Minimally invasive gingival phenotype modification in gingival recession associated with a non-carious cervical lesion using the root plastique technique (RPT). A quasi experimental one-group pretest-posttest study.

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Abstract:

Background: This study introduced the root plastique technique (RPT), the aim of which is to modify the gingival phenotype of sites with gingival recessions (GRs) associated with Non-Carious Cervical Lesions (NCCLs) prior to surgical treatment.

Methods: RPT was performed in 22 subjects with 53 RT1 A/B+ GRs. Changes in keratinized tissue thickness (KTT), keratinized tissue width (KTW), relative gingival recession (RGR), relative clinical attachment level (RCAL) and probing pocket depth (PPD) were measured at baseline (T0) and 2 months (T1) after the procedure was performed. All analyses were performed by means of hierarchical models.

Results: The study revealed statistically significant changes ($p<0.01$) in KTT (0.45 ± 0.04 mm), RGR (0.80 ± 0.13mm), KTW (0.67 ± 0.07mm), RCAL (-0.72 ± 0.16mm) and KTW (0.67 ± 0.07). No changes in PPD ($p>0.05$) were observed. Regression analyses of KTT increase and RGR reduction at T0 showed statistically significant correlation between the 2 variables ($p<0.05$). All the teeth with a KTT of <0.8mm at T0 (N=14) reached or surpassed this threshold at T1.

Conclusion: RPT increases KTT and KTW. In most of the sites, a reduction in GR was also achieved.

INTRODUCTION

Gingival recession (GR) is defined as the apical shift of the gingival margin with a consequent exposure of the root [1]. Mid-buccal GRs affect almost the entire population of the USA, with an approximate prevalence of 91% across the oral cavity and 70% when only the aesthetic zone is considered [2]. The attachment loss associated with GR may lead to an increased risk for root caries [3], tooth sensitivity, and compromised patient aesthetics [4, 5]. Multiple morphological and functional factors have been associated with the etiopathogenesis of GR including toothbrush-related trauma, inflammation induced by plaque accumulation, orthodontic tooth movement, and lack of keratinized tissue thickness (KTT) [6]. GRs are frequently concomitant with wedge-shaped, concave, flattened and irregular defects in the cervical area of the teeth known as non-carious cervical lesions (NCCLs) [6, 7]. The periodontal literature reports that the prevalence of NCCLs ranges from 11% to 62% in the general population [8, 9]. As a result of the combined defects with GRs, the probability of achieving complete root coverage following root coverage therapy is decreased [10]. The presence of an NCCL leads to alterations in both crown and root surface that often develop into root concavities (or steps) of variable extensions and depths, with the possibility of an indistinguishable cementoenamel junction (CEJ) [11]. Pini-Prato and coworkers classified the presence/absence of the CEJ as Class A (detectable CEJ) or Class B (undetectable CEJ), and the presence/absence of a cervical step at the apical extent of the NCCL into either the presence of a cervical step that is >0.5 mm (denoted as a +) or the absence of a cervical step (denoted as a -) [11]. As aforementioned, where an NCCL is present, surgical root coverage procedures possess a lesser chance of success [10]. In two randomized clinical trials investigating the surgical coverage of an NCCL, restorative materials in conjunction with a coronally advanced flap (CAF) with or without a subepithelial connective tissue graft (CTG) were used [12, 13]. The addition of a CTG demonstrated higher combined defect coverage (83%) compared with a CAF alone (43%), most likely due to the increased tissue thickness provided by the CTG [14]. However, multiple studies revealed that the use of a CTG could be avoided at sites with tissue thickness > 0.8 mm, where the use of a CAF alone is associated with similar clinical outcomes and superior aesthetics [8, 14, 15]. Hence, the aim of the present study is to present in a cohort of consecutively treated patients, a minimally invasive technique (root plastique technique; RPT) which aims to modify the gingival phenotype of sites with GRs associated with NCCLs prior to CAF.

MATERIALS AND METHODS

The one-group pretest-posttest design is a type of quasi-experiment study in which the outcome of interest is measured 2 times: once before and once after exposing a non-random group of participants to a certain intervention/treatment. This study was conceived as a proof of concept for a new treatment. While aware that a randomized controlled study would be needed to make a confirmatory claim, we realized that blinding the

examiner would be impossible in this case. Hence, we performed this exploratory trial to study the new treatment efficacy in detail by relying on the properties of its within-subject design, in which each patient is the control of himself. We designed this study specifying all the requirements needed for a future pivotal study. This investigation was conducted in agreement with the Helsinki Declaration of 1975 (World Medical Association, 1975) as revised in 2013 (World Medical Association, 2013). The protocol was approved by the Medical University of Tirana, Department of Dental Medicine, Institutional Review Board for Human Studies. Participants received an information sheet and provided their informed consent in accordance with the EU General Data Protection Regulation GDPR (UE) n. 2016/679 before beginning the rehabilitation. This prospective pilot study involved consecutive patients screened and treated in the time between February 2019 and October 2020.

Study population

The present study was conducted in combined defects (i.e., GR associated with an NCCL). The patients were selected according to the following eligibility criteria: 1) Presence of an ≥1 RT1 plus A/B+ NCCL combined defect; 2) Absence of caries and/or restorations in the area to be treated; 3) Absence of mucogingival surgery or surgical treatment for periodontal disease at the same site in the previous 12 months; 4) Full-mouth plaque score (FMPS) <20% and full-mouth bleeding score (FMBS) <20% [16]; and 5) Probing Pocket depth (PPD) of <4mm. Patients were excluded if: 1) they suffered any major systemic disease that may interfere with healing; 2) they were smokers or recreational drug users; 3) the defect was an RT1 GR without the presence of a gingival step 5) the GR was a class RT2 or RT3 defect; 6) there was evidence of trauma for occlusion in the area to be treated.

Clinical Assessments

Before data acquisition, each patient underwent a professional mechanical plaque removal session and individualized oral hygiene instructions. After this initial therapy, the following parameters were recorded:

1) Probing pocket depth (PPD): Assessed as the distance from the gingival margin to the apical end of the gingival sulcus.
2) Relative gingival recession (RGR): Measured as distance from the gingival margin to the incisal border of the tooth of the involved tooth [17].
3) Relative clinical attachment level (RCAL): Measured as distance from the apical end of the gingival sulcus to the incisal border of the involved tooth [17].
4) Relative Cervical Lesion Apical Step (RCLAS): Measured as distance from the lower margin of the NCCL to the incisal border of the involved tooth.
5) Non-carious cervical lesion depth (CLD): Distance between the deepest point on the facial wall of the NCCL and the coronal projection of the most external point of the apical border of the non-carious cervical lesion. This parameter was measured using an endodontic k file, a silicone stop fixed with a cyanoacrylate adhesive, and a digital caliper. [10]
6) Keratinized tissue width (KTW): Measured as the distance from the gingival margin to the mucogingival junction with a periodontal probe (University of North Carolina Probe).
7) KTT: Gingival thickness at baseline was measured 1.5 mm apical to the gingival margin using an endodontic k file inserted perpendicular to the tissue surface with a silicon stop over the gingival surface. The silicon disc stop was then placed in tight contact with the soft tissue surface and

fixed with a drop of cyanoacrylate adhesive, and a digital caliper used to measure the distance between the tip of the file and the stopper. [14]

Figure 1 schematically depicts the measurements performed in the present study. All measurements were performed at the time of the minimally invasive therapy (T0) under 5x magnification loops. All parameters were measured at baseline. PPD, RGR, RCAL, KTW and KTT were also measured at 2 months after the procedure was performed (T1) by a single examiner (LR). Prior to the beginning of the clinical study, the examiner (LR) measured the RGR and RCAL of all patients twice within 24 hours, with at least 60 min between examinations. The Kappa index was calculated for PD and KKT, resulting in 94% and 97% of reproducibility, and intraclass correlation was calculated for RGR, resulting in 92% of agreement [10].

Minimally invasive procedure

The technique performed is simple and fast, resonating the canons of minimally invasive therapies. After the administration of local anesthesia, the apical step of the NCCL (Fig. 2A) was eliminated using rotary burs mounted on a low-speed handpiece. The first cutter used was a rugby ball-shaped cutter with a diameter of 0.16 white ring, ultra-fine grain with a maximum grain size of 14 microns (Fig. 2B). The bur was inserted into the sulcus under abundant irrigation, until it invades the supracrestal tissue attachment (i.e., biological width). This was achieved by calculating the measurement of RBL and PPD. Eliminating the apical step was done in such a way to flatten the NCCL only in the apical portion. Namely, the coronal projection of the most external point of the apical border of the NCCL will be removed, reaching with the bur the bottom of the NCCL. In this way, the depth of the NCCL was eliminated (Fig. 2C). Subsequently, a final preparation drill with a diameter of 012 white ring, ultrafine grain with a maximum grain size of 14 microns was used (Fig. 2D). The bone crest was reached using the bur gently while checking the working depth with the probe. An ultra-fine finishing bur was used to ensure minimal cutting on the expense of the soft tissue and maximum smoothness of the root surface.

The use of this drill made it possible to refine and extend the preparation of the root from the mesial line angle to the distal line angle of the tooth affected (Fig. 2E). After the passage of the diamond burs, a flame Arkansas bur was used to refine and polish the root surface. The whole procedure was performed with the aid of a 5X magnifying lens. Through the invasion of supracrestal tissue attachment, the stimulation of bleeding was obtained and never required the use of sutures. This procedure was performed to eliminate any obstacle to the ascent of the gingival tissue and, at the same time, to create the space needed for the thickening of the soft tissues, a space that was initially filled only by the blood clot (Fig.2F). It was recommended that patients adopt a soft and cold diet following the procedure, avoiding biting any food with the treated teeth. All

patients were instructed to stop brushing the site and the two contiguous teeth (mesial and distal) for 30 days to avoid mechanical trauma to the tissues. The area was cleaned with chlorhexidine gel 2 times a day instead. The patient was reassured that the brownish discoloration of the tooth surface was extrinsic and reversible after a professional oral hygiene session. After 15 and 30 days the patient was re-evaluated (professional plaque control and oral hygiene instruction). After 2 months, all patients were examined, and clinical parameters reassessed (Fig. 2G-2H). A professional oral hygiene session was also performed. At that time, all the patients reinstated brushing in the affected area using an ultra-soft toothbrush for two weeks, then a soft toothbrush for another month.

Sample-size

According to our insight of the biological phenomenon, the population SD of the GT Gain in the most well-known treatments lies somewhere in between the 0.09 estimate of the CAF treatment[18], the one having the smaller probabilities of a substantial change, and the 0.47 of CTG treatment[19], which shows a greater variance probably because of the biological material addition. Even though an intermediate value seemed the most likely, the upper bound of this range was entered for the sample-size calculation, to warrant a sufficient power for the worst-case scenario. The minimal clinically important difference to detect was deemed 0.3 mm since it appeared the convenient size to bring cases with baseline thin phenotype around 0.5 mm up to the 0.8 mm threshold individuated by Cairo[14] as guarantee of a better prognosis against recession relapses. A sample-size of N=22 patients was calculated for a single-level analysis, setting alpha=0.05 and beta=0.2. Assuming a mean cluster-size=2.5, 22 patients ensure to the highest level of the mixed model an effective sample-size equivalent to 26 [20].

Statistical Analyses

All analyses were performed by means of Mixed Effect Models. A model with maximal random effects structure [21] entailed a time effect with two repeated measurements within tooth site at the first level, tooth site within patient at the second level and the patient at the third level. After the selection procedure, only an intercept per tooth site and an intercept and a time slope, constrained to be equal, per patient were added as random effects since any other parameter would result in over-specified models. For most regressions, the Time factor was the only included fixed effect. Additional predictors as age, sex and tooth type most often resulted in not significant coefficients, so they were dropped out from the relative models. Only for some response variables the tooth type main effect and time by tooth type interaction proved to be significant, so were included. The time by tooth type interaction, although not significant, was included in models aimed to

get more realistic effect predictions having the pre-post change as response and baseline values as covariate. The R 4.0.1 software package was used.

A power analysis by simulations (using the R package simr) was planned to know the range of possible effect-size with power equal or greater than 80% and the influence of patients' number on power.

RESULTS

Twenty-two subjects (12 males and 10 females) aged from 26 to 66 years (mean age of 45 years) were consecutively included in the present study. A total of 53 recessions were treated since each patient contributed with 1 to 4 recessions. Overall, 5 incisors, 17 canines, 23 premolars and 8 molars were included in the study. All the subjects healed uneventfully and none of them were excluded from the study.

The Power Analysis showed the study had power greater than 80% in detecting an effect right as small as 0.13mm and greater than 99% for effects larger than 0.25mm (see Figure S1 in online Journal of Periodontology)

Keratinized tissue thickness (KTT):

Overall, 98,5% of the sites exhibited KTT gain during the follow-up period. Statistically significant difference ($p<0.01$) was found from T0 to T1 for the KTT with a mean difference of 0.45 ± 0.04 mm (Table 1). Changes in the KTT by patient and recession are shown in Figure 3A. Tooth type, sex and age were not related to an increase in KTT from T0 to T1 ($p>0.05$). All the teeth with a KTT of <0.8mm at T0 (N=14) reached this threshold at T1 (Figure 3B) and 12 out of 14 (85.7%) passed this threshold. Regression analyses of increased tissue thickness by CLD at T0 showed statistically significant correlation between the 2 variables ($p<0.001$) (Figure 4). Furthermore, a statistically significant correlation was found between CLD depth and patient age ($p<0.01$) (see Figure S2 in online Journal of Periodontology).

Relative gingival recession (RGR):

Statistically significant difference ($p<0.01$) was found from T0 to T1 for the RGR reduction with a mean difference of 0.80 ± 0.132 (Table 1). Changes in the RGR by patient and recession are shown in Figure 5. Overall, 81% of the sites (n=43) had a RGR reduction, 15% (n=8) had no reduction and 3.8% (n=2) had an increase in the recession. Tooth type was related to a decrease of RGR ($p<0.001$) while age and sex were not ($p>0.05$). The mean RGR reduction was 0.51mm for the incisors, 1mm for the canines and 0.70 and 0.74mm for premolars and molars respectively (see Figure S3 in online Journal of Periodontology). Statistically significant difference ($p<0.01$) was found between canines-premolars, canine-molars and molars-incisors ($p<0.05$). A

strong trend was shown between premolar and incisors (p=0.056). Regression analyses of RGR reduction by CLD at T0 showed statistically significant correlation between the 2 variables (p<0,05) (Figure 5).

Keratinized tissue width (KTW):

Statistically significant difference (p<0.01) was found from T0 to T1 for the KTW increase with a mean difference of 0.67 ± 0.07 (Table 1). Changes in the KTW by patient and recession are shown in (see Figure S4 in online Journal of Periodontology). Tooth type, sex and age were not related to an increase in KTW (p>0.05) from T0 to T1. Linear regression analysis showed a correlation between the RGR reduction and increase of KTW (see Figure S5 in online Journal of Periodontology). None of the sites had a KTW of 0 at T0. Post-hoc Subgroup-analyses comparing changes in RGR, KTW and KTT according to the KTW at T0 (<2mm vs. ≥2mm) reported no statistically significant difference in any of the parameters (see Figure S6 in online Journal of Periodontology)

Relative clinical attachment level (RCAL):

Statistically significant difference (p<0.01) was found from T0 to T1 for the increase of RCAL with a mean difference of -0.72 ± 0.16 (Table 1). Changes in the RCAL by patient and recession are shown in (see Figure S7 in online Journal of Periodontology). Tooth type was related to a decrease of RGR (p<0.001) while age and sex were not (p>0.05). The mean RCAL increase was 0.59mm for the incisors, 1mm for the canines and 0.45 and 0.71mm for premolars and molars respectively (see Figure S8 in online Journal of Periodontology). Statistically significant difference (p<0.01) was found between canines-premolars and canine-molars (p<0.01).

Probing Pocket Depth (PPD):

No statistically significant difference (p>0.05) was found from T0 to T1 for the KTW increase with a mean difference of 0.23 ± 0.09 (Table 1). Changes in the PPD by patient and recession are shown in (Figure S9 in online Journal of Periodontology). Tooth type, sex and age were not related to an increase in PPD (p>0.05).

DISCUSSION

The present study evaluates whether the newly presented RPT led to an increased gingival phenotype (with a primary focus on the tissue thickness) after 2 months of follow-up. The result of the study clearly shows how this technique enables gingival thickness augmentation by diminishing root convexity apical to the NCCL. This provides a concave space for the formation and stability of a blood clot that will eventually lead to increased tissue thickness, and in most cases, partial reduction of the GR and increase in KTW. The described technique

has a similar biological foundation as the biologically oriented preparation technique (BOPT) previously introduced by Loi and Di Felice [22]. The BOPT is a protocol in which vertical (featheredge) tooth preparation in a flapless approach is performed, which eliminates the anatomical emergence profile of the crown corresponding to the CEJ. This allows the contiguous soft tissue to adjust its form and location to host the new prosthetic profile, thereby directing the healing, reattachment, and gingival thickness augmentation. A similar procedure and clinical concept has also been utilized in implant therapy by employing abutments with a shoulder-less design. The IBOPT abutments do not have a finish line, and it is the buccal gingival margin of the crown that creates the soft tissue form. The reduced buccal width of the abutment provides additional space to the gingival thickness thus promoting stability [23].

The first observation that can be extrapolated from the present study is that 98,5% of the sites exhibited KTT gain during the follow-up period with an average gain of 0.5 mm. Of high clinical relevance is that 85.7% of the recession with an initial KTT of <0.8 mm overcame this threshold during the follow-up period, while the remaining teeth reached exactly 0.8 mm. This possibly means that the current minimally invasive approach may facilitate root coverage procedures with promising results without the need of using a collagen matrix which would increase the cost of the procedure[24] or harvesting a CTG. Indeed, studies showed that CAF alone was associated with similar clinical outcomes and better final aesthetics compared with CAF + CTG at sites with tissue thickness ≥ 0.8 mm [14]. On the other hand, it should be said that the patient must undergo two different procedures (RPT first and CAF after). Another important consideration is that none of the sites presented with a KTW of 0 at T0. Hence, we could not examine if in complete absence of KTW, the clot formation after the procedure would have the ability to create keratinized gingiva. However, when we divided the sites with lowest KTW values (<2mm) and compared their performance (changes in RGR, KTW and KTT) with those with ≥2 mm, no difference was found, although these p-values>0.05 are not confirmatory results. As expected, REC reduction was observed only in the presence of KTW gain, whereas no reduction occurred when KTW did not show an increase. This finding is in accordance with the well-known concept of muco-gingival junction position stability over time [25].

Despite the primary goal of the RPT being to increase tissue thickness, most of the treated teeth displayed partial root coverage (81% of the sites with a mean root coverage of 0.8 mm). Another noteworthy observation is the statistically significant association noted between CLD and RGR. A biological explanation of this event is that in plastic surgery the convexity of the surface of the root may negatively impact the root coverage procedure. In this study, a higher CLD corresponded to a greater portion of root structure removed. Hence, the presence of a deep cervical lesion will lead to the creation of a greater space for the clot to form and, therefore, an increase in KTT and reduction in recession depth. The same concept could explain the higher mean root coverage achieved in the canine area, since the entire labial surface of this tooth is

markedly convex. Furthermore, the reported association between increased CLD and older age is a common finding in that the prevalence and severity of NCCLs appear to increase with age [8, 9, 26].

The present study is not exempt from limitations. Clinically, removal of root structure may lead to increased postoperative sensitivity. We did not collect the patient reported outcomes with a visual analog scale, but none of the patients included in the trial complained of excessive sensitivity. Since the RPT would serve as a preparatory (see Figure S10 A-E in online Journal of Periodontology10 A-E) procedure to be followed by CAF (see Figure S10 F in online Journal of Periodontology), sensitivity is expected to diminish after the surgical procedure. Moreover, the technique utilized here is essentially a flapless technique, which means it is technique sensitive and may involve a learning curve to attain the presented results. In this regard, it may be useful to consider future use of single-sided diamond tips mounted on piezo surgical handpieces to counterweight the technique sensitivity. Furthermore, RPT followed by CAF should be compared to CAF plus connective tissue graft (as the current gold standard therapy). This comparison would elaborate whether the two techniques achieve similar clinical results, with the advantage for the RPT of avoiding the need to harvest an autogenous graft when a GR defect is associated with an NCCL.

From the methodological perspective, five validity threats are usually indicated due to their potential to impact the one-sample pretest-posttest design. *History* is an implausible rival hypothesis since the trial was conducted under experimental conditions. *Maturation* would be in the opposite direction (decreased tissue thickness) compared to treatment [27]. *Patient selection* not representative of the population might have biased the size of the effect. With a standard deviation of 0.2 and a ρ=0.07 as in our sample, and with an assumed population averages ranging between 0.5 and 1.4 mm [27, 28], in a hypothetical scenario where all patients have baseline values below the cutoff of 0.9 mm, the bias due to the regression to the mean effect would not exceed 0.15 mm. This would guarantee an average treatment effect not smaller than 0.3 mm [29, 30] (see Figure S11 in online Journal of Periodontology). *Placebo effects* (when neither patients nor examinator are blinded) may be a cause of overoptimistic results; if any, this bias is expected to be bounded in the 0.1 mm rounding range.

CONCLUSION:

Within the limitations of the present study, it can be concluded that the RPT can increase keratinized tissue thickness and width. Moreover, in most of the teeth, an improvement in gingival recession was achieved. This procedure is especially recommended in patients with thin phenotype to reduce the need of using a collagen matrix or harvesting a CTG. Increased depth of the NCCL was related to better clinical outcomes after the RPT procedure.

Footnotes

¶: R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.]

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Table 1: Clinical variables change from T0 to T1

Parameter	Time 0 Mean ± SE (95% CI)	Time 1 Mean ± SE (95% CI)	Difference T1-T0 Mean ± SE (95% CI)
Keratinized Tissue Thickness	0.90 ± 0.045mm (0.812 to 0.988)	1.35 ± 0.056mm (1.238 to 1.461)	0.45 ± 0.043mm (0.366 to 0.533) p < 0.001***
Relative Gingival Recession	11.18 ± 0.214mm (10.755 to 11.602)	10.38 ± 0.242mm (9.901 to 10.861)	-0.798 ± 0.132mm (-1.057 to -0.538) p < 0.001***
Keratinized Tissue Width	3.01 ± 0.228mm (2.556 to 3.459)	3.68 ± 0.228mm (3.225 to 4.129)	0.67 ± 0.074mm (0.525 to 0.815) p < 0.001***
Relative Clinical Attachment Level	12.74 ± 0.219mm (12.308 to 13.177)	12.03 ± 0.255mm (11.521 to 12.534)	-0.715 ± 0.16 (-1.028 to -0.401) p < 0.001***
Bone Level	14.34 ± 0.186mm (13.970 to 14.709)	14.26 ± 0.187mm (13.894 to 14.634)	-0.076 ± 0.042mm (-0.157 to 0.006) p > 0.05
Probing Pocket Depth	1.58 ± 0.089mm (1.400 to 1.755)	1.55 ± 0.111mm (1.335 to 1.775)	-0.023 ± 0.09mm (-0.199 to 0.153) p > 0.05

The T1-T0 means differences are conditional to the estimated models, so result of a partial pooling.

Figure 1 A-B: Schematic drawings illustrating the clinical anatomic characteristics measured at the buccal aspect of the defect.

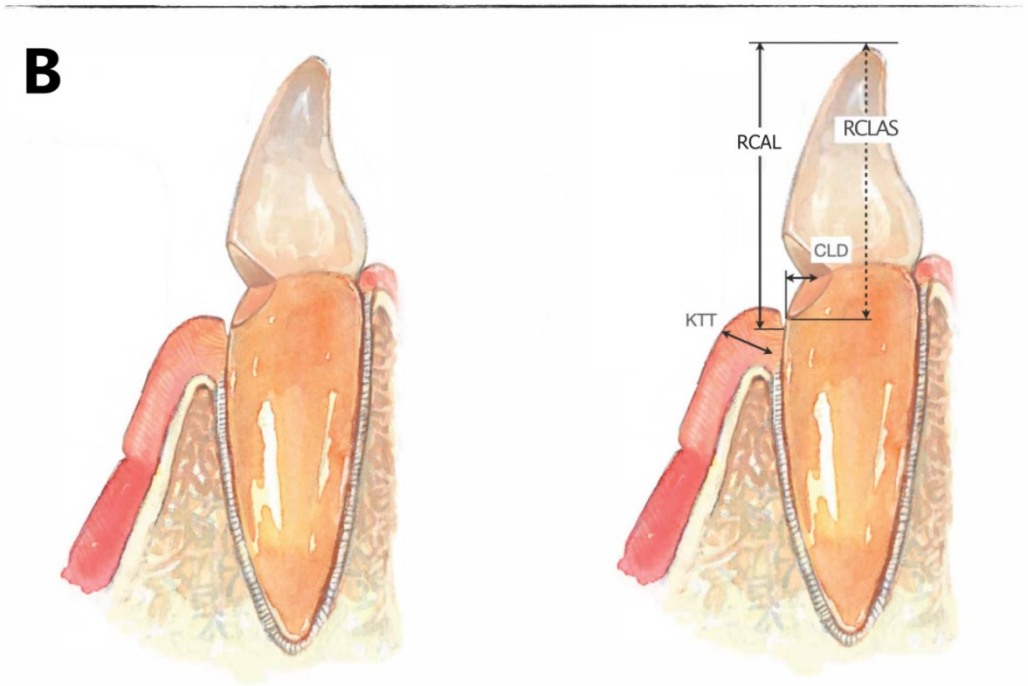

Figure 2: Step by step description of the root plastique technique.

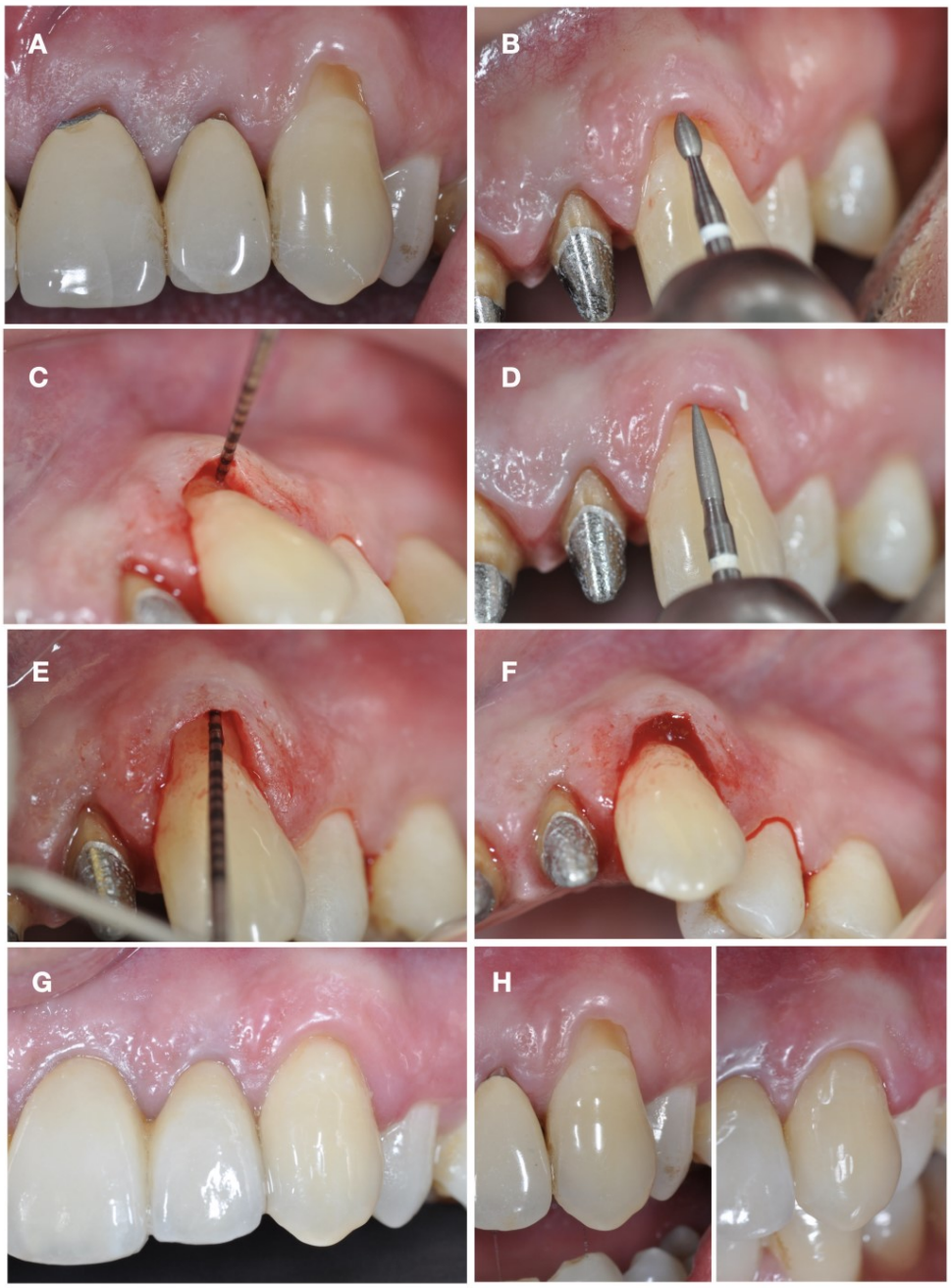

Figure 3: A) Changes in the keratinized tissue thickness by patient and recession from T0 to T1; B) Changes in the keratinized tissue thickness (only recession with initial KTT of <0.8mm) by patient and recession from T0 to T1

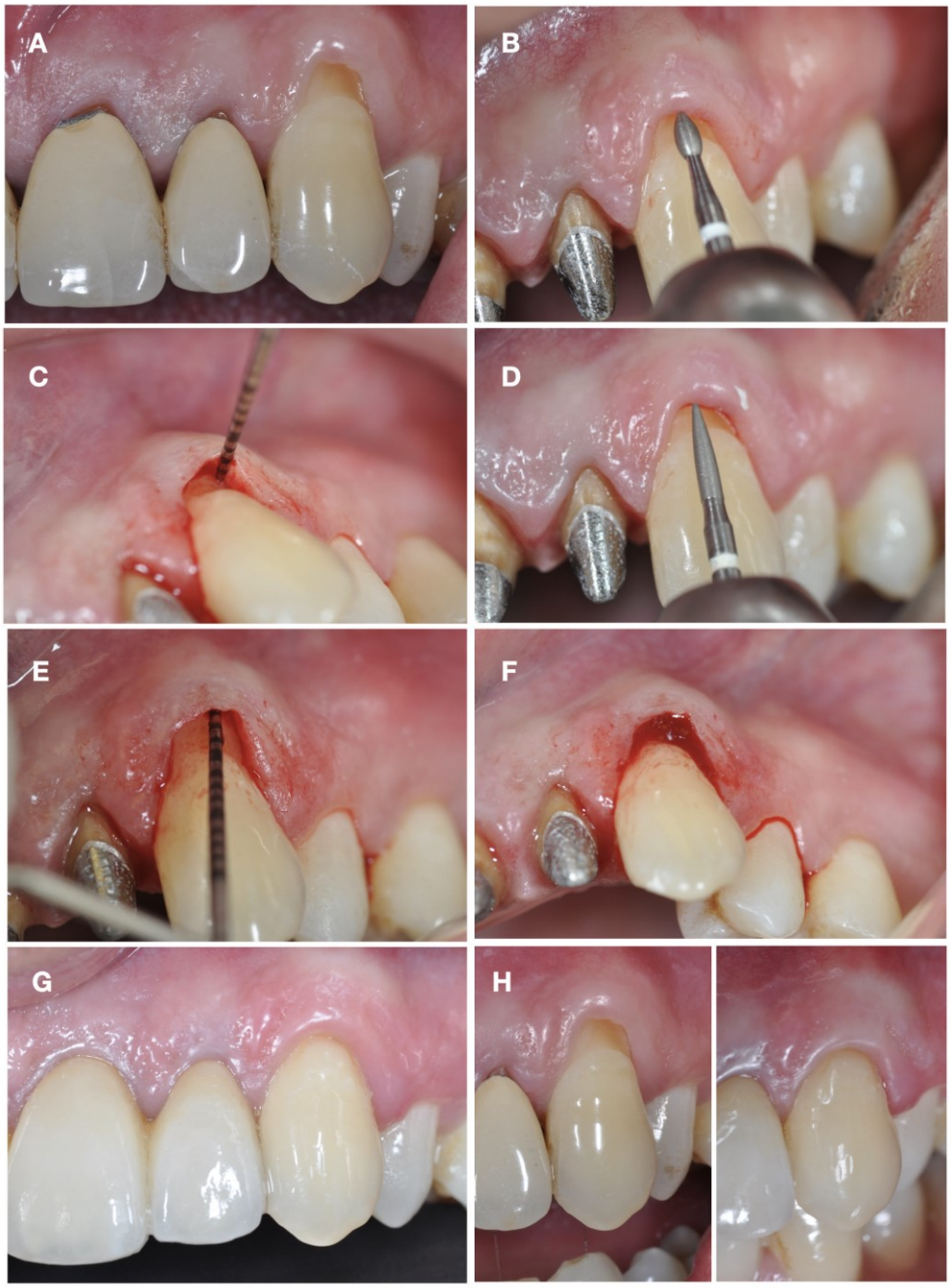

Figure 4: Regression analysis of the relative gingival recession reduction and keratinized tissue thickness increase by Non-carious cervical lesion depth at baseline

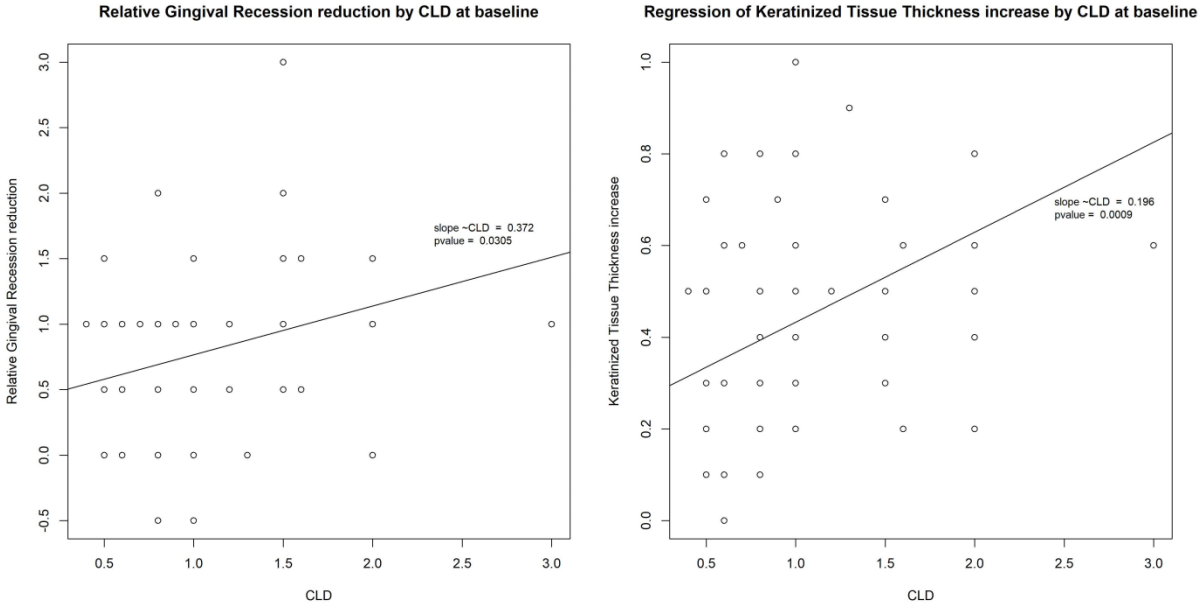

Figure 5: Changes in the relative gingival recession by patient and recession from T0 to T1