# SUMMARY OF SAFETY AND EFFECTIVENESS
# CELLPRO CEPRATE® SC STEM CELL CONCENTRATION SYSTEM

**PMA Number:** BP940001

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## INDICATION AND USAGE

The CEPRATE® SC System is indicated for the processing of autologous bone marrow to obtain a CD34+ cell enriched population which is intended for hematopoietic support after myeloablative chemotherapy. Infusion of the CD34+ enriched population results in a lower incidence of DMSO infusion-associated complications compared with infusion of unselected bone marrow cells. It is recommended that sufficient bone marrow be harvested to provide at least 1.2 x $10^6$ CD34+ cells per kg of patient body weight after CD34+ cell selection. Infusion of less than 1.2 x $10^6$ CD34+ cells/kg of recipient body weight has been associated with delayed platelet engraftment. (See Clinical Experience).

## DESCRIPTION

The CEPRATE® SC Stem Cell Concentration System consists of an instrument and a single-use, sterile, prepackaged kit containing disposable components. The CEPRATE® SC Disposables Kit consists of the following components:

(1) Avidin Column
(1) Precolumn
(1) Tubing Set
(3) Phosphate Buffered Saline (PBS), 1000 mL
(1) RPMI 1640 Cell Culture Medium, 1000 mL
(1) 40 μm Pall SQ40S Blood Filter
(1) Anti-CD34 Biotinylated Monoclonal Antibody (murine 12.8 antibody), 3.0 mL/vial

## PRINCIPLES OF OPERATION

The CEPRATE® SC System concentrates CD34+ cells using a proprietary, continuous-flow immunoadsorption technique. After marrow cell harvest and buffy-coat preparation, the cells are incubated with biotinylated murine anti-CD34 monoclonal antibody (MAb) which binds selectively to CD34+ cells. After a wash step to remove excess, unbound antibody, marrow cells are processed through the CEPRATE® SC System. The cells flow through a column containing beads coated with avidin. The biotinylated antibody-labeled CD34+ cells bind to the avidin-coated beads, and unlabeled cells are washed through the column. The CD34+ cells are then eluted by gentle mechanical agitation of the beads.

## BACKGROUND

Autologous bone marrow transplantation (ABMT) has been used in the treatment of patients with a wide variety of hematological malignancies and solid tumors; however clinical investigators have recognized serious and occasionally life-threatening toxicity that can be associated with the infusion of autologous marrow. Clinical studies have been performed that documented the specific toxicities that occur with marrow infusion (1-7) including several reports of deaths related to the infusion of autologous marrow (1, 3, 7).

Autologous marrow is typically cryopreserved so that the patient can recover from the marrow harvest and undergo high-dose chemotherapy prior to the actual marrow infusion. Most complications of marrow infusion are an indirect result of the cryopreservation and thawing procedures. These complications occur primarily for two reasons. First, the cryoprotectant, which is added to the cell suspension to inhibit crystallization of water during freezing, is associated with infusional toxicity. The agent used for this purpose is dimethyl sulfoxide (DMSO). The amount of DMSO infused is proportional to the volume of marrow. DMSO can cause a variety of mild to moderate side effects including nausea, vomiting, and diarrhea (4, 8, 9). DMSO also can induce histamine release and cause anaphylactoid reactions that range in severity from rashes and flushing to hypotension, bronchospasm, pulmonary edema and respiratory compromise (9-12). Hypertension may result from the effect on smooth muscle of DMSO (4, 10). DMSO is also known to have a negative chronotropic effect on the heart that can result in bradycardia, heart block and in severe cases, cardiac arrest (10, 11). DMSO is a diuretic and has been reported to contribute to decreased renal function and the onset of acute tubular necrosis when administered intravenously (13).

The second reason for complications resulting from cryopreservation is related to damage of marrow cells during the cryopreservation and thawing process. The cryoprotectant solution used to protect the marrow progenitor cells is hyperosmolar (approximately 2,000 mOsm). Although marrow progenitors are relatively resistant to this extreme osmolarity, other cells in marrow are not as resistant. Granulocytes, platelets and red blood cells (RBC) are particularly sensitive and readily lyse under these conditions. After standard buffy coat preparation, the marrow contains approximately ten billion granulocytes, one hundred billion platelets, and one trillion RBC (14, 15). Hemolysis results in the release of free hemoglobin which can cause renal damage, and in some cases, acute renal failure (1). Lysis of platelets, granulocytes and other nucleated marrow cells results in cell debris and cell aggregates which can lead to pulmonary emboli (5). Cell lysis products such as potassium, calcium or adenosine may also play a role in the development of bradyarrhythmias (16). Peritransplant cardiac and respiratory problems may also be exacerbated by the marrow infusion, which could have an adverse effect on the patient's transplant course. Children undergoing chemotherapy are considered to have lesser circulatory and renal reserves and may be at increased risk for toxicities due to marrow infusion (17).

Attempts to remove DMSO or unwanted cells after thawing marrow have been problematic due to losses of progenitor cells and other technical difficulties (8). In addition, such methods have been clinically ineffective in reducing side effects of marrow infusion (5).

There are several methods of decreasing the volume of bone marrow grafts, of which only two (buffy coat preparation and mononuclear cell (MNC) preparation) are commonly practiced in marrow processing laboratories. Less common are counterflow centrifugal elutriation and starch sedimentation. A typical bone marrow harvest consists of 1-3 liters of bone marrow. Following centrifugation to yield a buffy coat or total nucleated cell (TNC) suspension, the volume of the

marrow harvest is reduced to between 100 and 250 mL. In most marrow processing laboratories, buffy coats are cryopreserved in 10% (v/v) DMSO. This is accomplished by adding an equal volume of a cryopreservation medium, containing 20% DMSO, to the buffy coat. Thus, at the time of reinfusion, a typical marrow graft will have a volume of between 200 and 500 mL and contain between 20 and 50 mL of DMSO.

Some marrow processing laboratories apply the buffy coat to a density gradient, such as Ficoll® or Percoll®, centrifuge, and harvest the MNC layer at the interface between the density gradient medium and the supernatant. This procedure results in anywhere from a 0-50% reduction in volume, relative to the buffy coat. MNC suspensions are typically cryopreserved in 10% DMSO again by adding an equal volume of cryopreservation medium to the MNC suspension. Hence, at the time of reinfusion, a total of 10 to 50 mL of DMSO would be present in the graft.

Marrow can also be processed by starch sedimentation to yield a preparation similar in composition to a buffy coat. Since some starch remains in the "buffy coat," and starch is cryoprotective, less DMSO (5% v/v) may be used in the freezing medium. However, the users typically add up to 1/3 volume of 6% hetastarch to the product. This results in a large volume for infusion unless there is further concentration. Accordingly, a starch "buffy coat" from a 1-3 L marrow harvest consists of about 100-250 mL which, after cryopreservation would contain between 10 and 25 mL of DMSO.

In all of the methods described above, further volume reduction by centrifugation is limited by the concentration at which cells can be cryopreserved without clumping or aggregation that might compromise recovery.

When the CEPRATE® SC Stem Cell Concentration System is used to select CD34+ cells from a marrow buffy coat, the resultant cells are contained in a volume of approximately 5 mL. An equal volume of cryopreservation medium, containing 15% DMSO, is added to the CD34+ selected product to yield a final concentration of 7.5% (v/v) DMSO. Thus, at the time of reinfusion, the graft contains about 0.6 mL of DMSO.

Table 1 compares the post-processing volumes typically reinfused into the patient with each of the marrow processing methods discussed above. As stated above, DMSO is frequently present at 10% v/v. Marrow processing using CEPRATE® SC selection results in exposure of the patient to substantially less DMSO than other methods.

| Table 1<br>COMPARISON OF MARROW PROCESSING METHODS | | | | | |
|---|---|---|---|---|---|
| Processing Method | Starting Blood Product | Starting Volume | Post Processing Product | Post Processing Volume | DMSO Volume Infused |
| Centrifugation | Bone marrow | 1 - 3 L | Buffy Coat (TNC) | 100 - 250 mL | 20 - 50 mL |
| Density Gradient | Buffy Coat | 100 - 250 mL | Mononuclear cells | 50 - 250 mL | 10 - 50 mL |
| Elutriation | Bone marrow | 1 - 3 L | Large cell fraction | 400 - 450 mL | 80 - 90 mL |
| Starch Sedimentation | Bone marrow | 1 - 3 L | Buffy Coat (TNC) | 100 - 250 mL | 10 - 25 mL |
| CEPRATE® SC | Buffy coat | 1 L | CD34+ cells | 5 mL | 0.6 mL |

The utility of the CEPRATE® SC Stem Cell Concentration System is its ability to enrich for CD34+ cells for engraftment while reducing the amount of DMSO, contaminating cells, cellular debris, and volume of infusate. The potential therapeutic benefit explored in the clinical studies was reduction of marrow infusion toxicities as well as the need for monitoring and treating infusional toxicities.

## PRECLINICAL STUDIES

Various attempts to concentrate hematopoietic progenitors by exploiting differences in size and density between progenitor cells and other cellular elements in marrow have had minimal success (14, 15). An immunoadsorption technique relying on the high affinity interaction between the protein avidin and the vitamin biotin has been shown to be an effective method for purifying cells on a clinical scale. This method enables positive selection and concentration of hematopoietic progenitor cells while significantly reducing contamination of red blood cells, platelets, granulocytes and cellular debris. Initial preclinical studies focused on using avidin-biotin immunoadsorption to isolate hematopoietic progenitor cells for transplantation in dog and non-human primate models.

The anti-MHC class II monoclonal antibody (MAb) 7.2 was first used to isolate marrow cells, expressing class II antigens, for autologous transplantation in lethally irradiated dogs. Marrow cells from seven dogs were incubated with MAb 7.2, followed by biotinylated goat anti-mouse immunoglobulin (Ig). The cells were passed over columns of avidin-Biogel (polyacrylamide) beads, and adsorbed cells were recovered and infused into seven lethally irradiated canine autologous recipients. Sustained marrow engraftment and complete hematological recovery occurred in six dogs. The seventh dog died of infection, but marrow examination at necropsy showed trilineage engraftment. These studies demonstrated that avidin-biotin immunoadsorption was a feasible method for isolating a selected cell population capable of autologous engraftment. However, while it was known that numerous other, non-engrafting cells present in blood and marrow did express class II antigens, it was uncertain whether these antigens were expressed by human stem cells.

The MAb 12.8 appeared to be a good choice for clinical application as it recognizes the 115,000 dalton glycoprotein antigen CD34 present on 1-3% of human marrow cells including immature blast cells. This monoclonal antibody is a murine IgM (k) and has been designated as an anti-CD 34 monoclonal antibody by the Leukocyte Typing Workshop IV. The CD34 antigen is not detectable on most tumor cells or mature blood cells, such as lymphocytes, granulocytes, red blood cells, and platelets. Since MAb 12.8 reacts with a similar marrow population in baboons, studies were performed to determine whether CD34+ cells isolated by avidin-biotin immunoadsorption could reconstitute lethally irradiated baboons. For five animals, baboon marrow cells were treated successively with MAb 12.8 and biotinylated goat anti-mouse Ig, then passed over columns of avidin-Biogel. In three animals, 65-81% of the selected cells were CD34+. In two animals, the selected cells were further enriched by flow cytometry, yielding a cell population that was 85% to 91% CD34+. The animals were given 9.2 Gy total body irradiation, a myeloablative dose which has historically resulted in animal death from marrow aplasia without marrow transplant rescue. Following infusion of the autologous CD34+ selected cells, all five animals achieved granulocyte counts greater than or equal to $1000/mm^3$ and platelet counts greater than or equal to $20,000/mm^3$ by 13 to 24 days post transplant. The rate of engraftment was similar to that observed in two control animals infused with unfractionated marrow. Furthermore, two of the animals given CD34+ selected cells were followed for more than two years post transplant, remaining clinically well and with normal hematologic parameters. These data suggested that avidin-biotin immunoadsorption with MAb 12.8 could be used to isolate sufficient numbers of hematopoietic progenitors for rapid, sustained hematological recovery after myeloablative therapy.

Pre-clinical studies demonstrated that positive selection of CD34+ cells by avidin-biotin immunoadsorption results in enrichment of colony-forming cells from the marrow of patients with advanced cancer. When marrows from patients with stage IV breast cancer (n=22) or neuroblastoma (n=4), were fractionated using MAb 12.8 by the immunoadsorption procedure described above, the enriched cell populations contained approximately 65% CD34+ cells. Colony-forming cells are a useful measure of the concentration of early hematopoietic progenitor cells. The CD34+ selected fraction was enriched for colony-forming unit-granulocyte-macrophage (CFU-GM) by a mean of 80-fold compared to the starting population. Furthermore, the CD34-depleted (unselected) cells that passed through the column grew few, if any colonies, suggesting that the majority of hematopoietic progenitors detectable in culture were captured on the column.

The Master and Working Cell Banks employed in the manufacture of MAb 12.8 have been demonstrated to be free of adventitious agents. While these banks elaborate endogenous murine retroviruses, purification of MAb 12.8 has demonstrated sufficient clearance of murine retrovirus as well as challenge viruses. Purified MAb 12.8 has been shown to be pure, pyrogen free, and able to bind CD34+ cells. The purified MAb 12.8 is subsequently chemically coupled with biotin; the final purified biotin-12.8 conjugate has been demonstrated to be pure, sterile, pyrogen free and able to concentrate CD34+ cells from bone marrow.

As per the Tripartite Biocompatibility Guidance for Medical Devices, the total column and the pigtail components, extracted with sodium chloride USP, were tested *in vivo* and *in vitro* for cytotoxicity; blood compatibility (hemolysis); acute systemic toxicity; intracutaneous toxicity; muscle implantation reaction; antigenicity; mutagenicity; and sensitization reaction. No adverse effects were noted.

Leaching of the plasticizer di(2-ethylhexyl) phthalate (DEHP) occurs following storage of the tubing for at least three months. Analyses showed DEHP levels of less than 0.1 mg/mL in the bags containing the CD34+ enriched and CD34+ depleted populations and levels of 0.36-0.51 mg/mL in the waste bag. Based on an average body weight of 70 kg, the total approximate amount of DEHP infused is 0.013 mg/kg. This level is below that reported to be associated with any toxic effects.

## CLINICAL EXPERIENCE

**Description of Clinical Studies:**

Based on the results of a Phase 1/2 single-center, open-label clinical trial in 29 women with breast cancer and four patients (3 female and 1 male) with non-Hodgkin's lymphoma undergoing ABMT, the safety and effectiveness of the CEPRATE® SC System for hematopoietic reconstitution were evaluated in a Phase 3 trial in women with stage II - IV breast cancer undergoing ABMT. Ninety-five patients were enrolled. Ninety-two were transplanted, 45 received selected cells and 47 received unselected cells. Three transplanted patients were determined to be ineligible; one had a 5 mm metastatic brain lesion, the other two had received greater than two regimens of chemotherapy, contrary to the inclusion criteria. Eighty-nine patients were evaluable; 42 receiving selected cells and 47 receiving unselected cells.

The Phase 3 study had two primary objectives: to demonstrate equivalent neutrophil engraftment and to demonstrate a lower incidence of hemodynamic adverse events (that is, hypertension, bradycardia) occurring within 24 hours of infusion.

The randomized, controlled pivotal trial was conducted at five sites. The strong odor of DMSO in the control preparations and the small volume of the selected cells made blinding unfeasible. The formal study was designed to collect safety and efficacy data for each patient through day 100 after the last patient was transplanted. Further data on graft stability, immunologic reconstitution, disease free-survival, and overall survival were collected during the post-100-day period. The dose and schedule of chemotherapy as well as the standards of care related to antibiotics, anti-fungals, transfusion of platelets, transfusion of RBC, and criteria for hospital discharge varied between sites.

**Results of Clinical Studies:**

The mean values for the magnitude of change from baseline were significantly less in the selected arm than in the unselected arm for the following hemodynamic parameters: maximum increase in systolic blood pressure, maximum increase in diastolic blood pressure, and maximum decrease in heart rate (Table 2).

Table 2
Mean Changes in Hemodynamic Parameters within 24 hours Following Marrow Infusion (All Infused Patients)

| Hemodynamic Endpoint | Treatment Arm | | p-value* |
|---|---|---|---|
| | Selected | Unselected | |
| Maximum increase in systolic blood pressure (mm Hg) | 9 ± 11** | 22 ± 14** | <0.001 |
| Maximum increase in diastolic blood pressure (mm Hg) | 7 ± 7 | 16 ± 10 | <0.001 |
| Maximum decrease in heart rate (beats/min) | 9 ± 5 | 22 ± 11 | <0.001 |

* one-sided t-test
** mean ± std. dev.

The percentage of patients achieving neutrophil engraftment by day 20 was 89% in selected arm and 88% in the unselected arm. The engraftment rate on the selected arm minus that on the unselected arm was 1% with a 95% confidence interval of -11% to 15%.

Patients in the study experienced a variety of adverse events commonly associated with marrow infusion. Patients receiving marrow processed with the CEPRATE® SC System had a lower incidence of infusion-related toxicity compared to those receiving unselected marrow. All adverse events (grades 1-4) and severe/life threatening adverse events (grades 3 and 4) during the first 24 hours after infusion are listed in Table 3 for the 92 infused patients.

| Table 3<br>Percent of Patients with Infusion-Related Adverse Events* | | | | | |
|---|---|---|---|---|---|
| Organ system | | Selected | | Unselected | |
| | | All events | Severe/life threatening events | All events | Severe/life threatening events |
| Cardiovascular | Hypertension (systolic) | 9% | 2% | 47% | 0% |
| | Hypertension (diastolic) | 4% | 0% | 6% | 0% |
| | Cardiac rate/ rhythm | 33% | 0% | 87% | 0% |
| GI | Cramping | 9% | 2% | 21% | 6% |
| | Nausea | 51% | 22% | 70% | 30% |
| | Diarrhea | 36% | 2% | 38% | 4% |
| Other | Headache | 11% | 2% | 9% | 6% |
| | Hemoglobinuria | 29% | 0% | 89% | 9% |

* Grades of toxicity are modified SWOG toxicity criteria

The results of engraftment endpoints (up to day 100) are summarized in Table 4. No differences were noted between the two study arms, except in platelet recovery. The median number of days from infusion to date of platelet engraftment ($\geq$20,000/mm$^3$, without transfusion) was 8 days longer for the selected arm than the control arm (p=0.04); 28 days in the selected arm (95% C.I. = 23-32) and 20 days in the unselected arm (95% C.I. = 18-23), a difference of 8 days (95% C.I.: 1-11). The incidence of clinical sequelae that might be associated with delayed platelet engraftment, including number of units of transfused RBC and platelets, number of patients experiencing bleeding episodes, and number of Grade 3 or 4 bleeding episodes, was not significantly different between arms.

| Table 4<br>Engraftment Characteristics and Immediate Post-Transplant Course<br>(All Infused Patients) | | | |
|---|---|---|---|
| Endpoints | Treatment Arm | | |
| | Selected<br>median (range) | Unselected<br>median (range) | p-value |
| Percentage of patients with neutrophil engraftment at day 20 | 89% | 88% | 0.73 |
| Days to ANC $\geq$20,000/mm$^3$ | 13 (9-33) | 11 (8-48) | 0.16 |
| Days to platelet $\geq$20,000/mm$^3$ | 28 (11-68) | 20 (8-61) | 0.04 |
| RBC transfusions (units/patient) | 6 (2-30) | 8 (2-32) | 0.11 |
| Platelet transfusions (units/patient) | 64 (12-264) | 54 (8-570) | 0.44 |
| Days of Hospitalization | 18 (10-40) | 17 (10-50) | 0.50 |
| Percentage of patients with at least one infection | 53% | 47% | 0.68 |
| Percentage of patients with bleeding episodes | 20% | 26% | 0.62 |

Cox proportional hazards regression and multivariate regression analyses were used to assess the potential relationship between various laboratory variables and time to platelet engraftment in the 89 evaluable patients in the Phase 3 study. For each variable, cutoffs were defined at the 25th, 50th, and 75th percentiles of the data. Analysis of the lowest quartile showed that patients who received less than 13.8 x $10^9$ total nucleated cells (at time of cell harvest) and patients who had less than 1.2 x $10^6$ CD34+ cells/kg body weight prior to cryopreservation were at a significantly greater risk of delayed time to platelet engraftment. Similar analyses on the data from the 29 Phase 1/2 study patients who received selected CD34+ cells also demonstrated an increased risk of delayed platelet engraftment in patients who received less than 1.2 x $10^6$ CD34+ cells/kg.

**Long term follow-up Results:**

**Engraftment:**

Additional data were collected retrospectively in the Phase 3 study for the period beyond 100 days to two years after transplantation. There were no significant differences between the study arms with regard to median neutrophil count, leukocyte count, hemoglobin, or platelet count. There were no significant differences in number of platelet transfusions, infections and bleeding episodes.

With regard to stability of engraftment, one patient in the selected arm required infusion of back-up marrow at day 211 in order to achieve hematopoietic reconstitution. Five subjects were leukopenic (leukocytes < 2000 cells/ul) at 6 or 12 months post-transplantation, of these 4 were in the selected arm and 1 in the unselected arm. Eight patients required one or more platelet transfusions between day 100 and 12 months post transplantation, 5 were in the selected arm and 3 in the unselected arm.

Four patients in the selected arm were infused with close to the minimum number of selected CD34+ cells ($0.5 \times 10^6$ CD34+ cells/kg was the minimum requirement). Two of the four had unsustained neutrophil engraftment; one received back-up bone marrow cells post-transplant. A third patient experienced episodic neutropenia.

**Immune function studies:**

Between 6-12 months post-transplant there was equivalent immune reconstitution in patients in both study arms. Testing included mitogen responses, delayed hypersensitivity skin tests for Candida, Tetanus, and PPD (tuberculin), immunophenotyping for Natural Killer cells, CD4+, CD8+, CD19+ (B cells), CD4/CD8 ratios, serum immunoglobulins and antibodies to Cytomegalovirus, Herpes simplex virus and Vesicular Stomatitis virus.

**Progression-free survival and deaths:**

Progression-free survival and survival using Kaplan-Meier analysis showed a less favorable trend in the selected arm which was most marked in the early post transplant period. Median progression-free survival for the selected arm was 40 weeks; for the unselected arm 101 weeks. Median survival in the selected arm was 109 weeks; for the unselected arm the median survival was not yet obtained. Progression free survival curves for the two arms converged at 30 months. The differences between arms were not statistically significant.

**Results of Cell Processing:**

Processing of bone marrow on the CEPRATE® SC System results in an enrichment of $CD34^+$ cells and CFU-GM of 57-fold and 60-fold, respectively. This is accompanied by a 60% or greater decrease in total progenitor cell numbers. Processing results for all Phase 1/2 and Phase 3 patients who received selected bone marrow are shown in Table 5 below.

| Table 5<br>Composition of Autologous Marrow Before and After CEPRATE® SC System Processing | | |
|---|---|---|
| Enrichment<br>(Relative proportion of progenitor cells) | Prior to Selection | After Selection |
| Total $CD34^+$ cells as a percent of TNC<br>n = 73 | 1.3% (0.2 - 4.0) | 75% (16 - 92) |
| CFU-GM per $10^5$ nucleated cells<br>n = 61 | 28 (4 - 112) | 1694 (177 - 10,000) |
| Absolute Progenitor Cell Number | | |
| $CD34^+$ cells (x $10^6$)<br>n = 73 | 248 (53 - 908) | 91 (15 - 288) |
| $CD34^+$ cells (x $10^6$) per kg body wt.<br>n = 73 | 4.1 (0.7 - 15.9) | 1.4 (0.4 - 4.5) |
| CFU-GM (x $10^4$) per kg body wt.<br>n = 61 | 10 (0.9 - 60) | 4 (0.4 - 21) |

## ADVERSE EVENTS

**Infusion related toxicity:**

All adverse events (grades 1-4) and severe/life threatening adverse events (grades 3 and 4) during the first 24 hours after infusion are listed in Table 3. Adverse events were less frequent in the selected than in the unselected control arm (See Clinical Experience).

**Events Reported as Possibly Related to CD34 Selection:**

During the Phase 3 clinical study, five adverse events were reported in the immediate post-transplant period as possibly related to the use of the CEPRATE® SC Stem Cell Concentration System. Delayed platelet recovery was reported for three patients, however, platelet engraftment was ultimately observed in all three patients and all became platelet transfusion independent. In a fourth patient, neutrophil counts decreased on three occasions when granulocyte-colony stimulating factor (G-CSF) was stopped. Counts recovered when G-CSF was restarted; sustained counts were achieved following the third treatment and G-CSF was discontinued. A fifth patient reported experiencing a cool sensation during marrow infusion.

**Marrow Sterility:**

Bone marrow preparations were cultured in the 89 evaluable patients in the Phase 3 trial including cultures before and after CEPRATE® SC System processing. In 3 subjects, marrow that was culture negative prior to processing became culture positive after CEPRATE® SC System processing (2 *Staphylococcus*, coagulase-negative cultures and one *Aspergillus spp* culture). No clinical infections were associated with the positive bone marrow cultures.

**Human Anti-Mouse Antibody (HAMA):**

Processing of cells in the CEPRATE® SC Stem Cell Concentration System results in a theoretical possibility of exposing patients to murine protein; this possibility should be taken into consideration prior to infusion of selected cells into individuals with known hypersensitivity to products of murine origin. Sixty patients were evaluated at baseline, and at days 30 and 100 post-transplant for HAMA response. One of 29 in the selected arm and two of 31 in the control arm showed evidence of HAMA .

**Data Assessment:**

Post-infusional changes in blood pressure and heart rate were significantly lower (about 40% as large) in the group receiving CEPRATE® selected cells. These observed differences were not substantial enough to clearly equate with clinical benefit. However, they do provide proof of concept that use of CEPRATE® selected cells decreased the hemodynamic disturbances associated with bone marrow transplantation and thus they suggest that it can decrease the incidence and severity of adverse consequences sometimes associated with those disturbances. The data in Table 3 suggest that improvements of tolerability of bone marrow transplantation with CEPRATE® selected cells extends beyond the hemodynamic area.

Engraftment of neutrophils was quite similar in the selected and unselected arms and the 95% confidence intervals exclude an increase in engraftment failure rate at day 20 of 12%. By contrast, engraftment of platelets appeared to be delayed in the patients receiving CEPRATE® selected cells. The level of concern about delayed engraftment related to CEPRATE® selection is reduced by the fact that one can identify patients at risk for delayed engraftment based upon the cell yields after CEPRATE® selection.

In follow-up beyond day 100, no significant differences were observed. There was a suggestion that patients receiving low numbers of CEPRATE® separated cells were at risk for unsustained neutrophil engraftment and that patients receiving CEPRATE® selected cells had a higher incidence of late leukopenia. There was a moderate trend toward lower survival and progression-free survival in the treated arm for the first year or two, but curves converged at 2.5 years. The causes of death were nearly all related to breast cancer and not to bone marrow transplantation or its complications. While use of the CEPRATE® device to select cells for bone marrow transplantation theoretically could influence the course of breast cancer by impacting immunocompetence, no effect on immunocompetence was observed. The reasons for the observed survival differences are unclear and are believed to be due to chance rather than any effect of CEPRATE® selection.

**PANEL RECOMMENDATION:**

The Biological Response Modifiers Advisory Committee, meeting on February 28, 1996, found that the CEPRATE® device was safe and effective for selecting CD34+ cells from autologous bone marrow for hematopoietic reconstitution after chemotherapy.

**FDA DECISION:**

Satisfactory information was submitted to FDA in the following documents: PMA submitted January 3, 1994 and amendments submitted June 6, 1994, August 2, 1995, and January 15, 1996. CellPro was issued a letter stating that the device was "Approvable" on April 19, 1996. The FDA inspected the applicant's manufacturing facilities between May 16, 1996 and June 27, 1996. Several deficiencies in Good Manufacturing Practices (GMP) were identified. The applicant agreed to correct the deficiencies and submitted a written report on July 10, 1996 providing assurances that the deficiencies had been or would soon be corrected. As a result of this pre-market inspection, it was necessary to further evaluate the potential toxicity of a plasticizer leaching from the tubing. These issues were resolved and compliance clearance was issued on November 29, 1996.

**APPROVAL SPECIFICATIONS:**

See attached labeling (Attachment A) and Conditions of Approval (Attachment B)

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12/3/96
Date

Jawahar Tiwari, Ph.D.
Statistical Reviewer, CBER

12-3-96
Date

Marjorie Shapiro, Ph.D.
Product Reviewer, CBER

12/4/96
Date

Mercedes Serabian, M.S., DABT
Pharmacology/Toxicology Reviewer, CBER

12/3/96
Date

John Baxley, Ph.D.
Engineering Consult Reviewer, CDRH

12/3/96
Date

Elizabeth W. Shores, Ph.D.
Product Reviewer, CBER

12/4/96
Date

Liana Harvath, Ph.D.
Product Reviewer, CBER

12/4/96
Date