

## **Comparison of the transfusion effects of fresh and cryopreserved apheresis platelets.**

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### **Abstract**

**In this study, the clinical transfusion effects of fresh and cryopreserved apheresis leukocyte-reduced Platelets (PLTs) were examined. In total, 102 patients admitted to our hospital between August 2012 and August 2013 were enrolled, including 50 non-hematonosis patients and 52 hematonosis patients. Among them, 45 received fresh apheresis PLTs and 57 received cryopreserved PLTs. Patient blood PLT counts before and after transfusion were measured and the Percentage of Platelet Recovery (PPR) was compared among groups. The PPR for the cryopreserved PLT group was  $25.03 \pm 52.06\%$ , which was significantly lower than that of the fresh PLT group ( $47.06 \pm 42.94\%$ ,  $t=2.29$ ,  $P<0.05$ ). The PPR of the non-hematonosis group ( $51.01 \pm 65.10\%$ ) was significantly lower than that of the hematonosis group ( $21.40 \pm 24.23\%$ ;  $t'=2.92$ ,  $P<0.05$ ). Fresh apheresis leukocyte-reduced PLTs were safer and more effective than cryopreserved apheresis leukocyte-reduced PLTs. Transfusion was more effective in non-hematonosis patients than in hematonosis patients.**

**Keywords:** Apheresis, Cryopreservation, Hematonosis, Hemostasis, Platelet, Transfusion.

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### **Introduction**

Apheresis Platelets (PLTs) are mainly used to treat haemorrhagic diseases caused by dysfunctional or reduced PLTs to promote the recovery and maintenance of normal hemostasis and coagulation functions. In China, one standard therapeutic dose contains  $\geq 2.5 \times 10^{11}$  PLTs [1]. Fresh apheresis leukocyte-reduced PLTs are characterized by few erythrocytes and leukocytes, high purity, and good clinical effects. They could effectively reduce the occurrence of platelet refractoriness [2]. However, the preservation period is as short as 5 days. Cryopreserved apheresis leukocyte-reduced PLTs have a longer preservation period of 1 year and are suitable for emergency infusions. However, their transfusion effect is not as good as that of fresh PLTs. The Percentage of Platelet Recovery (PPR) is an important laboratory indicator for the accurate evaluation of the efficacy of PLT transfusion [3]. In this report, we utilized PPR to compare and evaluate the transfusion effects of fresh and cryopreserved apheresis PLTs.

### **Materials and Methods**

#### **Patients**

In total, 102 patients admitted to Yantai Yuhuangding Hospital between August 2012 and August 2013 were enrolled in the study, including 47 males and 55 females. All patients

complied with the PLT transfusion indications in the Technical Specifications of Clinical Blood Transfusion of China. All patients received a blood product for the first time. Patients were randomized, and this study was a retrospective analysis.

The patients were randomly divided into the following four groups: (i) the frozen PLT group, which included 28 non-hematonosis patients (18 that had cardiac surgery and 10 that had undergone other surgeries) and 29 hematonosis patients (13 with AML and 16 with MDS); (ii) a fresh PLT group, which contained 22 non-hematonosis patients (14 that received cardiac surgery and 8 that received other surgeries) and 23 hematonosis patients (14 with AML and 9 with MDS); (iii) a hematonosis group; and (iv) a non-hematonosis group. There were no statistical differences in age, sex, disease, and PLT count before transfusion among these groups ( $P>0.05$ ; Table 1).

This study was conducted in accordance with the declaration of Helsinki. This study was conducted with approval from the Ethics Committee of Yantai Yuhuangding Hospital. Written informed consent was obtained from all participants.

#### **Blood processing**

All blood products were sourced from the Yantai Central Blood Bank and met the GB18469-2012 quality requirements for

whole blood and blood components [4]. Fresh apheresis leukocyte-reduced PLTs were collected using an automatic blood component separator (CompoMaster), in which PLTs were separated from the whole blood in a closed state, leukocytes were removed, and they were suspended in plasma. PLTs were preserved in ACD-A preservative solution at 20-24°C with shaking at 60 rpm with 5 cm amplitude. Exposed apheresis PLTs or PLTs were stored in ordinary blood bags for no longer than 24 h. Unexposed PLTs or those stored in PLT-specific bags could be preserved for up to 5 days.

To produce cryopreserved PLTs, 5% (v/v) Dimethyl Sulfoxide (DMSO) was added at 1 ml/min to fresh apheresis leukocyte-reduced PLTs with shaking at 60-80 rpm under sterile conditions and then samples were stored horizontally at -80°C. For defrosting, bags of cryopreserved PLTs were incubated at 40-42°C in a water bath with gentle shaking until a cloudy suspension was formed. The blood bag was checked for any leakage or the formation of fibrous protein and PLT floccules.

### Calculations

Venous blood was collected from patients both before transfusion with apheresis leukocyte-reduced PLT and 12 hours after transfusion. PLT counts were obtained using the XT-1800 Blood Cell Analyser.

PPR (%)=(PLT count after transfusion-PLT count before transfusion) × blood volume/total number of PLT infused × 100 [4]. The blood volume was 75 ml/kg body weight. Cryopreserved apheresis PLTs were used within 120 days of preservation and the average PLT count should be approximately 84.1% of the fresh PLTs [5]. The defrosted cryopreserved PLTs used in this study all met this criterion.

### Statistical analysis

The data were normally distributed (Skewness=-0.453, Kurtosis=0.758). Statistical analyses were performed using SPSS11.5. The t-test was used and P<0.05 indicated a statistically significant difference.

## Results

### Comparison of PPR in the fresh and frozen PLT groups

The PPR of the frozen PLT group was significantly lower than that of the fresh PLT group (t=2.29, P<0.05) (Table 2).

### Comparison of PPR in the hematonosis and non-hematonosis groups

The PPR of the non-hematonosis group was significantly greater than that of the hematonosis group (t'=2.92, P<0.05) (Table 3).

**Table 1.** Grouping of the patients.

	Hematonosis group	Non-hematonosis group
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	M	F	M	F
Frozen PLT group	14	15	13	15
Fresh PLT group	12	11	8	14

**Table 2.** PPR comparison between frozen and fresh PLT group ( $\bar{x} \pm s$ ).

Group	No. of cases	PPR (%)
Frozen PLT group	57	25.03 ± 52.06
Fresh PLT group	45	47.06 ± 42.94*

\*Comparing to frozen PLT group, t=2.29, P<0.05.

**Table 3.** PPR comparison between hematonosis and non-hematonosis group ( $\bar{x} \pm s$ ).

Group	No. of cases	PPR (%)
Non-hematonosis group	50	51.01 ± 65.10
Hematonosis group	52	21.40 ± 24.23*

\*Comparing to non-hematonosis group, t'=2.92, P<0.05.

## Discussion

Complementary to apheresis leukocyte-reduced PLTs, cryopreserved apheresis leukocyte-reduced PLTs have only recently been used [6]. These PLTs benefit from the long preservation period [7]; accordingly, they resolve the conflict between the short preservation period of fresh apheresis PLTs and the urgency of clinical PLT transfusion [8]. Cryopreserved PLTs have satisfactory efficacy when used in emergency situations, including hemorrhage caused by cardiopulmonary bypass [9] and extremely low PLT counts in hematonosis patients [10]. In a related study, 5% DMSO was added to 12 portions of platelet-rich plasma and aliquots were stored at -80°C for 16 months. One aliquot was defrosted in a 37°C water bath every month at 1-6 months and then every 2 months at 8-16 months. Each aliquot was tested for PLT count, MPV, PDW, pH, PLT CD62p expression, thrombin-activated CD62p expression, thrombin-activated CD62p re-expression, PS expression, and aPACT. No significant differences in any of these parameters were detected during the 16 month preservation period (P>0.05), indicating that PLT with 5% DMSO could be stored for a relatively long time without quality deterioration and is safe for long-term PLT preservation [11].

During transportation, PLTs should be kept frozen at all times [12]. Defrosted PLTs should be used immediately or kept at 20-24°C with constant shaking for no longer than 2 hours. After the transfusion of cryopreserved PLTs, 3% of the infused DMSO metabolizes into dimethyl sulfide, which has a foul odor and could be exhaled [13]. Therefore, when a large volume of cryopreserved PLTs is transfused, a patient's breath could have a garlic-like odor [14]. DMSO is toxic to the human body. Therefore, the elderly, children, pregnant women, the infirmed, and patients with renal dysfunction should either not receive frozen PLTs [15] or should receive these PLTs with

extra care and precaution. Once the hemorrhage is controlled, infusion should be stopped immediately [16]. The effect of PLT transfusion is influenced by many factors. An accurate evaluation of the transfusion effect could effectively avoid platelet refractoriness; improve the utilization rate of platelets, and save precious blood resources. PPR is a common indicator for the laboratory evaluation of PLT transfusion effects. It is easily calculated and could objectively and accurately reflect the changes of PLT before and after transfusion [17]; therefore, PPR is widely applied clinically.

Based on our statistical analysis, the transfusion effect of fresh PLTs was superior to that of frozen PLTs; specifically, the former had a significantly higher PPR. This was probably due to the artificial damage caused by rough handling during the preservation and usage of PLTs [11]. The transfusion effect was superior in non-hematomia patients than in hematomia patients, as evidenced by the significantly higher PPR in the non-hematomia group, which can likely be attributed to repeated transfusion.

Cryopreserved PLTs have the advantage of a long preservation time, which addresses the limitations of fresh PLT resources in emergency situations; accordingly, their clinical value is undeniable [18]. Furthermore, chronic and repeated blood transfusions can easily lead to the formation of related antibodies in hematomia patients [19]. Therefore, under circumstances of limited PLT availability and to control medical expenses, these patients should receive a PLT cross-matching transfusion [20]. Choosing the optimal PLT based on the conditions of each patient is of the utmost importance in achieving the best clinical transfusion effect, and the potential strategies still require further exploration. Platelet supplies are currently inadequate in China. For an increasing number of hematomia patients, transfusion with cryopreserved platelets is the only option.

Owing to potential regional differences, the study results may reflect local situations. Additionally, the sample size was limited. The study was also limited with respect to the indicator chosen for the analysis; in future studies, we will compare various indicators.

### Conflict of Interest

Meiling Xing, Dehua Song and Yanbin Li declare that they have no conflict of interest.

### Compliance with Ethical Requirements

This study was conducted in accordance with the declaration of Helsinki. This study was conducted with approval from the Ethics Committee of Yantai Yuhuangding Hospital. Written informed consent was obtained from all participants.

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