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Coronally advanced flap adjunct with low intensity laser therapy: a randomized controlled clinical pilot study

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Abstract

Aim: Coronally advanced flap (CAF) technique and its modifications have been proposed in the literature. Low intensity laser therapy (LILT) is shown to increase wound healing. The aim of this split-mouth randomized controlled pilot study was to assess the effects of LILT with respect to root coverage after CAF procedure for the treatment of multiple-recession type defects (MRTD).

Material and Methods: Ten patients with symmetrical 74 Miller I and II gingival recessions were included in this study (37 in test, 37 in control group). A diode laser (588 nm) was applied to test sites before and immediately after surgery, and for 5 min. daily 7 days post-operatively. Comparisons of the surgical sites were made with clinical measurements.

Results: Statistically significant differences were observed between test and control sites in the gingival recession depth (GRD), gingival recession width (GRW) and width of the keratinized tissue (WKT) and clinical attachment level (CAL) measurements after 1 year ($p = 0.014$, $p = 0.015$, $p = 0.009$ and $p = 0.018$ respectively). The test group presented greater complete root coverage ($n = 7$, 70%) compared with the control group ($n = 3$, 30%) after treatment.

Conclusion: Within the limitations of this study, the results indicated that LILT may improve the predictability of CAF in multiple recessions.

Key words: aesthetic; coronally advanced flap; low intensity laser/therapy; Miller class I recessions; Miller class II recessions; root coverage

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Gingival recessions are commonly observed during periodontal practice, and surgical treatments of these defects are indicated when the recession results in root hypersensitivity and root caries or posses aesthetic concerns (Cairo et al. 2008, West 2008, Chambrone et al. 2010). The treatment of buccal gingival recession for aesthetics or root sensitivity is a frequent demand in patients with high oral hygiene standards. Various surgical procedures including pedicle flaps (envelope technique, coronally or laterally advanced flaps, double lateral bridging flap) (Bernimoulin et al. 1975, Caffesse & Espinel 1981, Raetzke 1985, Caffesse et al. 1987, Romanos et al. 1993, Harris 1996, Vergara & Caffesse 2004, Cairo et al. 2008, Zucchelli et al. 2009), free gingival grafts (Silva et al. 2010) and connective tissue grafts (CTG) (Raetzke 1985, Langer & Langer 1993, Vergara & Caffesse 2004, Moses et al. 2006, Rossberg et al. 2008, Nickles et al. 2010, Pini-Prato et al. 2010) are used with the aim of complete root coverage. Regardless of the surgical approach, the ultimate goal of a root-coverage procedure is the complete coverage of the recession defect and an optimal integration of the covering tissue with the adjacent soft tissue (Pini Prato et al. 1995, Cairo et al. 2008, 2009). Few case reports (Blanes & Allen 1999, Zabalegui et al. 1999), case series (Tinti & Parma-Benfenati 1996, Zucchelli & De Sanctis 2000, Cetiner et al. 2004,

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Carvalho et al. 2006) and some controlled and randomized (Zucchelli et al. 2009, Aroca et al. 2010) clinical trials have specifically addressed multiple adjacent gingival recessions showing complete root coverage in 35–90% of defects (Chambrone et al. 2009a, Pini-Prato et al. 2010).

Coronally advanced flap (CAF) technique is shown to be a predictable method for root coverage (Roccuzzo et al. 2002, Cairo et al. 2008) and it is a relatively easy procedure for the patient and the clinicians (Castellanos et al. 2006). Recent meta-analysis has shown that the percentage of root coverage with CAF varies between 34% and 86.67% (Cairo et al. 2008) Therefore, modifications that may increase the success and the predictability of the CAF technique, can possibly make this method the most preferred root-coverage procedure for patients and clinicians. Although CTGs when used together with CAF are shown to enhance the probability of complete root coverage (Matter 1999, Cairo et al. 2008, Chambrone et al. 2008), the need for a second surgical site, which may potentially cause post-operative pain and bleeding (Cortellini et al. 2009), can force the clinicians to perform less invasive techniques. Root conditioners (Cheng et al. 2007), enamel matrix derivatives (EMD) (Del Pizzo et al. 2005, Cheng et al. 2007), regenerative barrier membranes (Al-Hamdan et al. 2003), acellular dermal grafts (Gapski et al. 2005, De Queiroz Côrtes et al. 2006) and platelet rich plasma (PRP) (Aroca et al. 2009) are used together with CAF to promote healing and enhance clinical outcomes. As a new technique, Nd:YAG and Er:YAG lasers were used as root surface biomodifier for treatment of gingival recessions with subepithelial connective tissue grafts (SCTG) (Dilsiz et al. 2010a, b). The use of Nd:YAG laser as a root surface biomodifier negatively affected the outcome of root coverage with the SCTG (Dilsiz et al. 2010a), and the application of the Er:YAG laser for removing the smear layer from the root surfaces did not enhance the results when compared with SCTG alone (Dilsiz et al. 2010b).

Recently, low intensity laser therapy (LILT) has been used for getting better clinical results in periodontology. LILT was introduced by the work of Mester et al. 1971 who noted improvement in wound healing with the application of a low energy (1 J/cm^2) ruby laser. After these initial researches, various substrates have been used to create the lasers used for LILT. Initial research used lasers based on inert gases, including helium neon (HeNe: 632.8 nm), ruby (694 nm), argon (488 and 514 nm) and krypton (521, 530, 568, and 647 nm). Subsequent studies have used semiconductor laser diodes, including gallium arsenide (GaAs: 904 nm) and gallium aluminium arsenide (GaAlAs: 820 and 830 nm) devices. LILT is defined by several parameters such as power, wavelength, pulse rate, pulse duration, total irradiation time, intensity, interpulse interval and dose. Differences in the parameters used in various studies complicate the issue of making meaningful comparisons.

The basic principle of LILT is based on the biostimulation or biomodulation effect (Walsh et al. 1996, Walsh 1997, Damante et al. 2004), which consists of the fact that irradiation at a specific wavelength is able to alter cellular behaviour (Basford 1995, Hopkins et al. 2004, Posten et al. 2005). This effect is achieved by acting on the cellular mitochondrial respiratory chain (Silveria et al. 2007) or on membrane calcium channels (Alexandratou et al. 2002), which subsequently promotes an increase in cell metabolism and proliferation (Khadra et al. 2005a, b). In vitro and in vivo data suggest that LILT facilitates fibroblast and keratinocyte cell motility (Walsh et al. 1996, Yu et al. 1996), collagen synthesis (Pinheiro et al. 2005), angiogenesis and growth factor release (Tuby et al. 2006), which lead to increased wound healing (Hunter et al. 1984). In dentistry, LILT has been used for promoting wound healing and reducing pain after gingivectomy (Damante et al. 2004, Amorim et al. 2006), endodontic surgery (Kreisler et al. 2004), orthodontic treatment (Turhani et al. 2006) and as an adjunct after non-surgical (Kreisler et al. 2004, Qadri et al. 2005) and surgical (Ozcelik et al. 2008a, b) periodontal treatment. In addition, it has been also used for treatment of mucositis (Lara et al. 2007) and temporomandibular joint disorders (Venancio et al. 2005). Recently, it has been shown that LILT may improve the regenerative effects of EMD by reducing post-operative complications such as gingival recession (Ozcelik et al. 2008a). However, the basic mechanism on how LILT may affect periodontal wound healing still remains unknown and the use of LILT is still not widely accepted by the dental community due to the lack of sufficient number of controlled clinical trials.

With regard to these potential benefits in periodontal wound healing, the objective of this randomized, controlled, split-mouth clinical trial was to assess the effects of LILT on the clinical outcomes after CAF operations.

Material and Methods

Power analysis

Based on the study of Aroca et al. (2009), the means of GRD and gingival recession width (GRW) were determined as the expected primary outcomes of the study. GRW required bigger sample size and was used for the analysis. The sample size was calculated with an expected parameter estimate, assuming a mean of 3.7 GRW at baseline and a mean of 0.8 at 12th month with a 1.7 standard deviation. The minimum sample size for paired continuous data thus required 10 subjects per dependent groups within a 99% confidence and 80% power.

Patient and site selection

The sample of this split-mouth study was selected from patients who referred to the Department of Periodontology, Faculty of Dentistry, Cukurova University, between May 2008 and January 2009. Subject selection criteria were as follows: (i) the presence of at least two buccal adjacent Miller class I or II gingival recessions caused by traumatic toothbrushing on both sides of the maxillary or mandibular arch, (ii) No systemic diseases, (iii) No history of smoking and (iv) A full mouth plaque score 10% or less (Löe 1967). The study protocol was reviewed and approved by the institutional review board (IRB number: 2008-4-4) and informed written consent was obtained from all patients. In addition, patients received oral hygiene instructions to eliminate the wrong habits related to aetiology of the recession before the surgery. Six women and four men with a mean age of 34, who had a total of 74 recession type defects, were included in the study.

Clinical measurements

- The cemento-enamel junction (CEJ) was used as a reference point for the clinical measurements. In cases with unidentifiable CEJ, the margin of a restoration or a resin stent was used. The following clinical parameters were evaluated at baseline and post-operative 12 months.
- Gingival recession depth (GRD): defined as the distance from the CEJ to the free gingival margin (FGM) in millimetres mid-facially.
- Gingival recession width (GRW): defined as the width of recession at the most apical location of the CEJ in millimetres mid-facially.
- Probing depth (PD): defined as the distance from the FGM to the bottom of the sulcus in millimetres mid-facially.
- Probing clinical attachment level (CAL): defined as the distance from the CEJ to the end of the probe in millimetres mid-facially.
- Width of the keratinized tissue (WKT): defined as the distance from the gingival margin to the mucogingival junction (identified by Lugol staining) in millimetres.

The same-blinded examiner under took all of these measurements (SAD) with a UNC 15 manual probe. The calibration of the examiner was performed by the evaluation of all study parameters on two separate occasions on ten patients who were not enrolled in the study. Calibration was accepted if the measurements were similar at 90% level on these two examinations.

Clinical procedures and randomization

All patients received oral hygiene instructions and scaling and root planing at least 3 weeks before CAF procedures. In each patient, although one side of the jaw received conventional CAF (control sites) (37 defects), the opposite site received CAF + LILT (test sites) (37 defects). A total of 37 gingival recessions (29 Miller class I and eight Miller class II) treated with CAF consisted of six central incisors, four lateral incisors, 14 cuspids, eight-first pre-molars and 5 s pre-molars. For the test group; a total of 37 gingival recessions (28 Miller class I and nine Miller class II) consisted of six central incisors, four lateral incisors, 16 cuspids, seven-first pre- molars and 4 s pre-molars.

The same periodontist (S. O.) performed both operations (at test and control sites) during a single surgical session. Test and control sites were operated one-by-one. One site was operated until the suturing phase on which randomization concealment and laser application or simulation was performed by another staff (AD) by toss of a coin. In this way, the blindness of the operator was maintained and the operator finished the surgery by suturing after laser application or simulation by another staff. Immediately after this, the surgery of the other site was started and performed in the same manner.

Surgical procedure and LILT protocol

Both test sites and control sites underwent an identical CAF procedure, and surgical timing was similar for both sites. After local anaesthesia, root debridement and polishing of the exposed root surface was performed (Fig. 1a). No root conditioning was used. Two oblique, divergent beveled incisions extending beyond the mucogingival junction were made at the mesial and distal line angles of the most mesial and the most distal of teeth with gingival recessions. These incisions, together with the intrasulcular incisions along the mesial and distal recession margins, designed the external surgical papillae. Horizontal submarginal incisions, made interproximally at CEJ level, created the interdental surgical papillae. Then the intrasulcular incisions, which extended horizontally to the most distal and the most mesial of the involved teeth, were made, leaving the gingival margin of the non-affected adjacent teeth untouched. All surgical papillae were dissected, split-thickness, up to the probeable sulcular area, keeping the blade almost parallel to the root. The soft tissue apical to the root exposure (including the residual keratinized tissue) was elevated to full thickness by inserting a small periosteum elevator into the sulcus and proceeding in the apical direction to expose 3–4 mm of bone apical to the bone dehiscence. This was done to include the periosteum and the maximum soft-tissue thickness in the central portion of the flap covering the avascular root exposure. A gentle root debridement was performed by sharp curettes on the exposed root surfaces. The most apical portion of the flap was split-thickness to allow coronal repositioning of the flap without tension. To permit the coronal advancement of the flap, all mus-

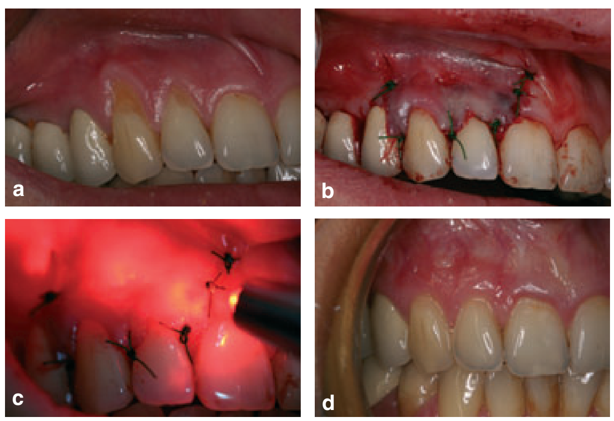

Fig. 1. Maxillary right lateral and canine, pre-surgical (a). Suturing of the flap in coronal position (b). Application of low intensity laser therapy immediately after sutures (c). Clinical results 1 year after surgery, significant increase in keratinized tissue is evident (d).

cle insertions present in the thickness of the flap were eliminated. This was done by keeping the blade parallel to the external mucosal surface. Coronal mobilization of the flap was considered adequate when the marginal portion of the flap was able to passively reach a level coronal to the CEJ of all teeth with the recession defects. The flap should be stable in its final coronal position even without the sutures. Once coronally advanced, the flap partially overlaid the soft-tissues mesial and distal to the receiving bed. These areas and the facial soft tissue of the anatomical interdental papillae were de-epithelialized to create connective tissue beds to which the surgical papillae and the peripherical portions of the CAF were sutured (Fig. 1d). A gentle root debridement was performed by sharp curettes on exposed root surfaces prior to suturing.

The laser irradiation protocol used in this study has been described by Ozcelik et al. (2008a, b). Briefly, before suturing (before flap closure), the exposed root surface and adjacent surgical area and the inner surface of the mobilized flap of the test sites were irradiated with low intensity laser for 5 min. Low intensity laser used in this study was a diode laser (ULOCKS, Russia) with a wavelength of 588 nm and output power of 120 mW and the power density for 5 min. was 4.0 J/cm^2, delivered by applying in continuous wave mode for 5 min. During irradiation, the tip of the laser probe was placed perpendicularly with slight contact on the area. The device used here was a safety class II laser. Safety goggles were worn by both patients and clinician during laser irradiation to avoid possible eye injury. For the control sites, the laser application was simulated, without pushing the start button. Then, the flaps were repositioned coronally and stabilized with 5.0 interrupted sutures on both test and control sites (Fig. 1b). LILT irradiation was repeated immediately after flap closure for the test sites for 5 min. (Fig. 1c). Pre-prepared customized impressions were used during the irradiation to standardize the irradiated area. The sutures were considered as the margins of the wound area and the laser was applied by slight contact with the tissue from the margins towards the centre of the wound in circular movements. The opposite control sites had laser simulation. No periodontal dressings were used. Post-operatively, LILT was repeated on the test sites 5 min. daily for 7 days as described, whereas the control sites had again laser simulation. The surgeon stayed blinded to the test and control sites both before and after flap closure. Entire laser energy used in each lased site was 4.0 J/cm^2 for before and after suturing and 28 J/cm^2 in the follow-up visits.

Post-operative maintenance

A protocol for the control of bacterial contamination consisting of doxycycline (100 mg bid for 1 week), 0.12% chlorhexidine mouth rinsing three times per day, and weekly prophylaxis was prescribed (Tonetti et al. 2002). Patients were requested not to chew rigorously and to avoid brushing and flossing in the treated area for a period of 2 weeks. Then, patients resumed full oral hygiene and they were placed on a 3-month recall system for 12 months (Fig. 1d).

Statistics

Non-parametric tests were chosen for continuous variables as the data were not distributed normally. Comparisons were done using the Mann–Whitney *U*-test between independent groups and using the Wilcoxon's rank sum test between the dependent groups. Measurements were compared among techniques-study groups using a mixed model analysis of variance to account the multiple measurements made on each subject. Subject was included as a random effect, and the study groups were included as fixed effects in the mixed linear model. Results were presented as mean ± SD. A *p*-value <0.05 considered as significant. Statistical analyses were performed using the statistical package SPSS v 16.0 (SPSS Inc., Chicago, IL, USA).

Results

All patients completed the study. No drop-outs occurred and no adverse events were reported during the follow-up period. Totally, 74 recession type defects were treated with CAF technique alone (control site 37 defects) or CAF + LILT (test site 37 defects). The treated teeth consisted of incisors (20), cuspids (30) and pre-molars (24).

Table 1 shows the baseline values and the changes in clinical parameters for the two groups, 1 year after surgical treatment. There were no statistically significant differences between groups at baseline. Statistically significant differences were observed between test and control sites in the GRD, GRW, WKT and CAL measurements after 1 year ($p = 0.014$, $p = 0.015$ $p = 0.009$ and $p = 0.018$ respectively). No statistical difference was observed between groups for PPD baseline and after 1 year (Table 1).

At baseline, the average depth of recession defects in CAF treated site was 3.03 ± 0.58, whereas the final gingival recession was 0.76 ± 0.46 ($p = 0.005$). In the CAF + LILT treated sites, the baseline gingival recession was 2.83 ± 0.64, whereas the final gingival recession was 0.26 ± 0.28 ($p = 0.005$) (Table 1).

Both treatment groups showed significant post-surgical improvement in the coverage of recession when compared with baseline for all parameters (GRW, GRD, WKT, PPD and CAL) (Fig. 2). However, there were no significant differences between groups according to mean differences between baseline and 1 year (Δ) for GRW; multivariate analyses revealed significant interaction between study groups for GRW ($F = 9.30$ $p = 0.003$). Significant interaction between study groups was also found for CAL ($F = 8.01$ $p = 0.006$). Multivariate analyses revealed no significant interaction between study groups for GRD ($F = 3.27$ $p = 0.225$), WKT ($F = 0.887$ $p = 0.348$) and PD ($F = 3.00$ $p = 0.086$).

The test group presented greater complete root coverage ($n = 7$, 70%) compared with the control group ($n = 3$, 30%) ($p < 0.07$).

Discussion

In this randomized controlled clinical trial, it was found that LILT could increase the predictability of CAF, with significantly higher percentages of complete root coverage at the post-operative first year. The success of periodontal plastic surgery mainly depends on patient-related (oral hygiene, traumatic tooth brush-

Table 1. The comparison of the clinical parameters of the CAF and the CAF + LILT-applied sites on the baseline and the post-operative 1 year

	Groups Mean ± SD		
	Control (CAF) n = 10	Test (Laser + CAF) n = 10	p**
Gingival recession depth			
Baseline	3.03 ± 0.58	2.83 ± 0.64	0.62
1year	0.76 ± 0.46	0.26 ± 0.28	0.014
Mean difference Δ	2.27 ± 0.48	2.57 ± 0.77	0.288
p*	0.005	0.005	–
Gingival recession width			
Baseline	2.610.27±	2.22 ± 0.48	0.053
1year	1.13 ± 0.57	0.63 ± 0.77	0.015
Mean difference Δ	1.48 ± 0.45	1.59 ± 1.15	0.36
p*	0.005	0.011	–
Amount of keratinized tissue			
Baseline	4.49 ± 0.31	4.51 ± 0.30	0.819
1year	4.71 ± 0.22	5.09 ± 0.38	0.009
Mean difference Δ	−0.22 ± 0.27	−0.58 ± 0.17	0.005
p*	0.034	0.005	–
Probing depth			
Baseline	1.89 ± 0.30	1.87 ± 0.29	0.638
1year	1.70 ± 0.33	1.53 ± 0.44	0.411
Mean difference Δ	0.20 ± 0.17	0.34 ± 0.31	0.277
p*	0.016	0.027	–
Clinical attachment level			
Baseline	4.92 ± 0.56	4.65 ± 0.50	0.412
1year	2.45 ± 0.50	1.83 ± 0.49	0.018
Mean difference Δ	2.47 ± 0.57	2.83 ± 0.85	0.362
p*	0.005	0.005	–
Complete root coverage at 1 year n (%)	3/10 (30.0)	7/10 (70.0)	0.07

Mean difference (Δ, Delta); the difference of the parameters between baseline and 6 months.
* > Wilcoxon test.
**Mann–Whitney U-test.
CAF, coronally advanced flap; LILT, low intensity laser therapy.

ing, smoking) (Silva et al. 2006), site-related (level of interdental bone, width and depth of the recession, flap thickness, post-operative gingival margin) (Berlucchi et al. 2005, Pini-Prato et al. 2005, Nieri et al. 2009) and clinician-related (experience) (Castellanos et al. 2006) factors. All of these factors subsequently affect post-operative wound healing, which relies on clotting, revascularization and maintenance of blood supply to the surgical area. It is possible that LILT application may have some immediate post-operative beneficial effects on these healing reactions.

The wound healing after mucogingival surgery mainly includes fibroblasts, keratinocytes and immune cells. In flap surgeries, immediately after suturing (up to 24 h), a connection between the flap and the tooth or bone surface is established by a blood clot, which consists of a fibrin reticulum with many polymorphonuclear leucocytes, erythrocytes, debris of injured cells, and capillaries at the edge of the wound (Cafesse & Ramfjord 1968). Previous studies suggest that LILT application may accelerate wound healing by increasing the motility of human keratinocytes and promoting early epithelization, by increasing fibroblast proliferation and matrix synthesis and by enhancing neovascularization. The major changes seen in wounds treated with LILT include increased granulation synthesis, enhanced neovascularization of tissue, increased fibroblast proliferation, maturation, attachment and matrix synthesis (Bisht et al. 1994, Mirsky et al. 2002). In addition, these biological effects of LILT may contribute to the higher tensile strengths of gingival flap margins and protection and stability of the granulation tissue and blood clot under the wound margins, which may subsequently prevent the collapse of healing wound, thus minimizing soft-tissue recession (Khadra et al. 2005a). These effects may also enhance the periodontal attachment gain.

Despite the listed beneficial effects of LILT, there are very few clinical studies about gingival surgery, which makes the comparison of our results impractical. Recently, it has been shown that LILT may increase the success of regenerative periodontal surgery by minimizing soft-tissue recession (Ozcelik et al. 2008a), which is in accordance with the results of the present study. In contrast, Almeida et al. (2009) has found no clinical improvement of LILT application after free gingival graft surgery. Research about application of LILT for periodontal use is complicated by discordance among the laser types used, the parameters selected and the subjects enrolled. To resolve questions regarding the possible benefits of LILT, several issues such as the establishment of the mechanisms underlying LILT's purported tissue effects and the type of the wound healing model need to be addressed. As wound healing is a temporal process, certain clinical studies are needed for LILT. The in vivo studies of LILT often attempt to quantify the surface area of an open wound and follow the change with time (Damante et al. 2004, Amorim et al. 2006, Ozcelik et al. 2008b). This, however, depends on intrinsic host factors, such as location and tension on the wound, as much as on the therapy itself. Ultimately, experiments need to be performed with large, randomized, double-blind, controlled samples to have convincing and reproducible results. The study design used in the present clinical trial was split-mouth; with two adjacent surgical sites in the same patient allowing each patient to have his/her own control. This structure allowed us the control of numerous factors, such as wound site, the age of the patient and the method of healing used. One of the two nearby wounds was randomized to receive LILT and the other had sham irradiation that allowed us to show the exact effects of LILT.

Fig. 2. The pre-operative and post-operative 1 year views of low intensity laser therapy applied site (a, b) and contralateral control site (c, d).

Complete root coverage was achieved in 70% of the gingival recession defects treated by the CAF + LILT and 30% of the gingival recession defects treated with the CAF. This result of CRC outcomes of the CAF + LILT is in accordance with the previous studies of CAF for single (De Sanctis & Zucchelli 2007) and multiple (89%) (Zucchelli & De Sanctis 2000, Zucchelli et al. 2009) gingival recessions in which similar surgical techniques were used. In addition, the result of the studies that include CAF + ADM (acellular dermal matrix) (Aichelmann-Reidy et al. 2001, Woodyard et al. 2004, Joly et al. 2007), CAF + PRP (Huang et al. 2005), CAF + EMD (enamel matrix derivative) (Modica et al. 2000, Del Pizzo et al. 2005, Spahr et al. 2005, Pilloni et al. 2006, Aroca et al. 2010), CAF + BM (barrier membrane) (Wang et al. 2001, Lins et al. 2003) and CAF + CTG (connective tissue greft) (Da Silva et al. 2004, Cortellini et al. 2009a) as a test root-coverage procedure were similar with the results of CAF + LILT technique used in this study.

Although significant improvements were achieved using LILT, this pilot study has a series of methodological limitations. First, the small sample size of the study may affect the reproducibility of the results and therefore these outcomes should be interpreted with caution. Other important limitations of the study include the lack of aesthetic assessment after surgery and the lack of patient outcomes in terms of satisfaction and discomfort. In addition, daily laser application for 7 days may be questionable in term of practicability for both patients and clinicians. Besides causing a possible increase in the final cost; the procedure takes a considerable amount of time that is required in addition to the time of surgery and may complicate the justification of the use of LILT with the relatively small additional benefits of this technique.

In this study, clinical measurements were taken to present the effects of LILT. Correlation of clinical effect with the mechanism could be best achieved through examination of biopsy samples of patients who were treated with LILT. These biopsies can be examined for evidence of collagen deposition, changes in proliferation of fibroblasts or macrophages or altered expression of cytokine factors, such as interleukins and growth factors. Therefore, further clinical, histological and/or immunohistological studies with larger study populations are required to evaluate the exact benefits of LILT on gingival healing and to correlate the clinical alterations with the findings at the cellular level.

The human body is vulnerable to the output of certain lasers, and under certain circumstances, exposure can result in damage to the eye and skin. To minimize the risk of laser accidents, especially those involving eye injuries, protective eyewear with appropriately filtering optics can protect the eyes from the reflected or scattered laser light, as well as from direct exposure to a laser beam. Eyewear must be selected for the specific type of laser, to block or attenuate in the appropriate wavelength range.

In conclusion, within the limitations of this study, the findings of this clinical pilot study have shown that the use of LILT may increase the success of CAF operations and result in more stable outcome.

References

Aichelmann-Reidy, M. E., Yukna, R. A., Evans, G. H., Nasr, H. F. & Mayer, E. T. (2001) Clinical evaluation of acellular allograft dermis for the treatment of human gingival recession. *Journal of Periodontology* **72**, 998–1005.

Alexandratou, E., Yova, D., Handris, P., Kletsas, D. & Loukas, S. (2002) Human fibroblast alterations induced by low power laser irradiation at the single cell level using confocal microscopy. *Photochemical and Photobiological Sciences* **1**, 547–552.

Al-Hamdan, K., Eber, R., Sarment, D., Kowalski, C. & Wang, H. L. (2003) Guided tissue regeneration-based root coverage: meta-analysis. *Journal of Periodontology* **74**, 1520–1533.

Almeida, A. L., Esper, L. A., Sbrana, M. C., Ribeiro, I. W. & Kaizer, R. O. (2009) Utilization of low-intensity laser during healing of free gingival grafts. *Photomedicine and Laser Surgery* **27**, 561–564.

Amorim, J. C., De Sousa, G. R., De Barros, S. L., Prates, R. A., Pinotti, M. & Ribeiro, M. S. (2006) Clinical study of the gingiva healing after gingivectomy and low-level laser therapy. *Photomedicine and Laser Surgery* **24**, 588–594.

Aroca, S., Keglevich, T., Barbieri, B., Gera, I. & Etienne, D. (2009) Clinical evaluation of a modified coronally advanced flap alone or in combination with a platelet-rich fibrin membrane for the treatment of adjacent multiple gingival recessions: a 6-month study. *Journal of Periodontology* **80**, 244–252.

Aroca, S., Keglevich, T., Nikolidakis, D., Gera, I., Nagy, K., Azzi, R. & Etienne, D. (2010) Treatment of class III multiple gingival recessions: a randomized-clinical trial. *Journal of Clinical Periodontology* **37**, 88–97.

Basford, J. R. (1995) Low intensity laser therapy: still not an established clinical tool. Review. *Lasers in Surgery and Medicine* **16**, 331–342.

Berlucchi, I., Francetti, L., Del Fabbro, M., Basso, M. & Weinstein, R. L. (2005) The influence of anatomical features on the outcome of gingival recessions treated with coronally advanced flap and enamel matrix derivative: a 1-year prospective study. *Journal of Periodontology* **76**, 899–907.

Bernimoulin, J. P., Lüscher, B. & Mühlemann, H. R. (1975) Coronally repositioned periodontal flap. Clinical evaluation after one year. *Journal of Clinical Periodontology* **2**, 1–13.

Bisht, D., Gupta, S. C., Misra, V., Mital, V. P. & Sharma, P. (1994) Effect of low intensity laser radiation on healing of open skin wounds in rats. *The Indian Journal of Medical Research* **100**, 43–46.

Blanes, R. J. & Allen, E. P. (1999) The bilateral pedicle flap-tunnel technique: a new approach to cover connective tissue grafts. *International Journal of Periodontics and Restorative Dentistry* **19**, 471–479.

Caffesse, R. G., Alspach, S. R., Morrison, E. C. & Burgett, F. G. (1987) Lateral sliding flaps with and without citric acid. *International Journal of Periodontics and Restorative Dentistry* **7**, 42–57.

Caffesse, R. G. & Espinel, M. C. (1981) Lateral sliding flap with a free gingival graft technique in the treatment of localized gingival recessions. *International Journal of Periodontics and Restorative Dentistry* **1**, 22–29.

Caffesse, R. G. & Ramfjord, S. P. (1968) Tissue reaction and periodontal healing in mucogingival and flap technics. *Revista de la Asociación Odontológica Argentina* **56**, 383–389.

Cairo, F., Pagliaro, U. & Nieri, M. (2008) Treatment of gingival recession with coronally advanced flap procedures: a systematic review. *Journal of Clinical Periodontology* **35**, 136–162.

Cairo, F., Rotundo, R., Miller, P. D. & Pini-Prato, G. P. (2009) Root coverage esthetic score: a system to evaluate the esthetic outcome of the treatment of gingival recession through evaluation of clinical cases. *Journal of Periodontology* **80**, 705–710.

Carvalho, P. F., Da Silva, R. C., Cury, P. R. & Joly, J. C. (2006) Modified coronally advanced flap associated with a subepithelial connective tissue graft for the treatment of adjacent multiple gingival recessions. *Journal of Periodontology* **77**, 1901–1906.

Castellanos, A., de la Rosa, M., de la Garza, M. & Caffesse, R. G. (2006) Enamel matrix derivative and coronal flaps to cover marginal tissue recessions. *Journal of Periodontology* **77**, 7–14.

Cetiner, D., Bodur, A. & Uraz, A. (2004) Expanded mesh connective tissue graft for the treatment of multiple gingival recessions. *Journal of Periodontology* **75**, 1167–1172.

Chambrone, L., Chambrone, D., Pustiglioni, F. E., Chambrone, L. A. & Lima, L. A. (2008) Can subepithelial connective tissue grafts be considered the gold standard procedure in the treatment of Miller Class I and II recession-type defects? *Journal of Dentistry* **36**, 659–671.

Chambrone, L., Lima, L. A., Pustiglioni, F. E. & Chambrone, L. A. (2009a) Systematic review of periodontal plastic surgery in the treatment of multiple recession-type defects. *Journal of the Canadian Dental Association* **75**, 203a–203f.

Chambrone, L., Sukekava, F., Araújo, M. G., Pustiglioni, F. E., Chambrone, L. A. & Lima, L. A. (2010) Root-coverage procedures for the treatment of localized recession-type defects: a Cochrane systematic review. *Journal of periodontology* **81**, 452–478.

Cheng, Y. F., Chen, J. W., Lin, S. J. & Lu, H. K. (2007) Is coronally positioned flap procedure adjunct with enamel matrix derivative or root conditioning a relevant predictor for achieving root coverage? A systematic review *Journal Periodontal Research* **42**, 474–485.

Cortellini, P., Tonetti, M., Baldi, C., Francetti, L., Rasperini, G., Rotundo, R., Nieri, M., Franceschi, D., Labriola, A. & Prato, G. P. (2009a) Does placement of a connective tissue graft improve the outcomes of coronally advanced flap for coverage of single gingival recessions in upper anterior teeth A multi-centre, randomized, double-blind, clinical trial. *Journal of Clinical Periodontology* **36**, 68–79.

Da Silva, R. C., Joly, J. C., De Lima, A. F. & Tatakis, D. N. (2004) Root coverage using the coronally positioned flap with or without a subepithelial connective tissue graft. *Journal of Periodontology* **75**, 413–419.

Damante, A. C., Greghi, S. L., Santana, A. C., Passanezi, E. & Taga, R. (2004) Histomorphometric study of the healing of human oral mucosa after gingivoplasty and low-level laser therapy. *Lasers in Surgery and Medicine* **35**, 377–384.

De Queiroz Côrtes, A., Sallum, A. W., Casati, M. Z., Nociti, F. H. Jr. & Sallum, E. A. (2006) A two-year prospective study of coronally positioned flap with or without acellular dermal matrix graft. *Journal of Clinical Periodontology* **33**, 683–689.

De Sanctis, M. & Zucchelli, G. (2007) Coronally advanced flap: a modified surgical approach for isolated recession-type defects: three-year results. *Journal of Clinical Periodontology* **34**, 262–268.

Del Pizzo, M, Zucchelli, G, Modica, F., Villa, R. & Debernardi, C. (2005) Coronally advanced flap with or without enamel matrix derivative for root coverage: a 2-year study. *Journal of Clinical Periodontology* **32**, 1181–1187.

Dilsiz, A., Aydin, T., Canakci, V. & Cicek, Y. (2010a) Root surface biomodification with Nd: YAG laser for the treatment of gingival reccesion with subepithelial connective tissue grafts. *Photomedicine and Laser Surgery* **28**, 337–343.

Dilsiz, A., Aydin, T. & Yavuz, M. S. (2010b) Root surface biomodification with Er:YAG laser for the treatment of gingival reccesion with subepithelial connective tissue grafts. *Photomedicine and Laser Surgery* **28**, 511–517.

Gapski, R., Parks, C. A. & Wang, H. L. (2005) Acellular dermal matrix for mucogingival surgery: a meta-analysis.. *Journal of Periodontology* **76**, 1814–1822.

Harris, R. J. (1996) Double pedicle flap – predictability and aesthetics using connective tissue. *Periodontology* **2000**, 39–48.

Hopkins, J. T., McLoda, T. A., Seegmiller, J. G. & David Baxter, G. (2004) Low-level laser therapy facilitates superficial wound healing in humans: a triple-blind, Sham-controlled study. *Journal of Athletic Training* **39**, 223–229.

Huang, L-H., Neiva, R. E. & Wang, H-L. (2005) Factors affecting the outcomes of coronally advanced flap root coverage procedure. *Journal of Periodontology* **76**, 1729–1734.

Hunter, J., Leonard, L. & Wilson, R. (1984) Effects of low energy laser on wound healing in a porcine model. *Lasers Surgery Medicine* **3**, 285–290.

Joly, J. C., Carvalho, A. M., Da Silva, R. C., Ciotti, D. L. & Cury, P. R. (2007) Root coverage in isolated gingival recessions using autograft versus allograft: a pilot study. *Journal of Periodontology* **78**, 1017–1022.

Khadra, M., Kasem, N., Lyngstadaas, S. P., Haanaes, H. R. & Mustafa, K. (2005a) Laser therapy accelerates initial attachment and subsequent behavior of human oral fibroblasts cultured on titanium implant material. A scanning electron microscope and histomorphometric analysis. *Clinical Oral Implants Research* **16**, 168–175.

Khadra, M., Lyngstadaas, S. P., Haanæs, H. R. & Mustafa, K. (2005b) Determining optimal dose of laser therapy for attachment and proliferation of human oral fibroblasts cultured on titanium implant material. *Journal of Biomedical Materials Research. Part A* **73**, 55–62.

Kreisler, M. B., Haj, H. A., Noroozi, N. & Willershausen, B. (2004) Efficacy of low level laser therapy in reducing postoperative pain after endodontic surgery a randomized double blind clinical study. *International Journal of Oral and Maxillofacial Surgery* **33**, 38–41.

Langer, L. & Langer, B. (1993) The subepithelial connective tissue graft for treatment of gingival recession. Review. *Dental Clinacs of North America* **37**, 243–264.

Lara, R. N., Da Guerra, E. N. & De Mola, N. S. (2007) Macroscopic and microscopic effects of GaAIAs diode laser and dexamethasone therapies on oral mucositis induced by fluorouracil in rats. *Oral Health and Preventive Dentistry* **5**, 63–71.

Lins, L. H., de Lima, A. F. & Sallum, A. W. (2003) Root coverage: comparison of coronally positioned flap with or without titanium-reinforced barrier membrane. *Journal of Periodontology* **74**, 168–174.

Löe, H. (1967) The gingival index, the plaque index and the retention index systems. *Journal of Periodontology* **38**, 610–616.

Matter, J. (1999) Free gingival grafts for the treatment of gingival recession. A review of some techniques. *Journal of Clinical Periodontology* **9**, 103–114.

Mester, E., Spiry, T., Szende, B. & Tota, J. G. (1971) Effect of laser rays on wound healing. *The American Journal of Surgery* **122**, 532–535.

Mirsky, N., Krispel, Y., Shoshany, Y., Maltz, L. & Oron, U. (2002) Promotion of angiogenesis by low energy laser irradiation. *Antioxidants and Redox Signaling* **4**, 785–790.

Modica, F., Del Pizzo, M., Roccuzzo, M. & Romagnoli, R. (2000) Coronally advanced flap for the treatment of buccal gingival recessions with and without enamel matrix derivative. A split-mouth study. *Journal of Periodontology* **71**, 1693–1698.

Moses, O., Artzi, Z., Sculean, A., Tal, H., Kozlovsky, A., Romanos, G. E. & Nemcovsky, C. E. (2006) Comparative study of two root coverage procedures: a 24-month follow-up multicenter study. *Journal of Periodontology* **77**, 195–202.

Nickles, K., Ratka-Krüger, P., Neukranz, E., Raetzke, P. & Eickholz, P. (2010) Ten-year results after connective tissue grafts and guided tissue regeneration for root coverage. *Journal of Periodontology* **81**, 827–836.

Nieri, M., Rotundo, R., Franceschi, D., Cairo, F., Cortellini, P. & Pini Prato, G. (2009) Factors affecting the outcome of the coronally advanced flap procedure: a Bayesian network analysis. *Journal of Periodontology* **80**, 405–410.

Ozcelik, O., Haytac, M. C., Kunin, A. & Seydaoglu, G. (2008b) Improved wound healing by low-level laser irradiation after gingivectomy operations: a controlled clinical pilot study. *Journal of Clinical Periodontology* **35**, 250–254.

Ozcelik, O., Haytac, M. C. & Seydaoglu, G. (2008a) Enamel matrix derivative and low-level laser therapy in the treatment of intra-bony defects: a randomized placebo-controlled clinical trial. *Journal of Clinical Periodontology* **35**, 147–156.

Pilloni, A., Paolantonio, M. & Camargo, P. M. (2006) Root coverage with coronally positioned flap used in combination with enamel matrix derivative: 18-month clinical evaluation. *Journal of Periodontology* **77**, 2031–2039.

Pinheiro, A. L., Pozza, D. H., Oliviera, M. G., Weissmann, R. & Ramalho, L. M. (2005) Polarized light (400–2000 nm) and non-ablative laser (685 nm): a description of the wound healing process using immunohistochemical analysis. *Photomedicine and Laser Surgery* **23**, 485–492.

Pini Prato, G. P., Clauser, C. & Cortellini, P (1995) Periodontal plastic and mucogingival surgery. *Periodontology* **2000**, 90–105.

Pini-Prato, G. P., Baldi, C., Nieri, M., Franseschi, D., Cortellini, P., Clauser, C., Rotundo, R. & Muzzi, L. (2005) Coronally advanced flap: the post-surgical position of the gingival margin is an important factor for achieving complete root coverage. *Journal of Periodontology* **76**, 713–722.

Pini-Prato, G. P., Cairo, F., Nieri, M., Franceschi, D., Rotundo, R. & Cortellini, P. (2010) Coronally advanced flap versus connective tissue graft in the treatment of multiple gingival recessions: a split-mouth study with a 5-year follow-up. *Journal of Clinical Periodontology* **37**, 644–650.

Posten, W., Wrone, D. A., Dover, J. S., Arndt, K. A., Silapunt, S. & Alam, M. (2005) Low-level laser therapy for wound healing: mechanism and efficacy. *Dermatologic Surgery* **31**, 334–340.

Qadri, T., Miranda, L., Tuner, J. & Gustafsson, A. (2005) The short-term effects of low-level lasers as adjunct therapy in the treatment of periodontal inflammation. *Journal of Clinical Periodontology* **32**, 714–719.

Raetzke, P. B. (1985) Covering localized areas of root exposure employing the "envelope" technique. *Journal of Periodontology* **56**, 397–402.

Roccuzzo, M., Bunino, M., Needleman, I. & Sanz, M. (2002) Periodontal plastic surgery for treatment of localized gingival recessions: a systematic review. *Journal of Clinical Periodontology* **29**, 178–194.

Romanos, G. E., Bernimoulin, J. P. & Marggraf, E. (1993) The double lateral bridging flap for coverage of denuded root surface: longitudinal study and clinical evaluation after 5 to 8 years. *Journal of Periodontology* **64**, 683–688.

Rossberg, M., Eickholz, P., Raetzke, P. & Ratka-Krüger, P. (2008) Long-term results of root coverage with connective tissue in the envelope technique: a report of 20 cases. *International Journal of Periodontics Restorative Dentistry* **28**, 19–27.

Silva, C. O., Ribeiro Eel, P., Sallum, A. W. & Tatakis, D. N. (2010) Free gingival grafts: graft shrinkage and donor-site healing in smokers and non-smokers. *Journal of Periodontogy* **81**, 692–701.

Silva, C. O., Sallum, A. W., de Lima, A. F. & Tatakis, D. N. (2006) Coronally positioned flap for root coverage: poorer outcomes in smokers. *Journal of Periodontology* **77**, 81–87.

Silveria, P. C., Streck, E. L. & Pinto, R. A. (2007) Evaluation of mitochondrial respiratory chain activity in wound healing by low-level laser therapy. *Journal of Photochemistry and Photobiology: B, Biology* **86**, 279–282.

Spahr, A., Haegewald, S., Tsoulfidou, F., Rompola, E. & Heijl, L. (2005) Coverage of Miller class I and II recession defects using enamel matrix proteins versus coronally advanced flap technique: a 2-year report. *Journal of Periodontology* **76**, 1871–1880.

Tinti, C. & Parma-Benfenati, S. (1996) The free rotated papilla autograft: a new bilaminar grafting procedure for the coverage of multiple shallow gingival recessions. *Journal of Periodontology* **67**, 1016–1024.

Tonetti, M. S., Lang, N. P., Cortellini, P., Suvan, J. E., Adriaens, P., Dubravec, D., Fonzar, A., Fourmousis, I., Mayfield, L., Rossi, R., Silvestri, M., Tiedemann, C., Topoll, H., Vangsted, T. & Wallkamm, B. (2002) Enamel matrix proteins in the regenerative therapy of deep intrabony defects. *Journal of Periodontology* **29**, 317–325.

Tuby, H., Maltz, L. & Oron, U. (2006) Modulations of VEGF and iNOS in the rat heart by low level laser therapy are associated with cardioprotection and enhanced angiogenesis. *Lasers in Surgery and Medicine* **38**, 682–688.

Turhani, D., Scheriau, M., Kapral, D., Benesch, T., Joke, E. & Bantleon, H. P. (2006) Pain relief by single low-level laser irradiation in orthodontic patients undergoing fixed appliance therapy. *American Journal of Orthodontics and Dentofacial Orthopedics* **130**, 371–377.

Venancio, R. A., Camparis, C. M. & Lizarelli, R. F. (2005) Low intensity laser therapy in the treatment of temporomandibular disorders: a double-blind study. *Journal of Oral Rehabilitation* **32**, 800–807.

Vergara, J. A. & Caffesse, R. G. (2004) Localized gingival recessions treated with the original envelope technique: a report of 50 consecutive patients. *Journal of Periodontogy* **75**, 1397–1403.

Walsh, L. J. (1997) The current status of low level laser therapy in dentistry. Part 1. Soft tissue applications. *Australian Dental Journal* **42**, 247–254.

Walsh, L. J., L'Estrange, P. R. & Seymour, G. J. (1996) High magnification in situ viewing of wound healing in oral mucosa. *Australian Dental Journal* **41**, 75–79.

Wang, H. L., Bunyaratavej, P., Labadie, M., Shyr, Y. & MacNeil, R. L. (2001) Comparison of 2 clinical techniques for treatment of gingival recession. *Journal of Periodontology* **72**, 1301–1311.

West, N. X. (2008) Dentine hypersensitivity: preventive and therapeutic approaches to treatment. *Periodontology* **2000**, 31–41.

Woodyard, J. G., Greenwell, H., Hill, M., Drisko, C., Iasella, J. M. & Scheetz, J. (2004) The clinical effect of acellular dermal matrix on gingival thickness and root coverage compared to coronally positioned flap alone. *Journal of periodontology* **75**, 44–56.

Yu, H. S., Chang, K. L., Yu, C. L., Chen, J. W. & Chen, G. S. (1996) Low-energy helium-neon laser irradiation stimulates interleukin-1 alpha and interleukin-8 release from cultured human keratinocytes. *Journal of Investigative Dermatology* **107**, 593–596.

Zabalegui, I., Sicilia, A., Cambra, J., Gil, J. & Sanz, M. (1999) Treatment of multiple adjacent gingival recessions with the tunnel subepithelial connective tissue graft: a clinical report. *International Journal of Periodontics and Restorative Dentistry* **19**, 199–206.

Zucchelli, G. & De Sanctis, M. (2000) Treatment of multiple recession-type defects in patients with esthetic demands. *Journal of Periodontology* **71**, 1506–1514.

Zucchelli, G., Mele, M., Mezzotint, C., Marzadori, M., Montebugnoli, L. & De Sanctus, M. (2009) Coronally advanced flap with and without vertical releasing incisions for the treatment of multiple gingival recessions: a comparative controlled randomized clinical trial. *Journal of Periodortology* **80**, 1083–1094.

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Clinical Relevance

Scientific rationale for the study: CAF is demonstrated to be effective in the treatment of MRTD and LILT has a bio-stimulatory effect, however, clinical data comparing CAF alone and CAF with LILT for the treatment of MRTD is currently not available.

Principal findings: It was found that LILT-applied sites had more favourable results after CAF operations compared with the control sites at the post-operative 1 year.

Practical implications: The result of this study may be of clinical relevance, in that LILT may be considered to be an adjunctive tool in the treatment of MRTD with CAF surgery.