Assessment of selected adhesion molecules and lymphocyte subpopulations in children with IgA nephropathy

Wierciński R[1], Zoch-Zwierz W[1], Stasiak-Barmuta A[2], Wasilewska A[1], Tomaszewska B[1], Winiecka W[1]

[1] 1st Department of Paediatrics, Medical University of Białystok, Poland
[2] Flow Cytometry Laboratory, Medical University of Białystok, Poland

Abstract

Purpose: The aim of the study was to assess the expression of selected adhesion molecules on mononuclear cells of peripheral blood and lymphocyte subpopulations in children with IgA nephropathy (IgAN).

Material and methods: 14 children with IgAN and 20 healthy controls were included in the study. Flow cytometry was used to determine the expression of such adhesion molecules as L selectin (CD62L), VLA-4 integrin (CD49d), intracellular molecule ICAM-1 (CD54) and cytotoxic lymphocyte molecule CTLA-4 (CD152), as well as the lymphocyte antigens: CD3, CD4, CD8, CD19, CD1656 (NK), CD4 and CD8 RO+ and RA+.

Results: The findings revealed that the expression of the adhesion molecules VLA-4 and CTLA-4 did not differ from that of the healthy controls ($p>0.05$). However, the expression of CD62L (L-selectin) was increased ($p<0.05$). The expression of ICAM-1 was reduced, but not significantly, compared to the control group ($p>0.05$). We found a decrease in the expression of NK cells (CD1656) and CD4/CD8 ratio, and an increase in CD8 cells ($p<0.05$). In the group of 9/14 children, with proteinuria over 1.0 g/24 hours, a decreased expression of CD4 was additionally found ($p<0.05$).

Conclusions: The children with IgAN show: 1. Changes in peripheral lymphocyte subpopulations involving an increase in CD8 cells and a decrease in CD1656(NK) cells, a reduction in the CD4/CD8 ratio, and additionally in cases with proteinuria a reduction in CD4 cell count, 2. Increased expression of L-selectin (CD62L) on periferal blood mononuclear cells.

Key words: IgA nephropathy, adhesion molecules, lymphocyte subpopulations.

ADDRESS FOR CORRESPONDENCE:
Wierciński R
1st Department of Paediatrics
Medical University of Białystok
ul. Waszyngtona 17
15-274 Białystok, Poland
Tel: +48 85 7450-823 Fax: +48 85 7421-838
e-mail: annwasil@interia.pl

Received 19.04.2004 Accepted 31.05.2004

Introduction

IgA nephropathy is one of the most common immunological diseases of renal glomeruli in adults. It differs from Schönlein-Henoch purpura, being more common in children in the lack of generalised changes. The final diagnosis of IgA nephropathy is based on renal biopsy and finding immune deposits in glomerular mesangium, which consist of complement and immunoglobulins with a quantitative predominance of immunoglobulin A (IgA). The immunological process leading to the formation of IgA deposits is stimulated mainly by viral and bacterial antigens [1]. These deposits induce a number of reactions with a release of cytokines, growth factors and adhesion molecules, and their interactions enhancing inflammation or repair processes. These processes result in the accumulation of the amorphous matrix in the glomeruli and their sclerosis [2]. Adhesion molecules play a special role in the persistent injury of renal glomeruli in different immunological diseases. The knowledge of the pathogenic role of adhesion molecules in IgA nephropathy can facilitate proper treatment. In the animal model it has been shown that blocking of the adhesion molecules with monoclonal antibodies tempers the course of such diseases as lupus erythematosus, rheumatoid arthritis or Graves–Basedow disease [3,4]. It has been revealed that adhesion molecules, i.e. selectins, integrins and intracellular adhesion molecules [1,7,10], are the sensitive markers of progression in many diseases [3-9]. The earlier studies, especially those using flow cytometry, have demonstrated the role of T-cell associated disturbances of cellular response in the pathogenesis of IgAN. First of all, the abnormalities affect CD4 and CD8 lymphocyte subsets, which has also been

Table 1. **Clinical and laboratory characteristics of examined children IgAN**

patient	age (years)	histhopathologic type (WHO)	erythrocyturia		proteinuria mg/kg b.v./24h	oedema
			micro	macro		
1	14	II	+		6.1	–
2	13	II	+	+	13.4	–
3	6	II	+		2.6	–
4	9	II	+		24.4	–
5	7	II	+	+	2.5	–
6	6	II	+	+	5.4	–
7	8	II	+		28.9	–
8	13	II	+		15.7	–
9	11	III	+	+	11.8	–
10	16	IV	+		9.9	–
11	14	IV	+	+	128.4	+
12	15	V	+	+	61.7	+
13	14	V	+		110.5	+
14	16	V	+	+	82.3	+

confirmed in our own study [11]. In recent years, the role of adhesion molecules in the etiopathogenesis and the course of glomerulonephritis was emphasised. According to literature survey, an increase in adhesion molecules has been shown in neoplastic diseases, in diabetes, hypertension, in patients with impaired renal function, and also in chronic glomerulonephritis [6-9,12]. However, very few data are available on the expression of different adhesion molecules in the subpopulations of peripheral mononuclear cells in the course of IgA nephropathy. Most of the studies are concerned with the assessment of particular adhesion molecules in serum or tissues.

In order to enrich the knowledge on the role of adhesion molecules in IgA nephropathy we decided to assess the expression of L-selectin, VLA-4 integrin, intracellular adhesion molecule ICAM-1 and the receptor of T-lymphocytes – CTLA-4 on the monoclonal cells of peripheral blood.

L-selectin (CD62L, LEC-CAM-1, LAM-1) is expressed on almost all leucocytes. Its role is to facilitate binding of leucocyte glycoprotein to endothelial cells and their passage through the blood capillary wall, and homing the peripheral lymphatic organs by T-lymphocytes. It is the main receptor of homing for circulating unstimulated lymphocytes [10,13,14].

VLA (CD49d/CD29) (very late antigen) is a subunit of integrin $\alpha 4\beta 1$, which possesses 4 α chains and 1 β chain and is distributed on lymphocytes, monocytes, granulocytes, platelets and other cells. It is a receptor localised on the surface of unstimulated lymphocytes, which is bound to the cytoplasmatic protein to transmit the signal to the cell. It has an affinity to fibronectin, which is an extracellular matrix protein, and is responsible for reciprocal adhesion of cells and their adherence to the extracellular matrix. During lymphocyte stimulation by antigens, VLA-4 production is elevated [2,9,10].

The ICAM-1 (CD54) is a vascular adhesion molecule situated mainly on high endothelial cells. It is a ligand for integrins. It is also a receptor which facilitates cell penetration by some viruses. Apart from endothelial cells, ICAM-1 molecules are expressed on lymphocytes, fibroblasts, epithelium and other cells [3,15-19,20].

The CTLA-4 is a co-stimulator, which blocks the lymphocyte proliferation. It is an antigen of cytotoxic T-lymphocytes. The stimulation of CTLA-4 receptors on T-lymphocytes leads to the inhibition of their proliferation, by inducing their apoptosis.

The aim of the study was to assess: 1. The expression of selected adhesion molecules on mononuclear cells of peripheral blood, 2. The subpopulations of peripheral blood lymphocytes in children with IgAN.

Material and methods

The study group consisted of 14 children (♀-3, ♂-11) aged 4-16 years (mean 11.6±3.7), with IgAN (group I). In 9/14 children (subgroup A) constant proteinuria (>1.0 g/24 h) was observed and in 4/14 nephrotic syndrome was found. The control group (C) consisted of 20 healthy children (♀-13 ♂-7) aged 6-17 years (mean 14.0±3.1), without erythrocyturia and proteinuria.

For analysis, 1 ml venous blood samples taken to probes containing K_2EDTA were used. White blood cell counts with smear, lymphocyte subpopulations and some adhesion molecules were determined. Flow cytometry, using a Coultier analyser and monoclonal antibodies (f. Becton Dickinson) was applied to assess peripheral blood lymphocyte subpopulations: CD3, CD4, CD4RO, CD4RA, CD8, CD8RO, CD8RA, CD19, CD56, and adhesion molecules: L selectin (CD62L), VLA-4 integrin (CD49d), intracellular adhesion molecules ICAM-1 (CD54) and cytotoxic lymphocyte molecules CTLA-4 (CD152) on mononuclear cells of blood. The study was performed before treatment of the children.

Statistical analysis was based on a computer program Statistica 6.0, with the t-Students test and Pearson correlation tests. A p value <0.05 was considered statistically significant.

The study was approved by the Ethical Committee, Medical University of Białystok.

Results

Tab. 1 presents clinical and laboratory characteristics the group of children with IgAN. Haevy proteinuria (>10 mg/kg

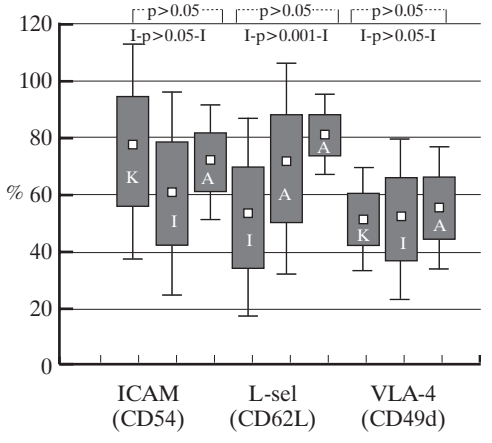
Table 2. **The mean values of lymphocyte subpopulations of peripheral blood in healthy children (C) and children with IgAN (I). Nine children with IgAN groups with heavy proteinuria are presented as subgroup A**

Group C n = 20	white cell count G/l	T CD3 %	T CD4 %	T CD8 %	B CD19 %	NK CD1656 %	CD4/CD8
X	6.20	70.56	43.50	27.26	13.56	12.30	1.58
SD	1.16	6.24	5.62	2.69	3.70	4.30	0.29
min–max	3.5-8.5	59.1-80.8	30.5-50.9	22.2-32.8	6.5-20.0	6.8-18.8	1.11-2.05
Group I n =14							
X	7.25	68.88	35.26	33.43	17.66	8.52	1.15
SD	1.49	8.53	12.10	8.12	9.12	4.13	0.54
min–max	5.2-9.6	51.6-82.8	12.7-52.3	17.8-44.0	6.5-41.0	2.9-14.2	0.3-2.0
p I z K	0.0887	0.8185	0.1203	0.0204	0.1548	0.0037	0.0421
Group A n = 9							
X		71.61	35.60	34.38	15.71	8.57	1.11
SD		7.99	10.29	6.29	7.22	3.85	0.49
min–max		57.0-82.8	20.3-47.4	24.0-44.0	6.5-26.7	3.5-14.2	0.5-1.9
p A z K		0.9132	0.03557	0.0051	0.6862	0.0419	0.0025

Table 3. **The expression the adhesion molecules on lymphocyte of peripheral blood in children with IgAN (I), subgroup IgAN with heavy proteinuria (A), and healthy children (C)**

Group C n = 20	L-selectin (CD62L) %	ICAM-1 (CD54) %	VLA-4 (CD49d) %	CTLA-4 (CD152) %
x	51.91	73.90	51.34	4.35
SD	16.97	18.39	8.79	1.91
min–max	18.1-76.1	16.6-98.1	28.4-66.0	1.6-7.3
Group I n = 14				
x	68.15	59.81	51.30	5.89
SD	18.03	17.39	13.71	3.05
min–max	32.7-87.6	25.7-90.0	16.7-72.7	2.2-12.2
Subgroup A				
x	79.91	65.21	58.30	5.97
SD	23.22	15.32	14.67	3.01
min–max	18.8-98.2	17.1-84.5	16.9-84.6	2.3-13.1

Figure 1. **Comparison of the adhesion molecules on lymphocyte of peripheral blood in children with IgAN (I), IgAN subgroup with heavy proteinuria (A) vs control group (C)**



% – percentage of the lymphocytes with adhesion molecules

b.v./24 h) was observed in 9 children. In 4 children was observed nephrotic proteinuria (> 50 mg/kg b.v./24 h). All the examined children revealed erythrocyturia and periodical haematuria as the predominant clinical symptoms. Renal biopsy showed: II degree of the disease progression in 8 children, III degree in 1 child, IV degree in 2 children, and V degree in 3 children, according to the histhopathological classification proposed WHO [21,22].

All the examined children with IgA nephropathy had normal white cell counts (7.2 ± 1.4 G/l), which did not differ from those of healthy controls ($p > 0.05$). Mean values of peripheral blood lymphocyte subpopulations in the examined groups of children are shown in *Tab. 1*. In group I, including all the children with IgAN, we found a reduced number of CD4 and CD1656 (NK) cells and elevated number of CD8 and CD19 (B lymphocytes), compared to the control group (C). However, the differences were statistically significant only for CD8 and CD1656 cells ($p < 0.05$), compared to healthy children ($p < 0,05$). The CD4/CD8 ratio was significantly depressed ($p < 0.05$). In subgroup A, including the children with IgAN, who had not only erythrocyturia, but also constant proteinuria (more than 1.0 g/24 hours), we additionally revealed a reduced CD4 lymphocyte count ($p < 0.05$).

The mean expressions of the adhesion molecules are presented in *Tab. 2*. In group I, the children with IgAN, showed an increased expression of CD62L cells (L-selectin) in comparison

with the healthy controls (C) ($p < 0.05$). However, the reduced expression of ICAM-1 molecule was not statistically significant. No differences were observed between the expression of integrin VLA-4 and CTLA-4 ($p > 0.05$). *Fig. 1* presents data concerning the expression of adhesion molecules in our patients (group I and subgroup A) vs. healthy controls (C). In subgroup A, including the IgAN children with constant proteinuria, increased expression of L-selectin (CD62L) ($p < 0.01$) and a statistically insignificant increase in integrin VLA-4 expression were found ($p > 0.05$). In the group of patients, no linear correlation was observed between L-selectin and B lymphocytes ($r = -0.183$), NK cells ($r = 0.111$), CD4 lymphocytes ($r = 0.089$), irrespective of the severity of pathological changes in the urine. However, a very weak negative linear correlation was found between the expression of L-selectin and CD4/CD8 ($r = -0.327$).

Discussion

Mononuclear cells play a very important role in the pathogenesis of IgA nephropathy. Their accumulation in the kidneys is conditioned by the adhesive interaction between the cells and the adequate ligands [15,21,23-27]. We assessed the expression of three main types of adhesion molecules and lymphocyte subpopulations in children with IgAN confirmed in the histopathological examination. The findings revealed that the expression of L-selectin on peripheral blood mononuclear cells in children with IgAN was increased and the expression of ICAM-1 was decreased, in comparison with healthy children. However, only the difference in the mean L-selectin expression was statistically significant. The expressions of VLA-4 and CTLA-4 were similar to those observed in the control group. Our findings suggest that L-selectin, which takes part in leucocyte migration and adhesion to endothelium, might play an important role in the pathogenesis of IgAN. Similar results were obtained by Kannel-de March et al. [13], who observed increased proportion of T and B lymphocytes and increased L-selectin expression on T-lymphocytes in their patients. The authors assume that overproduction of CD62 L cells may be related to the increased adhesion of lymphocytes and their homing in the lymphoid tissues in IgAN. Sakatsumi [28] used flow cytometry to assess the antigen expression of T-lymphocytes and monocytes in the urine of patients with different glomerulopathies, including IgAN. They showed the presence of $CD45RO^+$ and $CD62L^-$ T-effector cells, like in renal glomeruli. The authors believe that the analysis of antigen expression on T-lymphocytes in the urine of children with IgAN can facilitate monitoring of the disease activity. The highest expression of these cells was found in patients with heavy proteinuria and impaired renal function. However, $CD62L^+$ lymphocytes were present in the lymphatic follicles, where active cells change into memory cells.

Many reports indicate that vascular adhesins, represented by the intracellular adhesion molecule ICAM-1, also play an important role in the pathogenesis of IgAN [15-18,26,29]. The evidence for that fact is the expression of ICAM-1 on renal cells in patients with IgAN. The study of Arrizabalage [15] suggests that the cellular ICAM-1 in renal tubules and interstitial tissue takes part in leucocyte adhesion. The expression of ICAM-1 in renal tubules is induced by macrophages and interstitial and glomerular T-cells, especially CD8 cells [27]. It has been shown that such cytokins as TNF-α and interleukin 1 together with mononuclear cells induce the regulation of ICAM-1. The authors assume that the cellular expression of ICAM-1 in renal tubules and interstitial tissue may be a marker of tubulointerstitial damage in IgA nephropathy. Ootaka [29] showed that ICAM-1 took part in the induction of proteinuria in IgAN. Tomico [19] found an increased expression of ICAM-1 in the renal glomeruli of patients with IgAN. However the expression did not correlate with the immunological complex, containing IgA. Up to now, ICAM-1 expression on renal tissue has been mainly evaluated [5,15-17,19,26,29]. Few investigators have assessed serum levels of soluble ICAM-1 molecules in IgAN [20,30]. In our study, the expression of ICAM-1 on peripheral blood mononuclear cells was evaluated. A little decrease in ICAM-1 was found in the children with IgAN, compared to healthy controls. A decrease in ICAM-1 expression on peripheral blood mononuclear cells may indicate their utilisation in the immunological process. However, in our examination the decreased expression of ICAM-1 in the study group did not differ statistically significantly from the values in the control group.

In the immunological diseases, integrins are of pathogenic importance [10]. Namie and colleagues [2] estimated the expression of integrins on peripheral lymphocytes and monocytes of patients with IgAN. They found the increased expression of β integrins: VLA-4 and VLA-5. The activation of VLA-4 (fibronectin receptor) on peripheral mononuclear cells indicates its participation in the process of glomerulosclerosis in the course of IgAN. In our investigations the expression of VLA-4 on peripheral lymphocytes and monocytes in patients with IgAN did not differ from that of healthy children. We noted no changes in the expression of the CTLA-4 molecule, either.

The present study revealed a thymus-dependent lymphocyte activation in children with IgAN. This finding was supported by the increased number of CD8 T-cells and decreased CD4/CD8 ratio. Similar results were obtained in our earlier studies in children with Schönlein-Henoch nephropathy [11]. It was also confirmed by other authors, who studied IgAN [23,25,27]. The status of CD4 and CD8 T-lymphocytes is defined by the presence of the receptors $CD54RA^+$ and $CD54RO^+$ on their surface. CD4 or $CD8^+CD45RA^+$ cells are the naive lymphocytes, which have not got into contact with the antigens yet and which after the antigen stimulation change into CD4 or $CD8^+CD45RO^+$ memory lymphocytes [31-33]. In our examinations, antigen expression on lymphocytes did not differ from that observed in the healthy controls. On the other hand, a decreased count of CD1656 cells (NK) was found. One of the properties of NK cells is their participation in the cellular antibody-dependent cytotoxity. In that case a decreased number of NK cells on the peripheral cells in IgAN may indicate their involvement in the immunological process in the kidney [1,11,24,34]. In the urine of 9/14 examined children constant proteinuria (more than 1.0 g/24 hours) was found. A detailed analysis showed an increase in the expression of L-selectin and a reduction in the number of CD4 and CD8 cells – more pronounced than in the children without proteinuria. We noted increased expression of VLA-4 integrin, as well.

Our results confirm the observations of other authors that in IgAN children with concomitant proteinuria, lymphocyte subpopulations and some adhesion molecules, and particularly L-selectin are more involved in the immunological process than in patients with mild course of the disease.

Our results should be further evaluated in a larger study cohorts. There is still an open question: can inhibition of expression of adhesive molecules by neutralizing antibodies, he used as a therapeutic treatment of IgAN.

Conclusions

The children with IgAN show:

• changes in peripheral blood lymphocyte subpopulations involving an increase in CD8 cells and a decrease in CD1656 (NK) cells, a reduction in the CD4/CD8 ratio, and additionally in cases with proteinuria a reduction in CD4 cell count.

• increased expression of L-selectin (CD62L) on peripheral blood mononuclear cells.

References

1. Kowalski ML. Immunological respons in: Clinical Immunology. Mediton Łódź 2000; 1-32.
2. Namie S, Ozono Y, Harada T, Hara K. Expression and function of fibronectin receptors on peripheral mononuclear cells in IgA nephropathy. Nephrol Dial Transplant, 1995; 10: 1342-7.
3. Aoki S, Imai K, Yachi A. Soluble intercellular adhesion molecule-1 (sICAM-1) antigen in patients with rheumatoid arthritis. Scand J Immunol, 1993; 38: 485-90.
4. Sonet E, Massart C, Gibassier J, Allnnic H, Maugenore D. Longitudinal study of soluble intercellular adhesion molecule-1 (sICAM-1) in sera of patients with Graves disease. J Endocrinol Invest, 1999; 22: 430-5.
5. Chow J, Hartley RB, Jagger C, Dilly SA. ICAM-1 expression in renal disease. J Clin Pathol, 1992; 45: 880-4.
6. Kiliś-Pstrusińska K, Medyńska A, Wikiera-Magott J, Zwolińska D. Serum levels of soluble adhesion molecules in children with glomerulonephritis. Pol Merk Lek, 2001; 10: 247-9.
7. Krętowski A, Kinalska I. The immune markers of preclinical phase of type 1 diabetes mellitus. The role of cytokines, adhesion molecules and peripheral blood lymphocyte subpopulations in the pathogenesis and prediction of insulin-dependent diabetes. Przegląd Lekarski, 2000; 12: 763-72.
8. Martin-Henao GA, Quiroga R, Sureda A, Gonzalez JR, Moreno V, Garcia J. L-selectin expression is low on $CD34^+$ cells from patients with chronic myeloid leukemia and interferon-α up-regulates this expression. Hematologica, 2000; 85: 139-46.
9. Musiał K, Zwolińska D. The role of selected cell adhesion molecules in chronic renal failure patients on conservative treatment. Pol Merk Lek, 2001; 10: 209-12.
10. Jakóbisiak M, Obłąkowski P. Krążenie limfocytów, w: Jakóbisiak M, Immunologia. Wyd Nauk PWN Warszawa, 1996; 171-82.
11. Wierciński R, Zoch-Zwierz W, Wasilewska A, Tomaszewska B, Winiecka W, Stasiak-Barmuta A, Porowski T, Biernacka A. Lymphocyte subpopulations of peripheral blood in children with Schönlein-Henoch purpura and IgA nephropathy. Pol Mer Lek, 2001; 10: 244-6.
12. Bruijn JA, Dinklo NJ. Distinct patterns of expression of intercellular adhesion molecule-1, vascular cell adhesion molecule-1, and endothelia leukocyte adhesion molecule-1 in renal disease. Lab Invest, 1993; 69: 329-35.
13. Kennel-de March A, Bene MC, Renoult E, Kessler M, Faure GGC, Kolopp-Sarda MN. Enhanced expression of L-selectin on peripheral blood lympocytes from patients with IgA nephropathy. Clin Immunol Immunopathol, 1997; 84: 158-65.
14. Takei T, Iida A, Nitta K, Tanakat T, Ohnishi Y, Yamada R, Maeda S, Tsunoda T, Takeoka S, Ito K, Honda K, Uchida K, Tsuchiya K, Suzuki Y, Fujioka T, Ujiie T, Nagane Y Miyano S, Narita J, Gejyo F, Nihei H, Nakamura Y. Association between single- nucleotide polymorphisms in selectin genes and immunoglobulin A nephropathy. Am J Hum Genet, 2002; 70: 781-6.
15. Arrizabalaga P, Sole M, Quinto JL, Ascaso C. Intercellular adhesion molecule-1 mediated interactions and leucocyte infiltration in IgA nephropathy. Nephrol Dial Transplant, 1997; 12: 2258-62.
16. Arrizabalaga P, Sole M, Abellana R, Ascaso C, Cardesa A. Renal expression of intercellular adhesion molecule-1 in immunoglobulin A nephropathy tubulointerstitial injury and prognosis. Virchows Arch, 2001; 439: 27-34.
17. Lee TW, Park JK, Ahn JH, Ihm CG, Kim MG. Role of mononuclear cells of IgA nephropathy on ICAM-1 expresion in mesangial cells. Korean J Intern Med, 1998; 13: 27-32.
18. Nguyen TT, Shou I, Funabiki K, Shirato I, Kubota K, Tomino Y. Correlations among expression of glomerular intercellular adhesion molecule-1 (ICAM-1), levels of serum soluble ICAM-1, and renal histopathology in patients with IgA nephropathy. Ann J Nephrol, 1999; 19: 495-9.
19. Tomino Y, Ohumuro H, Kuramoto T, Shirato I, Eguchi K, Sakai H, Okumura K, Koide H. Expression of intercellular adhesion molecule-1 and infiltration of lymphocytes in glomeruli of patients with IgA nephropathy. Nephron, 1994; 67: 302-7.
20. Yokoyama H, Takaeda M, Wada T, Ohta S, Hisada Y, Segawa C, Furuichi K, Kobayashi K. Glomerular ICAM-1 expression related to circulating TNF-alpha in human glomerulonephritis. Nephron, 1997; 76: 425-33.
21. Schena FP. IgA nephropathies in: Oxford textbook of clinical nephrology. Cameron S University Press Oxford, 1992; 1: 339-62.
22. Sieniawska M, Kozak-Białasik D, Gura C, Wierzbowska-Lange B, Szymanik-Grzelak H, Girdwoyn A, Róg-Marczuk J, Wasiutyński A. Przebieg i wyniki leczenia nefropatii IgA u dzieci. Ped Pol, 1996; 71: 313-9.
23. Hori Y, Takamoto T, Nishida H, Ishizaki T, Yokoyama MM, Nomura G. Studies of cell-mediated immunity in mesangial proliferative glomerulonephritis – IgA nephropathy and non-IgA nephropathy. Nippon Jinzo Gakkai Shi, 1989; 31: 723-33.
24. Hotta O, Taguma Y, Ooyama M, Yusa N, Nagura H. Analysis of $CD14^+$cells and $CD56^+$ cells in urine using flow cytometry: a useful tool for monitoring disease activity of IgA nephropathy. Clin Nephrol, 1993; 39(6): 289-94.
25. Kashem A, Yamauchi F, Yano N, Miyazaki H, Endoh M, Nomoto Y, Sakai H. CD4 subsets in IgA nephropathy. Tokai J Exp Clin Med, 1992; 17: 173-7.
26. Ohmuro H, Tomino Y, Koide H. Effect of monoclonal antibody CD4 on expression of intercellular adhesion molecule 1 in renal tissues of ddY mice a spontaneous animal model of IgA nephropathy. Am J Nephrol 1994; 14: 136-41.
27. Shimamine R, Shibata R, Ozono Y, Harada T, Taguchi T, Hara K, Kono S. Anti-CD8 monoclonal antibody protects against spontaneous IgA nephropathy in ddY mice. Nephron, 1998; 78: 310-8.
28. Sakatsume M, Xie Y, Ueno M, Obayashi H, Goto S, Narita I, Homma N, Tasaki K, Suzuki Y, Gejyo F. Human glomerulonephritis accompanied by active cellular infiltrates shows effector T-cells in urine. J Am Soc Nephrol, 2001; 12: 2636-44.
29. Ootaka T, Saito T, Soma J, Yusa A, Abe K. Intercellular adhesion molecule-1/ leukocyte function associated antigen-1 mediated and complement receptor type 4 – mediated infiltration and activation of glomerular immune cells in immunoglobulin A nephropathy. Am J Kidney Disk, 1996; 28: 40-6.
30. Mrowka C, Heintz B, Sieberth HG. VCAM-1, ICAM-1, and E-selektin in IgA nephropathy and Schönlein-Henoch syndrome: differences between tissue expression and serum concentration. Nephron, 1999; 81: 1256-963.
31. Adamthwaite D, Cooley MA. $CD8^+$ CD – cell subsets defined by expression of CD45 isoforms. Immunology, 1994; 81: 253-6.
32. Bell EB, Sparshott SM. Interconversion of CD54R subsets of CD4 T-cells in vivo. Nature, 2000; 348: 163-6.
33. Ferreira C, Barthlott T, Garcia S, Zamoyska R, Stockinger B. Differential survival of naïve CD4 and CD8 T-cells. J Immunol, 2000; 165: 3689-94.
34. Barao J, Acsensao JL. Human natural killer cells. Arch Immun Therap Exp, 1998; 46: 213-29.