

Immunoglobulin Levels and Periodontal Diseases-A Clinical Immunological Study

Prathiba Chichurakanahalli Srinivasan*

¹DNSVK, Sri Venkateshwara Dental College, Bangalore, India

Abstract

There is little doubt that immunological mechanisms play an important role in the pathogenesis of periodontal diseases. Studies conducted so far have yielded contradicting results with regard to the immunoglobulin levels and also varying results after therapy. Hence, the present study was undertaken to study the levels of immunoglobulins-G, A and M in the serum and saliva of patients with periodontal disease. 40 systemically healthy subjects-10 cases of chronic periodontitis with 10 age and sex-matched controls and 10 cases of aggressive periodontitis with 10 age and sex-matched controls were included in the study. The serum and salivary Ig-G, A, and M levels were analyzed by immunoturbidimetry before and 6-8 weeks after Phase I therapy. In both the groups, there was increase in the immunoglobulin levels in cases compared to the controls, but individual variations were observed. There was a modest decline in the immunoglobulin levels after phase I therapy, but in some cases the levels increased after therapy. Therefore, further long-term studies with a larger sample population and with more definitive treatment procedures like periodontal surgery should be undertaken.

Keywords: Immunoglobulins; Periodontal diseases; Serum; Saliva

Introduction

Periodontal disease is considered to be a mixed infection wherein the pathogens act directly or indirectly in the destruction of the tooth-supporting tissues. The host reacts to this bacterial challenge by activating its defense mechanisms in an attempt to localize and eventually eliminate the pathogens [1]. The immune responses can be mediated either by antibodies (humoral) or by sensitized lymphocytes (cellular).

Antibodies belong to the third fastest migrating group of serum globulins, the gamma globulins. The term Immunoglobulin (Ig) refers to the immunity-conferring portion of the gamma globulin fraction [2]. Based on physicochemical and antigenic differences, five classes of immunoglobulins have been recognized—IgG, IgA, IgM, IgD and IgE [3]. These immunoglobulins contribute to the inhibition of bacterial adherence and colonization, enhance bacterial phagocytosis, and help detoxify bacterial toxins and thus play a major role in the defense against bacterial infections [4]. The inflammatory and immune responses clearly contribute to the maintenance of homeostasis between the host and the microbial biofilm of the periodontium [5]. For the host to maintain homeostasis within the oral cavity, three distinct but interrelated immune responses contribute to controlling the microbial challenge. These are the salivary and gingival tissue (local) and the serum (systemic) immune systems [6].

According to Lehner, immunological responses (through local secretory and systemic serum antibodies) can be mediated by three related fluid compartments: Saliva, crevicular fluid and blood. Hence, immunoglobulins if present, should be detected in these fluid compartments [7].

Studies with evaluation of either serum or salivary quantitation of immunoglobulins have provided varying results. Some studies revealed increased serum IgG, IgA and IgM in patients with periodontitis [8-14], while others showed no significant differences in serum Ig levels between periodontitis patients and healthy individuals [15-17]. A study conducted by Kaslick et al. [18] revealed increased levels of serum

IgA, IgG and IgM in periodontitis patients, but paradoxically 41% of patients had no increase in IgG, IgA or IgM levels.

Studies revealed increased salivary IgA in periodontitis patients [19-21], elevated salivary IgG and A levels in severe periodontitis patient [16], and another study showed salivary IgG and IgA to be elevated in juvenile periodontitis patients [22]. Contradicting these studies, study by Basu MK et al. [23] revealed decrease in salivary IgA in periodontitis patients compared to healthy individuals.

Study by Bratthal GT and Ellen RP [24] revealed elevated salivary and crevicular antibodies to periodontal pathogens after conventional gingivitis treatment. Reiff RL [7] stated that levels of salivary and serum Ig G and A declined after Phase I therapy, but in the same study, some study subjects revealed elevations in the immunoglobulin levels after therapy. Basu MK et al. [23] observed higher salivary IgG and lower salivary IgA levels in periodontitis patients before oral hygiene therapy. The concentrations of these immunoglobulins after periodontal therapy was comparable with those found in clinically normal individuals.

Since the above mentioned studies have yielded varying results, the present study is undertaken to estimate the total salivary and serum levels of Ig G, A and M in chronic and aggressive periodontitis cases followed by estimation of the same 6-8 weeks after phase I therapy and to compare the levels before and after phase I therapy.

Materials and Methods

Based on the criteria, 40 patients were recruited for the study. Since the study required collection of blood and saliva samples,

*Corresponding author: Prathiba CS, NSVK, Sri Venkateshwara Dental College, #25, 6th main, 13th 'A' Cross, Vyalikaval, Bangalore-560003, Karnataka, India, Tel: (+91) 9845485963; E-mail: csprati@yahoo.com

Received July 23, 2012; Published August 31, 2012

Citation: Srinivasan PC (2012) Immunoglobulin Levels and Periodontal Diseases-A Clinical Immunological Study. 1: 254. doi:[10.4172/scientificreports.254](http://dx.doi.org/10.4172/scientificreports.254)

Copyright: © 2012 Srinivasan PC. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

written informed consent from patients and ethical clearance from the institution were obtained.

Inclusion criteria

1. Male and female patients between 15 and 50 years of age.
2. Presence of loss of attachment and pocket probing depth greater than or equal to 4 mm prior to Phase I therapy.
3. Healthy individuals with no signs and/or symptoms of systemic disease.
4. Nonsmokers.
5. Individuals who have not undergone professional oral prophylaxis during the last one year.
6. Patients who have not received any antibiotic therapy 6 months prior to the commencement of the study.
7. Individuals included in the control group were systemically healthy individuals with good oral hygiene and no sign of periodontal disease, i.e., this group included subjects with a "healthy periodontium"—Gingival index (Loe and Silness 1963) of <0.5 and with no periodontal pockets exceeding 3 mm.
8. Patients who were compliant and willing to return after the Phase I therapy.

Exclusion criteria

1. Patients who are suffering from any systemic diseases (e.g. diabetes mellitus, connective tissue disorders like rheumatoid arthritis).
2. Patients who are immunocompromised (e.g. HIV positive, primary and secondary immunodeficiency disorders, malnutrition etc).
3. Patients with a history of upper respiratory diseases of recent occurrence (within 4 weeks), allergic disorders or autoimmune disorders, and
4. Patients who are on corticosteroid medications or on cytotoxic drugs.

A detailed medical and dental history was elicited from all the patients. The gingival index (Loe and Silness) values were determined and recorded. The Shick and Ash Plaque index was used to assess the plaque. Probing pocket depth (gingival margin to the base of the gingival sulcus or pocket) was measured to the nearest mm on 6 sites per tooth using a William's periodontal probe and recorded. Based on the clinical parameters assessed, the subjects were grouped as follows:

Group A: Comprised of 20 systemically healthy subjects- 10 cases of chronic periodontitis and an equal number of age and sex-matched controls who had no evidence of periodontal destruction.

Group B: Comprised of 20 systemically healthy subjects-10 cases of aggressive periodontitis and an equal number of age and sex-matched controls with no evidence of periodontal destruction. Before commencement of the treatment procedure, saliva and blood samples were collected from all the patients.

Method of collection and storage of saliva samples

The patients were informed not to eat or drink one hour before the collection of the saliva sample. Whole (mixed) resting/unstimulated

saliva was collected in a sterile glass jar. About 3-4 ml of saliva was collected by the "draining or spitting method". The subject was asked to accumulate saliva in the floor of the mouth and then expectorate saliva into a sterile glass jar. The saliva samples were centrifuged at 2500 g for 5 minutes to spin down the heavy mucous and other particles. The supernatant was pipetted into a sterile, dry screw-capped bottle and stored at -20 degree centigrade until it was analyzed.

Method of collection of blood samples, preparation and storage of serum

Blood was drawn by the Venipuncture technique from the antecubital fossa. The puncture site was cleaned with antiseptic and a tourniquet was placed around the upper arm 3-4 inches above the Venipuncture site to apply pressure and restrict the blood flow through the vein. A 5 ml syringe with a 21-gauge needle was used to draw about 2-3 ml of blood. Fasting blood samples were collected. The blood was transferred to a test tube and allowed to clot. The clotted blood samples were centrifuged at 3000 RPM for 10-15 minutes. The serum was pipetted into a sterile, dry screw-capped bottle and stored at -70 degree centigrade.

Estimation of immunoglobulin concentrations

Immunoturbidimetry, an automated procedure was employed to estimate the levels of immunoglobulins. The reagents provided in the kit that were specific for IgG, IgA and IgM were pipetted along with the serum and saliva samples separately for the estimation of IgG, IgA and IgM. The pipetting parameters specified by the manufacturer for each immunoglobulin was followed. The cassettes were then introduced into the THE HITACHI 704 ANALYSER. The results were obtained as a digital output on the computer monitor.

Phase I therapy comprising of scaling and root planning was performed for the patients. Oral hygiene instructions were given. The patients were then dismissed and recalled after 6-8 weeks for re-evaluation. The oral hygiene maintenance was assessed by the Shick and Ash Plaque index (score less than 1.0). If the patients had achieved a good level of plaque control then the samples of saliva and blood were collected again and analyzed for the levels of immunoglobulins.

Results

In the chronic periodontitis group, the serum IgG level in 8 out of the 10 cases was higher than their controls, in 2 cases below the levels in the control, but still within the normal range. After therapy, the levels increased in 4 out of the 10 cases (Table 1).

In 8 out of the 10 cases, the serum IgA levels were higher than the controls and in 2 cases lower than the controls. After therapy, the levels increased in 3 out of the 10 cases (Table 2). In 8 out of the 10 cases, the serum IgM levels were higher than the controls and in 2 cases the IgM levels were lower than the controls-in one patient the level was below the normal range. After therapy, the levels increased in 3 cases (Table 3).

In 9 cases, the salivary IgG levels were higher than the controls. In one case, the level was lower than the control. After therapy, the levels were increased in 5 cases (Table 4). In one case, the level of salivary IgA was lower than the control levels. After therapy, increased levels were observed in 3 cases (Table 5). In one case, the salivary IgM level was lower than the control. After therapy, the levels were increased in 3 cases (Table 6).

In the aggressive periodontitis group, the serum IgG levels in one case was below the control, but still within the normal range. After

SI No	Patient (cases)	OPD No	Age/Sex	IgG levels Before therapy	IgG levels after therapy	SI No	Patient (controls)	OPD No	IgG levels	Difference b/n cases & controls	Difference b/n Before and after therapy
1	Case 01	L-961	47/F	1986	1919	1	Control 01	L-993	1608	378	67
2	Case 02	N-505	36/F	2166	1987	2	Control 02	R-668	1668	498	179
3	Case 03	M-924	36/M	1073	1084	3	Control 03	L-458	1341	-268	+11(increased)
4	Case 04	N-491	43/F	2157	2136	4	Control 04	R-956	1834	323	21
5	Case 05	Q-358	45/F	1313	1326	5	Control 05	L-558	1430	-117	+13 (increased)
6	Case 06	S-684	40/F	1908	1900	6	Control 06	Y-28	1565	343	8
7	Case 07	S-804	40/F	1924	1911	7	Control 07	Z-202	1720	204	13
8	Case 08	T-17	40/M	2057	2068	8	Control 08	X-206	1320	737	+11(increased)
9	Case 09	S-416	40/F	1599	1602	9	Control 09	R-42	1313	286	+3(increased)
10	Case 10	T-43	40/M	1988	1982	10	Control 10	S-48	1328	660	6

In 8 out of the 10 cases, the IgG levels were higher than their controls, in 2 cases the levels were below the levels in the control, but still within the normal range. After therapy, the levels increased in 4 out of the 10 cases.

Table 1: Serum IgG levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

SI No	Patient (cases)	OPD No	Age/Sex	IgA levels Before therapy	IgA levels after therapy	SI No	Patient (controls)	OPD No	IgA levels	Difference b/n Cases & ontrls	Difference b/n before and after therapy
1	Case 01	L-961	47/F	356	349	1	Control 01	L-993	254	102	7
2	Case 02	N-505	36/F	343	338	2	Control 02	R-668	292	51	5
3	Case 03	M-924	36/M	262	268	3	Control 03	L-458	284	-22	+6 (increased)
4	Case 04	N-491	43/F	294	282	4	Control 04	R-956	268	26	12
5	Case 05	Q-358	45/F	209	219	5	Control 05	L-558	230	-21	+10 (increased)
6	Case 06	S-684	40/F	336	308	6	Control 06	Y-28	228	108	28
7	Case 07	S-804	40/F	342	338	7	Control 07	Z-202	242	100	4
8	Case 08	T-17	40/M	347	339	8	Control 08	X-206	218	129	8
9	Case 09	S-416	40/F	267	280	9	Control 09	R-42	209	58	+13 (increased)
10	Case 10	T-43	40/M	338	334	10	Control 10	S-48	226	112	4

In 8 out of the 10 cases, the IgA levels were higher than the controls and in 2 cases the IgA levels were lower than the controls. After therapy, the levels increased in 3 out of the 10 cases.

Table 2: Serum IgA levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

SI No	Patient (cases)	OPD No	Age/ Sex	IgM levels Before therapy	IgM levels After therapy	SI No	Patient (controls)	OPD No	IgM levels	Difference b/n cases & controls	Difference b/n Before and after therapy
1	Case 01	L-961	47/F	172	169	1	Control 01	L-993	136	36	3
2	Case 02	N-505	36/F	158	149	2	Control 02	R-668	123	35	9
3	Case 03	M-924	36/M	42	90	3	Control 03	L-458	160	-118	+48 increased
4	Case 04	N-491	43/F	196	183	4	Control 04	R-956	153	43	13
5	Case 05	Q-358	45/F	111	119	5	Control 05	L-558	136	-25	+8 increased
6	Case 06	S-684	40/F	154	148	6	Control 06	Y-28	128	26	6
7	Case 07	S-804	40/F	167	156	7	Control 07	Z-202	139	28	11
8	Case 08	T-17	40/M	187	179	8	Control 08	X-206	136	51	8
9	Case 09	S-416	40/F	127	154	9	Control 09	R-42	111	16	+27 increased
10	Case 10	T-43	40/M	156	151	10	Control 10	S-48	140	16	5

In 8 out of the 10 cases, the IgM levels were higher than the controls and in 2 cases the IgM levels were lower than the controls-in one patient the level was below the normal range. After therapy, the levels increased in 3 cases.

Table 3: Serum IgM levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

therapy, the levels increased in 4 cases (Table 7). The IgA levels in one case, was below the levels in the control, but still within the normal range. After therapy, the levels increased in 3 cases (Table 8). In all the 10 cases, the IgM levels were higher than the controls. After therapy, the levels were raised in 5 cases (Table 9). The Salivary IgG in all the 10 cases was higher than in the controls. After therapy, the levels were increased in 3 cases (Table 10). Likewise the salivary IgA and IgM levels in all the cases were higher than the controls. After therapy, the levels were raised in 2 cases (Table 11 and Table 12 respectively).

Discussion

It is pretty much agreed that the immune system is involved in

the pathogenesis of periodontal disease. The literature is replete with studies involving the immunoglobulin levels in different forms of periodontal diseases and these studies have yielded varying results.

In the present study, individual variations have been observed with regard to the immunoglobulin levels, i.e., although the serum and salivary levels are elevated in most of the cases, there are some exceptions where the levels were lesser than the controls, but still within the normal range. After therapy, in some cases there was a decline in the levels while in some others there was an increase.

The data collected in the study was analyzed statistically, by computing the necessary statistics like mean, standard deviation,

SI no	Patient (cases)	OPD No	Age/Sex	IgG levels Before therapy	IgG levels	SI No	Patient (controls)	OPD No	IgG levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 01	L-961	47/F	14.6	16.2	1	Control 01	L-993	11.92	2.68	+1.6(increased)
2	Case 02	N-505	36/F	17.9	15.2	2	Control 02	R-668	14.2	3.7	2.7
3	Case 03	M-924	36/M	2.19	3.1	3	Control 03	L-458	3.6	-1.41	+0.91 increased
4	Case 04	N-491	43/F	3	3.6	4	Control 04	R-956	2.67	0.33	+0.6 increased
5	Case 05	Q-358	45/F	8.1	7.2	5	Control 05	L-558	4.2	3.9	0.9
6	Case 06	S-684	40/F	18.2	17.92	6	Control 06	Y-28	16.9	1.3	0.28
7	Case 07	S-804	40/F	18.6	17.9	7	Control 07	Z-202	11.2	7.4	0.7
8	Case 08	T-17	40/M	9.3	9.8	8	Control 08	X-206	8.2	1.1	+0.5 increased
9	Case 09	S-416	40/F	27.1	29.2	9	Control 09	R-42	26.1	1	+2.1 increased
10	Case 10	T-43	40/M	10.1	9.8	10	Control 10	S-48	8.2	1.1	0.5

In 9 cases, the levels were higher than the controls. In one case, the level was lower than the control. After therapy, the levels were increased in 5 cases.

Table 4: Salivary IgG levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

SI no	Patient (cases)	OPD No	Age/Sex	IgA levels Before therapy	IgA levels after therapy	SI No	Patient (controls)	OPD No	IgA levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 01	L-961	47/F	38.2	30.6	1	Control 01	L-993	32.6	5.6	7.6
2	Case 02	N-505	36/F	30.2	28.6	2	Control 02	R-668	24.6	5.6	1.6
3	Case 03	M-924	36/M	31.19	32.65	3	Control 03	L-458	37.2	-6.01	+1.46 increased
4	Case 04	N-491	43/F	37.3	35.2	4	Control 04	R-956	32.5	4.8	2.1
5	Case 05	Q-358	45/F	39	40.1	5	Control 05	L-558	32.2	6.8	+1.1 increased
6	Case 06	S-684	40/F	29.1	28.1	6	Control 06	Y-28	26.2	7.6	1.2
7	Case 07	S-804	40/F	32.1	31	7	Control 07	Z-202	25.1	7	1.1
8	Case 08	T-17	40/M	32.3	30.6	8	Control 08	X-206	28.6	3.7	1.7
9	Case 09	S-416	40/F	42.8	56.3	9	Control 09	R-42	38.2	4.6	+13.5 increased
10	Case 10	T-43	40/M	33.8	32.6	10	Control 10	S-48	26.2	7.6	1.7

In one case, the levels of IgA were lower than the control levels. After therapy, increased levels were observed in 3 cases.

Table 5: Salivary IgA levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

SI No	Patient (cases)	OPD No	Age/Sex	IgM levels Before therapy	IgM levels after therapy	SI No	Patient (controls)	OPD No	IgM levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 01	L-961	47/F	9.8	9.1	1	Control 01	L-993	8.6	1.2	0.7
2	Case 02	N-505	36/F	11.6	10.2	2	Control 02	R-668	8.4	3.2	1.4
3	Case 03	M-924	36/M	10.6	11.2	3	Control 03	L-458	11.8	-1.2	+0.6 increased
4	Case 04	N-491	43/F	9.82	8.9	4	Control 04	R-956	8.1	1.72	0.92
5	Case 05	Q-358	45/F	11.4	11.6	5	Control 05	L-558	9.8	1.6	+0.2 increased
6	Case 06	S-684	40/F	9.2	8.6	6	Control 06	Y-28	8.2	1	0.6
7	Case 07	S-804	40/F	11.6	10.8	7	Control 07	Z-202	9.1	2.5	0.8
8	Case 08	T-17	40/M	8.6	7.9	8	Control 08	X-206	7.2	1.4	0.7
9	Case 09	S-416	40/F	9	9.8	9	Control 09	R-42	8.6	0.4	+0.8 increased
10	Case 10	T-43	40/M	9.1	8.8	10	Control 10	S-48	6.9	2.2	0.3

In one case, the IgM level was lower than the control. After therapy, the levels were increased in 3 cases.

Table 6: Salivary IgM levels (mg/dl) in Chronic Periodontitis patients before and after therapy and in their controls.

standard error of mean, and 95% confidence interval for mean. Unpaired student t-test (** in Tables 13-16) is employed to compare immunoglobulin levels between the cases and controls and paired t-test (* in Tables 13-16) is used to compare the immunoglobulin levels in the cases before and after therapy. The results are considered statistically significant whenever $p \leq 0.05$.

Figure 1 and Table 13 summarizes the serum immunoglobulin levels of chronic periodontitis cases before and after therapy. The levels of IgG ($p < 0.033$) and IgA ($p < 0.002$) were significantly higher than in the controls. The IgM levels were not significantly raised in the cases. ($p > 0.482$). This finding is in contrast to the results of the studies conducted by Tortelli A et al. [10], Seidlova et al. [12] and Anil S et al. [13] but is in agreement with the study conducted by Bokor-Bratic M [17]. The probable cause for increased IgG levels may be due to their increased production to neutralize bacterial toxins.

After therapy, there was no significant decline in the serum levels of IgG ($p > 0.202$) and IgA ($p > 0.323$), these results in agreement with the results of Reiff RL [7] and Papapanou et al. [25] for IgG, (the latter study concluded that despite successful periodontal therapy, titers remained elevated over a 30-month period) and in agreement with the study of Reiff RL [7] for IgA. There was no significant decline in IgM levels after therapy ($p > 0.665$). The mean IgM value before therapy was 147.00 and the value after therapy was 149.80. In 3 of the 10 of the cases the levels increased after Phase I therapy.

Figure 2 and Table 14 summarizes the levels of salivary immunoglobulins in chronic periodontitis cases before and after therapy. The salivary IgG levels in the cases was not significantly higher than in the controls ($p > 0.510$). This is in contrast to the results obtained in the study conducted by Basu MK et al. [23]. The salivary IgA levels

Sl no	Patient (cases)	OPD No	Age/Sex	IgG levels before therapy	IgG levels after therapy	Sl No	Patient (controls)	OPD No	IgG levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	1608	1501	1	Control 11	R-605	1436	172	107
2	Case 12	Q-481	36/F	1986	1990	2	Control 12	L-456	1656	330	+4 increased
3	Case 13	P-13	28/F	1194	1213	3	Control 13	R-998	1436	-242	+19 increased
4	Case 14	R-932	18/F	1644	1638	4	Control 14	Z-987	1236	408	6
5	Case 15	R-837	33/F	2125	2116	5	Control 15	X-889	1470	655	9
6	Case 16	R-810	33/F	1980	1946	6	Control 16	R-987	1525	455	34
7	Case 17	W-869	17/F	1612	1605	7	Control 17	L-334	1200	412	7
8	Case 18	Y-30	28/F	1592	1616	8	Control 18	L-887	1320	272	+24 increased
9	Case 19	Y-161	24/M	1482	1560	9	Control 19	R-779	1338	144	+78 increased
10	Case 20	L-774	18/M	1702	1693	10	Control 20	Y-23	1392	310	8

In one case, the levels were below the control, but still within the normal range. After therapy, the levels increased in 4 cases.

Table 7: Serum IgG levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

Sl no	Patient (cases)	OPD No	Age/Sex	IgA levels before therapy	IgA levels after therapy	Sl No	Patient (controls)	OPD No	IgA levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	263	250	1	Control 11	R-605	240	23	13
2	Case 12	Q-481	36/F	342	338	2	Control 12	L-456	272	70	4
3	Case 13	P-13	28/F	154	160	3	Control 13	R-998	202	-48	+6 increased
4	Case 14	R-932	18/F	407	400	4	Control 14	Z-987	186	221	7
5	Case 15	R-837	33/F	346	328	5	Control 15	X-889	268	78	18
6	Case 16	R-810	33/F	370	362	6	Control 16	R-987	298	72	8
7	Case 17	W-869	17/F	312	299	7	Control 17	L-334	200	112	13
8	Case 18	Y-30	28/F	282	294	8	Control 18	L-887	238	44	+12 increased
9	Case 19	Y-161	24/M	268	272	9	Control 19	R-779	192	76	+4 increased
10	Case 20	L-774	18/M	228	220	10	Control 20	Y-23	196	32	8

In one case, the levels were below the control, but still within the normal range. After therapy, the levels increased in 3 cases.

Table 8: Serum IgA levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

Sl No	Patient (cases)	OPD No	Age/Sex	IgM levels Before therapy	IgM levels after therapy	Sl No	Patient (controls)	OPD No	IgM levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	187	160	1	Control 11	R-605	156	31	27
2	Case 12	Q-481	36/F	156	168	2	Control 12	L-456	111	45	+12 increased
3	Case 13	P-13	28/F	148	154	3	Control 13	R-998	110	38	+6 increased
4	Case 14	R-932	18/F	209	198	4	Control 14	Z-987	108	101	11
5	Case 15	R-837	33/F	200	196	5	Control 15	X-889	140	60	4
6	Case 16	R-810	33/F	159	156	6	Control 16	R-987	137	22	3
7	Case 17	W-869	17/F	216	200	7	Control 17	L-334	148	68	+16 increased
8	Case 18	Y-30	28/F	166	168	8	Control 18	L-887	142	24	+2 increased
9	Case 19	Y-161	24/M	138	142	9	Control 19	R-779	118	20	+4 increased
10	Case 20	L-774	18/M	212	199	10	Control 20	Y-23	106	106	13

In all the 10 cases, the IgM levels were higher than the controls. After therapy, the levels were raised in 5 cases.

Table 9: Serum IgM levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

Sl no	Patient (cases)	OPD No	Age/Sex	IgG levels Before therapy	IgG levels after therapy	Sl No	Patient (controls)	OPD No	IgG levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	13.6	12.2	1	Control 11	R-605	9.2	4.4	1.4
2	Case 12	Q-481	36/F	18.2	21.3	2	Control 12	L-456	15.9	2.3	+3.1 increased
3	Case 13	P-13	28/F	24.5	26.8	3	Control 13	R-998	14.9	9.6	+2.3 increased
4	Case 14	R-932	18/F	29.6	24.3	4	Control 14	Z-987	19.2	10.4	5.3
5	Case 15	R-837	33/F	28.2	26.1	5	Control 15	X-889	14.9	13.3	2.1
6	Case 16	R-810	33/F	15.9	14.2	6	Control 16	R-987	12.6	3.3	1.7
7	Case 17	W-869	17/F	18.2	17.6	7	Control 17	L-334	14.2	4	0.6
8	Case 18	Y-30	28/F	16.8	18.2	8	Control 18	L-887	11.6	5.2	+1.4 increased
9	Case 19	Y-161	24/M	14.6	16.8	9	Control 19	R-779	9.1	5.5	2.2
10	Case 20	L-774	18/M	12.2	10.8	10	Control 20	Y-23	8.2	4	1.4

In all the 10 cases, the levels were above the controls. After therapy, the levels were increased in 3 cases.

Table 10: Salivary IgG levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

SI no	Patient (cases)	OPD No	Age/Sex	IgA levels before therapy	IgA levels after therapy	SI No	Patient (controls)	OPD No	IgA levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	43.8	40.6	1	Control 11	R-605	38.8	5	3.2
2	Case 12	Q-481	36/F	38.1	40.2	2	Control 12	L-456	28.2	9.9	+2.1 increased
3	Case 13	P-13	28/F	39	42.2	3	Control 13	R-998	40.2	-1.2	+3.2 increased
4	Case 14	R-932	18/F	41.7	39.6	4	Control 14	Z-987	38.6	3.1	2.1
5	Case 15	R-837	33/F	42	40.2	5	Control 15	X-889	38.8	3.2	1.8
6	Case 16	R-810	33/F	48.1	46.2	6	Control 16	R-987	32.8	15.3	1.9
7	Case 17	W-869	17/F	28.4	24.2	7	Control 17	L-334	19.2	9.2	4.2
8	Case 18	Y-30	28/F	29.2	26.1	8	Control 18	L-887	22.6	6.6	3.1
9	Case 19	Y-161	24/M	33.6	30.1	9	Control 19	R-779	28.2	5.4	3.5
10	Case 20	L-774	18/M	38.2	32.6	10	Control 20	Y-23	26.1	12.1	5.6

In all the cases the levels were higher than the controls. After therapy, the levels were raised in 2 cases.

Table 11: Salivary IgA levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

SI no	Patient (cases)	OPD No	Age/Sex	IgM levels before therapy	IgM levels after therapy	SI No	Patient (controls)	OPD No	IgM levels	Difference b/n cases & controls	Difference b/n before and after therapy
1	Case 11	M-927	30/F	10.18	10	1	Control 11	R-605	9.2	0.98	0.18
2	Case 12	Q-481	36/F	11.2	11.9	2	Control 12	L-456	9.1	2.1	+0.7 increased
3	Case 13	P-13	28/F	10	11.3	3	Control 13	R-998	9.1	0.9	+1.3 increased
4	Case 14	R-932	18/F	9.3	8.6	4	Control 14	Z-987	4.6	4.7	0.7
5	Case 15	R-837	33/F	11.4	10.6	5	Control 15	X-889	8.8	2.6	0.8
6	Case 16	R-810	33/F	12.9	10.2	6	Control 16	R-987	9.1	3.8	2.7
7	Case 17	W-869	17/F	8.2	7.6	7	Control 17	L-334	4.8	3.4	0.6
8	Case 18	Y-30	28/F	9.1	8.3	8	Control 18	L-887	6.2	2.9	0.8
9	Case 19	Y-161	24/M	10.2	7.9	9	Control 19	R-779	5.8	4.4	2.3
10	Case 20	L-774	18/M	9.8	7.6	10	Control 20	Y-23	4.9	4.9	2.2

In all the 10 cases, the levels were above the control levels. In 2 cases, the levels were raised after therapy.

Table 12: Salivary IgM levels (mg/dl) in Aggressive Periodontitis patients before and after therapy and in their controls.

		Sample size	Mean	Std. Deviation	Std. Error Mean	t-value	Df	p-value®
IgG	Before therapy	10	1817.10	369.325	116.791	1.377*	9	>0.202
	After therapy	10	1791.50	344.014	108.787			
	Before therapy	10	1817.10	369.325	116.791	2.314**	18	<0.033
	Control	10	1512.70	191.366	60.515			
IgA	Before therapy	10	309.40	49.063	15.515	1.046*	9	>0.323
	After therapy	10	305.50	42.212	13.349			
	Before therapy	10	309.40	49.063	15.515	3.587**	18	<0.002
	Control	10	245.10	28.380	8.975			
IgM	Before	10	147.00	44.719	14.141	0.447*	9	>0.665
	After	10	149.80	27.708	8.762			
	Before	10	147.00	44.719	14.141	0.729**	18	>0.482
	Control	10	136.20	13.935	4.407			

Table 13: Statistical inference based on t-test of disease-Chronic Periodontitis (serum).

is significantly higher than in the controls ($p < 0.059$). This finding correlates with the results of the study conducted by Guven et al. [19], Lindstrom et al. [20] and Nagasawa T et al. [21]. IgM levels in saliva of patients is significantly higher than the controls ($p < 0.024$). This result is in contrast to the study conducted by Yavuzilmaz E et al. [26].

Enzymes that cleave the IgG-the proteases, hypothesized as important virulence factors of bacterial pathogens [27]. This may be the reason for the decline in the IgG levels. Prolonged antigenic stimulus in an infectious condition like periodontal disease may also stimulate the local IgA immune system [20]. Local immunoglobulin synthesis in the inflamed gingiva in periodontitis patients produces mainly IgG and IgA, not IgM. Thus the elevated IgM levels may be due to the raised level of glandular secretory output or from increased leakage from the blood via the gingival sulcus [20].

The mean level of salivary IgG before therapy is 12.91 and after therapy is 12.99 ($p > 0.853$). In 3 out of the 10 of the cases the levels of salivary IgG and IgA ($p > 0.994$) increased after Phase I therapy. This finding is in agreement with the results obtained in the study conducted by Reiff RL [7], where there was a reduction in levels in most of the cases, but there was increase in levels as well in few cases following Phase I therapy. The results are in contrast to the results of the study conducted by Basu MK et al. [23]. There was a statistically significant decline in the salivary IgM levels in patients following Phase I therapy ($p < 0.035$). The most probable reason for this is the reduction in the acute phase of the infection.

Increase in the salivary Ig G, A and M levels in some cases after therapy can be attributed to the the following causes: Scaling itself may cause a transient rise in the blastogenic response [7] or inoculation of the microorganism into the host tissues resulting from scaling can

		Sample Size	Mean	Std. Deviation	Std. Error Mean	t-value	Df	p-value®
IgG	Before therapy	10	12.91	7.794	2.465	0.190*	9	>0.853
	After therapy	10	12.99	7.932	2.508			
	Before	10	12.91	7.794	2.465	0.672**	18	>0.510
	Control	10	10.66	7.169	2.267			
IgA	Before	10	34.59	4.481	1.417	0.008*	9	>0.994
	After	10	34.58	8.383	2.651			
	Before	10	34.59	4.481	1.417	2.014**	18	<0.059
	Control	10	30.34	4.942	1.563			
IgM	Before	10	10.07	1.148	0.363	2.473*	9	<0.035
	After	10	9.61	1.247	0.394			
	Before	10	10.07	1.148	0.363	2.468**	18	<0.024
	Control	10	8.67	1.382	0.437			

Table14: Statistical inference based on t-test of disease-Chronic Periodontitis (saliva).

		Sample size	Mean	Std. Deviation	Std. Error Mean	t-value	Df	p-value®
IgG	Before therapy	10	1692.50	273.827	86.592	0.318*	9	>0.758
	After therapy	10	1687.80	265.105	83.834			
	Before	10	1692.50	273.827	86.592	3.017**	18	<0.007
	Control	10	1400.90	135.749	42.927			
IgA	Before	10	297.20	74.171	23.455	1.632*	9	>0.137
	After	10	292.30	70.553	22.311			
	Before	10	297.20	74.171	23.455	2.556**	18	<0.020
	Control	10	229.20	39.735	12.565			
IgM	Before	10	179.10	29.065	9.191	1.342*	9	>0.213
	After	10	174.10	22.053	6.974			
	Before	10	179.10	29.065	9.191	4.701**	18	<0.0001
	Control	10	127.60	18.857	5.963			

Table15: Statistical inference based on t-test of disease-Aggressive Periodontitis (serum).

		Sample size	Mean	Std. Deviation	Std. Error Mean	t-value	Df	p-value®
IgG	Before therapy	10	19.18	6.123	1.936	0.428*	9	>0.679
	After therapy	10	18.83	5.660	1.790			
	Before	10	19.18	6.123	1.936	2.780**	18	<0.012
	Control	10	12.98	3.501	1.107			
IgA	Before	10	38.21	6.280	1.986	2.336*	9	<0.044
	After	10	36.20	7.417	2.345			
	Before	10	38.21	6.280	1.986	2.206**	18	<0.041
	Control	10	31.35	7.569	2.393			
IgM	Before	10	10.23	1.331	.421	2.035*	9	<0.036
	After	10	9.40	1.594	.504			
	Before	10	10.23	1.331	.421	3.958**	18	<0.001
	Control	10	7.16	2.059	.651			

Table 16: Statistical inference based on t-test of disease-Aggressive Periodontitis (saliva).

lead to elevated titers, or even elimination of the immunosuppressive microorganism can lead to elevated levels after therapy [28].

Figure 3 and Table 15 summarizes the immunoglobulin levels in serum of aggressive periodontitis cases before and after therapy. Serum IgG levels is significantly higher than the levels in controls ($p < 0.007$). This finding correlates with the results obtained in the studies of Lehner et al. [9], Kaslick RS et al. [18], Johnson RJ et al. [11], and Ranney RR et al. [16]. The serum IgA ($p < 0.020$) and the IgM ($p < 0.0001$) is significantly higher than the levels in controls. This finding correlates with the results obtained in the studies of Lehner et al. [9] and Kaslick RS et al. [18] for IgA, and the study by Lehner et al. [9] for IgM.

The elevation in serum IgG levels may be due to the increased antibody production to neutralize bacterial toxins. Brandtzaeg and Kraus reported an increased IgA content of the inflamed gingival, which may be the reason for the increased levels of IgA [13]. The

increased levels of serum IgM may reflect a response to the gram-negative bacteria, most commonly associated with periodontitis [9].

After therapy, the decline in the serum IgG ($p > 0.758$) and IgA ($p > 0.137$) levels in the cases was not statistically significant. This finding is in agreement with the results of the study obtained by Reiff RL [7]. Likewise, there was no significant decline in the serum IgM levels ($p > 0.123$) after therapy.

Figure 4 and Table 16 summarizes the levels of salivary immunoglobulins in aggressive periodontitis patients before and after phase I therapy. The salivary IgG levels ($p < 0.012$) in the cases is significantly higher than the controls and is in agreement with the studies conducted by Ranney RR et al. [16], Sandholm L et al. [22]. The above finding is in contrast to the results obtained in the study conducted by Saxen L et al. [29]. The salivary IgA levels in cases is

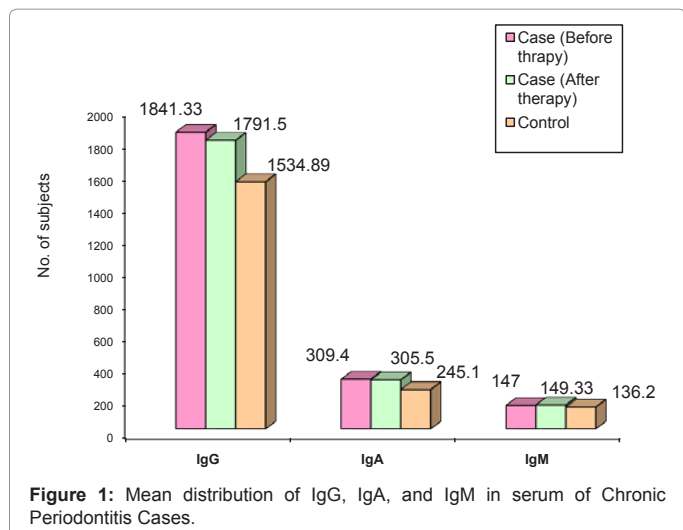


Figure 1: Mean distribution of IgG, IgA, and IgM in serum of Chronic Periodontitis Cases.

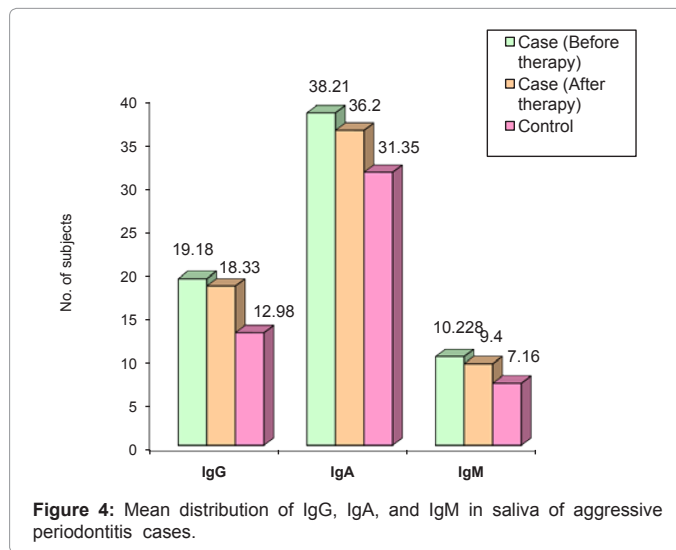


Figure 4: Mean distribution of IgG, IgA, and IgM in saliva of aggressive periodontitis cases.

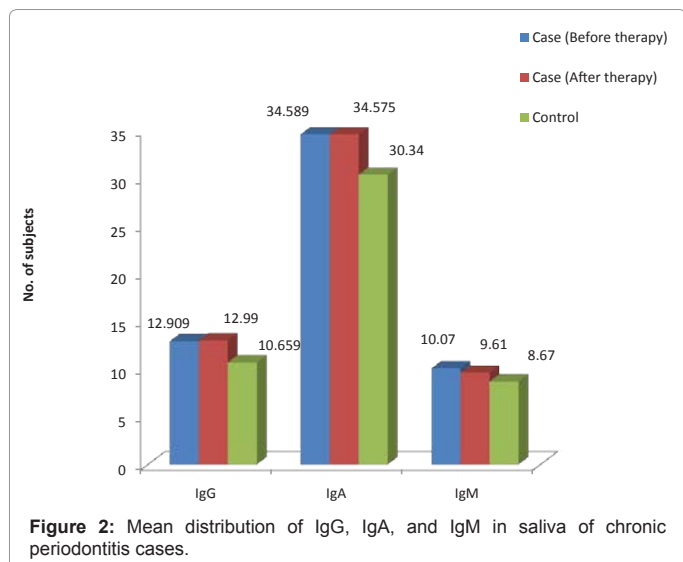


Figure 2: Mean distribution of IgG, IgA, and IgM in saliva of chronic periodontitis cases.

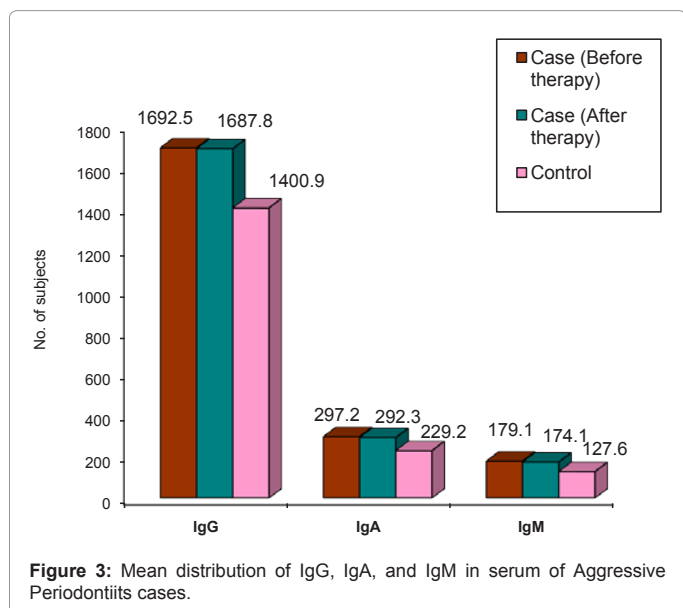


Figure 3: Mean distribution of IgG, IgA, and IgM in serum of Aggressive Periodontitis cases.

significantly higher than the controls ($p < 0.04$). This finding is in agreement with the results of the study conducted by Ranney RR et al. [16], but contradicts the results of study by Sandholm et al. [22] and Saxen L et al. [29]. The salivary IgM levels is significantly higher than the controls ($p < 0.001$). This finding correlates with the results obtained in the study conducted by Sandholm et al. [22], but is in contrast to the results obtained by Saxen L et al. [29].

The increased levels of IgG in saliva of patients with aggressive periodontitis may be due to the predominating synthesis of IgG and the transfer of this and serum-derived IgG from the gingival tissue to the oral cavity. The increased levels of IgA in whole saliva of patients with aggressive periodontitis may be due to the increased leakage of the serum via the inflamed pocket epithelium. Prolonged antigenic stimulus in an infectious condition like periodontal disease may also stimulate the local IgA immune system [7]. Elevated IgM levels may be due to the raised level of glandular secretory output or from increased leakage from the blood via the gingival sulcus [22].

After therapy, although there was a decline in the salivary IgG levels, it was not statistically significant ($p > 0.679$). This is in agreement with the study conducted by Reiff RL [7].

However, the decline in the salivary IgA ($p < 0.044$) and IgM ($p < 0.036$) was statistically significant. This finding is in contrast to the results of the study conducted by Reiff RL [7]. One of the possible causes for this may be the individual patient variation with respect to the oral microflora present at the time of sampling, the varying degrees of periodontal pathology and varying degrees of inflammation present at the site.

The role of immunoglobulins in the pathogenesis of periodontitis is not clear. Several questions remain unanswered. At what stage in the infection is the antibody detected? i.e., it is not clear at what point in the infection and subsequent disease process the initial seroconversion occurs. Once detected should it be considered as a sign of improvement in the condition or decline in the condition? How is the antibody associated with active disease? and Can early immune responses be detected prior to gross infection to enable early institution of therapeutic modalities? Also, the effects of therapy on the levels of immunoglobulins are not clear. Whether the raise in the immunoglobulin levels after Phase I therapy induced by the scaling

procedure is beneficial is unanswered. The long-term study of the disease process from its inception and in its various stages may provide answers to the questions raised.

Hence, further long-term studies with a larger sample population and with advanced immunological techniques have to be undertaken to study the role played by the immunoglobulins in the pathogenesis of periodontitis, to define at-risk population, and to use immunological data for diagnosis, classification and monitoring of periodontal diseases. Long-term follow-up studies will also shed light on the changes in the immunoglobulin levels following various treatment modalities employed for treatment of periodontal diseases.

References

1. Landi L, Amar S, Polins AS, Van Dyke TE (1997) Host mechanisms in the pathogenesis of periodontal disease. *Curr Opin Periodontol* 4: 3-10.
2. Sahingur SE, Cohen RE (2004) Analysis of host responses and risk for disease progression. *Periodontol* 2000 34: 57-83.
3. Ananthanarayan R, Paniker CKJ (1990) Antibodies-Immunoglobulins. In: *Textbook of Microbiology*. (4th edn), Orient Longman Ltd, Madras, PP: 84-91.
4. Albandar JM, DeNardin AM, Adesanya MR, Winn DM, Diehl SR (2002) Associations of serum concentrations of IgG, IgA, IgM and interleukin-1beta with early-onset periodontitis classification and race. *J Clin Periodontol* 29: 421-426.
5. Ebersole JL, Holt SC, Capelli D, Singer RE (1998) IgA antibody responses to periodontal pathogens in non-human primates. Symposium proceedings-IgA and Periodontal Diseases. Abstracts of IADR symposium Nice, France.
6. Ebersole JL (1996) Immune responses in Periodontal Diseases. In: Wilson TG, Kornman KS (eds) *Fundamentals of Periodontics*. Quintessence Publishing Co Inc, Illinois PP: 109-158.
7. Reiff RL (1984) Serum and salivary IgG and IgA response to initial preparation therapy. *J Periodontol* 55: 299-305.
8. Carvel RI, Halperin V, Wallace JH (1973) Immunological studies in chronic severe alveolar resorptive disease: a report of two young female patients. *J Periodontol* 44: 25-34.
9. Lehner T, Wilton JM, Ivanyi L, Manson JD (1974) Immunological aspects of juvenile periodontitis (periodontosis). *J Periodontal Res* 9: 261-272.
10. Törteli A, Backhausz R, Bruder S (1975) [Electrophoretic study of serum proteins in patients with periodontal diseases]. *Stomatol DDR* 25: 312-316.
11. Johnson RJ, Matthews JL, Stone MJ, Hurt WC, Newman JT (1980) Immunopathology of periodontal disease. I. Immunologic profiles in periodontitis and juvenile periodontitis. *J Periodontol* 51: 705-712.
12. Oláhanska-Seidlová A, Skarlandt P, Mikulecky M, Seymour G (1989) Some immunological findings in adult periodontitis. *Aust Dent J* 34: 417-420.
13. Anil S, Hari S, Remani P, Vijaykumar T (1990) Immunology of chronic generalized periodontitis. 1. Estimation of cellular and humoral immune status. *Indian J Dent Res* 2: 127-132.
14. Wilton JM, Hurst TJ, Sterne JA, Caves J, Tilley C, et al. (1992) Elevated levels of the IgG2 subclass in serum from patients with a history of destructive periodontal disease. A case-control study. *J Clin Periodontol* 19: 318-321.
15. Schenkein HA, Genco RJ (1977) Gingival fluid and serum in periodontal diseases. I. Quantitative study of immunoglobulins, complement components, and other plasma proteins. *J Periodontol* 48: 772-777.
16. Ranney RR, Ruddy S, Tew JG, Welshimer HJ, Palcanis KG, et al. (1981) Immunological studies of young adults with severe periodontitis. I. Medical evaluation and humoral factors. *J Periodontal Res* 16: 390-402.
17. Bokor-Bratić M (1998) The concentration of immunoglobulins A, G, and M in serum of patients with periodontal disease. *Med Pregl* 51: 310-314.
18. Kaslick RS, West TL, Singh SM, and Chasens AI (1980) Serum immunoglobulins in Periodontosis patients. *J Periodontol* 51: 343-344.
19. Güven O, De Visscher JG (1982) Salivary IgA in periodontal disease. *J Periodontol* 53: 334-335.
20. Lindstrom FD, Folke LEA (1973) Salivary IgA in Periodontal disease. *Acta Odontol Scand* 31: 31-34.
21. Nagasawa T, Aramaki M, Ishikawa I (1999) The role of salivary IgA antibody against periodontopathic bacteria. Symposium proceedings-IgA and Periodontal Diseases. Abstracts of IADR symposium 26 June 1998, Nice, France.
22. Sandholm L, Grönblad E (1984) Salivary immunoglobulins in patients with juvenile periodontitis and their healthy siblings. *J Periodontol* 55: 9-12.
23. Basu MK, Fox EC, Becker JF (1976) Salivary IgG and IgA before and after periodontal therapy. A preliminary report. *J Periodontal Res* 11: 226-229.
24. Tynelius-Bratthall G, Ellen RP (1985) Fluctuations in crevicular and salivary anti-*A. viscosus* antibody levels in response to treatment of gingivitis. *J Clin Periodontol* 12: 762-773.
25. Papapanou PN, Neiderud AM, Disick E, Lalla E, Miller GC, et al. (2004) Longitudinal stability of serum immunoglobulin G responses to periodontal bacteria. *J Clin Periodontol* 31: 985-990.
26. Yavuzylmaz E, Yamalik N, Calguner M, Ersoy F, Baykara M, et al. (1992) Clinical and immunological characteristics of patients with rheumatoid arthritis and periodontal diseases. *J Nihon Univ Sch Dent* 34: 89-95.
27. Gregory RL, Kim DE, Kindle JC, Hobbs LC, Lloyd DR (1992) Immunoglobulin-degrading enzymes in localized juvenile periodontitis. *J Periodontal Res* 27: 176-183.
28. Miyasaki KT (1996) Altered leukocyte functions and periodontal disease. In: Haake SK (ed) *Clinical Periodontology*. (8th edn) W.B. Saunders company, Philadelphia PP: 132-150.
29. Saxén L, Tenovuuo J, Vilja P (1990) Salivary defense mechanisms in juvenile periodontitis. *Acta Odontol Scand* 48: 399-407.