UC 577,1; 61 ISSN 0354-3447

Jugoslov Med Biohem 23: 55–58, 2004 *Originalni naučni rad*
Original paper

IS SOLUBLE INTERCELLULAR ADHESION MOLECULE 1 A MARKER OF DISEASE ACTIVITY IN BRONCHIAL ASTHMA?

Mirjana Bogić[1], Aleksandra Perić-Popadić[1], Žikica Jovičić[1], Snežana Kovačević[2], Sanvila Rašković[1], Vesna Tomić-Spirić[1], Nevena Savić[1], Miodrag Čolić[3]

[1]Institute of Allergology and Immunology, Clinical Center of Serbia, Belgrade, Serbia and Montenegro
[2]Internal Medicine, Clinical – Hospital Center »Dr Laza K. Lazarević« Šabac
[3]Institute of Medical Research, MMA, Belgrade

Summary: Increased number of neutrophils, eosinophils and lymphocytes in the airway mucosa in the course of asthma exacerbation develops simultaneously with increase of expression of the specific adhesion molecules (including intercellular adhesion molecule 1) on the postcapillary venulae of the endothelial cells. Our study was aimed at measurement of concentration of soluble intercellular adhesion molecule 1 (sICAM-1) in 7 patients with bronchial asthma in phase of exacerbation, and evaluation of correlation between the obtained value and sICAM-1 concentration obtained in the same patients in the stable state subsequent to seven-day glucocorticosteroid therapy, as well as in comparison to the healthy controls (10 subjects). Mean value of sICAM-1 concentration in patients with asthma exacerbation and healthy controls was 430.49 ng/mL and 260.9 ng/mL, respectively. sICAM-1 concentration was found to be statistically significantly higher in patients with exacerbating asthma in comparison to the healthy controls (Z = 2.246), as well as in comparison to the same patients in the stable state subsequent to seven-day glucocorticosteroid therapy (Z = 2.197).

Key words: bronchial asthma, exacerbation, intercellular adhesion molecule 1, glucocorticosteroids

Introduction

Intercellular adhesion molecule-1 (ICAM-1, CD54) is a member of the immunoglobulin gene superfamily with 5 extracellular Ig-like domains. It acts as a ligand for Lymphocyte Function Associated Antigen-1 (LFA-1) (CD11a/CD18) and MAC-1 (CD11b/CD18) which are members of the leukocytic integrin family (beta 2 integrins) and they are expressed on the leukocytes (1). ICAM-1 may be induced or upregulated on the fibroblasts and endothelial cells via the inflammation mediators such as IL-1, TNF alpha and IFN alpha. *In vitro* expression increase is initiated 6-8 hours after stimulations and it is maintained for at least 48 h (2).

Downregulation is performed via antiinflammatory agents such as glucocorticosteroids. In both monocytic (U937) and bronchial epithelial (H292) cell lines, dexamethasone potently suppresses basal and induced ICAM-1 expression (3). Soluble form of ICAM-1 (sICAM-1) may be found in the serum, cerebrospinal fluid and bronchoalveolar lavage and it is the result of proteolytic cleavage of the membrane ICAM-1 (mICAM-1) (4). Epithelial cells of the airways in the asthmatic patients may express increased quantities of the ICAM-1 molecules (5). Wegner et al (6) evidenced in allergic inflammation that application of anti ICAM-1 monoclonal antibody in monkeys results in reduction of eosinophilic lung infiltration and reduction of hyperresponsiveness of the airways, as well as reduction of upregulation of ICAM-1 expression evidenced in the endothelium, bronchial epithelium and eosinophiles suggesting important role of ICAM-1 in pathogenesis of the bronchial asthma.

Our study was aimed at measurement and comparison of soluble ICAM-1 concentrations in patients with bronchial asthma in phase of exacerbation and the same patients in the stable state after seven-day glucocorticosteroid treatment, as well as at establishment of correlation with healthy controls.

Address for correspondence

Mirjana Bogić, Institute of Allergology and Immunology
Clinical Center of Serbia, Koste Todorovića 2, Belgrade
Serbia and Montenegro
Tel.: 361 55 67

Material and Methods

Our study included seven patients with bronchial asthma diagnosed based on guidelines for diagnosis and treatment of asthma proposed by the International Expert Group with the National Heart, Lung and Blood Institute of Health, Bethesda, from May 1997, revised 2002 (7). Metacholine and/or Ventolin tests were positive in all the patients. All the patients underwent physical and medical examinations and their lung function was measured before blood sampling using the apparatus Autospir Discom-14 Chest Corporation Tokyo, Japan.

sICAM-1 was measured in blood samples obtained by puncture of the cubital vain using Vacutainer without EDTA addition. The sample was allowed to stand at room temperature for 60 120 minutes to be coagulated. The sample is thereafter centrifuged at 1300 g for 10 minutes at room temperature. Separated serum was stored till analysis at 20 °C.

Measurement of sICAM-1 serum concentration was performed using ELISA method (8). We used the commercial Parameter human sICAM-1 immunoassay (ELISA, R&D Systems Inc., Mineapolis, USA). Serum concentration of the sICAM-1 is expressed in ng/mL. Lower detection border of serum sICAM-1 was 0.35 ng/mL (the method according to Parameter human sICAM-1 immunoassay).

In 7 patients, averagely aged 46 years, in phase of asthma exacerbation diagnosed based on the symptoms and signs of the disease, degree of airway obstruction measured using FEV1 at the time of testing, sICAM-1 concentration was measured. After seven-day therapy based on inhalation glucocorticosteroids, beta-2 agonists, oral prednisone in dose of 30 mg/day, blood samples were repeatedly obtained and sICAM-1 concentrations were determined.

The control group included 10 healthy subjects averagely aged 37 years without any history of allergy, asthma, allergic rhinitis, atopic dermatitis or any other significant disease.

Statistics

Statistical analysis was performed using EPI INFO ver. 10 program package. Statistical differences were calculated according to non-parametric Mann-Whitney test. Correlations between different parameters were determined based on Spearman's rank correlation coefficient calculations (8, 9).

Results

Our group of healthy controls (without any history of allergy, asthma, allergic rhinitis, atopic dermatitis or any other significant disease) had mean sICAM-1 value of 226.64 ng/mL, which was consistent with the manufacturer's reference values (115 306 μg/mL). Mean value of sICAM-1 concentration in patients with exacerbating asthma (N=7) was 430.4 ng/mL. Correlation coefficient of sICAM-1 concentrations between groups of healthy controls and patients with asthma exacerbation was $Z = 2.246$ $p < 0.05$, indicating presence of statistically significant difference in sICAM-1 concentrations between the groups of subjects, i.e., sICAM-1 concentration was significantly higher in asthma patients with asthma exacerbation in comparison to healthy controls.

Mean sICAM-1 concentration after seven-day glucocorticosteroid therapy applied in the same subjects (asthmatic patients) was 352.30 ng/mL. Correlation coefficient of sICAM-1 concentrations between asthmatic patients in stable state (after therapy) and healthy controls was $Z = 2.051$ $p < 0.05$, indicating presence of statistically significant difference between the groups, i.e., sICAM-1 concentration was significantly higher in stable-state asthmatics in comparison to the healthy controls.

Correlation coefficient of sICAM-1 concentration between the groups before and after the therapy was $Z = 2.197$ $p < 0.05$, indicating presence of statistically significant difference in sICAM-1 concentrations before and after the therapy, i.e., in phase of disease exacerbation, sICAM-1 concentration values were significantly higher in patients with disease exacerbation in comparison to the same patients in the stable state (*Table I*).

Table I. Mean sICAM-1 concentrations in patients with bronchial asthma in exacerbation, before and after glucocorticoid therapy

sICAM-1 (ng/mL)		
n=7	before	after
Mean	430.4	352.3
SD	232.84	188.92
Med	355.17	299.23
Min	246.74	198.47
Max	930.65	754.21
$Z = 2.197$ $p < 0.05$		

Discussion

The mechanisms of asthma-associated airway inflammation include the cascade of events ultimately resulting in accumulation of inflammatory cells from circulation participating in upregulation of the endothelial adhesion molecules, including ICAM-1, as well as in reciprocal expression of their ligands on the leukocytes (Global initiative for asthma, 1997). Our study was aimed at measurement and comparison of

ICAM-1 concentrations in healthy controls and patients with bronchial asthma in phase of exacerbation and in stable state, as well as at establishment of correlation between sICAM-1 concentrations in the same group of patients with asthma exacerbation and in stable state seven days after glucocorticosteroid therapy administration.

Mean value of sICAM-1 concentration in our group with bronchial asthma in phase of exacerbation was 430.4 ng/mL, in stable state 352.30 ng/mL, while in healthy controls it was 226.64 ng/mL. Shiota et al (10, 11) evidenced mean value of sICAM-1 concentration in healthy volunteers of 260.9 ng/mL, which was somewhat higher in comparison to the mean value of sICAM-1 concentration evidenced in our controls. The difference in number of healthy controls (10 vs. 39) may have contributed to this inconsistence.

Analysis of sICAM-1 concentration correlation between the groups with asthma exacerbation and healthy controls evidenced correlation coefficient of $Z = 2.246$ $p < 0.05$ indicating presence of statistically significant correlation between ICAM-1 values, i.e., that the patients with asthma in phase of exacerbation had higher sICAM-1 serum concentrations in comparison to healthy controls. Our results are consistent with the data published in the reference literature evidencing that adult patients with asthma exacerbation (in the course of the acute attack) had higher sICAM-1 concentrations in comparison to the healthy controls (10 14). Tang et al (15) also reported in their study that asthmatic children in the phase of exacerbation and rhinitis also had higher sICAM-1 serum concentrations in comparison to the healthy controls. Data indicating higher sICAM-1 concentrations in stable-state patients with asthma in comparison to those measured in healthy controls were also consistent with data from the reference literature (16). These results clearly indicated the role of the adhesion molecule ICAM-1 in pathogenesis of bronchial asthma, particularly as a disease activity (exacerbation) marker.

Out of 7 patients with asthma exacerbation, three were atopic and four non-atopic. Severe asthma was diagnosed in 2 patients, while moderately severe one in phase of exacerbation in 5 patients. Mean value of sICAM-1 concentration, before introduction of the therapy, was 430.49 ng/mL to be reduced to 352.30 ng/mL after 7-day therapy. Only in one of the patients, sICAM-1 concentration value was mildly higher after 7-day therapy (355 ng/mL vs. 362 ng/mL). Application of a non-parametric test (Wilcox's rank test), evidenced correlation coefficient of $Z = 2.197$ $p < 0.05$ indicating presence of statistically significant difference between sICAM-1 concentrations before and after therapy, i.e., that the same patients has statistically significantly higher sICAM-1 concentrations in phase of exacerbation of bronchial asthma in comparison to those evidenced in the stable state (remission). The later was one of the most important observations consistent with data reported by the Japanese authors (10, 16, 17), which pointed out to the significance of application of inflammation mediators and markers in monitoring of the therapy as well as that sICAM-1 concentration level may be modulated by glucocorticosteroid therapy (3).

Finally, the results obtained in our study led to conclusion that seven-day glucocorticosteroid therapy results statistically significant reduction of sICAM-1 concentrations in patients with bronchial asthma, as well as that sICAM-1 is an obvious marker of activity in bronchial asthma patients.

DA LI JE SOLUBILNI INTERCELULARNI ADHEZIVNI MOLEKUL 1 MARKER AKTIVNOSTI BOLESTI KOD BRONHIJALNE ASTME?

Mirjana Bogić[1], Aleksandra Perić-Popadić[1], Žikica Jovičić[1], Snežana Kovačević[2], Sanvila Rašković[1], Vesna Tomić-Spirić[1], Nevena Savić[1], Miodrag Čolić[3]

[1]Institut za alergologiju i imunologiju, Klinički centar Srbije, Beograd
[2]Interna medicina, Kliničko-bolnički Centar »Dr Laza K. Lazarević« Šabac
[3]Institut za medicinska istraživanja VMA, Beograd

Kratak sadržaj: Povećan broj neutrofila, eozinofila i limfocita u mukozi disajnih puteva za vreme pogoršanja astme događa se paralelno sa povećanjem ekspresije specifičnih adhezivnih molekula (među kojima je i intercelularni adhezivni molekul 1) na postakapilarnim venulama endotelnih ćelija. Cilj ovog rada je bio da se izmeri koncentracija solubilnog intercelularnog adhezivnog molekula 1 (sICAM-1) kod 7 bolesnika sa bronhijalnom astmom u pogoršanju i korelira dobijena vrednost s koncentracijom sICAM-1 kod istih bolesnika u stabilnom stanju nakon sedmodnevne terapije glukokortikosteroidima, kao i u odnosu na zdrave osobe (10 ispitanika). Srednja koncentracija sICAM-1 u bolesnika s pogoršanjem astme iznosila je 430,49 ng/mL, a u zdravih ispitanika 260,9 ng/mL. Bolesnici sa astmom u pogoršanju imali su statistički značajno veću koncentraciju sICAM-1 u odnosu na zdravu kontrolu ($Z = 2,246$), kao i u odnosu na iste bolesnike u stabilnom stanju nakon sedmodnevne upotrebe glukokortikosteroidne terapije ($Z = 2,197$).

Ključne reči: bronhijalna astma, pogoršanje, intercelularni adhezivni molekul 1, glukokortikosteroidi

References

1. Rothlein R, Ckajkowski M, O Neill MM, Mailin SD, Mainolfi E, Merluzzi VJ. Induction of intercellular adhesion molecule 1 on primary and continuous cell lines by pro-inflammatory cytokines. J Immunol (1988): 141: 1665 9.
2. Hirata N, Kohrogi H, Iwagoe H, Goto E, Hamamoto J, Fujii K, Yamaguchi T, Kawano O, Ando M. Allergen exposure induces the expression of endothelial adhesion molecules in passively sensitized human bronchus: time course and the role of cytokines. Am J Respir Cell Mol Biol 1998; 18 (1): 12 20.
3. Van der Saag PT, Caldenhoven E, van de Stolpe A. Molecular mechanisms of steroid action: a novel type of cross-talk between glucocorticoids and NF-kappa B transcription factors. Eur Respir J Suppl 1996; 22: 146s 53s.
4. Van de Stolpe A, van der Saag PT. Intercellular adhesion molecule-1 (ICAM-1). J Mol Med 1996; 74 (1); 13 33.
5. Holgate S. Mediator and cytokine mechanisms in asthma. Thorax 1993; 48: 103 9.
6. Wegner CD, Gundel RH, Reilly P, Haynes N, Letts G, Rothlein R. Intercellular adhesion molecule-1 (ICAM-1) in the pathogenesis of asthma. Science 1990; 247: 456 9.
7. Global Strategy for Asthma Management and Prevention. National Heart Lung and Blood Institute National Institutes of Health. NHLBI/WHO Workshop Report NIH Pub No 95 3659, 1995, Revised 2002, 2.
8. Jekel JF, Elmore JG, Katz DL. Epidemiology Biostatics and Preventive Medicine. Philadelphia, WB Saunders Company, 1996.
9. Erić-Marinković J, Dotlić R, Janošević S, Kocev N, Gajić M, Ille T, Stanisavljević D, Babić D. Statistika za istraživače u oblasti medicinskih nauka. Beograd, Medicinski fakultet 2001.
10. Shiota Z, Sato T, Ono T. Serum levels of soluble ICAM-1 in asthmatic patients. Arerugi 1993; 42 (12): 1782 7.
11. Shiota Z, Wilson JG, Marukawa M, Ono T, Kaji M. Soluble intercellular adhesion molecule 1 (ICAM-1) antigen in sera of bronchial asthmatics. Chest 1996; 109 (1): 94 9.
12. Bagnato G, Gulli S, Altavilla D, Squadrito F, Giacobbe O, Purello D' Ambrosio F. Circulating adhesion molecules in bronchial asthma. J Investig Allergol Clin Immunol 1998; 8 (2): 105 8.
13. Adachi T, Chihara J. Test for bronchial asthma. Rinsho Byori 2000; 48 (12): 1125 9.
14. Kokuludag A, Sin A, Terzioglu E, Saydam G, Sebik F. Eleveation of serum eosinophil cationic protein, soluble tumor necrosis factor receptors and soluble intercellular adhesion molecule-1 levels in acute bronchial asthma. J Investig Allergol Clin Immunol 2002; 12 (3): 211 4.
15. Tang RB, Chen SJ, Soong WJ, Chung RL. Circulating adhesion molecules in sera of asthmatic children. Pediatr Pulmonol 2002; 33 (4): 249 54.
16. Hashimoto S, Imai K, Kobayashi T, Ammemiya E, Takahashi Z, Tomita Z, Iwata T, Suguro H, Yamaguchi M, Yachi A, et al. Elevated levels of soluble ICAM-1 in sera from patients with bronchial asthma. Allergy 1993; 48 (5): 370 2.
17. Kobayashi T, Hashimoto S, Imai K, Amemiya E, Yamaguchi M, Yachi A, Horie T. Elevation of serum soluble intercellular adhesion molecule-1 (sICAM-1) and sE-selectin levels in bronchial asthma. Clin Exp Immunol 1994; 96 (1): 110 5.

Received: October 15, 2003

Accepted: December 18, 2003