ARTICLE

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Non-cryopreserved autologous peripheral blood stem cell transplantation for multiple myeloma and lymphoma in countries with limited resources: practice considerations from the Worldwide Network for Blood and Marrow Transplantation

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Autologous peripheral blood stem cell (PBSC) transplantation is a standard treatment of multiple myeloma (MM), Hodgkin lymphoma and various subtypes of non-Hodgkin lymphoma. Cryopreservation of hematopoietic stem cells is standard practice that allows time for delivery of conditioning regimen prior to cell infusion. The aim of this Worldwide Network for Blood & Marrow Transplantation (WBMT) work was to assess existing evidence on non-cryopreserved autologous transplants through a systematic review/meta-analysis, to study feasibility and safety of this approach. We searched PubMed, Web of Science and SCOPUS for studies that utilized non-cryopreserved autologous PBSC transplantation. Identified literature was reviewed for information on mobilization, apheresis, preservation and viability, conditioning regimen, engraftment, response, and survival. Results highlight collective experience from 19 transplant centers (1686 patients), that performed autologous transplants using non-cryopreserved PBSCs. The mean of infused CD34+ was 5.6×10^6/kg. Stem cell viability at transplantation was >90% in MM and >75% in lymphomas, after a storage time of 24–144 h at +4 °C. Mean time-to-neutrophil engraftment was 12 days and 15.3 days for platelets. Pooled proportion estimates of day 100 transplant-related mortality and graft failure were 1% and 0%, respectively. Non-cryopreservation of apheresed autologous PBSCs appears feasible and safe.

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INTRODUCTION

Autologous hematopoietic progenitor cell transplantation represents a supportive rescue therapy that allows the use of high-dose chemotherapy or chemoradiotherapy in hematological malignancies and some solid tumors [1–3]. As growth factors such as Granulocyte Colony Stimulating Factor (G-CSF) became available in the early 1990s, peripheral blood stem cells (PBSC) collected by leukapheresis have replaced harvested bone marrow (BM) as a source of hematopoietic stem cells in almost all patients [4]. Several studies have demonstrated a survival benefit of autologous PBSC following high-dose therapy versus conventional chemotherapy in multiple myeloma (MM) and malignant lymphoma [5–8]. The standard of care for autologous PBSC transplantation involves a two-step procedure. The first step involves the collection of mobilized autologous PBSCs facilitated by growth factors alone or in combination with priming chemotherapy followed by stem cell cryopreservation. The second step involves administration of high-dose conditioning therapy and infusion of the cryopreserved stem cells after being thawed. However, cryopreservation of stem cells requires financial and personnel resources including equipment for programmed freezing and liquid nitrogen availability for storage, in addition to temperature-monitoring capabilities and trained staff. Quality control during thawing is also required. However, these steps are time-consuming and expensive [9].

Several attempts have been made to consider alternative strategies to circumvent the need for cryopreservation, including maintaining PBSCs at 4 °C in a conventional refrigerator. The use of non-cryopreserved BM as a source of hematopoietic progenitor cells in animal models was first described in the 1950s [10]. This field has witnessed progress in the use of non-cryopreserved BM and PBSCs as a source of stem cells in clinical autologous stem cell transplant studies [11–14].

There are no published randomized controlled studies on the use of non-cryopreserved versus cryopreserved autologous PBSCs; and published reports are limited to retrospective single-center and observational comparative studies. To our knowledge, only two literature reviews have been published on this topic. The first by Wannesson et al. in 2007 on autologous stem transplantation (BM and PBSC) in hematological malignancies and solid tumors [15] and the second by Al-Anazi et al. in 2012 on the autologous PBSCs transplantation in MM [16].

The objective of this Worldwide Network for Blood & Marrow Transplantation work is to study the feasibility of using non-cryopreserved ("fresh") hematopoietic stem cells for autologous PBSC transplants in countries with limited resources where cryopreservation facilities may not be available and to provide practice considerations.

METHODS

Search and study selection

We searched the literature (PubMed, Web of Science, and SCOPUS) for original articles, and case series using the keywords "autologous" and "stem cells" and "non-cryopreserved" and "MM" and "lymphoma." All identified articles reporting on autologous transplantation using non-cryopreserved PBSCs in patients with MM and malignant lymphomas were included. The search included an end-date limit of February 2022. The selected studies were analyzed in terms of stem cell mobilization, apheresis, storage and viability, conditioning regimen, engraftment, disease response and survival. Given the anticipated heterogeneity of these studies pertaining to baseline characteristics, the results of this work are presented descriptively. To be eligible for inclusion in the systematic review/meta-analysis (SR/MA), studies must have enrolled a minimum of ten patients.

We collected information regarding the location and time period of the included studies whenever this information was available and excluded studies with obvious overlap. Disagreements were resolved by consensus with two other authors.

Data collection

We extracted data on clinical outcomes including graft failure (GF), time to platelet and neutrophil engraftment, CD34+ cells, and transplant-related mortality (TRM). Factors affecting non-cryopreservation outcomes such as stem cell mobilization, storage, cell viability, and conditioning regimens were also extracted. Methodologic quality of the included studies was evaluated using the Newcastle–Ottawa Scale adapted for single-arm cohort studies where all items and overall assessment was summarized as either high, low or unclear risk [17].

Statistical analysis

The meta-analysis was performed utilizing the DerSimonian and Laird random-effect model to account for variations in treatment effects across studies in order to estimate the inter-study treatment effect variance [18, 19]. Continuous variables from each study were extracted and summarized by calculating the pooled mean and standard error. Binary variables were extracted as events and total sample and pooled as proportions with corresponding 95% confidence intervals (95% CI). However, given that all studies reported continuous outcomes utilizing order statistics such as medians, ranges, and interquartile ranges, the Wan method [20] was employed to transform these order statistics into means and standard deviations.

Pooled proportion estimates for binary outcomes were obtained using the double arcsine transformation variance-stabilizing transformation method, as proposed by Freeman and Tukey [21] to adjust for skewness [22].

Forest plots were utilized to display the point and pooled estimates of study effects together with their corresponding confidence intervals [23].

Heterogeneity among studies was evaluated using the I^2 test based on criteria set by Higgins et al. [24]. Low, moderate, and high heterogeneity were categorized as $I^2 < 30\%$, >30%, and >60%, respectively. Sensitivity analysis was performed to identify influential studies with substantial impact on the pooled estimates. This was accomplished by examining the impact of eliminating individual studies one at a time on the pooled estimates. Statistical analysis was performed using RStudio 2023.09.1 Build 494 © 2009–2023 Posit Software, PBC with *meta* and *metafor* packages. This review follows the PRISMA principles and adheres to the PRISMA guidelines [25].

RESULTS

Search results and characteristics of eligible studies

Our search strategy identified 108 manuscripts and twenty abstracts published between 2000 and 2021 (Fig. 1). Only twenty met our inclusion criteria and were thoroughly analyzed. Countries of origin for these publications, by alphabetical order, were Algeria, Brazil, Chile, Colombia, Egypt, Greece, India, Iran, Mexico, Morocco, and Thailand. The majority focused on MM and malignant lymphomas, except for three studies that included patients with acute leukemia and solid tumors (Table 1) [26–44]. Over a period of 21 years, a total of 16886 autografts with non-cryopreserved stem cells were reported. Results are described below. Characteristics of the eligible studies are reported in Table 1.

Assessment of methodologic quality

Details on interventions (i.e., exposure) was determined from secure records for all studies, and appropriate precautions were taken to ensure that outcomes of interest were absent before the study started. All studies had appropriate follow-up time for outcomes of interest (>1 year). Table 2 summarizes the methodological quality of included studies.

Mobilization. Most centers ($N = 17$) performed PBSCs mobilization using G-CSF alone, while two groups used G-CSF in combination with chemotherapy.

Storage. The collected PBSCs were stored in a refrigerator at +4 °C for a period ranging from 1 to 6 days depending on the type of conditioning regimen to be used. For MM patients,

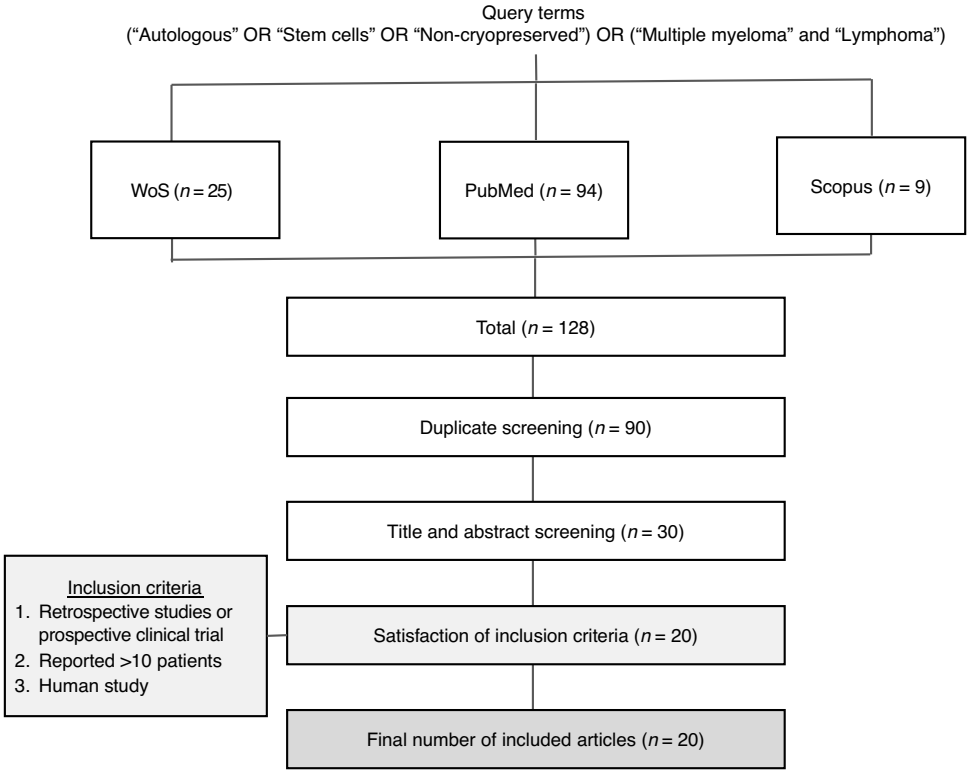

Fig. 1 Study selection process.

storage time ranged from 1 to 2 days, whilst it was 3 to 6 days for lymphoma cases (Table 1) [12, 14, 34, 36, 44].

Cell viability. The viability of PBSCs was calculated by both trypan blue technique and by flow cytometry [45]. Only twelve studies reported the viability; the method and the way of reporting was very heterogeneous among studies (Table 1). The lowest reported viability value was 75% and the highest 98.5%.

Conditioning regimens. The conditioning regimen was dependent on the diagnosis. In MM, all studies used melphalan at a dose range of 140–200 mg/m^2 [16, 31, 32, 34, 35]. In malignant lymphoma, the protocols types varied: melphalan, CBV (cyclophosphamide, carmustine, etoposide) [46], BEAM (carmustine, etoposide, cytarabine, melphalan) [47], CEAM (lomustine, etoposide, cytarabine, melphalan), EAM (etoposide, cytarabine, melphalan) [48], CEC (cyclophosphamide, etoposide, carboplatin), MEL/VP16 (melphalan, etoposide) and their duration ranged from 3 to 6 days (Table 1).

Outcomes

Apheresis. PBSCs apheresis was performed using devices such as Haemonétics®, Cobe Spectra® or Optia®. Normality test for CD34+ data showed no significant deviation from normality; therefore, the Wan method was directly applied to transform medians, min, max, first quartile (Q1), and third quartile (Q3) into means and SDs. Pooled CD34+ mean estimate was 5.6×10^6 kg (95% CI = 5.13×10^6/kg; 6.116×10^6/kg). The *Q*-test showed significant heterogeneity among studies, with a *p* value of <0.01. The between-study variance, represented by T^2, was 0.99 (95% CI = 1; 5.6. 97.6% of the variation was attributed to between-study variance, as shown by an $I^2 = 97.6\%$ (95% CI = 97.0%; 98.1%) (Fig. 2). Sensitivity analysis showed no change in I^2 when excluding studies one by one.

Engraftment. Engraftment was defined as attainment of an absolute neutrophil count (ANC) ≥ 0.5×10^9/L and a platelet count ≥ 20×10^9/L, except for one study in which the threshold was 25×10^9/L platelets [27]. The results of engraftment in different studies are shown in Table 3. Normality test for time to ANC engraftment data showed significant deviation from normality (p < 0.001). Therefore, the Box-Cox transformation method was applied prior to the pooled mean estimation. The pooled mean estimate of the time-to-ANC engraftment was12 (95% CI = 11.3; 12.5). Significant heterogeneity was identified using *Q*-test, $p < 0.001$. T^2 value of 1.9 (95% CI = 1.8; 8.4) suggests a substantial level of heterogeneity. Nevertheless, 96.5% (95% CI = 95.6%; 97.3%) of the heterogeneity was attributed to between-studies variance as indicated by I^2 (Fig. 3). Sensitivity analysis showed no effect of study removal.

The pooled mean estimate of the time to platelet engraftment was 15.2 (95% CI = 14.2; 16.2) days. The *Q*-test showed significant heterogeneity, T^2 of 4.2 (95% CI = 4.2426; 20.9), *p* value < 0.001. Moreover, as indicated by I^2, 95.8% (95% CI: 94.5%; 96.7%), of heterogeneity was attributed to between-studies variance (Fig. 4). Sensitivity analysis revealed no effect of removing studies.

TRM

Pooled proportion of TRM was 13.5% (95% CI = 10.2% to17.5%). The total heterogeneity test revealed a significant heterogeneity using *Q*-test, $p = 0.008$. However, low level of heterogeneity was attributed to between-study variance as indicated by T^2 of 0.002 (95% CI = 0.0002 to 00.0079). Only 45% of heterogeneity was attributed to between-study variance as evidenced by I^2 of 45% (95% CI = 5.9; 71.3%) (Supplementary Fig. 1).

Graft failure

Pooled proportion of GF was 0 with non-significant heterogeneity, $p = 0.9$. Only two of 20 studies reported non-zero GF (Supplementary Fig. 2).

Post-transplant survival outcomes. Analyzed studies were heterogeneous in terms of diagnosis, pre-transplant therapy, and conditioning regimen prescribed. The aim of these studies was

Table 1. Patient-, disease-, treatment- and storage-related characteristics.

Author	*N*	Age (years)	Diagnosis	Conditioning regimen	CD34 cell dose × 10^6/kg	Storage time at + 4 °C (h)
Papadimitriou et al. [26]	72	46–68	MM /NHL /HL/Others	Mel140–180 /Mel-VP16	3 (0.8–2.78)	24–60 h
Ruiz-Argüelles et al. [27]	46	8–69	MM/NHL/HL /AML/ALL/ Others	Mel200	4.68	24–72 h
Cuellar-Ambrosi et al. [28]	47	12–67	MM/NHL/Others	CBV/CTX-TBI /Mel200	3.9 (0–16.9)	24–144 h
Mabed et al. [29]	28	16–50	HL	CTX/VP16 /Carboplatin	6.4 (3.8–24.6)	24–72 h
Mabed et al. [30]	32	17–55	NHL	CBDA/VP16 /CTX	NR	24–72 h
Lopez-Otero et al. [31]	26	42–66	MM	Mel200	7.56 (0.92–14.8)	24–72h
Ramzi et al. [32]	45	16–50	HL	CEAM	3.4 (1.9–9)	72 h
Ramzi et al. [33]	38	31–70	MM	Mel140/Mel200	3.6 (2.4–5.8)	48 h
Bekadja et al. [34]	54	35–65	MM	Mel200	3.60 (1.90–10.52)	24 h
Kayal et al. [35]	92	22–65	MM	Mel200	2.9 (0.9–7.67)	24–120 h
Bekadja et al. [51]	45	17–46	HL	CBV/BEAM/BeEAM/ EAM	3.61 (2.90–21.05)	72–144 h
Bekadja, et al. [36]	240	35–65	MM	Mel140/Mel200	5.7 (1.90–10.52)	24 h
Sarmiento M, et al. [37]	42	22–68	MM/NHL/HL	Mel200	5.1 (2.5–5.6)	48–144 h
Karduss-Urueta et al. [38]	359	59 (34–68) 34 (14–64)	MM/ Lymphoma	Mel200/BEAM/CBV	3.6	48–144 h
Naithani et al. [39]	76	56 (34–68) 34 (14–64)	MM Lymphoma	Mel200/140 BEAM	2.56 (1.22–17.9)	48–144h
Kulkarni et al. [40]	224	50 (23–68)	MM	Mel200	4.87 (1.15–23.7)	24–72 h
Bittencourt [41]	45	53.8	MM	Mel200/140	3.5	24–48 h
Jennane [42]	55	43 (37–67)	MM	Mel140/Mel200	4.5 (2–12.2)	24–48 h
Piriyakhuntorn [43]	26	55.7	MM	Mel200	3.8 (2.0–16.5)	24–48 h
Bekadja [44]	94	29 (17–60)	HL/NHL	CBV/BEAM/BeEAM/ EAM	4.12 (3.4–5.4)	144 h

HDCT high-dose chemotherapy, *MM* multiple myeloma, *NHL* non-Hodgkin lymphoma, *HL* Hodgkin lymphoma, *AML* acute myeloid leukemia, *ALL* acute lymphoblastic leukemia, *Mel* melphalan, *VP16* etoposide, *CBV* cyclophosphamide, carmustine and etoposide, *CTX* cyclophosphamide *TBI* total body irradiation, *BEAM* carmustine, etoposide, cytarabine, melphalan, *NR* Not reported.

to focus on feasibility of autologous transplantation without cryopreservation rather than assessing long-term outcomes such as overall survival (OS) and progression-free survival (PFS). A formal statistical evaluation could not be performed to assess OS and PFS. However, selected studies have reported the long-term outcomes which are summarized in Supplementary Table 1.

Comparative studies. A limited number of non-randomized studies compared the use of cryopreserved and non-cryopreserved stem cells in autografts. A number of these significant studies are listed in Supplementary Table 2. Sarmiento et al. compared 74 patients from the "Institut Catala d'Oncologia Hospitalet" in Barcelona, Spain receiving cryopreserved PBSCs with 42 patients from the "Pontificia Universidad Católica" (Santiago, Chile) receiving non-cryopreserved PBSCs. Results showed a faster neutrophil engraftment and shorter hospitalization duration in the non-cryopreserved group [37]. Similar findings were reported by Bittencourt et al. who compared 63 patients receiving cryopreserved PBSCs with 45 patients receiving non-cryopreserved PBSCs and showing faster neutrophil engraftment and lower toxicity in the non-cryopreserved cohort [41]. Furthermore, their cost-benefit analysis of the latter study revealed a total cost of US $1300 for one cryopreserved PBSC unit but only US $300 for one non-cryopreserved unit. Garifullin et al. [49] reported findings of 78 patients with MM receiving PBSCs and reported no difference between non-cryopreserved and cryopreserved PBSCs.

In contrast, a recent study from Algeria compared results of a matched-pair analysis of autografts performed using non-cryopreserved PBSC with those from the EBMT group using cryopreserved cells in patients with lymphoma, showing faster ANC engraftment in cryopreserved PBSCs on day 10 (48% vs. 17%) [44]. However, all patients in both groups had ANC engraftment by day 20 and there was no difference in TRM, relapse, PFS or OS.

RECOMMENDATIONS AND PRACTICE POINTS

The limited availability of transplantation in resource-limited countries highlights the need for efficient fund allocation, such as the use of non-cryopreserved autologous PBSC transplants, to better allocate available resources and increase transplant activity.

A significant number of autologous transplant procedures using non-cryopreserved PBSC take place in nonacademic centers in low to middle income countries; and it is likely that they do not get reported. However, based on published literature, we can provide a number of practice recommendations regarding non-cryopreserved autograft planning to guide practitioners in areas of limited resources.

Table 2. Risk of bias assessment.

Domains	Comparability		Outcomes							
Study	Representativeness of cohort	Selection of the non-exposed cohort	Ascertainment of exposure	Absence of outcomes at start of study	Comparability of cohorts	Score	Assessment of outcome	Sufficient Follow-up duration	Adequacy of follow-up	Final assessment
Papadimitriou et al. [26]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Ruiz-Argüelles et al. [27]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Cuellar-Ambrosi et al. [28]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Unclear risk	Unclear risk	High risk
Mabed et al. [29]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Mabed et al. [29]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Lopez-Otero et al. [31]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Ramzi et al. [32]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Ramzi et al. [33]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Bekadja et al. [34]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Kayal et al. [35]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Bekadja et al. [51]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Bekadja, et al. [36]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Sarmiento et al. [37]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Karduss-Urueta [38]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Naithani et al. [39]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Kulkarni et al. [40]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Bittencourt [41]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Jennane S [42]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Piriyakhuntorn [43]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk
Bekadja [44]	Low risk	NA	Low risk	Low risk	NA	NA	Low risk	Low risk	Unclear risk	Low risk

NA not available or not applicable.

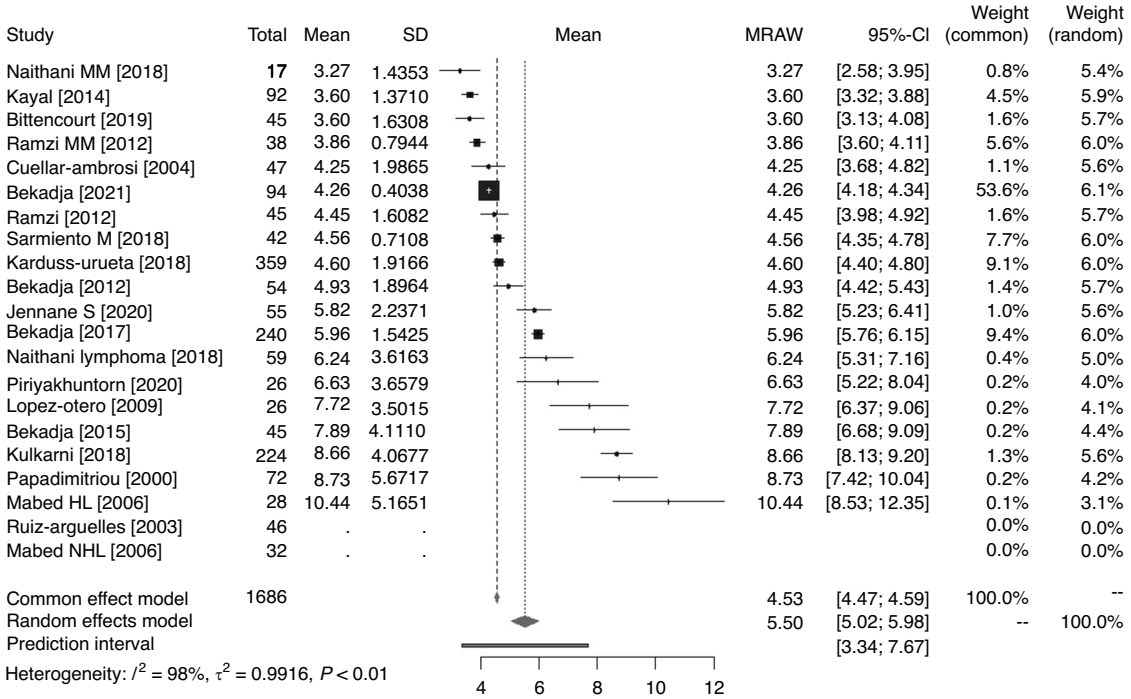

Fig. 2 Pooled mean estimates of CD34+ cell dose. MRAW—represents the untransformed means along with confidence intervals; common and random shows the weightage given to each study in pooling of summary result from each study.

Table 3. Results of engraftment with non-cryopreserved autologous peripheral blood progenitor cell transplantation.

Author	*N*	Neutrophil engraftment, median (range)	Platelet engraftment, median (range)	TRM (%)	Graft failure (%)
Papadimitriou et al. [26]	72	9 (6–16)	5 (0–89)	0	0
Ruiz-Argüelles et al. [27]	46	14 (0–86)	24 (0–102)	2	0
Cuellar-Ambrosi et al. [28]	47	13 (10–17)	15 (14–20)	NR	0
Mabed et al. [29]	28	13 (7–18)	15 (7–20)	0	0
Mabed et al. [30]	32	12 (8–17)	14 (7–19)	0	0
Lopez-Otero et al. [31]	26	27 (0–53)	37 (0–73)	9.6	0
Ramzi et al. [32]	45	11 (8–18)	14 (11–29)	2.2	0
Ramzi et al. [33]	38	11 (9–21)	13 (10–31)	0	0
Bekadja et al. [34]	54	10 (6–17)	13 (9–24)	0	0
Kayal et al. [35]	92	10 (8–27)	14 (9–38)	3.2	0
Bekadja et al. (2015)	45	11 (8–12)	13 (10–24)	3	0
Bekadja et al. [36]	240	10 (6–17)	13 (9–24)	1.3	0
Sarmiento et al. [37]	42	9 (9–16)	11 (10–19)	0.5	0
Karduss-Urueta et al. [38]	359	13 (9–39)	16 (7–83)	NR	0
Naithani et al. [39]	76	12 (9–35)	13 (9–65)	5.3	1.7
Kulkarni et al. [40]	224	12 (9–22)	17 (10–44)	3.1	0.44
Bittencourt et al. [41]	45	10	NR	2	0
Jennane et al. [42]	55	12 (7–19)	14 (9–32)	3.6	0
Piriyakhuntorn et al. [43]	26	12 (10–19)	14 (10–23)	3.8	0
Bekadja et al. [44]	94	14 (12–32)	17 (15–28)	9	0

N number of patients, *TRM* transplant-related mortality, *NR* not reported.

Mobilization, storage, and target dose of CD34+ cells

There is no established optimal dose of CD34+ cells necessary for hematopoietic reconstitution. Yet, a minimum of $2.0–3.0 \times 10^6$ CD34+ cells/kg is recommended. A mean CD34+ cell dose of 3.85×10^6/kg was achieved with this approach, hence exceeding the minimum requirement. Storage in a refrigerator at a temperature of +4 °C showed a minimum viability of 75% with storage up to six days and a viability above 90% when the storage is within two days or less. Based on these results, the non-cryopreserved approach does not appear

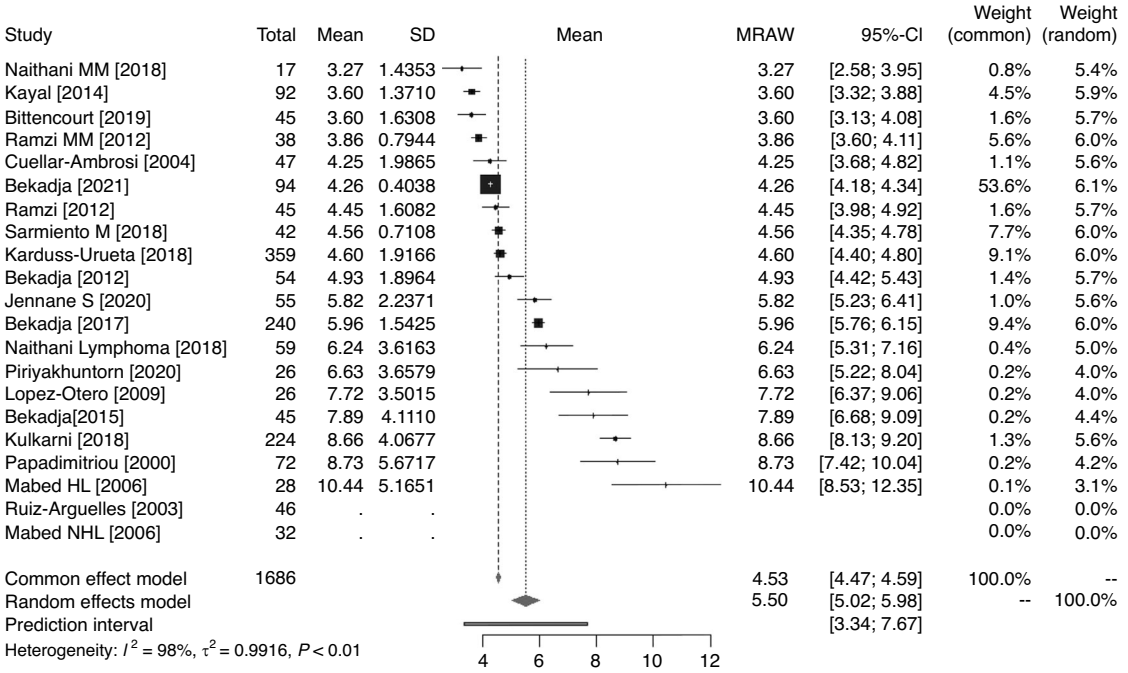

Fig. 3 Pooled mean estimates of ANC. MRAW—represents the untransformed means along with confidence intervals; common and random shows the weightage given to each study in pooling of summary result from each study.

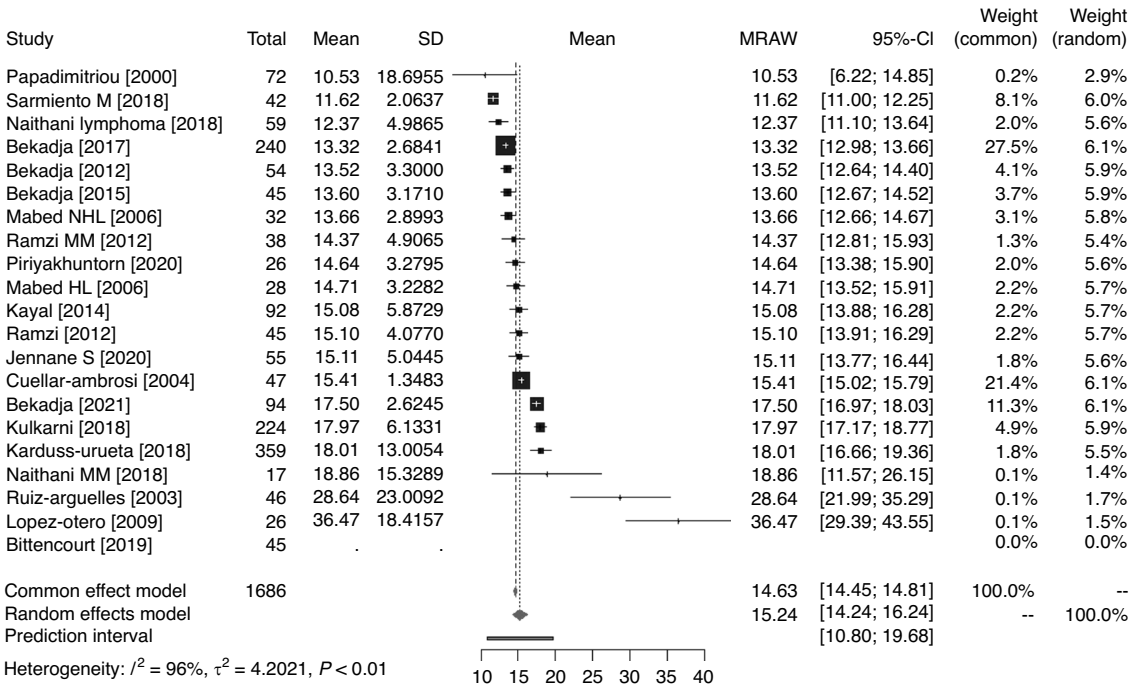

Fig. 4 Pooled mean estimates of platelets recovery. MRAW—represents the untransformed means along with confidence intervals; common and random shows the weightage given to each study in pooling of summary result from each study.

to jeopardize the cell dose, however we recommend targeting a higher CD34+ collection when planning longer storage for conditioning regimens that need more than two days to deliver.

Conditioning regimen in relation to CD34 cell dose

As the standard conditioning regimen for MM is high-dose melphalan on day −1, storage of PBSCs at +4 °C for only 24–48 h does not appear to significantly affect cell viability. As a result, we believe that a minimal target dose of CD34+cells such as 2×10^6/kg could be sufficient to ensure engraftment. For lymphoma patients, however, conditioning regimens are delivered over 3 to 6 days, hence, intuitively a higher number of CD34+ cells (4×10^6/kg) should be collected to account for potential viability loss expected with prolonged storage at +4 °C. Programs can adopt or tailor a conditioning regimen that serves the purpose according to local needs and logistics.

Safety of non-cryopreserved cells

Existing data on 1641 performed non-cryopreserved autografts showed a mortality rate comparable to that of autologous transplants using cryopreserved stem cells. As a result, there does not appear to be a need for extra-safety precautions in the non-cryopreserved setting.

Engraftment kinetics

Data derived from 1641 non-cryopreserved autografts showed comparable median times to neutrophil and platelet engraftments compared to autografts using cryopreserved PBSCs for both MM and lymphoma [50] Only two cases of GF were reported.

CONCLUSION

Significant cost reduction represents a clear advantage of non-cryopreserved transplants. This is relevant in countries with limited resources. Non-cryopreservation circumvents issues related to storage capacity and potential complexities associated with discarding unused cryopreserved stem cells. Another potential benefit of non-cryopreservation is reducing the time between the last induction or salvage chemotherapy regimen and the start of high-dose therapy, especially in patients at high(er) risk of relapse. Moreover, the application of non-cryopreserved autologous PBSCs would have a significant impact on expanding the number of centers that may offer high-dose therapy to patients in need of this treatment modality. This is especially relevant in countries with limited resources. Distance traveled to and from the transplant center can be a significant burden in some of these countries and so performing the entire transplant in one step can be an important consideration for increasing access to transplants and for overall compliance.

Nevertheless, non-cryopreserved transplants have some unique challenges. For instance, "fresh" cell autologous transplants require a vigorous logistical coordination effort to achieve all required steps in a timely manner. Also, some conditioning regimens for lymphoma require administration over several days posing a challenge with prolonged stem cell storage. This limitation is further compounded if the stem cell infusion needs to be postponed for any clinical reason during or after the conditioning regimen has been initiated. Another limitation of non-cryopreserved stem cells is the inability to consider the option of a second or tandem autologous transplant in MM patients; however, emergence of novel anti-myeloma therapies is obviating the need for a second or tandem transplant in most cases.

We acknowledge several inherent limitations to this work, related to use of retrospective/observational data with no data from prospective randomized controlled trials. Yet, performing autologous PBSC transplants without cryopreservation is possible and appears safe and it is certainly affordable. This practice will make it possible for countries with limited resources to build their transplant capacities, hence offering these procedures to a larger population in need of this treatment. Ideally, prospective controlled studies are desirable to validate this approach for wider applicability.

DATA AVAILABILITY

All data generated or analyzed during this study are included in this published article.

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AUTHOR CONTRIBUTIONS

Study concept: MAB, MAK-D, REF, MA. Study design: MAB, MAK-D, REF, AK, TE, MA. Data collection: TE. Statistical analysis: TE. Interpretation of results: MAB, DN, MAK-D, REF, LG, IY-A, HG, DJW, SG, SOA, CC, SKH, AR, UG, AB, NH, AA, MP, AH, JS, YK, AK, TE, DMc, NW, RG, MM, YA, MK, AS, DR, MA, WR. Manuscript writing: MAB, DN, MAK-D, REF, LG, IY-A, HG, DJW, SG, SOA, CC, SKH, AR, UG, AB, NH, AA, MP, AH, JS, YK, AK, TE, DMc, NW, RG, MM, YA, MK, AS, DR, MA, WR.

COMPETING INTERESTS

MAB, DN, REF, DJW, SG, AR, AB, AH, YK, AK, TE, MM, WR declare no conflicts of interest MAK-D: declares research/grant from Bristol Myers Squibb, Novartis, and Pharmacyclics, and lecture/honoraria from Kite Pharma; LG: declares relationship with Bristol Myers Squibb, Sanofi, Janssen, Pfizer; IY-A: declares honoraria from Kite Pharma, Novartis and Bristol Myers Squibb; HG: declares speaker bureau and consultancy for Therakos, Gilead, Novartis, Stemline, Neovii, Sanofi, and Takeda; SOA: declares advisory board with Kite Pharma and Novartis, and speaker honoraria from Kite Pharma, Novartis, and Johnson & Johnson; CC: declares honoraria (personal and institutional) for lectures and advisory boards from Bristol Myers Squibb, Kite Pharma/Gilead, Janssen, Jazz, Novartis, and Miltenyi Biotec; SKH: declares educational/travel grants from Novartis, Pfizer, Janssen, Therakos, Vertex, MSD, Roche; UG: declares consultancy/Honoraria :Kite Pharma, Incyte, Astellas, Jazz,and Vor; NH: honoria from Janssen, Novartis, Takeda, Abbvie, Roche, Astellas, Bigene; AA: declares lecture-Advisor /honoraria from Kite Pharma, Novartis, Takeda and Janssen; MP: declares research with Bristol Myers Squibb, Janssen, Kite Pharma, Novartis, and consultancy for Bristol Myers Squibb, Novartis, and honoraria from Gilead; JS: declares consultancy for Sanofi and ADRx, honoraria from Sanofi, Alexion, AstraZeneca Rare Disease, Prevail Therapeutics (Eli–Lilly), Pfizer, Sobi Pharmaceuticals, and Novartis, advisory committees for Sanofi, AstraZeneca Rare Disease, Prevail Therapeutics (Eli–Lilly), Pfizer, Sobi Pharmaceuticals, Novartis, speaker bureau for Sanofi, AstraZeneca Rare Disease, Prevail Therapeutics (Eli–Lilly), Pfizer, Sobi Pharmaceuticals, Novartis; DMc: declares lecture/honoraria from GSK, Novartis, Abbvie. Research funding from Imago Biosciences; NW: declares speakers fees from BMS Celgene, Kite Gilead, Novartis, Pierre Fabre, Sanofi Genzyme, Therakos Mallinckrodt, Travel reimbursement from Jannsen, Pierre Fabre; RG: declares speaking honoraria from Biotest, Pfizer, Medac, Neovii and Magenta; YA: declares lecture/honoraria from Otsuka Pharmaceutical Co., Ltd, Chugai Pharmaceutical Co., Ltd., Novartis Pharma KK, Meiji Seika Pharma Co., Ltd, Janssen Pharmaceutical K.K., and consultancy fee from JCR Pharmaceuticals Co., Ltd and Kyowa Kirin Co., Ltd; MK: declares non-specified relationship with Kite Pharma, Takeda and Gilead; AS: declares honoraria from Takeda, Bristol Myers Squibb /Celgene, MSD, Janssen, Amgen, Novartis, Gilead Kite, Sanofi, Roche, Genmab, AbbVie, Jazz Pharmaceuticals, consultancy from Takeda, Bristol Myers Squibb/Celgene, Novartis, Janssen, Gilead, Sanofi, Genmab, AbbVie, speaker bureau for Takeda and Research support from Takeda; MA: declares lecture/honoraria from Kite Pharma and Vertex Pharma.

ADDITIONAL INFORMATION

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