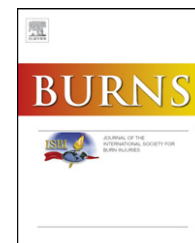


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Preservation methods of allografts and their (lack of) influence on clinical results in partial thickness burns

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ABSTRACT

Allografts, cadaver skin and amnion membrane are considered the golden standard in the management of partial thickness burns. However, debate on whether the tissue needs to be viable is on-going, since many believe that viable grafts result in better healing.

The objective of this literature survey was to analyse the evidence on the method of preservation of allografts (cadaver skin or amnion membrane, glycerol, cryopreservation, lyophilisation) having a clinical impact on the healing of partial thickness burns. The survey focussed on preservation techniques and clinical outcomes (reepithelialisation) in partial thickness burns, as well as on differences in viability, immunogenicity and antimicrobial properties of the preservation methods.

Most studies on allograft treatment of partial thickness burns are observational, with only one study of a (historical) comparative nature. A true meta-analysis was not performed and the results of this survey are observational in nature as well: they indicate that there is no evidence that viability of the graft influences healing outcomes. Thus, instead of viability, other aspects, such as intrinsic antimicrobial safety of the preservation method and cost should be the primary criteria for the choice of preservation method to be used for allografts.

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1. Introduction

Allografts, also called homografts, are tissues or organs transplanted from a donor of the same species but of a different genetic constitution. In wound care in general, and burn care in particular, the primary types of allografts used are amnion membrane and cadaver skin.

With initial routine use dating as far back as the 1950s [1–5] the use of allografts is still a mainstay in the treatment of burns [6,7].

The main indication for allografts is partial thickness burns [8,9] where they are known to promote reepithelialisation [10,11] and pain relief [12–15]. Human allografts are also widely used for wound bed preparation [16,17] after excision of deep dermal or full thickness burns and as an overlay over

autografts in the sandwich or intermingled techniques [18,19]. Although less commonly used than in burn care, allografts are also part of the armamentarium utilized in non-thermal trauma [20] and skin ulcer care [5,21].

To assure reliable availability allografts are often stored in tissue banks [22]. Most commonly, glycerol and cryopreservation are used as storage and preservation methods. Both techniques have their own advantages and disadvantages but an essential difference between cryopreservation and 85% glycerol preservation is the level of viability of the preserved tissues [23–25]: glycerol preservation preserves the morphology of the cells but they are non-viable, whereas cryopreservation allows for a certain level of viability after the tissues are thawed.

Secondary analysis of the results of two surveys, conducted with 9 years separating them, on the type of allografts used in

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burn care indicates that cryopreservation techniques are primarily used in the United States, while most Western European burn centres prefer glycerol preservation [8,9]. In many discussions with clinicians we largely have observed the same dichotomy. The “rest of the world” does not seem to have such a clear preference.

Those who prefer viable cells often state that the growth factors and other compounds delivered from these cells into the wound lead to superior clinical performance. Using the hypothesis that increased viability is reflected in better clinical performance, we have undertaken a review of the literature to analyse if any evidence exists that this hypothesis is, indeed, valid. We also looked at other aspects of preservation methods, such as antimicrobial and inflammatory properties that have the potential to contribute positively or negatively to healing results.

2. Methods

An extensive literature search was initiated, primarily on whether different preservation techniques used for amnion and cadaver skin lead to different clinical outcomes, with reepithelialisation speed, percentage of healing and long term results as the primary criteria.

We also searched for data on secondary aspects of preservation techniques which may have an influence on the primary outcomes, such as viability and immunogenicity of the tissues, antimicrobial properties and the potential of allografts to “sterilise a wound”.

Search criteria used for online resources (i.e. PubMed) included, but were not limited to, homograft, allograft, donor skin, cadaver skin, amnion, amniotic membrane, burns, partial thickness, 2nd degree, mid dermal, deep dermal, cryopreserved, cryopreservation, deep frozen, nitrogen, glycerol, glycerolised, epithelialisation, reepithelialisation and healing.

We only analysed data on partial thickness burns and excluded cultured epithelial grafts since their physical and biological functions are different from human tissues (lack of dermis, for example). For similar reasons xenografts and biosynthetic or synthetic materials were excluded as well.

The use of allografts in full thickness burns (as wound bed preparation after excision or as biological dressings over autografts) was not analysed: for this indication too many additional variables (i.e. timing of excision, type of excision) contribute to the success or failure of the procedures.

Since the use of amnion membrane and cadaver skin is rare in ulcer care and since the aetiology of skin ulcers is very diverse, we did not look at the performance of the biological materials in these indications either.

An initial survey did not identify any prospective randomized controlled studies. Therefore, we changed our criteria and included any article in which clinical results on partial thickness burns, treated with allografts, were presented and where the study population had a minimum size of 5 patients. Because of our interest in viability, immunogenicity and antimicrobial aspects of the preservation techniques we also searched for, and included any articles on preclinical and clinical results in which these topics were discussed.

2.1. Harvesting and preservation techniques

Skin allografts most commonly are harvested from cadavers but may also be obtained from living donors, i.e. from an abdominoplasty or mammoplasty [26].

Skin donor sites are prepared with one or more topical antimicrobial solutions [27]. Amnion membranes are cleaned and washed extensively in similar solutions and/or with antiseptics such as sodium hypochlorite [28]. Several serological and skin samples are taken from the donor and analysed for the presence of bacterial and viral content [29]. Usually, the allografts are incubated with antibiotics prior to preservation, although some centres also use fresh allografts.

The two main ways of preservation and storage are cryopreservation in liquid nitrogen or glycerol preservation, although lyophilisation also has been used [30].

Details of cryopreservation differ [31–33] but all methods use a controlled freezing process with compounds such as dimethylsulfoxide Me(2)SO [34] (DMSO) or glycerol [35] as a cryoprotectant. Cryopreservation is sometimes combined with radiation [36,37]. In excised murine wounds with primary take as criterion for clinical efficacy cryopreserved human cadaveric skin (CPA), showed that performance decreased not significantly for up to 5 years of storage when compared to fresh skin [38].

Glycerol preservation uses rinsing with glycerol solutions in concentrations increasing from 50 to 85%. For each concentration the cadaver skin is agitated at 33 °C for 3 h. Glycerolised allografts (GPA) are then stored at 2–8 °C for a minimum of 3 weeks: bacterial killing increases with exposure time [39]. At the EuroTissue bank,¹ the primary provider of GPA, trimmings (the by-product of cutting the pieces of skin to size after glycerolisation is complete) are separately incubated and bacteriologically tested at set intervals: results of the cultures of the trimmings are used to determine the level of bacterial kill in the main product, and, consequently, its readiness for release for clinical usage. GPA storage is limited by the pharmacopeia guidelines for glycerol and set at a maximum of 2 years.

Glycerolised or cryopreserved allografts are available in full sheet as well as in meshed formats [39–41]. Generally, glycerol preservation is considered more cost effective than cryopreservation since the method itself, but particularly also the storing facilities (i.e. household refrigerators) are simple and relatively low-cost [42].

Preservation of amnion membrane is essentially done in the same two ways as human cadaver skin, either using the glycerol or the cryologic technique.

2.2. Viability and morphology of grafts

Viability is considered important by many since cells with a higher level of viability are assumed to deliver more “beneficial growth hormones and cytokines into the healing wound.” Thus, a great deal of research has gone into assessing the influence of preservation methods and different cryoprotectants on tissue viability.

In general, for cryopreserved cadaver skin the method of thawing does not influence viability of the skin [43] but the

¹ Beverwijk, The Netherlands.

type of preservation does [44], as does increasing the age of the donor [45].

For non-frozen skin, the viability of human split skin grafts was shown to be influenced by the storage solution [23]. Using tetrazolium reduction and oxygen consumption assays [25] it was shown that human skin cryopreserved with DMSO retained significantly higher viability than GPA [34] and similar results were shown in a murine experiment [24].

In GPA, the high concentration (85%) of glycerol replaces virtually all the intracellular water: this helps avoiding degradation of the skin during storage [46]. Thus, the cells are dead but the structural integrity of the skin is preserved [39,47].

Although no specific information was available, it is likely that lyophilised cadaver skin [30] is not viable.

2.3. Antimicrobial properties of storage method and of the allografts

Various biological dressings, such as human fresh and cadaver skin grafts, have intrinsic antimicrobial properties, albeit it to a different degree [48]: they help reducing the bacterial load of the recipient site although, when used as wound bed preparation prior to skin grafting, the recipient site is not always completely free from microbial contamination [49]. In vitro studies indicate that, amongst other factors, antimicrobial effects depend on whether the grafts are fresh, frozen, or irradiated, while the preservation medium also plays a role [50].

Cryopreserved allografts may have the potential to act as a bacterial [51] or viral [52,53] vector from the donor to the recipient [54]. Suspected transmission of HIV to the recipient [52] and cytomegalo-seroconversion [55] have been reported.

Several studies indicate that significant percentages (ranging from 4.9% to 19% [29,56,57]) of cryopreserved allografts have to be discarded upon finding positive cultures and/or serology, either from the donor or after initial antimicrobial treatment of the graft itself. The percentages depend on the type of donor preparation and the type of bacteriological and viral testing done [27].

Preservation and storage of cadaver skin in 85% glycerol has very strong antimicrobial effects. The percentage of glycerol, the temperature, and the exposure time are of influence on the sterilisation process. In one study, 10.1 + 4.1% of the cadaver skin showed initial bacterial contamination, but after prolonged storage in glycerol all skin samples eventually showed no bacterial growth [57]. After incubation with glycerol 85% of the mean survival time of *P. aeruginosa* strains in glycerol 85% at 24 °C was 2.6 days, 14.7 days for different *Staphylococcus* species and 29.6 days for three vegetative *Bacillus* species [58]. Glycerol 85% also has been used to resterilise cryopreserved allografts which, upon thawing, showed positive cultures [59].

In addition to its antibacterial effects, glycerol 85% also has strong virucidal effects as shown with tests with herpes simplex virus 1 and polio virus: similar to the situation with bacteria, the effect is related to concentration and exposure time [60,61]. Other experiments show a strong virucidal influence on HIV [62]. Thus, according to some, the risk of HIV transmission is not a drawback anymore for the use of glycerolised skin [63]: indeed, glycerol preservation, but not cryopreservation can inactivate both intracellular and extra-cellular HIV-1 [62].

2.4. Immunogenicity

Immunogenicity and, consequently, the type of rejection reaction by the recipient are related to the viability of an allograft.

Essentially because they are dead, 85% glycerol preserved grafts elicit a less dramatic and slower response in the recipient than GPA [46]: experiments in a full thickness porcine wound model showed that rejection of glycerol treated allogeneic skin grafts was delayed for up to 6 days. Viable, untreated allogeneic skin grafts were rejected predominantly by CD8 positive T-cells whereas in the 85% glycerol treated grafts the influx of host cells was lower and the majority of the cells were macrophages: this process is less disturbing for the outgrowth of autologous cells in sandwich grafting [64]. Additional research suggests that after transplantation of glycerol preserved skin an inflammatory process mediated by infiltrating host monocytes occurs, rather than a rejection process mediated by T-cells [46].

However, the clinical observations that the glycerolisation procedure results in decreased immunogenicity of donor skin was not supported in a mixed lymphocyte culture test in a rat model in which vital allografts were compared to GPA [65].

2.5. Clinical outcomes

Cost effectiveness in wound care is becoming an important outcome and amnion membrane as well as cadaver skin are reported to be cost effective, particularly when compared to synthetic dressings [66]. Particularly, a reduced number of required dressing changes [67] (when compared to antimicrobial creams) and a reduction in length of stay [68] may contribute to lower costs of care.

Healing outcomes may be defined in different ways: the most common criteria used are the percentage of reepithelialisation within a certain time frame or the time to complete reepithelialisation. Other outcomes used are the percentage of patients that, after treatment with an allograft, have to undergo secondary (excision and) grafting, the percentage of patients that develop hypertrophic scarring, or the length of stay for a given cohort of patients.

Unfortunately, we found little consistency in published healing outcomes and the way they are reported. Even the depth of the burns and/or their location is sometimes missing from publications.

In all studies in which allografts were compared to antimicrobial creams, the allograft, whether amnion (i.e. compared to Furacine [69]) or cadaver skin (i.e. compared to silver sulfadiazine [70,71]), performed better. However, we were not able to find one single randomized controlled study in which different types of allografts were prospectively

Table 1 – Accumulative number of burns per allograft category.

Glycerol preserved cadaver skin	247
Cryopreserved cadaver skin	161
Lyophilised cadaver skin	25
Amnion	263
Total	696

Table 2 – Assumed or documented superiority of preservation techniques (X indicates the superior technique).

	Cryopreservation	Glycerol preservation	Comment
Harvesting technique			Similar
Preparation prior to preservation (i.e. antibiotics treatment)			Similar
Viability of cells	X		
Inherent antimicrobial property		X	
Immunogenic response		X	Literature somewhat conflicting
Average time to healing			Similar
Average percentage of burns healed within set time frame			Similar: literature documents different time frames
Cost of preservation		X	Glycerol technique presumed superior because of simpler technique and equipment

compared to each other. Given the often expressed opinion that allografts are the standard of care, the number of published, randomized, clinical trials with any type of allograft is actually quite low.

The total number of partial thickness burns, enrolled in published clinical trials, that were treated with GPA, CPA, lyophilised cadaver skin, and amnion membrane is 247, 161, 25 and 263 respectively (Table 1). The documented or presumed superiority of the preservation techniques is presented in Table 2 and a summary of results of the management of partial thickness burns with different types of allografts is presented in Table 3.

2.6. Glycerol preserved cadaver skin

In one historical-control study, 106 patients with partial thickness burns were treated with CPA and 57 with GPA: the GPA group fared considerably better with regard to the number of necessary secondary grafting procedures (26.3% versus 39.6% respectively) [72].

Vloemans et al. describes an average time to 95% reepithelialisation with GPA of superficial, mixed and deep partial burns of 8.5 days in a study where GPA was compared to a synthetic dressing (N = 40) [73]. Horch compared silver sulfadiazine treatment with GPA treatment in patients with superficial and deep partial thickness burn of the face (N = 5 in both groups) and found an average reepithelialisation time of 10.5 days for GPA, with a significantly improved scar outcome ($p < 0.05$) for the GPA treated burns as well [71].

Hermans, in a non-comparative trial, reports an average healing time of 11.7 days in 57 patients with superficial and deep partial thickness burns, primarily of the arm and the thorax [74]: all patients were treated with GPA.

Brans et al. analysed the long term outcome (2–5 years post burn) retrospectively of 45 children whose partial thickness burns were treated with GPA [75]. In 21 patients (47%), the wounds healed spontaneously and in 24 patients remaining defects were closed by a split skin autograft in the third week post burn. The author reported healing without scar formation in 53%, with moderate scars in 21% and with severe scar formation in 26% of all patients.

Peeters et al. state in a published discussion that the incidence of necessary grafting is approximately 31% with the use of cadaver allografts, versus an estimated 50% prior to the

introduction of allografts in their respective clinics discussion [76]. Khoo et al. describes an average healing time of 19 days in his patients with partial thickness burns, treated with GPA (N = 43) [16].

2.7. Cryopreserved cadaver skin

Rose et al. report an average healing time of 19 days in a group of 27 young patients with partial thickness burns, treated with CPA [12]. Eldad et al. compared 12 deep partial thickness flame burns, treated with CPA, with similar burns in the same patients, treated with silver sulfadiazine: he reports a healing percentage of 76, with good cosmetic results within 3 weeks post burn for the cryopreserved treated patients versus 40% healing for the silver sulfadiazine wounds in the same patients [70]. In both studies many different anatomical locations were included.

13 patients with large (>40% TBSA) partial thickness burns were treated with debridement and silver sulfadiazine and compared to 16 patients with similar burns treated with debridement and fresh or cryopreserved allografts. While the authors do not report specific reepithelialisation time, allograft treatment significantly decreased the length of stay [68].

2.8. Lyophilised cadaver skin

In a trial in which 25 patients with partial thickness scalds were treated with lyophilised cadaver skin, 15 (60%) showed complete reepithelialisation on PBD 13 [77].

2.9. Amnion

The healing of burns in a porcine model showed no difference amongst fresh human, fresh bovine and acellular amnion. Wound cultures in the control groups in this study (polyurethane foams) showed a higher level of contamination [78].

Singh et al. report the results of two groups of patients (N = 25 for each group) in which gamma radiated glycerolised amnion membrane was compared with non-radiated glycerol preserved amnion. The burns were mostly located on the face and thorax and for both groups the average healing time was 10–14 days [79].

Branski et al. [67] have compared patients with partial-thickness burns of the face, neck and head, treated with amnion, either disinfected but fresh or cryopreserved (N = 53),

Table 3 – Healing results with different types of cadaver skin and amnion membrane in partial thickness burns.

Primary author	Publication	Type of cadaver skin	Number of burns	Indication: depth of burn	Location	Outcomes/comments
Hermans [74]	Burns (1989)	GPA	57	Partial thickness	Primarily arm and thorax	Average reepithelialisation time: 11.7 days
Brans et al. [75]	Burns (1994)	GPA	45	Superficial and deep partial thickness	Primarily upper thorax and upper limbs	47% complete reepithelialisation within 14 days
Peeters et al. [76]	Burns (1994)	Allograft in general				Significant reduction in number of required/indicated secondary grafting after GPA instituted, from >50 to 31% (note: transcript of discussion)
Vloemans et al. [72]	Burns (2002)	GPA	57	Superficial and deep partial thickness	Miscellaneous	26% of burns requiring secondary grafting (not healed on PBD 14)
Vloemans et al. [73]	Burns (2003)	GPA	40	Superficial, mixed and deep partial thickness	Miscellaneous	68% spontaneous complete reepithelialisation within 14 days. 15% late excision and grafting
Horch et al. [71]	Burns (2005)	Early debridement and GPA	5	Superficial and deep partial thickness	Face	Average reepithelialisation time: 10.5 days
Khoo et al. [16]	Burns (2010)	GPA	43	Partial thickness	Not reported	Average reepithelialisation time: 19 days
Rose et al. [12]	JBCR (1997)	CPA	27	Partial thickness	Miscellaneous	Average reepithelialisation time: 19 days
Eldad et al. [70]	Burns (1997)	CPA	12	Deep partial thickness	Miscellaneous	76% reepithelialisation within 21 days post burn
Vloemans et al. [72]	Burns (2002)	CPA	106	Superficial and deep partial thickness	Miscellaneous	39.6% of burns requiring secondary grafting (not healed on PBD 14)
Naoum et al. [68]	Burns (2004)	Debridement and CPA or fresh allograft	16	Partial thickness burns	Miscellaneous	Significant decrease in length of stay
Liecht et al. [77]	Burns (1989)	Lyophilised cadaver skin	25	Partial thickness burns	Miscellaneous	60% complete reepithelialisation on PBD 14.
Sawhney [80]	Burns (1989)	Fresh	15	Superficial partial thickness	Miscellaneous	Average reepithelialisation time: 9.3 days
			15	Intermediate partial thickness		Average reepithelialisation time: 15.7 days
			15	Deep dermal		Average reepithelialisation time: 27.5 days
Lorrusso et al. [81]	Annals of the Mediterranean Burn Club (1989)	Cryopre-served	11	Partial thickness	Miscellaneous	Average reepithelialisation time: 10.7 days
Ravishanker et al. [15]	Burns (2004)	Glycerolised	71	Superficial partial thickness	Miscellaneous, face excluded	Average reepithelialisation time: 7–10 days
Singh [87]	Burns (2007)	Glycerolised	25	Partial thickness	Mostly face and thorax	Average reepithelialisation time: 10–14 days
		Glycerolised and radiated	25	Partial thickness	Mostly face and thorax	Average reepithelialisation time: 10–14 days
Branski et al. [67]	Burns (2008)	Fresh or Cryopre-served	53	Partial thickness	Face and neck	Average reepithelialisation time: 6 days
Bujang-Safawi et al. [82]	Burns (2010)	Dried and irradiated	33	Superficial partial thickness	Face	Average reepithelialisation time: 5.4 days

with topical antimicrobials as control ($N = 49$). Healing in both amnion groups was 6 + 2 days versus 8 + 2 days in the control group. Time to healing, length of stay and the development of hypertrophic scarring was not different between the groups. Patients in the amnion group had significantly fewer dressing changes than in the control group ($p < 0.05$).

Superficial partial thickness burns treated with amnion ($N = 15$) reepithelialised on average in 9.3 days versus 12.5 days ($N = 15$) for silver sulfadiazine treatment. For intermediate burns healing time was 15.7 (amnion) and 23.9 (silver sulfadiazine) days ($N = 15$) and for deep dermal burns ($N = 15$) 27.5 and 37.5 days respectively [80]. Lorusso et al. treated superficial partial thickness burns in 11 patients with frozen amnion (and compared this treatment to Biobrane²) and obtained an average healing time of 10.7 days for the amnion treated burns [81].

Dried irradiated human amniotic membrane was used for superficial facial burns in 33 patients, with an average healing time of 5.4 days (range: 2-14 days) [82].

In a group of 71 patients, glycerol preserved amnion was reported to lead to complete healing of superficial partial burns within 7-10 days and in mid dermal burns the same result was obtained within 20 days [15].

3. Discussion and limitations

In total, 17 studies were found on partial thickness burns, treated with different types of allograft, with a total of 696 burns (Table 1).

Given that many consider allograft treatment the "golden standard [10,11,21,71]," the number of published clinical trials is small. Moreover, the methodology of most of the trials was poor and outcomes studied diverse and ranging from days of hospitalization, reepithelialisation percentage and time, percentage of patients that had to undergo secondary grafting of their partial thickness burns, to long term outcome with regard to scarring. In addition, in some articles the authors make a distinction between superficial, mid dermal and deep dermal burns while others group all partial thickness burns together. In some reports only certain anatomical locations are included whereas in others all anatomical areas could be the target of a certain type of treatment.

None of the studies compared the different preservation methods in a prospective, randomized manner and most studies were, in fact, observational. Consequently, the level of evidence according to the Oxford Centre for Evidence Based Medicine [83] ranks 2a at best (1 study, direct, historical comparison of the two conservation techniques) and 3a or lower for most studies.

The lack of scientific evidence also indicates a major limitation of this literature review: the different patient cohort and treatment regimens are not comparable and, consequently, the analysis and conclusions are observational rather than evidence based.

Still, the most frequently reported outcomes are average reepithelialisation time (13 studies) and percentage of complete reepithelialisation within a defined period (4 studies).

When these criteria are used for superficial and mid dermal partial thickness burns, amnion membrane seems to offer the most favourable results, irrespective of the preservation technique used. However, many of the amnion trials primarily or only included the face which heals consistently faster than any other anatomical area. Eliminating this aspect, the actual differences amongst the different types of allografts, whether amnion or cadaver skin, fresh, glycerolised, lyophilised, cryopreserved and/or irradiated, are not significant. In the large majority of publications the reepithelialisation time for partial thickness burns, deep dermal ones excluded, seem to be within the 2-3 weeks' time frame.

With regard to the percentages of burns healed within a defined time frame, two different periods (2 and 3 weeks respectively) were taken as criteria. These datasets are not comparable since different standards are used for secondary intervention (excision and grafting): some clinics do not allow spontaneous healing to continue after 2 weeks, while others extend this period to 3 weeks. In addition, it can be argued that burns that take 3 weeks to heal spontaneously were not entirely superficial or mid dermal partial thickness in the first place: it is likely that this type of lesion contained at least some deep dermal or full thickness patches or that secondary deepening has occurred [84-86].

The number of analyses on long term results and on required percentages of secondary interventions is too small to draw any conclusions on outcomes differences amongst the different types of allograft.

Viability and immunogenicity levels were not shown to have any influence on the clinical performance of the allografts. Therefore, these preservation-dependent properties should not be the primary drivers for choosing a specific type of allograft.

Other arguments, such as the superior intrinsic antimicrobial properties of glycerol preservation should drive the choice of preservation technique. In addition, although no comparative data were found in the literature, it is likely that glycerol preservation is less expensive since simple equipment (a household refrigerator) is used for storage. Consequently, the cost involved with preservation technique should be a driver of choice as well.

4. Conclusion

The literature on allografts and clinical outcomes is of poor quality. The data collected in the studies are too diverse to allow for a true scientific comparison or statistical analysis. This is particularly surprising because of the existing convictions about superiority of one preservation technique over another. It is also because of these strong convictions that we felt publishing this overview was worthwhile, although we realize that the analysis of the literature itself does not follow all the guidelines provided by the Cochran Collaboration or similar organisations.

The type of preservation influences the level of immunogenicity, viability, and intrinsic antimicrobial properties of allografts, both cadaver skin and amnion membrane. Many assume that a higher level of viability is an important advantage of cryopreservation since, supposedly, this con-

² UDL Laboratories, Rockford, IL, USA.

tributes to better healing. This literature review does not provide evidence for this assumption.

Thus, rather than viability, antimicrobial safety and cost should be the primary driver for determining which type of preserved allograft to use for the treatment of partial thickness burns. Everything else being equal, these arguments seem to favour glycerol preservation over cryopreservation.

Conflict of interest

None declared.

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