Pathogen inactivation/reduction of platelet concentrates: turning theory into practice

B. S. Gathof, M. E. Tauszig & S. M. Picker
Department of Transfusion Medicine, University of Cologne, Cologne, Germany

Background  Pathogen reduction technology (PRT) has been proven to reduce the residual risk of transmission of infectious agents. Reduction of various contaminating bacteriae, viruses and parasites by few to several log steps and efficiency to prevent GVHD has been shown.

Aim  To evaluate and compare advantages and disadvantages of PRT available for practical application in platelets.

Materials and Methods  PRT for the treatment of platelets is currently offered by two formats: Amotosalen (INTERCEPT, Cerus, Concord, CA, USA) and vitamin B2 (Mirasol, Caridion, Denver, USA). Results from different studies and our own experiences with the two techniques are compared and discussed.

Results and Discussion  For both technologies, different groups of investigators have shown acceptable in-vitro results with respect to functional and storage data for platelets stored for up to 5 days after production and before transfusion. Initial clinical studies showed no inferiority of the treated platelets in comparison to untreated controls in thrombocytopenic patients. However for both techniques a tendency towards lower CCI has been reported, which may be more pronounced in the platelets treated with the Intercept process. For introduction of PRT many countries require not only CE mark but licensing with the respective authorities since treatment for pathogen reduction is regarded as creating a ‘new’ blood product. With respect to a platelet loss during pathogen reduction it seems recommendable to increase the lower limit of platelet content of the product to $2.5 \times 10^{11}$. Particularly for the Intercept system, where a considerable amount of platelets is lost in the purification of the product from Amotosalen, a change in the production process to increase the platelet yield may be necessary. Data from our group show a tendency for improved functional and storage parameters for platelets treated with the Mirasol process. Compared to conventional manufacturing of platelets by apheresis or pooling of buffy coats, pathogen reduction requires additional labour, space, and quality control. Shelf life of platelets is limited in most countries because of the risk of bacterial contamination (in Germany presently to 4 days). A prolongation to 5 or more days after pathogen reduction seems feasible but remains a topic for future studies.

Conclusion  Results of in vitro and clinical studies of pathogen reduced platelets are promising. Larger clinical trials will help to determine whether PRT proves to be beneficial (reduction of transmission of infections, less alloimmunisation) and overall cost effective (bearing in mind that additional costs may be compensated for by omission of gamma irradiation and potential longer shelf life).

Key words: bacterial contamination of platelets, INTERCEPT Blood System, Mirasol-PRT, pathogen reduction technology.

Correspondence: Birgit S. Gathof, Department of Transfusion Medicine, University of Cologne, Cologne, Germany
E-mail: birgit.gathof@uk-koeln.de
The transfusion of blood products is a cornerstone of medical care. However, the threat of disease transmission casts a shadow on its therapeutic benefits. Through years of research and test development, the risk of transfusion-transmitted infections has dropped enormously. Improvements have largely been based in the areas of donor selection, serologic and nucleic acid testing, leukoreduction, bacterial testing, skin sterilization and aseptic phlebotomy techniques.

Currently, the most important risk of platelet (PLT) transfusion is bacterial growth in the product due to storage at 22°C, the temperature at which bacteria can quickly multiply [1]. Bacterial contamination in PLT transfusions may cause about one fatality in every 60,000 transfusions, a 50-fold greater risk than for red-blood-cell (RBC) transfusions [2,3]. Severe sepsis is estimated to occur in about 1 per 20,000 transfusions, and bacterial contamination, more pronounced after longer storage [4], is at 1 of 3000 units [1]. Routine sterility testing seems to be an imperfect tool. Because of the small number of bacteria in a freshly donated unit, the sample tested for sterility may not actually contain any bacteria, and thus, produce a false-negative result [5]. In addition, bacterial culture results are often not available until a product has already been transfused, as shelf life is limited to 4–5 days depending on specific country’s recommendations [6,7].

In addition, the potential for newly emerging pathogens or mutations not yet detectable by current testing practice could endanger the blood supply. West Nile virus, SARS, Dengue virus, Chikungunya virus, Trypanosoma cruzii parasite and Plasmodium falciparum parasite represent present examples [8]. The epidemic of Chikungunya virus in La Reunion, France in 2005–2006 caused far reaching consequences and massive mobilization to maintain the blood supply. Furthermore, despite advances in nucleic acid and serologic testing of known pathogens such as HIV and the Hepatitis viruses, mutations may occur which escape current test.

Historically, the transfusion medicine community has responded to emerging infectious disease by adding new donor deferral criteria and screening tests. However, this kind of reaction may no longer be supportable in the face of a dwindling donor supply and ever-increasing costs of laboratory analyses.

Pathogen inactivation/pathogen reduction (PI/PR) technologies provide a potential solution to these issues. Furthermore, and of huge practical consequence, the storage life of PLT products could be increased from 4 to 5 days to 7 or more days. This would, in turn, prevent many units from expiring and reduce the lack of PLT products. In theory, there are several methods for pathogen inactivation that can be used for individual fractionated components of whole blood—red blood cells, PLTs and plasma. In practice, however, governing body approval (FDA in the United States, PEI in Germany and EMEA in Europe) has only been given to plasma products and PLT concentrates (PC).

The ideal PR/PI technology would eliminate all types of pathogens, whether viral, bacterial, fungal, protozoan or even prion. It would cause no damage to cellular and molecular elements of transfused blood. Furthermore, a technology, which implicates the addition of a chemical compound, would cause no long-term toxic, mutagenic or carcinogenic effects in the recipient nor would it cause neoantigenicity. So far no technology has been able to meet all of these demands. However, extensive investigations and clinical studies have led to market approval for platelet PLT/PR in Europe, as noted by the CE (Conformite Europeenne) mark. This article will concentrate on market approved or near-market approval technologies.

**Practical aspects of PR/PI**

Two different technologies, based on heterocyclic compounds with photoillumination, have been developed and studied in an attempt to bring PR/PI into practice for PC.

**Psoralen-based technology**

**Method of action**

The system developed by INTERCEPT (Cerus, Concord, CA) currently has market approval in Europe. Platelets are suspended in approximately 65% platelet additive solution (PAS) (InterSol, Fenwal, Inc, Lake Zurich, IL) and 35% plasma, to which amotosalen, a synthetic psoralen, is added. The product is illuminated with 3 Joules (J)/cm² of UV-A light at 320–400 nm, causing amotosalen to irreversibly cross-link complementary nucleic acids (either DNA or RNA), thus preventing replication and eventual elimination of the pathogen. The residual amotosalen and byproducts must be adsorbed via the Compound Adsorption Device (CAD) for 4–16 h, to minimize toxicity [9].

**Results of clinical trials**

INTERCEPT platelets were examined for several in vivo parameters in clinical trials. In the euroSPRITE trial, as reported by van Rhenen *et al.* in 2003, thrombocytopenic patients (*n* = 103) were double-blindly randomized to either a standard buffy-coat PLT preparation or an amotosalen-UVA-treated PLT arm. The goal was to examine the efficacy of the photochemically treated product in terms of a corrected count increment (CCI), while also monitoring haemostasis and adverse events. This trial showed at 1 h and 24 h no statistically significant difference in CCI; although a tendency in favour of the controls was shown. Time between transfusions, clinical haemostasis,
haemorrhagic and thrombotic adverse events were similar between test and control groups [10].

In the SPRINT trial, a US-based, prospective, randomized study haemostasis in thrombocytopenic patients with WHO grade 2 or higher bleeding was examined. Adverse events and CCI were also monitored in this two-arm trial \((n = 645)\). No difference in final haemostasis between the two arms was observed. However, 1- h CCI and total number of transfused products varied significantly, favouring the control arm. Closer examination revealed that products treated with amotosalen-UVA had a lower concentration of platelets \(<3 \times 10^{11}\). Interestingly, the amotosalen-UVA-treated platelets were found to be associated with a lower number of transfusion reactions than controls [11–13]. Cerus, manufacturer of INTERCEPT, also performed a radiolabelled PLT study in healthy volunteers comparing its product to a gamma-irradiated control. Results from these studies showed a 15–20% decrease in photochemically treated platelets. A lower CCI, however, did not result in a difference in bleeding, when compared to control [14,15].

Interim data on a study examining the clinical effectiveness and safety of pooled PC in Haemato-Oncology patients were recently released by Kerkhoffs et al. The multicenter randomized controlled trial was divided into three arms: PLT stored in plasma, PLT stored in PAS III and PLT stored in PAS III and treated with amotosalen-UVA \((n = 199)\). Results revealed a significant decrease in 1-hour CCI of 34.2% as well as 24-h CCI of 33.5%. Furthermore, 24 patients in the amotosalen-UVA arm experienced bleeding, when compared to 14 in the plasma control arm. The study was halted prior to expectation in light of these results, with the conclusion that amotosalen-UVA-treated platelets showed significant inferiority [16].

**Research aspects**

Human PLTs, although terminally differentiated and enucleated, are not completely free of nucleic acids. Messenger RNA is detectable in the cytosol, and functional proteins of the respiratory chain are coded by mitochondrial DNA. Thus, many essential platelet functions could be affected by use of PR/PI techniques.

Because psoralens bind non-covalently to plasma proteins and lipids, the plasma ratio must remain within well-defined limits to ensure optimal efficiency of PR/PI. In addition, the performance can be affected by red-blood cells because of haemoglobin-related UVA light absorption. Therefore, INTERCEPT requirements are more stringent than those demanded by national guidelines: 30–45% for plasma ratio and \(<4 \times 10^9\) for RBC contamination, 2.5–5.0 \(\times 10^{11}\) for platelet dose and 300–390 ml for volume [17]. We conducted a two-arm *in-vitro* study, comparing these quality parameters of pooled PCs. Amotosalen-UVA-treated platelets \((n = 25)\) were tested against controls \((n = 25)\). After 7 days of storage, the PLT count decreased by 7–10% in both study arms, a finding confirmed by multiple research teams [17–20]. The PLT dose in amotosalen-UVA-treated PLTs is lower in part because of repeated transfers into different containers and to CAD filtration. Compared to controls, there was an 11% loss of volume and platelets alike in treated units [21]. We suggested to increase the PLT to approximately \(3.4 \times 10^{11}\) before amotosalen-UVA treatment to achieve the required platelet dose \((3 \times 10^{11})\) per unit.

This finding may serve as an explanation for problems observed in the initial clinical evaluations such as the reduction of CCI and haemorrhagic complications. It has been shown that these issues may be overcome by increasing initial platelet concentration or transfusing more PLT units. However, on a practical level, it would cause a burden on the limited PLT supply and also increase costs. In addition, with the more stringent requirements of the INTERCEPT system, there would be an additional loss of units that would otherwise have been available to patients. In the aforementioned study, 6 units of 25 (25%) initially fulfilled European quality requirements before but not after INTERCEPT treatment [21].

The effects of INTERCEPT on *in-vitro* parameters to detect platelet storage lesion have been examined. Research shows that amotosalen-UVA treatment increases cellular activation, cytokine liberation, as well as glycolytic flux thereby decreasing the final pH of the product. This may have caused impaired mitochondrial-based respiration, resulting in significantly lower maintenance of ATP and cell viability, thereby also lowering final PLT counts [22].

**Experience in routine use**

Greater than 300 000 PLT units treated with INTERCEPT technology have been transfused in several hospitals in some European countries [23]. A respective hemovigilance programme reported equal clinical tolerance and safety profile for treated and untreated PLTs. There were fewer incidences of acute transfusion reactions, possibly because of the use of PAS replacement of donor plasma [24,25]. Another study showed that there were a comparable number of units transfused prior to INTERCEPT implementation and thereafter in similar patient groups [26]. These results are supported by similar independent studies in France and Germany [27,28].

**Riboflavin-based technology**

Another CE marked system is examined in clinical studies (CaridianBCT Biotechnologies Mirasol Platelets, Lakewood, CO). This system is based on riboflavin, an essential nutrient, also known as vitamin B2. PLTs are suspended in
plasma or plasma/PAS combination. After the addition of riboflavin, 6.23 J/cm² of UV light is added for 10 min, causing irreversible damage to the nucleic acids [8]. In contrast to INTERCEPT, no postillumination removal step is required.

Riboflavin is most effective for lipid-enveloped viruses. West Nile virus and the non-enveloped parvovirus B19 also showed reductions, as did some bacteria and protozoa [29]. Because of its function as vitamin and many historic studies on toxicity levels, there are few concerns about potential toxicity in minute amounts. Yet, studies were conducted to further explore the coactivity of riboflavin with UVB light in terms of toxicity, revealing no differences between test and controls [30].

**Results of clinical trials**

Riboflavin-UVB-treated PLTs have also been tested in clinical trials, although not as extensively as the amotosalen-UVA-treated ones. In one of the larger studies, radio-labelled, riboflavin-UVB-treated cells were reinfused into healthy donors after 5 days of storage. Testing for viability revealed that the treated PLTs were up to 25% lower than controls but remained within the acceptable range. In addition, no adverse events were noted with the use of treated PLTs [31].

A clinical trial in France (n = 80) revealed that patients receiving riboflavin-UVB-treated PLTs had a lower 1-h CCI, although not at 24 h. Also, more transfusions were noted in the treatment arm. However, all other remaining variables such as number of PLT transfusions per patient, total PLT dose, per cent refractory patient, number of red cells per patient, number of bleeding events and serious adverse events were not statistically different, when compared to controls [32].

**Research aspects**

The effect on Mirasol treatment on platelet in vivo parameters has been extensively studied, because of concern of effect on integral messenger RNA and mitochondrial DNA necessary for proper PLT function. Parameters used to study PLT storage lesion development were examined. The molecular basis of PLT storage lesion is considered to be related to inadequate metabolic support, PLT activation and/or the accumulation of bioactive substances released from contaminating white-blood-cells or PLTs.

In a direct comparison of the INTERCEPT and Mirasol processes that also included a control arm, the amotosalen-UVA-based technique was associated with impaired mitochondrial-based respiration [22]. This, as discussed earlier, caused significantly lower intracellular ATP content and impaired maintenance of cell viability. Although storage variables clearly showed effects of riboflavin-UVB treatment, apheresis PLTs retained cell quality during 5 days of storage without loss of mitochondria-based oxidative respiration. Riboflavin-UVB treatment increased glycolytic flux as well as respiratory/ enzymatic activity but did not alter functional cell or mitochondrial integrity. This is in direct contrast to amotosalen-UVA treatment. Because of increased glycolytic flux, there was evidence for accelerated glucose consumption, thereby increasing PLT acidity because of higher lactate production rates [22,33]. Furthermore, the riboflavin-UVB-treated platelets remained comparable to untreated controls throughout 7 days of storage for ATP maintenance and in-vitro function (swirl, HSR, aggregation, annexin A5 release, adhesion under flow conditions).

**Experience in routine use**

Practical experience with the riboflavin-UVB technology is limited. Several hundred Mirasol-treated PLT products have been transfused in hospital patients, and no adverse reactions have been reported [8]. Additional clinical studies are expected for 2010/2011 in different European countries.

**Common practical aspects of INTERCEPT and Mirasol**

Universal leukoreduction has been introduced in Europe countries around the year 2000. In many other countries and the United States, it has been a topic of debate. Many centres favour removing residual white-blood-cells (WBC) because it reduces the risk of postoperative infection and improves post-transfusion survival through reduction in transfusion-related immunomodulation (TRIM) [34]. Leukoreduction is also beneficial to patients prone to receive multiple transfusions or a transplant as there are lower rates of alloimmunization, decreased episodes of refractoriness to platelet transfusions and fewer febrile reactions [35,36]. Multiple studies have proven that leukoreduction prevents CMV transmission [37]. Leukoreduction by filtration is commonly used, reliable, yet time consuming. Some research teams have investigated the possibility of using PL/PR systems as a means of reducing or inactivating leukocytes within the PC. Specifically, the riboflavin-UVB treatment is able to functionally prevent immunological consequences because of WBC presence. Mirasol PRT treatment inhibited activation in response to stimuli or allogeneic stimulator cells. Mirasol PRT treatment also prevented the cells’ ability to act as antigen-presenting cells and the ability to produce cytokines in response to stimuli such as LPS or anti-CD3+ anti-CD28 [38]. Clinical research observed fewer acute transfusion reactions in INTERCEPT-treated PLTs [39].

An additional benefit of PL/PR would be for the prevention of graft-versus-host disease. Presently, PLTs are gamma irradiated to prevent transfusion-associated (TA)-
GVHD. Although the benefits are proven, this practice can be associated with problems including neglect to irradiate, relabelling, time, cost and increasing security requirements. In a study on the effects of riboflavin-UVB treatment on CD3+ cells in Rag2−/−γ−− double knockout mice (xenogeneic), GVHD induction rates in Mirasol-treated and untreated cells were determined. 85.7% of control mice (12/14) developed xenogeneic GVHD, compared to 0% (0/14) of riboflavin-UVB-treated mice [40]. A review article summarizes that in-vitro and clinical practice observations have shown that non-irradiated yet INTERCEPT- and Mirasol-treated blood components do not cause TA-GVHD [41].

Conclusion

The inevitability of emerging pathogens coupled with the continuing risk of bacterial contamination of blood products is stimulating new methods to increase safety of blood supply. PI/PR technologies are pro-active, rather than the historical reactive strategies. Turning theory into practice is filled with challenges. Laboratory research has shown that platelets counts are lowered by the procedure, most likely because of a combination of apoptosis, platelet storage lesion, as well as loss consequent to processing. This, in turn, has translated into lower corrected count increments (CCI) and higher transfusion rates in some clinical studies. However, PI/PR holds promise of many practical advantages such as immediate protection from emerging pathogens before identification and defence against bacterial contamination, thus leading to an improved safeguard of the blood supply. In addition, there are advantages such as the elimination of gamma-irradiation of platelets, possibly prolonged storage of platelets for up to 7 days and decreased adverse transfusion reactions. Thus, implementation into routine use may confer manifold rewards but will first necessitate further study, both in the laboratory as well as the hospital.

Disclosures

None.

References

1 Hillyer C, Josephson C, Blajchman M et al.: Bacterial contamination of blood components: risks, strategies, and regulation. Joint ASH and AABB Educational Session in Transfusion Medicine. Hematology 2003; 575–589
2 Brecher ME, Hay SN: The role of bacterial testing of cellular blood products in light of new pathogen inactivation technologies. Blood Therapies Med 2003; 3:49–55
3 Blajchman MA: Incidence and significance of the bacterial contamination of blood components. Dev Biol 2002; 108:59–67
4 Yomotovian R, Lazarus HM, Goodnough LT et al.: A prospective microbiologic surveillance program to detect and prevent transfusion of bacterially contaminated platelets. Transfusion 1993; 33:902–909
5 Montag T: Strategies of bacteria screening in cellular blood components. Clin Chem Lab Med 2008; 46:926–932
6 Council of Europe: Guide to the preparation, use and quality assurance of blood components, 8th edn. Strasbourg, Council of Europe Publishing, 2002
7 Lockwood WB, Leonard J, Liles S: Chapter 9: Storage, Monitoring, Pretransfusion Processing, and Distribution of Blood Components. Technical Manual, 16th edn. AABB, pp. 283–298
8 Stramer S, Hollinger FB, Katz LM: Emerging infectious disease agents and their potential threat to transfusion safety. Transfusion 2009; 49(Suppl.):15–29S
9 Lin L, Hanson CV, Alter HJ et al.: Inactivation of viruses in platelet concentrates by photochemical treatment with amotosalen and long-wavelength ultraviolet light. Transfusion 2005; 45:580–590
10 van Rhenen D, Gulliksson H, Cazenave JP et al.: Transfusion of pooled buffy coat platelet components prepared with photochemical pathogen inactivation treatment: the euroSPRITE trial. Blood 2003; 101:2426–2433
11 McCullough J, Vesole DH, Benjamin RJ et al.: Therapeutic efficacy and safety of platelets treated with a photochemical process for pathogen inactivation: the SPRINT trial. Transfusion 2005; 45:1864–1875
12 Snyder E, McCullough J, Slichter SJ et al.: Clinical safety of platelets photochemically treated with amotosalen HCl and ultraviolet A light for pathogen inactivation: the SPRINT trial. Transfusion 2006; 46:24–33
13 Murphy S, Snyder E, Cable R et al.: Platelet dose consistency and its effect on the number of platelet transfusions for support of thrombocytopenia: an analysis of the SPRINT trial of platelets photochemically treated with amotosalen HCl and ultraviolet A light. Transfusion 2006; 46:24–33
14 Snyder E, Raife T, Lin L et al.: Recovery and life span of 111indium-radiolabeled platelets treated with pathogen inactivation with amotosalen HCl [S-59] and ultraviolet A light. Transfusion 2004; 44:1732–1740
15 Slichter SJ, Raife TJ, Davis K et al.: Platelets photochemically treated with amotosalen HCl and ultraviolet A light correct prolonged bleeding times in patients with thrombocytopenia. Transfusion 2006; 46:71–740
16 Kerkhoffs JH, Te Bockhorst PA, Schipperus MR et al.: Clinical effectiveness and safety of pooled, random donor platelet concentrates, leucoreduced and stored up to seven days in either plasma or additive solution with and without pathogen reduction in hematopoietic patients. Abstract. Scientific Section. AABB. Transfusion 2009;49 (Suppl. 2A): 2311–2318.
17 van Rhenen D, Vermeij J, Mayaudon V et al.: Functional characteristics of S-59 photochemically treated platelet concentrates derived from buffy coats. Vox Sang 2000; 79:206–214
18 Janetzko K, Klinger M, Mayaudon V et al.: Storage characteristics of split double dose platelet concentrates derived from

© 2010 The Authors.
Journal compilation © 2010 International Society of Blood Transfusion, ISBT Science Series (2010) 5, 114–119
apheresis and treated with amotosalen-HCl and UVA light for pathogen inactivation. *Infusion Therapy and Transfusion Medicine* 2002; 29:193–198

19 Knutsen F, Alfonso R, Dupuis K et al.: Photochemical inactivation of bacteria and HIV in buffy-coat derived platelet concentrates under conditions that preserve in vitro platelet function. *Vox Sang* 2000; 78:209–216

20 Lin R, Alfonso R, Behrman B et al.: Photochemical treatment of platelet concentrates with a novel psoralen and UVA to enhance the safety of platelet transfusions. *Infusion Therapy and Transfusion Medicine* 1998; 25:39–48

21 Picker SM, Speer R, Gathof BS: Evaluation of processing characteristics of photochemically treated pooled platelets: target requirements for the INTERCEPT Blood System comply with routine use after process optimization. *Transfus Med* 2004; 14:217–223

22 Picker SM, Schneider V, Oustianskaia I, Gathof BS: Cell viability during platelet storage in correlation to cellular metabolism after different pathogen reduction technologies. *Transfusion* 2010; in press

23 Cerenus Presentation. ISBT (Nagoya, Japan). November 15, 2009

24 Osselaer JC, Messe N, Hervig T et al.: A prospective observational cohort safety study of 5106 platelet transfusions with components prepared with photochemical pathogen inactivation treatment. *Transfusion* 2008; 48:1061–1071

25 Osselaer JC, Cazenave JP, Lambermont M et al.: An active haemovigilance programme characterizing the safety profile of 7437 platelet transfusions prepared with amotosalen photochemical treatment. *Vox Sang* 2008; 94:315–323

26 Osselaer JC, Doyen C, Defoin L et al.: Universal adoption of pathogen inactivation of platelet components: impact on platelet and red blood cell component use. *Transfusion* 2009; 49:1412–1422

27 Cazenave JP, Waller C, Mendel I et al.: Clinical experience with conventional versus INTERCEPT platelet concentrates transfused to all patients during two-one-year periods: reduction of adverse events with equivalent use of blood products. *Vox Sang* 2008; 95(Suppl.):305–306

28 Schlenke P, Hagenah W, Andresen S et al.: Platelet components treated with pathogen inactivation using the INTERCEPT Blood System: clinical outcomes of the first single-centre clinical trial in Germany. *Vox Sang* 2007; 93(Suppl. 1):171

29 Ruane PH, Edrich R, Gampp D et al.: Photochemical inactivation of selected viruses and bacteria in platelet concentrates using riboflavin and light. *Transfusion* 2004; 44:877–885

30 Reddy HL, Dayan AD, Cavagnaro J et al.: Toxicity testing of a novel riboflavin-based technology for pathogen reduction and white blood cell inactivation. *Transfusion Medicine Review* 2008; 22:133–153

31 AuBuchon JP, Herschel L, Roger J et al.: Efficacy of apheresis platelets treated with riboflavin and ultraviolet light for pathogen reduction. *Transfusion* 2005; 45:1335–1341

32 Goodrich R, Roberts T, Föllea G: Clinical evaluation of Mirasol PRT treated apheresis platelets in thrombocytopenic patients. (Abstract). *Transfusion* 2008; 48:1061–1071

33 Picker SM, Oustianskaia I, Schneider V, Gathof BS: Functional characteristics of apheresis-derived platelets treated with ultraviolet light combined with either amotosalen-HCl (S-59) or riboflavin (vitamin B2) for pathogen reduction. *Vox Sang* 2009; 97:26–33

34 AuBuchon JP: Chapter 20: Hemotherapy Decisions and Their Outcomes. Technical Manual, 16th edn. Bethesda, MD, AABB, 2008. 569–611.

35 Leukocyte reduction and ultraviolet B irradiation of platelets to prevent alloimmunization and refractoriness to platelet transfusions. The Trial to Reduce Alloimmunization of Platelets Trial Study Group. *N Engl J Med* 1997; 337:1861–1869.

36 King KE, Shirey RS, Thoman SK et al.: Universal leukoreduction decreases the incidence of febrile nonhemolytic transfusion reactions to RBCs. *Transfusion* 2004; 44:25–29

37 Preiksatis J: The cytomegalovirus-“safe” blood product: is leukoreduction equivalent to antibody screening? *Transfusion Medicine Review* 2000; 14:112–136

38 Fast LD, DiLeon G, Li J et al.: Functional inactivation of white blood cells by Mirasol treatment. *Transfusion* 2006; 46:642–648

39 Cazenave JP: Presentation at ISBT macao 2008. The european experience: intercept platelets in routine use. http://ne.edge-castcdn.net/000380/2008/Segment2/segment2_final.swf

40 Fast LD, DiLeon G, Cardarelli G et al.: Mirasol PRT treatment of donor white blood cells prevents the development of xenogeneic graft-versus-host disease in Rag2-/- gamma c-/- double knockout mice. *Transfusion* 2006; 46:1553–1560

41 Mintz PD, Wehrli G: Irradiation eradication and pathogen reduction. Ceasing cesium irradiation of blood products. *Bone Marrow Transplant* 2009; 44:205–211

© 2010 The Authors. Journal compilation © 2010 International Society of Blood Transfusion, *ISBT Science Series* (2010) 5, 114–119