EFFICACY OF NONCULTURED MELANOCYTES TRANSPLANATATION VERSUS MINIGRAFTING IN VITILIGO

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Abstract

Background: Surgery is one of the recognized lines in treating vitiligo. Comparative studies are lacking.

Objective: The present study was designed to compare between transplantation of non-cultured melanocytes (MT) and minigrafting (MG).

Methods: MG and MT were concomittantly performed on 105 patients.

Results: There was no significant difference in the repigmentation response for MG (65.7%) and MT (73%). However, MG resulted in a significantly better extent of repigmentation (77.2% ± 24.6) compared to MT (56.3% ± 23.6). The repigmentation response for MG-treated patches was significantly better for localized (100%) compared to generalized vitiligo (62.1%). This was not for MT-treated patches (localized 100%, generalized 71.3%). The extent of repigmentation for localized vitiligo was better than generalized for both procedures, implicating the role of autoimmune response in hindering melanocyte proliferation and function. Younger patients responded better to both procedures. On comparing the response and extent of repigmentation of selected body sites, for MG, all were comparable, but for MT, elbows and fingers showed no repigmentation. Moreover, hands and feet showed a lower extent of repigmentation than the rest of the body for MT.

Conclusion: MG is an easier procedure, with a better cosmetic outcome compared to MT. It is suitable for most body sites including skin overlying joints and periorbital areas. MT could be considered for larger patches present on the trunk and legs.

Key Words

Vitiligo, treatment, autologous grafting, minigraft, melanocyte transplantation.

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Introduction

Vitiligo patches resistant to medical treatment in exposed areas repesent a therapeutic problem to physicians and an aesthetic problem to dark patients. Surgical procedures for correcting leukoderma have been reported since the sixties of the last century and included Thiersch's grafts[1], minigrafts[2,3], epidermal grafts[4], and the development of transplantation of cultured[5] or noncultured[6] melanocytes. All employ melanocytes from the patient's normal skin for covering depigmented patches. The experience with vitiligo, especially the generalized type was on limited numbers of patients, this may be due to the notion that transplantation therapy did not succeed to the same degree as in localized vitiligo[7-12]. Comparative studies between any of these procedures are lacking[13].

Our aim was to use minigrafts and the transplantation of non-cultured melanocytes in a large series of generalized vitiligo patients. A minigraft test as suggested by Falabella[14] was not performed, since the procedures were considered as test and therapy at the same time. A positive repigmentation response is a positive test for either procedure. In the present study, we compared two surgical procedures, minigrafting (MG) and non-cultured melanocyte transplantation (MT) regarding the rate of success of each procedure and in relation to the type of vitiligo (generalized or localized), the state of activity of the disease, the site of the lesion that gives the highest extent of repigmentation as well as the feasibility of either technique considering expenses, healing time, complication and cosmetic appearance.

Patients and Methods

One hundred and eleven patients (111) with vitiligo were included in the present study. Six patients were lost to follow up. The clinical data of patients completing the study are given in table I.

Since various conditions, such as disease activity[11] and type of vitiligo[15,16], affect the outcome of surgery, MG and MT tested in the present study were performed on the same patient. In every patient two procedures were performed in two different areas, and a third patch was left as a control.

Included in the present study were patients with stable and moderately progressive vitiligo who wished to repigment resistant vitiligo patches or to achieve a rapid

repigmentation. Ideal size of lesion chosen was from 0.5 x 0.5 cm to 8 x 5 cm for minigrafts and from 1 x 2 cm to 10 x 10 cm for transplantation of noncultured melanocytes. Patients were instructed to stop any previous treatment at least 4 weeks prior to surgery. From every patient full medical history was taken, examination for the extent of body involvement with vitiligo was performed and consent was obtained. Prophylactic broad-spectrum antibiotics (Velosef ; cephradine; first generation cephalosporine; Squibb) were given to the patients starting intramuscularly one hour before operation and continuing orally for 3 days postoperatively.

MG was performed according to the principles of Falabella[12] with few modifications. Punch grafts (2 mm) were obtained from the medial aspect of the thigh under local anesthesia. The minigrafts were transferred to a saline soaked piece of gauze and excess dermis and subcutaneous fat were trimmed using iris scissors and non-toothed Adson's forceps. Minigrafts were implanted in equal-sized recipient sites previously prepared and set 4-5 mm apart from each other. After the grafts have been pressed firmly, a broad-spectrum antibiotic spray was applied (Tribiotic Spray , which contains neomycin, bacitracin and polymyxin; 3M Health Care, UK) followed by Op-site spray (Smith & Nephew Med. Corp. ltd., UK). Micropore adhesive tape was then placed as a dressing directly on the grafted area. Gauze and plaster were then applied in order to keep the micropore in place. This was followed by Crepe bandage to increase the contact of the grafts to the recipient skin. Dressing was removed after 15 days.

MT was performed according to Gauthier and Surleve-Bazeille[6]. Treatment was carried out in two steps on two consecutive days. On the first day one or several superficial skin samples (2 cm^2) were obtained using a dermatome with a razor blade (Silver's dermatome, Padgett instruments, UK) from the medial aspect of the thigh under local anesthesia. Skin pieces were immersed in sterile phosphate buffered saline solution (PBS 0.1% without calcium or magnesium, Seromed, Germany). The skin samples were cut into small squares (0.5 x 0.5 cm) under completely aseptic conditions in a sterile Petri dish. They were then incubated for 18 hours (16-20 h) in 0.25% trypsin solution at 4 C. Blisters were induced in the recipient vitiliginous area by the application of liquid nitrogen with a cotton-tipped applicator for 15-25 seconds in several areas separated by 1-2 cm. On the following day, the epidermis was separated from the dermis and vigorously pipetted to detach the cells. Excess trypsin was washed off by PBS (repeated 3 times, centrifugation for 10 minutes at 1000 rpm). Finally, epidermal cells were counted by a hemocytometer and were adjusted to obtain a concentration of 1 million cells per mm^3 fluid . Since melanocytes represent 2-4% of epidermal cells, then 20-40 thousand melanocytes were present per ml fluid . The cellular suspension was then aspirated in an insulin syringe with a 25 gauge-needle. The recipient area was cleaned with 70% alcohol, the viscous blister fluid was partially aspirated and 0.1 ml of the epidermal cell suspension was injected in each blister. The average number of melanocytes seeded were 25-100 melanocytes per mm^2. After the injection, the patient remained still for 20 minutes to allow sedimentation of the cellular suspension. The grafted areas were then bandaged. The tops of the blisters served to hold the transplanted cells in place.

Minigrafts healed and blisters dried within 2 weeks. The follow up was then set at biweekly interval. Photographs were taken every two months. Success of MG was characterized by preservation and spread of color from the donor area, seen about 3-4 weeks postoperatively. Pigment spread continued to reach its maximum at 4 months[14], hence the predetermined observation period. Success of MT was characterized by appearance of color within 4-5 weeks postoperatively in injected areas that increased in its size during the observation period. Loss of color from the donor graft in MG was considered negative repigmentation response. Failure of appearance of pigment at the site of injection of MT, or appearance followed by loss of pigment during the observation period was also considered a negative response.

At the end of the follow-up period, the extent of repigmentation was recorded as the surface area of repigmentation in relation to the original size of the patch and was reported as excellent (75-100%), good (50-75%), fair (25-50%), poor (0-25%).

Statistical Analysis

The clinical data were recorded on an investigative report form. These data were then transferred to IBM card using IBM-PC with statistical program "Microstat V2" to obtain: I) Descriptive statistics: a) Mean (x), b) Standard deviation (SD) & c) Range (min-max). II) Analytical statistics: a) Student's t-test: to compare between 2 independent means. Difference was considered significant at $p<0.05$, b) Chi-square test for qualitative data.

Results

Three hundred and twenty seven (327) vitiligo sites in one hundred and five (105) patients were studied in the present work. 222 sites were surgically treated and 105 sites were left as control. MG and MT were performed in two different locations in 100 patients and a third area was left as a control. Five patients were treated by minigrafts only, as either the site (periorbital) or the size of patches (< 2 cm^2) was unsuitable for MT therapy. Fifty-nine patients were followed up for up to one year and 46 were followed up for at least 4 months only. The clinical data of patients included in the present study are listed in table I.

1. Response to MG and MT:

a) Out of 105 patients treated with MG, 69 patches (65.71%) showed positive repigmentation with an average extent of repigmentation of 77.2% ± 24.6. The extent of repigmentation was excellent in 47 (68.1%), good in 12 (17.4%), fair in 9 (13%) and poor in one (1.5%) patient (Figure 1).

b) On the other hand, out of 100 patients treated with MT, 73 patients (73%) showed positive repigmentation with an average extent of repigmentation of 56.3% ±23.6. The extent of repigmentation was excellent in 18 (24.871%), good in 25 (34.2), fair in 25 (34.2) and poor in 5 (6.8%) patients (Figure 1 & 2). No significant difference in the repigmentation response between MG and MT was detected (p>0.05) (Figure 3), whereas the extent of repigmentation attained with MG was significantly higher than MT (p<0.05) (Figure 4, Figure 5).

2. Correlation of results of MG and MT to clinical data:

Although there was no significant relation between the repigmentation response for MG and MT and the patients' sex, the age of patients showing positive repigmentation was significantly lower compared to those failing to respond to both MG and MT (p<0.05). Mean age of responders with MG was 22.5 + 9.3y vs. 28.4 + 10.2 y to nonresponders and 23.2 + 9.1 y for responders with MT vs. 29.6 + 11 y for nonreponders. On comparing the positive repigmentation response of MG to MT as regards age and sex of the patient, the difference was statistically insignificant (p>0.05). (data not shown).

3. Comparison of the results of MG andMT in relation to type of vitiligo:

The repigmentation response in MG-treated patches was significantly greater in patients with localized vitiligo (100%) when compared to generalized vitiligo (62.1%) (p<0.05). In contrast, repigmentation response in MT-treated areas showed no significant association with the type of disease (p>0.05) (Figure 6). The repigmentation response in MG compared to MT was nonsignificant as regards the type of vitiligo (p>0.05).

On comparing the extent of repigmentation achieved with MG for generalized to localized vitiligo, the difference was statistically significant. Localized vitiligo showed greater extent of repigmentation (90%± 8.5) in comparison to generalized vitiligo (75.1% ± 25.8) (p<0.05). Similarly, the extent of repigmentation achieved with MT for localized vitiligo (80.7%±20.9) was significantly better than for generalized vitiligo (53.7% ±22.5)(p<0.05) (Figure 7). On the other hand, on comparing the extent of repigmentation achieved with MG to MT, the difference was nonsignificant for localized vitiligo (p>0.05), but significant for generalized vitiligo (p<0.001) (Figure 8).

4. Results of MG and MT according to body site:

a) Repigmentation respone: The results of successful repigmentation response and extent of repigmentation of MG and MT in different body sites are summarized in Table II. Comparing the response of pigmentation with MG in face to trunk, face to hands, face to foot, face to fingers and face to elbows, all sites showed insignificant differences in their repigmentation response (p>0.05) (Figure 9). Comparing the same sites with each other for the appearance of pigment after MT, the response of pigmentation in face to trunk, face to foot and face to hand was statistically insignificant (p>0.05). However, comparing the repigmentation response of face to fingers and face to elbows the repigmentation response in face was significantly better (p<0.05). The fingers and elbows showed practically no repigmentation response (Figure 10). When the repigmentation response of MG was compared with that of MT in selected body sites, face, trunk, hands, feet and fingers showed nonsignificant difference (p>0.05), whereas in elbows MG was significantly better

than MT ($p<0.05$) (Figure 11 & Table II).

b) Extent of repigmentation: When the extent of repigmentation achieved with MG in face, trunk, hands and feet was compared to each other, all sites showed insignificant differences ($p>0.05$) (Figure 12). When areas treated with MT were compared to each other, the face to trunk and face to foot were statistically insignificant ($p>0.05$), whereas face to hand ($p<0.001$), trunk to hand ($p<0.001$), trunk to foot ($p<0.05$) and hand to foot ($p<0.05$), revealed significant difference. The hand always showed the least extent of repigmentation and the trunk being better than the foot (Figure 13).

On comparing the extent of repigmentation achieved with MG and MT in the same selected sites, we found that the extent of repigmentation acquired in face and trunk were similar with both treatment modalities, the difference was statistically insignificant ($p>0.05$). However, the extent of repigmentation attained with MG in foot and hand were significantly better than that attained with MT (foot $p<0.05$, hand $p<0.001$) (Figure 14 & Table II).

5. Relation of positive repigmentation response of MG and MT to disease activity:

Vitiligo was considered stationary, when there were no new lesions appearing or no increase in the size of existing lesions for 6 months or more before performing our surgical procedures. Moderately prgressive vitiligo is ascribed to cases in whom a slow increase in size or number of vitiligo macules has occurred over the last 6 months with a minimal time interval of 2 months since the last change of vitiligo condition. There was no significant difference between patients repigmenting and those failing to repigment with MG and MT and the duration of disease inactivity ($p>0.05$) (Table III).

6. Side effects:

Side effects are listed in Table IV, 5% of patients had more than one. None of these side effects significantly affected the repigmentation response of either MT or MG.

7. Feasibility and Practicality:

Aspect	MG	MT
Operation time for a 30 cm^2 vitiligo patch	1° hours	3° -4 hours
Days taken to perform the procedure	1 day	2 days
Healing time for grafts	2 weeks	2-3 weeks
Repigmentation time: start	3-4 weeks	4-5 weeks
Repigmentation time: total	4 months	4-6 months

Discussion

The reason for choosing MG and MT in the present study was that the first procedure represents an office procedure that could treat small vitiligo patches [3]. The second procedure involves a laboratory procedure that represents the first steps of cell culture without the need for culture media, incubators or specialized and trained staff. Besides, transplantation of noncultured melanocytes could be a more suitable treatment modality than minigrafts for larger vitiligo patches (up to 100 cm^2) [6]. Moreover, noncultured melanocytes were reported to be more capable of migration and proliferation than the cultured ones [17].

In the current study, pigment started spreading from MG 3-4 weeks postoperatively as previously reported[10,12,14,18]. In cases of MT, pigment appeared after 4-5 weeks and was in the form of minute brownish spots 1-2 mm in diameter. This interval is longer than that mentioned in the preliminary study by Gauthier & Surleve-Bazeille [6], where stippling appeared after 3 weeks. The time taken in studies using cultured melanocytes is even much shorter, where pigment started as early as 6 days in one study [19] and took 10-21 days in the other [20]. The longer time taken in the current study could be due to the lower number of melanocytes present within the epidermal cell suspension in contrast to those present in studies using cultured melanocytes. Falabella et al.[20] mentioned the multiplication of the number of melanocytes by 10 times in cultures, whereas the number of melanocytes applied by Loentz et al. [19] were 10-20 million cells/ml. In the present study only 20-40 thousand melanocytes were present per ml fluid (25-100 cells were seeded / mm^2). This calculation is based on the fact that melanocytes represent 2-4% of the epidermal cell population [21]. The low number of cells may, accordingly, be the cause of the longer duration of repigmentation. Fluid present inside the bullae may have also hindered proper growth of melanocytes. Loentz' and Falabella's studies mentioned above applied melanocytes directly on the denuded areas without intervening fluid [19,21].

Pigment in MG and MT treated areas gradually increased in size, an observation that could be due to melanocyte proliferation and migration under the influence of several mitogens secreted by surrounding keratinocytes [22,23]. Therefore, melanocytes taken from a small donor area could pigment a much larger recipient area amounting to 16 times its size [14]. Full repigmentation was achieved in MG within 3-4 months, results comparable to previous studies [10,13]. Appreciable repigmentation took a longer time with MT (4-6 months). Most previous studies mentioned that in 3-4 months maximum amount of pigmentation was reached [20,24]. Pigment spread was at first rapid and then slowed down. The pigment acquired in MG-treated areas was not lost over one year, which represents the entire observation period. This was not the case in MT-treated patches, where loss of pigment was observed in 2 patients who later developed activity.

The repigmentation response in all treated patients was comparable in the two techniques, however, as regards the type of vitiligo, the failure rate of repigmentation in cases with generalized disease receiving MG was statistically higher than localized disease. These results are comparable or may be even better than those obtained by Falabella et al. [16] in a MG-test study of patients attempting to perform melanocyte transplantation and by Boersma et al. [25]. Both groups observed that less than half the patients with generalized vitiligo (13/27 in the first study [16] and 24/59 in the second study [25]) showed a positive MG test. On the contrary, almost all patients of the first study with localized disease (19/20) had a positive MG test [16].

On the other hand, the repigmentation response of MT in cases with generalized vitiligo was found to be statistically insignificant in comparison to those with localized vitiligo. The reason for the discrepancy between the results of MT and MG among the two types of vitiligo is unknown. Loentz et al. [19] claimed that the activity of the disease, which primarily reflects the immune response in generalized vitiligo, is not the determining factor in the success of take of cultured melanocytes and that the site to which the cells are transplanted is more important. This could be also the case with non-cultured melanocytes.

On comparing the extent of repigmentation, MG-treated patches were significantly better than MT at 4 months. Cases with generalized vitiligo resulted in a significantly lower extent of repigmentation than localized cases both in MG- and in MT-treated patches. Comparing the repigmentation response in cases of generalized vitiligo treated with MG to those treated with MT (generalized MG: generalized MT) the difference was not significant. Similar results were obtained on comparing the repigmentation response in localized vitiligo. On the contrary, the extent of repigmentation achieved in generalized vitiligo was significantly lower in MT-treated patches compared to MG-treated patches, while the difference between the two procedures was insignificant in localized disease. The lower results obtained with generalized vitiligo infer that the immune response hinders both the existance and function of melanocytes[26]. The presence and level of antimelanocyte antibodies correlate positively with the activity and extent of generalized vitiligo [27,28].

Noticeably, results obtained in the present study using MT in localized vitiligo are comparable to those of Gauthier & Surleve-Bazeille [6]. They treated 11 patients with localized vitiligo (2 segmental and 9 focal vitiligo) and one case with nevus depigmentosus with MT. The repigmentation response in their series was 66.6% (8/12), including the patient with nevus depigmentosus, with an average extent of pigmentation of 85% ±12.8. The 11 patients with localized vitiligo studied in the present work showed a better repigmentation response (100%) with a comparable extent of repigmentation (80.7%± 20.9). Our results show, however, a lower extent of repigmentation compared to those obtained in studies on transplantation of cultured melanocytes [19,24]. It is likely that low melanocyte concentration, presence of fluid in the bullae and wide spacing between bullae are the reasons for the lower extent of repigmentation for MT at 4 months. A further evidence for these speculations is the significant amelioration of the extent of repigmentation observed with time with MT (data not shown). Falabella et al. [15] reported that curettage applied to the recipient area greatly improves melanocyte take. Changing the transplantation technique and application of curettage may yield better extent of repigmentation.

The age of patients showing a positive response to MG and MT was significantly lower compared to patients with a negative response. Similar results were obtained by Falabella and his co-workers [16] in patients with generalized vitiligo. Olsson and Juhlin [24], however found no influence of age of the patients on the outcome of therapy. The association of the lower age with a positive response might be explained by the observation that melanocytes of patients older than 45 years have a lower

potential to proliferate and produce pigment in culture. A similar behavior could also be present in vivo [19]. There was no statistically significant relation in the repigmentation response of MG compared to MT as regards the age and sex of the patient or the duration of disease inactivity, probably because both procedures were performed on the same patient.

In the current study, vitiligo was considered stationary, when there were no new lesions or increase in the size of existing lesions for 6 months or more before performing the surgical procedures. In order to test the assumption of Loentz and his colleagues [19] that the activity of disease played no role in the repigmentation response after transplantation of cultured melanocytes, we included 39 patients in the present study with moderately active vitiligo. There was no significant difference between responders and non-responders treated with either procedure in relation to disease activity. These findings may stress the importance of a minigraft test before treatment of larger patches [6, 15, 16]. MG test helps, in addition, to make the patient understand the procedure, the time taken for repigmentation, in addition to testing for the development of Koebner phenomenon or keloids at the donor area [14, 16]. The significance of MG test has been questioned by Boersma et al. [25]. They found that in 2 patients with a previous positive MG test, Koebner phenomenon developed in the donor area on performing minigrafting of the whole vitiligo area, with depigmentation of the transplanted grafts, indicating reactivation of the disease process.

As regards body sites, MG could be performed on any site in the body except for the finger tips and periungual areas due to thickness of the skin [29]. MT was unsuccessful in elbows and fingers due to their mobility and probably also, corrugation of the skin hindering proper seeding of melanocytes. Furthermore, lips and perioral areas could not be treated by MG or MT due to their mobility during grimacing or eating and hydration produced from drinking and salivation. Besides, these areas in addition to the nose are sensitive to liquid nitrogen application for MT as severe pain and edema are a usual consequence [30].

Statistical comparison was performed between selected body sites, which were face, trunk, dorsa of the hands and feet, elbows and fingers. The face represents sun-exposed highly vascular site, trunk represents the sun protected site, the dorsa of the hands and feet are sun-exposed, less vascular sites which are prone to friction and trauma, elbows and fingers are mobile sites which are also prone to friction and trauma. All these MG-treated sites showed insignificant differences in their response and in the extent of pigmentation when compared to each other. This indicates that different anatomical sites whether sun exposed or nonexposed, fixed or mobile respond similarly to MG, and that the presence or absence of sun does not change the repigmentation response.

For MT face, trunk and hands showed comparable repigmentation response. In contrast, areas overlying joints as the elbows, knees as well as the hands and fingers resulted in significantly lower repigmentation response. The best extent of repigmentation achieved with MT was in lesions present on the trunk, leg and face. MT could, therefore, be used for the treatment of relatively large patches commonly present on the trunk and leg, which are less sensitive to pain than the face. Again, the extent of repigmentation in positively repigmenting patches of the hands and feet were lower than the rest of the MT-treated patches. Similar results were obtained in studies using cultured melanocytes [19,24], where success was least in hands, feet, fingers and elbows.

On comparing the anatomical sites treated with MG to similar ones treated with MT, the repigmentation response was lower only on elbows receiving MT. Unfortunately, the number of patients treated with MT on the fingers was too small (2 patients) to be compared to MG. Generally, MG yielded an extent of repigmentation comparable to MT in all sites treated except for the hands and feet, which were significantly higher in minigrafted areas. MG is therefore, especially useful in treating mobile areas of the body, such as dorsa of the fingers, elbows and knees as well as for the periorbital area. The results of other surgical procedures as transplantation of noncultured [6] or cultured melanocytes [15, 19, 24] and epidermal grafting using the tops of suction blisters [31] are less favorable.

The main side effects encountered are mainly cosmetic problems. Cobble stoning resulting from MG tended to disappear by time. Variegate coloration which is more pronounced with MT also improved by time. Scars at donor area make epidermal grafting superior to both in that aspect. Koebner at donor was rare and indicated activity [24].

To conclude, MT needs a specialized laboratory and is time consuming and tedious compared to MG. Furthermore, its cosmetic outcome is less than that obtained with MG. On the other hand, MG can be easily performed by most dermatologists using simple instruments present in every office. MG heal rapidly with a favorable cosmetic outcome.

Table I. Clinical data of patients receiving MG & MT:

Item	Data
Age	10-50y (Median 22y)
Sex	66 females:39 males
Skin Type	III=12, IV=66, V=26, VI=1
Type of Vitiligo	94 Generalized:11 Localized
Duration of disease inactivity	2 months-15 years (1.9y ± 2.5)

MG=minigraft, MT=noncultured melanocyte transplantation

Table II. Results of repigmentation response and extent of repigmentation of MG and MT in relation to site of the body treated:

Body site	Repigmentation response		Extent of repigmentation	
	MG	MT	MG	MT
Face	68.4 %*	100%	81.2% ± 20.6*	66.7%±15.1
Nape	100%	100%	90% ±7.1	66.7 ± 25.2
Trunk	72.7%*	69.2%	76.3% ± 21.8*	73.3 ± 20.6
Arm	33.3%	66.7%	80%± 0	63.3% ± 5.8
Leg	83.3%	81%	75% ± 28.5	59.1% ± 25
Hand	41.2%*	33.3%	90% ±26.5**	17.5± 5.0
Wrist	80%	80%	72.5% ± 9.6	65% ± 21.2
Fingers	33.3%*	0%	37.5% ±31.8	0%
Elbows	66.7%**	0%	66.25% ± 35	0%
Knee	60%	33.3%	100% ± 0	60% ± 15
Foot	76.9%*	79.3%	70.4% ± 26.3**	50% ± 23

**= Nonsignificant difference found in the selected body sites, **= Significant difference found in the selected body sites when MG was compared to MT.*

MG=minigraft, MT=noncultured melanocyte transplantation

Table III. Comparisons of the number of patients failing to respond to surgery at different length of time since the last appearance of activity all are nonsignificant

Interval	No. of patients	Interval	No of patients	Chi sq
< 2 mo	8	> 2mo	28	2.1
< 4 mo	12	> 4 mo	24	0.9
<6 mo	13	> 6 mo	23	1.0
< 8 mo	18	> 8 mo	18	1.1
< 12 mo	16	> 12 mo	20	0.02

Table IV. Side effects encountered with surgical treatment of vitiligo:

Side Effects	No of pts (%)
a) Liquid Nitrogen application (MT): i) Pain ii) Paraesthesia iii) Localized edema and swelling iv) Persistant erythema v) Depressed scar	 All 3% 5.8% 10% 2%
b) Injection of melnocyte-containing suspension (MT): i) Loss of fluid from bullae ii) Hemorrhage iii) Infection	 5% 10%
c) MG: i) Itching ii)Infection iii) Miliaria pustulosa	 5.7% 1.9% 4.8%
d) Cosmetic Problems: i) Discoloration of the grafts: Hyperpigmentation Variegate coloration ii) Cobble stone appearance of MG	 MT>MG 44.5%

MG=minigraft, MT=noncultured melanocyte transplantation, pts=patients

Fig. 1&2: Extent of repigmentation acquired in minigraft and melanocyte transplantation treated areas.

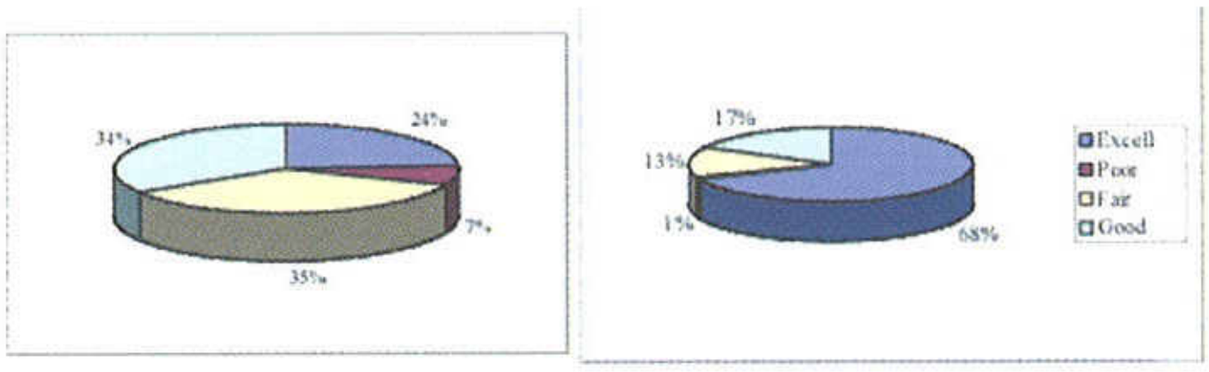


Fig. 4&5: Comparison of the extent of repigmentation of minigraft and melanocyte transplantation.

Fig. 7: Comparison of the extent of repigmentation of generalized to localized vitiligo in minigraft and melanocyte transplanted areas.

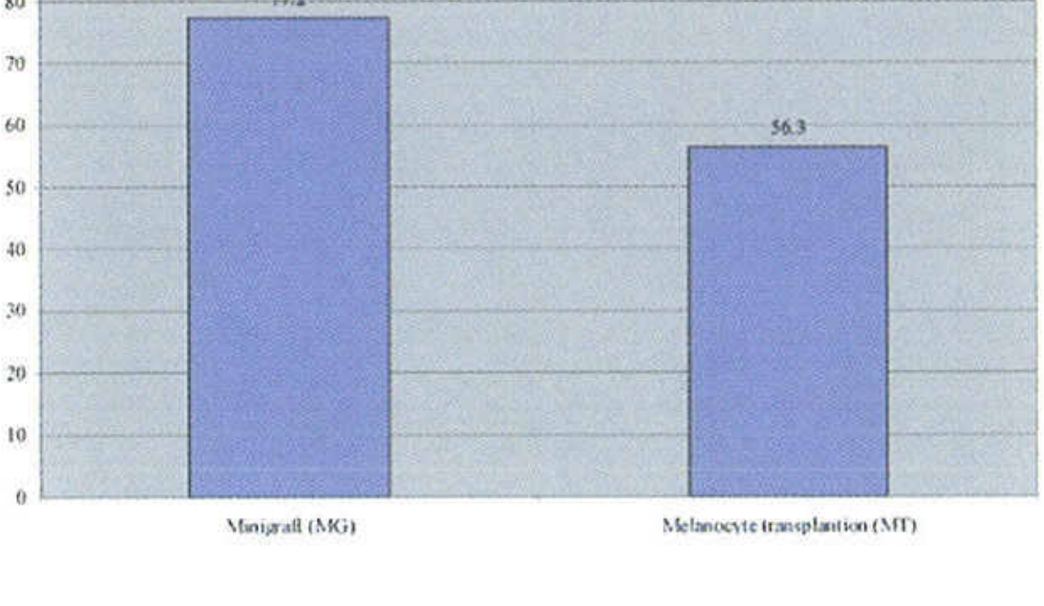


Fig. 3: Comparison of the repigmentation response of minigraft versus non-cultured melanocyte transplantation.

Fig. 6: Comarison of the repigmentation response in relation to type of vitiligo.

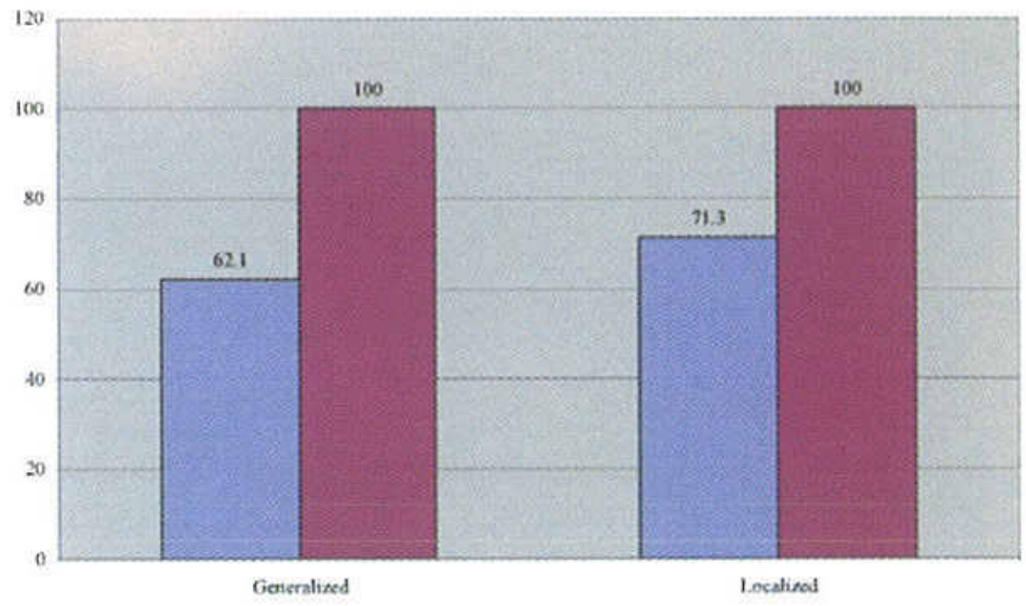
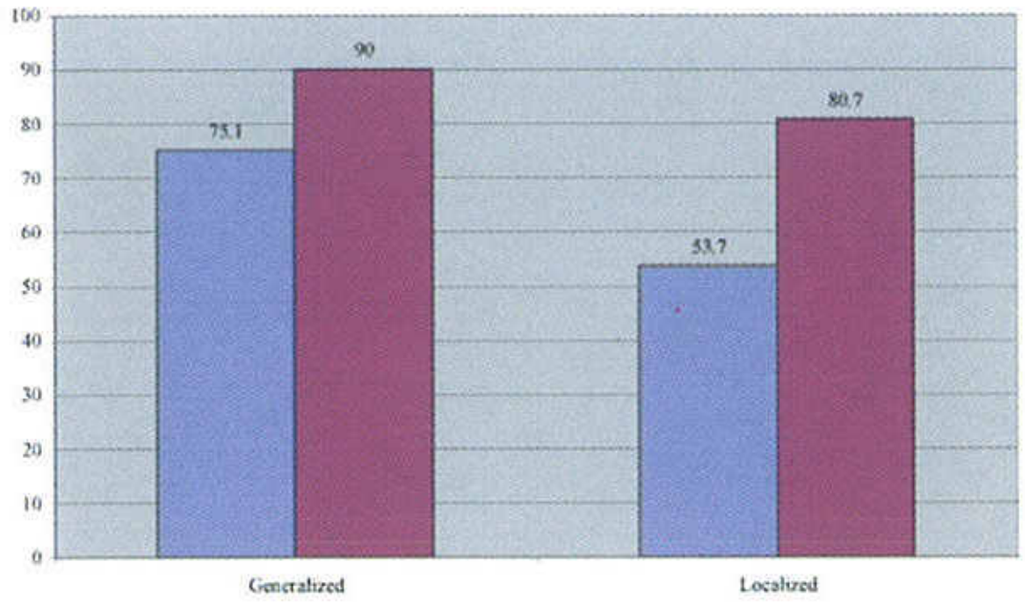


Fig. 8: Comparison of the extent of repigmentation of minigraft to melanocyte transplantation-treated areas in relation to the type of vitiligo.



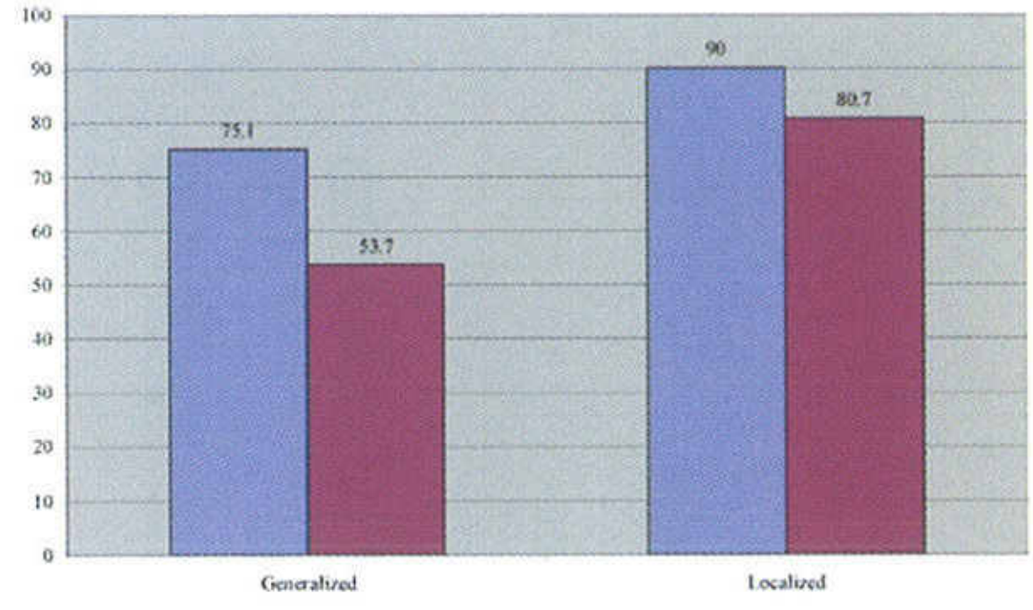
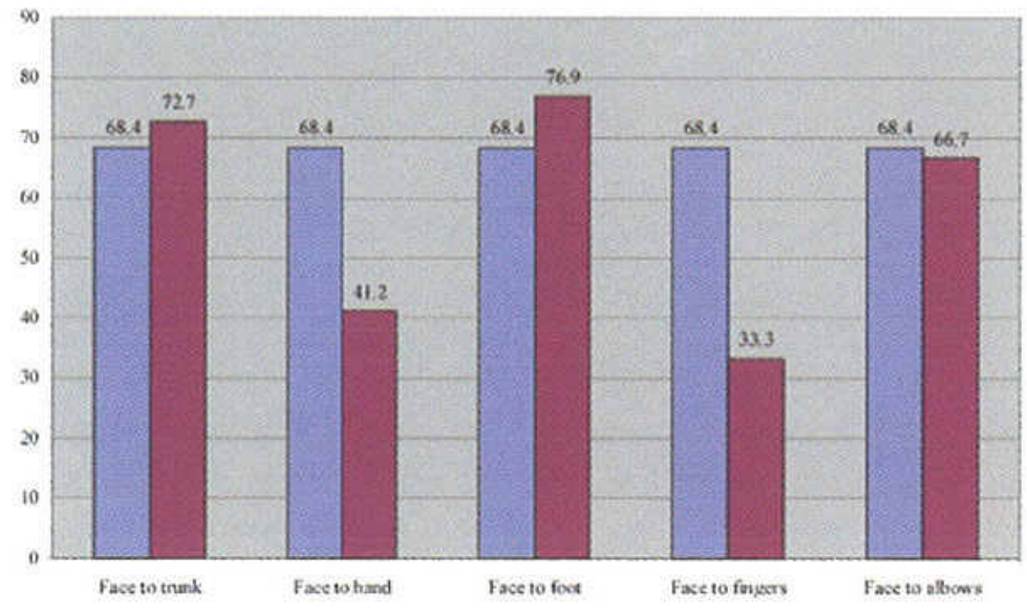


Fig. 9: Comparison of the repigmentation response of minigraft-treated areas in selected body sites.

Fig. 10: Comparison of the repigmentation response of melanocyte transplanted areas in selected body sites.



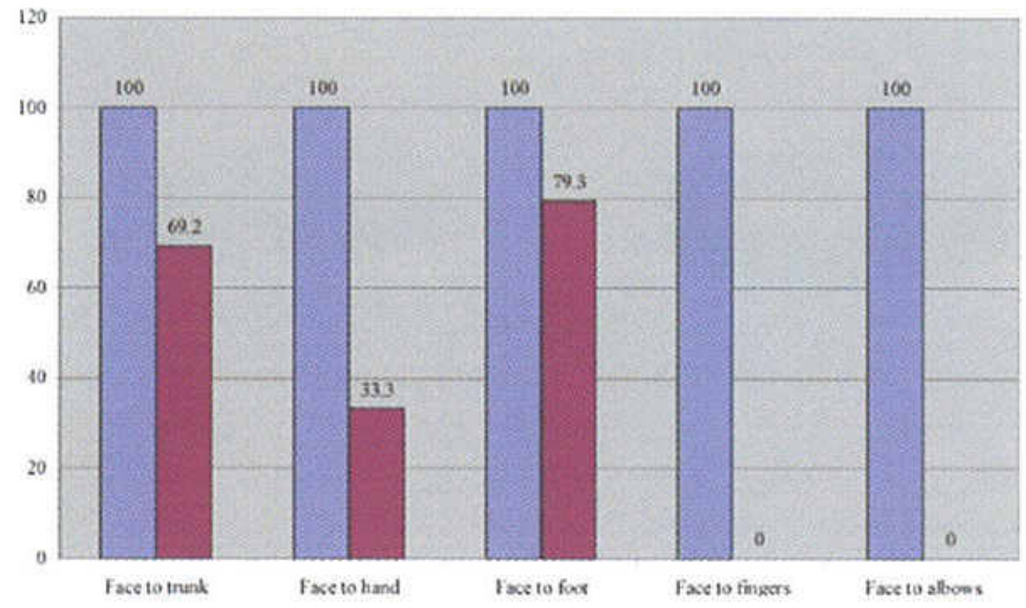


Fig. 11: Comparison of the repigmentation response of minigraft to melanocyte transplanted areas in relation to the site of the body.

Fig. 12: Comparison of the extent of repigmentation of minigraft in selected body sites.

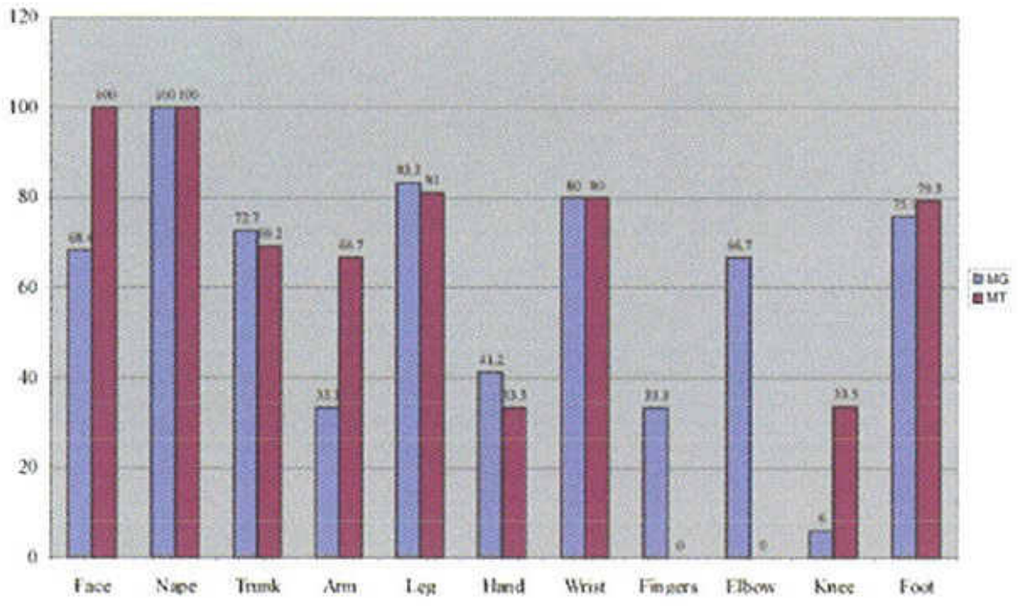
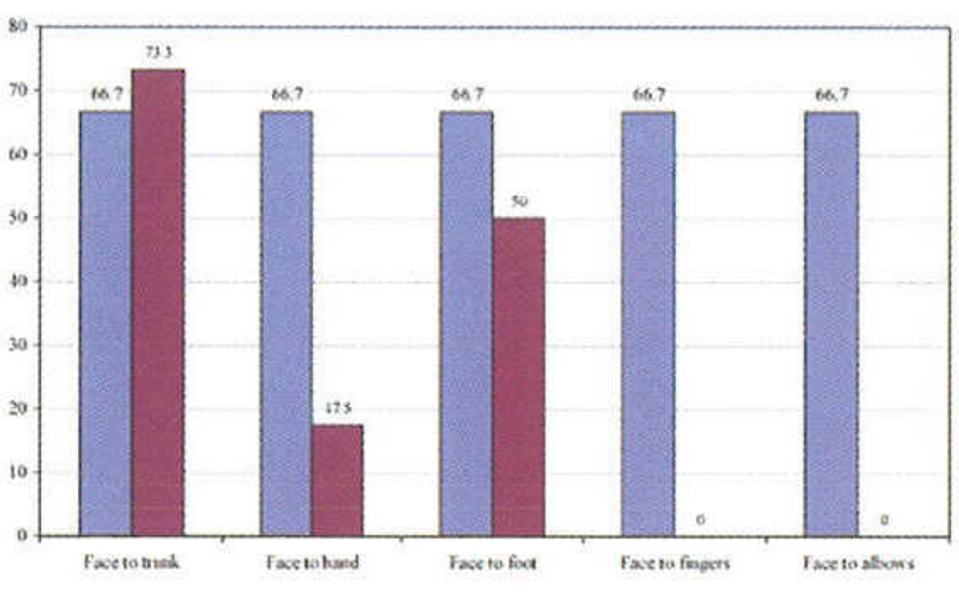
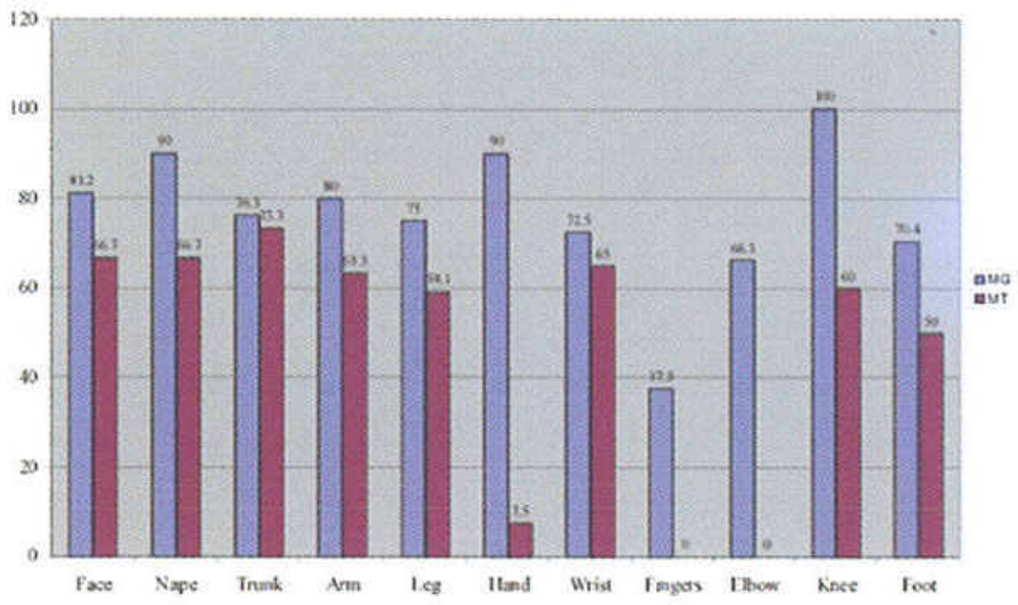


Fig. 13 : Comparison of the extent of repigmentation of MT in selected body sites

Fig. 14: Comparison of the extent of repigmentation of minigrafting to melanocyte transplantation in relation to body site treated.





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