Salivary IL-1β: A Biochemical Marker that Predicts Periodontal Disease in Orthodontic Treatment

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Recent research has shown that IL8, IL6, IL-1β and TNFα played significant roles in the first stages of tooth movement, but not in the linear stage. After treatment, the values of IL–1β dropped significantly more in the case of patients who received combined treatment (periodontal and orthodontic) compared to the patients that received only periodontal treatment. We must underline the fact that at the start of the treatment the patients whose periodontium was affected and who were about to receive periodontal treatment associated with orthodontic treatment had higher values of IL–1β (but not statistically significant). The comparative study of IL-1β values post-treatment shows that in the case of associated treatment the post-treatment values drop significantly more for all Angle classes, being higher in the patients who had class III Angle.

Keywords: IL-1β, salivary biomarker, periodontal disease, orthodontic treatment, cytokines

Ren and associates have demonstrated [1] that IL8, IL6 IL1β and TNFα played significant roles in the first stages of tooth movement, but not in the linear stage. This study suggests that once the micro-environment in the periodontal ligament is activated by orthodontic forces, several key proinflammatory cytokines are produced and they start a cascade of cellular events [1].

The objective of this study was to evaluate comparatively the levels of IL-1β in patients wearing orthodontic devices who had a stabilised pre-existing periodontal pathology (chronic periodontitis localised in minimum three teeth) compared to patients with the same pathology but who were not receiving orthodontic treatment.

The aim of this research was to describe the biological processes that take place at molecular level upon applying orthodontic forces on an affected periodontium, to establish the role of orthodontic treatment in periodontal treatment and, last but not least, to identify the markers that can indicate in the most accurately the level of inflammation in periodontal tissues.

Dental movement is effected through processes of bone resorption and apposition that occur subsequent to the existence of an inflammatory process localised at this level. IL-1β is an interleukin with a major proinflammatory role and therefore can be used as an inflammation marker. [2,3]

Experimental part

The objective of the study was to determine the level of IL-1β in the saliva of teeth that are orthodontically tractioned. A comparison was also made between the levels of IL-1β in these teeth and the teeth upon which no orthodontic force is applied.

The authors obtained the authorisation of the research ethics committee of the Grigore T. Popa University of Medicine and Pharmacy in Iasi for the collection of saliva samples; the 60 selected patients gave informed consent for the study.

The 60 patients considered in the study were divided into three groups, as follows: **group A**, the control lot, consisting of 16 patients without periodontal disease, without clinical gingival modifications; **group B**, 22 patients with periodontal disease (chronic periodontitis localized it at least three teeth), who received periodontal treatment; **group C**, 22 patients with periodontal disease (chronic periodontitis localized it at least three teeth) who received combined orthodontic and periodontal treatment.

Saliva was collected in two rounds from the patients in groups B and C, as follows:

-for **group B,** an initial collection and a second collection six months after the end of periodontal treatment

- for **group C,** an initial collection and a second one six months after periodontal stabilisation and start of orthodontic treatment

Both for the patients in group B and for those in group C periodontal treatment similar and consisted in supra- and sub-gingival scaling and root planing.

The results were interpreted through high-accuracy statistic methods and tests.

Results and discussion

The values of IL-1β determinations in the three groups : B (periodontal treatment), C (periodontal treatment associated with orthodontic treatment) and the control group A, are systematically presented in the table below.

The values of IL-1β dropped significantly both in the patients who had only periodontal treatment (F = 26.69, p<<0.01, 95%CI) as well as in those who had periodontal treatment associated with orthodontic treatment (F = 16.49, p <<0.01).

The values of IL-1β before treatment in the two lots did not show significant differences ($p_{Newman-Keuls}$ = 0.061), but they are significantly higher than the values in the control lot.

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Table 1

STATISTICAL INDICATORS OF IL-1β [ng/mL] IN PATIENTS WITH PERIODONTAL DISEASE (MINIMUM THREE TEETH AFFECTED) WHO
RECEIVED PERIODONTAL TREATMENT ASSOCIATED (YES/NO) WITH ORTHODONTIC TREATMENT

		Average IL-1β [pg/ml]	Average -95%	+95%	Std.dev.	Std.err.	Min	Max	Q25	Median	Q75
Group B (periodontal treatment)	before	0.70	0.65	0.74	0.10	0.02	0.51	0.85	0.62	0.68	0.77
	after	0.49	0.41	0.56	0.16	0.03	0.21	0.81	0.34	0.46	0.60
Group C (periodontal treatment + OT)	before	0.76	0.71	0.80	0.11	0.02	0.57	0.94	0.67	0.76	0.83
	after	0.38	0.33	0.44	0.12	0.03	0.19	0.61	0.27	0.39	0.48
Group A (control)	before	0.21	0.16	0.25	0.07	0.02	0.11	0.35	0.17	0.19	0.23
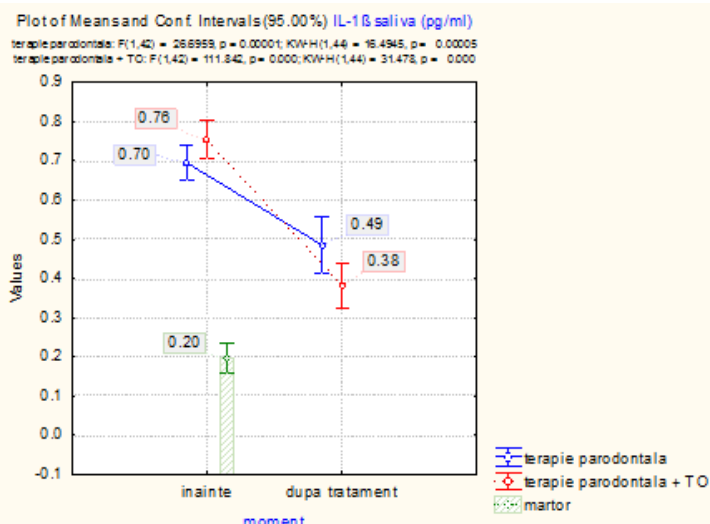
Fig. 1. Statistical indicators of IL-1β [ng/mL] in patients with periodontal disease

	F (95% confidence interval)	p
Kuskal-Wallis Test	70.30598	0.000000

Table 2

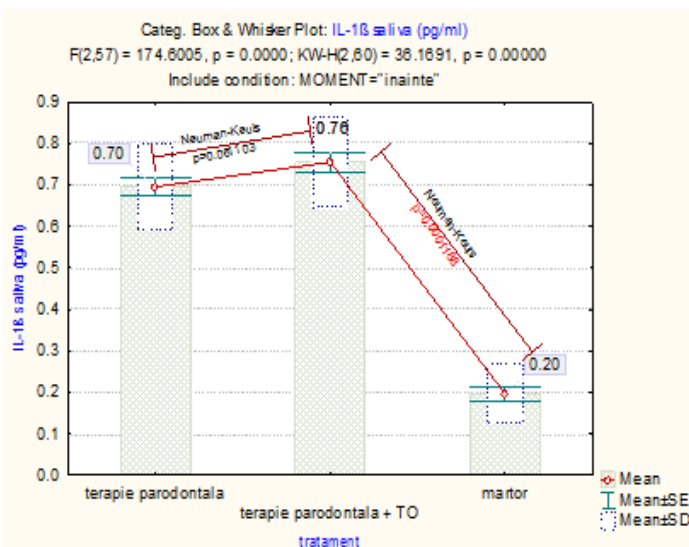TEST FOR COMPARING THE VALUES OF IL-1β [ng/mL] IN PATIENTS WITH PERIODONTAL DISEASE

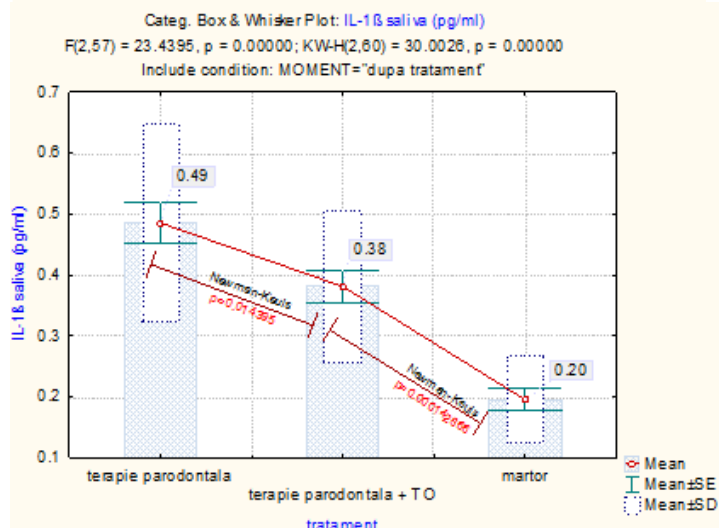Fig. 2. Statistical indicators of IL-1β in patients with periodontal disease before treatment

Fig. 3. Statistical indicators of IL-1β in patients with periodontal disease after treatment

Although both patient groups showed significant drops in IL-1β, after treatment it can be noticed that in the case of patients who received periodontal treatment associated with orthodontic treatment the values of IL-1β (Median$_{IL-1â}$=0.38±0.12DS) are significantly lower than those that correspond to the patients who received only periodontal treatment (Median$_{IL-1â}$ =0.49±0.16DS) (F=23.43, p<<0.01, 95%CI). In both lots the values of IL-1β remain significantly higher post-treatment, compared to the control lot.

Evaluation of IL-1β in relation to Angle Class

The patients with periodontal disease had significantly higher values (F=37.43, p<<0.01, 95%CI) of IL-1β compared to the control lot, but there were no significant differences depending on the type of treatments and Angle class (F$_{associated treatment}$=0.64, p=0.59; F$_{periodontal treatment}$=0.401, p=0.804).

After treatment, the values of IL-1β in the patients who received associated treatment dropped significantly, but they had significantly higher values in the case of the

Table 3
STATISTICAL INDICATORS OF IL-1β IN PATIENTS WITH PERIODONTAL DISEASE WHO RECEIVED PERIODONTAL TREATMENT ASSOCIATED WITH ORTHODONTIC TREATMENT, IN RELATION TO ANGLE CLASS

Before treatment	Angle	Average IL-1β [pg/ml]	Average -95%	Average +95%	Std.dev.	Std.err.	Min	Max	Q25	Median	Q75
periodontal treatment	1	0.68	0.52	0.85	0.10	0.05	0.58	0.82	0.62	0.67	0.75
	2	0.66	0.45	0.87	0.13	0.06	0.51	0.82	0.58	0.65	0.74
	3	0.76	0.38	1.14	0.15	0.09	0.59	0.85	0.59	0.84	0.85
	4	0.70	0.56	0.84	0.11	0.05	0.57	0.85	0.61	0.72	0.73
	5	0.70	0.63	0.76	0.06	0.02	0.62	0.77	0.64	0.69	0.77
periodontal treatment +OT	1										
	2	0.70	0.52	0.88	0.11	0.06	0.60	0.83	0.61	0.68	0.79
	3	0.74	0.59	0.88	0.12	0.05	0.58	0.88	0.67	0.75	0.81
	4	0.77	0.64	0.90	0.14	0.05	0.57	0.94	0.65	0.77	0.93
	5	0.79	0.73	0.86	0.06	0.03	0.71	0.88	0.75	0.79	0.86
control	1	0.20	0.16	0.23	0.07	0.02	0.08	0.35	0.16	0.18	0.22

Plot of Means and Conf. Intervals (95.00%) IL-1ß saliva (pg/ml)
terapie parodontala: F(4,17) = 0.4018, p = 0.8047; KW-H(4,22) = 1.4636, p = 0.8331
terapie parodontala + TO: F(3,18) = 0.6419, p = 0.5979;
Include condition: MOMENT="inainte"
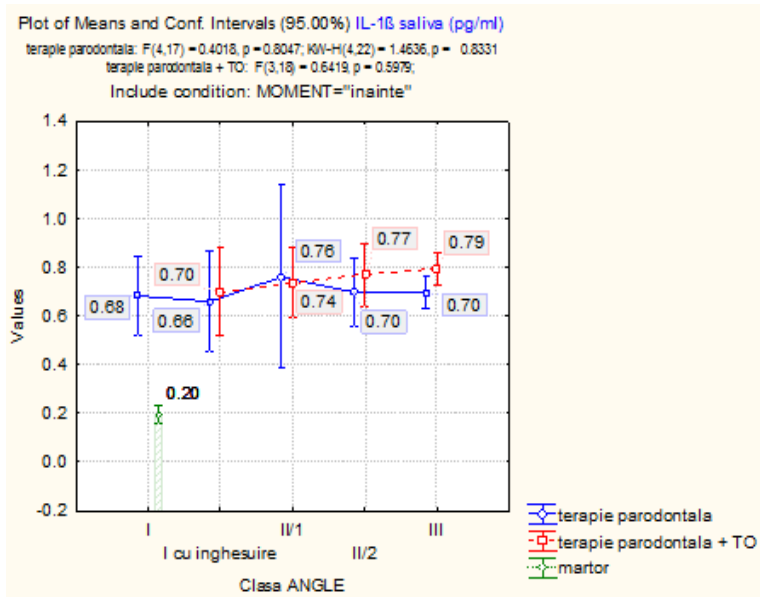
Fig. 4. Statistical indicators of IL-1β in patients with periodontal disease vs. Angle Class

	F (95% confidence interval)	p
Kuskal-Wallis Test	8.669860	0.000000

Table 4
TEST FOR COMPARING THE VALUES OF IL-1β IN PATIENTS WITH PERIODONTAL DISEASE VS. ANGLE CLASS

Table 5
STATISTICAL INDICATORS OF IL-1β IN PATIENTS WITH PERIODONTAL DISEASE (MINIMUM THREE TEETH AFFECTED) WHO RECEIVED PERIODONTAL TREATMENT ASSOCIATED (YES/NO) WITH ORTHODONTIC TREATMENT VS. ANGLE CLASS

After treatment	Angle	Average IL-1β [pg/ml]	Average -95%	Average +95%	Std.dev.	Std.err.	Min	Max	Q25	Median	Q75
periodontal treatment	1	0.37	0.28	0.46	0.06	0.03	0.34	0.45	0.34	0.34	0.40
	2	0.50	0.15	0.86	0.22	0.11	0.29	0.81	0.36	0.45	0.65
	3	0.61	0.13	1.08	0.19	0.11	0.39	0.77	0.39	0.66	0.77
	4	0.46	0.20	0.72	0.21	0.09	0.21	0.70	0.29	0.50	0.60
	5	0.51	0.42	0.61	0.09	0.04	0.39	0.63	0.45	0.52	0.58
periodontal treatment +OT	1										
	2	0.32	0.17	0.46	0.09	0.05	0.19	0.42	0.25	0.33	0.38
	3	0.37	0.22	0.52	0.12	0.05	0.21	0.50	0.27	0.41	0.43
	4	0.31	0.22	0.40	0.10	0.04	0.20	0.47	0.23	0.31	0.38
	5	0.52	0.46	0.58	0.06	0.02	0.44	0.61	0.48	0.52	0.56
control	1	0.20	0.16	0.23	0.07	0.02	0.08	0.35	0.16	0.18	0.22
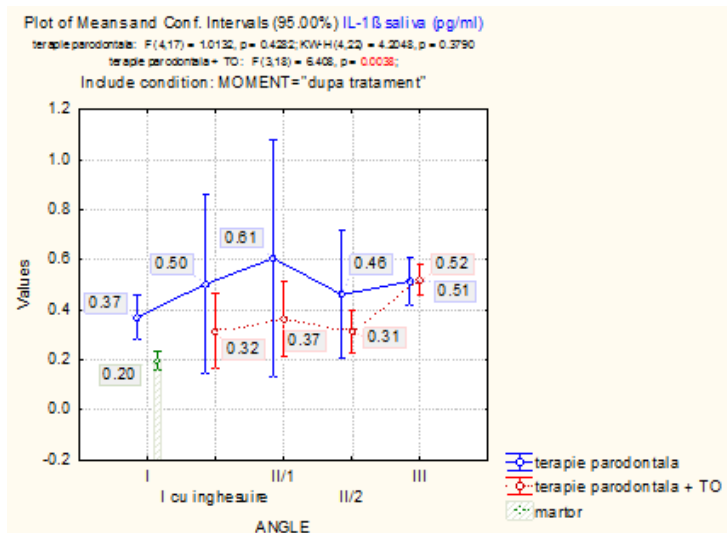
Fig.5. Statistical indicators of IL-1β in patients with periodontal disease (after treatment)

Table 6
TEST FOR COMPARING THE VALUES OF IL-1β IN PATIENTS WITH PERIODONTAL DISEASE

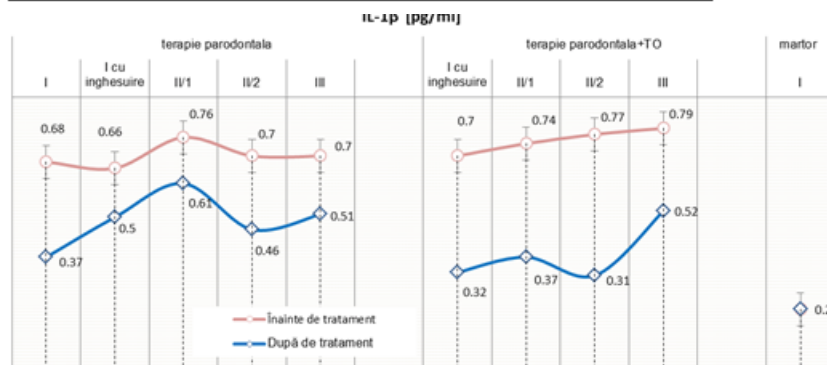|	F (95% confidence interval)	p
Kuskal-Wallis Test	8.669860	0.000000

Fig. 6. Statistical indicators of IL-1β in patients with periodontal disease vs. Angle class

patients with Class III Angle (F=6.408, p=0.0038). In the patients who received only periodontal treatment, post-treatment there were no significant differences found depending on the Angle Class (F=1.013, p=0.42). Depending on the type of treatment, the values of IL-1β were significantly lower post-treatment in the case of associated treatment for all the Angle classes, less class III, for which comparable values of IL-1β were obtained.

A comparative analysis of the IL-1β values depending on the type of treatment and Angle class shows that, after the associated treatment, the values of IL-1β drop significantly more, although they were significantly higher prior to treatment.

After treatment, the values of IL–1β dropped significantly more in the patients who received combined treatment compared to the patients who received only periodontal treatment. It is worth noting that at the start of the treatment, the patients with periodontal pathology who were about to start periodontal treatment associated with orthodontic treatments had higher values of IL–1β (but not statistically significant).

An important aspect is pointed to by the comparative study of IL-1β values post-treatment: it shows that in the case of combined treatment the post-treatment values drop significantly more for all Angle classes, being higher in patients with Angle Class III. Previous studies on saliva were evaluating the statistical correlation between PGE2 increase and Angle Class [4].

Multiple studies have been carried out, especially using crevicular fluid and venous blood, but the research of inflammatory cytokines in saliva offers broad perspectives that deserve to be explored.

Jaedicke and associates published in 2016 an extensive article based on longitudinal and transversal studies, in which they underline the importance of IL-1 β and IL 6

interleukins as main salivary inflammatory biomarkers associated with periodontal disease [5].

De Lima and associates carried out a meta-analysis based on 905 studies found in literature. Of the 905 studies, four were included in a quantitative and qualitative analysis. The results pointed out that a biomarker, namely the macrophage inflammatory protein 1 alpha (MIP1α), showed excellent accurate diagnostic properties. Another two inflammatory markers IL-1 beta and IL-6 showed acceptable diagnostic properties. The research characterises MIP1α based solely on one study, fact that affects the credibility and robustness of results [6].

The results published by De Lima are in agreement with the results obtained by the authors of this paper in terms of the diagnostic value of 1 beta interleukin. [5]

Previous studies pointed out that circulatory inflammatory marker increase in periodontitis [7]. In our research we have made a comparative analysis of salivary IL-1β values depending on the type of treatment and Angle class and we have found that after associated treatment the values of IL-1β drop significantly more, although they were higher prior to treatment.

The values of IL–1β dropped significantly more in the patients who received combined treatment compared to the patients who received only periodontal treatment. It is important to mention the fact that at the start of the treatment the patients with periodontal pathology who were about to start periodontal treatment associated with orthodontic treatment had higher values of IL–1β. In the same time the biological limits of the periodontal tissues should not be exceed by using heavy forces [8].

Conclusions

After combined treatment (periodontal treatment and orthodontic treatment) the values of IL–1β in saliva

decreased significantly more compared to the values found after periodontal treatment alone.

An important aspect that was noticed in the comparative study of IL-1β values post-treatment underlines the fact that in the case of combined treatment (periodontal and orthodontic) the values drop significantly more for all Angle classes. Higher values were recorded tough, in the patients who presented Angle class III.

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