Turkish Journal of Medical Sciences

Volume 31 | Number 4 Article 13

1-1-2001

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Turk J Med Sci
31 (2001) 365-369

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The Effect of Fixed Restoration Materials on the IL-1β Content of Gingival Crevicular Fluid

Received: August 03, 2000

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Abstract: Cast alloys used in dentistry come into close and prolonged contact with the gingiva and the oral mucosa, and certain alloys have been claimed to cause inflammation of gingival and periodontal tissues. However, only little information is available on the molecules mediating the mechanism of such an effect. For this reason, the aims of this study were to determine 1L-1β levels in the gingival crevicular fluid (GCF) before and after the placement of base, high-noble metal-ceramic and all-ceramic crowns.

Thirty patients requiring an artificial crown for a maxillary second premolar were suitable for the present study; 18 were male and 12 female and their mean age was 24.3 years. Six months prior to participating in the study, all patients were free of systemic disorders, did not take any drugs and had clinically healthy gingiva.

Ten patients (group 1) were treated with CrNiMo-based ceramic crowns (Remanium CS, Dentaurum, Germany), 10 (group 2) were treated with Inceram (Vita, Germany) ceramic crowns, and 10 (group 3) were treated with AuPtIn-based ceramic crowns (Pontostar, Bego, Germany). GCF samples were collected with paper strips before and 4 months after the placement of crowns. The samples were shocked at -70ºC. The remaining maxillary second premolars in the opposing arch acted as controls.

1L-1β levels increased after the placement of CrNiMo crowns ($p<0.05$), but for the other crowns no statistically significant changes were recorded. On the other hand, concentrations of 1L-1β did not vary with any of the crowns ($p>0.05$).

These results reconfirmed the hypothesis that base alloys cause more gingival inflammation than high-noble and all ceramic crowns.

Key Words: Gingival crevicular fluid, casting alloys, interleukin-1 beta

Introduction

Many alloys are used in fixed prosthodontics. Evaluation of the performance of the individual alloys and different alloy systems is difficult, not only technically, but also biologically and clinically. Limited data are available to assess the biological effects and the clinical efficacy of all these alloys.

Dental casting alloys vary markedly in composition and some of them contain allergenic and potentially toxic elements, such as nickel, cobalt, lead, cadmium and beryllium. Certain dental alloys have been claimed to cause gingival and periodontal inflammation. However, little information is available on the molecules mediating the mechanism of gingival and periodontal inflammation (1).

Dysregulated cytokine and immunogloblobin production at local disease sites have been considered major contributors to the development of inflammatory diseases, such as lichen planus, autoimmune disorder and some neoplastic processes. Among the numerous cytokineses involved in the induction and regulation of host responses in inflammation, interleukin-1 seems to play a central role in the inflammatory reaction. Monocytes and macrophages are the main producers. Production of IL-1 can be induced by a variety of agents: microorganisms, microbial products, inflammatory agents, plant lectins and antigens. IL-1 has been shown to enhance various immune responses in vitro, including β lymphocyte differentiation, antibody secretion, T lymphocyte proliferation and acute phase protein synthesis, and to cause endogenous pyrogen-induced fever, fibroblast proliferation, bone resorption, and collagenase and prostaglandin E production by fibroblasts and chondrocytes (2). Masada et al. (3) showed that both 1L-1α and 1L-1β were produced and released locally in

periodontal disease at concentrations sufficient to mediate tissue inflammation and bone resorption (3). They found that the culture supernatants from gingival samples biopsied from inflamed gingival tissues contained significantly higher IL-1 activities than those from healthy ones.

The immunological basis for the side - effects of dental casting alloys is poorly understood. Therefore, the aim of the present study was to determine the amount of 1L-1β in the GCF at crown sites, which were of different alloys and all ceramic.

Materials and Methods

1. Experimental groups and criteria for selection

Thirty patients (age range 19-30 years, mean 24.5 years), requiring artificial crowns for maxillary second premolars were selected for this study. They were divided into three groups. The 10 patients in group 1 were treated with CrNiMo-based ceramic crowns. Group 2 consisted of 10 patients treated with Allceram ceramic crowns and group 3 of 10 patients treated with AuPtIn based ceramic crowns. The remaining natural teeth in the opposing arch acted as controls. All patients were free of systemic disorders, had healthy gingival tissues and did not take any drugs during the 6 months prior to participating in the study. Oral hygiene instructions were given, and patients were encouraged to follow their oral hygiene regime throughout the study.

Prior to prosthetic treatment, clinical data were obtained from control and test teeth after fluid sampling. The following parameters were assessed by one examiner sequentially: plaque index (P1I) (4), gingival index (GI) (5), probing depth and bleeding index (6). The readings for P1I, GI, probing depth and bleeding index (BI) were taken only from the palatal or lingual surface of each tooth.

This procedure was repeated after 4 months and individuals who did not have healthy gingival tissue were excluded from the study. All of the patients were treated by one dentist with a chamfer preparation, approximately 1 mm beneath the gingival crest, followed by impressions with addition reaction silicone (Provil, Bayer, Germany). Artificial crowns were fabricated by the same dental technician for each prepared tooth.

2. Sampling Of Gingival Crevicaler Fluid (GCF)

Prior to treatment, GCF was collected from the teeth which required artificial crowns and from the control teeth. They were isolated from saliva with cotton wool rolls and dried with short air blasts. GCF was collected with periopaper strips (Periopaper, Proflow Inc. Amityville) (Fig. 1). The pre-weighed strips (2 mm x 7 mm) were left within the sulcus for a standardized period of three minutes. The amount of GCF on the strips was measured by weighing the accumulated fluid (7). The strips containing GCF were placed in pre-weighed, sealed plastic tubes and weighing was repeated immediately

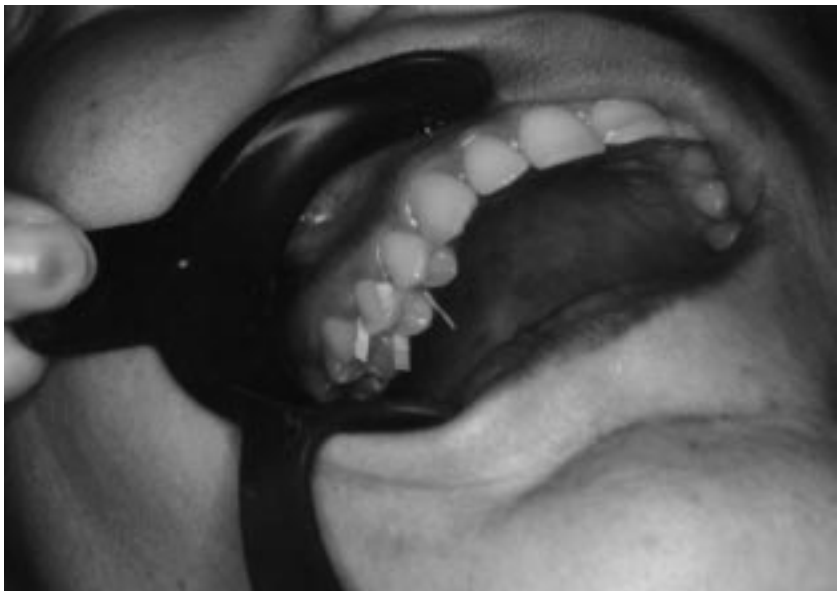

Fig. 1. A case depicting the GCF samples, which were collected with periopaper strips.

after collection. The difference between the two weighings gave the volume of fluid collected, assuming a specific gravity of approximately 1 (8). Samples were then wrapped in aluminium foil and the foil packet was placed in a plastic tube and stored at -70ºC until it was analyzed.

3. Extraction of GCF and analysis of IL-1β

The amount of 1L-1β in GCF samples was assayed by enzyme-linked immunosorbent assay (ELISA) with recombinant 1L-1β monoclonal antibody as a standard (ELISA kit, R-D systems, Minneapolis, MN). All assay procedures were carried out according to the manufacturer's instructions and optical densities were measured in a spectrophotometer (EL 312 Bio-Tek) at 450 nm. The total IL-1β in the sample was determined in picograms (pg) and calculation of the IL-1β concentration in each sample was performed by dividing the amount of IL-1β by the volume of the sample (pg/µl).

Statistical Analysis

Data analysis was performed using the statistical software package SPSS for Windows 10.0. The statistical significance of 1L-1β levels and clinical parameters (before the preparation and four months after the crown insertion) was analyzed using the Wilcoxon rank sum test. The Mann-Whitney U test was used to determine the significance of all parameters for all of the groups. The correlation between the 1L-1β levels and the clinical parameters was used in the Spearman correlation test. $P < 0.05$ was considered statistically significant.

Results

Data regarding the clinical parameters are given in Table 1. No significant statistical differences were detected before the preparation and four months after the crown insertion except in group 1. The laboratory results are shown in Table 2. The volume of GCF was statistically different in group 1 ($p<0.05$). As for the other groups, no change was recorded. 1L-1β levels increased in group 1, but in the other groups no statistically significant differences were recorded (Fig. 2). On the other hand, the concentration of crevicular 1L-1β was not statistically significant when comparing before and after 4 months of treatment (Table 2).

There is only a positive significant correlation between the clinical parameters and total 1L-1β levels in group 1 ($r=0.548$, $p<0.05$). No correlation was found in groups 2 and 3 (respectively $r=0.254$, $p>0.05$, $r=0.197$, $p>0.05$).

Discussion

This study investigated whether marginal gingiva in contact with the margins of CrNiMo based-ceramic crowns would exhibit more inflammation than gingiva in contact with the margins of Inceram and AuPtIn-based ceramic crowns. The reason for increased gingival inflammation at CrNiMo-based ceramic crowns could have been the increased IL-1β levels in the GCF.

Table 1. Periodontal condition before and after the treatment .

		GROUP 1		GROUP 2		GROUP 3	
		TEST (Mean ±SD)	CONTROL (Mean ±SD)	TEST (Mean ±SD)	CONTROL (Mean ±SD)	TEST (Mean ±SD)	CONTROL (Mean ±SD)
PRIOR TO THE	GI	0.42 ± 0.21	0.57 ± 0.25	0.45 ± 0.13	0.53 ± 0.24	0.47 ± 0.18	0.59 ± 0.24
TREATMENT	P1I	0.62 ± 0.24	0.71 ± 0.19	0.73 ± 0.20	0.75 ± 0.17	0.65 ± 0.26	0.64 ± 0.21
	B1	0.66 ± 0.15	0.64 ± 0.33	0.68 ± 0.27	0.65 ± 0.15	0.69 ± 0.25	0.61 ± 0.29
	Probing Depth (mm)	1.27 ± 0.30	1.33 ± 0.47	1.31 ± 0.22	1.27 ± 0.38	1.25 ± 0.42	1.28 ± 0.36
AFTER 4	GI	0.71 ± 0.22 #	0.52 ± 0.21 *	0.52 ± 0.27	0.54 ± 0.17	0.54 ± 0.25	0.56 ± 0.14
MONTHS	P1I	0.79 ± 0.33 #	0.64 ± 0.15 *	0.69 ± 0.24	0.74 ± 0.24	0.65 ± 0.26	0.67 ± 0.19
	B1	1.12 ± 0.42 #	0.79 ± 0.22 *	0.77 ± 0.32	0.75 ± 0.31	0.74 ± 0.30	0.68 ± 0.23
	Probing Depth (mm)	1.55 ± 0.20 #	1.48 ± 0.34 *	1.41 ± 0.37	1.37 ± 0.43	1.40 ± 0.39	1.37 ± 0.38

# P< 0.05. between prior to the treatment and after 4 months

* p< 0.05. between test and control groups

Table 2. Results of total 1L-1β levels, GCF volume, 1L-1β concentration of the test and control teeth in groups 1, 2 and 3.

	Group 1 (n=10)			Group 2 (n=10)			Group 3 (n=10)		
	Test	Control	P	Test	Control	P	Test	Control	P
Total IL-1β (pg)	72.2±18.2	47.5±20.3	<0.05	61.4±34.9	58.1±30.4	>0.05	57.6±24.5	57.4±25.9	>0.05
Volume (ml)	0.81±0.23	0.67±0.3	<0.05	0.66±0.25	0.57±0.23	>0.05	0.63±0.29	0.58±0.33	>0.05
Consant. (pg/μl)	95.31±29.19	74.81±22.18	>0.05	93.63±45.06	101.5±34.045	>0.05	103.4±54.34	108.4±32.15	>0.05

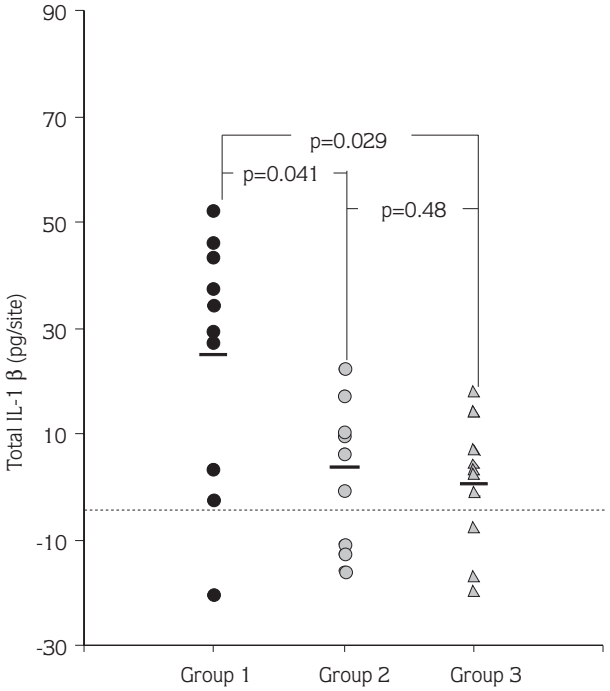

Fig. 2. The changes in 1L-1β levels in groups 1, 2 and 3.

The IL-1β levels were measured in GCF by comparing teeth with crowns and opposing natural teeth to monitor the effect of oral hygiene on gingival inflammation. Measurements obtained from natural teeth revealed no significant differences during the experiment, so it was concluded that the clinical conditions of the natural teeth and the restored teeth were similar during the experiment.

The measurement of the IL-1β is based on the prediction that the quantity of sulcular fluid is a parameter of inflammation of the marginal gingiva. According to basic studies, the collection and analysis of GCF offers a noninvasive means of evaluating the host response in periodontal disease (10). IL-1β has been selected for numerous investigations because of its important role in the host response in periodontal disease (8,9).

Total IL-1β levels of the restored teeth in group 1 were significantly higher than those of the other groups. This result reconfirmed the hypothesis that marginal gingiva in contact with the margins of group 1 crowns exhibit more inflammation than gingiva coming in contact with the other group of crowns.

There was a wide range in the values of IL-1β detected in our GCF samples. This range might reflect the degree of inflammation at the time of sampling, which is difficult to grade by clinical parameters alone. This explanation was supported by the correlation between the amounts of IL-1β from each sampling site and the clinical indices. These data indicate that the amounts of crevicular IL-1β from each sampling site correspond well with the clinical severity of gingival health.

Masada (3) reported a correlation between IL-1β concentration and GCF volumes or clinical indices including probing depth, plaque index and bleeding index. Since the GCF volume correlated with the degree of gingival inflammation, the greater increase in GCF flow over the production of IL-1β by locally infiltrated inflammatory cells may in fact result in a decrease in IL-1β concentration in the GCF samples. Obviously, the total amount of IL-1β, but not the concentration, could be highly relevant to the clinical status of periodontal health.

No studies have recorded each patient's periodontal status before crown insertion or linked the initial periodontal tissue condition to the response after restoration insertion (11). Few studies have been

published on the reaction of marginal periodontal tissues and little information is available on the molecules mediating the mechanism of such an effect (1,2).

Kourkouta et al. (12) evaluated the response of microbial plaque and gingival inflammation to the placement of porcelain laminate veneer on anterior teeth. They measured the volume of GCF, level of neutral proteolytic enzyme activity, GI, P1I and plaque bacteria vitality after placement of veneers. They found no statistically significant differences in proteolytic enzyme activity or GI ($p>0.05$). Our results support the findings of Kourkouta et al.

These results reconfirmed the hypothesis that dentists should select alloys that have the lowest release of elements. This goal can be achieved by using high noble or noble alloys. Further research in this field is required, over longer periods of time, to assess the clinical performance of dental casting alloys and all-ceramics and their influence on gingival health.

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