

Minutes

XXIXth Meeting of the Biomedical Excellence for Safer Transfusion Collaborative

Rome, Italy
March 11-12, 2005

These minutes represent presentations and discussions at the BEST meeting on March 12 as well as a synopsis of the team meetings held immediately before. They include some detail and discussions that occurred only in the team meetings in order to provide all members with as much information as possible about past and planned studies. Additional details can be obtained from the leader of each team, who prepared the summary of the team's activity given below, often with assistance from presenters.

Jim AuBuchon opened the meeting with a welcome to all attendees. First-time attendees were introduced, and a special welcome was extended to NovoNordisk as BEST's newest manufacturer member. He also extended heartfelt thanks to Dr. Jane Hassan of the Istituto Superiore di Sanità and her team their assistance in arranging the logistics of the meeting. The participation in the meeting by F. Decary, ISBT president, and W. Mayr, editor-in-chief of *Vox Sanguinis*, were noted with appreciation.

The BEST Band's performance during the BEST dinner was also noted with thanks. P. Rebullia, S. Murphy, T Takahashi, C Smit Sibinga and G. Andreu – along with the dancing of J. Miripol and M. Contreras - made it a memorable evening indeed!

S. Murphy asked AuBuchon to provide the financial report. The group was referred to the updated financial information in the BEST book that had been distributed. No financial problems were noted. BEST now has 15 manufacturer members.

In response to a question, AuBuchon clarified that BEST was now incorporated as a non-for-profit corporation in the State of New Hampshire and had applied for 501(c)(3) status with the US IRS. BEST operates independently of the ISBT. This may (or may not) be of financial benefit to manufacturers.

AuBuchon reviewed for the group the leadership renovation and membership renovation processes that would begin with the next meeting. At BEST XXX, Manufacturer Members and Scientific Members will elect 8 of their own to serve on the Nominating Committee. This Committee will provide a slate of team leader and chair candidates for voting on at BEST XXXI (Spring, 2006). The new chair and team leadership group will select the renovated list of Scientific Members around the time of the BEST XXXII meeting (September, 2006). The new leadership and renovated BEST group will hold its first meeting as BEST XXXIII (Spring, 2007). Those interested in serving on the Nominating Committee were asked to let AuBuchon know of their interest.

A further step in team leadership evolution was noted as occurring with this meeting. G. Moroff had expressed his desire to step down from Cellular Therapy team leader responsibilities after this meeting, to be replaced in team leadership by Z. Szczepiorkowski. However, a viral illness prevented Moroff from attending the meeting, so the transition was occurring a bit earlier than anticipated. AuBuchon thanked Moroff for all of his contributions to BEST and noted that appropriate recognition would be planned for the next meeting at which Moroff could attend.

The next BEST meeting (XXX) will be held immediately after (rather than before) the AABB Annual Meeting (in order to avoid conflicts with the Jewish holy days). Team meetings will be Wednesday, October 19, the BEST dinner will be that evening, and the BEST meeting (and Nominating Committee elections) will be on the morning of October 20. With the shortened BEST meeting schedule, noon departures should be possible. The meeting location will not be the Seattle Hyatt as originally anticipated, and another downtown location (close to the convention center) is being sought. The location will be posted on the website as soon as available.

BEST XXXI will be approximately one year from now. M. Luzano has been able to identify new contacts for us that may allow us to set up a meeting in Barcelona. If this is not feasible, another European site will be identified.

BEST XXXII will be immediately before the ISBT Congress in Cape Town, South Africa.

AuBuchon explained the new (shortened) format of the BEST meeting. Team leaders would be providing brief 20-minute summaries of their teams' meetings to avoid repetition of the previous presentations. This would also provide additional time (at lunch) for discussions and informal planning. (At this meeting, some of this time was utilized for a demonstration of the new BEST website (www.bestcollaborative.org) by the web designer, Eileen Selogie of eNetAnswers, who happened to be in Rome and offered to provide this assistance.) Comments on the revised meeting format were encouraged.

AuBuchon provided a brief overview of the content and navigation of the BEST website. Suggestions for improvements were encouraged to be submitted to him. If members wish to use the website for other purposes, such as surveys, they were encouraged to discuss the idea with the appropriate team leader. Presentations at BEST will be posted in the Members Only section of the website as pdf files. AuBuchon expressed appreciation for presenters' willingness to make their files available. All were cautioned that the slides and the data they contained remained the intellectual property of their creators and must not be copied or distributed beyond BEST without the creator's approval. This is particularly important since many slide sets contain original data that have not yet been published.

S. Dzik was applauded for his interesting puzzler that demonstrated many important and early advances in medicine – including transfusion medicine – that had occurred in Italy. The answers to the puzzler were (1) Giovanni Riva; (2) Francesco Folli; (3) Michele Rosa. The tie breaker pertained to the movie *Roman Holiday* starring Gregory Peck and Audrey Hepburn. Winners included M. Lozano, N. Heddle, C. Prowse, Z. Szczepiorkowski and A Thomson (with other partially correct responses). M. Blajchman noted the John Hess' puzzler from the Frankfurt meeting has been transformed into the first of a series of historical vignettes he hopes to see submitted to *Transfusion Medicine Reviews*; he encouraged others to write up their puzzlers or to write (auto)biographies to capture the history and personages of transfusion medicine.

J. Hess reported that BEST had been elected to the Global Collaborative for Blood Safety. The executive Committee will consider what role we may be able to play in this organization.

Report: Transfusion Safety Team

S. Dzik, M. Murphy

The Transfusion Safety team began by welcoming new members to BEST including Silvano Wendel (Sao Paolo, Brazil), Amanda Thomson (Sydney, Australia) and Alan Tinmouth (Ottawa, Canada as alternate for Paul Hébert). The meeting began by a review of the Canadian Consensus Conference on TRALI presented by Dr Morris Blajchman. The Canadian group is developing a registry of TRALI cases and Dr Blajchman suggested that BEST may wish to consider a future project to coordinate and consolidate various TRALI registries that are being established in different regions—including UK SHOT program, the European Hemovigilance program, and the French, Italian, and US FDA reporting programs.

The team next heard a report from Christopher Prowse who with Silvano Wendel has conducted a survey of current practices used for bacterial screening of platelet concentrates. The survey involved numerous programs and represented data on over 1 million units. Data on the different systems used for screening, rates of false positive results, and operational considerations of quarantine and date of testing were reported. The data remain PRELIMINARY and must be validated by those who submitted results. More detailed analysis will also be undertaken. A preliminary draft of the manuscript based on this survey should be available for the Seattle meeting.

The team next received a report on progress made since Edinburgh on a study of the potential adverse clinical consequences of transfusion with stored RBCs. Alan Tinmouth presented progress made on protocol development of a study to be called the ABLE study (Age of Blood Evaluation). The hypothesis is that fresh RBCs are associated with better clinical outcomes than standard RBCs. The study will compare 30 day mortality among a large number of patients randomized to receive either fresh (< 8 day) or standard age RBCs. The group was pleased with the progress made by the Ottawa group but acknowledged that this study was too large in scope to be done by the BEST group.

Sunny Dzik then reviewed two possible future topics for the Transfusion Safety Team. The first would evaluate the use of radiofrequency identification tags on blood bags and on patient wristbands as a means to reduce the risk of mis-transfusion. Dzik is beginning a pilot study in Boston using this technology in the Operating Room. Based on the results of the pilot, he may bring the topic for consideration as a BEST project. The second future topic addressed the “Decision to transfuse”. Dzik briefly reviewed the scope of the issue, identifying several possible areas for BEST study. Focusing on the methods used to assess the decision to transfuse, Dzik reviewed prior studies on blood utilization highlighting strengths within the BEST group for further study of utilization methods. He reviewed the system in use at his hospital in Boston as an example of what BEST might learn from a survey of current practice in this area. The next step would be draft a pilot survey and test this on 3 or 4 BEST members prior to consideration of a full BEST study.

Mike Murphy updated the group on the Prevention of Bedside Errors in Transfusion Medicine (PROBE-TM) trial. This is a cluster randomized controlled trial of a simple intervention (a label attached to the ports of blood bags stating ‘STOP – CHECK THE PATIENT’S WRISTBAND’). The study will involve direct observational audits of 270 transfusions in 16 BEST sites in 7 countries. The primary endpoint is the percentage of transfusions correctly checked. The labels and study packs will be mailed to participating centers soon after this meeting. It is expected that preliminary data will be presented at the Seattle meeting.

Neil Beckman updated the group on an approach his group is exploring to apply standard principles of Statistical Process Control (SPC) to the labeling of patient samples. He pointed out that the development of a practical method for monitoring the sample collection process was a natural extension of the BEST Collaborative COBS Study. This work may lead to a practical recommendation for laboratories everywhere on tracking the performance of sample collection. The next step is to seek input from experts on the best method for tracking process control of blood sample labeling, which could be provided by BEST Manufacturing members who have this expertise. Following this, the best approach would be selected, adapted for this setting and tested by BEST members.

Jane Hassan (of the host institute in Rome) spoke about hemovigilance in Italy, including the development of a centralized method for tracking rates of infectious markers in blood donors. She also provided information about the establishment of a national scheme for collecting data on adverse transfusion events in Italy. This will provide scope for improvement in transfusion practice in Italy, and for comparing data with other hemovigilance programs throughout the world.

Report: Conventional Components Team

H. Gulliksson, J. Hess

Gulliksson welcomed the team and opened the meeting of the Conventional Components Team by presenting the agenda. The team meeting started with a “red cell hour” chaired by Hess.

In a summary, Hess presented activities continuing in five areas of RBC storage:

1. Högman and Meryman presented a manuscript suggesting ways to improve RBC storage and standardization (please see summary below):
 - a. Establish a “Standard Unit” (40-45g Hb), needed to facilitate double unit collection
 - b. Design storage solutions for standard units to optimize viability and function
 - c. Manufacturers should develop cell washers to facilitate optimal storage
 - d. Adjust storage life to minimize non-functional RBC (42 days not necessary)Dzik recommended that Högman and Meryman publish the manuscript while further discussion and fact-finding takes place. Hess concluded that that the proposal is controversial. Shared opinions of members include both suggestions that the unit size is too small and too large.
2. The BEST #26 Study, “Interlaboratory comparison of RBC ATP, DPG and haemolysis measurements” – The manuscript was accepted by Vox Sanguinis in February 2005 for publication in June 2005.

3. Hess and Greenwalt have filed a patent for the 8-week conventional volume additive solution EAS-81 and similar solutions. Clinical trials of a commercially manufactured version are planned for this summer.
4. Hebert has proposed a trial called "ABLE", the Age of Blood Evaluation, to determine in a multi-country large simple prospective trial if administration of old RBC, those stored longer than 8 days, are associated with increased rates of complications and death. Because of the size of the proposed study, BEST cannot assume a significant role according to the Blood Safety Team.
5. At the request of the several of the manufacturing members, Hess will attempt to develop BEST standard protocols for 51-Cr and 99m-Tc labeling of RBC and for the measurement of RBC recovery and survival.

Suggestions for standards for RBC products.

Högman and Meryman raised the question whether it may be time to revise current standards for red blood cell units. They pointed out that great variation exists with respect to viability and function of fresh and stored RBCs as well as of the contents of RBC hemoglobin in individual units. New technology is available for improved preparation as well as storage of RBCs. They proposed the following:

The establishment of a standard unit of blood based on hemoglobin content should be a high priority goal. Manufacturers of blood collection equipment should provide suitable technology for collecting a standard unit based on donor hemoglobin concentration.

Major organizations concerned with the collection and distribution of blood components should agree on the criteria for a standard unit of red blood cells based on hemoglobin content and for the collection of double units.

Efforts should be directed at the design of storage solutions acceptable for transfusion that maximize the maintenance of both red cell viability and function during storage. The ideal storage protocol would require sterile, high pH solutions containing both glucose and electrolytes.

Manufacturers of blood processing equipment should address the challenge of economical red cell washing both to facilitate the use of optimal storage solutions and to eliminate adverse transfusion reactions resulting from plasma contamination.

Discussions regarding the merits of this approach ensued.

BEST #23 - In vitro pH Effects on In vivo Recovery and Survival of Platelets.

This study was presented by Dumont. It will be examining the hypothesis that in vivo recovery of autologous, radiolabeled platelets is not affected by in vitro $\text{pH}_{22^\circ\text{C}} > 7.4$. This is especially critical in light of recent EU Directives. We are conducting a metanalysis of available data from platelet studies conducted in the past 12 years. The initial invitations for participation were promulgated in January 2005, and we have begun to receive data at the data coordinating center (over 200 transfusion so far). We are targeting to have the database consolidated, checked, and coded by the end of May. Data analysis and distribution of results to the team should be completed by the end of June 2005. The manufacturing members of BEST have been very supportive in providing permission to use their data in this study.

BEST #24 - Interruption of Agitation of Platelet concentrates - Effects of shipping on platelet function and biochemical analyses. Multi-center in-vitro study.

This study, also presented by L Dumont, is an extension of other studies conducted by BEST members on the effects of interruption of agitation. The study extends those results by conducting true shipping studies. One objective is to determine the effect of platelet shipping on metabolic and functional parameters. $\text{pH}_{22^\circ\text{C}}$ should be maintained at greater than or equal to 6.5 at the end of the storage period. Six (6) laboratories are participating.

Each has prepared and shipped platelet products beginning day 3 or 4 of storage. Platelet products at low and high concentrations were prepared from the same pool. Concurrent, paired controls were maintained in the laboratory under standard conditions with continuous agitation. Various in vitro platelet characteristics were determined prior to and following shipment out to 7 days of storage. Detailed methods were previously published in the BEST meeting book, Feb. 2004 Bermuda. All studies are completed and data submitted. L Dumont is in the process of consolidating and checking the database. A completed analysis should be ready by the end of the summer. Preliminary results suggest platelet response to true shipping is very similar to simple static studies conducted in the laboratory: 1. Platelets held without agitation upregulate glycolysis resulting in accumulation of lactate and CO₂ and reduced pH; 2. Glycolysis rate returns to control values following re-establishment of agitation; 3. This effect is enhanced at high platelet concentrations; 4. There are no significant effects on ESC, HSR, P-selectin as a function of shipping; 5. pH of platelets are maintained for shipments over at least 24 hours.

The BEST platelet questionnaire, platelet fixation study and platelet send-around study.

Van der Meer presented the status of those projects. A platelet counting questionnaire was sent to BEST laboratories to make an inventory of current practices in platelet processing, counting and quality control. Van der Meer presented results based on data from 44 laboratories (in 9 countries), using 55 platelet analyzers in total. Most of those analyzers use impedance measurement for platelet enumeration, but differences occurred in instrument settings ('fitted' versus 'smoothed' platelet distribution curves). This may affect the results when low numbers of red cells are present. The analyzers had been validated with a great variation in requirements, and using a wide range of platelet samples. All centers performed daily quality control and most centers participated in external quality assurance schemes. While most blood centers use their analyzer for platelet counting, generally only fixed 'normal' whole blood samples were available for controls.

Blood processing is done within 8 hours (PRP method) or 24 hours (buffy coat (BC) method). For the BC methods some variation was seen in time-to-processing. Van der Meer showed data suggesting that a longer storage period was associated with higher platelet yields. A wide variation was observed for time-to-sampling: while BC-platelet concentrates are usually sampled immediately after preparation, apheresis- and PRP-platelet concentrates may be sampled any time during their storage period up to 6 days after collection. A platelet loss of 2% per day may be expected, potentially giving a 10% lower value after 5 days. About 75% of the centers used EDTA tubes for their platelet counts, the others used dry tubes, and accounting for 5-10 lower platelet counts in dry tubes.

In conclusion, many practices are used for platelet counting, timing of sampling and sample handling. Practical guidelines and requirements (based on current international guidelines) should need to be developed by the group.

Further steps are development of a stable, fixed platelet preparation, to be used for a platelet send around study, and validation of a 'gold' standard for platelet counting on the flow cytometer.

Introduction to the BEST supplement on platelet radiolabeling.

AuBuchon announced the inclusion in the Rome book of a manuscript in draft form entitled "The rationale for a standardized approach to assessment of platelet kinetics" (AuBuchon, Snyder). Draft manuscripts for the radiolabeling supplement from Murphy and Dumont are also included in the BEST book for comment. Although the manuscripts will require additional work by the authors before submission, comments on the present contents are appreciated.

Storage of Platelets at +4°C.

The introduction and background presentation by Gulliksson announced in the agenda unfortunately had to be excluded due to lack of time. It will be summarized at the next Conventional Components Team meeting together with additional studies on the storage of platelets at +4°C. The presentation is available at the BEST website member's area. Please note that "(pre)" in the figures means pre-incubation of sample at +37°C for one hour before analysis. This is an approach claimed to restore discoid shape of platelets.

Clinical biology of galactosylated platelets

The presentation by Thomas Stossel, MD, guest speaker reviewed the well-known phenomenon discovered by Murphy and Gardner of rapid clearance of chilled platelets following transfusion and the equally well appreciated consequences of this effect for platelet transfusion technology: short shelf life and deterioration in platelet structure and function. Less appreciated is the fact that recent clinical studies evaluating hemorrhage in patients receiving prophylactic platelet transfusions document 20 – 60% bleeding rates. This prevalent bleeding is rarely lethal, but represents an improvement opportunity. The presentation summarized a research program that evolved from the incorrect assumption that cold-induced platelet shape changes explain clearance to a new mechanism involving recognition of exposed β N-acetyl glucosamine (β GlcNac) residues on N-glycans of clustered Gp1ba subunits of the von-Willebrandt receptor complex by α M β 2 integrin receptors on hepatic macrophages. An externally disposed β 1,4 galactosyltransferase enzyme on the platelet catalyzes the transfer of galactose from UDP-galactose to the exposed β GlcNac residues, thereby masking them, preventing phagocytosis of chilled human platelets by macrophages *in vitro* and accommodating the prolonged circulation of chilled platelets in mice. Studies with human platelet concentrates demonstrate that galactose transfer is feasible, stable and inhibits phagocytosis of cold platelet by macrophages *in vitro* over 14 days of storage. Stossel claimed that refrigeration storage of platelets preserves *in vitro* functions markedly better than room temperature storage.

Gulliksson then adjourned the meeting after thanking the members for excellent contributions.

Report: Clinical Studies Team

L. Williamson, N Heddle

1. Survey of buffy coat (BC) platelet production amongst 14 BEST members in Europe and Canada-Scott Murphy.

As a prelude to presenting this survey, S Murphy drew the group's attention to a publication of his (Transfusion 1992; 32: 9-16), in which BC platelets had been stored for 15 days in $\frac{2}{3}$ Plasmalyte/ $\frac{1}{3}$ plasma with excellent storage characteristics.

The technological prerequisites for producing BC platelets were:-
sterile connecting device, platelet additive solution (although plasma possible), gas permeable bags, bottom and top packs and methods for separation & pooling, of which Orbisac was the latest.

The survey covered 1.4×10^6 adult doses, with total production methods of:-

Apheresis	48.6%	>60% Austria, Belgium, Germany, France
BC pools	44.1%	>80% in Denmark, Finland, Netherlands
PRP	7%	

Most pool on day 1; some on day 0 <8hrs. 10/13 use PAS II.

Canada, in which 75% of platelet production is from whole blood, will switch from PRP → BC later this year (this led to a cultural digression into ice hockey/the Canadian national anthem).

Advantages of BC platelets:-

- ↓ plasma → patient
- well suited to pathogen reduction and bacterial testing
- advanced automation
- maximal use of donor's gift

Disadvantages:-

- ↑ donor exposure relative to apheresis
- 13% loss of red cells compared to PRP

The survey revealed diversity in:

- whether pooling was done before test results available
- whether bacterial testing
- whether small pools were made for children
- whether groups A & O only were made
- 5/7 provide RhD neg pools (2 don't)
- 3/7 provide CMV neg pools

There was another brief cultural digression, this time on the subject of de Tocqueville.

2. Ralph Vassallo- Review of different methods of analysis of recovery and survival studies.

Vassallo (from S Murphy's group) presented R&S data on apheresis platelets to illustrate the differences which could arise from various methods of analysis.

- (1) Non-inferiority (Dumont) or FDA method
- (2) One-sided vs 2-sided t test (1-sided more stringent)
- (3) % required compared to fresh platelets (50 vs 66%)
- (4) Confidence intervals

These determine no. subjects, exposure to radionuclide in population, and costs, so should be set to allow a minimum number of subjects to be tested.

There was discussion around the clinical relevance of the 66% recovery/50% survival figures and whether these had been/could be calibrated against criteria immediately relevant to patients. These may determine different requirements for prophylaxis and for treating bleeding.

3. Sherrill Slichter. Recovery and survival studies of PRP platelets.

The respective relevance of recovery and survival was discussed. In studies performed some time ago (Slichter & Harker, Clin Haem 1978;7:523), it was shown that when the platelet count is $<100 \times 10^9/L$, there is shortened platelet survival. 7-8000 platelets/ $\mu l/day$ are lost to maintain endothelial integrity. In the TRAP trial, the mean days to next transfusion was $1.8 \pm 1.3d$.

Slichter first presented data on apheresis platelets (labelling in bags for Chromium, not Indium, Haemonetics + Cobe in plasma.

Pooled R = 71%) i.e. pass on Murphy's Law at 8 days
 Data S = 88%) but do not pass at 9 days.

She then presented new PRP data using new BEST method compared with fresh platelets and tube labeling.

		Recovery as a % of fresh (Mean \pm SEM)	Survival as a % of fresh (Mean \pm SEM)	Pass Murphy's law?
n so far	8	48	45	No
	7	67	50	No
	6	75	60	No

Projecting back, even 5 day plts will probably fail the 66% survival criterion!

Taking the TRAP data requirement of 1.8d to next Tx, then 8 day apheresis platelets, showing survival of 6.1 days are 3.4 x patient needs, and 6 day PRP platelets, with 7 day survival, are >2 x patient needs. Platelet recovery is also critical, especially for surgery.

Slichter reported that the FDA might need a clinical trial. There was also discussion of the impact of the difference between PRP and apheresis platelets on what would be an appropriate dose.

4. Lisa Cooke –Validation of single buffy coat platelets and recovery and survival studies.

Cooke, working in Williamson's group, had validated single BC against pools using a number of *in vitro* parameters, as a prelude to R&S studies. The method for production was provided in the BEST book. Although some quantitative differences were observed between single BC and pools, the data were adequate to allow the use of single BC platelets for R&S studies (details are posted on the website).

Data from 6 donors using single BC in plasma at day 7, using the BEST method, did not pass the 66/50 criteria. However, data for fresh platelets were approximately 10% below expected for both recovery and survival, suggesting that the technique needed to be refined. AuBuchon kindly offered to host a training visit. Other variables included COST vs local programme, and the use of CLX packs.

5. Nancy Heddle - StoP study.

24 patients had so far been recruited. The study needs 550-600 altogether. Ottawa have gained supercentre status with 14 patients recruited so far. Some minor protocol changes have now been agreed. Heddle will develop an adjudication committee from within BEST members.

Slichter reported that the US Transfusion Clinical Trials network is doing a similar study, and have recruited ~150 patients so far, the intention being to have some outcome in 2 years.

Dumont asked whether there would be stratification as a 2^o endpoint by product type, but this is not planned for the US trial. The StoP study had not included any BC platelets so far.

6. Nancy Heddle - Tool to measure bleeding severity.

This study is now funded by CBS, and Heddle expressed her thanks to the BEST members who helped with suggestions. Kathryn Webert is returning from maternity leave in April and will continue the study. The critical features are the questions, so the next step is item generation, which will be done via BEST, local clinicians, and the literature. There will then be a process of item reduction with ?grouping/omission – BEST members can help with this. There will also be development of measurement criteria eg dichotomous versus scale/severity, and examination of the psychometric properties of the instrument. The whole process may take up to 2 years to validate.

KW will be in touch with BEST members for help.

There was considerable interest in this study and many questions.

Corash asked how would be applied, and could it be applied retrospectively?

Heddle confirmed that this could be applied to raw data already collected.

Start/stop of bleeds/change of grade is the most difficult to assess.

Slichter reported that because such data cannot be blinded, data on the US trial will be sent to the Coordinating Centre.

7. Simon Stanworth, Oxford, UK. Patient self-assessment of bleeding

Stanworth, working in M Murphy's group, described a patient self-reporting tool for assessing bleeding, which gets round data collection problems which depend on nurses. The aim of the study was to validate the method by comparing self-assessment with nursing assessment. Patients were trained before the plt count fell to <50. Two medical assessors plus the patient assessed bleeding.

13 pts have been studied so far (+2 exclusions), giving 250 observer days to report, with 120 'self and medical' days. There were 105 days with self assessment only (at home or weekends), and 30 days with medical assessment only (due to patient illness). There were some discrepancies at the 'mild' end of the bleeding scale but it was as yet uncertain whether these would have been enough to change the WHO bleeding grade. In conclusion, this is feasible and useful, and is particularly good for weekends/time at home, and in defining days

of no bleeding. This has also highlighted the limitations of the WHO bleeding scale. It is very cost-effective as it needs medical/nursing input <20% of the time.

There was strong encouragement from the group to continue. Comments during discussion included a reminder that a similar approach had been taken in haemophilia studies (although data quality was uncertain), concern whether this might reduce the threshold for requesting a platelet transfusion (no evidence of this so far), and whether having some responsibility for self-recording might improve patient's perception of their treatment experience. Tight definitions during training would also be important.

7. Nancy Heddle/Kathryn Webert. Can patients do their own bleeding assessment?

Heddle reported a similar study of 34 patients, with 513 matched assessment forms of which 428 were complete (4-28 forms/patient). Many types of bleeding showed 100% agreement, with <90% agreement in:

- petechiae	med>pt
-small bruises	pt>med
-large bruises	med>pt

There was a problem with cerebral bleeding, where there was 98% agreement. In some cases, the patient had been told they 'may have had a stroke', but the health care professional didn't record it. This is such an important end point that the aim should be 100% concordance.

Again this generated much useful discussion:-

- Where there is disagreement how to ascertain which is right?
- Might the use of this method bias platelet trials towards study of less sick patients, and could you then generalise results to very sick patients?
- The problem of immigrant populations with language difficulties also need to be tackled.

It was agreed that there was scope for the Oxford and McMaster groups eventually collaborating to produce a joint questionnaire.

8. Nancy Heddle. Can we improve clinical research reporting?

While undertaking a review of studies on platelet dose, Heddle had observed considerable variability of reporting, as illustrated by 4 studies published in Blood or Transfusion. The main variables observed were:

- study design
- failure to state precise purpose of study
- reports of inclusions/exclusion
- data on the platelet products used
- clear statement of summary statistics.

SO – Is there interest in producing a guideline for reporting patient clinical studies?

There was lively discussion, with a number of helpful comments, including:-

- There are already CONSORT guidelines for reporting trials- it would be helpful to use them as a basis to develop a guideline for all transfusion studies.
- This concept would be worth discussing with editors of the key transfusion journals. Papers which need statistical review may not always get it.
- There were limits on what data you can collect from patients who have not given consent.
- It might be worth making funding bodies aware of such guidelines.

It was agreed to form a working group to take this forward. Dzik, AuBuchon, M Murphy, Williamson, Blajchman, J Miripol offered to help Heddle take this forward.

9. Lorna Williamson. BEST #33. Tracking of transfused platelets in patients using HLA discrepancy and flow cytometry

Due to lack of time, this was not discussed in detail. A draft protocol has been posted on the website and members should provide comments to Williamson within a month.

Williamson thanked all the speakers and attendees for a lively and productive meeting.

Report: Cellular Therapy Team

G. Moroff, H. Eichler

1. Administrative Issues and Update on Cellular Therapy Team Projects and Manuscripts (Hermann Eichler)

Hermann Eichler welcomed all participants of the team meeting and announced that Gary Moroff would unfortunately not be able to join the meeting in Rome. Prior to the Rome meeting, Gary Moroff had already informed the team members that he would step down as co-team leader immediately after the Rome meeting. Ziggy Szczepiorkowski will follow him as co-team leader. All participants signed a special diploma making Gary an Honorary Co-Leader.

Hermann Eichler gave an overview of the agenda of the current cellular team projects. The revision of the BEST Study #15 manuscript by Tsuneo Takahashi has now been completed and will be forwarded to Gary Moroff until the end of March for submission to *Cytotherapy*. The results of BEST #19 have now been summarized in the manuscript "Multiple laboratory comparison of in-vitro assay utilized to characterize hematopoietic cells in cord blood". Gary Moroff submitted this draft manuscript to all team members prior to the meeting. Comments and suggestions should reach Gary Moroff by the end of March so that the manuscript can be finalized and submitted to *Transfusion*.

2. Guest Presentation: Challenges of Post-characterization of Cord Blood Products (Michael Creer)

Dr. Michael Creer, an Assistant Prof of Pathology and Pediatrics, Wash University and director of St. Louis Cord Blood Bank located in St. Louis, Missouri, presented a very eloquent and informative talk entitled "Challenges of Post-Thaw Characterization of Cord Blood Products". The presentation in its entirety is available on the BEST website. A few highlights of general interest are provided in this summary. Dr. Creer discussed challenges associated with cryopreservation and thawing of cord blood units. He noted that there are no guidelines or standards for interpretation of cooling curves. What is even more concerning transplantation centers (at least in 50% of cases) do not evaluate CD34 recovery post thawing. Thus, it becomes critical that the Cord Blood Bank has appropriate quality measures to assure that the cell content is high enough to minimize a risk of non-engraftment. In his laboratory Dr. Creer investigated an effect of different variables such as cooling rate, propanediol vs. DMSO, on TNC recovery, CD34 recovery, CFUs and viability. For example, cooling rates in excess of -5C/min profoundly affect cell recovery and CFUs. Interestingly, viability measured using trypan blue was only marginally affected. Dr. Creer advocated the use of CFUs as well as CD34/7-AAD for assessment of cord blood units. However, even CD34/7-AAD viability did not correlate well with loss of CFUs. All of which are important measures of potency of cord blood units. Importantly, thawed cells need additional time to regain their ability to pump out a dye (7-AAD), thus timing of the assay may be critical. It is also noteworthy that the viability measured using trypan blue staining tends to be higher than one measured using 7AAD. In addition, Dr. Creer discussed transient warming event (TWE) which is related to water condensation. Based on preliminary data he noted that exposure of frozen units to TWE for less than one minute does not affect their potency as measured by CFU-C. However, it is advisable to minimize the risk of TWE of any longer duration. And finally, an interesting send-out experiment, akin to many BEST projects, was presented. Several cord blood samples were sent to four different Cord Blood Bank in the US to compare cell viability in the segment and bags. The products were either frozen using Thermogenesis System or CBS Cryo System. Similar results were obtained on segments and products, it was noted that there is a significant and highly variable loss of viability of CD34+ cells (10-70%) after thaw. Results obtained following controlled-rate freezing at fixed volume (Thermogenesis) do not differ from those obtained with variable-volume controlled-rate freezing (CBS Cryo. System). After Dr. Creer's presentation a lively discussion ensued. One of the conclusions of the discussion was that our group might be interested in inviting an expert in cryobiology to its meeting in Seattle to discuss basic as well as more complex aspects of cryopreservation. This may lead to additional BEST studies.

3. Cord Blood Assay Study – Results with Frozen/Thawed Samples (BEST 30) (Anneke Brand)

Dr. Anneke Brand presented preliminary results of the most recent send around. In this study three cord blood specimens were sent-out to 9 centers. There was a fairly good correlation between centers in measuring nuclear, however, there were significant differences in reported differentials as well as CD34 content (see full presentation on the web site). Unfortunately, none of the centers could detect any CFUs. Anneke discussed different potential phenomena explaining these results. Upon thorough investigation, it is possible that the cells were warmed up significantly on their way from Leiden to Mannheim that despite reasonable viability reported by some centers no CFUs were observed. Interestingly, reported viability correlated with different techniques used with centers using trypan blue reporting the highest viability followed by AO/EB and 7 AAD (the lowest). After extensive discussion, it was proposed to repeat this run with the same participants and potentially, if sufficient specimen number of cells available, add new participants (Sue Armitage, Francine Decary and Michael Creer). The protocol will be also amended to add specific cell populations to study and clarify potential mathematical challenges encountered by some of the centers.

4. Best #28: Inter-laboratory Pilot Study on Quality Characteristics of Dendritic Cells (Hermann Eichler)

Hermann Eichler presented the preliminary analyses of BEST #28 performed since the last meeting in Edinburgh. The presentation, including descriptive analyses results, can be reviewed in detail on the BEST web-site. Primary objective of this pilot study was to compare non-functional DC quality control assay results obtained from multiple laboratories testing identical DC samples. Overall, three identical DC samples were tested in parallel by eight participating laboratories in Europe, US and Japan. All DC samples were prepared and shipped from the study center in Mannheim/Germany between October and December 2004. Data analyses showed a possible difference in nucleated cell counting of thawed DC samples tested by various hematology analyzers. Viability testing determining 7-AAD negative cells by flow cytometry showed significantly different results between sites. Furthermore, the testing of antigen expression on mature dendritic cells also showed alterations between the sites. Storage stability of the final mature DC product was also included into the study protocol. It showed that each laboratory used different final storage containers. A high variability could be observed between sites testing DC viability over time. The follow-up plan is to obtain completed information from each site and to re-analyze FACS data files of antigen expression measurements. Hermann Eichler will then summarize the information by preparing a draft manuscript for publication. The draft is to be presented for review at the next BEST meeting in Seattle.

5. Mesenchymal Stem Cells – Consideration of Multi-center Study based on Pilot Study (Jo-Anna Reems)

Jo-Anna Reems presented the results of a pilot study on the expansion of mesenchymal stem cells isolated from frozen/thawed bone marrow performed in the laboratories in Seattle and Mannheim. This study was performed after the meeting in Edinburgh. Objective was to compare the expansion of obtained MSC from both laboratories since both were using the same source of material. In this pilot study, bone marrow was harvested from a cadaverous organ donor. It was then frozen without a further processing step. Vials of frozen bone marrow were then shipped on dry ice to the center in Mannheim. The expansion was performed in Seattle and Mannheim using the pertinent methods. Differences in expansion capacity and CFU-F content could be observed as well as differences using different expansion media. Nevertheless, this pilot study provided relevant information for planning BEST study #32 with the objective to determine the optimal conditions to achieve maximum MSC expansion. Until the next meeting in Seattle, Jo-Anna Reems and the study group will finalize the draft study protocol for BEST study #32 that was included in the BEST book for Rome. This finalized protocol should then be discussed at the next meeting. BEST members interested in participating in such a MSC study are Jo-Anna Reems, Anneke Brand, Ron Sacher, Paolo Rebulli, Sam Coker, Tsuneo Takahashi, David McKenna, Derwood Pamphilon and Hermann Eichler.

6. Release Criteria Utilized with Cellular Therapy Products: Review of commentary on first questionnaire and review of second questionnaire (BEST 29) (Ronald Sacher/Ziggy Szczepiorkowski)

Ziggy Szczepiorkowski presented his and Ron's thoughts on the next steps related to this project (see presentation for more details). The results of questionnaire #1 were presented by Ron in Edinburgh. Three major concerns were identified: small pool of participants, lack of information on participants (i.e. size, IND, etc), and

the narrative nature of questions made interpretation difficult. Three potential options for the use of the obtained data were suggested [*a letter to the editor outlining the results or a manuscript summarizing the data (suggested in Edinburgh) or a review article on release criteria explaining the lack of standardization supported by the data from the first questionnaire*]. After a short discussion it has been decided to prepare a review on release criteria (Draft by Ziggy/Ron by Seattle Meeting) as well as prepare more specific second questionnaire improved by using Web-based collection system, minimize narratives, offer incentives for completion of the survey (involve AABB, ISCT, FACT, JACIE, international sites), collect information on responder's facility size, scope of activities, regulatory framework (i.e. IND or equivalents; accreditation status and include product specific questions (Draft to be presented in Seattle).

The group members were also interested in a survey to poll their interests and additional study ideas. It will be designed and distributed by Hermann and Ziggy before Seattle Meeting. It was noted that the meeting was very well attended with lively discussion and at least 21 attendees.

Conclusion

At the conclusion of the meeting, AuBuchon reviewed the meeting format, and the consensus was that it was an improvement. He thanked all for their contributions and participation and adjourned the meeting.

Minutes of the Meeting of the Executive Committee

March 12, 2005

Sala Concilio, Palace Hotel, Rome, Italy

The Executive Committee convened immediately after the BEST XXIX meeting. Present were J. AuBuchon (chair), S. Murphy (treasurer), W. Dzik, H. Eichler, H. Gulliksson, N. Heddle, J. Hess, M. Murphy, Z. Szczepiorkowski, L. Williamson and S. AuBuchon (staff).

J. AuBuchon opened the meeting by noting the considerable and appreciated contributions of G. Moroff on his "retirement" from team leadership, and he welcomed Z. Szczepiorkowski to leadership of the Cellular Therapy team.

AuBuchon and S. Murphy reviewed the financial situation of BEST, and there was consensus that it was sound. No long-term outstanding dues matters remain. No financial difficulties are anticipated for BEST in the next year.

AuBuchon reviewed the not-for-profit status of BEST, its required five-year report to the State of New Hampshire and submission of a 501(c)(3) application to the US Internal Revenue Service to gain national not-for-profit standing. No difficulties are anticipated in this. Upon completion of these efforts, legal expenses are expected to decline sharply.

The team leaders reviewed the status of the membership slots and participation of their teams. The ability of team leadership to alter the associate scientific membership composition of the team to address particular efforts was discussed. Plans for beginning the elections for next round of leadership were discussed.

Various options for travel reimbursements were discussed. The high cost of meeting in some venues (such as Rome) was noted, and the Executive Committee agreed unanimously to increase the sessional indemnity for BEST XXIX for Associate Scientific Members to \$1500.00. (This will not strain BEST finances.) The Committee then considered how to address the issue in the future with rising costs of travel and a geographically dispersed membership and agreed unanimously to return to the arrangement that had been used previously, specifically: (1) Scientific and Associate Scientific Members will receive the same sessional indemnity per meeting; (2) If the meeting is held on the continent of residence of the individual, reimbursement will be \$1,500.00 per meeting; (3) If the meeting is held on a continent other than the one of residence of the individual, reimbursement will be \$3000.00 per meeting.

The Executive Committee reaffirmed policies regarding attendance of associate/members at meetings and guests. Although each member is allowed to request the attendance of a guest, limitation of request to adhere with this policy has at times been difficult. If transgression of this policy becomes habitual, BEST would expect the member to pay for additional costs associated with the extra attendance.

The Committee reviewed the conflict of interest policies. There was unanimous agreement to add an additional clause to the statement submitted by Scientific Members pertaining to corporate support of their research (in any amount).

The Committee expressed satisfaction with the newly unveiled BEST website. A suggestion was made to include the BEST membership in the public pages. At this time, the status of manufacturer members (primary and Associate Manufacturer Members) will be clarified. In addition, the list of BEST meetings and minutes will be placed on the public website in order to provide access to the discussions widely among those who might be able to utilize the information. The presentations (with appropriate admonitions about the impropriety of further distribution) will be placed in the Members Only section.

J. Hess provided information on the Global Collaborative for Blood Safety into which BEST has just been elected last November. The question that has been considered by BEST previously is how to best harness the group's interest and expertise in transfusion science for developing countries and their limitations. Hess suggested that one way that BEST could start is to simply take into consideration conditions in developing

countries when considering conditions to be included in a trial – such as one looking at the effect of discontinuing platelet agitation or storing at different temperatures.

There was agreement that the format of a shortened BEST meeting with team summaries being presented was an improvement. This will be utilized again at the next meeting.

AuBuchon thanked team leaders for assembling the presentations made in their teams' meetings and passing them to him. He requested that team leaders provide their minutes to him by April 18.

AuBuchon detailed the problems that had developed unexpectedly in dealing with the Hyatt Hotel in Seattle for BEST XXX and the efforts that were being made to find another appropriate venue close to the convention center. This is expected to be resolved shortly, and notice will be placed on the website.

The plans for future meetings were discussed. For BEST XXXI, the consensus was to utilize the offer made by M. Lozano to arrange meeting space in Barcelona for March 17-18 or 24-25, 2006. If this cannot be secured under reasonable terms within a defined period (of a few weeks), the next choice of a meeting site was Dublin. The meeting site for BEST XXXII is expected to be September 1-2, 2006, in Cape Town, South Africa, immediately before the next ISBT Congress. The new BEST leadership will plan the BEST XXXIII meeting for early 2007.

There being no further business to conduct, the meeting was adjourned at 5:30 pm.