Journal of Clinical Periodontology
Official Journal of The European Federation of Periodontology
Founded by the British, Dutch, French, German, Scandinavian, and Swiss Societies of Periodontology

J Clin Periodontol 2009; 36: 868–876 doi: 10.1111/j.1600-051X.2009.01460.x

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Clinical evaluation of a new *collagen matrix* (Mucograft® prototype) to enhance the width of keratinized tissue in patients with fixed prosthetic restorations: a randomized prospective clinical trial

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Sanz M, Lorenzo R, Aranda JJ, Martin C, Orsini M. Clinical evaluation of a new collagen matrix (Mucograft® prototype) to enhance the width of keratinized tissue in patients with fixed prosthetic restorations: a randomized prospective clinical trial. J Clin Periodontol 2009; 36: 868–876. doi: 10.1111/j.1600-051X.2009.01460.x.

Abstract

Aim: The aim of this study was to test a new collagen matrix (CM) aimed to increase keratinized gingiva/mucosa when compared with the free connective tissue graft (CTG).

Material and Methods: This randomized longitudinal parallel controlled clinical trial studied 20 patients with at least one location with minimal keratinized tissue (⩽1 mm).

Main Outcome Measure: The 6-month width of keratinized tissue. As secondary outcomes, the aesthetic outlook, the maintenance of periodontal health and the patient morbidity were assessed pre-operatively at 1, 3 and 6 months.

Results: At 6 months, the CTG attained a mean width of keratinized tissue of 2.6 (0.9) mm, while the CM was 2.5 (0.9) mm, these differences being insignificant. In both groups, there was a marked contraction (60% and 67%, respectively) although the periodontal parameters were not affected. The CM group had a significantly lower patient morbidity (pain and medication intake) as well as reduced surgery time.

Conclusions: These results prove that this new CM was as effective and predictable as the CTG for attaining a band of keratinized tissue, but its use was associated with a significantly lower patient morbidity.

Key words: aesthetics; collagen; connective tissue graft; keratinized tissue

Accepted for publication 22 June 2009

The keratinized gingiva (KG) is a specialized mucosa covered with keratin or parakeratin that includes the free and the attached gingiva and extends from the gingival margin to the mucogingival junction. The width of the KG around teeth may vary between 1 and 9 mm depending on the subject's age and the jaw and tooth location and position. For decades, there was a controversy regarding the need for an ''adequate'' width of KG in order to preserve periodontal health, although a general clinical impression still remains that the presence of a certain width of keratinized tissue is important in maintaining periodontal health and preventing soft tissue recession. Some classic studies suggested a minimum of 2 mm of KG to maintain gingival health (Lang & Löe 1972), corresponding to 1 mm of attached gingiva. However, several clinical and experimental investigations

Conflict of interest and source of funding statement

The authors declare that there are no conflict of interests in this study.
This study was partially supported by a research grant from Geistlich Pharma.

have shown that the absence of attached keratinized tissue is compatible with the maintenance of periodontal health (Dorfman et al. 1980, Wennstrom et al. 1981, Wennstrom 1983, Wennstrom & Lindhe 1983, Kennedy et al. 1985). In a similar way, it has not been demonstrated that the presence of keratinized mucosa is a prognostic factor for the survival of dental implants (Adell et al. 1986).

In spite of this controversy, there are clinical situations, however, where a mucogingival surgical procedure may be considered, because a thin gingiva might be less resistant in the presence of inflammation or toothbrushing trauma and could lead to gingival recession. Several clinical studies have shown that this event may occur more likely in situations when the absence of keratinized tissue is combined with orthodontic treatment (Karring et al. 1971, Rateitschak et al. 1979, Ericsson & Lindhe 1984); when vestibular depth is needed in patients with removable partial dentures, and in patients with fixed prosthetic restorations, when margins are placed subgingivally (De Trey & Bernimoulin 1980, Hangorsky & Bissada 1980). A similar clinical behaviour has been reported in relation to the mucosa around dental implants. Experimental studies in monkeys have demonstrated that the lack of keratinized mucosa around implants increases plaque accumulation and the ensuing long-term effect of undisturbed plaque-derived inflammation caused bone loss around the implants in these animals (Warrer et al. 1995). Clinical studies in humans have shown similar results with regard to the significantly higher amounts of plaque and inflammation at implant sites without keratinized tissue, when compared with sites with adequate amounts, although its possible impact on the survival of the implants was not demonstrated (Chung et al. 2006). Recently, Zigdon & Machtei (2008), in a retrospective clinical study, have shown that both the peri-implant width and thickness of the mucosa had a significant negative correlation with the recession of the mucosa.

Many surgical techniques have been utilized to augment gingival tissue dimensions, the free gingival graft (Sullivan & Atkins 1969) being the most frequently used. Longitudinal studies have shown that procedures using pedicle and free gingival grafts are predictable and effective for providing newly created keratinized tissue up to 4 years (Dorfman et al. 1982). Histologically, heterotopic gingival and alveolar mucosal transplants in animal models have demonstrated that transplanted tissues always retained their original structure and specificity even after 1 year postoperatively (Karring et al. 1971). When using epithelialized grafts, healing, however, often results in compromised aesthetics (''patch-like area''). Alternatively, free connective grafts have been proposed (Edel 1998) with similar clinical predictability, but with better results in terms of aesthetics and colour matching (Roccuzzo et al. 2002, Orsini et al. 2004). Both techniques are, however, associated with significant patient morbidity due to the need to create a wound at the palatal donor site.

In order to avoid this patient morbidity, acellular dermal allografts have been used as a substitute for the palatal donor tissue, demonstrating the feasibility of using allograft materials (Park 2006, Yan et al. 2006, Imberman 2007). However, because this allograft material is derived from human cadavers, it is associated with ethical concerns and the possible risk of disease transmission. An alternative option, both avoiding the need for palatal donor tissue and allograft material, is the use of collagen membranes of porcine origin, which are already standard in oral wound-healing procedures in association with bone augmentation (Hämmerle et al. 2002, Wallace & Froum 2003). Recently, a new collagen matrix (CM) of similar porcine origin (Mucograft® Prototype) has been produced by Geistlich Pharma AG (Wolhusen, Switzerland). Even though its qualitative properties and safety have been evaluated according to the procedures established in ISO 14971 and ISO 10993-1, no clinical trial data are as yet available for this CM graft material. The objective of this clinical trial is, therefore, to test the efficiency of this new CM (Mucograft® Prototype) to build up a clinically sufficient width of newly formed KG and secondarily to assess the aesthetic outcomes and post-operatory morbidity, when compared with the standard treatment, the free connective tissue graft (CTG).

Material and Methods

Patients

Twenty patients were selected from those attending the Periodontal Postgraduate Clinic at the Faculty of Odontology in the University Complutense of Madrid, if they fulfilled the following criteria:

Inclusion criteria

- Older than 18 years and periodontally and systemically healthy.
- Presenting at least one location with minimal or no keratinized tissue ($\leqslant 1$ mm). The selected tooth/implant must be part of a fixed partial restoration.
- The patient should demonstrate good plaque control (FMPS $< 20\%$) and should be able to comply with all procedures related to the study.

Patients were excluded if they were heavy smokers (> 10 cigarettes per day), had any systemic disease that would negatively influence wound healing or known allergy to collagen.

The point of enrolment, following recruitment, was the time the subject signed the informed consent form. This happened after a full periodontal examination with registration of probing pocket depths (PPD), bleeding scores (FMBS) and plaque scores (FMPS), after checking whether the patient meets the inclusion criteria, and most importantly, after the patient has received thorough information from the investigator and has signed the ethics committee-approved informed consent form.

Experimental design

This study was designed as a longitudinal parallel-designed controlled clinical trial comparing the CM (Mucograft® Prototype) with the free CTG. The study protocol was approved by the Ethics Committee of San Carlos Clinical Hospital in Madrid and by the Spanish Ministry of Health who authorized this clinical trial.

Four weeks before enrolment, all patients received periodontal treatment consisting of oral hygiene instructions, a professional prophylaxis and, if needed, scaling and root planning. A new toothbrush was given to each patient to practice oral hygiene according to the given instructions. Four weeks after this initial treatment, all subjects underwent a full periodontal examination with recording of PPD and registration of FMBS and FMPS that enabled patients to be included in the study. For proper standardization between baseline and follow-up data, the probes were labelled

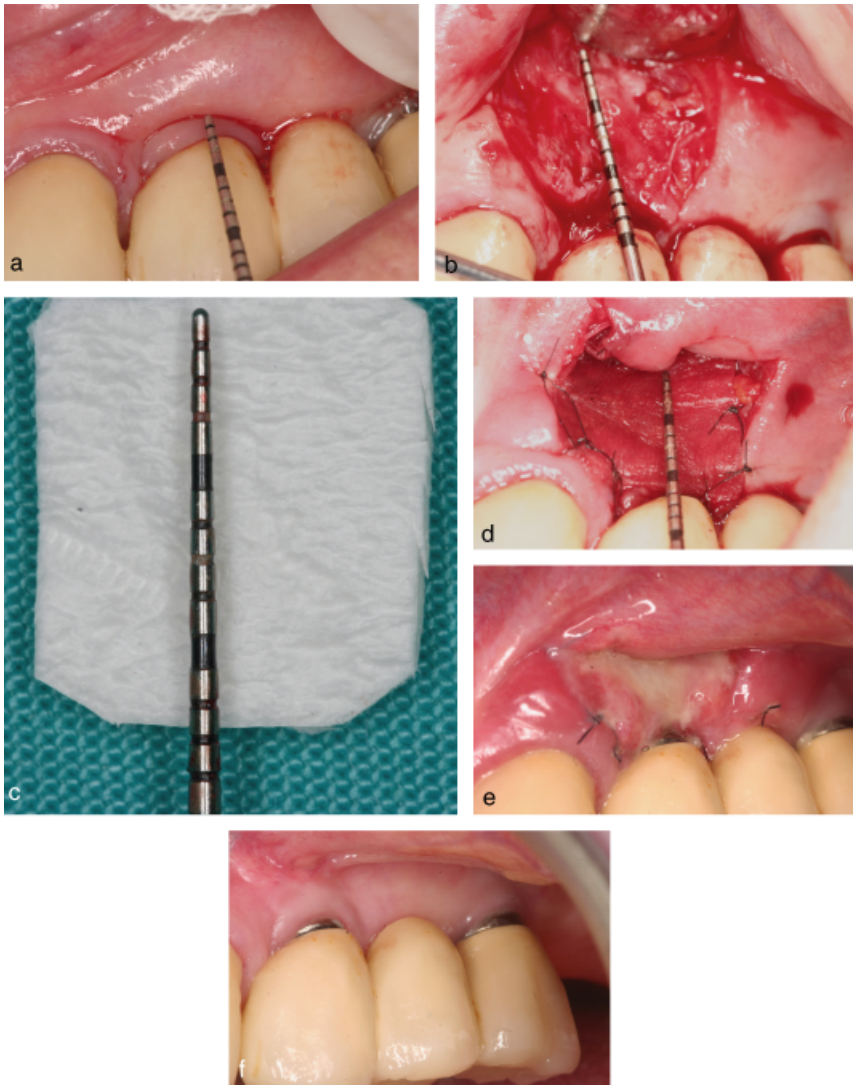

Fig. 1. (a) Pre-surgical image from an experimental site. Note the minimal amount of keratinized tissue around an implant supported restoration. (b) Split-thickness flap elevated to prepare the surgical bed for the Mucograft® in the experimental sites. (c) Experimental soft tissue substitute, a three-dimensional collagen tissue matrix (Mucograft® Prototype). (d) Experimental collagen matrix sutured on the prepared surgical bed. (e) Healing of the collagen matrix at 10 days post-surgery. (f) Presence of a band of keratinized gingival/mucosa (3 mm) at 6 months in the experimental site treated with the collagen matrix.

and the same probe was always used in the same patient for all measurements and the same examiner different from the surgeon took all the measurements (R. L.).

Surgical procedure

At the time of the surgery, the appropriate local anaesthesia was administered and recorded and the surgical procedure was performed as follows:

After patient selection, the control and test groups underwent a mucogingival surgical procedure in order to enlarge the area of keratinized tissue. All surgeries were performed by the same two calibrated surgeons (M. O. and J. J. A.). Immediately before the surgery, the following clinical parameters were recorded (Figs 1a and 2a):

- The width of the KG/mucosa measured from the free gingival margin to the mucogingival junction, using a North Carolina University probe.
- Periodontal indexes of the adjacent teeth [gingiva index (GI), plaque index (PI), PPD, and clinical attachment levels (CAL)].
- Clinical photographs of the surgery area to register baseline colour characteristics.

The surgical technique used consisted of the following steps:

Using a #15 blade, an intra-sulcular incision was made and a mucosal partial-thickness flap was raised. The recipient site was prepared by sharp dissection in order to create a periosteal bed free of any muscle attachment. The resulting flap was excised or sutured at the base of the newly created vestibule with 5-0 non-resorbable T-mattress braided nylon sutures (Figs 1b and 2b).

At this point, the treatment groups were assigned by means of sealed envelopes containing a code derived from a randomized list, to receive

- free CTG – control group (Fig. 2c).
- CM – test group (Fig. 1c).

In the control group, once the size of the graft was pre-determined using a tin foil stent prepared over the recipient site, a free CTG was harvested from the palate, following the classical procedure by Langer & Langer (1985). The CTG thickness varied depending on the patient's palate availability, but ranged between 1 and 3 mm. The CTG obtained was then sutured in the recipient bed with 5-0 non-resorbable braided nylon interrupted single sutures (Fig. 2d).

In the experimental group, the trimmed CM was measured with a probe and was then sutured in the recipient bed with 5-0 non-resorbable braided nylon interrupted single sutures (Fig. 1d).

The surgery time was recorded in both groups to the closest minute from the start of the first incision to the accomplishment of the last suture.

Patients were then instructed to rinse twice daily with a chlorhexidine mouth rinse (0.12%) for 2 weeks. Anti-inflammatory therapy (Ibuprofen 400 mg) was prescribed and patients were given instructions to take this drug in case of pain or swelling. The patients were prompted to record the dosage used in a customized form retrieved at the follow-up visits. Sutures were removed after 10 days and clinical photographs were taken to document the healing process (Figs 1e and 2e). All patients were asked to fill out a first pain questionnaire.

At 1, 3 and 6 months after the surgery, follow-up visits took place with examination of the gingiva or mucosal condition. Clinical photographs were taken and the following parameters were measured and recorded: widths of

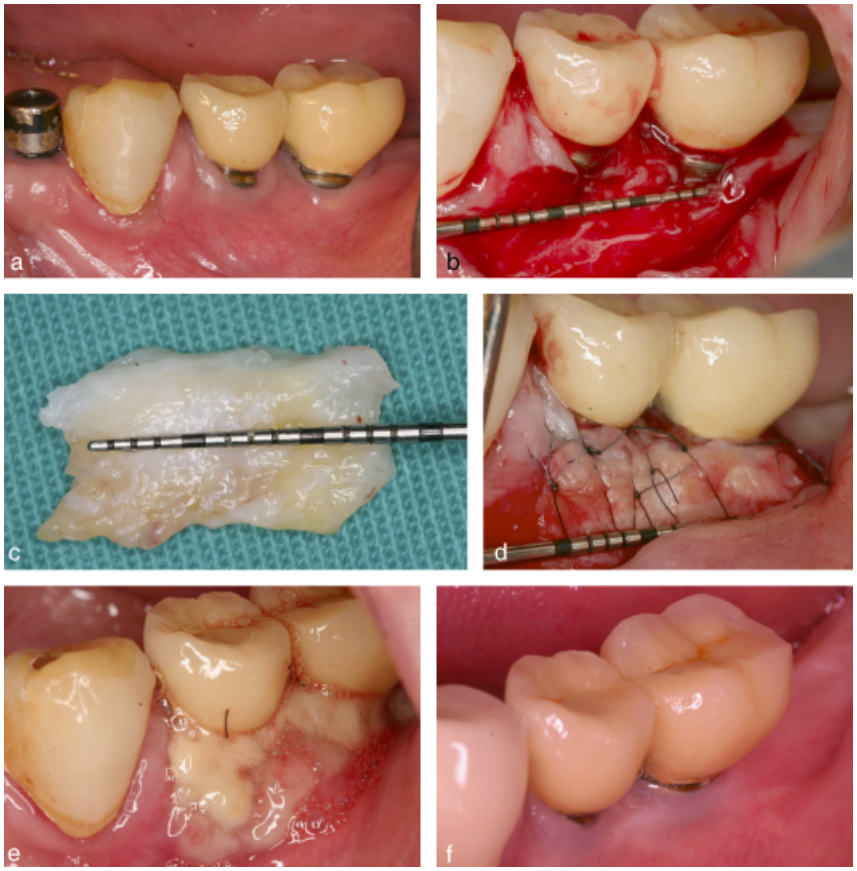

Fig. 2. (a) Pre-surgical image from a control site. Note the minimal amount of keratinized tissue around an implant supported restoration. (b) Split-thickness flap elevated to prepare the surgical bed for the connective tissue auto-graft in the control sites. (c) Dimension of the connective tissue graft retrieved from the patient's palate. (d) Connective tissue graft sutured on the prepared surgical bed. (e) Healing of the connective tissue graft at 10 days post-surgery. (f) Presence of a band of keratinized gingival/mucosa (2 mm) at 6 months in the control site treated with the connective tissue graft.

KG, PPD, CAL, GI and PI (Figs 1f and 2f).

During each of these visits, the patient filled out a pain questionnaire and returned the anti-inflammatory medication forms.

Experimental product information

The CM is a class III medical device according to the Medical Device Directive 93/42 (EEC definitions: 1.1. long-term implant; 1.2. implantable; 8: resorbable and 17: animal origin). It has not achieved CE labelling yet, as it is a new device with distinctive features.

The structure of the CM consists of two functional layers: a cell occlusive layer consisting of collagen fibres in a compact arrangement and a porous layer. Its general architecture is similar to the clinically available collagen membrane for bone regeneration, Bio-Gide® (Geistlich Pharma, Wolhusen, Switzerland); however, the CM porous layer is thicker in order to achieve more keratinized tissue by inducing a space-creating effect and by favouring blood clot formation (Fig. 1c).

Outcome measurements

An adequate width of the KG was selected as the primary endpoint of the study. The clinical evaluation of this outcome was performed by measuring the distance from the free gingival (mucosal) margin to the mucogingival junction, using a North Carolina University probe, pre-operatively and 1, 3 and 6 months after the treatment (Figs 1f and 2f).

As secondary endpoints, we selected the aesthetic outcome, the maintenance of periodontal (peri-implant) health in the affected teeth (implant) and the patient morbidity after the surgical procedure.

For the assessment of the periodontal and marginal health status, the following parameters were measured:

- graft size: size of the CTG or the cut CM in mm, measured using a periodontal probe.
- GI according to Loe and Silness.
- PI, according to Silness and Loe.
- PPD, measured using a periodontal probe in millimetres.
- CAL, measured using a periodontal probe in millimetres.

The periodontal indexes were measured before the surgery (baseline) and at the follow-up visits, 1, 3 and 6 months after the surgery.

The aesthetic outcome was assessed from standardized photos taken of the augmented sites during each visit, by judging the colour blending of the grafted site with the adjacent tissues through a qualitative questionnaire carried out by an independent examiner for the aesthetic evaluation.

The patient morbidity was assessed through a questionnaire filled out by the patient at each visit for pain assessment using a visual analogue scale (0–10). The investigator, using a specified form, recorded the presence of complications, additional treatments and medication in connection to the surgical treatment.

Data analysis

For the power analysis the values for the control (CTG) were taken from the test group of (Orsini et al. 2004), and in the test group it was assumed that a similar amount of increase in the width of keratinized tissue as well as graft shrinkage would be achieved. We thus assumed that 1 mm was the maximum, clinically non-relevant difference between both procedures. For non-inferiority of the CM with respect to the CTG, a sample size $n = 19$, was then estimated and taking into account a drop-out rate of 5%, a total of 20 patients were treated.

The study was monitored by an external agency in accordance with ISO 14155-1 and the source data obtained were verified for correctness and completeness before statistical analysis.

Descriptive statistics were generated for both the primary and the secondary outcome measurements as means, standard deviations and 95% confidence intervals. The normality of the distribution of these parameters was tested and

except for the variable recession, the rest proved not to be normally distributed and hence, non-parametric statistical tests were used for all comparisons. The intra-group comparisons across the different evaluation times were tested with the Friedman test, and if proven to be statistically significant, Dunn's Multiple Comparison Test was used to identify the statistical significance for the different intervals. The two-way analysis of variance for repeated measurements tested the inter-group comparisons across the different evaluation times. Mann–Whitney's non-parametric test was used for comparing the different variables between the control and the test groups at baseline and at 6 months. For all these comparisons we used a level of significance of 0.05.

Table 1. Patient description and clinical outcome measurements at baseline

	Baseline (mm)		
	group (CTG)	group (CM)	*p*
Patients			
Age (mean–range)	59.2 (39–62)	64.3 (57–79)	NS
Gender (female/male)	8/2	7/3	NS
Sites			
Tooth/implant	4/6	2/8	NS
Anterior/posterior	3/7	3/7	NS
Clinical outcome measurements			
Keratinized tissue	0.20 (0.42)	0.40 (0.52)	0.36 (NS)
Gingival index	0.1 (0.31)	0.30 (0.67)	0.54 (NS)
Probing pocket depth	2.0 (0.47)	2.12 (0.83)	0.68 (NS)
Recession	0.9 (0.87)	0.80 (1.45)	0.59 (NS)

NS, not significant; CTG, connective tissue graft; CM, collagen matrix.

Table 2. Primary outcome: increase in keratinized tissue

Patient #	Keratinized Tissue							
	group CTG				group CM			
	0 day	30 days	90 days	180 days	0 day	30 days	90 days	180 days
1	1	3	3	3	1	2	2	2
2	0	3	3	2	1	3	3	3
3	0	4	4	4	1	5	4	3
4	0	3	3	3	0	4	4	4
5	0	2	2	2	0	3	3	3
6	0	5	4	4	0	2	2	2
7	0	3	3	3	0	2	2	2
8	0	3	2	2	1	3	3	2
9	0	2	2	1	0	2	2	2
10	1	3	2	2	0	2	2	2
Mean	0.20	3.10	3.10	2.60	0.4	2.8	2.6	2.5
SD	0.42	0.87	0.87	0.96	0.52	1.03	0.7	0.7
95% CI	0.03–0.77	2.06–3.53	2.10–3.10	1.99–3.00	0.03–0.77	2.06–3.53	2.10–3.10	1.99–3.00

Differences between 0 and 30 days, 90 and 180 days were statistically significant in both groups.
Differences between 30 and 90 days, between 30 and 180 days and between 90 and 180 days were not statistically significant in any of the groups.
Differences between Group A and Group B were not statistically significant, both accross the study (two-way ANOVA analysis) or at any of the evaluation visits (Mann–Whitney analysis).
CTG, connective tissue graft; CM, collagen matrix.

Results

The study population consisted of 20 patients, 10 in the Control Group (CTG) and 10 in the Experimental Group (CM), recruited between September (2007) and February (2008). All patients fulfilled the protocol and attended all the follow-up visits. No patient in any of the groups developed any significant complication. In the control group (CTG), three patients showed partial necrosis of their CTGs at 10 days post-surgery; however, in no case was there a total loss of the graft. The rest of the patients in this group and all the patients in the experimental group (CM) healed uneventfully.

At baseline, both groups were well balanced with regard to the patient characteristics, location of selected sites and the clinical parameters assessed, demonstrating lack of significant gingival inflammation and the presence of shallow sulci (Table 1).

The changes in the primary outcome of this study (increase in keratinized tissue) are shown in Table 2. This table depicts the results for all the study patients. The mean width of keratinized tissue at baseline in the control and the test group was 0.2 (0.42) and 0.4 (0.51) mm, respectively. After the surgical procedure, there was a statistically significant increase in both groups at 30 days, being 3.1 (0.8) and 2.8 (1.0) mm, respectively, but the differences between groups were not statistically significant. Between day 30 (1 month) and day 180 (6 months), there was a contraction in the grafted area, although differences in the width of keratinized tissue were not statistically significant in any of the groups. At 6 months, Group CTG attained a mean width of keratinized tissue of 2.6 (0.9) mm, while the corresponding figure in Group CM was 2.5 (0.9) mm, this difference not being statistically significant (Fig. 3). The amount of graft contraction in both groups is shown in Table 3. There was a marked contraction in both groups between surgery and the 1-month evaluation (60% in group CTG and 67% in group CM). Between 30 and 180 days, this contraction continued in both groups although in a small percentage (17% and 8%, respectively) (Figs 1f and 2f).

The surgical procedure in both groups did not alter significantly the periodontal parameters in the affected teeth (implants). In Group CTG, the GI changed from 0.1 (0.3) at baseline to 0.3 (0.4) at 6 months. The corresponding figure in Group CM was 0.3 (0.6) and 0.2 (0.4), respectively. There were no statistically significant differences between the groups in any of the evaluation intervals. As can be seen in Fig.

Table 3. Mean graft contraction

	Day 0–30	Day 30–90	Day 90–180
CTG (%)	59.7	8.6	8.3
CM (%)	67.2	2	5.8

CTG, connective tissue graft; CM, collagen matrix.

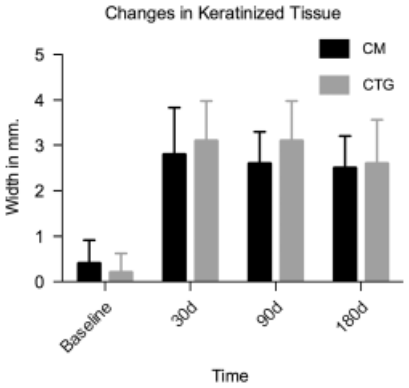

Fig. 3. Changes over time in the amount of keratinized tissue. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG).

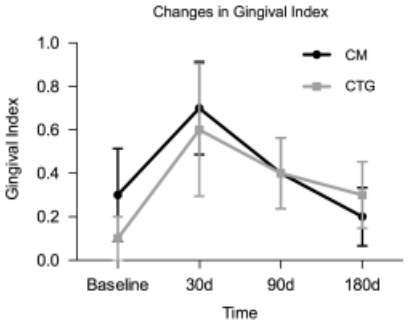

Fig. 4. Changes over time in the gingival index. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG).

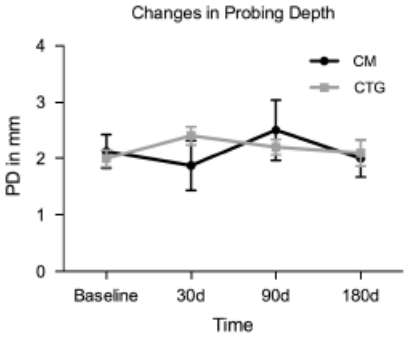

Fig. 5. Changes over time in the periodontal probing depths. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG).

4, between the surgical procedure and the one-month evaluation, there was an increase in gingival inflammation, similar in both groups, probably due to the post-surgical healing process. Similarly, the PPD in the affected teeth remained stable during the study in both groups. Figure 5 depicts these changes, without evidencing significant differences between the groups in any of the evaluation

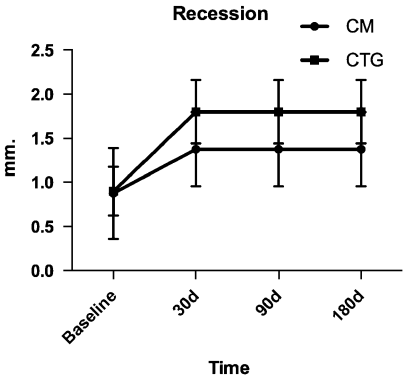

Fig. 6. Changes over time with regards to gingival recession. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG).

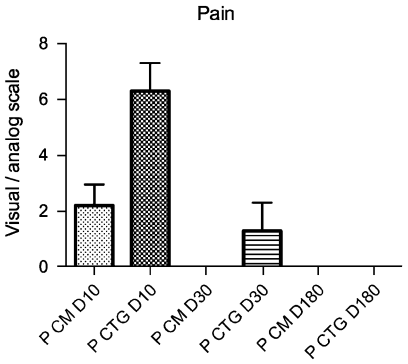

Fig. 7. Changes over time in patient's pain. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG).

periods. The changes in the position of the gingival (mucosal) margin (recession) are shown in Fig. 6. Although group CTG demonstrated a higher recession post-operatively, when compared with group CM, these differences were not statistically significant at 6 months.

The evaluation of the clinical photographs provided similar results in aesthetics and colour blending with the adjacent tissues in both groups. The blind evaluators were not able to distinguish between both procedures in terms of colour or aesthetic outlook.

In spite of the similar results obtained in both groups for the clinical parameters, statistically significant differences were detected between Groups CTG and CM for the outcome variables measuring the post-operative morbidity. The amount of pain referred by the patient was measured through a visual analogue scale, filled by each patient at the post-operative visits at 10 and 30 days. Figure 7 depicts these changes. At

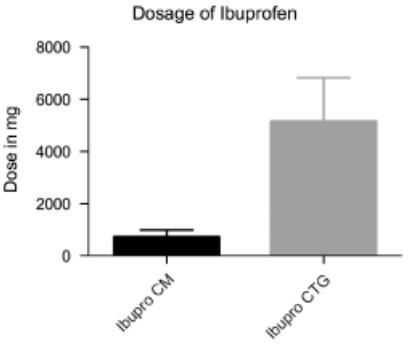

Fig. 8. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG) with regards to the amount of pain and anti-inflammatory medication taken (Ibuprofen®).

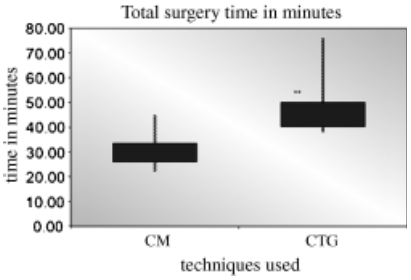

Fig. 9. Comparison between the collagen matrix (CM) and the connective tissue graft (CTG) with regards to the time needed to do the surgical procedure.

10 days patients in Group CTG had a mean pain score of 4.01 (8.5), while in the CM group the corresponding score was 2.30 (2.39), these differences being statistically significant ($p = 0.0002$). At 30 days, no patient in the CM group had any pain, while in the CTG group there were still patients suffering from pain, with a mean pain score of 1.30 (3.19). The amount of pain was also measured indirectly by the amount of pain and anti-inflammatory medication (Ibuprofen®) needed during the post-operative period by the patients. These data are depicted in Fig. 8. In the CTG group, the measured total dose at 10 days was 5140 (5336) mg, while the corresponding figure for group CM was 720 (860) mg, these differences being statistically significant ($p = 0.0013$).

The total surgery time spent in both surgical procedures was also different when both groups were compared. The CTG surgeries lasted a mean of 47.20 (10) min., while the CM surgeries lasted a mean of 30.80 (7) min. (Fig. 9). These differences were statistically significant ($p = 0.0006$).

Discussion

Although the controversy regarding the need for an ''adequate'' width of KG around teeth in order to preserve periodontal health still exists, there are clinical situations where the presence of a certain width of keratinized tissue may be important in maintaining periodontal health and preventing soft tissue recession, such as in areas around fixed prosthetic restorations, when margins are placed subgingivally. In fact, Orsini et al. (2004) showed in a 1-year longitudinal clinical study that the presence of a wide band of keratinized tissue favoured proper plaque control and reduced gingival inflammation in teeth abutments of fixed restorations. Augmentation of gingival keratinized tissue has been carried out traditionally using the free gingival graft. Rateitschak et al. (1979) evaluated this surgical technique in a 4-year longitudinal study. The transplanted grafts demonstrated an average shrinkage of 25%, while the gingival margin remained stable during the observation period. In order to avoid the patient morbidity problems associated with the donor site when using this graft technique, Edel (1998) demonstrated that the use of free CTGs could be a feasible alternative procedure, avoiding an open wound in the palate and attaining better colour matching with adjacent tissues. Using this surgical procedure, Orsini et al. (2004) reported a mean shrinkage of the graft of 40% at 1 year, attaining a wide band of KG (around 5 mm) and excellent colour blending with the surrounding gingiva.

There is also scarcity of information concerning the importance of the keratinized mucosa around dental implants and its effect on the peri-implant tissue health. An experimental study using implants in monkeys with minimal or no keratinized mucosa and plaque accumulation demonstrated significantly more recession and bone loss in these implants when compared with implants surrounded by keratinized mucosa (Warrer et al. 1995). These findings have been confirmed in humans (Block & Kent 1990, Brägger et al. 1997, Chung et al. 2006), demonstrating that the lack of keratinized mucosa around implants correlated with plaque accumulation and soft tissue inflammation. Moreover, recent studies have shown a positive correlation between lack or minimal amounts of keratinized mucosa and mucosa recession (Artzi et al. 2006, Chung et al. 2006, Zigdon & Machtei 2008), although a direct relationship with bone loss around implants has not been demonstrated (Adell et al. 1986).

The results from this investigation demonstrate a statistically significant amount of keratinized tissue achieved with both surgical procedures, the CTG and the CM, with mean widths of 2.60 and 2.50 mm, respectively. Although these results are more modest compared with those reported by Orsini et al. (2004) using the CTG, these differences may be due to the location, since in this study, most of the treated sites were posterior tooth/implant sites with shallow vestibules and high muscle attachments, which makes the establishment of a wide band of keratinized tissue difficult. In this study, most of the graft contraction occurred within the first month of healing, both with the CTG and the CM (60% and 67%, respectively). In fact, the differences between 1 month and 1 year were small in both groups (CTG 17% *versus* CM 8%). These findings are also in agreement with Orsini et al. (2004), who reported 37% contraction at 4 weeks and then 43% at 1 year. In spite of this significant increase in KT with both grafting materials, a small although visible recession was not prevented, evidencing the clinical margins of the restorations and implant abutments in the affected sites.

Although the clinical outcomes obtained with both surgical procedures in both studies were similar, there were significant differences in terms of patient morbidity, evaluated by the subjective patients pain perception and by the amount of pain and inflammatory medication needed. Most of this morbidity was associated with the need for a second surgical procedure in the palate to obtain the donor tissue in the CTG technique. In fact, other investigators have used other soft tissue substitutes in order to achieve a significant amount of KG or mucosa without the need for procuring a graft from the palate. One of these substitutes has been the use of an acellular dermal matrix (ADM) allograft. This allograft was originally intended for covering burn wounds. It is a structurally integrated basement membrane complex and extracellular matrix in which collagen bundles and elastic fibers are the main components. This allogenic graft will eventually degrade by the production of new connective tissue and will become completely replaced by host tissues (Wei et al. 2002). Several clinical studies have evaluated this allograft for its capability to increase the width of keratinized tissue around dental implants (Wei et al. 2000, Park 2006, Yan et al. 2006, Imberman 2007). Park (2006), in a prospective case series evaluating ADM to increase the width of keratinized mucosa around implants, obtained satisfactory results, with a mean increase of 2.2 mm at 6 months, although the contraction of the grafted area between 3 and 6 months was significant (58%). Wei et al. (2000) compared the clinical efficacy of ADM with CTG in achieving increased attached keratinized tissue around implants. Although there was a statistically significant increase in both groups, this gain in keratinized mucosa was significantly higher in the CTG when compared with ADM (5.5 *versus* 2.5 mm) and also the contraction associated with this allograft was substantial (71%). Moreover, the use of allografts derived from human cadavers may be associated with ethical concerns and the risk of disease transmission. To date, besides ADM, there are no other soft tissue substitutes available for this clinical indication and therefore, this study is the first clinical trial reporting the efficacy of a three-dimensional CM to increase the band of keratinized tissue. Its intended mechanism of action is by acting as a three-dimensional caffold that allows the in-growth and re-population of fibroblasts, blood vessels and epithelium from surrounding tissues, eventually being transformed into keratinized tissue. In this clinical investigation, this matrix has demonstrated a good healing pattern and clinical behaviour, attaining similar clinical outcomes in terms of increase of keratinized tissue, maintenance of the marginal tissue health and colour blending when compared with the standard CTG. Moreover, it has shown excellent handling properties with a significant reduction in surgery time and patient morbidity.

In conclusion, the results of this study prove that this new three-dimensional CM, when used as a soft tissue substitute aiming to increase the width of KG or mucosa, was as effective and predictable as the CTG, but its use was associated with a significantly lower patient morbidity.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Supporting information in accordance with the CONSORT Statement 2001 checklist used in reporting randomized trials.

Please note: Wiley-Blackwell is not responsible for the content or functionality of any supporting materials supplied by the authors. Any queries (other than missing material) should be directed to the corresponding author for the article.

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Clinical Relevance

Scientific rationale for the study: Although the importance of the presence of keratinized tissue is discussed controversially in the literature, most clinicians would seek a stable soft tissue around fixed restorations, free of inflammation. There are clinical situations, therefore, where there is an indication to increase the amount of keratinized tissue. The objective of this clinical trial is to test the efficiency of a new CM to build up a clinically sufficient width of newly formed KG when compared with one of the standard treatments: the placement of a free CTG.

Principal findings: This study has shown that a similar amount of KG/mucosa was achieved with the new CM. This outcome, however, was achieved with less post-operative morbidity and using less surgical time.

Practical implications: This new CM may be a useful soft tissue substitute in those clinical situations requiring an adequate amount of keratinized tissue.