half-life, whereas those with eight have almost no biologic effect. This led to the hypothesis that if the number of sialic acids on epoetin alfa were increased, its T1/2 would be even longer. By creating two additional N-linked glycosylation sites on epoetin alfa, darbepoetin alfa was created and contains a maximum of 22 sialic acids versus the 14 on epoetin alfa (Figure 35.3). Adding these sialic acids minimally reduced binding to the EPO receptor, but markedly increased the T1/2 from 8.5 hours to 25.3 hours, thereby increasing the biologic effect. Darbepoetin alfa (MW = 39,000 Da) is made in CHO cells and is 51% carbohydrate. It comes in vials and prefilled syringes with various doses. It is FDA approved for:

- the treatment of anemia due to chronic kidney disease (CKD), including patients on dialysis and patients not on dialysis; and
- the treatment of anemia in patients with nonmalignancies where anemia is due to the effect of concomitant myelosuppressive chemotherapy, and upon initiation, there is a minimum of two additional months of planned chemotherapy.

The starting dose for renal failure is of 0.45 mcg/kg weekly; for oncology, either 100 mcg weekly or 200 mcg every two weeks. Doses are subsequently adjusted every four weeks according to response.

Effects and adverse effects of erythropoietin administration
After the administration of epoetin alfa or darbepoetin alfa to healthy individuals, the reticulocyte count starts to rise on day 3 and peaks on day 10, while the hematocrit starts to rise on day 8 and peaks at days 20–25.14 Importantly, the iron saturation and transferrin begin to fall by day 4 and drop 74% by day 16.14

The major side effects of darbepoetin alfa or epoetin alfa administration are listed in Table 35.1. Of these, thrombosis and potential adverse outcomes of cancer patients deserve further detail.

Thrombosis
There is an approximately twofold increased rate of thromboembolism in renal failure and cancer patients receiving these agents. A meta-analysis of 91 trials with 20,102 cancer patients showed that the risk ratio (RR) of thromboembolic complications was increased in patients receiving these drugs compared to controls (RR, 1.52; 95% confidence interval [CI], 1.34–1.74; 57 trials, N = 15,498).15 In a meta-analysis of 27 trials (10,452 patients), chronic kidney disease patients at a higher hemoglobin target (median, 13.0 g/dL; IQR, 12.0–14.0 g/dL) had higher risks for stroke (RR, 1.51; 95% CI, 1.03–2.21), hypertension (RR, 1.67; 95% CI, 1.31–2.12), and vascular access thrombosis (RR, 1.33; 95% CI, 1.16–1.53) compared with those at a lower hemoglobin target (median, 10.1 g/dL; IQR, 9.2–11.0 g/dL). However, there were no statistically significant differences in the risks for mortality (RR, 1.09; 95% CI, 0.99–1.20), serious cardiovascular events (RR, 1.15; 95% CI, 0.98–1.33), or end-stage kidney disease (RR, 1.08; 95% CI, 0.97–1.20).16

Tumor progression and cancer mortality
The second major concern with the erythropoietin growth factors is that they may increase tumor progression and mortality in cancer patients. In one study, head and neck cancer patients treated with erythropoietin to a target hemoglobin of 14 to 15 had worse progression-free survival (RR, 1.62; 95% CI, 1.22–2.14; p = 0.0008), loco-regional control (RR 1.69; 95% CI 1.16–2.47; p = 0.007), and overall survival (RR 1.39; 95% CI 1.05–1.84; p = 0.02) than placebo.17 However, this study and others reporting similar worsened outcomes were not sufficiently structured or powered to assess cancer progression or survival. In the absence of adequate clinical trials, a massive meta-analysis of cancer patients treated with epoetin alfa or darbepoetin alfa has been conducted of 21,102 subjects in 91 trials.15 Erythropoietin growth factors significantly reduced the relative risk of red cell transfusions (RR, 0.65; 95% CI, 0.62–0.68; 70 trials, N = 16,093). On average, patients in the erythropoietin arms received one unit less blood than the control group with a suggestion of increased quality of life. However, erythropoietin growth factors increased mortality during active study (HR, 1.17; 95% CI, 1.06–1.29; 70 trials, N = 15,935) and might have decreased overall survival (HR, 1.05; 95% CI, 1.00–1.11; 78 trials, N = 19,003). There was no evidence that erythropoietin affected tumor response (RR, 1.02; 95% CI, 0.98–1.06; 15 trials, N = 5012).

Clinical use of erythropoietin growth factors
The erythropoietin growth factors have been used in a wide range of medical conditions ranging from renal failure to hemochromatosis (Table 35.2).

Chronic renal failure
In patients with chronic renal failure, the use of erythropoietin growth factors has been shown to increase hematocrit, decreased transfusion needs, and increase quality of life. For most such patients, the hemoglobin target is no higher than 11–12 g/dL.19 This is based upon clinical studies that showed that patients experienced greater risk for death and serious cardiovascular events with a target hemoglobin of >13 g/dL; those with a target of 11.5–13 g/dL had no better or worse outcomes, but possibly improved quality of life compared to those with a lower hemoglobin level.16,19

Table 35.1 Adverse effects of erythropoietin growth factors

<table>
<thead>
<tr>
<th>Effect</th>
<th>Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypertension</td>
<td>renal failure patients</td>
</tr>
<tr>
<td>Seizures</td>
<td>renal failure patients</td>
</tr>
<tr>
<td>Allergic reactions</td>
<td>rare</td>
</tr>
<tr>
<td>Antibody formation leading to pure red cell aplasia</td>
<td>European formulation: very rare in United States</td>
</tr>
<tr>
<td>Thrombosis</td>
<td>twofold increase in all patients</td>
</tr>
<tr>
<td>Increased cardiovascular complications and death at Hgb &gt; 11</td>
<td>renal failure patients</td>
</tr>
<tr>
<td>Decreased survival if target Hgb &gt; 12</td>
<td>cancer patients (breast, head-neck, cervical, and lymphoma) on chemotherapy</td>
</tr>
<tr>
<td>Decreased survival: cancer patients not receiving chemotherapy</td>
<td></td>
</tr>
</tbody>
</table>
Cancer patients

The use of erythroid growth factors has been shown to decrease the need for red cell transfusions in most populations studied; the exceptions are anemic cancer patients who are not receiving chemotherapy and those with EPO levels >500 IU. Given the concerns over tumor progression and survival, current guidelines encourage a thorough discussion of the risks and benefits of these agents before their use in all patients receiving myelosuppressive chemotherapy. Furthermore, these agents are not to be used in cancer patients not receiving myelosuppressive chemotherapy or in those for whom the treatment is expected to be curative. Dosing is not started unless the hemoglobin is <10 g/dL, with only the lowest dose being used to avoid transfusions, and treatment is discontinued when chemotherapy ends. It should be remembered that improvement in quality of life reaches a plateau once the hemoglobin rises above 11 g/dL (range, 11–13 g/dL).

HIV infection

Clinical benefit has been demonstrated in anemic (Hct <30) HIV-infected patients with endogenous EPO levels ≤500 IU/mL undergoing treatment with zidovudine at doses ≤4200 mg/week. In a pooled analysis of four trials, 297 AIDS patients treated with zidovudine received either epoetin alfa or placebo for 12 weeks. No benefit was seen in those with EPO levels >500 IU, but in those with lower levels, epoetin alfa therapy decreased the mean number of units of blood transfused per patient compared with placebo (3.2 units vs. 5.3 units, respectively; p = 0.003) and increased the mean hematocrit from the baseline level (4.6 vs. 0.5 percentage points, respectively; p < 0.001). Overall quality of life improved in patients on epoetin alfa but was not statistically significant (p = 0.13).

Preoperative anemia

In patients who are not candidates for autologous blood transfusion, epoetin alfa has been shown to increase the hemoglobin and decrease the need for subsequent allogeneic transfusions when given to patients with hemoglobin between 10 and 13 prior to therapy. In two studies of 461 patients undergoing major orthopedic surgery, 358 received epoetin alfa (100–300 IU/kg sq daily for 15 days [10 days preoperative] or 600 IU/kg weekly for four weeks). The use of allogeneic transfusions was significantly reduced (p < 0.001).

An important issue associated with use of erythroid growth factors

The response to erythroid growth factors depends on the availability of adequate reserves of bone marrow and iron. Because the ferritin falls dramatically after administration of EPO, iron supplementation has been shown to be critical in amplifying the hemoglobin response to erythroid growth factors in chronic kidney disease patients and in cancer chemotherapy patients. In most studies, intravenous iron was superior to oral iron repletion.

Implications for transfusion medicine

The impact of the erythroid growth factors on blood resource utilization has received little attention. With the recent guideline changes for erythroid growth factors in chemotherapy patients, their use has dropped dramatically with an estimate that RBC transfusions will increase 1% nationally. Similar regulatory changes have also reduced the use of these agents in chronic kidney disease and have been accompanied by increased rates of transfusion. These changes have the potential to increase the pressure on available blood supplies.

Although neither epoetin nor darbepoetin act sufficiently rapidly to replace RBC transfusions in acute settings, the newly described sotatercept (ACE-011) may fulfill this role. This recombinant human fusion protein contains the extracellular domain of the human activin receptor IIA, and it binds to and inhibits activin and other members of the transforming growth factor-β (TGF-β) superfamily. Administration of sotatercept to healthy volunteers led to a rapid and sustained increase in hematocrit in phase I clinical trials by an as-of-yet unclear mechanism.

The erythroid growth factors remain the only option for many patients who cannot receive RBC transfusions for religious or medical reasons. Administration of 140 IU/kg epoetin alfa three times a week for three weeks along with oral iron three times a day to 45 Jehovah’s Witness patients increased the hemoglobin and allowed all patients to undergo major cardiac procedures without blood transfusion.

Finally, the use of erythroid growth factors has been demonstrated to increase the preoperative collection of RBCs. In a randomized study of 47 patients scheduled for orthopedic surgery, patients received 600 IU/kg or placebo twice a week for 21 days (along with oral iron) during which time up to six units of RBCs were scheduled to be collected. Epoetin alfa–treated patients collected a mean (±SD) of 5.4 ± 0.2 units compared with 4.1 ± 0.2 for the placebo group.

Myeloid growth factors

While deficiencies in RBCs and platelets can be readily treated with transfusion, neutropenia may be a severe, life-threatening disorder that is not readily amenable to transfusion. Although neutrophil transfusions may be arranged, they are neither readily available nor of demonstrated benefit. With the FDA approval of both G-CSF (filgrastim) and GM-CSF (sargramostim) in early 1991, treatment and prevention of neutropenia became a reality.

Structure, function, and physiology

G-CSF is a 174-amino-acid glycosylated protein (MW = 25,000 Da) and is encoded by a single gene (17q21.1). G-CSF is synthesized by macrophages, monocytes, endothelial cells, and fibroblasts, and its production can be vastly increased when other inflammatory molecules (TNFα, IL1, IL3, interferon γ, and IL4) are present. It is made without a storage form and immediately released into the circulation, where it is cleared by neutrophils via their G-CSF receptor (Kd = 65 pM) and less so by the kidney. In the noninfected individual, production is constant, and circulating levels are inversely proportional to the ANC by way of neutrophil clearance. With infection, production is stimulated by inflammatory cytokines. Normal G-CSF levels are <39 ng/L, but rise to a different

---

**Table 35.2 Clinical uses of erythroid growth factors**

<table>
<thead>
<tr>
<th>Use</th>
<th>Ch. 35.2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chronic renal failure</td>
<td></td>
</tr>
<tr>
<td>HIV infection</td>
<td></td>
</tr>
<tr>
<td>Cancer chemotherapy</td>
<td></td>
</tr>
<tr>
<td>Preoperative anemia: hemoglobin 10–13</td>
<td></td>
</tr>
<tr>
<td>(Myelodysplastic syndromes [MDS])</td>
<td></td>
</tr>
<tr>
<td>(Anemia in rheumatoid arthritis)</td>
<td></td>
</tr>
<tr>
<td>(Anemia of chronic disease)</td>
<td></td>
</tr>
<tr>
<td>(Anemia in congestive heart failure)</td>
<td></td>
</tr>
<tr>
<td>(Treatment of anemia in patients whose religious beliefs forbid transfusion)</td>
<td></td>
</tr>
<tr>
<td>(Anemia due to hepatic G disease)</td>
<td></td>
</tr>
<tr>
<td>(Mobilization of iron in hemochromatosis)</td>
<td></td>
</tr>
<tr>
<td>(Improved harvesting of autologous blood)</td>
<td></td>
</tr>
<tr>
<td>(Mobilize peripheral blood progenitor cells [PBPCs])</td>
<td></td>
</tr>
<tr>
<td>(Neuroprotection—studies in progress)</td>
<td></td>
</tr>
</tbody>
</table>

Uses in parentheses denote lack of FDA approval.
extant depending on the type of infection: bacterial, 799 ± 1501 ng/L; viral, 58 ± 34 ng/L; and mycoplasma, 60 ± 33 ng/L.

G-CSF has many biological effects (Table 35.3). For existing neutrophils, it promotes demargination and release from bone marrow reserves and increases neutrophil survival. Neutrophil production is markedly increased, with the maturation time reduced from six days to three days, an increase in "left shift," and often the appearance of Dohle bodies and toxic granulation. Neutrophil motility is altered. Neutrophil function (phagocytosis, O2 generation, endomitosis, antibody-dependent cell-mediated cytoxicity [ADCC], and FcγRI receptors) is increased, an underappreciated effect. Finally, G-CSF mobilizes peripheral progenitor cells. In animals lacking G-CSF,39 neutrophils are 20–30% of normal, neutrophil precursors are 50% of normal, and there is decreased neutrophil mobilization into the circulation.

Although G-CSF is necessary for the normal production and maturation of myeloid precursors into neutrophils, GM-CSF has no physiologic importance for hematopoiesis. In GM-CSF-deficient mice, neutrophils, macrophages, and their precursors are normal but the mice develop pulmonary problems.40

GM-CSF is a 127-amino-acid (23,000 Da) glycosylated protein made from a single gene (5q31.1). It is made by T cells, macrophages, monocytes, endothelial cells, and fibroblasts. Significant levels are not detected in the circulation, and amounts of this protein do not vary inversely to the ANC. GM-CSF does not increase in amount during infection, but its production by bone marrow stromal cells may be decreased by interferon 1β. It is normally cleared by the GM-CSF receptor on neutrophils and monocytes, with less than 40% being renally cleared.

The biologic effects of GM-CSF on neutrophil survival, production, and mobility are comparable to those of G-CSF but are extended to eosinophils and monocytes (Table 35.3). In addition to increasing neutrophil function like G-CSF, it also increases the destruction of Trypanosoma cruzi, Mycobacterium avium, Influenza A, Candida, and T. cruzi. GM-CSF has increased TNFα and IL1, which may explain some of its clinical adverse effects.

### Clinically relevant myeloid growth factors

#### Filgrastim (Neupogen)

Filgrastim is a 175-amino-acid protein (MW = 18,800 Da) identical to the native molecule except for an added N-terminal methionine (r-met hG-CSF) and a lack of glycosylation since it is made in Escherichia coli. Vials and prefilled syringes of filgrastim are available in doses of 300 and 480 mcg. For most uses, the dose is 5 mcg/kg/day (~230 mcg/m2/day). Remembering the log-linear dose–response curve for hematopoietic growth factors, one rounds down to the nearest vial or syringe size. Although doses for stem cell mobilization are usually higher, much lower doses (0.4 mcg/kg/day) are often adequate in neutropenic patients with HIV, idio-pathic neutropenia, or drug-induced neutropenia. Filgrastim has a T1/2 of 3.5 hours and is metabolized mostly by mature neutrophils and less so by the kidney. Like most hematopoietic growth factors, subcutaneous administration gives a better response than intravenous administration. In chemotherapy patients, filgrastim is usually started 24 hours after the end of chemotherapy and stopped at least 24 hours before the next chemotherapy dose. In chemotherapy patients, prescribing information suggests stopping once the ANC is >10,000, but it is often stopped once the ANC is >2000. Filgrastim is FDA approved for:
- decreasing the incidence of infection, as manifested by febrile neutropenia, in patients with nonmyeloid malignancies receiving myelosuppressive anticancer drugs associated with a significant incidence of severe neutropenia with fever;
- reducing the time to neutrophil recovery and the duration of fever following induction or consolidation chemotherapy treatment of adults with acute myelogenous leukemia (AML);
- reducing the duration of neutropenia and neutropenia-related clinical sequelae (e.g., febrile neutropenia) in patients with non-myeloid malignancies undergoing myeloablative chemotherapy followed by marrow transplantation;
- mobilizing hematopoietic progenitor cells into the peripheral blood for collection by leukapheresis;
- chronic administration to reduce the incidence and duration of sequelae of neutropenia (e.g., fever, infections, and oropharyngeal ulcers) in symptomatic patients with congenital neutropenia, cyclic neutropenia, or idiopathic neutropenia; and
- increase survival in patients acutely exposed to myelosuppressive doses of radiation.

#### Tbo-filgrastim (Granix)

Tbo-filgrastim is a protein identical to filgrastim, but with slightly different concentrations of excipients. Its pharmacokinetics and neutrophil response are identical to filgrastim in clinical trials in chemotherapy, stem cell mobilization, and stem cell transplantation. Although it has been regarded as a "bio-similar,"41 it has gone through the full FDA approval process just like filgrastim. It is available as 300 and 480 mcg prefilled syringes and used just like filgrastim. It is FDA approved only for reducing the duration of severe neutropenia in patients with nonmyeloid malignancies receiving myelosuppressive anticancer drugs associated with a clinically significant incidence of febrile neutropenia.

#### Pegfilgrastim (Neulasta)

Pegfilgrastim is a longer acting form of filgrastim produced by adding a 20,000 Da polyethylene glycol (PEG) moiety to the amino-terminus of the filgrastim protein. It has a MW of 39,000 Da and is available only as a 6 mg prefilled syringe that is usually administered every two weeks. Its action is prolonged because it is cleared mostly by neutrophils, not the kidney. Its T1/2 of 33 hours (varies from 15 to 80 hours depending upon the ANC) is at least 10 times longer than that of filgrastim. It is usually given one day after the end of the chemotherapy cycle and is not to be given in the period 14 days before to 24 hours after chemotherapy. Its major attribute is that of convenience; one 6 mg pegfilgrastim dose every two weeks is equivalent to 10–14 daily doses of filgrastim.42 It is FDA approved only to decrease the incidence of infection, as manifested by febrile neutropenia, in patients with nonmyeloid malignancies receiving myelosuppressive anticancer drugs associated with a clinically significant incidence of febrile neutropenia.
Sargramostim (Leukine)
Sargramostim has a structure identical to that of native GM-CSF except that it has a leucine at amino acid 123. This glycosylated protein is prepared in yeast and has three different molecular forms of 19,500, 16,800, and 15,500 Da. It is available in vials of 250 and 500 mcg with a chemotherapy dose of 250 mcg/m2/day. It is more effective when given subcutaneously than intravenously and is usually given 24 hours after the end of the chemotherapy and not in the 24 hours before chemotherapy starts. It is recommended that sargramostim be stopped after chemotherapy when the ANC is >10,000, but many stop at lower ANCs. It is FDA approved for:

- use following induction chemotherapy in older adult patients with AML to shorten time to neutrophil recovery and to reduce the incidence of severe and life-threatening infections and infections resulting in death;
- mobilization of hematopoietic progenitor cells into peripheral blood for collection by leukapheresis. Myeloid reconstitution is further accelerated by administration of sargramostim following peripheral blood progenitor cell transplantation;
- acceleration of myeloid recovery in patients with non-Hodgkin’s lymphoma (NHL), acute lymphoblastic leukemia (ALL), and Hodgkin’s disease undergoing autologous bone marrow transplantation (BMT);
- acceleration of myeloid recovery in patients undergoing allogeneic BMT from HLA-matched related donors; and
- patients who have undergone allogeneic or autologous bone marrow transplantation (BMT) in whom engraftment is delayed or has failed.

Effects and adverse effects of G-CSF and GM-CSF administration
The neutrophil response to the filgrastims and sargramostim in healthy volunteers are comparable in some ways:

- **15–30 minutes**: Neutrophils decrease modestly and then return to baseline, probably due to transient sequestration/margination.
- **1–36 hours**: The neutrophils gradually rise due to de-margination and release from bone marrow stores.
- **>36 hours**: Increased production of neutrophils.

But, in other ways, the filgrastims and sargramostim differ greatly with much of the white blood cell (WBC) response of the latter coming from increases in eosinophils. Most of the GM-CSF effect on neutrophils is to increase their survival, but not their production (Table 35.4).

The side effects of the myeloid growth factors are listed in Table 35.5. The three G-CSFs are comparable in terms of their side effects, but GM-CSF has a somewhat expanded repertoire. These commonly include myalgias, but capillary leak syndrome and a first-dose phenomenon of hypotension, tachycardia, and dyspnea may be seen at high doses of GM-CSF in the transplant setting. If myeloid growth factors are administered concurrent with chemotherapy or radiation therapy, subsequent neutropenia is usually worsened.

**Table 35.4 Effects of G-CSF and GM-CSF on neutrophil kinetics**

<table>
<thead>
<tr>
<th>Neutrophil Production</th>
<th>Normal</th>
<th>GM-CSF</th>
<th>G-CSF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum count (×10⁶/mL)</td>
<td>5.2</td>
<td>17.0</td>
<td>35.0</td>
</tr>
<tr>
<td>Appearance in peripheral blood (days)</td>
<td>4–7</td>
<td>4.5–6.5</td>
<td>1–2</td>
</tr>
<tr>
<td>Peripheral t½ (h)</td>
<td>8</td>
<td>48</td>
<td>7.6</td>
</tr>
<tr>
<td>Amplification factor</td>
<td>1</td>
<td>1.5</td>
<td>9.4</td>
</tr>
<tr>
<td>Extra divisions</td>
<td>0</td>
<td>0.6</td>
<td>3.2</td>
</tr>
</tbody>
</table>

Source: Lord et al. (1992). Reproduced with permission of Wiley.

The use of myeloid growth factors in chemotherapy patients may be associated with a small increased risk of treatment-related acute myeloid leukemia or myelodysplasia.44,45

Clinical uses of myeloid growth factors
Table 35.6 lists the uses of myeloid growth factors. Most of these have been studied only with G-CSF, but current National Comprehensive Cancer Network (NCCN) guidelines draw no strong distinction yet between GM-CSF and G-CSF.46

Chemotherapy-induced neutropenia
Filgrastim, tbo-filgrastim, and pegfilgrastim have all shown marked ability to stimulate neutrophil production and to mitigate chemotherapy-induced neutropenia. In small cell lung cancer patients undergoing chemotherapy,47 neutropenic fever for all chemotherapy cycles was decreased from 77% with placebo to 40% (p < 0.001) with filgrastim; hospital days were shortened from 4.2 to 2.3 for all cycles. Antibiotic use and days with an ANC <500 were also markedly decreased. NCCN guidelines46 suggest using myeloid growth factors for primary prophylaxis of febrile neutropenia when the expected incidence of such is greater than 20%. Additionally, myeloid growth factors can be considered for primary prophylaxis for patients receiving chemotherapy where

**Table 35.5 Adverse effects of myeloid growth factors**

<table>
<thead>
<tr>
<th>Effect</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bone pain</td>
<td>(15–39% on G-CSF vs. 0–21% on placebo)</td>
</tr>
<tr>
<td>Exacerbation of preexisting inflammatory conditions (eczema, psoriasis, and vasculitis)</td>
<td></td>
</tr>
<tr>
<td>Allergic reactions at injection sites (rare)</td>
<td></td>
</tr>
<tr>
<td>Sweet's syndrome (acute, febrile neutrophilic dermatosis)</td>
<td></td>
</tr>
<tr>
<td>Antibody formation (none are neutralizing)</td>
<td></td>
</tr>
<tr>
<td>Splenic rupture, acute respiratory distress syndrome, and precipitate sickle cell crisis (all rare)</td>
<td></td>
</tr>
<tr>
<td>Capillary leak syndrome</td>
<td></td>
</tr>
<tr>
<td>Alveolar hemorrhage and hemoptysis</td>
<td></td>
</tr>
<tr>
<td>Risk MDS and AML in patients receiving chemotherapy or with congenital neutropenia</td>
<td></td>
</tr>
<tr>
<td>↑ LDH, uric acid, LAP; ↓ cholesterol</td>
<td></td>
</tr>
<tr>
<td>Myalgias, fever</td>
<td></td>
</tr>
</tbody>
</table>

“First dose phenomenon”: hypotension, tachycardia, and dyspnea due to transient pulmonary leukocyte sequestration (very high doses)

Italics denote effects seen only with GM-CSF.

**Table 35.6 Clinical Uses of myeloid growth factors**

<table>
<thead>
<tr>
<th>Use</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary prophylaxis of FN if incidence &gt;20%</td>
<td></td>
</tr>
<tr>
<td>Primary prophylaxis considered if FN incidence &gt;10–20% (usually “high-risk” patients)</td>
<td></td>
</tr>
<tr>
<td>Primary prophylaxis if FN incidence &lt;10% (rarely)</td>
<td></td>
</tr>
<tr>
<td>Secondary prophylaxis of FN to keep dose intensity</td>
<td></td>
</tr>
<tr>
<td>All high-risk chemotherapy patients admitted with FN</td>
<td></td>
</tr>
<tr>
<td>After allogeneic or autologous stem cell txp</td>
<td></td>
</tr>
<tr>
<td>PBPC mobilization</td>
<td></td>
</tr>
<tr>
<td>MDS with neutropenia and recurrent infection</td>
<td></td>
</tr>
<tr>
<td>AML induction chemotherapy (≥35 years old)</td>
<td></td>
</tr>
<tr>
<td>Severe chronic neutropenia</td>
<td></td>
</tr>
<tr>
<td>(Drug-induced neutropenia—low dose [0.4–5 μg/kg])</td>
<td></td>
</tr>
<tr>
<td>(Aplastic anemia)</td>
<td></td>
</tr>
<tr>
<td>(Hairy cell leukemia)</td>
<td></td>
</tr>
<tr>
<td>(Acute lymphoblastic leukemia)</td>
<td></td>
</tr>
<tr>
<td>(Agranulocytosis)</td>
<td></td>
</tr>
</tbody>
</table>

FN, febrile neutropenia; PBPC, peripheral blood progenitor cells; MDS, myelodysplastic syndrome; AML, acute myeloid leukemia. Uses in parentheses denote lack of FDA approval.
febrile neutropenia risk is 10–20% if patients are high risk (e.g., age >65, prior chemotherapy or radiation therapy, preexisting neutropenia or bone marrow involvement or infection, poor performance status, and/or HIV infection). Phylactic myeloid growth factors are rarely considered for chemotherapy patients where the rate of febrile neutropenia is expected to be <10%.

In chemotherapy patients experiencing febrile neutropenia in a chemotherapy cycle not supported by myeloid growth factor, subsequent use of myeloid growth factor (secondary prophylaxis) is recommended to maintain dose intensity.

Unless considered high risk, myeloid growth factor treatment is not indicated for chemotherapy patients (not already on myeloid growth factor therapy) who develop severe neutropenia without fever or those currently hospitalized with febrile neutropenia.

Severe chronic neutropenia

The clinical benefit of long-term myeloid growth factor administration for patients with severe congenital neutropenia (including Kostmann syndrome), severe idiopathic neutropenia, or cyclic neutropenia has been well established. In a large randomized clinical trial of patients with severe chronic neutropenia, the occurrence of fever, oropharyngeal ulcers, infections, hospitalization, and antibiotic use were all significantly reduced with filgrastim treatment. The quality of life and activity profiles of patients also improved. The only complication of therapy was the increased risk of developing myelodysplasia and acute leukemia in patients with severe congenital neutropenia. This risk appears to be disease specific with substantially lower or no risk for patients with cyclic or idiopathic neutropenia. Those with severe congenital neutropenia need to be monitored with regular white counts, clinical observation, and possibly bone marrow examinations.

Mobilization of peripheral blood progenitor cells

Four to six days after administration of G-CSF, peripheral blood progenitor cells (PBSCs) increase as much as 100-fold. This has allowed for the marked improvement of collection of such cells in patients or donors for stem cell transplant. The basic physiologic principles governing mobilization of PBSCs probably involve breakage of molecular bonds between the adhesive marrow elements, that is, CXCR-4 on the progenitors and stromal-cell-derived factor 1 (SDF-1) on the marrow stromal cells. Administration of G-CSF expands myeloid and progenitor mass, and there is good evidence that the proteases released from neutrophils disrupt key adhesive bonds holding the progenitors in the marrow and allow their mobilization from marrow spaces.

Implications for transfusion medicine

The use of myeloid growth factors to mobilize peripheral blood progenitor cells is a standard transfusion medicine practice. They can also be used to ameliorate drug-induced neutropenia such as that occurring after the administration of IVIG. Widespread use of myeloid growth factors in oncology may reduce the need for hospitalization and antibiotics. Unfortunately, their use to provide neutrophils for transfusion has not met with much success. Indeed, a recent heroic effort was unable to show a significant effect of neutrophil transfusion on the outcomes of infected, neutropenic patients.

Thrombopoietic growth factors

Although Sir James Homer Wright described how bone marrow megakaryocytes produced platelets in 1902, it was not until 1958 that Kelemen proposed that a “thrombopoietin” regulated this process. This last of the major hematopoietic growth factors was not purified until 1994. Initial clinical studies with two types of recombinant TPO (recombinant human TPO [rhTPO] and pegylated recombinant human megakaryocyte growth and development factor [PEG-rhMGDF]) showed they were quite effective in raising the platelet count in healthy subjects and increasing platelet apheresis yields. They raised the platelet count nadirs and decreased platelet transfusions after non-myoeloblastic chemotherapy, but had minimal effect on the recovery of platelets to >20,000 or need for platelet transfusions in patients undergoing leukemia induction chemotherapy or stem cell transplantation. There was a modest platelet count increase when given to myelodysplastic syndrome (MDS) patients. Unfortunately, these studies were terminated when antibodies developed against one of these recombinant molecules (PEG-rhMGDF) that cross-reacted with native TPO and produced thrombocytopenia. Subsequently, two TPO receptor agonists, romiplostim and eltrombopag, were not antigenic and proved to be potent stimulators of platelet production. Both were FDA approved in 2008 for the treatment of immune thrombocytopenia (ITP).

Structure, function, and physiology

TPO is encoded by a single gene (3q27.1) that produces a 332-amino-acid (MW = 95,000 Da) glycoprotein of which the first 153 amino acids are 23% homologous with EPO, and probably 50% similar if conservative amino acid substitutions are considered. This region also contains four cysteine residues like those in EPO and is responsible for binding to the TPO receptor. But, despite these similarities, TPO does not bind the EPO receptor and EPO does not bind the TPO receptor. The rest of the molecule is rich in carbohydrates that increase its half-life.

TPO is the key regulator of platelet production. In mice deficient in both genes for either TPO or the TPO receptor, the platelet count, bone marrow megakaryocytes, and megakaryocyte precursors are 10–15% of normal. These animals also have reduced levels of erythroid and myeloid precursors but with no subsequent anemia or neutropenia. TPO is made primarily in the liver. Whether its basal rate of production is regulated remains unclear. Multiple studies of animals with ITP or after chemotherapy have shown no increase in the hepatic TPO mRNA levels. However, in a recent study, hepatocytes were found to increase TPO mRNA production after desialylated platelets bound to the Ashwell–Morell receptor. How this affects normal physiology remains unclear at the present time.

TPO levels are inversely related to the rate of platelet production. Once in the circulation, TPO is bound and cleaved by high-affinity platelet TPO receptors leaving a small basal amount of TPO. This primitive normal feedback loop is shared with other hematopoietic growth factors such as M-CSF and G-CSF; there appears to be no specific sensor of the platelet count in the body. In aplastic anemia patients, TPO levels are >2000 pg/mL (normal: 7–99 pg/mL). In animals or humans transfused to platelet counts above normal, TPO is suppressed below basal levels. In patients with hepatic cirrhosis, TPO production is reduced.

TPO has no carrier molecule and works by binding to TPO receptors on target hematopoietic cells. TPO is necessary for the viability of stem cells; humans born without it are thrombocytopenic at birth and eventually become pancytopenic. TPO is necessary for the viability of precursors of all lineages (Figure 35.1) but only amplifies the megakaryocyte lineage by promoting the mitosis of megakaryocyte colony-forming cells, increasing the rate of
megakaryocyte endomitosis and maturation, and thereby increasing platelet production (Figure 35.4).

**Clinically available thrombopoietic growth factors**

**Romiplostim (Nplate)**

Romiplostim is a TPO receptor agonist composed of an IgG1 heavy chain carrier molecule into which have been inserted four identical 14 amino acid peptides that activate the TPO receptor. This peptide (IEGPTLRQWLAAA) has no sequence homology with TPO but was found to bind and activate the TPO receptor. If dimerized, it was as potent as thrombopoietin in vitro but given its short half-life had minimal activity in vivo. By inserting this peptide into the IgG carrier construct, romiplostim has a T1/2 of ~120 h, three times longer than native TPO. Although romiplostim binds to the TPO receptor with an affinity 25% of that of TPO, it is a very potent activator of platelet production. Romiplostim is available as vials containing 250 or 500 mcg of lyophilized drug. It is administered as a subcutaneous injection of 1–10 mcg/kg once a week for the treatment of ITP. There is no effect upon white cell or red cell production. Romiplostim is currently FDA approved only for the treatment of thrombocytopenia in patients with chronic ITP who have had an insufficient response to corticosteroids, immunoglobulins, or splenectomy.

**Eltrombopag (Promacta, Revolade)**

Efforts to identify small molecules that bound and activated the TPO receptor successfully identified a number of compounds. One was subsequently modified to enhance its pharmacological and biological properties, resulting in eltrombopag. Eltrombopag binds the TPO receptor at a transmembrane site distant from where TPO binds. It thereby activates the TPO receptor differently than TPO or romiplostim: JAK and STAT phosphorylation are less, and there is no effect upon the AKT pathway. Nonetheless, eltrombopag increases megakaryocyte growth and maturation to increase platelet production.

Eltrombopag is available as 12.5, 25, 50, 75, or 100 mg tablets. Eltrombopag is primarily metabolized by the liver, and dose adjustments are necessary for patients with reduced metabolism of this drug due to East Asian ancestry or liver dysfunction. It is usually taken orally once a day and distant from calcium-containing compounds or food stuffs that would neutralize its activity if co-administered. Eltrombopag is currently FDA approved for the treatment of:

- thrombocytopenia in adult and pediatric patients 1 year and older with chronic ITP who have had an insufficient response to corticosteroids, immunoglobulins, or splenectomy;
- thrombocytopenia in patients with chronic hepatitis C to allow the initiation and maintenance of interferon-based therapy; and
- patients with severe aplastic anemia who have had an insufficient response to immunosuppressive therapy.

**Effects and adverse effects of thrombopoietin administration**

After giving a single dose of romiplostim or 10 daily doses of eltrombopag to healthy volunteers, platelet counts begin to rise on day 5, peak at days 10–15, and return to baseline by day 28. Eltrombopag is probably one-eighth as potent as romiplostim:

---

![Figure 35.4](image-url) **Figure 35.4** Scheme of megakaryocyte maturation from stem cell to mature platelet. Stem cells and megakaryocyte colony-forming cells (Meg-CFCs) undergo mitosis, but at some stage they stop mitosis and undergo endomitosis during which nuclear and cytoplasmic division do not occur, giving rise to polyploid early megakaryocytes that contain 2–16 times the normal diploid (2N) amount of DNA. The early megakaryocytes then stop endomitosis and mature into morphologically identifiable megakaryocytes and then shed platelets. TPO plays a major role in all but the last process. Source: D. Kuter. Reproduced with permission.
at the maximum dose of 75 mg/d, eltrombopag increases the platelet count by about 150,000 above baseline compared with a rise of 1,400,000 for romiplostim. Nonetheless, this difference in potency has not translated into any different response rate for eltrombopag compared to romiplostim in clinical trials. With continued administration of either molecule, peak platelet counts can be maintained indefinitely with no effect on the RBCs or WBCs. The platelets so produced have normal function.

The TPO receptor agonists have not demonstrated any tachyphylaxis or significant long-term complications.23,24 The potential and real adverse effects are listed in Table 35.7. A few deserve additional comment.

**Thrombosis**

Although both TPO receptor agonists can significantly increase the platelet count, thrombosis has not been clearly associated with either in placebo-controlled ITP studies. Rather, what has been uncovered is that ITP is itself a prothrombotic disorder25 whose rate of thrombosis does not appear to be exacerbated by either agent. Compared with placebo treatment, thrombosis was not increased by recombinant TPO treatment in earlier trials in cancer patients. Platelet function studies have shown no increase in platelet activation with either agent despite the finding that romiplostim (but not eltrombopag) reduces the activation threshold for some agonists (eg, ADP) by about 50% in platelet aggregation tests.

**Tumor progression and cancer mortality**

Unlike the controversy over erythroid growth factors, solid tumor cells lack the TPO receptor.76 Furthermore, in cancer chemotherapy studies with recombinant thrombopoietins in the 1990s, there is no effect on tumor progression or survival.58

**Antibody formation**

Although antibody formation ended the development of the recombinant TPO molecules, no clinically relevant antibodies have been developed against the TPO receptor agonists. Three patients have developed non-neutralizing antibody against romiplostim, and none have developed antibodies against eltrombopag.

**Hepatic toxicity**

Eltrombopag is metabolized by the liver, and in one ITP study 13% of patients developed abnormal liver function tests. In general, these were mild and reversible, and did not often require cessation of medication. Intermittent monitoring of liver function tests is recommended for eltrombopag.

**Rebound thrombocytopenia**

The major concern with TPO receptor agonists occurs in ITP patients in whom the drug is abruptly stopped. Current prescribing information is incorrect and states that either agent be stopped when the platelet count exceeds 400,000. Unfortunately, in 10–20% of such situations, stopping the drug results in a rebound thrombocytopenia with the platelet count rapidly dropping below prior baseline values 7–10 days later and a markedly increased bleeding risk.62 Such patients are better treated by a gradual dose reduction over several weeks.

**Reticulin fibrosis**

Reticulin is a normal component of the bone marrow and may increase in patients with ITP77 and in those receiving TPO receptor agonists.78 This reticulin is generally mild, reversible, and clinically silent, and does not portend progression to the disease myelofibrosis. Recent prospective bone marrow studies of ITP patients receiving TPO receptor agonists have shown infrequent increases in reticulin over three years of treatment.79

### Clinical use of thrombopoietic growth factors

Table 35.8 provides a complete list of conditions that have been treated with thrombopoietic growth factors.

**Immune thrombocytopenia**

ITP is a disease of increased platelet destruction and inappropriately low platelet production.80 Although standard therapies such as immunosuppression, rituximab, and splenectomy decrease platelet destruction, thrombopoietic growth factors increase platelet production, thereby mitigating thrombocytopenia. Both TPO receptor agonists increase the platelet count >50,000 in more than 85% of patients, accompanied by reduced bleeding and need for other therapies.81,82 Long-term use of both molecules is successful73,74 and may be associated with increased numbers of T-regulatory lymphocytes and occasional disease remission.

**Hepatitis C–related thrombocytopenia**

Hepatitis C produces two forms of thrombocytopenia that may be difficult to distinguish in any one patient. One is an ITP-like condition that responds to ITP therapies; the second is TPO deficiency due to hepatic injury. Nonetheless, in patients with platelet counts <70,000, antiviral therapy with ribavirin and interferon may be contraindicated. When such patients with a mean baseline platelet count of 55,000 were treated with eltrombopag 75 mg/d for four weeks, the mean platelet count rose to 209,000 compared with 54,000 in those receiving placebo.83 With continued eltrombopag support, 65% of patients could finish antiviral treatment compared with 6% receiving placebo. In a large phase III study, hepatitis C patients with platelet counts under 100,000 were all treated with eltrombopag and 95% increased their platelet count to over 100,000 by week 9. They were then randomized to continue eltrombopag or receive placebo during the next 24–48 weeks of

### Table 35.7 Adverse effects of thrombopoietic growth factors

<table>
<thead>
<tr>
<th>Condition</th>
<th>Risk Factor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thrombocytosis</td>
<td>Increased bone marrow reticulin</td>
</tr>
<tr>
<td>Thrombosis</td>
<td>Stimulation of leukemic blasts</td>
</tr>
<tr>
<td>Elevated levels of reticulin</td>
<td>Reduction in threshold for platelet activation</td>
</tr>
<tr>
<td>Elevation of thrombocytopenia</td>
<td>Rebound worsening of thrombocytopenia with</td>
</tr>
<tr>
<td></td>
<td>discontinuation in ITP patients</td>
</tr>
</tbody>
</table>

### Table 35.8 Clinical uses of thrombopoietic growth factors

<table>
<thead>
<tr>
<th>Condition</th>
<th>Risk Factor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chronic ITP</td>
<td>(Liver failure)</td>
</tr>
<tr>
<td>Hepatitis C–related thrombocytopenia</td>
<td>(Liver failure)</td>
</tr>
<tr>
<td>Aplastic anemia</td>
<td>(Myosin heavy chain 9–related disease)</td>
</tr>
<tr>
<td>Pediatric ITP</td>
<td>(Stem cell mobilization)</td>
</tr>
<tr>
<td>(ITP initial therapy)</td>
<td>(Stem cell transplant, failed engraftment)</td>
</tr>
<tr>
<td>(Chemotherapy–induced thrombocytopenia)</td>
<td>(Acute leukemia)</td>
</tr>
<tr>
<td>(Presurgical thrombocytopenia)</td>
<td>(Platelet apheresis)</td>
</tr>
<tr>
<td></td>
<td>(Mylodysplastic syndromes)</td>
</tr>
</tbody>
</table>

Uses in parentheses denote lack of FDA approval.
antiviral treatment.84 Sustained virologic response was seen in 19–23% of eltrombopag patients compared with 13–15% on placebo (p < 0.02), with more patients maintaining a platelet count >50,000 (69–81% vs. 15–23%, respectively).

**Aplastic anemia**

Although TPO levels are markedly elevated (>2000 pg/mL) in most aplastic anemia patients, eltrombopag produced a platelet count rise in 9/25 patients with a trilineage response in 6/25 patients.85 In responders, marrow cellularity increased and response was maintained long-term with some coming off treatment.86 These striking results were not seen in prior studies with recombinant TPO58 and suggest that eltrombopag may not be working through the TPO receptor.

**Presurgical treatment of thrombocytopenic patients**

Thrombocytopenia complicates many illnesses, and such patients are often given aggressive platelet transfusions or even refused needed surgical procedures. Several studies have shown that romiplostim and eltrombopag can increase platelet counts in such patients, reduce bleeding, and allow procedures to be performed. In one study of 17 thrombocytopenic patients treated with romiplostim, all were able to raise their platelet count and undergo surgery with no excessive bleeding and with minimal need for transfusion.87 In a second study of thrombocytopenic cirrhotic patients being prepped for liver biopsy, platelet transfusion was avoided in 72% of patients receiving eltrombopag compared with 19% of those receiving placebo (p < 0.001); there was no difference in bleeding (17% vs. 19%, respectively).88 However, this study was terminated early because 6/145 eltrombopag patients had portal vein clot versus 1/147 on placebo. The study was weakened by the absence of pretreatment assessment for portal vein clot.

**Chemotherapy-induced thrombocytopenia**

This remains a major challenge, and there are as yet no completed studies with the TPO receptor agonists. Early studies in patients receiving non-myeloablative chemotherapy for ovarian cancer89 or lung cancer90 showed that treatment with rhTPO and PEG-rhMGDF increased the nadir platelet count, decreased the need for platelet transfusions, and allowed chemotherapy to be given on time.88 However, neither TPO showed benefit in patients receiving myeloablative chemotherapy for acute leukemia induction or for stem cell transplant.

**Myelodysplastic syndrome**

Thrombocytopenia commonly complicates the care of patients with MDS. In a study of thrombocytopenic (<20,000 or history of bleeding and platelets ≥20,000) low-risk/intermediate-1-risk MDS patients, romiplostim or placebo was administered for 58 weeks. Overall, clinically important bleeding events were not significantly reduced with romiplostim (HR, 0.83; 95% CI, 0.66–1.05; p = 0.13), but in those with platelet counts ≥20,000 significant reductions were seen (HR, 0.34; 95% CI, 0.20–0.58; p < 0.0001). Romiplostim reduced bleeding events (RR, 0.92) and platelet transfusions (RR, 0.77) and increased platelet response (OR, 15.6). This study was stopped at an interim analysis because of a perceived increase in AML rate (HR, 2.51) with romiplostim, but final analysis showed AML rates of 6% with romiplostim and 4.9% placebo (HR, 1.20; 95% CI, 0.38–3.84) and similar survival rates.

**Other thrombopoietic growth factors**

**Interleukin-11 (Oprelvekin, Neumega)**

IL11 is not a normal regulator of platelet production.91 In mice in which this gene is deleted, all blood counts are normal.92 But administration of IL11 does increase the platelet count93 and has been found to reduce the transfusion rate from 96% to 70% in patients who had experienced thrombocytopenia in a prior chemotherapy cycle.94 Oprelvekin is FDA approved for the prevention of severe thrombocytopenia and the reduction of the need for platelet transfusions following myelosuppressive chemotherapy in adult patients with nonmyeloid malignancies who are at high risk of severe thrombocytopenia. It is not indicated following myeloablative chemotherapy. Unfortunately, orelvekin has a fairly large number of side effects that make it unwise to recommend for any patient.

**Implications for transfusion medicine**

This area is still early in its development. TPO receptor agonists clearly decease the use of IVIG and platelet transfusions in ITP patients.81 They can also reduce or eliminate the need for transfusions in thrombocytopenic patients undergoing major surgery. They have helped a number of Jehovah’s Witness patients or those with severe platelet alloimmunization to undergo surgery without platelet transfusion.87

The bigger issue is whether TPO receptor agonists can decrease the thrombocytopenia associated with chemotherapy and reduce the need for platelet transfusions. Early data showed that rhTPO reduced the rate of platelet transfusion from 75% to 25% in women being treated for ovarian cancer.89 It is anticipated that studies with the current TPO receptor agonists will show similar benefit.95 Whether TPO receptor agonists will ever play a role in stimulating platelet apheresis donors remains unclear. Prior studies showed that PEG-rhMGDF increased the median platelet count from 248,000 to 602,000, with an increase in median (range) apheresis yield from 3.8 × 10^11 (1.3 × 10^11 – 7.9 × 10^11) to 11.0 × 10^11 (7.1 × 10^11 – 18.3 × 10^11) platelets.59 Once transfused, these products were hemostatically active and provided a dose-dependent rise in platelet count and increased transfusion-free interval.90 Such rhTPO mobilized platelets have also been frozen and later transfused into alloimmunized chemotherapy patients.80

**General conclusions**

The hematopoietic growth factors have markedly affected the practice of medicine over the past 30 years. Beginning with erythropoietin and then extended with myeloid growth factors and now with thrombopoietic growth factors, there is an ability to increase specific blood cell production in patients who have anemia, neutropenia, or thrombocytopenia. These molecules are generally safe and effective and with adequate time will raise the blood count. Unfortunately, none is immediately active, and most take days for the onset of activity. None of these will replace the need for transfusion in acute situations of anemia or thrombocytopenia. However, all three offer the opportunity to prevent subsequent transfusion therapy or infection. In some settings, they have been demonstrated to reduce medical care costs.
Key references
A full reference list for this chapter is available at: http://www.wiley.com/go/simon/transfusion