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Original Article

To study the effects of gamma irradiation on single donor apheresis platelet units by measurement of cellular counts, functional indicators and a panel of biochemical parameters, in order to assess pre-transfusion platelet quantity and quality during the shelf life of the product

Surg Capt R.S. Mallhi ^{a,*}, Maj A.K. Biswas ^b, Col J. Philip ^c,
Brig T. Chatterjee ^d

^a Associate Professor, Department of Immunohaematology & Blood Transfusion, Armed Forces Medical College, Pune 411040, India

^b Resident, Department of Immunohaematology & Blood Transfusion, Armed Forces Medical College, Pune 411040, India

^c Commanding Officer, Eastern Command Transfusion Centre, Kolkata, India

^d Commandant, 166 Military Hospital, C/O 56 APO, India

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ABSTRACT

Background: The occurrence of transfusion associated graft versus host disease can be prevented by gamma irradiation of blood components. This study was undertaken to assess the effects of gamma irradiation on single donor platelet (SDP) concentrate units.

Method: SDPs were collected by a continuous flow apheresis technique ($n = 400$). The SDPs from each donor were divided into two parts, one gamma-irradiated with 25 Gy and the other used as a non-irradiated control. Swirling and morphological features, cellular counts, biochemical parameters including blood gas analysis, and platelet activation levels (CD62P: p-selectin) by flow cytometry were analyzed on Day 1 and on Day 5.

Results: Swirling and morphology were maintained in all products, in both the groups throughout the shelf life. No significant change was seen in both groups, on the first and fifth day, as far as pO_2 , pCO_2 , Na^+ , K^+ , HCO_3^- & Ca^{2+} were concerned. However, lactate increased and glucose decreased significantly in irradiated products over 5-day storage period. A small but significant decrease in pH and platelet count was found in the irradiated PCs after 5-day storage. The mean proportion of platelets expressing CD62P over 5-day storage increased significantly.

Conclusion: After an overall assessment of all our in vitro parameter results and observations, a few of which were significant, while most were not significant, we concluded that a well-preserved quality of gamma irradiated apheresis platelets is maintained throughout

* Corresponding author. Tel.: +91 9370538174.

E-mail address: pragraj@yahoo.com (R.S. Mallhi).

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the entire 5-day shelf life of the platelet product, with minimal difference compared to non-irradiated platelets.

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Introduction

Transfusion associated graft versus host disease (TA-GVHD) is a rare but a highly lethal complication of cellular blood transfusion. It is defined as a delayed immune transfusion reaction caused due to an immunologic attack by viable donor lymphocytes contained in the transfused blood component against the transfusion recipient.¹ According to UK haemovigilance system severe hazards of transfusion (SHOT), TA-GVHD accounts for 0.2% of complications associated with transfusion of blood products.² The occurrence of TA-GVHD can be prevented by gamma irradiation of cellular blood components with a minimum of 25 Gray (Gy) delivered to the central portion of the product and a minimum of 15 Gy dose elsewhere. It acts by inhibiting the proliferation of T-lymphocytes.¹

The transfusion of irradiated blood components is mainly indicated for transfusion in infant or foetal patient populations (in case of intra-uterine and exchange transfusions), in patients with malignancies or compromised immune systems, individuals receiving cellular blood components from blood relatives and also in patients receiving HLA-matched products, e.g. haematopoietic stem cell transplant recipients.³

The exact number of residual leucocytes (T-lymphocytes) in leuco-reduced blood products that can cause TA-GVHD is unknown.⁴ Hence, it is important to irradiate blood components including platelet concentrate (PC) units, whose demands are growing day by day and for which a large number of PCs are produced by apheresis procedures.⁵ Until now, only a few relevant studies on the influence of gamma radiation on single donor platelet concentrate units (SDPs) obtained by plateletpheresis have been performed, which show conflicting results with regard to in vivo and in vitro platelet characteristics.^{5–10}

Even though some of the earlier studies attempted to evaluate various quantitative and qualitative aspects of platelets as well as their in vivo properties (by radiolabelling studies) in random donor platelet (RDPs) concentrate units and SDPs,^{5–18} the sample size chosen was too small to reach to a conclusion with certainty.

This study was undertaken to assess the effects of gamma irradiation on single donor apheresis platelet units by measurement of its cellular counts, functional indicators and a panel of biochemical parameters, in order to assess pre-transfusion platelet quantity and quality during the entire approved shelf life of the product (i.e. 5 days), so as to unfold any significant deleterious effects occurring due to irradiation.

Material and methods

Study design

This was a prospective, analytical study carried out in the Department of Immunohaematology and Blood Transfusion,

over a period of 2 years from December 2012 to December 2014. The study was carried out after taking prior approval from the Institutional Ethical Committee and informed consent from the plateletpheresis donors.

Donor selection

Platelets were collected by standard apheresis procedures from four hundred (400) healthy voluntary donors, who met the Food and Drug Administration (FDA),¹⁹ Directorate General of Health Services (DGHS) India,²⁰ and American Association of Blood Banks Guidelines and recommendations for plateletpheresis.²¹ It was ensured that none of the subjects were receiving medications known to affect platelet function or kinetics.

Platelet collection

For the study, SDP units were collected on a continuous-flow cell separator, Fenwal Amicus with software version 3.5 (Baxter Healthcare, Deerfield, IL, USA) using the single needle plateletpheresis kit (PL 2410). Acid-citrate-dextrose (ACD-A) was used as anticoagulant agent. The instrument was programmed to collect 3×10^{11} platelets in a volume of 300 ml. On the basis of the entered donor parameters (such as sex, weight, height, haematocrit and pre-procedure platelet count) and the ratio of whole blood to ACD, the instrument calculated the total blood volume to be processed. Contaminating white blood cells were removed by the in-built leucoreduction system of the cell separator.

Storage

After completion of the procedure, each platelet suspension was equally divided into the two integral attached polyolefin containers. This was done with the help of an electronic weighing scale and a tube sealer. A 10 ml representative sample was collected from each of the two bags to analyze the baseline values of the parameters to be measured in the study. The product bags were stored as per laid down guidelines in a platelet agitator/incubator (Terumo Penpol, India) at 22–24 °C with continuous agitation, at a frequency of 60 horizontal agitations/min.

Gamma irradiation

One part of the product was then subjected to gamma irradiation of 25 Gray (Gy) with a ¹³⁷Cs source blood irradiator (Gammacell 3000 Elan, Nordion, Canada), as per manufacturer's directions. The irradiation instrument canister was loaded with single platelet concentrate unit at one time. An irradiation indicator (Rad-Sure type 25Gy, International Specialty Products, Wayne, NJ) was applied to each unit and

irradiated to document a dose delivery of at least 25 Gy to the irradiated product.

Sampling

A 10 ml representative sample was collected 2 h after irradiation on the first day of the shelf life of the product. Another 10 ml representative sample was collected on the fifth (last) day of the shelf life of the product. Similar samples were taken on the first and fifth day, from the second part of the product which was not subjected to gamma irradiation, for comparison.

All samples were drawn aseptically through sampling-site couplers, in 10 ml disposable syringes. Just prior to all sample collection, the content of the bags was gently mixed, so as to get a homogeneous representative sample from the bags.

In vitro investigations carried out on samples

The following parameters were measured on the samples collected from the irradiated and non-irradiated products on the first as well as on fifth day of storage, for comparison. The readings were analyzed to assess any changes in the qualitative and quantitative parameters of platelets pre- and post-gamma irradiation.

(i) Visual assessment of swirling:

Both the product bags (i.e. irradiated as well as non-irradiated) were assessed individually by inspecting against a bright background and swirling was graded according to Singh et al.,²² as given below:

Score 0: Homogeneously turbid and not changed with pressure.

Score 1: Homogeneous swirling only in some parts of the bag and is not clear.

Score 2: Clear homogeneous swirling in all parts of the bag.

Score 3: Very clear homogeneous swirling in all parts of the bag.

(ii) Morphological assessment of platelets:

The morphological assessment of platelets was done at 100× magnification using a Nikon binocular microscope. A smear was made from a small drop of the product on a clean glass slide and stained with Leishman-Giemsa stain. The structural features of platelets were evaluated by a morphology score devised by Kunicki et al.²³ and is based on the following sequence of changes:

- (1) **Discs** – those platelets that have retained the characteristic discoid shape of fresh platelets.
- (2) **Spheres** – the spherical shape seen after refrigeration of platelets, after addition of certain aggregating agents, and often after storage of platelets at room temperature for several days.
- (3) **Dendrites** – platelets that have developed pseudopodia and/or dendritic processes.
- (4) **Balloons** – platelets that have undergone swelling after losing the capacity to maintain an osmotic gradient across their membrane.

The percentage of each morphologic type, assessed by microscopy, was multiplied by a series of arbitrary factors

as follows: discs × 4, spheres × 2, dendrites × 1 and balloons × 0. The morphology score was defined as the total of the four numbers thus derived. At least 200 platelets were scanned each time platelet morphology was assessed. The most superior morphology, or 100 percent discs, was scored as 400.

(iii) Determination of platelet counts:

Platelet count in each sample was determined using an automated blood cell counter (Sysmex KX-21 haematology analyzer, Sysmex Corporation, Japan).

The platelet count in the whole product was calculated as:

$$\text{Platelet count/bag} = \text{platelet count } (\times 10^3)/\mu\text{l} \\ \times \text{volume of the product (ml)} \times 1000 \mu\text{l/ml}$$

The volume of the product was calculated by dividing the net weight of the SDPs by the specific gravity of the platelets (1.026). Weights were measured on an electronic weighing scale.

(iv) Assessment of biochemical parameters:

Various biochemical parameters, such as pH, pO₂, pCO₂, bicarbonate, Na⁺, K⁺, Ca²⁺ and lactate, were measured quantitatively in all the samples using Eschweiler Combiline blood gas analyzer, as per manufacturer's instructions.

(v) Glucose estimation:

Glucose levels in all the samples were estimated using Erba XL 600 biochemical auto-analyzer (Transasia, Erba Mannheim), as per manufacturer's instructions.

(vi) Flow cytometric estimation for platelet activation:

In this study, CD62P (P-selectin) expression was chosen as the marker of platelet activation in the platelet samples. It was assessed with the help of a BD FACS Calibur flow cytometer (Becton Dickinson, NJ, USA) using the CellQuest Pro software, version 5.1, as per the manufacturer's instructions. Staining of samples was carried on fixed samples for which the samples were first fixed with 1% paraformaldehyde solution and stained with a fluorochrome-coupled CD62P-PE reagent (clone AC 1.2) (BD). After 15 min of incubation, 1 ml of PBS was added to the samples and immediately analyzed in the flow cytometer. The staining protocol was developed by BD research and development wing, USA and was validated by BD India representatives on BD FACS Calibur flow cytometer at our institute.

The instrument was set to measure forward and side light scatter in a logarithmic mode. A minimum of 50,000 events or cells were counted by the instrument. The cell population of interest was then gated on the dot plot diagram. The percentages of activated platelets expressing CD62P among the total number of platelets 'gated' (which is taken as 100%) were analyzed with the help of software (Fig. 1).

Statistical analysis

All the calculations were done using SPSS version 20 (IBM, Armonk, NY, USA). Descriptive statistics (mean and standard deviation) were calculated to study the nature of data.

Fig. 1 – Flow cytometer generated report of CD62P estimation on a paired SDP sample (non-irradiated and irradiated) on Day 5 of storage. Notes: (i) In first dot plot, FSC and SSC were selected on x-axis and y-axis, respectively which showed cell distribution based on cell size and complexity of the unstained sample. (ii) Markers (M1) were put for identifying the CD62P marker at 0.4% on x-axis of first histogram, which was for the unstained sample (which acts as a negative control) and hence, acted as a baseline value. (iii) The remaining two histogram plots were for antibody testing, i.e. the second histogram was for stained non-irradiated sample and the third one was for stained irradiated sample.

Group comparisons were performed by using student's paired two-tailed sample t-test. Chi-square test was used to compare the achievement of swirling score of the platelet units. A *p* value of <0.05 was considered, to indicate significant differences between groups.

Results

For each product, homogeneous paired samples for analysis were taken from irradiated and non-irradiated parts of the

Table 1 – Day 5 swirling parameter, of 400 apheresis derived platelet concentrates, irradiated with 25 Gy on Day 1.

Swirling score	Non-irradiated (n = 400)	Irradiated (n = 400)	Chi-square	p value
3	356 (89%)	348 (87%)	0.76	0.384
Less than 3 (2, 1, 0)	44 (11%)	52 (13%)		

product bags, derived from the same donor. Various in vitro (qualitative as well as quantitative) parameters were assessed in both the irradiated and their respective control groups and are shown in Tables 1–3.

Swirling and morphology were maintained in all the products of both groups, throughout their shelf life. Also, there was no significant change seen, either in the non-irradiated or the irradiated products, on the first as well as on the fifth day, as far as pO_2 , pCO_2 , Na^+ , K^+ , HCO_3^- and Ca^{2+} were concerned.

The irradiated suspensions showed a significantly lower pH on the 5th day of storage ($p < 0.05$). However, pH was within accepted range (>6.0) during the whole storage period for both groups of PCs. Glucose decreased and lactate increased significantly on Day 5 of storage in irradiated PCs compared to non-irradiated PCs ($p < 0.05$).

In our study, the platelet activation levels measured by CD62P expression were found to be significantly higher ($p < 0.05$) in the irradiated group from Day 1 through Day 5. The platelet count decreased slightly but significantly ($p < 0.05$) in the irradiated PCs on the fifth day of storage, but it still remained well above the minimum mandatory quality control requirement for platelet concentrates, i.e. $\geq 3 \times 10^{11}$ per unit.

Discussion

The clinical demand for PCs has grown markedly in recent years. TA-GVHD is a serious and most often, lethal side effect of transfusion of PCs to immunosuppressed recipients. For this reason, the PCs are irradiated before their transfusion to

Table 2 – Day 1 in vitro parameters, of 400 apheresis derived platelet concentrates, irradiated with 25 Gy on Day 1.

Parameters	Mean $\pm$ SD (95% CI)		p value (2-tailed)
	Non-irradiated (control)	Irradiated	
Morphology score	338 $\pm$ 17 (321–355)	338 $\pm$ 17 (321–355)	0.537
Platelet counts	1363 $\pm$ 134 (1229–1497)	1362 $\pm$ 134 (1228–1496)	0.072
pH	6.84 $\pm$ 0.31 (6.53–7.15)	6.82 $\pm$ 0.21 (6.61–7.2)	0.095
pO_2	30 $\pm$ 9.9 (20.1–39.9)	29.9 $\pm$ 10.2 (19.7–40.1)	0.331
pCO_2	59.6 $\pm$ 8.2 (51.4–67.8)	59.6 $\pm$ 7.5 (52.1–67.1)	0.921
Na^+	150.9 $\pm$ 10.4 (140.5–161.3)	151.1 $\pm$ 10.4 (140.7–161.5)	0.269
K^+	2.75 $\pm$ 0.26 (2.49–3.01)	2.74 $\pm$ 0.25 (2.49–2.99)	0.383
HCO_3^-	14.6 $\pm$ 1.7 (12.9–16.3)	14.5 $\pm$ 1.5 (13–16)	0.104
Ca^{2+}	6.75 $\pm$ 0.3 (6.45–7.05)	6.76 $\pm$ 0.3 (6.46–7.06)	0.424
Lactate	3.143 $\pm$ 0.7 (2.443–3.843)	3.145 $\pm$ 0.5 (2.645–3.645)	0.894
Glucose	423 $\pm$ 36 (387–459)	425 $\pm$ 31 (394–456)	0.130
CD62P	26.21 $\pm$ 7 (19.21–33.21)	27.71 $\pm$ 7.2 (20.51–34.91)	$<0.001^a$

^a p value < 0.05 is considered to be significant.

Table 3 – Day 5 in vitro parameters, of 400 apheresis derived platelet concentrates, irradiated with 25 Gy on Day 1.

Parameters	Mean $\pm$ SD (95% CI)		p value (2-tailed)
	Non-irradiated	Irradiated	
Morphology score	288 $\pm$ 10 (278–298)	288 $\pm$ 10 (278–298)	0.112
Platelet counts	1225 $\pm$ 152 (1073–1377)	1156 $\pm$ 124 (1032–1280)	$<0.001^a$
pH	6.57 $\pm$ 0.25 (6.32–6.82)	6.56 $\pm$ 0.25 (6.31–6.81)	$<0.001^a$
pO_2	27.7 $\pm$ 10.2 (17.5–37.9)	27.6 $\pm$ 10.7 (16.9–38.3)	0.141
pCO_2	62.9 $\pm$ 8.2 (54.7–71.1)	62.7 $\pm$ 8.2 (54.5–70.9)	0.622
Na^+	146.4 $\pm$ 11.3 (135.1–157.7)	146.4 $\pm$ 11.5 (134.9–157.9)	0.825
K^+	3.18 $\pm$ 0.45 (2.73–3.63)	3.19 $\pm$ 0.5 (2.69–3.69)	0.234
HCO_3^-	13.6 $\pm$ 1.8 (11.8–15.4)	13.6 $\pm$ 1.8 (11.8–15.4)	0.206
Ca^{2+}	6.47 $\pm$ 0.9 (5.57–7.37)	6.47 $\pm$ 0.9 (5.57–7.37)	0.087
Lactate	4.16 $\pm$ 1.4 (2.76–5.56)	4.52 $\pm$ 1.5 (3.02–6.02)	$<0.001^a$
Glucose	395 $\pm$ 26 (369–421)	384 $\pm$ 38 (346–422)	$<0.001^a$
CD62P	27.7 $\pm$ 6.8 (20.9–34.5)	29.41 $\pm$ 7 (22.41–36.41)	$<0.001^a$

^a p value < 0.05 is considered to be significant.

prevent the occurrence of TA-GVHD. Although irradiated PCs have been transfused routinely over the last couple of decades, only a limited number of studies have examined how irradiation influences the properties of human platelets, with conflicting results.⁵

The available data from previous studies give an impression that an examination of the influence of gamma radiation on apheresis platelet components is unnecessary, because there appears to be sufficient data on the effect of irradiation on platelet products. However, a detailed analysis of all these studies shows that the sample size of all these studies was extremely small and the results may not truly reflect the actual platelet quality, since tests were carried out on a limited number of samples.

In this day and age, where the indications for irradiation of blood components are expanding, thereby increasing the number of patients affected to a larger extent, the issue of effects of irradiation on platelets should be settled once and for all. This is also required for strict implementation of the current concept of total quality management (TQM) of blood components in transfusion medicine.⁵ This will ensure that the ideal quality of irradiated platelet concentrate is transfused to patients in whom such a product is indicated.

In view of above, our study was undertaken to find out the effect of gamma irradiation on the pre-transfusion quality of apheresis-derived platelet concentrate units by exhaustive multi-parameter assays to estimate any deleterious effects of gamma irradiation that can occur to the apheresis-derived platelet products, which in turn would affect the *in vivo* survivability of the platelets.

In our study, there were no significant differences found between the non-irradiated and irradiated platelet groups, throughout the shelf life of the product in the following parameters: swirling and morphological score, blood gases (pO_2 , pCO_2) and supernatant Na^+ , K^+ and HCO_3^- concentrations. Other than Na^+ and K^+ concentrations which were not evaluated by van der Meer et al.¹⁸ on RDPs, the remaining parameters are in consensus with this study.

Lin et al.⁸ on the other hand found a significantly increased mean pO_2 , and mean concentrations of K^+ , after 5-day storage of irradiated SDP products. In addition, the pCO_2 levels decreased significantly, along with the concentrations of HCO_3^- , in the irradiated group. Some findings of the above study such as increase in mean pO_2 and decrease in pCO_2 appear somewhat discordant.

In our study, comparable value of Ca^{2+} concentration was found in both the non-irradiated and irradiated groups. This parameter has never been evaluated in any of the previous studies till date.^{5–18} We included this parameter, as a surrogate marker for platelet activation since calcium ion imbalance predisposes to platelet activation and further enhances platelet aggregation, thereby compromising platelet function.

We found a small but significant decrease in pH in the irradiated PCs, after 5-day storage, when compared with the non-irradiated control PCs. This is in accordance with previous reports by Sweeney et al.⁷ and Bessos et al.¹¹ There are, however, also studies that have reported no differences in pH between irradiated and non-irradiated PCs.^{5,6,15,16,18} In our study, even though the pH decreased significantly in the irradiated PCs on the fifth day of storage, it still remained well

above the minimum mandatory quality control requirement for platelet concentrates, i.e. more than 6. The irradiated PCs, therefore, still remained suitable for transfusion throughout the shelf life, as far as pH is concerned. One of the possible explanations for the significant but slight decrease in pH during 5-day storage may be attributed to increased lactate production as a result of anaerobic glycolytic metabolism. Even though the platelets rely mainly on aerobic oxidative metabolism for ATP synthesis²⁴ *in vivo*, anaerobic glycolytic pathways may also contribute towards ATP production *in vitro*, due to a relatively limited supply of oxygen and other nutrients within the platelet concentrate bags. However, these changes are not related to a failure to maintain normal energy metabolism, as found out by Sweeney et al.,⁷ in which they have found out that the total ATP levels were not affected in the irradiated group.

In our study, glucose decreased and lactate increased significantly in irradiated products on Day 5. This is in consensus with other studies.^{6–9} On the other hand, van der Meer et al.¹⁸ found similar trend in both lactate and glucose levels, but this was not found to be significant. Reduced glucose and increased lactate can be attributed to the ongoing cellular metabolic mechanisms described above. This appears to be slightly more in the irradiated product, probably due to a relatively stressful state induced by irradiation producing some degree of compromise in the cell membrane integrity and mitochondrial DNA. These effects are known to be produced in varying degrees by gamma radiation.

We found the platelet activation levels (measured by CD62P expression) to be significantly higher in the irradiated group from Day 1 through Day 5. These findings are in consensus with those of Tynngård et al.⁹ Studies done by Sweeney et al.⁷ and van der Meer et al.¹⁸ showed a similar trend of CD62P levels as our study, although not significant. On the contrary, Zimmermann et al.⁵ and Lin et al.⁸ found highest CD62P expression in the non-irradiated products when compared to irradiated products. This seems to be at a variance with the overall trend and is difficult to explain.

In both the non-irradiated and irradiated products, platelets get activated as a consequence of the effects of platelet storage lesions. Platelet activation occurs as a result of lysis of intra-cytoplasmic granules and is measured in the form of platelet surface CD62P expression. This mechanism of platelet activation may be amplified as a result of stress due to various biochemical changes induced by irradiation, as discussed earlier. Hence, the CD62P expression was found to be significantly higher in the irradiated group from Day 1 through Day 5. However, the difference between the platelet activation levels in both irradiated and non-irradiated products were found to be very minimal, and may not significantly affect the *in vivo* platelet survivability.

In our study, the platelet count of the irradiated product decreased slightly but significantly on the fifth day of storage. This is at variance with previous studies on the subject.^{5–9,15–18} However, in our study although the platelet count decreased slightly but significantly in the irradiated PCs on the fifth day of storage, it still remained well above the minimum mandatory quality control requirement for platelet concentrates, i.e. $\geq 3 \times 10^{11}$ per unit, making the product perfectly suitable for transfusion even on the fifth day of storage. The probable

explanation for the slight decrease in the platelet count can be the end result of all the deleterious effects observed in our study such as decrease in pH, increase in lactate, decrease in glucose and increase in activation, measured by CD62P expression, thereby possibly leading to a compromise in membrane integrity.

In our study, even though there were some statistically significant differences found between non-irradiated and irradiated PCs in some of the essential in vitro quality control parameters, such as pH and platelet counts, it can be very well be appreciated that the differences found were very small and all platelet products in both the groups qualified the minimum mandatory quality control requirements, as approved by governing agencies. Other parameters measured in our study which were statistically significant between the two groups but which are not a part of the standard quality criteria of platelet concentrates are lactate, glucose and CD62P expression. However, the variations in values were not substantial between the two groups. The overall findings, therefore, indicate that an adequate pre-transfusion quality of apheresis platelet concentrates is maintained after irradiation on Day 1.

The above findings can be taken as guidelines for a standard schedule for irradiating platelet concentrates. In case of a resource-equipped centre, “universal irradiation” on Day 1 should be performed, since irradiation on Day 1 does not appear to have significant deleterious effect on the 5-day shelf life of the platelet product. So in a tertiary care centre (with a large clientele of patients requiring irradiated platelet concentrates), irradiation should be done on Day 1 of storage for optimal patient benefit, by almost eliminating the chances of TA-GVHD.

Another very important aspect to be highlighted for many smaller transfusion centres, which are not equipped with onsite facilities for blood irradiation, is that provision of Day 1 irradiated blood products to such centres in the vicinity of a central collecting facility would greatly supplement the quality of platelet concentrates held in their inventory.¹⁶ This practice would improve their efficiency in providing this specialized blood product and would prevent the administration of unirradiated platelet concentrates to immunocompromised patients, especially in emergency situations.

Hence, in view of above findings, it is recommended that “universal irradiation” on Day 1 should be performed in a resource-equipped centre for optimal patient benefit, as it is a onetime investment with no significant recurring expenditures and will lead to the optimal utilization of the gamma irradiator instrument. Further, placement of Day 1 irradiated platelet concentrate units in the inventory can also assist smaller transfusion centres, by eliminating administrative and technical inadequacies, which could lead to denial of the irradiated products to patients where they are indicated.

Our study though armed with strength of a large sample size and exhaustive in vitro parameters, had some limitations of excluding certain highly sophisticated and expensive in vitro parameters such as electron microscopy, platelet aggregation, markers of membrane integrity (e.g. LDH, HSR), cytokine levels (e.g. IL-8, IL-1 beta, TNF-alpha, TxB₂), ATP levels, other markers of platelet activation (e.g. PF3, beta-thromboglobulin) and in vivo platelet survival by radiolabeling studies. However, it is not feasible for a single study to

incorporate all the above-mentioned parameters. Future studies can attempt to evaluate these parameters also, by sharing the labour-intensive and expensive work by way of multi-centric research, which will ensure feasibility, while ensuring a large sample size. Further such studies can also explore the possibility of the role of recently developed platelet additive solutions (PAS) in improving the quality of stored irradiated platelet products.

Conclusion

After an overall assessment of all our in vitro parameter results and observations, a few of which were significant, while most were not significant, we concluded that a well-preserved quality of gamma irradiated apheresis platelets is maintained throughout the entire 5-day shelf life of the platelet product, with minimal difference compared to non-irradiated platelets.

Conflicts of interest

The authors have none to declare.

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