# **ANALYSIS OF THE VARIATION IN BIOCHEMICAL PARAMETERS IN ROOT COVERAGE WITH** ENAMEL MATRIX DERIVED PROTEINS ASSOCIATED WITH CONNECTIVE TISSUE GRAFT

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# BACKGROUND

Gingival recessions refers to the apical displacement of the gingival margin towards the cemento-enamel junction, resulting in an exposure of the tooth 's root surface (Jepsen et al., 2018). The aim of treating these recessions is to achieve total and permanent regeneration of the soft tissues over the exposed roots in the long term, particularly when patients have symptoms such as tooth sensitivity, aesthetic concerns or a high risk of the lesion progressing (Zuhr et al., 2021). Despite the various techniques that have been proposed, the association between the coronally advanced flap (CAF) and autologous connective tissue graft (CTG) has established itself as a reference approach for root coverage (Stefanini et al., 2018). Proteins derived from the enamel matrix (whose commercial name is Emdogain (EMD))

influences tissue regeneration and healing in this type of surgery, but there is little evidence of its benefits when combined with CAF+CTG (Roman et al., 2013), (Casillas et al., 2015), (Discepoli et al., 2019),

(Aydinvurt et al., 2019), (Mercado et al., 2020).

## **OBJECTIVES**

The aim of this research was to assess the behaviour of two inflammatory biomarkers, interleukin 8 (IL-8) and vascular endothelial growth factor (VEGF), in the gingival crevicular fluid (FCG) of patients undergoing periodontal root coverage surgery using the CAF+CTG technique, with or without the additional application of EMD. Tracking the temporal profile of these biomarkers in the FCG could provide relevant information on the inflammatory and healing responses associated with each treatment approach.

### **RESULTS AND DISCUSSION**

# MATERIALS AND METHODS

The study included 24 patients from the Egas Moniz Dental Clinic, divided into two groups: the test group (n=13) treated with EMD and the control group (n=11). A total of 94 FCG samples were collected using paper strips (Periopaper®) at four different times during the healing process, i.e., immediately after surgery (day 0) and after 7, 14 and 21 days. The volume of each sample was assessed using an electronic device (Periotron<sup>®</sup> 8010). IL-8 and VEGF concentrations were quantified using enzyme-linked immunosorbent assay (ELISA) kits at the Molecular Biology LAB of the Egas Moniz School of Health and Science. This analysis was carried out blindly, without any access to patient data prior to surgery. Statistical analysis was carried out using the non-parametric Mann-Whitney U test for inter-group comparisons and Friedman's two-way analysis of variance by ranks for intra-group comparisons (over time), since the data did not follow a normal distribution for significance level of 95%.

#### Table 1 - Mean values of IL-8 concentration (µg/ml)

#### Table 2 - Mean VEGF concentration values (µg/ml)

Time (t)	Control Group	Test Group	Time (t)	Control Group	Test Group
	(CAF+CTG)*	(CAF+CTG+EMD)*		(CAF+CTG)*	(CAF+CTG+EMD)*
Der: 0	22.24 + 16.9	25.22 + 16.0	Day 0	$12.07 \pm 0.4$	$14.16 \pm 7.2$

A slight increase in IL-8 (Table 1 and Figure 1) was observed on day 7, which was only significant in the test group (p=0.023). Thereafter, the levels returned to the baseline values until day 21 with a significant decrease observed on day 14 in the control group. These patterns are indicative of the anticipated inflammatory response in the immediate postoperative period. However, the variations were not found to be significantly different between the two groups analysed. For VEGF (Table 2 and Figure 2), a similar pattern was observed, yet the test group exhibited sustained elevated levels until day 14, a finding that diverged significantly from the control group (p = 0.015). These results do not permit the detection of differences in the use of EMD, as was also concluded from the clinical results (Figures 3, 4 and 5). Although the test group showed better results than the control group in reducing gingival recession (RedRec) and achieving partial/total coverage (Rrp/Rrt) (1.2 mm and 0.8 mm; 50% and 33.2%, respectively), these

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Day 7	86,97 ± 51,9	137,99 ± 157,6	Day 7	$24,93 \pm 13,6$	22,99 ± 12,1
Day 14	$18,41 \pm 16,3$	78,00 ± 101,8	Day 14	$11,18 \pm 8,6$	21,73 ± 12,9
Day 21	$26,42 \pm 30,4$	$42,20 \pm 57,1$	Day 21	$14,90 \pm 17,7$	17,08 ± 12,5

\* The values presented correspond to the mean value ± standard error

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Figure 2 – Mean VEGF concentration values (µg/ml)







**Figure 5** – Post-operative photograph after 3 months

# CONCLUSIONS

The findings of this investigation do not demonstrate the potential benefits of IL-8 e VEGF as markers of the inflammatory status following CAF+CTG. The present work is thus positioned as a relevant pilot

study, allowing the estimation of the variability of biomarkers and providing necessary data for future large-scale investigations.

**KEYWORDS:** gingival recession, CAF, biomarkers, IL-8, VEGF.

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