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Comparison between two surgical techniques for root coverage with an acellular dermal matrix graft

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Abstract

Aim: The aim of this randomized, controlled, clinical study was to compare two surgical techniques with the acellular dermal matrix graft (ADMG) to evaluate which technique could provide better root coverage.

Material and Methods: Fifteen patients with bilateral Miller Class I gingival recession areas were selected. In each patient, one recession area was randomly assigned to the control group, while the contra-lateral recession area was assigned to the test group. The ADMG was used in both groups. The control group was treated with a broader flap and vertical-releasing incisions, and the test group was treated with the proposed surgical technique, without releasing incisions. The clinical parameters evaluated before the surgeries and after 12 months were: gingival recession height, probing depth, relative clinical attachment level and the width and thickness of keratinized tissue.

Results: There were no statistically significant differences between the groups for all parameters at baseline. After 12 months, there was a statistically significant reduction in recession height in both groups, and there was no statistically significant difference between the techniques with regard to root coverage.

Conclusions: Both surgical techniques provided significant reduction in gingival recession height after 12 months, and similar results in relation to root coverage.

Key words: aesthetics; clinical trials, controlled; clinical trials, randomized; comparison studies; gingival recession/surgery; gingival recession/therapy; grafts, gingival; matrix, acellular dermal; surgical flaps

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Treatment of exposed root surfaces has become an important therapeutic issue due to patients' increasing aesthetic demands. Successful coverage of exposed roots has been the objective of various mucogingival procedures (Wennström 1996, Zaher et al. 2005). Several surgical techniques have been proposed to treat gingival recession areas such as a free gingival graft, a laterally positioned flap, a double papilla flap, a semilunar flap, a coronally positioned flap, a subepithelial connective tissue graft (SCTG), and guided tissue regeneration (Wennström 1996, Müller et al. 1998, 2000, Del Pizzo et al. 2002, Roccuzzo et al. 2002, Zucchelli et al. 2003, Burkhardt & Lang 2005, Leknes et al. 2005, Trombelli et al. 2005, De Sanctis & Zucchelli 2007). Recently, the association of the enamel matrix derivative with a coronally positioned flap has also been evaluated for the root coverage (Hägewald et al. 2002, Del Pizzo et al. 2005). Among these procedures, the SCTG has achieved high levels of success and predictability for root coverage, and furthermore, it increases the width and thickness of keratinized tissue; however, the amount of donor material that is necessary limits the number of teeth that can be treated in a single surgery. Furthermore, this technique involves a certain degree of discomfort for the patient due to the second surgical site: the donor area, increasing

Conflict of interest and source of funding statement

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the risk of pain and haemorrhage postoperatively.

In dentistry, the acellular dermal matrix graft (ADMG), which is obtained from human skin, has been used as a substitute for the connective donor tissue to increase the width of keratinized tissue around teeth or implants (Wei et al. 2000, Harris 2001, Buduneli et al. 2003), for the treatment of alveolar ridge deformities (Batista et al. 2001), and for root coverage procedures (Aichelmann-Reidy et al. 2001, Novaes et al. 2001, Paolantonio et al. 2002, Barros et al. 2004, De Queiroz Côrtes et al. 2006, Felipe et al. 2007), eliminating the disadvantages described above for the autogenous donor graft. Also, the ADMG has been used to eliminate gingival melanin pigmentation (Novaes et al. 2002b), as a membrane for guided bone regeneration (Novaes & Souza 2001, Novaes et al. 2002a, Luczyszyn et al. 2005) and, furthermore, was recently evaluated as a membrane for guided tissue regeneration in the treatment of mandibular Class II furcation lesions in dogs (Andrade et al. 2007).

Previous studies compared the ADMG with the SCTG for the treatment of gingival recession areas, demonstrating similar results between both surgical techniques (Aichelmann-Reidy et al. 2001, Novaes et al. 2001, Paolantonio et al. 2002). However, these grafts exhibit different healing processes, due to their distinct cellular and vascular structures. The ADMG is a non-vital allograft, which contains principally collagen bundles and elastic fibres. While this material acts as a scaffold for the proliferation of epithelial cells, fibroblasts, and blood vessels from the recipient site to achieve reorganization (Batista et al. 2001), the SCTG contains some vessels and cells, and hence, the healing and vascularization of this graft are based on the anastomosis between the vessels of the receptor site and the graft's vessels (Oliver et al. 1968, Janson et al. 1969). Thus, according to Barros et al. (2004), a larger blood supply may be necessary for the allograft when compared with the SCTG, and based on this a new surgical technique for the root coverage with the ADMG was proposed. This was based on an extended flap, in which the releasing incisions were displaced with to the adjacent teeth, to favour the incorporation of the allograft so that more blood vessels and consequently more nutrients and a better source of cells may be present in the flap. The technique proposed by Barros et al. (2004) was compared with the routinely used surgical technique, which was designed by Langer & Langer (1985) for use with the SCTG and not with the ADMG. In this SCTG technique, the releasing incisions are placed on the proximal surfaces of the tooth with the gingival recession area, and it may not be adequate for the use of ADMG. The close proximity of the incisions to the allograft limits the blood supply and source of cells, and, as a result, may compromise the amount of root coverage. Barros et al. (2004) concluded that the modified technique, in which the releasing incisions were displaced to the adjacent teeth, is more suitable for root coverage of localized gingival recession areas with the ADMG, because it had statistically significantly better clinical results.

However, releasing incisions are used in the surgical technique proposed by Barros et al. (2004), which could decrease the blood supply to the flap, impairing the incorporation of the ADMG. Theoretically, it seems possible that the absence of releasing incisions may favour the blood supply to the graft, and as consequence may accelerate the healing process, and the root coverage (Bruno 1994).

Thus, the aim of this study was to evaluate, clinically in humans a new surgical technique for root coverage with the ADMG, in which releasing incisions were not performed, comparing it with the technique proposed by Barros et al. (2004).

Material and Methods

Population study

After the approval of the Institution's Human Research Committee (protocol 200501.934.58.1), patients were recruited at the Department of Periodontology of Faculty of Dentistry of Ribeirão Preto, University of São Paulo. Fifteen volunteers (nine males and six females) aged 23–54 were selected for the study. The entry criteria were: (1) non-compromised systemic health and no contraindications for periodontal surgery; (2) no previous periodontal surgical treatment on the involved sites; (3) non-smokers; and (4) bilateral Miller Class I or II $\geqslant$2 mm maxillary or mandibular gingival recession areas located on homologous teeth. Radiographs were taken to evaluate inter-proximal alveolar bone level to aid the gingival recession classification of teeth exhibiting recession defects. Teeth with loss of inter-dental papilla height, tooth rotation, and tooth extrusion were not selected. A total of 30 recession areas were treated. All the risks and benefits involved in the procedures were explained to the patients before they signed an informed consent form in agreement.

The patients initially completed a plaque control programme, including oral hygiene instructions to eliminate habits related to the aetiology of the recession area, scaling and root planing, and crown polishing.

Before surgery, in each patient, one of the two teeth with a gingival recession area was randomly assigned, by a coin toss, to the test group (TG) and the contra-lateral tooth to the control group (CG) (Zucchelli et al. 2003, Del Pizzo et al. 2005, De Queiroz Côrtes et al. 2006). The randomization was performed by one investigator 1 h before surgery. Both groups were treated with the ADMG (Alloderm, LifeCell, The Woodlands, TX, USA) as a subepithelial graft. In the CG, this allograft was used with the technique proposed by Barros et al. (2004), in which an extended surgical flap and releasing incisions were performed (Fig. 1a). In the TG, a modification of the Bruno technique (Bruno 1994), which consisted of an extended surgical flap without releasing incisions, was used (Fig. 1b).

In the current study, we tested the hypothesis that the absence of releasing incisions may provide better root

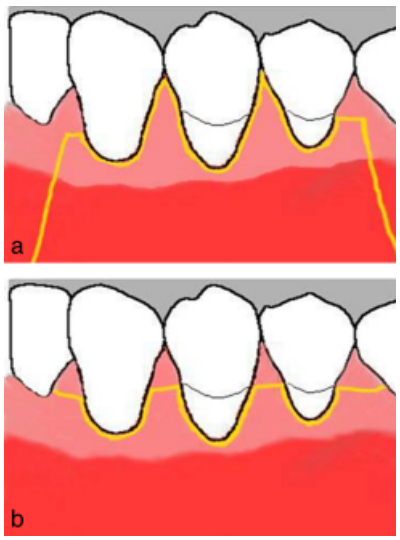

Fig. 1. (a) Control group. (b) Test group.

coverage, considering that the presence of releasing incisions may theoretically impair the blood supply to the allograft.

Clinical assessments

The clinical parameters evaluated were: gingival recession height (GR); probing depth (PD); relative clinical attachment level (RCAL), measured as the distance from a fixed point in a stent to the bottom of the pocket; width of keratinized tissue (KT); and thickness of keratinized tissue (TKT). All clinical parameters were recorded by one blinded experienced periodontist (M. E. F.) at the mid-buccal point of teeth scheduled for surgery and also of the adjacent teeth at baseline and after 12 months. An acrylic stent, in which reference marks were present, corresponding to the mid-buccal point of the teeth involved, was used to determine the exact measurement site at baseline and 12 months after surgery.

The GR, PD and RCAL were evaluated using an automated periodontal probe (Florida Probe, Gainesville, FL, USA). This periodontal probing system has the advantages of a constant probing force of 25 g, precise electronic measurement to 0.1 mm, and computer storage of the data, which allows to perform measurements more accurately. The WKT, distance between the gingival margin and the mucogingival junction (MGJ), was measured, using a compass and the measurement determined by the compass was recorded by a slide caliper (0.05 mm resolution). The TKT was measured 1 mm coronally to the MGJ. In order to ensure that the TKT was measured in the same location during clinical examination 1 and re-evaluation, the distance between the acrylic stent and the location of this measurement was recorded. The TKT was measured with an anaesthesia needle attached to a silicone disc stop. The needle, placed perpendicular to the mucosal surface, was introduced into the soft tissue with light pressure until a hard surface was felt. The silicone disc stop was then placed in close contact with the external soft tissue surface. After carefully removing the anaesthesia needle, the penetration depth was measured with a caliper with a 0.05 mm resolution. Each clinical measurement was performed three times by the examiner, and the mean from these three values was calculated to represent the GR, PD, RCAL, KT, and TKT.

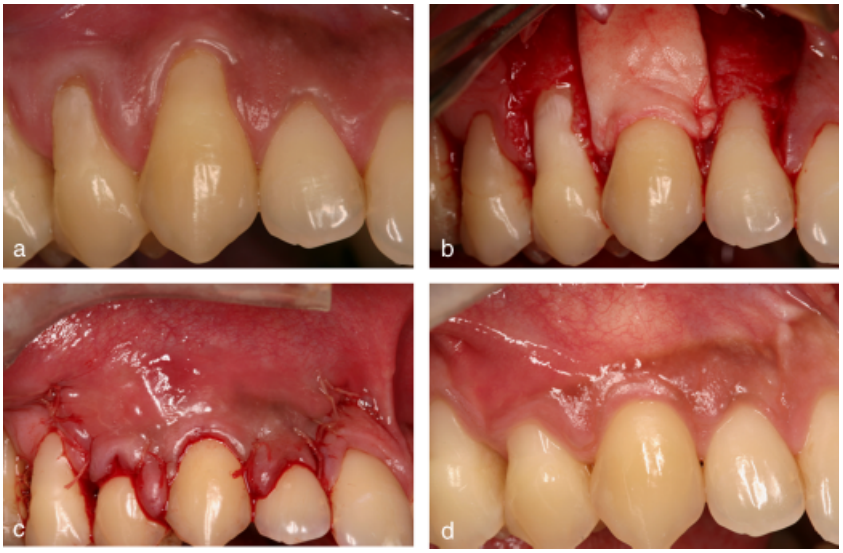

Fig. 2. Clinical sequence of the control group. (a) Pre-operative gingival recession on a maxillary right canine. The restorations on the root surfaces were removed before the surgery. (b) Flap elevated with a partial-thickness dissection and the acellular dermal matrix graft sutured in place. (c) Flap coronally sutured covering the entire graft. (d) Post-operative image of the treated area after 12 months.

Fig. 3. Clinical sequence of the test group. (a) Pre-operative gingival recession on a maxillary left canine. The restorations on the root surfaces were removed before the surgery. (b) Flap elevated with a partial-thickness dissection and the acellular dermal matrix graft sutured in place. (c) Flap coronally sutured covering the entire graft. (d) Post-operative image of the treated area after 12 months.

Surgical procedures

All surgical procedures were performed by a single experienced operator (P. F. A.), and each pair of recession areas was treated in the same surgical session (Figs 2 and 3). After local anaesthesia, the flaps were designed to accommodate the ADMG subepithelially. In the CG, the two releasing incisions were placed on the mesial and distal line angles of the adjacent teeth, distant from the recession area, providing a broader flap. Sulcular incisions were made through the bottom of the crevice, uniting the releasing vertical incisions, and the papillae were included in the flap. In the TG, horizontal incisions were made in the inter-proximal tissues at the base of the papilla at the level or slightly coronal to the cemento-enamel junction (CEJ) of the tooth with the gingival

recession area, and also in the papillae on the mesial and distal adjacent teeth. These incisions were connected by sulcular incisions. A partial-thickness flap was then reflected as close to the periosteum as possible by mesio-distal and apical sharp dissection, parallel to the mucosa, and extended beyond the MGJ, so that the flaps could be passively positioned coronally over the defects without tension. In the TG, all papillae involved in the flap were de-epithelialized.

Following flap elevation, the exposed root surfaces were gently planed with sharp curettes (Hu-Friedy, Chicago, IL, USA) and conditioned with a 24% EDTA gel preparation for 2 min. with subsequent rinsing with sterile saline. The ADMGs were aseptically re-hydrated in sterile saline according to the manufacturer's instructions and were trimmed to cover the exposed root surfaces of the involved teeth, and extended about 3 mm of the surrounding bone. The ADMGs were positioned at the level of CEJ, and the basement membrane side was placed facing the flap, while the connective tissue side was placed in contact with the bone and the root surface. The allografts were sutured over the defects with sling sutures, using 5-0 bioabsorbable sutures. The previously reflected partial-thickness flaps were coronally positioned to cover the entire grafts and sutured without tension into place with sling sutures, also using 5-0 bioabsorbable sutures. In the CG, interrupted sutures were placed in the releasing incisions, and in the TG, the sutures were performed in the papillae of the adjacent teeth. No periodontal dressing was applied.

Post-surgical care

All patients were instructed to avoid trauma and to discontinue toothbrushing at the surgical site. A 0.12% chlorhexidine digluconate solution rinse twice a day for the first 15 days, amoxicillin (500 mg, t.i.d.) for 7 days, starting 24 h before surgery, and a non-steroidal anti-inflammatory agent (nimesulide, 100 mg – twice a day, for 3 days) for control of post-operative pain were prescribed.

The sutures were removed after 15 days. The patients were instructed to clean the surgical area with a cotton pellet soaked in a 0.12% chlorhexidine digluconate solution twice a day for 30 days. All patients were recalled for control and prophylaxis after 1, 2, 3, and 4 weeks and, subsequently, once a month, until the final examination (12 months).

Statistics

Quantitative data were recorded as mean and standard deviations. In order to verify the normality of the data, the kurtosis and skewness curves were used. Because all data were considered to be normal for the parameters analysed, Student's paired *t*-test was used for intra-group (baseline × 12 months) and inter-group (CG × TG) comparisons. The level of significance of 0.05 was used in all statistical comparisons. Initially, a statistical power analysis was not performed for sample size determination, but an initial sample of 15 volunteers was included in the trial ($n = 15$). The $n = 15$ was estimated in clinical studies, which have used a similar methodology in the treatment of gingival recession areas (Barros et al. 2004, Burkhardt & Lang 2005, Del Pizzo et al. 2005, De Queiroz Côrtes et al. 2006). According to the comparison between treatments for the clinical parameter of GR at 12 months, a statistical power estimate was carried out.

Results

Fifteen patients with bilateral Miller Class I gingival recession areas ($n = 15$ patients/30 sites analysed) were treated with ADMG, using the technique designed by Barros et al. (CG: 15 sites) or the technique proposed in the present study (TG: 15 sites). The recessions were located on 12 canines (10 maxillary and two mandibular) and 18 pre-molars (10 maxillary and eight mandibular). Healing was uneventful for all patients, and none was excluded from the study. Therefore, all patients completed the 12 months of follow-up. Considering that the experiment had a >99% power, the sample of 15 patients was considered to be sufficient to detect differences of 1.0 mm in GR between the groups.

Table 1. Clinical parameters (mean values ± SD) at baseline and 12 months – intra-group comparison

Parameters	Test			Control		
	baseline	12 months	*p*-value	baseline	12 months	*p*-value
GR (mm)	2.88 ± 0.81	0.72 ± 0.65	0.000*	2.73 ± 0.76	0.46 ± 0.53	0.000*
PD (mm)	1.52 ± 0.39	1.63 ± 0.33	0.438	1.69 ± 0.54	1.56 ± 0.51	0.353
RCAL (mm)	11.23 ± 2.50	9.50 ± 2.39	0.004*	11.51 ± 2.30	9.77 ± 2.13	0.003*
KT (mm)	1.94 ± 1.66	2.10 ± 1.01	0.598	2.17 ± 1.42	2.58 ± 0.94	0.146
TKT (mm)	0.48 ± 0.29	1.04 ± 0.44	0.001*	0.55 ± 0.21	1.15 ± 0.45	0.000*

*Statistically significant difference at $p \leqslant 0.05$.
GR, gingival recession height; PD, probing depth; RCAL, relative clinical attachment level; KT, width of keratinized tissue; TKT, thickness of keratinized tissue.

Table 2. Frequency of root coverage between the groups after 12 months

Group	100%	99–50%	49–0%
Test	6	7	2
Control	8	7	0

There were no statistically significant differences between the two groups at baseline with regard to GR, PD, RCAL, KT and TKT. At 12 months, there was no statistically significant difference between the two groups for the GR. At 12 months, there was only a statistically significant difference between the CG and TG in the KT, favouring the CG. At baseline, the KT in the TG was 1.94 ± 1.66 mm, and was 2.17 ± 1.42 mm ($p = 0.432$) in the CG; however, at 12 months, the KT in the TG was 2.10 ± 1.01 mm, and 2.58 ± 0.94 mm ($p = 0.007$) in the CG.

There was a statistically significant reduction in GR, a gain in RCAL, and an increase in TKT between baseline and the 12-month post-operative visit for both groups, while the changes in the PD and in the KT were not statistically significant after 12 months (Table 1).

After 12 months, the mean recession height reduction of the CG was superior to that obtained in the TG. The percentage of root coverage was 83.28% for the CG and 74.32% for the TG; however, this difference was not statistically significant. In the TG, there was a root coverage of 100% in six sites, 99–50% in seven sites, and 49–0% in two sites. In the CG, there was a root coverage of 100% in eight sites, 99–50% in seven sites, and no cases had 49–0%. The distribution of sites according to the percentage of root coverage is shown in Table 2.

Table 3. Clinical parameters (mean values ± SD) of adjacent teeth at baseline and 12 months – intra-group comparison

Parameters	Test			Control		
	baseline	12 months	*p*-value	baseline	12 months	*p*-value
GR (mm)	1.48 ± 1.30	0.83 ± 0.80	0.000*	1.61 ± 1.58	0.88 ± 1.04	0.001*
PD (mm)	1.58 ± 0.44	1.63 ± 0.47	0.626	1.36 ± 0.37	1.47 ± 0.52	0.294
RCAL (mm)	10.41 ± 2.50	9.45 ± 1.89	0.011*	10.40 ± 2.56	9.29 ± 1.79	0.001*
KT (mm)	2.13 ± 0.86	2.15 ± 1.03	0.106	2.46 ± 1.07	2.53 ± 1.09	0.175
TKT (mm)	0.70 ± 0.27	0.91 ± 0.32	0.002*	0.66 ± 0.30	0.86 ± 0.33	0.008*

*Statistically significant difference at $p \leqslant 0.05$.
GR, gingival recession height; PD, probing depth; RCAL, relative clinical attachment level; KT, width of keratinized tissue; TKT, thickness of keratinized tissue.

The adjacent teeth of both groups were also evaluated. There were no statistically significant differences between the clinical measurements at baseline and at 12 months between the control and test sites. In both groups, there was a statistically significant reduction in GR, a gain in RCAL, and an increase in TKT between baseline and the 12-month period. There were no significant changes in the other parameters evaluated (Table 3).

Discussion

In this randomized, controlled, clinical study, two surgical techniques for root coverage, using the ADMG as a subepithelial graft, were compared and evaluated after 12 months with regard to root coverage. In the CG, the ADMG was used with the technique proposed by Barros et al. (2004), in which an extended surgical flap and releasing incisions were performed, while in the TG, a modification of the Bruno technique (Bruno 1994), which also consisted of an extended surgical flap, but without releasing incisions, was used. The root coverage was similar between the surgical techniques.

Because the main gingival blood supply is directed caudo-cranially from the vestibule to the gingival margin (Mormann & Ciancio 1977), a broader flap increases the number of cells and blood vessels available to participate in the healing process. This fact is even more relevant when the ADMG is used, because this allograft is non-vital, depending on cellular and vascular proliferation to achieve its reorganization and incorporation into the host tissue (Batista et al. 2001). Based on this, Barros et al. (2004) proposed a surgical technique for root coverage with the ADMG, which consisted in a modification of the technique described by Langer & Langer (1985) for the SCTG. In the surgical technique proposed by Barros et al. (2004), the two releasing incisions were displaced to the mesial and distal line angles of the adjacent teeth, distant from the recession area, creating a broader flap. This technique was compared with the conventional technique (Langer & Langer 1985), and had better clinical and statistically significant results. However, theoretically, it seems possible that the absence of releasing incisions may favour the blood supply to the allograft, favouring its incorporation into the host tissue, and as a consequence may result in more root coverage. However, in the present study, there was no statistically significant difference between the CG (technique proposed by Barros et al. 2004) and the TG (surgical technique, which also consisted of an extended surgical flap, but without releasing incisions) regarding root coverage after 12 months. Both techniques may have impaired the blood supply to some extent, the CG with the releasing incisions, and the TG with the horizontal incisions through the papillae. Comparisons between the present study with those of others may have limited value, because only Barros et al. (2004, 2005) used a similar methodology. In the present study, the percentage of root coverage was 74.32% for the TG and 83.28% for the CG and this result was equivalent to the study of Barros et al. (2005), which achieved 82.5% after 12 months.

There was a statistically significant difference in KT between the CG and TG after 12 months, favouring the CG. In both groups, the flaps were coronally positioned to cover the entire grafts and sutured without tension. However, in the CG, the releasing incisions seemed to provide a more coronal displacement of the flap over the defects, and thus, a greater extension of alveolar mucosa could have been converted in keratinized tissue, and furthermore, perhaps, a tendency of the mucogingival line to regain its original position may explain this difference between the groups (De Queiroz Côrtes et al. 2006). There were no statistically significant differences regarding an increase in the KT between baseline and after 12 months in both groups, and this is in disagreement with other studies, which demonstrated a statistically significant increase in KT after 6 or 12 months after the use of the ADMG for root coverage (Aichelmann-Reidy et al. 2001, Paolantonio et al. 2002, Barros et al. 2005, De Queiroz Côrtes et al. 2006). In the present study, because the gingival tissues were not stained with Schiller's iodine solution to determine the KT, and the tissue covering the graft showed clinical characteristics of alveolar mucosa, it was difficult to determine the precise position of the MGJ during the initial period of healing. However, at baseline and after 12 months of healing, the MGJ was visible, and thus it was possible to measure the KT. It is important to consider that the time required for additional gain in the amount of keratinized tissue is greater for the ADMG than for the connective tissue procedures (Novaes et al. 2001). This observation may be explained by the fact that ADMG is a non-vital, acellular material that acts as a scaffold for cells from the surrounding tissues, and is subsequently substituted by the host tissues. Thus, this biological process may require an additional period of time, and to delay the keratinization of the epithelium. In addition, differences in the acellular dermal matrix orientation (basement membrane side in contact with the flap or with the periosteum) may influence the cellular healing dynamics of this material in terms of keratinization of the overlying epithelium. Additional studies to show whether and how changes in the acellular dermal matrix orientation influence the healing process and the KT would be interesting (Novaes et al. 2001). Only long-term treatment outcomes of root coverage with ADMG with different orientations of matrix will clarify these assumptions.

Although, in both groups, there were no statistically significant differences with regard to KT between baseline and after 12 months, there was a statistically significant increase in TKT in the

selected teeth, and also in the adjacent teeth. This is particularly important because, although it has been admitted that a minimal amount of gingival tissue can be compatible with a healthy periodontal condition (Miyasato et al. 1977), the marginal tissue thickness is a critical determinant of future recession areas (Novaes et al. 1975). Hence, an increase in gingival thickness, as obtained with ADMG, could help to prevent future recession areas in sites with a thin periodontal phenotype (Paolantonio et al. 2002).

The small increase in the PD in the group observed in the present study should not be considered to be clinically relevant because all cases showed a healthy sulcus with no bleeding on probing after 12 months. There was a statistically significant gain in the RCAL in the two groups between the baseline and after 12 months. This is in accordance with the study performed by Cummings et al. (2005), which demonstrated histologically in humans a combination of long junctional epithelium and connective tissue adhesion after the use of the ADMGs for root coverage.

Because both surgical techniques included the adjacent teeth in the flap design, their clinical parameters were also assessed. It is important to emphasize that the results of this study showed that the two surgical techniques were not detrimental to the adjacent teeth and this is in accordance with the studies performed by Barros et al. (2004, 2005); instead, statistically significant improvements in GR reduction, a gain in RCAL, and an increase in TKT from baseline to the 12-month examination were found. The reduction in GR and the gain in RCAL are probably a result of the coronal displacement of the flaps (Barros et al. 2005), and we speculate that the increase in TKT in the adjacent teeth may be attributed to the space created between the flaps and the periosteum by the ADMGs, which was occupied by the blood clot.

In conclusion, both surgical techniques, which used the acellular dermal matrix allograft as a subepithelial graft, provided significant reduction in GR after 12 months, and there was no statistically significant difference between the groups regarding root coverage. Hence, both surgical techniques were found to be equally effective and suitable for root coverage procedures with the acellular dermal matrix allograft. However, there was a statistically significant difference between the surgical techniques after 12 months for the KT, favouring the group with releasing vertical incisions.

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Clinical Relevance

Scientific rationale for the study: Treatment of exposed root surfaces has become an important issue due to patients' increasing aesthetic demands. This study compared two surgical techniques, using the ADMG, to evaluate which could provide better root coverage. One technique included vertical incisions, while the other did not.

Principal findings: After 12 months, there was no statistically significant difference between the techniques regarding root coverage, and both provided a statistically significant reduction in gingival recession height.

Practical implications: Both surgical techniques were found to be equally effective and suitable for root coverage procedures with the acellular dermal matrix.