

The IgG subclass responses in the phenotypic subsets of the early-onset periodontitis

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I. Introduction

The studies on the host-parasite interactions in the early-onset periodontitis(EOP) have been reviewed by several authors¹⁻⁴⁾. Total serum IgG and IgG subclass responses to various antigenic preparations of these organisms have also been examined in the EOP patients. Many authors reported the elevated IgG or IgG2 subclass levels against the 29kd protein, lipopolysaccharides(LPS), or carbohydrate moiety of the LPS of *Actinobacillus actinomycetemcomitans* (Aa) in the localized juvenile periodontitis(LJP)⁵⁻¹⁷⁾. Elevated IgG subclass such as IgG2 against to Aa antigens has been considered to play a modulating role in localizing the disease^{8,18)}. In the rapidly progressing periodontitis(RPP), the total IgG or IgG2 subclass have also been found to be elevated against LPS or total cell preparations of *Porphyromonas gingivalis*(Pg)¹⁹⁻²⁴⁾. The protective role of the IgG2 subclass in RPP has been questioned due to its poor capacity to fix the complements and the low avidity. The investigators postulated that the failure to successfully eliminate the

infecting organisms might have resulted in the disease progression. The characteristic increase in the IgG2 subclass to these organisms has also implicated the significant role of carbohydrate antigens in the EOP^{16,17,25-27)}.

Despite the considerable informations on the humoral immune responses in the periodontal diseases, there seem to be no extensive studies on the responsiveness of each IgG subclass or various combinations of the IgG subclasses in the EOP patients against the periodontopathic bacteria. Moreover, most of the results are still controversial and inconclusive to our understanding of the immunological mechanisms in the pathogenesis of the EOP. This may be due to the complexity of the disease including the heterogenic nature in clinical manifestations^{28,29)}, thus making it difficult to interpret the laboratory data^{5,29-31)}. Realizing this problem, we have previously attempted to show that the classical EOP could be grouped into four phenotypic subsets which have been demonstrated to be more homogeneous by radiographic criteria we have devised³²⁾. Through the studies based on a

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more defined clinical subsets, one may be able better clarify the immunological aspects occurring in the patients suffering from EOP.

The aim of the present study was to see the total IgG and four IgG subclass responses against *Aa* and *Pg* in the four EOP subforms of EOP or the adult periodontitis(AP).

II. Materials and methods

1. Patients Selection and Serum Sampling

Patients whose clinical profiles have been revised by a newly radiographic criteria have been assigned into four homogeneous subforms as previously described³². 6 patients consisting of 3 patients from subform I(distinctive LJP pattern), 19 from subform II(post-juvenile periodontitis pattern), 16 from subform III(LJP pattern but rapidly progressing), 24 from subform IV(distinctive RPP pattern), and 8 from age-matched AP(20-40 years of age) have randomly been selected for the measurements of the total IgG and each IgG subclass against to *Pg* and the IgG subclass against *Aa*, respectively. For the race and age-matched controls, 50 subjects exhibiting no evidences of the destructive periodontal diseases have been selected. The clinical measurements included the mean bone level(BL) and the radiographic ratio(Ratio) have been performed according to the methods previously described. Peripheral blood was drawn by the venipuncture and the serum samples have be collected by centrifugation and stored at -20°C until used.

2. Antigen Preparations and the Determination of Antibody Titers

Briefly, *Aa* Y4 and *Pg* 381 [kindly provided by Dr,

Katsuji Okuda, Tokyo Dental College, Japan) have been plated on TSBV plate and blood agar plate and grown in CO₂ incubator and anaerobic chamber(Coy Co., MI), respectively. After harvesting, the bacterial cells were formalinized to be used as a whole cell surface antigens. ELISA procedures for the determination of the serum antibody titers have been performed. Briefly, 0.2 ml of bacterial antigens appropriately diluted in the buffer I(0.039 % sodium carbonate, 0.671% sodium bicarbonate, 0.02% sodium azide) were added into each well, incubated for 4 hours at 37°C and stored overnight at 4°C. After washing with the buffer II(0.9 % sodium carbonate, 0.5 ml/L of Tween 20) three times and 0.1 ml of serum samples diluted in the buffer II were added into each well and incubated for 2 hours at room temperature. The plate was washed three times with the buffer II and 0.1 ml of mouse antihuman IgG1, IgG2, IgG3, and IgG4(affinity-purified monoclonal antibody, gamma-chain specific, Sigma Chemicals, Ohio, USA) diluted in the buffer III(0.27 % sodium phosphate dibasic, 0.028 % sodium phosphate monobasic, 0.875% sodium chloride, 0.02 % sodium azide, 0.5 ml/L of Tween 20) were added into each well and incubated for 2 hours at room temperature. After washing three times with buffer II, 0.1 ml goat anti-mouse IgG(heavy/light-chain specific, affinity purified, alkaline phosphatase-conjugated, Calbiochem, Basel, Switzerland) diluted in the buffer III were added into each well and incubated for overnight at room temperature. After the washing plates, 0.2 ml of nitrophenyl phosphate(1 mg/ml) were added into each well and incubated for 30 minutes and finally 0.1 ml of 1N NaOH were added to stop the color reaction. The optical density were measured by ELISA reader with wavelength set at 492 nm. For the total IgG titer, basically the same procedure has been applied except that the goat antihuman IgG(affinity purified, gamma-chain spe-

cific, Calbiochem, Basel, Switzerland) and the rabbit anti-goat IgG (heavy and light chain specific, affinity purified, alkaline phosphatase-conjugated, Sigma Chemicals, Ohio, USA) have been used as the 2nd and the 3rd antibodies, respectively.

To determine the serum IgG antibody titers, optical densities(O.D.) were plotted according to the various dilution factors for the regression analysis and reciprocals of the serum dilution factors at the X-axis intersection of O.D.=1 were expressed as the ELISA unit of each sample. The antibody titers greater than the two-fold values of the mean IgG titers of the control subjects have been regarded as the elevated.

3. Statistical Analysis

For the comparison of the total IgG and the IgG subclass titers among the four EOP subforms and adult periodontitis, analysis of variance (ANOVA) and Tukey test have been performed to seek the statistical significance.

III. Results

Table 1 summarizes the clinical profiles of the patients which showed characteristic features of the four EOP subforms and the adult periodontitis reported in our study³². Mean values and the ranges of the total IgG and the four IgG subclasses to *Pg* in the four EOP subforms and AP are demonstrated in the Table 2. The total IgG titers against to *Pg* of the subforms I & III had a significantly higher values than subforms II and IV($p < 0.05$). Among the IgG subclasses, only the IgG3 levels were significantly higher in the subform I than the subform IV($p < 0.05$). Wide ranges of the antibody titers were noted in all of the EOP subforms and the AP. The prevalence of the patients showing the elevated responses in the total IgG and each IgG subclass were also depicted in the Table 2. Elevated antibody responses were found between 50 % to 100 % of the patients according to the disease types. However, theme values were either greater or smaller with respect to the IgG subclass responses.

Table 1. Distributions of age, gender, mean radiographic bone levels, and ratios of four subforms and adult periodontitis group (69 blood sampling patients)

Subforms	I	II	III	IV	AP
n	3	19	15	24	8
mean age	23.5	34.6	34.5	34.4	36.4
range	22-25	25-40	28-40	24-40	31-40
gender					
male(%)	33.3	42.0	60.0	66.7	62.5
female(%)	66.7	58.0	40.0	33.3	37.5
mean BL*	4.08	4.96	5.85	6.73	4.05
range	3.27-4.61	3.26-7.50	4.42-7.43	3.89-8.90	2.11-6.61
mean Ratio**	2.08	1.51	1.39	1.17	1.11
range	1.11-3.28	1.02-2.45	0.95-2.23	0.80-1.67	0.50-1.92

* represents the mean bone level(BL) of total present in a disease group. Mean bone level of an individual tooth is calculated from the distal interproximal alveolar bone crests(see the Materials and Methods³²)

** represents the mean BL of 1st molars relative to the mean BL of neighboring teeth(premolars and canines, see the Materials and Methods³²)

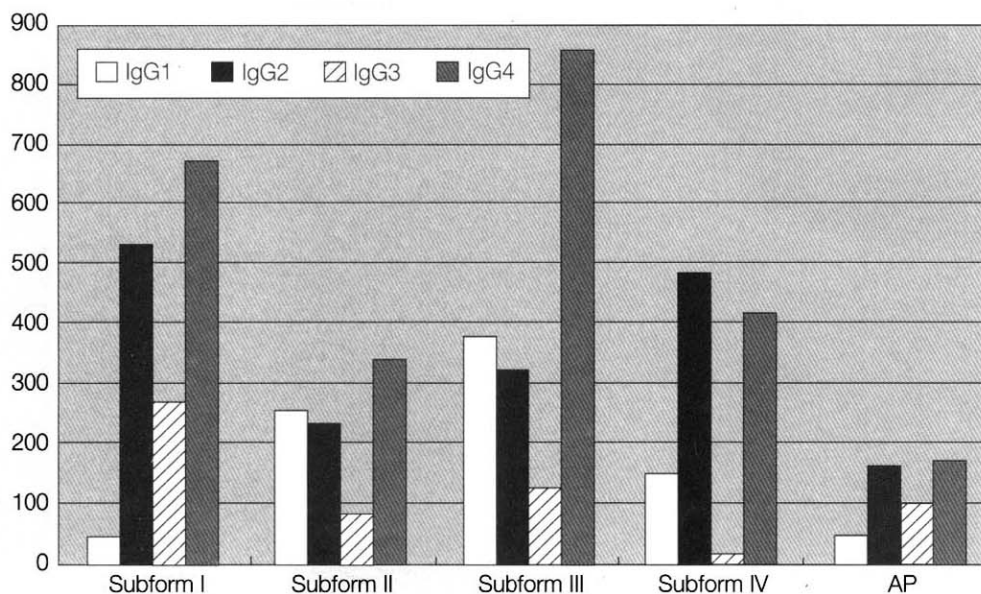


Figure 1. Diagrammatic representation of the mean IgG subclass titers against *Porphyromonas gingivalis* 381 according to the EOP subforms.

Table 2. Mean total IgG and IgG subclass titer to *Porphyromonas gingivalis* and the prevalence of patients showing elevated responses in each subform and the adult periodontitis (range)

	Subforms								AP	
	I n=3		II n=19		III n=15		IV n=24		n=8	
	M	p**	M	p	M	p	M	p	M	p
IgG1*	45.0 (0-134)	0/3	256.6 (0-1403)	8/19	381.3 (0-2190)	7/15	154.7 (0-463)	9/24	53.5 (0-428)	1/8
IgG2*	531.0 (44-1264)	2/3	233.2 (0-844)	9/19	321.3 (49-791)	11/15	488.1 (16-2840)	16/24	65.4@ (0-496)	3/8
IgG3*	270.0 (167-411)	2/3	84.1 (0-485)	4/19	128.1 (0-545)	4/15	16.3 (0-312)	1/24	0.0 (0-0)	0/8
IgG4*	761.0 (487-1006)	3/3	339.4 (0-814)	12/19	859.1 (0-6968)	11/15	419.5 (0-3213)	13/24	178.3 (0-513)	2/8
Total§	15502.3 (11002-17891)	3/3	5590.3 (0-11065)	15/19	10100.3 (1862-25520)	12/15	4172.3 (77-13738)	12/24	7876.9 (4031-12181)	8/8

** prevalences of the patients showing the elevated antibodies

* no significant differences among the four subforms

significantly different between the subform I and the subform IV (p<0.05)

§ significantly different between the subforms I & III and the subforms II & IV (p<0.05)

@ significantly lower than those of the four subforms (p<0.05)

When we looked closely into the IgG subclass titers in the spreading forms(subform II, III, IV), except for the subform I which was typical of localized form, the IgG2 subclass levels to *Pg* gradually became higher in accordance with the subforms II, III and IV(Figure 1, Table 2). If the antibody levels of the EOP we compared with those of AP, both the IgG2 and the IgG4 levels of all four subform were significantly higher, while other subclasses were not. IgG subclass levels were compared according to the different age groups consisting of 18-26, 26-36, and 36-40, respectively(data not shown). All of the four IgG subclass levels to *Pg* were consistently found to be higher in the younger age group around 20. The levels found to be low around the thirties and then gradually became higher at the ages of late thirties. The IgG2 titer to *Aa* in the subform I was significantly higher than those of any other subforms(Table 3). A further analysis has been performed to elucidate the various patterns of ele-

vated IgG subclasses(Table 4). Combinations of IgG1+2+4 were the most frequently found to be elevated followed by the IgG4 only, the IgG2 only, the IgG2+4, the IgG2+3+4, and the IgG1 only, in the descending order. In no patient was a single IgG3 subclass shown to be elevated. When we tested whether or not a single or group of IgG subclasses contributed to the increase in the total IgG of a given patient, this was true for almost all of the cases, though there were a few exceptions(data not shown).

To test the possible relationships of the four IgG subclass responsiveness to *Pg* or the IgG2 responsiveness to *Aa* with the early events occurring in the localized type EOP, we selected thirteen patients from the subforms I and II whose mean bone levels were lower than 5 mm. These patients were subdivided into two groups based on the mean radiographic ratio(Ratio; see the materials and methods). One subgroup consisted of seven patients

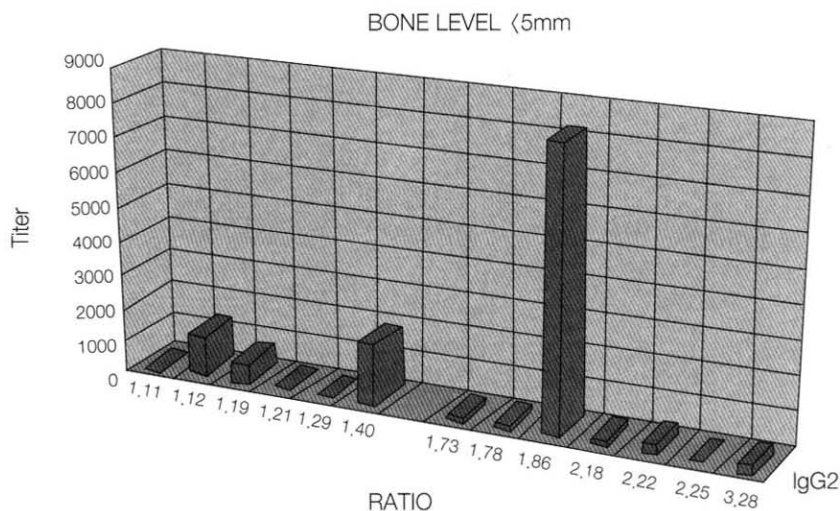


Figure 2. Graphic representation of the IgG2 subclass titers against *Actinobacillus actionmycetemcomitans* Y4 of each patient of the EOP subform I and II patients whose mean bone level < 5mm. Patients were divided into two groups according to the radiographic bone ratio(Ratio); one group having the Ratio greater than 1.5 and the other group having the Ratio smaller than 1.5, respectively.

Table 3. Mean IgG2 titers against *Actinobacillus actionmycetemcomitans* in the EOP subforms(\pm s.d.)

Subforms							
I n=3		II n=19		III n=15		IV n=24	
M	prev**	M	prev	M	prev	M	prev
2867,0*	1/3	966,3	10/19	1539,1	9/15	1783,0	13/24

* significantly higher than all the other subforms($p < 0,05$)

** prevalences of the patients showing the elevated antibody responses

Table 4. Distribution of patients demonstrating various combinations of the elevated IgG subclass to *Porphyromonas gingivalis* according to the four subforms and the adult periodontitis

Subclasses	EOP Subforms				AP	Total
	I	II	III	IV		
1 only	0	2	2	0	0	4
1+2	0	0	0	2	1	3
1+3	0	0	0	0	0	0
1+4	0	0	1	0	0	1
1+2+3	0	1	0	0	0	1
1+2+4	0	3	3	6	0	12
1+3+4	0	0	0	0	0	0
1+2+3+4	0	2	1	1	0	4
2 only	0	1	1	5	1	8
2+3	0	0	0	0	0	0
2+4	0	1	3	2	1	7
2+3+4	2	1	3	0	0	6
3 only	0	0	0	0	0	0
3+4	0	0	0	0	0	0
4 only	1	5	0	4	1	11
No elevations	0	3	1	4	4	12
Total	3	19	15	24	8	69

whose mean radiographic ratios(Ratio; see the materials and methods) were greater than 1,5(suggesting the rapid progression of the disease into the localized areas) and those of the other subgroup consisted of six patients whose ratios were smaller than 1,5. All the IgG subclass levels to *Pg* were much higher in most, if not all, of the former subgroup than in the latter one(data not shown). When the comparison was made for the IgG2 levels to *Pg*, these association

could not be demonstrated(Figure 2).

IV. Discussion

We have performed the present study as one of the series experiments to clarify the immunological events occurring in the various subforms of EOP which we have to revised into the more homogeneous phenotypes³². Wide ranges of the antibody

titers were noted in all of the EOP subforms and the AP. The prevalence of the patients showing the elevated responses in total IgG and each IgG subclass were also depicted in the Table 2. The elevated antibody responses were found between 50 % to 100 % of the patients according to the disease types. However, these values were either greater or smaller with respect to the IgG subclass responses. As we have attempted to subdivide the classical forms of the RPP into a more homogeneous two subforms III or IV (distinctive RPP pattern), the percentage of patients showing elevated total IgG to *Pg* were relatively low (50 %), while much higher percentages (66.7%) of patients demonstrated a high IgG2 responses. The relatively low percentage was attributed in part from extremely low prevalence of patients with the elevated IgG3.

Total serum IgG and the four IgG subclass levels to *Pg*, and the IgG2 levels to *Aa* have been determined respectively against the whole cell surface antigens. The results were mostly consistent with the previous reports by others.^{5-17,19-24} The elevated IgG2 responses against whole cell surface antigens of these two bacteria were also similar to the findings of others in the LJP or the RPP.^{8,9,11,14,16,17,20,23,24} However, we could not demonstrate any significant differences in the single IgG subclass levels against *Pg* among the four EOP subforms, except for the IgG3. These results may support the concept of the continuous development of the various forms of the EOP (i.e. from the localized types into the more generalized ones)^{32,33-38}. Among the several factors responsible for the development of the EOP, the IgG2 responsiveness to the organisms has been considered to play a modulating role.^{8,18,23,24} This concept was based upon the functional aspects of the IgG2 which has a weak complement-fixing capability as well as the low avidity^{19,20,39}. The gradual increase in the IgG2 antibody against *Pg* in accor-

dance with the subform II, III, and IV might be supporting this concept. However, the hypothesis needs to be verified further by the systematic longitudinal investigations on the multiple factors including the environmental and the genetic components.

The IgG2 levels to the bacteria have been reported to increase drastically during the circumpubertal period^{8,14}. All of the four IgG subclass levels to *Pg* were also consistently found to be high in the younger age group around twenties, low around the ages of the early thirties and again high in the group of late thirties.

Mean IgG3 to *Pg* have been significantly elevated in the subform I compared with that of subform IV ($p < 0.06$). However, in no patient was a single IgG3 subclass shown to be elevated. The elevated IgG3 responses in the subform I patients were always accompanied by the concomitant elevations of the IgG2 and the IgG. Lu et al.⁸ demonstrated the elevated IgG3 titers against *Aa* in LJP which was less robust than the IgG2 differences. As the patients number of the subform I was too small, our results were not conclusive to establish its functional role in the LJP. This together with the report of Lu et al. prompted us to look into the IgG2 subclass levels to *Aa* in the four EOP subforms. It was significantly higher in the subform I (distinctive LJP pattern) than any other subforms which was similar to the findings of others^{8,18}.

Most of the previous reports on the role of the IgG2 responsiveness in the localizing the disease process did not consider the disease activity occurring around the affected teeth.^{8,18} though Ebersole et al.³ reported the elevated IgG responses to *Aa* in disease-active LJP patients. This fact led us to an additional analysis on the possible functional role of the IgG subclasses to *Pg* in modulating the disease activity around the first molars of localized EOP subforms (I and II). We selected the 13 patient from the

subform I and II patients whose overall bone destruction were the minimal (mean BL <6 mm) and then divided into the two subgroups; one group with the advancing diseases occurring in the localized areas and the other group with the low disease activities (see Materials and Methods). All the IgG subclass titers against *Pg* were significantly higher in most, if not all, of the former group patients compared with the latter group. This strongly indicated that the increased IgG subclass responsiveness to *Pg* were closely associated with the disease progression around the 1st molars (and possibly around the incisors) in the subforms I and II at their early stages of phenotypic development. When we looked into the IgG2 levels to *Aa* in the above-mentioned two subgroups. Similar patterns could not be demonstrated as in the case of *Pg*. Consequently, though the IgG2 responsiveness to *Aa* might be closely associated to the disease localization process, this may not be associated with the earlier destructive events occurring in the localized areas of the subforms I or II. It is interesting to note that the IgG2 was highest in the subform I not only to *Aa* but to *Pg*. This finding together with the above-mentioned results suggest the strong host-*Pg* interactions in the subform I (LJP). Vandcsteen et al.³⁵⁾ in their study of a family with a high prevalence of EOP have also reported that almost all the LJP members had an antibody specific to *Pg*. Moreover, not all of the JP patients had detectable *Aa*, nor did they have detectable antibody levels to the organism.

Lu et al⁸⁾ and Schenkein¹⁸⁾ have reasoned that increased IgG2 responsiveness to *Pg* with the development of the generalized pattern of the EOP such as rapidly progressing periodontitis. Elevated levels of IgG2 was also found to be associated with the history of destructive periodontitis⁴⁰⁾. Therefore we have examined the possible association in the modulating the phenotypic development of the more

generalizing subforms (i.e. II, III and IV) in addition to the functional role in the subform I. Our findings suggest that the increased levels of IgG2 (and possibly IgG4 as well) might strongly play a modulating role in the development of the rapidly generalizing EOP phenotypes. With this in mind and because there have been a variety of patterns in the different groups of the elevated IgG subclasses against *Pg* even within the same subforms, a further analysis has been performed to elucidate the various patterns of elevated IgG subclasses. Of all the patterns in the four EOP subforms, combinations with IgG1+2+4 were the most frequently found one, the next being IgG4 only, IgG2 only, and IgG2+4, etc., in the descending order. When we focused on the IgG2 or the IgG4 subclasses, or combinations of these two, 49 out of total 61 patients (80.3%) showed the elevated responses in either one or combination of these two subclasses. These 49 patients comprised 92.5% of 53 patients who had the elevated antibody in at least one IgG subclass. Again these patients were more frequently found in most severe subforms of the EOP (III and IV) than the subform II. These findings strongly indicated that the IgG2 or the IgG4 subclass responsiveness to *Pg* may be important in the rapidly disseminating process of the EOP.

Several authors postulated the bacterial LPS or carbohydrate moiety of LPS as immunodominant antigen(s) of *Aa* or *Pg* may act as immunodominant antigen eliciting exclusively the IgG2 responses. We may reason the possible role of the capsular polysaccharides or the LPS as others^{16,17,25-27,51)}. Along with these concepts, the functional role of the IgG2 in the disease protection has been suspected due to its low capacity to fix the complements³⁹⁾ and the low avidity found in the RPP patients^{20,21)}.

As mentioned above, the IgG4 may also play an important role in the pathogenesis of EOP. There

are only a few reports regarding the elevated IgG4 responses in the other forms of the periodontal diseases. Ogawa et al⁽⁴¹⁾, reported that the number of the IgG2- and the IgG4-secreting cells increased with the disease severity. They also found that the number of the IgG1-secreting cells decreased with the increase in the IgG4-secreting cells in the advanced forms of the adult periodontitis.⁽⁴²⁾ The highest anti-*Pg* LPS antibody was the IgG2 followed by the IgG4 in AP,⁽³⁶⁾ suggesting the switching pathway by the immunoglobulin genes clustered in 3' region($\gamma 2$ - $\gamma 4$ - ϵ - $\alpha 2$) involved in LPS specific responses⁽⁴²⁾. Reinhardt et al⁽⁴³⁾, also found the elevated GCF IgG4 levels in the sites showing disease activity. However these results were found in the adult periodontitis patients. Our results on the IgG4 responses to *Pg* in the EOP were in contrast with most of the studies which consistently demonstrated the low IgG4 concentration in the various forms of the periodontal diseases other than the EOP^(20,21,23). There may be the two reasons responsible for the elevated IgG4 responses. One is that IgG4 antibody share some common antigenic structures with IgG2 molecules resulting in the cross reactions in patients having high IgG2 titers. This possibility has been postulated by several authors.⁽⁴⁴⁻⁴⁶⁾ Moreover our results demonstrated the frequent findings of the concurrent elevations of the IgG4 and the IgG2 in the same subjects(Tables 2 and 4). When we compared the mean IgG4 of the patients with the elevated IgG2 titers with those without, the value was significantly higher in the former group in consistent with other studies.^(45,47,48) Another reason would be due to the prolonged stimulation by the proteineous antigens which might gradually have converted the IgG1-dominated responses into the IgG4-dominated ones^(42,49). This might have resulted in the gradual disease progression due to the low protective activity of the antibody with the weak complement-fixing

capability like IgG2 antibody. In the same line with this probability, we calculated the relative value of the IgG1 to the IgG4 in all the EOP patients. The values were less than 1 in all the four subforms and again it gradually became smaller with the increasing ages and were smallest in the subform IV implicating its another possible relationship with the disease severity in the EOP.

Considering that the specific IgG subclass responses are elicited against the various bacterial antigens, there would be differences in the immunodominant antigen(s) recognized by the various sets of the elevated of IgG subclasses shown in the present study. Thus it is tempting to consider both the capsular polysaccharides(and possibly LPS) and proteineous antigen(s) of *Pg* as candidates for the immunodominant antigens in the severe subforms of the EOP, while confirming the carbohydrate antigens of *Aa* in the EOP subform I. Based on our findings of the elevated IgG subclasses, we are currently under the experiments on the immunodominant antigen(s) of *Pg* and *Aa* in terms of the various combinations of the elevated IgG subclasses to answer these questions.

The normal IgG subclass concentrations and the IgG subclass responsiveness to bacterial antigens as well as to vaccine preparations are thought to be under the control of the genetic markers for each immunoglobulin allotype(Gm and Km),^(45,50-60) which are also race-dependent and inheritable.⁽⁶¹⁾ Based on these concepts, we have initiated a series of experiments to see the race-dependent immunogenetic control over the diversely elevated IgG subclass responsiveness as well as the various patterns of immunodominant antigens of *Pg* recognized by each of the EOP subform patients.

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조기발병형 치주염의 표현형적 소집단의 IgG Subclass에 대한 연구

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본 연구는 조기발병형 치주염의 서로 다른 4가지 표현형에 있어서 *Porphyromonas gingivalis*(Pg) 381과 *Actinobacillus actinomycetemcomitans*(Aa) Y4에 대한 상승된 IgG subclass의 양상을 평가하기 위해 시행하였다. Subform I(distinctive localized juvenile periodontitis pattern)에서 3명, subform II(post juvenile periodontitis pattern)에서 19명, subform III (localized but rapidly progressing pattern)에서 15명, subform IV(distinctive rapidly progressing periodontitis pattern)에서 15명의 환자를 조사하여 Pg에 대한 그들의 total IgG level과 각각의 IgG subclass level 및 Aa에 대한 IgG level을 검사했다. Pg에 대한 total IgG level은 subform II와 IV보다 subform I과 III에서 훨씬 높게 나타났다. IgG3 level이 subform I과 IV사이에서 현저한 차이가 있다는 점을 제외하고는, 다른 IgG subclass level에서 subform 사이에 아무런 차이가 없었다. Pg에 대한 IgG subclass는 single class 혹은 다양한 group에서 상승되어 나타났으며, IgG1+2+4가 가장 흔하게 발견되었고, 다음으로 IgG4 단독, IgG2 단독, IgG2+4, IgG2+3+4의 순으로 발견되었다. IgG2와 IgG4가 빈번히 상승되어 발견되었는데, 특히 severe form(subform III & IV)에서 그러했다. 뿐만 아니라, IgG level은 subform II, III, IV와 일치하여 점차적으로 증가하였고, 반면에 IgG1/IgG4 ratio는 그와 일치하여 감소되었다. 이러한 ratio의 감소는 단백질성의 오래된 항원의 과부하로 인해 immunoglobulin gene의 전환을 가능하게 한다는 것을 나타내고 있다.

Aa에 대한 IgG2 level은 다른 유형보다 subform I에서 상당히 높았다. Pg에 대한 IgG2 levels이 subform I의 국소 부위에서 발생하는 disease activity와 밀접한 관련이 있으며, Aa의 경우에는 이러한 관련성이 나타나지 않았다. Pg에 대한 IgG2 level은 18-25세에서 훨씬 높은 동시에 26-35세에서는 감소했으며 결국 30대 후반에서는 더 높은 수치로 되돌아갔다.

이러한 결과는 Pg에 대한 IgG2 및 IgG responsiveness (single 혹은 combined)가 EOP의 severe form의 발달에 중요하게 작용하며 IgG2 levels은 IgG1/IgG4 ratio와 더불어 EOP의 localized type이 generalized type으로 계속 진행되는 것을 조절하는 역할을 하는 것으로 보인다는 것을 강하게 시사하였다.

Key Words : IgG subclass, Early-Onset Periodontitis, *Porphyromonas gingivalis*, *Actinobacillus actinomycetemcomitans*