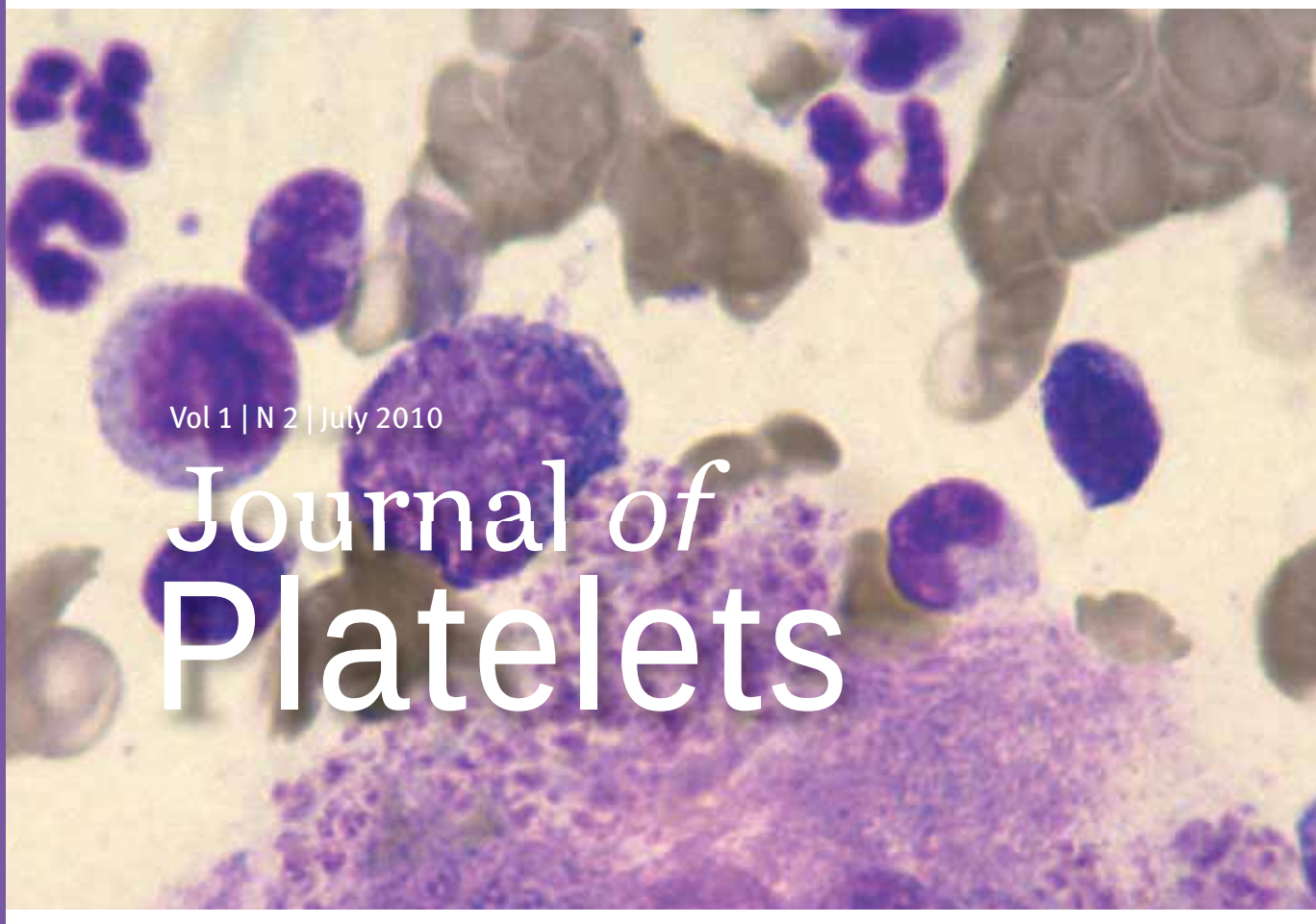

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Welcome to the second issue of the Journal of Platelets. In the same vein as the inaugural issue, the articles in this second issue continue to offer a broad and multidisciplinary perspective on platelets and platelets disorders.

In this issue, we provide excellent articles that cover highly valuable information and scientific data to not only enhance our existing knowledge of platelet disorders, but we are also provided with useful guidelines and updates to enhance the treatment practices that practitioners among us use to manage and treat these disorders.

Following on from the excellent article in the inaugural issue, which discussed the standardization of definitions and terminology in immune thrombocytopenia (ITP),[Rodeghiero F. and Ruggeri M., 2010] Prof. Rodeghiero opens the second issue with a critical appraisal of a new international consensus report on the diagnosis and management of primary ITP.[Rodeghiero F., 2010]

The association between clonal lymphoid expansions and ITP, whether subclinically or overtly malignant, represents an interesting model for further evaluating the pathophysiology of ITP.[Nassi L. and Gaidano G., 2010] In addition, the ability to identify clonal lymphoid expansions may help to provide information that can potentially be used in tailoring individual patient therapy that primarily targets specific cellular subsets.

Thrombocytopenia is a common hematological condition in patients with chronic hepatitis C virus (HCV) infection, and several mutually exclusive pathophysiological mechanisms are known to contribute to its development.[Gianni E.G. and Savarino V., 2010] These mechanisms include, primarily, portal hypertension and decreased hepatic thrombopoietin synthesis, although bone marrow suppression, and viral- or antiviral therapy-related immune-mediated mechanisms are also implicated in some patients.

Several important case reports are presented in this issue, covering the necessity to perform a careful and thorough differential diagnosis for ITP, in order to avoid inappropriate or unnecessary treatment;[Veneri D. *et al*., 2010] a demonstration of how persistent thrombocytopenia can negatively impact on the treatment of a chemosensitive disease, highlighting the case of a patient with Ewing's sarcoma;[Colombi F. *et al*., 2010] and an analysis of the economic burden of chronic ITP in Italy.[Rodeghiero F. and Gerzeli S., 2010]

This issue concludes with exciting reports, focussing specifically on new developments in ITP, from the recent annual meetings of the European Association for the Study of the Liver (EASL)[Giannini E.G., 2010], held in Vienna, the European Hematology Association (EHA)[Ruggeri M., 2010] meeting, held in Barcelona, and the American Society of Clinical Oncology (ASCO), held in Chicago.[Rossi V. and Gallo S., 2010]

Professor Michele Baccarani

A new international consensus report on the diagnosis and management of primary immune thrombocytopenia: a critical appraisal

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Abstract

Previously available guidelines for the diagnosis and treatment of primary immune thrombocytopenia (ITP) have recently been updated and published by a consensus panel of international clinicians with expertise in the area. The new international consensus report (ICR) considers new data and provides consensus-based recommendations relating to the diagnosis and treatment of ITP in adults, children and during pregnancy. The aim of this current article is to provide an overview of the key aspects of the ICR, as they pertain specifically to adults, and to provide a critical appraisal of the potential impact that these new guidelines will have on clinical practice.

Introduction

Until very recently, few randomized clinical trials have been conducted in immune thrombocytopenia (ITP) and most current clinical decisions are still not evidence-based. In this regard, evidence- and/or consensus-based international or national guidelines may have a positive impact. Such documents are intended to help the practising physician to utilize the different grades of evidence for specific diagnostic or treatment options. In those cases where there is insufficient evidence to dictate the most appropriate intervention, clinicians may base their choice of individual patient treatment on the general recommendations proposed by "experts" for similar clinical situations. To date, two main guidelines are available: the 1996 American Society of Hematology guidelines (ASH-GL),[1] and the 2003 British Committee for Standards in Haematology guidelines (BCSH-GL).[2]

Recently, an international panel of 20 members with recognized clinical and/or scientific experience in ITP proposed an updated systematic review of the literature and provided consensus-based guidance for the diagnosis and treatment of ITP.[3] This international consensus report (ICR) is restricted to primary ITP and builds upon ASH-GL and BCSH-GL, taking into account recent major progress in the field.

Indeed, during the last decade we have witnessed the availability of new treatments for ITP, including the use of anti CD-20 monoclonal antibodies and the introduction of the second generation thrombopoietin (TPO) receptor agonists, purposely developed for the correction of thrombocytopenia. This progress has been made possible through an improved understanding of the pathophysiology of ITP, currently recognized as being due to both increased peripheral destruction and suboptimal platelet production.[4]

Furthermore, the etiological and pathogenetic role of *Helicobacter pylori* infection has also been consolidated by several prospective studies, along with effective therapy that eradicates *H. pylori* infection and induces sustained responses in about one third of cases with clinically relevant ITP.[5]

Finally, recognizing the confusion with terminology that plagues this field, an international working group (IWG) recently proposed new terminology and more clinically meaningful and homogeneous response criteria that are becoming widely adopted.[6,7] The ICR has adopted these proposals. Accordingly, the ICR classifies primary ITP as an acquired immune-mediated disorder characterized by isolated thrombocytopenia, defined as a peripheral blood platelet count less than 100 x 10^9/L, and by the absence of any obvious initiating and/or underlying cause of the thrombocytopenia. In addition, whenever applicable, all of the new consensus definitions of the IWG for the different phases of ITP have also been adopted.

The aim of this current overview is to offer a critical appraisal of the incorporation by the ICR of the above

Table 1. Recommendations for the diagnosis of ITP in adults (and children).[3] Tests indicated in italics were considered differently, or not included, in previous ASH-GL. Reproduced with permission[3]

Basic evaluation	Tests of potential utility in the management of an ITP patient	Tests of unproven or uncertain benefit
Patient history	Glycoprotein-specific antibody	TPO
Family history	Antiphospholipid antibodies (including anticardiolipin and lupus anticoagulant)	Reticulated platelets
Physical examination	Antithyroid antibodies and thyroid function	PaIgG
Complete blood count and *reticulocyte count*	*Pregnancy test in women of childbearing potential*	Platelet survival study
Peripheral blood film	*Antinuclear antibodies*	Bleeding time
*Quantitative immunoglobulin level measurement**	*Viral PCR for parvovirus and CMV*	Serum complement
Bone marrow examination (in selected patients; refer to text)		
Blood group (Rh)		
Direct antiglobulin test		
H pylori†		
HIV†		
HCV†		

Refer also to supplemental Document 2 in the electronic appendix.[3]

Rh indicates rhesus; *H pylori, Helicobacter pylori;* HIV, human immunodeficiency virus; HCV, hepatitis C virus; PCR, polymerase chain reaction; CMV, cytomegalovirus; TPO, thrombopoietin; and PaIgG, platelet-associated immunoglobulin G.

* Quantitative immunoglobulin level measurement should be considered in children with ITP and is recommended in those children with persistent or chronic ITP as part of the reassessment evaluation.

† Recommended by the majority of the panel for adult patients regardless of geographic locale.

mentioned novelties and to try to highlight the impact that these new guidelines may have in clinical practice. The original 20-page printed version of the manuscript[3] covers a wide spectrum of diagnosis and treatment in children, adults, and during pregnancy, and includes 10 tables and 207 references, most of them entirely new. Unfortunately, due to space limitations imposed by the publisher, valuable information including several "recommendation boxes" are available only through the online version of the article. The present discussion is limited to the adult population.

Diagnosis of primary ITP

Increased awareness of the many secondary forms of ITP, and of their differing natural history and clinical relevance,[8] prompted the panel to carry out an extensive literature review; unfortunately, there is no available improvement in the definition of a positive diagnosis for primary ITP. Therefore, in line with previous guidelines, the ICR reaffirms that the diagnosis of primary ITP remains one of exclusion on the basis of detailed personal and familial history, physical examination, complete blood count, reticulocyte count, peripheral blood film examination and a few selected laboratory tests, and that there is no "gold standard" test that can reliably establish the diagnosis. At variance with previous guidelines, a few specific blood tests were considered mandatory for the basic evaluation of, or of potential utility for, the management of primary ITP (Table 1). The inclusion or exclusion of specific tests appears to be somewhat disputable: e.g., inclusion of a direct antiglobulin test, or routine HIV, or viral PCR for parvovirus, or exclusion of an HVB investigation. The contentious issue of bone marrow examination mirrors ASH and BCSH guidelines.

It should be noted that some of the tests listed in Table 1 are grouped differently to those in ASH-GL, making a comparison difficult. In fact, the connotation for lack

Table 2. Recommendations for first-line treatment (initial treatment for newly diagnosed patients). Taken from supplemental Document 8, Recommendation box 3 of the online version, and reproduced with permission[3]

Corticosteroids (prednisone 0.5–2 mg/kg/day) is the standard first-line treatment for adults with ITP who need treatment and do not have a relative contraindication to its use (e.g. diabetes, psychiatric disorders). Prednisone is continued at full dose for 10–28 days then tapered.
Use of IVIg or IV anti-D may be appropriate in patients with bleeding, at high risk of bleeding, or who are unresponsive to prednisone.
Certain patients may have contradictions to high-dose steroid therapy (e.g. insulin-dependent diabetes) and may be managed with only IVIg or IV anti-D as first-line therapy.

Table 3. Recommendations for second-line therapy in adults – medical. Modified, with permission, from supplemental Document 8, Recommendation box 5 of the online version[3]

TPO-receptor agonists have provided excellent responses in both splenectomized and non-splenectomized patients (Grade A recommendation, Evidence level Ib). Response to TPO-receptor agonists persists for up to 4 years and often allows other ITP therapy to be reduced or discontinued. Cessation of treatment will lead to return of thrombocytopenia in most cases.
Evidence from a systematic review of multiple uncontrolled trials shows a response to a first-generation humanized anti-CD20 monoclonal antibody in over half relapsed/refractory patients. Long-term durable responses occur in 15–20% of patients (Grade B recommendation, Evidence level IIa). Hepatitis B status needs to be determined prior to treatment (Grade C recommendation, Evidence level IV).
Immunosuppressive agents, including mycophenolate mofetil, cyclophosphamide and azathioprine may be used in patients failing other therapies. Danazol and dapsone are 'corticosteroid-sparing' agents that may be particularly useful in elderly patients and in those in whom splenectomy is contraindicated (Grade B recommendation, Evidence level IIa/IIb).
Cyclosporin A (2.5–3 mg/kg/day) increases the platelet count as a single agent or in combination with prednisone. In some patients, the side-effect profile restricts its use (Grade B recommendation).

of consensus is "Tests for which necessity/appropriateness of routine testing is uncertain" or in the case of consensus against testing "Tests that are unnecessary/inappropriate to establish diagnosis of ITP in all patients at presentation". BCSH-GL do not provide a clear-cut distinction of the diagnostic value of the different tests and are vague in considering different scenarios.

As highlighted in table 1, ITP diagnosis is based principally on the exclusion of other causes of isolated thrombocytopenia using patient history, physical examination, blood count and evaluation of the peripheral blood film. If therapy is administered, platelet count should be closely monitored for response as a diagnostic aid. Bone marrow examination is appropriate in patients >60 years old (Evidence level IIb, Grade B recommendation), in those relapsing after remission, in patients not responding to first-line therapy options, and where splenectomy is considered (Evidence level III, Grade C recommendation). This examination should ideally include an aspirate, biopsy, flow cytometry and cytogenetics (Evidence level IV, Grade C recommendation). The detection of *H. pylori* infection, with the urea breath test or the stool antigen test, should be included in the initial work-up of adults in appropriate clinical settings (Evidence level IIa, Grade B recommendation). The majority of authors routinely tested for HIV and HCV in all adult patients (Evidence level IIb). Quantitative Ig level testing is indicated to exclude an immune deficiency syndrome (Evidence level IV, Grade C recommendation), or when treatment with intravenous immunoglobulin is considered.

Who should be treated?

The ICR states that treatment is rarely indicated in patients with platelet counts above 50 x 10^9/L in the absence of the following: bleeding due to platelet dysfunction or another hemostatic defect, trauma, surgery, comorbidities for bleeding, anticoagulation therapy, or in individuals whose profession or lifestyle predisposes them to trauma. Patient preference must also be considered when discussing treatment options. More clear-cut indications were provided in ASH-GL, e.g. withholding treatment in patients with less than 20 x

10^9/L was stated as inappropriate. The ICR offers detailed consensus-based recommendations regarding target platelet counts during surgery in adults, a relevant aspect in clinical practice. In general, compared to BCSH-GL, higher target platelet counts are recommended for dentistry.

First line therapies

No relevant new therapies were found to justify recommending any changes to previous guidelines with regard to first line treatment (corticosteroids, intravenous Ig, anti-D) [Table 2].[3]

Second line therapies

In the printed ICR recommendations,[3] potential second-line treatment options are listed alphabetically, thus not implying a preferred treatment option by the panel. The panel was unable to reach a consensus on the most appropriate, and preferred sequence of, treatment. Therefore, it is up to the treating physician to individualize management to meet patient needs, after discussing the pros and cons of each option with the patient. Treatment options include (in alphabetical order): azathioprine, cyclosporine A, cyclophosphamide, danazol, dapsone, mycophenolate mofetil, a first-generation humanized anti-CD20 monoclonal antibody, splenectomy, TPO receptor agonists, vinca alkaloids. For ITP patients who have failed first- and second-line therapies, TPO receptor agonists represent an option where sufficient clinical data exist (category A). Campath-1H, combination of first and second line therapies, combination chemotherapy, or HSCT, represent options with minimal clinical data and the potential for considerable toxicity (category B).

At the time of preparing the ICR recommendations, mature data were available on the efficacy and short- and medium-term safety of a first-generation humanized anti-CD20 monoclonal antibody,[9-11] together with the results from pivotal trials that led to the approval of new TPO receptor agonists for the treatment of ITP.[12-20] Consequently, the most significant addition of the ICR recommendations should have been centred around these novel therapies, and a discussion of their place compared with the time-honored practice of splenectomy for patients with chronic ITP at risk of bleeding. In current practice, clinicians and patients are now faced with new questions such as: what is the role of splenectomy in patients with persistent or chronic ITP? Should TPO receptor agonists or a first-generation humanized anti-CD20 monoclonal antibody be used before splenectomy? In which order should splenectomy, the TPO receptor agonists and a first-generation humanized anti-CD20 monoclonal antibody be used? What are the long-term consequences of these treatments? In its analysis, the panel took into account the lack of randomized studies regarding the efficacy and safety of splenectomy and, in doing so, inevitably attributed a level of evidence and a strength of recommendation lower for the studies on splenectomy than for those available for TPO receptor agonists (level I, grade A); this is despite the evidence available for splenectomy that is derived from dozens of studies involving thousands of patients. Clearly this approach appears disputable and somehow arbitrary. The panel thus avoided making a direct comparison of the pros and cons of splenectomy versus TPO receptor agonists, setting splenectomy aside as a "second line surgical" option. No doubt, there is an increasing reluctance of both clinicians and patients to advise or undergo splenectomy and to prefer medical treatments for patients failing corticosteroids and requiring continuous treatment for their bleeding risk.[21] However, the fear of peri-procedural morbidity (no mortality is reported with the introduction of laparoscopic splenectomy), and the lifelong risk of fatal sepsis, have been overemphasized. With concerns relating to splenectomy, several authors have explored the off-label use of a first-generation humanized anti-CD20 monoclonal antibody in these patients, aiming to avoid surgery with

Table 4. Recommendations for second-line therapy – surgery. Taken from supplemental Document 8, Recommendation box 6 of the online version, and modified with permission[3]

Splenectomy remains the treatment option with by far the highest likelihood of producing cure. In general, it is recommended to wait at least 6 months from diagnosis before performing splenectomy due to the chance of spontaneous remission (Grade C recommendation, Evidence level IV)
When available, indium-labeled autologous platelet scanning may be useful prior to splenectomy to confirm that the spleen is the main site of platelet sequestration (Grade B recommendation, Evidence level III).
Accessory splenic tissue is common and should be sought in those who relapse after an initial durable response to splenectomy (Grade C recommendation).
Appropriate vaccination against *S pneumoniae*, *N meningitidis* and *H influenzae* must be provided; recent treatment with a first-generation humanized anti-CD20 monoclonal antibody may impair vaccination efficacy.

Table 5. Pros and cons of new treatments, compared to splenectomy, in patients failing first-line therapy and/or (some) medical second-line therapies

	Splenectomy	First-generation humanized anti-CD20 monoclonal antibody	TPO-receptor agonists
Type of treatment	Surgical (one-shot)	Medical (one-shot)	Medical (continuous)
ITP-specific	No, time-honored	No, off-label use	Yes, FDA and EMA approval
Response rate	› 80%	Long-term 15-20% of initially treated	› 80%
Response prediction	No reliable assay	No reliable assay	No reliable assay
Curative potential	Yes, up to 60% of patients	Uncertain. Possible in rare cases	Not expected, but possible in rare cases
Short term toxicity	Perioperative morbidity	Allergic reactions Fluid overload	No or minimal
Medium and long term toxicity	Small life-long risk of overwhelming sepsis	Cases of persistent leukemia or hypogamma globulinemia reported Risk of reactivation of hepatitis B Lack of efficacy of vaccination during the first months after exposure Rare cases of PML reported	No or minimal but to be fully evaluated
Follow up after response	On clinical ground	On clinical ground	Strict
Cost	Affordable (also in developing countries)	High	Highest

suboptimal outcomes.[22] It is now evident[10,23] that long-term responses are less than 30%, a much lower figure than that obtained with splenectomy which yields an 80% immediate response and long-term responses of 60-70%.[24] Moreover, the recent report of cases of progressive multifocal leukoencefalopathy (PML) in patients with autoimmune disorders including ITP is of concern.[25] Only TPO receptor agonists can parallel the favorable results obtained with splenectomy and their short- and medium-term safety is now well established. However, on cessation of treatment, all patients relapse (with only rare exceptions), as expected on the basis of the mechanism of action of these agents.

Tables 3 and 4, derived from recommendation boxes in the electronic appendix of the ICR guidelines, offer more direction to clinicians facing these difficult choices.

Table 5 summarizes the advantages and disadvantages in the author's opinion of using a first-generation humanized anti-CD20 monoclonal antibody, TPO receptor agonists or splenectomy; these considerations may represent the expectations of practising physicians to be clarified by practice guidelines. Many of these controversial aspects have not yet been definitively clarified in the published literature or in current practice recommendations. So, although the ICR may be a very valuable platform for the clinician to have a whole and balanced view of the state-of-the–art for ITP,

Table 6. Recommendations for patients failing first- and second-line therapies. Modified, with permission, from supplemental Document 8, Recommendation box 7 of the online version[3]

TPO-receptor agonists have produced excellent response rates in both splenectomized and non-splenectomized patients including those with relapsed/refractory disease unresponsive to numerous other approaches (Grade A recommendation, Evidence level Ib).

Other therapies that have been used as last resorts include combination chemotherapy, campath-1H and HSCT. The side effects of these treatment options may be severe and the data supporting their use are limited (Grade B recommendation; Evidence level IIb).

clinicians must still make final decisions based on their own judgement.

Furthermore, the ICR offers clearcut recommendations for patients failing first- and most second-line therapies, including/excluding splenectomy (Table 6).[3]

Finally, Table 7 provides a summary of the main characteristics from the ICR recommendations compared with those from the earlier ASH and BCSH guidelines.

Table 7. Summary of the main characteristics of the ICR compared with previously available guidelines

Guideline (Year of publication)	Composition of the panel*	Method for analysis consensus	Pros	Cons	Utility in difficult cases
ASH-GL (1996)	15 professional members only from US and Canada. Conflicts of interest not disclosed	Explicit, by voting. Rank of agreement provided	Detailed and complete. Endorsed by ASH	Not of immediate readability. Offers clear directions to clinicians. National guideline	Major in most circumstances
BCSH-GL (2003)	9 professional members only from UK. Conflicts of interest not disclosed	Not explicit	Detailed and complete. Updating from ASH-GL. Endorsed by BCSH	Not of immediate readability. National guideline	Major in most circumstances
ICR (2010)	20 professional members. International representation. Conflicts of interest fully disclosed	Not explicit	Detailed and complete only when appendix is considered. Offers the most updated summary of available evidence. International guideline	Not of immediate readability. Less directive in providing advise to practising physicians	Major in most circumstances Requires consideration of the electronic appendix for full appreciation of its utility.

* In addition to professional members (clinicians or scientists) one or more patient representative was included.

Conclusion

The ICR represents the most updated reference for the diagnosis and treatment of primary ITP. I had the privilege to work with the members of the ICR panel and to appreciate the efforts made for the completeness of the review, classification of the evidence and grading of recommendations. Many answers are still not available due to the lack of appropriate studies. In addressing this limitation, the ICR has favored a balanced assessment of all available evidence, abstaining from making a formal recommendation where consensus was limited. This approach often leaves the physician in charge of the patient with regard to final decision-making. The ICR offers extensive information for making appropriate clinical decisions and offering patients a full understanding of the pros and cons of current treatments. My personal view is that the expert panel should perhaps have undertaken the challenge of offering even more guidance. However, this may well reflect a personal perspective in practising medicine that favors medical direction over patient preferences in complex situations. With this approach, however, whilst never undervaluing a patient's personal values or concerns, but also avoiding taking a neutral stance in controversial areas, it is important to be fully aware of patient concerns about being well-informed.

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Clonal lymphoid expansions and immune thrombocytopenia

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Abstract

Purpose: There is well established evidence documenting an association between immune thrombocytopenia (ITP), an autoimmune disease characterized by reduced platelet count due to autoantibodies, and B-cell clonal malignancies, namely chronic lymphocytic leukemia (CLL), non-Hodgkin's lymphoma and, less frequently, multiple myeloma.

Summary: In these settings, anti-platelet autoantibodies are frequently produced by B-cells that do not belong to the tumor clone and that have emerged due to the immune dysregulation caused by the malignancy. In patients with ITP and a B-cell neoplasm, ITP should be treated *per se* and therapy against the B-cell clonal malignancy is not always required. ITP has also been described in patients with monoclonal gammopathy of uncertain significance with an incidence higher than that observed in the general population. The novel entity of monoclonal B-cell lymphocytosis (MBL) has been recently defined to include light-chain restricted lymphocytosis with an absolute lymphocyte count lower than that required for diagnosis of CLL. While it was initially felt that MBL may also predispose to ITP, available evidence from several MBL series has not validated this hypothesis. In addition to B-cell disorders, ITP may also be linked to T-cell large granular lymphocyte leukemia (T-LGL), and its presence is considered as a condition for initiating T-LGL treatment. Apart from T-LGL, there is scant evidence linking ITP and subclinical T-cell clonalities, although this issue is currently under scrutiny.

Conclusion: The observation that subclinical monoclonal expansions of B- and T-cells underlie the development of ITP in a fraction of patients, suggests a need for studies aimed at exploring whether the occurrence of pathogenetically relevant lymphoid clones is a more general feature of ITP. The identification of such clonal lymphoid expansions may be helpful for individually tailoring therapy with drugs primarily targeting specific cellular subsets.

Introduction

Immune thrombocytopenia (ITP) is a relatively frequent autoimmune disease, characterized by a persistently low platelet (PLT) count (‹150 x10^9/L) due to the presence of anti-PLT antibodies determining an increased rate of PLT destruction in the reticuloendothelial system. From a clinical standpoint, ITP is characterized by an increased risk of hemorrhagic complications, after surgery or dentistry, and spontaneous bleeding.[1]

The pathogenesis of ITP is highly heterogeneous; it may be related not only to the presence of anti-PLT autoantibodies but also, in a fraction of cases, to impaired PLT production. Increasing evidence suggests that the balance between these two pathogenetic mechanisms, i.e. immune PLT destruction and impaired PLT production, varies markedly between patients.[2] ITP is associated with a disruption of normal immune function that may induce the development of self-reactive autoantibodies. Accordingly, patients with ITP frequently display a CD4+ T helper $(Th)_0/Th_1$ activation profile along with reduced Th_2 and T regulatory cells, a pattern associated with improved antigen presentation (Andersson 2009, Cines 2009).[3,4]

First-line treatment of ITP consists of corticosteroids, high-dose intravenous immunoglobulins, or a combination of the two. Splenectomy is reserved as a second-line option, together with immuno-suppressive agents (e.g., cyclosporine, azathioprine, cyclophosphamide, mycophenolate mofetil), danazol or a first-generation humanized anti-CD20 monoclonal antibody, which shows efficacy even at doses markedly lower (100 mg/m^2) than those utilized in lymphoma.[5,6] Recently, thrombopoietin (TPO) receptor agonists have been developed as oral or subcutaneous formulations with very promising results. These novel drugs

may provide an innovative tool for the treatment of relapsed ITP with a highly favorable therapeutic index.[7,8]

ITP and Clonal Lymphoid Expansions

Due to the current absence of solid and reproducible markers of the disease, either molecular or serological, the diagnosis of ITP in clinical practice is largely based on exclusion of possible causes of secondary immune thrombocytopenia. These secondary causes include *Helicobacter pylori* infection, hepatitis C virus (HCV), human immunodeficiency virus (HIV), drugs, and autoimmune diseases, such as systemic lupus erythematosus or antiphospolipid syndrome.

Clonal lymphoid B-cell expansions, including B-cell lymphoproliferative disorders, represent a clinical condition that may associate with, and in some cases predispose to, ITP as well as other hematological immune cytopenias, namely autoimmune hemolytic anemia (AIHA). The co-occurrence of ITP and AIHA in a single patient is also known as Evans syndrome.[9]

ITP and overt B-cell malignancies

Among overt B-cell malignancies, ITP is known to associate with chronic lymphocytic leukemia (CLL)[10] and B-cell non-Hodgkin's lymphoma (NHL).[11] An association between ITP and multiple myeloma, though infrequent, has also been described in the literature.[12] ITP may precede the diagnosis of CLL and B-cell NHL or may be diagnosed simultaneously with the underlying malignancy. Occasionally, identification of ITP drives the diagnosis of a B-cell malignancy that would otherwise remain undiagnosed. Importantly, both staging systems commonly applied to CLL, i.e the Binet[13] and the Rai[14] staging systems, do not differentiate between thrombocytopenia due to bone marrow infiltration and thrombocytopenia due to autoimmune destruction. As a consequence, independent of the mechanism causing PLT reduction, CLL associated with thrombocytopenia is classified as stage C CLL according to Binet or high-risk CLL according to Rai.[10]

In CLL, the production of anti-PLT autoantibodies is not directly correlated to the CLL malignant clone but is probably due to immune dysregulation caused by the neoplastic disorder.[15] On these grounds, the treatment of ITP associated with CLL, and possibly with other B-cell malignancies, is not necessarily directed against the underlying B-cell neoplasm. Rather, treatment of ITP associated with CLL exploits drugs also used in "primary" ITP, including corticosteroids and intravenous immunoglobulins as first-line therapies. However, with this approach, the response rate of CLL-associated ITP is lower than that of primary ITP not associated with CLL.[16] Although some patients with CLL-associated ITP who fail first-line therapy may respond to second-line strategies, in clinical practice the presence of refractory ITP in the context of CLL is frequently considered to be an indication for initiating CLL-targeted treatment.[10] A similar therapeutic approach may be followed for patients with ITP associated with B-cell NHL, most frequently represented by indolent lymphoma. However, in NHL patients with ITP resistant to first-line therapy, the necessity for antineoplastic treatment is more pronounced, and an ITP remission is frequently achieved only after the administration of lymphoma therapy.[11]

ITP and other clonal lymphoid expansions

The co-occurrence of ITP and subclinical clonal B-cell populations has been known for the last twenty years.[17] Since pivotal evidence demonstrated the association of ITP with B-cell expansions, improvements in flow cytometry techniques led to the possibility of identifying very small clones in the peripheral blood of apparently healthy individuals. Today, the flow cytometric detection of a light-chain restricted lymphocytosis is defined as a monoclonal B-lymphocytosis (MBL) when fitting the diagnostic criteria described by Marti *et al.*,[18] subsequently revised by Hallek *et al.*,[10] for the International Workshop on CLL. MBL is phenotypically distinguished into three categories according to the expression of CD5 and CD23: i) no-CLL MBL (CD5-); ii) atypical CLL-like MBL (CD5+, CD23-, ^{bright}CD20+); and iii) CLL-like MBL (CD5+, CD23+, ^{dim}CD20+). Considering the absolute lymphocyte count, MBL may be distinguished into clinical MBL (cMBL) and low-count MBL. Individuals with cMBL are characterized by the presence of lymphocytosis identifiable by a routine blood cell count examination. The B-lymphocyte count distinguishing cMBL from CLL is 5000 x10^9/L. The identification of cMBL has led to a "downstaging" of a large number of patients who would be otherwise categorized as CLL, thus removing the diagnosis of leukemia, with its negative psychological impact, from patients who might never progress to overt leukemia. The definition of low count MBL applies to individuals with a normal blood cell count who exhibit clonal B-populations when investigated using highly sensitive flow cytometry approaches, usually restricted to research studies.[18]

The prevalence of MBL in the general population has been studied by several groups. In people aged over 60 years, the detection rate of MBL exceeds 5% when studied with 4-color flow cytometry,[19] and exceeds 10% when studied with highly refined 8-color flow cytometry techniques.[20] The highest prevalence of MBL is detected in relatives of patients with CLL.[21] In prospective studies of individuals with a diagnosis of CLL-like cMBL, the risk of progression to CLL has shown a certain degree of variability, ranging from 1-2% per year[22,23] to 10% per year in the first 6 years, followed thereafter by 3% per year, with the absence of a plateau.[24]

Clinical management of individuals with cMBL is still a matter of debate, particularly for those with atypical CLL-like and non-CLL MBL. Individuals with cMBL should be evaluated by a hematologist, with a review of family history, examination for B-symptoms, and physical examination with particular attention to potential lymph node enlargement or organomegaly. In the case of atypical CLL-like and non-CLL MBL, a thorough examination should be performed to exclude NHL, including computed tomography with contrast medium, a bone marrow biopsy with FISH analysis

for t(11;14) and immunohistochemical staining for cyclin D1.[25,26]

Given the well known association between ITP and CLL, it was initially felt that MBL might predispose to ITP. This hypothesis, however, is not supported by current evidence. Rawstron *et al*.[22] studied a cohort of 185 individuals with CLL-like MBL, with a median observation time of 6.7 years. Twenty-eight out of 185 patients progressed to CLL, and the presence of anemia or thrombocytopenia was described in 4 of these 28 patients. Considering the PLT count at the initial evaluation of MBL, individuals with CLL-like low count MBL, i.e. those with a normal blood count, showed a median PLT number of 259000 x 10^9/L (range 142000 - 492000 x 10^9/L), while subjects with CLL-like cMBL, i.e. those with a lymphocytosis, showed a median PLT number of 221000 x 10^9/L (range 67000 - 487000 x 10^9/L). Importantly, in this study, PLT count was not a prognostic factor for either progressive lymphocytosis or death. Overall, this large series failed to document an association between MBL and ITP.

The Mayo Clinic published an independent series of cMBL, that were compared to subjects with Rai 0 CLL, with a median follow-up for vital status of 34 months.[23] The authors excluded subjects with cytopenia due to marrow infiltration, and consequently the median PLT count was 226000 (184000 x 10^9/L in quartile 1) in the 302 individuals with cMBL and 208000 x 10^9/L (177000 x 10^9/L in quartile 1) in the 94 patients with Rai 0 CLL. In this series, no patients had thrombocytopenia at the time of diagnosis – confirming the results of other series.[22] During follow-up, the development of AIHA, but not of ITP, was reported as an indication for starting treatment.

The third large series of cMBL available in the literature has been published by Rossi *et al*.[24] This series comprised 460 consecutive individuals with CLL phenotype B-lymphocyte expansions, collected since 2006 at the Division of Hematology of the Amedeo Avogadro University of Eastern Piedmont and at the University of Siena, Italy, with a median follow-up of 42.7 months. The aim of this study was to evaluate clinical progression, and also to determine the biological prognostic factors of CLL, such as IGHV-D-J rearrangements, HCD3 clustering, TP53 mutations, flow cytometric expression of CD38 and CD49d, and FISH analysis for aneuploidy of chromosome 12, del13q14, del17p13, del11q22-q23. Subjects were reclassified as cMBL (123 cases) or CLL (337 cases, 154 with Rai stage 0). The study concluded that cMBL and Rai 0 CLL patients differ in their prognosis and biological profile, with a multivariate analysis indicating that the presence of +12 or del17p13 were the only independent risk factors for future requirement of treatment in cMBL individuals progressing to CLL. Median PLT count at the time of diagnosis was 221000 x 10^9/L (range 188000 - 261000 x 10^9/L) in subjects with cMBL and 207000 x 10^9/L (range 172000 - 253000 x 10^9/L) in those with Rai 0 CLL. Consistent with other published series,[22,23] no cMBL case presented a PLT count diagnostic for ITP.

Overall, the results of these three large series of cMBL do not point to an association between cMBL and ITP. Given the recent recognition of MBL as an independent entity and, consequently, the relatively short follow up of these studies, it cannot be excluded that ITP may represent a late event in the clinical history of cMBL. Longer follow up of the available series, in addition to prospective studies, are needed to clarify this issue and to correctly determine the relationship between ITP and cMBL.

ITP and plasma cell disorders

Monoclonal gammopathy of uncertain significance (MGUS) is a condition characterized by the presence of a monoclonal component (MC) not fulfilling the diagnostic criteria for multiple myeloma and occurring in the absence of end-organ damage (hypercalcemia, renal failure, anemia, and bone lesions).[27] Several series have described an association between MGUS or multiple myeloma and ITP.[12,28,29] Rossi *et al*. reported on the occurrence of thrombocytopenia in a large series of 228 consecutive patients with MGUS.[29] Causes of secondary thrombocytopenia were excluded in all patients. The type of MC was IgG in 163 cases (71.8%), IgA in 17 cases (7.4%), and IgM in 41 cases (18.9%). A double MC was observed in 6 cases (2.6%) and light chain only (λ) was seen in 1 case (0.4%). All cases tested negative for B-lymphocyte clonality by flow cytometry on bone marrow aspirate.

Overall, after an observation period of 681.33 patient-years, 7/228 (3.1%) patients with MGUS presented with or developed ITP. In particular, 6 of these patients had ITP at the time of diagnosis, and another patient developed ITP during follow-up. The crude incidence rate of ITP in patients with MGUS was 146.8 per 100,000 patient-years, this equates to a crude incidence of ITP approximately 55- to 90-fold higher than in the general population.[29] Of the 7 patients with ITP and MGUS, none progressed to multiple myeloma after a median follow-up of 22 months; 3 patients required ITP treatment, and the response was not associated with the level of the monoclonal component.[29]

As is the case for ITP associated with B-cell clones, the target for treatment in patients with ITP associated with MGUS is represented by ITP and not MGUS. As observed in ITP associated with other B-cell disorders, the risk of resistant/refractory ITP in MGUS appears to be higher than in primary ITP.[12]

ITP and monoclonal T-cell expansions

T-cell large granular leukemia (T-LGL) is a rare clonal T-cell disorder associated with a proliferation of CD8+ T-lymphocytes.[30] T-LGL is more common in the elderly, and is characterized by neutropenia, thrombocytopenia, and an increased incidence of infections and autoimmune diseases. Thrombocytopenia occurs in approximately 20% of T-LGL patients.[31] Analogous to CLL, the pathophysiology of ITP occurring in the context of T-LGL is multifactorial, and a role for anti-PLT autoantibodies has been hypothesized. Thrombocytopenia with a PTL count less than 50 x 10^9/L is considered to be criteria for initiation of treatment directed against the T-LGL

clone; for example, an immunosuppressive treatment generally consisting of cyclosporine with or without corticosteroids.[31]

The association of ITP with subclinical T-cell expansions is being independently investigated. Sabnani *et al*.[32] recently described 7 patients with ITP and a concurrent clonal T-cell expansion not fulfilling the diagnostic criteria for T-cell malignancies and therefore defined as T-cell clonopathy of unknown significance. These patients displayed particularly resistant disease, characterized by failure of several treatments for ITP, including corticosteroids, intravenous immunoglobulins, splenectomy, and a first-generation humanized anti-CD20 monoclonal antibody but had anecdotal success with azathioprine. Based on these results, the authors suggested that patients with resistant or refractory ITP should be evaluated for T-cell clonality and considered for treatment options directed against cytotoxic T-cells rather than therapies, such as a first-generation humanized anti-CD20 monoclonal antibody, that specifically target B-cells.[32]

Conclusions

ITP represents a possible complication of B- and T-cell clonal diseases, such as CLL, B-cell NHL and T-LGL, and should be suspected when patients affected by these disorders display thrombocytopenia that cannot be attributed to other causes, including bone marrow infiltration or toxicity due to chemotherapy. In many of these patients, only bone marrow examination allows conclusive differentiation between ITP and thrombocytopenia secondary to marrow infiltration.

Collectively, the data on MGUS and the identification of ITP patients with T-cell clonopathy of unknown significance, indicate that ITP may also associate with subclinical lymphoid proliferations. However, available data has failed to document such an association between ITP and MBL. The recent recognition of MBL and the limited follow-up of MBL cohorts, suggest that long-term data are necessary to draw a definite assessment regarding the putative relationship between ITP and MBL.

Overall, the association between ITP and clonal lymphoid expansions, either subclinical or overtly malignant, represents an interesting model for understanding the pathophysiology of ITP and possibly also for treatment stratification. The observation that subclinical monoclonal expansions of B- and T-cells, as documented by MGUS and T-cell clonopathy of unknown significance, underlie the development of ITP in a fraction of patients, suggests a need for studies aimed at exploring whether the occurrence of pathogenetically relevant lymphoid clones is a more general feature of ITP. From a clinical standpoint, given the availability of multiple therapeutic agents for ITP, the identification of such clonal lymphoid expansions may be helpful for individually tailoring therapy with drugs primarily targeting specific cellular subsets.

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Pathophysiological mechanisms of thrombocytopenia in patients with chronic hepatitis C virus infection

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Abstract

Purpose: Thrombocytopenia is a hematological abnormality commonly encountered in patients with chronic hepatitis C virus (HCV) infection. This review discusses the various pathophysiological mechanisms associated with decreased platelet count in these patients.

Summary: In patients with chronic HCV infection, thrombocytopenia can be caused by portal hypertension, decreased thrombopoietin synthesis by the liver, immune-mediated phenomena or depressed bone marrow activity. In the individual patient, it may be difficult to identify the main pathophysiological mechanism responsible for decreased platelet count, as more than one mechanism may be implicated in the development of thrombocytopenia. Furthermore, interferon-based antiviral therapy may be associated with a decrease in platelet count, secondary to inhibition of late-stage maturation of megakaryocytes.

Conclusions: In patients with chronic HCV infection, several pathophysiological mechanisms may be responsible for thrombocytopenia. Portal hypertension and decreased hepatic synthesis of thrombopoietin are the main mechanisms responsible, although virus- or antiviral therapy-related immune-mediated phenomena and bone marrow suppression may play a role in a subset of patients.

Introduction

Thrombocytopenia (decreased platelet count) is a hematological abnormality frequently observed in patients with chronic hepatitis B virus (HBV) or hepatitis C virus (HCV) infections.[1] It is more common among patients with HCV infection than those infected with HBV.[2] Indeed, chronic HCV infection is a well established cause of thrombocytopenia, even in the absence of overt liver disease,[3] and HCV testing should be considered in patients with unexplained low platelet count.[2] The finding that patients with HCV infection have higher rates of thrombocytopenia than patients with HBV infection suggests that the etiology of thrombocytopenia in HCV patients involves factors other than advanced liver disease.[4] Recently, several studies evaluated the pathophysiological mechanisms associated with the development of thrombocytopenia in patients with chronic HCV infection. Many of these studies considered special populations and therefore may be subject to selection bias. Nevertheless, taken together, they confirm the hypothesis that several pathophysiological mechanisms may contribute to decreased platelet count in HCV patients. This review will detail these mechanisms, and comment upon the major findings on this topic.

Pathophysiology of thrombocytopenia in patients with chronic HCV infection

The pathophysiology of thrombocytopenia in patients with HCV-related chronic liver disease is not yet completely understood. Several mechanisms have been implicated in this process, including: i) increased splenic pooling of platelets due to portal hypertension (hypersplenism);[5] ii) decreased hepatic synthesis of thrombopoietin (TPO) due to increased fibrosis and decreased hepatocellular functioning mass;[6,7] iii) immune-mediated reactions involving anti-platelet antibodies, formation of immune-complexes, or antigen mimicry between HCV and platelet glycoproteins (as reviewed by Stasi 2009);[8] iv) suppression of bone marrow activity either by HCV or by interferon (IFN)-based antiviral therapy (as reviewed by Cines *et al.* 2009[9] (Figure 1). In a given patient, more than one mechanism may be responsible for the development of thrombocytopenia.

Figure 1. Main pathophysiological mechanisms responsible for thrombocytopenia in patients with chronic hepatitis C virus infection

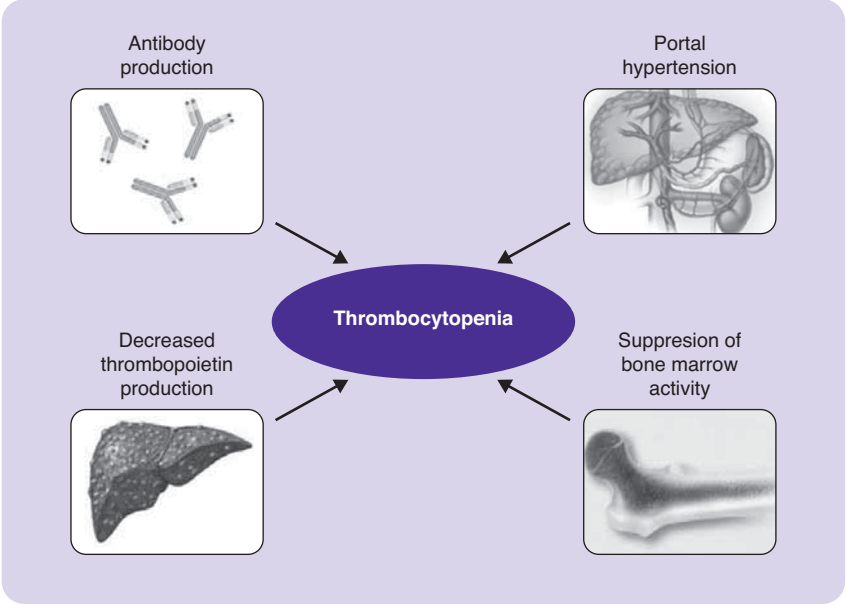
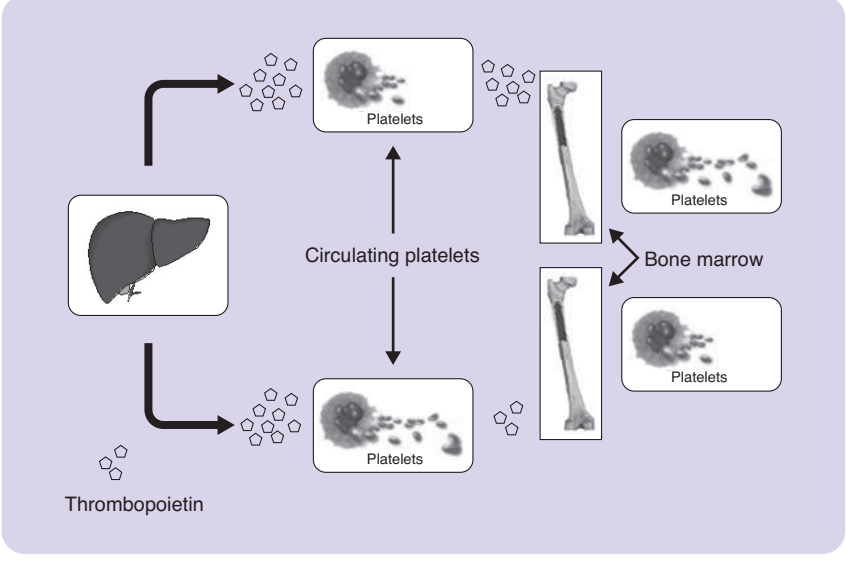


Figure 2. Mechanisms of thrombopoietin and platelet mass feedback



Hypersplenism

Hypersplenism (pooling of platelets within an enlarged spleen and subsequently their clearance from circulation) was the first mechanism proposed to explain the pathogenesis of thrombocytopenia in patients with chronic liver disease.[5] However, splenic sequestration of platelets does not fully account for the occurrence of thrombocytopenia in these patients. While up to one-third of patients with splenomegaly have normal platelet counts,[6] hypersplenism is not a consistent clinical finding in patients with HCV and thrombocytopenia.[3,10] Correction of portal hypertension (the main pathophysiological cause of hypersplenism), either by surgical decompression or insertion of a transjugular intrahepatic portosystemic stent-shunt, is not necessarily associated with platelet count recovery.[11] Notably, partial splenic embolisation, an interventional radiological technique that decreases splenic volume by delivering embolizing agents through the splenic artery and therefore reducing its pooling capacity, is associated with an increase in platelet count that is only partially due to improved portal hemodynamics. Improved liver function, increased TPO synthesis and decreased TPO uptake by pooled platelets within the spleen seem to be the relevant factors leading to an increase in platelet count after this procedure.[12]

Decreased hepatic synthesis of thrombopoietin

TPO is the growth factor that primarily regulates megakaryocyte maturation and platelet formation.[13] It is normally produced by the liver at a constant rate, and is cleared from the circulation upon binding to its receptor (c-Mpl), which is present on both megakaryocytes and platelets.[14] Platelets are able to bind, internalize and degrade TPO. Therefore, total platelet mass, including platelets sequestered in the spleen, is the main physiological regulator of serum TPO levels.[13] Conditions associated with a decrease in total platelet mass lead to a greater availability of TPO, stimulating megakaryocytopoiesis and platelet production, thus restoring platelet homeostasis. When platelet count increases, excess TPO is scavenged by the circulation upon its binding to the platelet receptor, with a consequent decrease in its serum levels (Figure 2). This feedback is impaired in patients with chronic liver disease. In patients with chronic viral hepatitis without splenomegaly, increasing degrees of liver fibrosis are associated with lower serum levels of TPO.[6] Indeed, among untreated patients with chronic HCV infections, those with advanced fibrosis have significantly lower TPO serum levels than those with no or mild fibrosis.[7] These findings are not due to liver fibrosis *per se* but rather to the deleterious effect of liver fibrosis on liver function. In fact, there is a direct correlation between the hepatocellular function-

ing mass and TPO serum levels (r_s=0.52, p=0.01) in these patients.[15] Overall, in these patients serum TPO levels may be normal or reduced, although they are too low for the observed degree of thrombocytopenia (as reviewed by Nichol 1998).[16] Lastly, studies carried out in patients undergoing liver transplant have provided the most solid evidence for the role TPO plays in the development of thrombocytopenia in patients with advanced liver disease. These studies have consistently shown that, after successful orthotopic liver transplantation, there is an increase in serum TPO levels, which is accompanied by an increase in reticulated platelets, and followed by an increase in platelet count 7–10 days after the increase in TPO levels, eventually leading to the restoration of normal platelet counts.[17-19]

Immune-mediated platelet destruction

In a study in Japanese patients infected with HCV or HBV, the prevalence of either any thrombocytopenia (platelet count ‹150 × 10^9/L) and severe thrombocytopenia (platelet count ‹10 × 10^9/L) was higher in HCV than HBV patients (41% versus 19% and 10% versus 2%, respectively).[4] In this study, the prevalence of abnormally elevated platelet-associated immunoglobulin G was also higher in HCV than HBV patients (88% versus 47%). Despite these results, and the evidence for a possible pathogenic role of antibodies directed against platelet surface glycoproteins,[20] the pathogenic significance of anti-platelet antibodies in patients with liver disease is still uncertain. Indeed, anti-platelet antibodies can be detected in up to 66% of anti-HCV-positive patients.[21] Notably, anti-platelet antibodies, mainly directed against glycoprotein IIb/IIIa, are not related to platelet counts, stage of liver disease or viral genotype, and are not influenced by treatment with IFN-α despite an increase in platelet count.[21]

Although a definite clinical relevance of anti-platelet antibodies is lacking, their likely pathogenic role in thrombocytopenia in patients infected with HCV has been the subject of various studies. Several mechanisms have been proposed to explain the pathophysiological basis of immune-mediated thrombocytopenia in these patients. de Almeida *et al*. have shown that HCV-RNA is detected in platelets significantly more commonly in thrombocytopenic (60%) than non-thrombocytopenic patients (35%, p=0.017), suggesting that HCV may be directly involved in the pathogenesis of thrombocytopenia.[22] HCV can bind to the platelet membrane,[23] leading to subsequent binding of anti-HCV antibodies to the HCV-platelet membrane complex, eventually leading to phagocytosis of platelets. Antigen mimicry can also lead to immune-mediated thrombocytopenia in patients with HCV. The HCV core protein can induce formation of antibodies that cross-react with an epitope of platelet glycoprotein IIIa, and can therefore induce thrombocytopenia.[24] This evidence is supported by the substantial increase in platelet count reported after successful antiviral therapy in patients with HCV and thrombocytopenia.[25]

Bone marrow suppression

HCV infection affects platelet production in two ways: either directly through inhibition of megakayocytopoiesis or indirectly through the myelosuppressive effect of antiviral drugs, such as standard or pegylated IFN-α.

Thrombokinetic data show that, in a well identified subset of patients with HCV-associated thrombocytopenia and without more easily identifiable causes for this hematological abnormality, underproduction of platelets secondary to bone marrow suppression due to HCV may play a causative role in the development of thrombocytopenia.[26]

Antiviral therapy aimed at eradicating HCV infection may be associated with some degree of myelosuppression. Both IFN and its pegylated form can induce a decrease in peripheral platelet count.[27] Inhibition of megakaryocytopoiesis is more marked in regimens that do not contain ribavirin, as ribavirin-induced anemia increases endogenous erythropoietin production, stimulating megakaryocytopoiesis due to its aminoacidic sequence homology with TPO.[27] IFN-induced thrombocytopenia is caused by suppression of late-stage megakaryocytopoiesis, while megaryocytes endomitosis and proliferation are preserved.[28]

Thrombocytopenia occurs commonly during pegylated-IFN and ribavirin antiviral therapy. In patients with advanced fibrosis and cirrhosis treated with full-dose pegylated IFN-α-2a, Heathcote *et al*. found that platelet count fell below 75 × 10^9/L at some point during the study in 47% of patients, and platelet count below 50 × 10^9/L was observed in 19% of the patients assigned to this treatment regimen.[29] Although this is rarely associated with relevant bleeding, thrombocytopenia that requires a reduction in drug dosage may lead to lower sustained virological response rates.[30] A blunted TPO response to thrombocytopenia may contribute to more severe decreases in peripheral platelet counts during IFN-based antiviral therapy, especially in patients with cirrhosis.[31] It remains to be established whether therapeutic administration of TPO-receptor agonists may be associated with not only lower rates of treatment-induced thrombocytopenia but also with higher rates of viral eradication.

Conclusions

Infection with HCV is one of the most common causes of chronic liver disease. Thrombocytopenia is a common hematological abnormality in these patients. Several mechanisms can contribute to its development, including splenic sequestration of platelets due to portal hypertension, decreased hepatic synthesis of thrombopoietin secondary to impaired liver function, immune-mediated platelet clearance and defective production of platelets in the bone marrow. These mechanisms are not mutually exclusive, and may each contribute to the development of thrombocytopenia in various subsets of patients.

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Idiopathic thrombocytopenia: the importance of a correct differential diagnosis

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Abstract

Idiopathic thrombocytopenia (ITP) is an autoimmune disease. In the absence of reliable diagnostic tests for ITP, the possibility of secondary thrombocytopenia must first be evaluated. The aim of this study was to evaluate the differential diagnosis of otherwise healthy individuals presenting with thrombocytopenia and normal peripheral white and red blood cell counts to the Department of Hematology at the Azienda Ospedaliera Universitaria Integrata di Verona. From July 1991 to June 2005, 161 consecutive adult patients aged 16 to 82 years presented with unexplained thrombocytopenia. A diagnosis of ITP was confirmed in 109/161 (67.7%) patients. Of the remaining 52 patients, 35 patients (21.7%) were diagnosed with pseudothrombocytopenia and a further 17 patients (13.5%) had confirmed secondary thrombocytopenia. These findings highlight the importance of a careful differential diagnosis for thrombocytopenia in order to avoid unnecessary or inappropriate treatment.

Background

Thrombocytopenia may be inherited or acquired.[1] While the inherited forms are usually associated with platelet dysfunction and/or physical malformation, they are rare and primarily occur in infancy or in young adults. The acquired form of thrombocytopenia may be detected at any age and may or may not be of autoimmune origin.[2-4]

Idiopathic thrombocytopenia (ITP) is an autoimmune disease in which autoantibodies bind to the platelet surface, leading to platelet destruction.[1] ITP is defined by the presence of thrombocytopenia (platelets ‹100 x 10^9/L) and megakaryocytic hyperplasia in the bone marrow aspirate.[4] The disease is relatively common in children, with an estimated incidence of 5 cases per 100,000 persons per year, while the incidence in adults is somewhat lower (2.7 per 100,000 persons per year).[4] The criteria for the diagnosis of ITP are based on the exclusion of other causes of thrombocytopenia, such as drugs, type IIB von Willebrand disease (VWD) or other congenital thrombocytopenias, disseminated intravascular coagulation, viral infections (i.e., human immunodeficiency virus [HIV], hepatitis C virus [HCV], Epstein-Barr virus, cytomega-lovirus), thrombotic microangiopathies (i.e., thrombotic thrombocytopenic purpura and hemolytic uremic syndrome) and autoimmune disorders (i.e., systemic lupus erythematosus [SLE] and antiphospholipid syndrome [APS]). Thus, in the absence of reliable diagnostic laboratory tests for ITP, the possibility of secondary thrombocytopenia must be evaluated.

The aim of this study was to evaluate the different diagnoses performed at our institutions, in otherwise healthy individuals presenting with thrombocytopenia and with normal peripheral white and red blood cell counts.

Patients and Methods

From July 1991 to June 2005, 161 consecutive cases of unexplained thrombocytopenia in adult patients aged 16 to 82 years (median age 53 years; male/female ratio 0.6) were referred to the Department of Hematology at the Azienda Ospedaliera Universitaria Integrata di Verona. In all cases, physical evaluation, peripheral blood smear examination and blood cell counts in sodium citrate were performed. Examination of bone marrow aspirate, and laboratory tests including, screening for HCV and HIV infections, antinuclear antibodies (ANA) and extractable nuclear antigen (ENA), antiphospholipid antibodies (lupus anticoagulant and anticardiolipin antibodies, anti-β2 glycoprotein 1), prothrombin time (PT) and activated partial thromboplastin time (aPTT) were also performed.

Results

A diagnosis of thrombocytopenia was confirmed in 109/161 (67.7%) cases. Of the remaining 52 cases, 35 patients (21.7%) were diagnosed with pseudothrombocytopenia, and 17 patients

(13.5%) were diagnosed with secondary thrombocytopenia, including: 1 case of acute lymphoblastic leukemia, 1 case of congenital macrothrombocytopenia, 2 cases of myelodisplastic syndrome, 2 cases of HIV infection, 3 cases of HCV infection and 2 cases of SLE were detected. In four cases, a prolonged aPTT was assessed, and 3 cases of APS and 1 case of type IIB VWD were detected. In two patients nodal enlargement was revealed upon abdomen ultrasonography, and a diagnosis of bone marrow involvement by follicular non Hodgkin B lymphoma was confirmed after bone marrow trephine biopsy.

Discussion

Of those patients presenting with thrombocytopenia, pseudothrombocytopenia was diagnosed in 22%; however, it is likely that the real prevalence of pseudothrombocytopenia may be underestimated due to the fact that not all detected cases are sent to a referral center. In a further 13.5% of cases the detection of thrombocytopenia led to a diagnosis other than ITP. On the basis of these findings, a correct diagnosis of ITP was made in approximately two-thirds of patients (109/161) with thrombocytopenia of unknown origin.

Conclusions

Our data underline the necessity of a careful differential diagnosis for thrombocytopenia in order to avoid unnecessary or inappropriate treatment. This is particularly the case in pediatric patients in whom inherited thrombocytopenia may be more frequently encountered.

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Long-lasting thrombocytopenia in a patient with Ewing family of tumors

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Abstract

Thrombocytopenia often occurs in cancer patients and is usually caused by antineoplastic agents. However, thrombocytopenia may arise from other causes, such as hematological conditions. These conditions include heparin-induced thrombocytopenia (HIT), disseminated intravascular coagulation (DIC), thrombotic thrombocytopenic purpura (TTP)/hemolytic-uremic syndrome (HUS), immune thrombocytopenia, viral thrombocytopenia, and hypersplenism. Thrombocytopenia may determine both the dose-density and dose-intensity of chemotherapy that can be administered to a patient and can, therefore, negatively impact on clinical outcome. We present a case of persistent thrombocytopenia in a young woman diagnosed with Ewing's sarcoma in which continuous delays in administering chemotherapy due to low platelet counts had a negative impact on disease prognosis.

Background

In general, thrombocytopenia is defined as a platelet level below 150 x 10^9/L. It is a multifactorial condition that can be classed as either primary, due to intrinsic bone marrow disease, or secondary, caused by chemotherapy, tumor-induced coagulopathy, thrombotic thrombocytopenic purpura (TTP)/hemolytic-uremic syndrome (HUS),[1] disseminated intravascular coagulation (DIC),[2] heparin-induced thrombocytopenia (HIT),[3] viruses,[4] autoimmunity, and hypersplenism.[5]

Thrombocytopenia in cancer patients is important because it impacts not only on clinical management but also prognosis. The presence of thrombocytopenia may necessitate dose modification in the form of dose delay or dose reduction, transfusions and generally increases the patient's requirement for medical support.

Over the past decade, myeloid and erythroid hematopoietic growth factors have helped to reduce complications associated with anemia and leucopenia. Although extensively studied, the phenomenon of megakaryocytopoiesis has been much more difficult to elucidate. Indeed, many cytokines are involved in this process, such as interleukin (IL)-11, IL-6 and thrombopoietin.[6] However, to date none of these agents have proven to be an effective treatment of thrombocytopenia. However, thrombopoeitin receptor agonist, which demonstrates a good toxicity profile,[7] is now available but is only approved for the treatment of patients with chronic refractory immune thrombocytopenic purpura post-splenectomy. Several ongoing studies aim to better define its optimum administration schedule and impact on survival, if any, in cancer patients.

The following case illustrates the negative impact on clinical outcome of thrombocytopenia in a patient with a solid tumor.

Patient and Methods

In November 2008, a 22-year old woman with back pain consulted her family physician. She was prescribed a non-steroidal anti-inflammatory drug but without any benefit. In January 2009, MRI revealed a mass of the right hip which was then biopsied. The histological diagnosis was Ewing's sarcoma/peripheral neuroectodermal tumor (PNET). Cytogenetic analysis showed positive rearrangement of the EWSR gene at 22q12.

Radiological evaluation was then performed. Brain, chest and abdominal CT, CT-PET and bone scan revealed the involvement of the right pelvis, with a longitudinal extension of 20cm infiltrating gluteal, ileopsoas, and paravertebral muscles. Secondary lesions involved the vertebral column, femur, lungs and kidneys. A bone marrow biopsy revealed normal features without involvement of the sarcoma. Baseline laboratory tests

Table 1. Protocol ISG-AIEOP/VHR-EW 02. Each step is preceded by complete radiological assessment of the disease

Stage 1	Two cycles of cisplatin 120 mg/m^2
Stage 2	Eight cycles of intensive chemotherapy (using different combinations of ifosfamide, vincristine, doxorubicin, etoposide, cyclophosphamide + peripheral blood stem cell harvest after cyclophosphamide/etoposide) + radiotherapy Surgery if possible
Stage 3	If partial response or stable disease have been achieved and if an HLA-compatible relative is available, non-myeloablative treatment followed by allogeneic stem cell transplantation

Table 2. Chemotherapy dosage and administration

Treatment regimen	Agent and doses
Cisplatin (every 3 weeks)	Cisplatin 60 mg/m^2 days 1 and 2
IVAdr (every 3 weeks)	Ifosfamide 3g/m^2 days 1-3 MESNA 3g/m^2 days 1-4 Vincristine 1.5 mg/m^2 (max 2mg total dose) day 1 Doxorubicin 45 mg/m^2 days 1 and 2
VAC (every 3 weeks)	Cyclophosphamide 1200 mg/m^2 day 1 Doxorubicin 40 mg/m^2 days 1 and 2 Vincristine 1.5 mg/m^2 (max 2mg total dose) on day 1
CE (every 3 weeks)	Cyclophosphamide 4 g/m^2 day 1 Etoposide 100 mg/m^2 days 2-4
VIDE (every 3 weeks)	Ifosfamide 3 g/m^2 days 1-3 MESNA 3 g/m^2 days 1-4 Vincristine 1.5 mg/m^2 (max 2mg total dose) day 1 Doxorubicin 20 mg/m^2 days 1-3 Etoposide 150 mg/m^2 days 1-3
Vincristine (weekly)	Vincristine 1.5 mg/m^2 (max 2mg total dose) day 1
Topotecan/ cyclophosphamide (every 4 weeks)	Topotecan 0.75 mg/m^2 days 1-5 Cyclophosphamide 250 mg/m^2 days 1-5
Methotrexate (weekly)	Methotrexate 7.5 mg/m^2 day 1

revealed normal complete blood count (platelets 418 x 10^9/L; white cell count 13.6 x 10^9/L; haemoglobin 8.99 g/dL due to β-thalassemic trait), and normal organ function (lactate dehydrogenase 860 U/L).

The patient was treated according to the protocol ISG-AIEOP/VHR-EW.02 for very high risk Ewing's family of tumors (Table 1). Initial chemotherapy treatment was started with 2 cycles of cisplatin as "window therapy" followed by 2 cycles of intravenous doxorubicin and 1 cycle of VAC (see Table 2 for all chemotherapy regimen doses). At the end of "window therapy", the patient had experienced a partial response (CT longitudinal extension of the mass had decreased to 13cm) and clinical benefit, as demonstrated by a marked reduction in opioid analgesic therapy. During the first two cycles of chemotherapy, the patient experienced grade 3 thrombocytopenia (platelet count at nadir was 27 x 10^9/L) with subsequent slow improvement.

When the platelet count had recovered to 76 x 10^9/L, a cycle of CE was administered with the aim of mobilising CD34 positive hematopoietic precursors into the peripheral blood. Even though this cycle was associated with a further improvement in the clinical situation, it did not mobilize peripheral blood stem cells (PBSC), making PBSC collection impossible. Moreover, it also induced further thrombocytopenia, requiring platelet transfusions and lasting longer than expected (48 x 10^9/L 4 weeks after the cycle). Further cycles of chemotherapy were, therefore, postponed.

Despite the patient's young age and lack of bone marrow disease involvement, we were faced with the puzzling situation of a woman with unexpected thrombocytopenia that was causing important delays in treatment. Possible causes of thrombocytopenia other than chemotherapy were investigated.

A blood smear was performed in order to exclude pseudo-thrombocytopenia. After confirming the diagnosis of true thrombocytopenia, subsequent analysis permitted us to exclude the following etiologies: immune (heparin-induced, autoimmune); viral, peripheral destruction (hypersplenism, DIC, TTP); and bone marrow invasion. None of the investigations led to a definitive pathogenetic explanation of the thrombocytopenia (Table 3). However, bone marrow trephine biopsy showed severe megakaryocytic hypoplasia despite erythroid hyperplasia (Figure 1).

Fifty-five days after the cycle of CE, by which time the platelet count had risen

Table 3. Investigations for differential diagnosis of thrombocytopenia

Parameter	Value	Normal range
Hematology		
WBC (x10^9/L)	1.48	4.4–11.0
ANC (x10^9/L)	0.77	2.0–8.0
Hb (g/dL)	9.13	12.0–16.0
MCV (fl)	79	80–98
Platelets (x10^9/L)	48	150–400
Reticulocytes (x10^9/L)	53	20–100
PT (%)	104	70–120
INR	0.97	0.86–1.30
aPTT (sec)	58.3	25–42
Fibrinogen (mg/dL)	387	200–400
AT-III (%)	111	80–120
D-Dimer (µg/mL)	0.29	0.01–0.5
Biochemistry		
Aptoglobin (g/L)	0.78	0.5–2.0
Bilirubin (mg/dL)	0.42	0.2–1.3
Creatinine (mg/dL)	0.5	0.3–1.1
LDH (U/L)	246	240–480
	Other investigations	
Autoantibodies	Negative for antiplatelet autoantibodies Negative for antiheparin autoantibodies	
Blood smear	Schistocyte screen negative	
Bone marrow biopsy	Histologic: negative for neoplastic involvement Immunophenotype: no pathologic features Viral screen EBV/CMV/Parvovirus/Aspergillus DNA negative Megakaryocytic hypoplasia	
Chest-abdomen CT	No hypersplenism or other findings that might explain thrombocytopenia	
Complete physical exam	No pathologic features related with thrombocytopenia or its causes	
Virus on peripheral blood	EBV/CMV/Aspergillus DNA negative	

to 114 x 10^9/L, a second cycle of cyclophosphamide was administered. We omitted the etoposide with the aim of reducing bone marrow toxicity. Clinical benefit was seen, as measured by the patient no longer requiring analgesics, but PBSC mobilization again failed. The platelet nadir was 15 x 10^9/L. Due to the unsuccessful PBSC harvest, the patient was excluded from further treatment with the protocol ISG-AIEOP/VHR-EW.02.

The patient's platelet count slowly increased and reached 100 x 10^9/L 40 days later. At that time, a cycle of VIDE was administered. Thirty days after this cycle, grade 3 thrombocytopenia was still present (platelet count 53 x 10^9/L) and increasing pain was reported by the patient.

A CT scan in September 2009 demonstrated local progression of the disease. Hypofractionated radiotherapy (20 Gy of 4 Gy/fraction) was administered to the right pelvis and produced a clinical benefit, allowing interruption of analgesic drug therapy. Weekly intravenous vincristine, chosen for its low bone marrow toxicity, was commenced when the platelet count reached 101 x 10^9/L. The vincristine dose was subsequently reduced to 70% (5 of the 7 planned cycles were administered). Short lasting disease stability was achieved, but the disease progressed 3 months after the last radiological assessment.

Two cycles of topotecan/cyclophosphamide were administered as soon as the platelet count reached 80 x 10^9/L. Persistent grade 4 thrombocytopenia that required repeated platelet transfusions followed. Thereafter, only palliative therapy could be administered and the patient died from progressive disease in March 2010.

The trend in platelet count during the clinical history is shown in Figure 2.

Discussion

This case demonstrates how persistent thrombocytopenia can negatively impact on the treatment of a very chemosensitive disease. Dose-dense and high-dose treatments have been shown to improve the prognosis of patients with Ewing's sarcoma/PNET.[8] This young girl presented with an initially responsive disease, even though it was metastatic at the time of diagnosis. However, she was not able to receive adequate chemotherapy because of thrombocytopenia. This condition resulted in a significant delay of chemo-

Figure 1. Bone marrow showing severe megakaryocytic hypoplasia (10 x magnification, May Grunwald Giemsa)

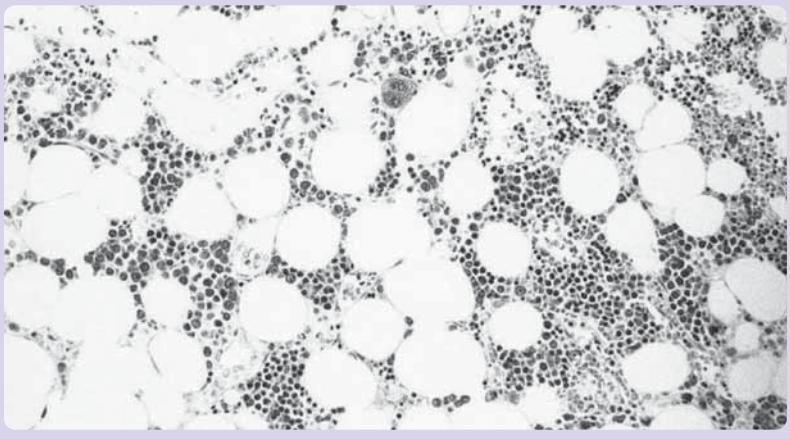
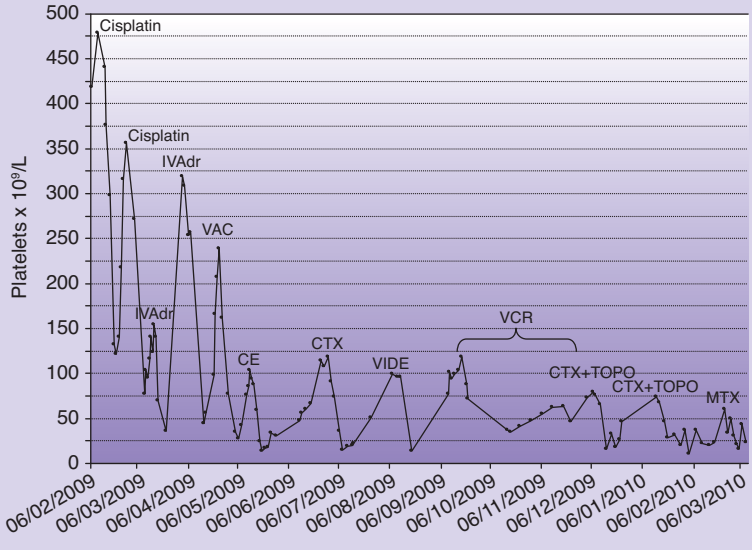


Figure 2. Platelet levels over time and chemotherapy administered



therapy cycles which ultimately affected the clinical outcome. Two different cycles aimed at PBSC mobilization failed to collect enough CD34+ cells and this prevented the administration of high-dose chemotherapy. Such a strategy has been shown to improve clinical outcome.[9]

Beyond the failure to collect PBSC, any further attempt to administer chemotherapy at a reasonable dose intensity failed. Only palliative therapy could be given and the patient subsequently died.

Normal platelet recovery during treatment could have permitted the adequate dose-dense and/or high-dose chemotherapy associated with a higher response rate. Response is a crucial step to progression-free and overall survival. At present, the management of thrombocytopenia is based on platelet transfusion and treatment of the underlying disease. A choice between chemotherapy dose reduction or a delay in subsequent cycles must, therefore, be considered when the platelet count does not recover to acceptable levels. Unfortunately, chemotherapy dose-intensity and dose-density may be important to patient outcome, particularly when more aggressive and potentially curative regimens are used to treat chemosensitive malignancies. The recent development of safe and manageable compounds within the setting of immune thrombocytopenia is a breakthrough. This achievement represents the long awaited opportunity to enhance thrombopoietic support during chemotherapy treatment,[10] as is currently the case with G-CSF and erythropoietin for white and red cell counts, respectively.

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Economic burden of chronic immune thrombocytopenia in Italy

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Abstract

Although chronic immune thrombocytopenia (ITP) incurs substantial medical resource utilization, information on the cost of illness of this disease is still quite scant. This study aims to estimate the economic impact of ITP and to quantify the medical resource utilization of the treatment of ITP in Italy, as well as the costs associated with the management of this condition.

This was a non-interventional, retrospective chart review study of patients diagnosed with chronic ITP. The study was performed in seven Italian hematology centers and lasted 12 months. Data on medical resource utilization were collected.

In total, 158 patients were included in the study. Overall, the results show that that medication and hospitalization costs account for a large part of the economic burden of ITP, with the greatest expenses due to splenectomies and to recurrent hospital admissions. Moreover, the average cost per patient per year increased as the number of previous lines of therapy increased. The cost per patient per year of treating ITP was highly variable, likely because of the unpredictable course of the disease. In particular, the highest cost and the largest use of medical resources were reported in patients who underwent splenectomy during the study period, thus suggesting that the disease management period culminating in splenectomy is highly resource-intensive and expensive.

Background

The epidemiology of chronic immune thrombocytopenia (ITP) is poorly described. The prevalence of ITP in adults and children range from 9.6 to 23.6 per 100,000 persons.[1] The estimated annual incidence of ITP ranges from 1.6 to 3.2 per 100,000 persons.[2,3] In Italy, the estimated incidence in the adult population ranges from 800 to 1600 new cases per year. Women in childbearing age develop ITP twice more frequently than men.[4]

Noteworthy, ITP incurs substantial medical resource utilization because of the onset of bleeding events and the need for hospitalization. However, information on the cost of illness of ITP is still quite scant.

On this basis, this study estimates the economic impact of ITP and quantifies the medical resource utilization of the treatment of ITP in Italy, as well as the costs associated with the management of this condition.

Patients and Methods

Design and patients

This was a non-interventional, retrospective chart review study of patients diagnosed with chronic ITP. The study was performed in seven Italian hematology centers and covered 12 months of chronic ITP treatment history.

Patients with chronic ITP seen by hematologists at each study site in the period 01.01.2005-31.03.2007 were included in the study if they were aged ≥18 and were diagnosed with chronic ITP according to international disease classification. Pregnant women and patients participating in a clinical trial during the observation period were excluded to avoid possible bias in the results.

Data on medical resource utilization were collected, including physician and clinic visits, hospitalization, surgical procedures, treatment for ITP and laboratory tests. Cost analyses were performed from the National Healthcare Service perspective.

Patient subgroups were stratified by their splenectomy status before the observation period (patients splenectomized prior to the inclusion in the study and non-splenectomized patients); non-splenectomized patients at the start of the observation period were stratified

Table 1. Stratification of patients according to treatment before the observation period and medical resource utilization during the observation period

Patient characteristics	N	Splenectomy	Only visit/ laboratory tests	Refractory to splenectomy[b]	Continuous treatment[c]
Splenectomized patients at start of study period[a]	28 (17.72%)	0	7	21	0
Treatment-naïve patients	8 (5.06%)	2	5	0	1
Patients with one line of treatment	67 (42.40%)	4	29	0	34
Patients with two lines of treatment	42 (26.58%)	5	5	1	31
Patients with three lines of treatment	13 (8.23%)	1	3	0	9
Total patients	158 (100%)	12 (8%)	49 (31%)	22 (14%)	75 (47%)

[a]Patients splenectomized prior to the start of the observation period
[b]Patients who did not control the platelet count after splenectomy and who needed retreatment with other medications
[c]Patients who are treated with medications during the observation period

according to the number of treatments received prior to the inclusion (1, 2 and 3 lines of treatment) and/or according to the need for splenectomy.

Medical/resource utilization

Data on resource utilization related to the treatment of chronic ITP or of associated adverse events were collected and analysed. Resource use data from patient medical charts included outpatient visits (to hematology center or other clinicians), emergency room admission, day-clinic, hospitalizations, medications prescribed for ITP, platelet/blood transfusion, splenectomy or ITP-associated adverse events, examinations and procedures performed.

The medical costs were included for outpatient and inpatient charges. In the outpatient setting, ambulatory visits, day-clinic visits, specialist visits, diagnostics, laboratory analysis, procedures and examinations were included. Inpatient costs included hospitalizations costs.

Unit costs for resources were retrieved from official sources: Diagnosis Related Groups (DRGs) for the cost of hospitalization, specialist tariff for physician visits, laboratory analysis and Drug National Formulary for the costs of medications. All costs were adjusted to 2009.

Cost analyses included only resources consumed during the 12-month observa-

Table 2. Average cost per patient per year in different subpopulations

Patients	N	Mean cost/ patient/year (€) (SD)	Range of cost (€)
Patients splenectomized prior to the observation period	28	1,124 (2,156)	4 – 7,054
Treatment-naïve patients without splenectomy	6	98 (105)	4 – 274
Treatment-naïve patients with splenectomy	2	39,008 (18,652)	20,356 – 57,660
First-line patients without splenectomy	63	1,250 (3,284)	4 – 16,317
First-line patients with splenectomy	4	10,034 (5,327)	4,826 – 17,705
Second-line patients without splenectomy	37	2,263 (6,943)	4 – 42,206
Second-line patients with splenectomy	5	42,300 (72,274)	4,573 – 186,784
Third-line patients without splenectomy	12	3,434 (8,684)	27 – 30,391
Third-line patients with splenectomy	1	5,053	n/a

Figure 1. Medical resource utilization during the observation period

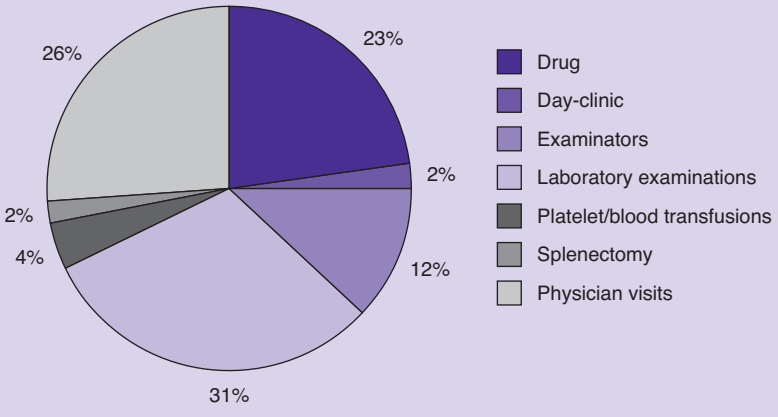
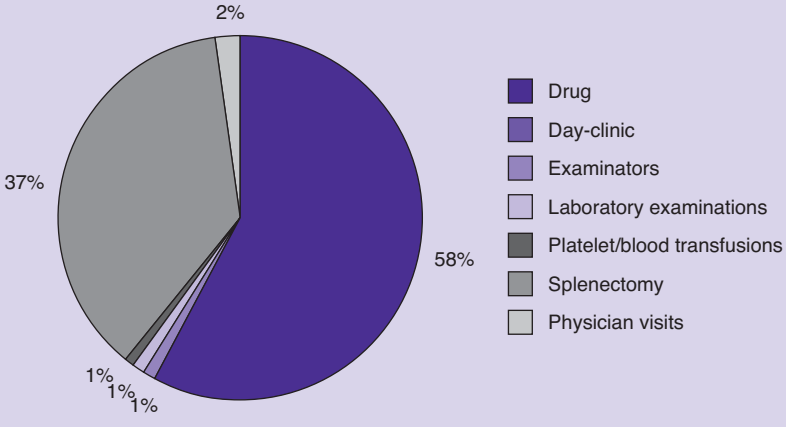


Figure 2. Cost breakdown of medical resource utilization during the observation period



tion period and only related to the treatment of chronic ITP or to the management of ITP treatment-related adverse events. Data were analyzed with descriptive statistics.

Results

Patient characteristics

In total, 158 patients were included in the study (mean age: 55 years, range: 19-89; median time from first diagnosis: 28 months). The stratification of patients according to treatment before the observation period is summarized in Table 1.

Medical resource utilization and costs

Overall, 69% of patients received a treatment during the observation period (Table 1): in particular, 8% were treated with medications for ITP and then underwent splenectomy because of lack of response; 14% were patients refractory to splenectomy who required further treatment; and 47% received medical therapy throughout the observation period and some switched to other treatments, with a more frequent treatment switch in patients on second- and third-line therapy. The remaining 31% of patients were in a 'watch-and-wait' phase. During the year of analysis they received only routine laboratory test or physician examinations; this situation was more prevalent in treatment-naïve patients and in those who received only a single line of treatment. Medical resource utilization during the observation period and the cost breakdown of medical resource utilization are represented in figures 1 and 2, respectively.

When considering the whole population, the mean cost per patient per year was €3,634 (±16,145).

Medical resource utilization cost by subgroups

The analysis of patient subgroups indicated that the cost per patient increased with the number of lines of therapy received before and during the observation period (Table 2). Moreover, cost was higher in patients who underwent splenectomy than in those who did not need this intervention (Table 2).

Limitations

It must be acknowledged that this study presents several limitations. First, it was a retrospective data collection from patient records; therefore, data may be incomplete or errors in data extraction may have occurred. Second, analyses are based on an overall limited number of patients (in particular, the number of patients undergoing splenectomy during the observation period was relatively small), and thus results must be interpreted with caution. Third, from a pharmacoeconomic perspective, DRG reimbursement tariffs are not necessarily a true cost to the health service; moreover, indirect costs, such as limited productivity and cost burden to the patient, were not taken into account.

Discussion

Overall, this study shows that medication and hospitalization costs account for a large part of the economic burden of ITP. The greatest expenses were due to splenectomy interventions and to recurrent hospital admissions for intensive/rescue medica-

tions. Moreover, an increase in the average cost per patient per year was observed as the number of previous lines of therapy increased; the average cost reported in the 12-month period was markedly increased in those patients who had been repeatedly treated before the study initiation. These refractory patients were moved on from corticosteroids and on to the more expensive rituximab and IVIg therapies.

The cost per patient per year of treating ITP varied widely, ranging from €4 to €186,000. This finding is likely due to the variable and unpredictable course of the disease. Further research could explore the average duration of intensive treatment and the cost of any period of intensive treatment, rather than computing the cost of disease management on an annual basis.

The highest cost and the largest use of medical resources were reported in patients who underwent splenectomy during the study period, with an average cost of €27,892 and a range from €4,573 to €186,784. This finding suggests that the disease management period culminating in splenectomy is highly resource-intensive and expensive.

Acknowledgements

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Report from EASL – 45th Annual Meeting of the European Association for the Study of the Liver, Vienna, Austria, April 14-18, 2010

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Abstract

Purpose of review: Platelet count is an important diagnostic and prognostic hematological parameter in patients affected by chronic hepatitis C, although little is known with regard to its relevance during antiviral therapy. In addition, thrombocytopenia can be an obstacle to the performance of invasive procedures in patients with chronic liver disease. This article reviews three recent studies that were presented at the 45th Annual Meeting of the European Association for the Study of the Liver (held in Vienna on April 14 - 18, 2010) that evaluated: 1) the use of an orally-administered thrombopoietin (TPO) receptor agonist in thrombocytopenic patients with chronic liver disease who were undergoing invasive procedures (the ELEVATE study); and 2) the role of platelet count and its modification during antiviral therapy in patients with chronic hepatitis C.

Recent findings: ELEVATE demonstrated that an orally-administered TPO receptor agonist was able to increase platelet count and avoid platelet transfusions in a significantly greater proportion of patients as compared to placebo, but was associated with an increase in the incidence of thrombotic events in some actively-treated patients. Higher baseline platelet counts were associated with early viral clearance of the hepatitis C virus during pegylated interferon and ribavirin therapy in an interim analysis of "real-world" patient cohorts in PROPHESYS, and greater decreases in platelet count during antiviral therapy with pegylated interferon alpha-2a and ribavirin were associated with a greater likelihood of achieving viral eradication at the end of treatment in a retrospective analysis.

Summary: The three studies highlighted in this report describe: 1) the important therapeutic use of orally-administered TPO receptor agonists in patients with thrombocytopenia and chronic liver disease; and 2) underscore the peculiar role of platelet counts before and during antiviral therapy in patients with chronic hepatitis C.

Thrombocytopenia, invasive procedures, and prophylaxis with oral TPO receptor agonists

Thrombocytopenia is the most common hematological abnormality in patients with chronic liver disease,[1] and may hinder the performance of invasive diagnostic and therapeutic procedures.[2,3] Furthermore, the presence of thrombocytopenia has been associated with an increased risk of bleeding after invasive procedures.[4-6] To date, the only therapeutic option available for this condition is prophylactic platelet transfusion, but this treatment may be associated with side-effects and worse outcomes after liver transplantation.[7]

ELEVATE was a multinational study that evaluated the ability of an orally-administered thrombopoietin (TPO) receptor agonist to reduce the need for platelet transfusion in thrombocytopenic (platelet count ‹50 x 10^9/L) patients with chronic liver disease undergoing an elective invasive procedure.[8] After stratification for bleeding risk of the planned procedure, patients were randomised to once-daily treatment with the oral TPO receptor agonist 75 mg or placebo for 14 days prior to their planned invasive procedure, which was performed within 5 days of the last dose of the study drug. The primary study endpoint was the proportion of patients avoiding platelet transfusion before, during and up to 7 days post-procedure; secondary endpoints included the number of bleeding events and platelet counts up to 30 days post-procedure. The study was terminated early due to an imbalance in thrombotic events in the active treatment arm so as to allow a complete analysis of data. Of the 292 enrolled patients, platelet transfusion was avoided by 104 (72%) oral TPO receptor agonist recipients and 28 (19%) placebo recipients

(p<0.0001). Furthermore, the average platelet count at day 14 was 103 x 10^9/L in the oral TPO receptor agonist group compared with 39 x 10^9/L in the placebo group (p<0.0001). Bleeding events were reported in 17% of oral TPO receptor agonist recipients versus 23% of patients in the placebo group, thus meeting the non-inferiority secondary endpoint of the study. The incidence rates of overall adverse events (55% vs. 59%) and serious adverse events (13% vs. 12%) were similar between the oral TPO receptor agonist and placebo groups. However, thrombotic events appeared to be more frequent in the oral TPO receptor agonist group (6 vs. 2 patients; odds ratio 2.827, 95% confidence interval 0.695–11.501). An increase in platelet count >200 x 10^9/L was significantly associated with a greater risk of occurrence of thrombosis. All TPO events in the active treatment arm occurred in the portal vein system. This study confirms the efficacy of an orally-administered TPO receptor agonist in increasing platelet counts in thrombocytopenic patients with chronic liver disease, and shows that its administration may reduce the need for platelet transfusion in a significant proportion of patients who are undergoing invasive procedures. However, further analyses are needed to better characterize the safety profile of this drug in this population of patients.

Platelets and antiviral therapy in patients with chronic hepatitis C

The interplay between platelets and the outcomes associated with pegylated interferon alpha antiviral therapy in patients with hepatitis C virus infection is complex. In these patients, platelet count along with other parameters has been shown to be a significant predictor of disease staging, which may reduce the need for liver biopsy as a diagnostic tool.[9-11] Chronic hepatitis C can be difficult to treat and the use of interferon therapy is often associated with thrombocytopenia, which can lead to dose reductions or treatment discontinuation. Indeed, pegylated interferon and ribavirin therapy may be associated with decreased platelet counts and thrombocytopenia even when these hematological findings are not present at baseline;[12] this may occur as a result of bone marrow suppression and the inhibition of late-stage megakaryocytopoiesis.[13]

PROPHESYS comprised three multinational, non-interventional cohort studies that evaluated the predictive value of virologic response at Week 4 (rapid virologic response, RVR) and Week 12 (early virologic response, EVR) on sustained virologic response to pegylated interferon alpha and ribavirin in treatment-naïve, mono-infected hepatitis C virus patients in a "real-world" setting.[14] An interim analysis of pooled data from all three cohorts (n=5,292) evaluated patients who had a documented baseline and Week 12 serum HCV-RNA result.[14] Main baseline clinical, virologic, and histologic findings of the study patients closely mirrored the picture of those encountered in everyday clinical practice. Accordingly, the rates of RVR and EVR in the various viral genotypes reflected common practice, with genotype 1 patients showing a lower likelihood of RVR (21%) to antiviral therapy compared with genotype 2 (73%) and 3 (66%) patients. At baseline, 13% of the patients had a platelet count of <140 x 10^9/L. Multivariate analyses demonstrated that a baseline platelet count of ≥140 x 10^9/L was independently associated with viral clearance (RVR/complete EVR) in both genotype 1 or 4 (p<0.0001) and 2 or 3 patients (p=0.0241). Interestingly, the predictive value of platelet count was shown to be independent of the presence of cirrhosis, regardless of genotype.

Patients with chronic hepatitis C undergoing pegylated interferon and ribavirin therapy may frequently experience varying degrees of myelosuppression and hematologic abnormalities such as anemia, neutropenia and thrombocytopenia. These parameters often determine the dosing of the drugs and have also been associated with response to antiviral treatment.[15] A retrospective pooled analysis of 1,778 patients from 4 trials was recently conducted to determine the association between virologic response and host pharmacodynamic effects following antiviral therapy with pegylated interferon alpha-2a and ribavirin in patients with chronic hepatitis C.[16] The analysis found that maximum decreases from baseline in hematologic variables and weight loss during therapy were associated with virologic response. After adjusting for drug exposure, only decreases in neutrophils and platelet counts were independently associated with virologic response, indicating that anemia and weight loss may be more susceptible to drug exposure. In particular, cirrhosis-adjusted average maximum decreases from baseline in platelet counts were 76.1 x 10^9/L in non-responders, 89.0 x 10^9/L in sustained virologic responders (p<0.0001 versus non-responders), 92.5 x 10^9/L in relapsers (p<0.0001 versus non-responders), and 88.0 x 10^9/L in patients with breakthrough (p<0.01 versus non-responders). It should also be noted that decreases in platelet counts were still significantly different between patients who achieved a sustained virologic response and those who did not, after adjusting for drug exposure. Racial and ethnic differences were also assessed in this analysis, which showed smaller decreases in both neutrophil and platelet counts in African Americans compared with Latino and non-Latino Caucasians. Therefore, the results of this analysis suggest that there is a close relationship between the myelosuppressive effect of pegylated interferon alpha-2a/ribavirin therapy and maximal induction of the host antiviral state.

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Report from EHA – 15th Annual Congress of the European Hematology Association, Barcelona, Spain, June 10-13, 2010

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Abstract

Purpose of review: This article reviews studies on immune thrombocytopenia (ITP) presented at the 2010 Annual Congress of the European Hematology Association.

Recent findings: Several studies were presented on the second-generation thrombopoietin receptor agonists. Studies included both subcutaneous and oral agents, and many were concerned with safety issues (bone marrow reticulin increase and thrombotic complications in particular) from long-term and extension trials. Both subcutaneous and oral agents demonstrated a good risk-benefit ratio, being effective in chronic patients and without any new safety issues. Studies on a first-generation humanized anti-CD20 monoclonal antibody in ITP were also presented. Very few clinical studies focused on pediatric patients.

Summary: TPO receptor agonists demonstrated durable efficacy and favorable long-term tolerability in the treatment of ITP. Post-marketing registry studies would be desirable. The use of anti-CD20 antibodies may be an option for refractory cases.

Introduction

Primary immune thrombocytopenia (ITP) was the subject of several presentations at the recent meeting of the European Hematology Association (EHA), held in Barcelona, Spain, 10–13 June 2010. In this article we will summarize the most important abstracts for pediatric and adult patients. Similar to the EHA congress and American Society of Hematology meeting held in 2009, studies on second-generation thrombopoietin (TPO) receptor agonists (both subcutaneous and oral), were reported extensively, with a particular focus on safety issues.

Childhood ITP

To analyze the natural course of childhood chronic ITP, a retrospective cohort study from Kyungpook National University Hospital was conducted.[1] The medical records of 244 ITP children from 1988 to 2009 were reviewed. Fifty-nine cases were chronic (male:female ratio: 30:29) in spite of initial treatment with steroids, immunoglobulins (Ig), or anti-D Ig. The mean initial platelet count was 23 x 10^9/L and the mean age at diagnosis was 5.4 (0.4–14.7) years. The mean follow-up period was 5.2 (0.7–16.8) years. At the last follow-up, 21/57 children (36.8%) had a normal platelet count (mean: 207 x 10^9/L [150–317]), with a mean time to recovery of 3.5 (0.7 –8.3) years and the mean age at cure of 8.6 (1.2–18.6) years. Twenty children (35.1%) maintained a mean platelet count of 81 x 10^9/L (50–128), whereas 16 children had a mean platelet count of 24 x 10^9/L (9–45). The latter subgroup was closely observed during follow-up. This paper confirms the possibility of a chronic evolution of ITP in children.

A single-centre prospective randomized study, presented by the Mandelli group (Rome), was designed to evaluate the short-, medium- and long-term efficacy of two doses of prednisone (PDN) in children with untreated severe ITP.[2] A total of 91 patients (40 males, 51 females) aged ›12 months to ‹20 years with previously untreated ITP and platelets ‹20 x 10^9/L (or ›20 x 10^9/L if bleeding was present) were stratified by age (‹10 and ›10 years) and randomized to receive PDN 0.25 mg/kg/day (Arm A) or PDN 2 mg/kg/day (Arm B) for 3 weeks. Treatment was discontinued during the fourth week. Treatment evaluation was performed at the 3rd, 4th and 8th week (short-term), 6th and 12th month (medium-term), 5th year and in October 2009 with a follow-up questionnaire (long-term). Treatment response was defined as: complete (CR) - platelet count ›150 x 10^9/L; partial (PR) - platelet count ≥50 x 10^9/L and ‹150 x 10^9/L; and no response (NR) - platelet count ‹50 x 10^9/L. Relapse was defined as a reduction in platelets to ‹50 x 10^9/L among patients with CR or PR. Forty-

seven patients were randomized to Arm A and 44 to Arm B. The short-term evaluation showed response in 37/44 patients (84%) in Arm B compared with 12/43 patients (28%) in Arm A ($p<0.01$) at the 3rd week. At week 8, the response rate increased to 56% in Arm A, compared with 79% in Arm B (p=0.06). No differences in response rate were observed between Arms A and B at the 6th and the 12th month. Time to response was 5 days to 18 months in Arm A and 5 days to 5 months in Arm B ($p<0.01$). Five patients (Arm A) were lost to follow-up. Relapse occurred in 25/81 (31%) patients (Arm A: 14; Arm B: 11). The median time to relapse in these patients was 11 months (range: 2–88). Thirteen patients (Arm A: 9; Arm B: 4) underwent splenectomy (at 9 months to 10 years) and 14/86 (16%) patients (Arm A: 8; Arm B: 6) developed an autoimmune disorder. By year 5, the number of patients with an overall response was 39/46 (85%) in Arm A and 42/43 (98%) in Arm B (p=0.01). Among the 43 patients who agreed to be interviewed in October 2009 (median time from diagnosis: 17.6 years); 38 (88%) were in CR (equal distribution between Arms A and B). No long-term side effects were observed. In conclusion, high-dose corticosteroids induced a rapid platelet count increase and long-term response in children with ITP.

Adult ITP

Cohort Studies

One study[3] estimated the incidence of isolated thrombocytopenia and ITP in the Asturian population in Spain. The incidence of isolated thrombocytopenia was 138.6/million people/year and the incidence of ITP was 66.8/million people/year. The distribution of ITP according to gender and age was: >60 years – 17 males/10 females; <60 years – 14 males/25 females (p=0.02). The diagnosis was causal in 28.78% of the cases.

A retrospective study in 350 ITP patients (males: 41%; median age: 49 years) was presented.[4] Patient demographics, medical history, current treatments and adverse events (AEs), together with medical resource utilization data, were retrieved from patient medical charts for the 12 months prior to their most recent visit. Prior to the observational period, 35% of patients had been splenectomized and the most frequently reported treatment was corticosteroids. During the observational period, 72% of all patients were treated (corticosteroids: 71% of treatments; Ig: 13%; azathioprine: 5%; splenectomize: 6%). In total, 35% of patients required hospitalization, 8% of them in an intensive care unit. Overall, 89% of hospitalizations were due to ITP (low platelet count: 69%; bleeding: 31%) and 7% were due to AEs associated with ITP treatment. Mean hospitalization duration was 10.3 days. A study[5] reported the results of second-line therapy, reviewing the outcomes of 114 ITP patients treated with second-line therapies, after steroid failure/relapse. Forty-eight patients were re-treated with corticosteroids (response rates: 54% and 23% for CR and PR, respectively; mean duration of second remission: 18 months). Corticosteroids as third (or higher) line of treatment were used in 40 patients with CR (45%) and PR (20%); mean duration of response was 21 months. Thirty-nine patients underwent splenectomy, of whom 36 (92%) responded. Of these 36 patients, 13 (36%) relapsed (median time to relapse: 4 [1–60] months). The 5-year probability of response was 64% with a median duration of response of 120 months (95% confidence interval [CI]: 60–180). Of the patients who relapsed after splenectomy, 5 responded to low-dose steroids. A first-generation humanized anti-CD20 monoclonal antibody was administered in 32 patients as a second- or higher-line treatment (375 mg/m^2 weekly for 4 weeks). Nineteen patients (60%) had a CR, with a mean and median response duration of 31 and 39 months, respectively (95% CI: 24–55). Ten patients (31%) maintained long-term (2 to 5 years) responses to the first-generation humanized anti-CD20 monoclonal antibody. Re-administration of the drug in 3 cases resulted in new responses, of similar duration. No long-term AEs were observed. Vinca alkaloids (vincristine/vinblastine) were used in 46 patients, approximately half of whom experienced some degree of short response (median 5 months). Ten patients received danazol (response rate: 58%, sustained on treatment only). Other treatments among relapsed patients were as follows: anti-Rhesus globulin (n=3), cyclosporine (n=3), cyclophosphamide (n=5) and thrombopoetin analogs (n=2). Finally, in another cohort evaluation,[6] therapy outcome was retrospectively analysed in 168 adults with ITP. Steroids were the first-choice treatment, with a response in 84% of patients; relapse occurred in one-third of cases, and splenectomy was performed in 25 cases.

First-line treatment

Zaja and colleagues[7] reported the results of an observational study examining the activity and safety of high dose dexamethasone (HD-DXM; 40 mg/day for 4 days every 15 days for 3 cycles) in 20 newly diagnosed adult patients with ITP.[7] Median platelet count at baseline was 10 x 10^9/L. Response was evaluated according to the rate of initial OR and CR (defined as platelet level ≥50 x 10^9/L and ≥100 x 10^9/L after the end of the third HD-DXM course) and the rate of relapse. Fifteen patients completed the therapeutic program. The rate of initial OR and CR was 40% for each (8/20). Two of these 8 patients experienced relapse. One patient developed pneumonia during HD-DXM. Thus, HD-DXM did not seem to yield further benefit beyond that expected with standard steroid therapy.

Interim data from an ongoing multicenter study in Denmark were presented.[8] This study is comparing a first-generation humanized anti-CD20 monoclonal antibody + DXM vs DXM monotherapy in newly-diagnosed ITP. Patients ≥18 years of age with confirmed ITP and platelet count ≤25 x10^9/L (or ≤50 x10^9/L in the presence of bleeding symptoms) were randomized 1:1 to receive either DXM (40 mg/day for 4 consecutive days, repeated every 1–4 weeks for up to 6 treatment cycles) in combination with a first-generation humanized anti-CD20 monoclonal antibody (375 mg/m^2 repeated once-weekly for a total of 4 weeks) or DXM monotherapy. The primary endpoint is sustained CR (platelets ≥100 x10^9/L) or PR (platelets ≥50 x10^9/L) at 6 months.

Secondary endpoints include duration of relapse-free time and rates of splenectomy. Ninety-four patients were included in the efficacy and tolerability analysis (median age: 51 years [range: 18–84]; 59% women). Sustained response (CR or PR) at 6 months' follow-up was achieved in 55% and 34% of patients in the first-generation humanized anti-CD20 monoclonal antibody + DXM and DXM monotherapy groups, respectively (p=0.1). No significant difference in splenectomy rates was noted between the two groups (5 patients in the first-generation humanized anti-CD20 monoclonal antibody + DXM group; 2 patients in the DXM group; p=0.3). AEs were mild to moderate. Overall, 74 % in the first-generation humanized anti-CD20 monoclonal antibody + DXM group and 85% of patients in the DXM group reported grade 1–2 AEs (p=0.32). Grade 3 AEs were reported in 9 and 4 patients, respectively; 3 of these patients had 2 events (p=0.32).

TPO receptor agonist: efficacy

The effects of an oral TPO receptor agonist on platelet response, bleeding symptoms and reduction in concomitant ITP medication were evaluated in a 6-month placebo-controlled, phase 3 study in 97 ITP patients with baseline platelet count ≤15 x 10^9L (67 receiving active drug and 30 receiving placebo) as a part of the RAISE trial.[9] Analysis of this subset showed significant clinical benefit from oral TPO receptor agonist treatment; the odds of responding were significantly higher compared with placebo (p<0.0001). Response to oral TPO receptor agonist (75 mg) was observed in 56% of patients who required dose escalation and 10% of patients in the placebo group. The odds of bleeding were significantly reduced with active treatment compared with placebo (p<0.001). Among the 29 patients in the active group who were receiving concomitant ITP medications at baseline, 19/29 (66%) discontinued or reduced at least 1 concomitant ITP medication (vs 4/15 [27%] in the placebo group). Of the 19 active treatment patients, 14 (74%) did not require any rescue medication after a sustained reduction or permanent discontinuation of a baseline ITP medication, compared with one patient (25%) in the placebo group. The incidence of AEs in the overall population was similar between oral TPO receptor agonist and placebo; approximately 90% of patients in each group experienced ≥1 AE. Serious AEs were noted in 9% of patients receiving oral TPO receptor agonist and 30% of patients receiving placebo.

The effect of individualized drug-dose modifications on platelet response was assessed in two phase 3 studies (a 6-week study [TRA100773B] and a 6-month study [RAISE]) in adult patients with chronic ITP.[10] In TRA100773B, the dose of oral TPO receptor agonist was increased from 50 mg to 75 mg for insufficient platelet response (‹50 x 10^9L) in 46% (35/76) of patients; 31% of these patients (11/35) subsequently responded to 75 mg. In RAISE, patients who did not achieve platelet counts ≥50 x10^9/L on oral TPO receptor agonist 50 mg had the dose increased to 75 mg. The percentage of patients receiving the 75 mg dose ranged from 29% to 53% from day 29 to the end of treatment; response to 75 mg was observed after 1 week in ›15% of patients and in 30% to 46% of patients throughout the remainder of the 6-month study. Throughout the study, approximately 20% of patients at each visit were on a ≤25 mg dose. Individualized dosing enabled lower doses in patients sensitive to oral TPO receptor agonist. Dose increases were associated with increased platelet count over a 6-month period.

TPO receptor agonist: safety

The safety of long-term subcutaneous TPO receptor agonist treatment was analyzed using data from 13 clinical studies in patients with ITP (n=718 [59% female], corresponding to over 1000 patient-years).[11] Patients received subcutaneous TPO receptor agonist, placebo, or standard of care (SOC); data from placebo/SOC patients were pooled. The mean platelet count at baseline was 20 x 10^9/L (SD: 16 x 10^9/L). In total, 580 patients received subcutaneous TPO receptor agonist, 65 received placebo/SOC, and 73 received placebo/SOC in the parent study and active treatment in the subsequent study. The median duration of exposure to drug was 52 weeks (range: 1–250 weeks). The proportion of patients discontinuing their parent ITP study was 15% for subcutaneous TPO receptor agonist treated patients and 22% for placebo/SOC-treated patients. AEs were reported in 92% and 94% of actively-treated and placebo/SOC patients, respectively. The most frequent AEs (rates per 100-patient years, drug vs placebo) were headache (79 vs 58), contusion (60 vs 50), and epistaxis (46 vs 53). Reductions in the rate of bleeding events were observed in the subcutaneous TPO receptor agonist group compared with the placebo/SOC group, but the confidence intervals overlapped. AEs of interest in the treatment of ITP with TPO mimetics included thrombosis events, bone marrow reticulin events, neoplasms, and hematopoietic malignancies. The rate of thrombotic events was comparable between the two groups. Bone marrow reticulin was detected in 12 patients who received subcutaneous TPO receptor agonist. In patients for whom post-treatment follow-up biopsies were available (n=4), reticulin grade either decreased or remained the same after drug withdrawal. The rate of all neoplasms and of hematopoietic malignancies was lower with active treatment than with placebo. Two patients developed neutralizing antibodies to subcutaneous TPO receptor agonist but not to TPO. In conclusion, no new safety concerns were identified with the long-term use of subcutaneous TPO receptor agonist.

The possible effect of oral TPO receptor agonist on the increase of reticulin fibres and collagen in the bone marrow of patients with ITP was evaluated in the EXTEND study, an ongoing, long-term open-label ITP study in which patients who had completed previous trials received oral TPO receptor agonist at a starting dose of 50 mg.[12] In this trial, an annual bone marrow biopsy was requested during treatment. Reticulin was quantified by means of the modified myelofibrosis (MF) scale.[13] As of February 1, 2010, 135 patients (median treatment duration at the time of the procedure: 12 months [range: 1–32 months]) had bone marrow biopsies evaluated for reticulin/collagen. A reticulin grade of MF-3 was not observed in any patient. Three patients had collagen. Eleven patients had

a reticulin grade of MF-2, however none showed clinical signs or symptoms that would indicate bone marrow dysfunction. A second on-treatment biopsy was performed after ≥24 months in 11 of the 135 patients. Among these 11 patients, compared to the first on-treatment biopsy, 8 patients had no change in reticulin grade, 1 experienced an increase of MF-1 to MF-2, and 2 experienced a decrease in reticulin grade (MF-2 to MF-0 and MF-1 to MF-0). No biopsies were prompted by clinical symptoms or a blood smear suggestive of MF.

Anti-CD 20 antibodies studies

The efficacy of low-dose first-generation humanized anti-CD20 monoclonal antibody in patients with persistent/chronic ITP was assessed in two studies. The first study[14] evaluated 10 adult ITP patients treated with the first-generation humanized anti-CD20 monoclonal antibody (100 mg weekly) for four consecutive weeks. Median time between diagnosis and start of the first-generation humanized anti-CD20 monoclonal antibody was 1.6 years (0.5–12.4). All patients had received at least one line of therapy (median 2 [1–3]) before initiating the first-generation humanized anti-CD20 monoclonal antibody therapy. These therapies included standard-dose PDN, pulsed HD-DXM, Ig or splenectomy. The median platelet count was 17 x 10^9/L (5–30 x 10^9/L) at the start of treatment with the first-generation humanized anti-CD20 monoclonal antibody therapy. Response was defined as follows: CR - platelet count ≥100 x 10^9/L; PR - ›50 ‹100 x 10^9/L; minimal response (MR) - ›30, ≤50 x 10^9/L; NR - ≤30 x 10^9/L. After completing therapy, platelet count was evaluated after 1 and 3 months, and thereafter every 3 months. After 1 month, 7 responses (4 CR, 2 PR, 1 MR; 70%) and 3 NR (30%) were observed. At month 3, 6 patients had responded (4 CR, 2 PR 60%), 2 patients had NR (20%) and 2 patients had relapsed (20%). At the last control, 6 patients had sustained response (4 CR, 2 PR; 60 %), while 4 had NR. Only one AE (a case of influenza-like illness) was observed.

In the second study,[15] 48 adult patients (median age: 41 years; male:female ratio: 18:30; median platelet count: 30 x 10^9/L), were treated prospectively with intravenous first-generation humanized anti-CD20 monoclonal antibody (fixed dose of 100 mg weekly) for 4 weeks. Results were evaluated according to OR and CR rates (platelet count ≥50 and 100 x 10^9/L), time to OR and CR (TTR and TCR: time to platelet count ≥50 and 100 x 10^9/L), relapse rate, relapse-free survival (RFS: interval between initial response and the loss of the best response previously achieved) and treatment-free survival (TFS: interval between initial response and the necessity to begin rescue therapy). Twenty nine (60%) patients achieved OR and 19 (40%) achieved CR. In responders, the median TTR and TCR were 35 days (range: 7–112) and 51 days (range: 7–150). The median time of observation in responders was 18 months (range: 3–41 months). Relapse was noted in 16/29 responders overall (55%), 10/19 CR (53%) and 6/10 PR (60%). Fourteen patients required further treatment. The cumulative RFS and TFS probabilities were 61% and 70%, respectively, at 12 months, and 45% and 51%, respectively, at 24 months. Treatment was generally well tolerated. Two patients experienced mild chills during the first infusion of the first-generation humanized anti-CD20 monoclonal antibody and 1 patient developed a transient interstitial pneumonia 1 month after the end of first-generation humanized anti-CD20 monoclonal antibody therapy. Compared with standard doses, salvage therapy with low-dose first-generation humanized anti-CD20 monoclonal antibody is associated with similar initial response rates, longer TTR and TCR, worse long-term efficacy and a higher incidence of relapse.

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Report from ASCO – Annual Meeting of the American Society of Clinical Oncology, Chicago, IL, USA, June 4-8, 2010

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Abstract

Purpose of review: This brief report discusses the clinical activity of currently developed thrombopoietins, and highlights recent news from clinical studies evaluating the treatment of chemotherapy-induced thrombocytopenia (CIT), that were presented at the 2010 Annual Meeting of the American Society of Clinical Oncology (ASCO).

Recent findings: Current chemotherapy used to treat solid and hematologic cancer often causes bone marrow arrest and thrombocytopenia. Platelet transfusion is commonly used to manage CIT but it has clinical limitations. A new study evaluating prophylactic recombinant human thrombopoietin (rhTPO) showed that it can attenuate the severity, and shorten the duration, of CIT in advanced cancer patients. The use of genetically engineered human interleukin-11 mutein (mIL-11) is also being evaluated, particularly with regard to its thrombopoietic activity and, compared with standard recombinant human interleukin-11(rhIL-11); there are suggestions that mIL-11 may have potential in reducing CIT. The role of TPO receptor agonists in the treatment of thrombocytopenia in solid and hematologic cancer is still undergoing clinical evaluation.

Summary: Prophylactic rhTPO administered after chemotherapy attenuates severe CIT. mIL-11 has demonstrated good thrombopoietic activity. Several studies evaluating TPO receptor agonists are currently in progress.

Introduction

Chemotherapy-induced myelosuppression results in varying degrees of neutropenia, anemia, and thrombocytopenia and related complications that can lead to hospitalization, and an impaired quality of life.[1] While myeloid growth factors[2] reduce neutropenia and the incidence of neutropenic fever,[3] and erythropoietic agents reduce anemia and transfusions,[4] there still remains a critical need to treat chemotherapy-induced thrombocytopenia (CIT).

Thrombocytopenia increases the risk for bleeding complications,[5] the need for platelet transfusions, and the frequency of chemotherapy dose reductions and/or treatment delays, which may compromise treatment outcomes. Platelet transfusions are also limited by cost, supply, and associated risks including transfusion reactions, infection, alloimmunization and platelet refractoriness. However, the identification of a hematopoietic growth factor that is capable of stimulating platelet production has proved to be an even greater challenge. Since the 1994 discovery of thrombopoietin (TPO), there has been much progress in the clinical development of a number of novel TPO molecules.[6] However, despite extensive clinical development efforts with various thrombopoietic agents in the past decade, recombinant human interleukin-11 (rhIL-11) is the only agent currently approved by the US Food and Drug Administration for the treatment of CIT. Unfortunately, the use of this agent is limited due to its narrow therapeutic index.

Genetically engineered human interleukin-11 mutein

Recently, a group from China compared genetically engineered human interleukin-11 mutein (mIL-11) with standard rhIL-11.[7] Efficacy and tolerability data were presented at the 2010 ASCO meeting but, in general, were disappointing. Indeed, the incidences of mIL-11-related adverse events were lower than those with rhIL-11, but there was no statistically significant difference between the two agents with regard to clinical efficacy.

Recombinant TPOs

As has been previously established, promising biologic activity has been observed with recombinant TPOs in

nonmyeloablative clinical settings; however, clinical development was stopped due to the presence of neutralizing antibodies. More satisfactory results are expected with the new recombinant human thrombopoietin (rh-TPO). Lu and collaborators evaluated the clinical efficacy of prophylactic rh-TPO in patients with advanced non-small cell lung cancer.[8] This approach resulted in a shorter duration of thrombocytopenia after chemotherapy than in the control group, and it also attenuated the severity of thrombocytopenia.[8]

TPO receptor agonists

Recently, a number of novel TPO receptor agonists have been developed with promising clinical activity and a reduced potential for immunogenicity. Several of these second-generation platelet-stimulating agents, including nonpeptide mimetics, are currently in clinical development, and have been shown to be active and safe in autoimmune thrombocytopenia. At the 2010 ASCO meeting, a new session was devoted to clinical trials that are currently in progress. Two ongoing clinical trials are aiming to optimize the dose and schedule of an oral TPO receptor agonist in order to ameliorate chemotherapy-induced thrombocytopenia.[9,10]

Conclusions

In conclusion, there is much enthusiasm for the use of thrombopoietic growth factors, in lieu of platelet transfusions, in patients with cancer and/or hematological conditions, and there is much interest in developing agents that might stimulate platelet shedding from existing megakaryocytes. Although the data from clinical trials presented at ASCO 2010 are currently insufficient to change clinical practice, the results reported are promising and it is likely that, in the near future, new strategies will be available to counteract thrombocytopenia secondary to chemotherapy.

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