Characterization of Skin Allograft Use in Thermal Injury

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This study provides objective data on the practice of allograft usage in severely burned patients. Furthermore, gaps in our knowledge are identified, and areas for further research are delineated. Using an institutional review board–approved protocol, active duty military patients injured while deployed in support of overseas contingency operations and treated at our burn center between March 2003 and December 2010 were identified. Their electronic medical records were reviewed for allograft use, TBSA burned, injury severity score, anatomic distribution of burns, operative burden, length of stay, transfusions, and outcome. Among 844 patients, 112 (13.3%) received allograft and 732 (86.7%) did not. The amount of allograft used per patient varied and was not normally distributed (median, 23.5; interquartile range, 69.5). Patients received allograft skin an average of 12.75 times during their admission. Allografted patients sustained severe burns (μ, 53.8% TBSA); most were transfused (71.2%) and grafted frequently, averaging every 7.45 days. Most commonly, allograft was placed on the extremities (66.5%) followed by the trunk (44.2%); however, the vast majority of allografted patients also had concomitant burns of the head (91.1%) and hands (87.5%). All-cause mortality among the allografted patients was 19.1%. In conclusion, allograft is commonly used in the surgical treatment of severe burns. Although there are no anatomic limitations to allograft placement, there are distinct patterns of use. Given the role of allograft in the acute management of large burns, there is need for further investigation of its effect on mortality, morbidity, and antigenicity. (J Burn Care Res 2013;34:168–175)

Although autograft is the optimal therapy for deep burns that require excision, there exists a population in whom autografting is impossible, often related to a large wound burden. These patients frequently benefit from the use of skin allotransplantation and particularly from the use of cadaveric skin allograft. Pollock's[1] original 1871 description of his—the first—experimental allograft for a burn wound detailed the natural course of graft incorporation, improved reepithelialization, and finally a gradual although certain rejection of the graft. This T-cell–mediated rejection is of unknown significance to future formation of percent reactive antibody (PRA) in a population that is a target for vascularized composite tissue allotransplantation (VCA).

Initially, efforts were made to extend the viability of allografts with the goal of permanent incorporation into the host; however, as early as 1952[2] it was recognized that cadaveric allotransplantation was best applied as a temporizing measure to obtain biologic closure, and that rejection was inevitable. More recently, Wendt et al[3] demonstrated that skin allografts have potential for long-term clinical survival in patients receiving conventional immunosuppressive therapy for renal allotransplantation. Clinically, predicted rejection and staged regrafting clearly remains the norm.

As successful procedures for the safe banking of cadaveric skin were developed, initially by the U.S. Navy,[4] the burden of harvesting skin in-house was reduced. This propelled allograft into its current role as the primary alternative to autograft. Today, allograft is widely used to achieve temporary biologic closure in large burns,[5] to optimize wound beds for planned future autografting,[6] as a test of wound bed viability before placing an autograft,[4] and as an adjunct to reconstruction.[7] Thus, with this versatility and availability, allograft has facilitated the advancement of early excision and grafting, which has played an essential role in burn management since Cocoanut Grove in 1942.[8]

Despite its 60-year history as a fixture in the surgical armamentarium, there exists a relative paucity of literature to characterize the use of skin allograft. Several recent articles detailed the indications,[9,10] and one described a novel technique for estimating needs during a mass casualty incident;[11] however, none attempt to describe, in a broad sense, the current practice of skin allograft usage in a busy burn center.

Although cadaveric skin allotransplantation is a long-standing practice, the modern preparations of cadaveric skin allograft have only recently been established. Cryopreserved allograft (CPA) skin, first introduced in 1979, carries the putative benefit of viability. Glycerol-preserved allograft (GPA), in use since the mid-1980s, is nonviable[12] but is inherently antimicrobial and is presumed to be less immunogenic.[13,14] Despite these differences, a thorough review of the current literature by Hermans[15] found that there were essentially no randomized controlled head-to-head trials prospectively comparing CPA with GPA.

In this study, we have described the patients, features, characteristics, and outcomes that define the use of CPA within our burn center and unique patient population.

METHODS

Subjects

All subjects were active duty military patients who received treatment for burn injury at our institution between March 2003 and December 2010.

Data Source

Under an approved protocol, our burn registry and the Joint Theater Trauma Registry were queried for active duty service members who sustained burns, including the demographics, date of injury, length of intensive care unit (ICU) stay, hospital stay, TBSA, the relative distribution of burns, mechanism of injury, and mortality. The hospital's electronic medical record system was used to obtain information about the number of operations, the use of CPA, and the quantity of transfusion.

Statistical Analysis

Basic univariate analyses were used to analyze the resulting data sets. All variables that demonstrated statistical significance $(P \geq .05)$ in univariate analyses or those deemed to be potentially relevant to clinical outcomes were included in the logistic regression model for more in-depth analysis. For bivariate comparisons between cases and controls, two-tailed χ^2 tests were used to test for statistical significance when the expected frequencies were >5 per group. In the small propensity-matched population, the Wilcoxon test was used to determine significance, and in the specific case of TBSA in comparison of frequency of allograft use, a one-way analysis of variance was used. In all other cases, Fisher's exact test was used. Data were analyzed with commercially available statistical software (SAS version 8.1, SAS Institute, Inc., Cary, NC).

RESULTS

Military service members treated in our center were identified by searching the burn registry and electronic medical records. Of the records searched, 844 military patients had data sufficient for our analyses. Of the 844 patients identified, 112 (13.3%) had been treated with CPA during the study period and 732 (86.7%) were managed without the use of CPA. The 112 patients treated with CPA were overwhelmingly male, with a mean age of 26 ± 7.3 years. The injuries suffered were predominantly the result of combat-related blast causing primary flash or secondary flame burns (87%), followed by flame injury not related to blast (7%) and electrical injury (3%); the remaining injuries were due to aircraft mishaps and unknown causes. The distribution of burns in our population is more concentrated with the smaller burns, as expected, but increased CPA usage is seen with increasing TBSA (Figure 1).

Patients who received CPA during their acute hospitalization demonstrated a mean burn size of $53.8 \pm 20.1\%$ TBSA, a mean injury severity score (ISS) of 34.7 ± 14.1, and an all-cause mortality rate of 19.1%. Furthermore, 71.2% of those treated with CPA received transfusions with an average packed red blood cell volume of 11.2 ± 12.2 l. This degree
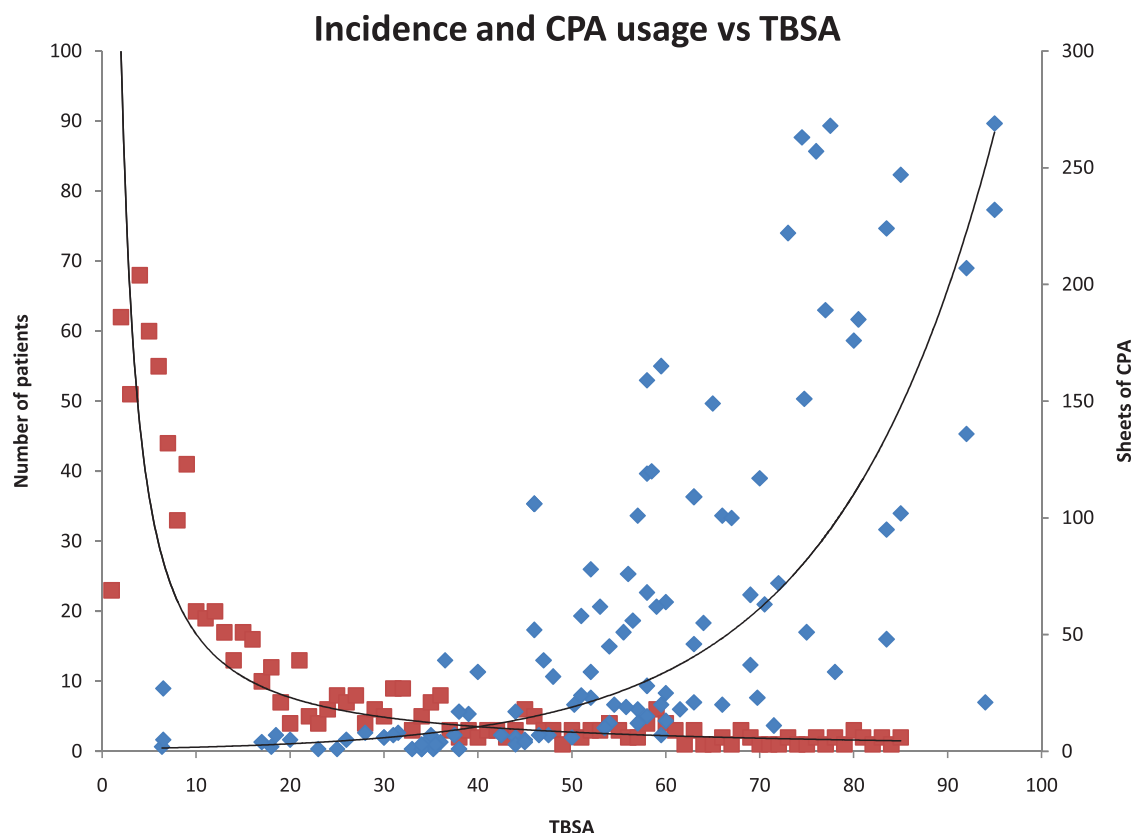
Figure 1. A graph showing the relationship of TBSA to incidence and cryopreserved allograph (CPA) usage. Data fitted with power and exponential linear regressions, respectively. Incidence vs TBSA, $y = 222.39$, $x = 1.122$, $R^2 = 0.80$; CPA usage vs TBSA, $y = 1.015e0.059x$, $R^2 = 0.58$.

of injury resulted in an average ICU length of stay (LOS) of 52 ± 68 days, with 13 (11.6%) patients' ICU LOS exceeding 100 days. During the ICU stay, these patients underwent an operation every 7.5 ± 4.4 days, on average, and underwent an average of 7.3 ± 7 operations during their ICU admission. When examining the hospital stay after discharge from the ICU, the interoperative interval (IOI)

increased to 42.5 ± 45.6 days, with an average LOS of 94.3 ± 88.7 days (Table 1).

The overall frequency of CPA use among our patients was 13.3%. The number of sheets of CPA used per patient was not normally distributed (Figure 2). The median number of CPA sheets used per patient was 23.5, with an interquartile range of 69.5. Patients with 30 to 40% TBSA burns received

Table 1. Clinical features of patients treated with allograft

Variable	No.	Mean ± SD (Range)
Age, years	112	26.11 ± 7.34 (19–54)
TBSA (%)	112	53.78 ± 20.07 (6.37–95)
Injury severity score	112	34.67 ± 14.12 (1–75)
PRBC transfusions (ml)	79	11198.57 ± 12246.40 (300–71557)
Total number of operations	112	12.99 ± 9.89 (1–45)
ICU number of operations	100	7.27 ± 6.95 (1–45)
ICU interoperative interval (days)	85	7.45 ± 4.40 (1–21)
ICU length of stay (days)	112	52.13 ± 68.13 (0–426)
Total length of stay (days)	112	91.56 ± 88.30 (2–502)
Mortality (% died)	106	19.10

PRBC, packed red blood cells; *SD*, standard deviation.

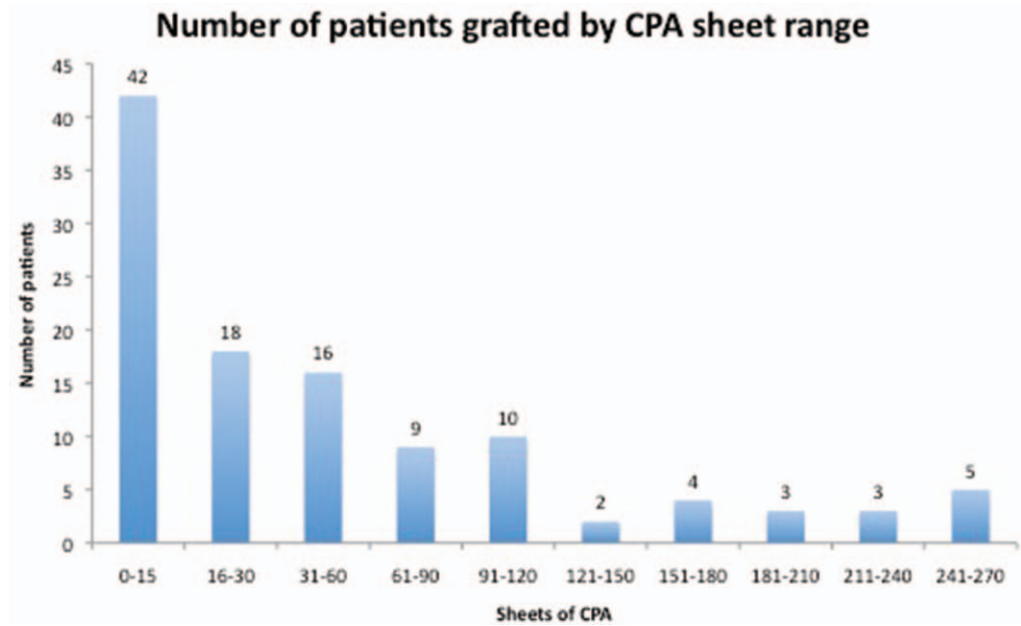## Number of patients grafted by CPA sheet range

Figure 2. A histogram showing the frequency of grafting by amount of cryopreserved allograft (CPA) grafted (median, 23.5; interquartile range, 69.5; μ: 57.2 sheets/patient; σ: 71.7).

CPA almost as frequently as not (49.4%). As TBSA increased, the proportion of patients treated with CPA also increased, as demonstrated by CPA grafting of 91.7% of patients with 50 to 69% TBSA and 92.0% of patients with >70% TBSA ($P < .0001$). In Figure 3, of the five patients who sustained burns of >50% TBSA and did not receive CPA, three died before visiting the operating room for complete excision and grafting, and two survived (57.5 and 53.5% TBSA).

When distribution of burns was considered, we found that among the initial 112 patients treated with CPA there was a high likelihood to have sustained concomitant burns to the head (91.1%) and hands (87.5%). This relationship persisted when 33 additional subjects who originally had been excluded on grounds of insufficient data not related to anatomic distribution were included, demonstrating 127 (87.6%) who sustained head and neck burns and 125 (86.2%) who had burns to the hands. When compared with patients who were not treated with CPA, the pattern of distribution is similar; however, there is a markedly increased prevalence in burns to the genitals (34.5 vs 4.7%) and feet (24.1 vs 5.3%) among those who received CPA (Figure 4).
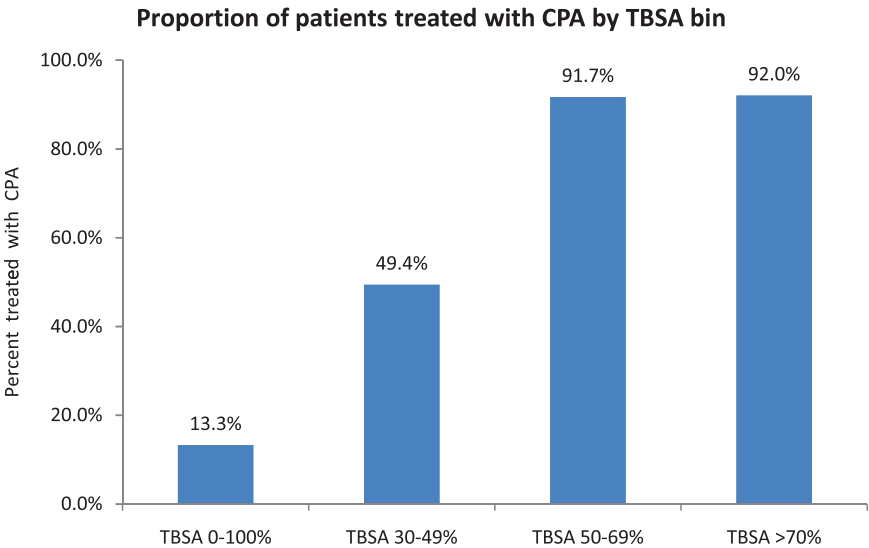
Proportion of patients treated with CPA by TBSA bin

Figure 3. A graph comparing the anatomic location concomitant burns in CPA recipients and non-CPA recipients.

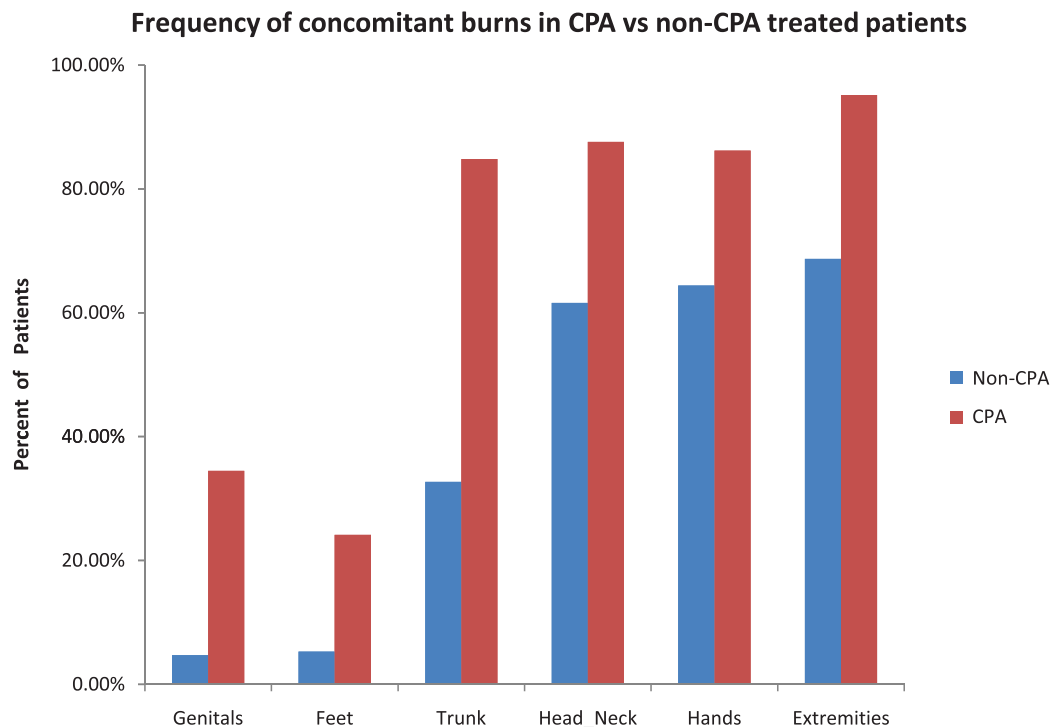**Frequency of concomitant burns in CPA vs non-CPA treated patients**

Figure 4. A graph showing the proportion of patients treated with cryopreserved allograft by TBSA.

Additional analysis was done to determine where CPA is most commonly placed. Of the 988 operative encounters that used CPA, 66.5% involved the placement of CPA on the extremities, and in 44.2% of these operations CPA was placed on the trunk. The head and neck (7.8%) and hands (5.6%) are far less frequently grafted with CPA, and the feet (1.4%) and groin (0.5%) together have CPA placed at <2% of all engraftments (Figure 5).

When propensity matched for TBSA ($N = 72$) and compared across endpoints, patients treated with CPA had more than double the average ICU LOS than those not treated with CPA (30.6 vs 12.9 days; $P < .0001$), longer overall LOS (76 vs 43.3 days; $P = .005$), and twice the operative burden (12.16 vs 6.06 operations; $P = .005$), whereas differences in ISS, ICU IOI, and hospital IOI were not statistically significant. Although the TBSA-matched CPA patients had a lower mortality (8.33 vs 13.79%; $P = .691$) the difference, as in the case of red blood cell transfusion volume ($P = .224$), failed to reach statistical significance (Table 2).

DISCUSSION

CPA skin provides the surgeon with a means to establish biologic, albeit temporary, wound coverage for patients with large burns that exceed the limits of available autograft donor sites. This approach to temporary wound closure reduces insensible fluid and heat loss, thereby partly mitigating the hypermetabolic response that accompanies burn wounds.[16] Furthermore, in a population in which up to 27% of patients who die are killed by a bacterial infection,[17] the attributable decrease in infection[18] secondary to allograft use is clearly advantageous. The advantages of CPA use also extend to fewer dressing changes, pain reduction from decreased donor site surface area and an adherent physical barrier over the burned areas, and improved overall wound healing.[19] These factors, when combined with the induction of revascularization,[6] facilitate closure via both reepithelization in less severe burns and by preparation of the wound bed for delayed autografting in severe burns.

Although our burn center treats both civilian and military burn injuries, this analysis was limited to our active duty service members injured in theater, which increases the presence of burns with superimposed polytrauma far above that found in the conventional civilian burn center. Although isolated flame injury is the most common form of thermal injury in the civilian population, our study has a high number (87%) of patients exposed to combat-related blast and burned as a consequence of quaternary blast effects or who were secondarily burned as a result of involvement in fires after a blast. Despite this, our
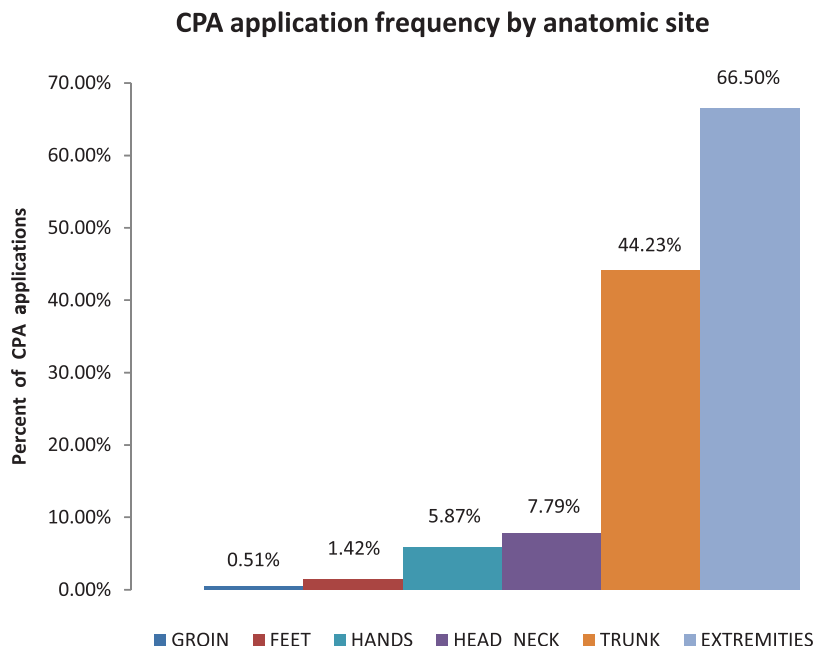
CPA application frequency by anatomic site

Figure 5. A graph showing frequency of cryopreserved allograft (CPA) application to specific anatomic sites by percentage of all CPA applications.

overall study population shows an expected distribution of TBSA, with most (75%) patients treated in our center sustaining burns of <25% TBSA.

Although not confined to the severely burned, the use of CPA increases with TBSA. This relationship is certainly because of limited donor site availability. Thus, CPA often is used to await donor site maturation or the availability of cultured autografts. Given the limited availability of autograft in patients with large burns, it is reasonable to conclude that, when needed, donor autograft may be reserved for functionally and aesthetically privileged areas of the body such as the hands and face. As a consequence, CPA tends to be placed primarily on the trunk and extremities or as an adjunct over widely meshed autograft.

From our data, patients with burns involving more than 30% TBSA demonstrated a greater likelihood that an allograft skin substitute will be needed. This is important because it describes a threshold below which management with autograft alone can be expected and above which the use of skin substitute is common. This relationship is further exaggerated when TBSA reaches 50% and nearly all patients (91.7%) receive CPA. It is interesting to note that, of the five patients with burns >50% TBSA who did not receive CPA, three died before complete excision and grafting could be performed, suggesting that this value may be artificially low.

The current literature supports a time to allograft rejection of between 11 days[5] and 4 weeks[20]; however, our data suggest that in practice, the average

Table 2. Comparison of nonallografted and allografted patients propensity matched on TBSA

Variable	No.	Nonallograft	N	Allograft	P
TBSA	36	34.83 ± 18.74 (0.5–90)	36	35.14 ± 17.01 (6.37–85)	.941
ICU length of stay (days)	36	12.86 ± 13.03 (0–56)	36	30.56 ± 28.31 (0–130)	<.0001
Length of stay (days)	36	43.25 ± 43.95 (1–243)	36	75.97 ± 68.68 (9–367)	.005
Total operations (n)	33	6.06 ± 4.84 (1–19)	36	12.16 ± 9.62 (1–43)	.005
Injury severity score	36	27.44 ± 14.55 (1–75)	36	28.36 ± 12.23 (4–59)	.765
Interoperative interval (days)	33	55.18 ± 70.15 (0–313.75)	36	54.36 ± 54.13 (0–212.86)	.483
ICU interoperative interval (days)	15	4.83 ± 3.32 (1–10.5)	22	5.58 ± 4.15 (1–18)	.539
PRBC transfusions (ml)	21	4745.05 ± 7247.32 (700–33673)	26	6466.65 ± 6683.52 (361–29820)	.224
Mortality (% died)	29	13.79	36	8.33	.691

Values provided as mean ± standard deviation (range).

PRBC, packed red blood cells.

time interval for allograft exchanges or change to autograft is shorter. We found an ICU interoperative interval of 7.5 ± 4.4 days, which indicates that grafting occurs more frequently than previously thought. These data closely approximate the work of Khoo et al,[10] who found a mean duration of GPA adherence of 7.9 ± 2.0 days. Although there exists some weakness in our database caused by nondiscrete text entries that fail to consistently delineate between reoperation for CPA rejection and for poor take, we feel that given the concordance of our data to that of Khoo et al[10] and the wide range of previously published times to rejection, our findings are noteworthy and warrant further investigation.

Anatomically, it is not surprising to find that CPA-treated patients are more likely to suffer burns to the genitalia and feet. These are traditionally well-protected areas that, unless specifically targeted and burned in isolation, are most commonly injured in continuity with burns covering a large BSA. Therefore, it stands to reason that this relationship is maintained by a difference in severity of burns between patients treated with and without CPA than by a pure difference in anatomic prevalence.

Despite this, it is important to recognize that although >30% of CPA-treated patients suffered burns to the genitals, barely more than one half of 1% of all CPAs were applied onto the genitals. We also found that CPA was seldom applied to other functionally or aesthetically sensitive areas such as the feet, hands, and, to a lesser degree, the face. This finding is not unexpected because one of the main functions of CPA is to replace large areas of burn eschar burden. The face and hands often are excised last because the palmar and facial dermis is thick and is given more time to self-epithelialize. In the case of full-thickness loss to the face and hands, however, the burn surgeon should consider the use of CPA for wound bed preparation before the application of a large sheet graft in a patient already deprived of donor skin.

After propensity matching, the persistent differences found in ICU LOS, overall LOS, and operative burden may suggest that the CPA patients suffered deeper burns requiring more extensive surgical management and longer hospitalization. Unfortunately, our data did not delineate the depth of burn and described only TBSA. Despite this shortcoming, the lack of a difference in ISS suggests that the degree of injury was, in fact, similar between groups. Although there is a difference in mortality that favors CPA, this did not reach statistical significance. This indicates that further analysis of TBSA-matched populations, most likely in the 30 to 40% TBSA range, is needed,

but our findings are almost certainly due to a lack of statistical power. The same statement can be made for the requirement of red blood cell transfusion.

Considering that CPA-treated patients often have concomitant burns to the face and hands, questions have been raised about the potential host development of new allo-antibodies from exposure to many CPA donors. This is potentially problematic should these patients become candidates for facial[21] and hand[22] VCA. Given the large concentrations of antigen-presenting cells and the presence of extracellular matrix glycoproteins, CPA and blood transfusions are certainly potential inducers of antigenicity. This is reflected in the finding that epidermal Langerhans cells from the allograft migrate to local host lymph nodes, activating a T-cell–mediated immune response that ultimately results in rejection.[23] Unfortunately, the effect the unique antigenic stimulation caused by CPA has on future PRA and, consequently, the role prior CPA use might play in human leukocyte antigen matching for VCA remains unknown.

Limitations

This retrospective review of allograft usage is subject to all the limitations of a traditional observational study. In practice, the decision to use allograft currently is determined by the operating surgeon and is influenced by past clinical experiences rather than by discrete clinical criteria and is thus prone to bias. In addition, burn extent as described by TBSA, although a universal language endorsed by the burn community, can grossly overestimate or underestimate the extent of burn. Similarly, burn depth assessment, an important intraoperative "call" of whether grafting is necessary, depends on a subjective interpretation.

This study was also conducted with existing clinical databases not originally intended for research; hence, the recording of data initially was done with the goal of providing clinically useful information than statistically discrete data. Accordingly, data used to determine anatomic location were recorded in nondiscrete fields using text entry. Advantages of this method include potential for high anatomic resolution, for example, "right antecubital fossa," ease of text input, and the widest possible library of terms to describe the site. In a related fashion, this freedom of entry allowed for vague terms such as *arm*, for specific terms that bridge anatomic regions, for example, *axilla*, and for errors in data entry such as misspelling or transcription errors. In addition, these entries were dependent on subjective observation by the recorder.

CONCLUSION

The use of CPA skin is a well-established practice among surgeons providing acute care for patients with severe burns. CPA is used to provide temporary biologic closure, allowing alternative options for permanent closure to be used. Within our population, CPA is almost universally used for patients with a TBSA >50%. This is important because it establishes a population with significant burns who may be able to be managed with autograft alone (30–49% TBSA) and a threshold above which management with CPA should be expected. There is also correlation for high ISSs, increased ICU and overall LOS, increased operative, and high frequency of concomitant burns to the face, hands, genitals, and feet. Although there are no limitations to where CPA can be placed, it is most commonly placed on the extremities and trunk. Future studies with greater statistical power are needed to investigate mortality, morbidity, and reconstructive needs. Furthermore, the implications of CPA-induced allogenic stimulation on PRA and future VCA donor-matching deserves attention. Additional randomized, controlled, head-to-head trials of CPA vs GPA, engineered skin substitutes, or staged autografting would contribute greatly to the literature.

ACKNOWLEDGMENTS

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