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CLINICAL AND IMMUNOLOGICAL EVALUATION OF PATIENTS WITH SELECTIVE IgA DEFICIENCY*

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Among the most frequent forms of primary immunodeficiency diseases is that called selective IgA deficiency which is characterized by absence of serum IgA or a serum IgA level of less than 5 mg/dl[1]. In cases with serum IgA levels sharply below the normal values but above 5 mg/dl, partial IgA deficiency should be considered. The etiology of selective IgA deficiency is unclear but it has been assumed that various immunopathogenic mechanisms may cause the disease. In most cases an intrinsic defect in B lymphocytes leads to IgA deficiency[2-4]. However, in some patients the increase in IgA specific suppressor T lymphocyte activity is likely to cause the disease[5,6]. The clinical features of selective IgA deficiency vary greatly, of which chronic recurrent infections, allergic diseases, hematological disorders, gastrointestinal manifestations, autoimmune diseases and malignancies have been frequently observed[7,8].

In this study patients with selective IgA deficiency are evaluated in terms of their clinical findings and T and B cell-mediated immunity.

Material and Methods

Fourteen patients, 12 with selective and two with partial IgA deficiency were included in this study. Thirteen of the patients had been diagnosed during the measurement of the serum immunoglobulin levels of patients who suffered from recurrent infections, allergic disorders or hematological diseases. One patient, the mother of Case 1 (UU), was diagnosed during the family screening of serum IgA levels in seven patients. The patients' ages, with the exception of one (BU), who was 34 years-old, ranged between two-eleven years.

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Immunolgical studies:
Serum immunoglobulins (IgG, IgM, IgA) were determined quantitatively by using immunodiffusion plates (Behring-Werke, W. Germany)[9]. Delayed hypersensitivity skin tests were performed by applying the following antigens in seven patients: PPD (5 U/0.1 ml), phytohemagglutinin (PHA) (10 mg/0.1 ml; Burroughs Wellcome), SK/SD (final concentration 50 U SK, 12.5 U SD/0.1 ml, SK/SD-varidase; Lederle) and *Candida* (1 PNU/0.1 ml; Hollister-Stier). Induration was determined at 48 h. The E-rosette test was performed in all patients using the method described by Jondal et al[10].
In vitro blastogenic response of lymphocytes to PHA was evaluated using a previously described method with some modifications in which the whole blood of 12 patients and 11 age-matched controls was studied[11,12]. The results were expressed as a stimulation index (SI= cpm (count per minute) of stimulated cells/cmp of unstimulated cells). The SI values above 20 were accepted as normal.
Surface immunoglobulin positive B cells were determined by fluorescent conjugated antisera against Ig isotypes (polyvalent, anti-IgG, IgM, IgA sera; Behring Werke, W. Germany) in eight patients and 19 healthy age-matched controls[13].
T lymphocyte subpopulations (Pan T, helper, suppressor T cells) were investigated in seven patients and 19 age-matched healthy controls by using monoclonal antibodies (OKT_3, OKT_4, OKT_8; Behring Werke, W. Germany) and the standard indirect immunofluorescence technique[14].
Anti-nuclear, anti-reticulum, anti-cytoplasmic, anti-parietal cell, anti-smooth muscle, and anti-mitochondrial antibodies were determined in ten patients, utilizing heterologus tissue and the indirect immunofluorescence technique.
The results were expressed as a Mean ± Standard Deviation and the Mann-Whitney-U test was used in statistical analyses.

Results

One of our patients (BU) with selective IgA deficiency was asymptomatic. Six patients, four with selective and two with partial IgA deficiency had allergic diseases (five patients with bronchial asthma and one with food allergy; EÇ, VN, KD, TT, KY, EA). Three patients had hematologic disorders, two with idiopathic thrombocytopenic purpura (FB, GD) and one with autoimmune hemolytic anemia (HA). The other four patients had chronic or recurrent sinopulmonary infections (Table I).
The serum IgA level was 0 in 12 patients, and 16 mg/dl and 22 mg/dl in two patients. The serum IgG levels were high in four patients and the IgM levels were high in two patients (Table I). Total lymphocyte counts were above 2000/mm^3 in all patients.

TABLE I: Clinical Data and Immunglobulin Levels of the Patients

Patients	Sex	Age (Yrs)	Clinical Picture	Serum Ig Levels (mg/dl) IgA	IgM	IgG	Autoantibody
1. UU	M	9	Recurrent infections	0	85	1220	(–)
2. BU	F	34	Asymptomatic	0	148	960	(–)
3. EÇ	M	9	Allergic diseases+ recurrent infections	0	59	700	(–)
4. GC	M	2	Chronic infections	0	77	700	nd
5. VN	M	9	Allergic diseases	0	129	1630	(–)
6. HŞ	M	8	Chronic-recurrent infections	0	138	1240	nd
7. FB	F	6	ITP	0	130	3150	nd
8. AB	F	6	Chronic-recurrent infections	0	319	3490	(–)
9. GD	M	3	ITP	0	89	1560	(–)
10. HA	F	5	Recurrent infections+ hemolytic anemia	226	508	(–)	
11. KD	M	9	Allergic diseases+ chronic infections	0	90	2340	(–)
12. TT	M	10	Allergic disease	0	145	2260	(–)
13. KY	M	7	Allergic disease	16	132	1250	(–)
14. EA	M	5	Allergic disease+ recurrent infections	22	128	710	nd

• Idiopathic thrombocytopenic purpura.

Mean Values of Normal Serum Ig's for the Various Age Groups•

Age Group	IgA (mg/dl)	IgM (mg/dl)	IgG (mg/dl)
2-3	66 ± 16	118 ± 35	889 ± 228
4-6	80 ± 31	130 ± 46	970 ± 186
7-11	130 ± 28	133 ± 39	1191 ± 282

nd : not done

• : The mean values of healthy children (Pediatric Immunology Unit, Hacettepe University Institute of Child Health).

Skin tests in which PHA and *Candida* were applied showed a positive reaction in seven patients. One patient reacted positively to the SK/SD test (Table II).

The percentages of E-rosette forming cells were found to be between 64-78 (mean value 72 ± 4). There was no significant difference observed between the patient and control groups.

In vitro blastogenic response to PHA was found to be low in only one patient (HA). The mean value of the stimulation index which was 40.6 ± 24.8 in the patients, was not significantly different from the controls (Table II).

TABLE II: Cell-Mediated Immunity of Patients

Patients	E-Rosette Forming Cells (%)	In Vitro PHA Response SI	Delayed Hypersensitivity Skin Test PHA	Candida	SK/SD	PPD
1. UU	74	23.2	+	+	–	–
2. BU	73	30.6	nd	nd	nd	nd
3. EÇ	64	53.0	nd	nd	nd	nd
4. GC	70	51.1	+	+	–	–
5. VN	78	nd	nd	nd	nd	nd
6. HŞ	69	31.5	+	+	–	–
7. FB	74	95.4	+	+	–	–
8. AB	78	34.2	+	+	–	–
9. GD	74	22.7	+	+	–	–
10. HA	76	10.6	nd	nd	nd	nd
11. KD	74	57.4	nd	nd	nd	nd
12. TT	71	nd	nd	nd	nd	nd
13. KY	78	29.0	+	+	–	–
14. EA	67	49.6	nd	nd	nd	nd
Patients (n = 14)	72 ± 4	40.6 ± 24.8				
Controls (n = 19)	69 ±6	42.1 ± 17.0				

The mean percentage of B lymphocytes was 18.4 ± 5.8 in eight patients. The percentages of surface Ig bearing cells were as follows: $sIgM^+$ lymphocytes 11 ± 6.2, sIG^+ lymphocytes 7.6 ± 3.5, and $sIgA^+$ lymphocytes 1.8 ± 1.4. There was no significant differences found when comparing the mean values of the patients with those of the controls. Only two patients had no $sIgA^+$ bearing cells (Table III).

The percentages of T lymphocytes (CD^+_3) in 7 patients were between 54-81 and the mean value was 69.4 ± 10.9. The percentages of CD^+_4, CD^+_8 T cells and the CD^+_4/CD^+_8 ratios of the patients were 43.7 ± 11.5, 26.9 ± 6.1 and 1.75 ± 0.88 respectively. The CD^+_4/CD^+_8 ratio was found to be low in two patients. There was no significant difference between the mean values of the patients and the controls (Table III).

Autoantibodies were negative in 10 patients.

TABLE III: Surface Immunoglobulins and T Cell Subpopulations

Patients	B Lymphocytes (Total) sIg+	sIgM+	sIgG+	sIgA+	T Lymphocytes (%) CD^+_3	CD^+_4	CD^+_8	CD^+_4/CD^+_8
1. BU	23	19	7	2	77	49	30	1.63
6. HŞ	13	6	6	3	54	26	24	1.08
7. FB	15	6	7	1	73	38	36	1.05
8. AB	17	10	12	2.5	54	38	29	1.31
9. GD	13	3	5	0	nd	nd	nd	nd
11. KD	15	9	6	0	75	42	23	1.82
12. TT	28	16	14	2	72	62	17	3.64
13. KY	23	19	4	4	81	51	29	1.75
Patients (n = 8)	18.4 ±5.8	11.0 ±6.2	7.6 ±3.5	1.8 ±1.4	69.4 ±10.9	43.7 ±11.5	26.9 ±6.1	1.75 ±0.88
Controls (n = 19)	22.2 ±5.0	8.2 ±4.2	9.1 ±3.6	4.5 ±3.1	66.9 ±13.3	45.6 ±5.2	27.0 ±6.9	1.80 ±0.52

Discussion

Selective IgA deficiency has been postulated to have both autosomal recessive and autosomal dominant forms of inheritance[7,8]. The parents and siblings of seven patients were screened for IgA deficiency, and only one case of selective IgA deficiency was found in these seven families. This patient (BU) was asymptomatic. The absence of IgA in the mother and in one of her children suggests an autosomal mode of inheritance in this family. It has been proposed that a compensatory increase of IgG and IgM in the serum and increased levels of secretory IgM and innate humoral factors may play an important role in obstructing the onset of symptoms in asymptomatic patients[15].

Chronic, recurrent viral or bacterial infections are the most frequent findings in selective IgA deficiency. Recurrent chronic infections were the main complaints in four of our patients. Four other patients who had allergic or hematologic disorders also presented with recurrent and/or chronic infections. The absence of secretory IgA is considered to be the main cause of infection.

Six of our IgA deficient patients had allergic diseases. When compared to the normal population allergic disorders are observed more frequently in individuals with selective IgA deficiency[3,7,8]. Although the cause of the frequent occurrence of allergic diseases is unknown, the absence or deficiency of serum and secretory IgA may lead to impaired neutralization of antigens resulting in a significant increase of IgE synthesis.

Of our three patients with hematologic diseases, two had ITP and one had autoimmune hemolytic anemia. Autoimmune hematologic disorders are seen frequently in selective IgA deficiency but this association cannot be explained by co-existence[16,17].

An increased frequency of autoimmunity in selective IgA deficiency has been observed as in some other primary immunodeficiencies. Amman and Hong[18] found various autoantibodies in ten of 15 patients studied with selective IgA deficiency. In our study, two patients had ITP, and one patient had autoimmune hemolytic anemia. Autoantibodies (anti-nuclear, anti-reticulum, anti-cytoplasmic, anti-parietal cell, anti-smooth muscle, anti-mitochondrial) were found to be negative in the serum of 10 of our patients which corresponds to the findings of the 50 patients evaluated by Burgio et al[3]. The following hypotheses have been proposed to explain the frequent occurrence of autoimmune diseases with selective IgA deficiency[15]: I. Due to the absence of secretory IgA, environmental antigens enter the organism from the mucous membranes. The antibodies produced to react against these antigens may act as autoantibodies because of cross-reactivity. II. The association of autoreactivity with a thymic defect may lead to IgA deficiency. III. Susceptibility to viral infections may stimulate the autoreactivity.

As we observed in some of our patients, the serum IgG and IgM concentrations have been found to be increased in selective IgA deficiency. These increases in the serum IgG and IgM levels occur in compensation for the absence of IgA or originate from associated disease, like chronic infections[19].

The majority of IgA deficient patients have an intrinsic defect in B lymphocytes. Various mechanisms have been assumed in its immunopathogenesis. Of these, an arrest in the development of B cells associated with decreased synthesis or release of IgA rather than the absence of IgA^+ B lymphocytes, or an arrest in the earlier stage of immunoglobulin-producing B cells following normal sequential development of IgM to IgG have been suggested[7,20-24]. In two of our patients, IgA bearing B lymphocytes were not detected. An early intrinsic defect in the maturation of B lymphocytes has been suggested as the cause for the absence of IgA bearing B lymphocytes[20].

However, in some cases, an increase of IgA specific suppressor T lymphocyte activity inhibits the production of IgA by normal B lymphocytes. It has been shown that B lymphocytes in these individuals produce IgA when co-cultured in vitro with normal T lymphocytes[5,6,25,26,27]. In our study the CD^+_4/CD^+_8 ratio was found to be low in two patients which was caused by a decrease of CD^+_4 cells in one patient (HS), and an increase of CD^+_8 in the other (FB). Since quantitative changes of T cell subsets may not always correlate with functional activity, functional studies are needed on this subject.

Although there may be impairment in some cases, cell-mediated immunity is usually normal in selective IgA deficiency[3,23]. In our study, the percentage of E-rosette forming cells was normal in all the patients and delayed hypersensitivity skin tests were positive at least when two antigens were applied to the patients. The evaluation of in vitro lymphoblastic response to PHA showed that the stimulation index was low in only one patient which was probably due to a viral infection that the patient had during the time of the study.

The heterogeneity of clinical and immunological features in these patients suggests that various mechanisms may be responsible for immunopathogenesis of IgA deficiency.

Summary

The clinical and immunological features of 14 patients including 12 with selective and two with partial IgA deficiency are presented. One patient was asymtomatic, six patients had allergic diseases, three patients had hematologic disorders and the remaining patients had chronic-recurrent infections. The IgG and IgM serum concentrations were high in four and two patients, respectively. Delayed hypersensitivity skin tests were applied to seven patients, and were found positive in all. E-rosette forming cells were within the normal range. In vitro lymphoblastic transformation with PHA was normal in all the patients, except for one. Two patients had no $sIgA^+$ B cells. The percentage of CD^+_4 T cells was decreased in one patient and the percentage of CD^+_8 T cells was increased in another, both of whom had a low CD_4/CD_8 ratio. The heterogeneity of clinical and immunological features in these patients suggests that various mechanisms may be responsible for the immunopathogenesis of IgA deficiency.

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