Online Submissions: wjg.wjgnet.com
wjg@wjgnet.com
doi:10.3748/wjg.15.3916

World J Gastroenterol 2009 August 21; 15(31): 3916-3919
World Journal of Gastroenterology ISSN 1007-9327

BRIEF ARTICLES

Soluble intercellular adhesion molecule-1, D-lactate and diamine oxidase in patients with inflammatory bowel disease

Wei-Bing Song, Yong-Hui Lv, Zhen-Shu Zhang, Ya-Nan Li, Li-Ping Xiao, Xin-Pei Yu, Yuan-Yuan Wang, Hong-Li Ji, Li Ma

Wei-Bing Song, Ya-Nan Li, Li-Ping Xiao, Xin-Pei Yu, Department of Gerontology, Guangzhou General Hospital of the PLA, Guangzhou 510010, Guangdong Province, China
Yong-Hui Lv, The Traditional Chinese Medical Hospital of Guangzhou City, Guangzhou 510010, Guangdong Province, China
Zhen-Shu Zhang, Yuan-Yuan Wang, Hong-Li Ji, Institute for Digestive Diseases, Nanfang Hospital, Southern Medical University, Guangzhou 510515, Guangdong Province, China
Li Ma, Suzhou River Cadre's Sanatorium, Second Artillery Rest of the Cadres of the PLA, Beijing 100086, China
Author contributions: All authors contributed to the writing and approved the final manuscript.
Correspondence to: Dr. Wei-Bing Song, MD, Department of Gerontology, Guangzhou General Hospital of the PLA, Guangzhou 510010, Guangdong Province, China. swb535@126.com
Telephone: +86-20-36653497 Fax: +86-20-38848945
Received: May 13, 2009 Revised: July 1, 2009
Accepted: July 8, 2009
Published online: August 21, 2009

Abstract

AIM: To study the levels of serum soluble intercellular adhesion molecule-1 (sICAM-1), plasma D-lactate and diamine oxidase (DAO) in patients with inflammatory bowel disease (IBD), and the potential clinical significance.

METHODS: Sixty-nine patients with IBD and 30 healthy controls were included in this study. The concentration of sICAM-1 was detected with enzyme-linked immunosorbent assay, the level of D-lactate and DAO was measured by spectroscopic analysis, and the number of white blood cells (WBC) was determined by routine procedure.

RESULTS: The levels of sICAM-l, DAO, and WBC in IBD patients were significantly higher than those in the control group ($P < 0.01$). sICAM-l in IBD patients was found to be closely related to the levels of DAO and D-lactate (212.94 ± 69.89 *vs* 6.35 ± 2.35, $P = 0.000$), DAO 212.94 ± 69.89 *vs* 8.65 ± 3.54, $P = 0.000$) and WBC (212.94 ± 69.89 *vs* 7.40 ± 2.61, $P = 0.000$), but no significant difference was observed between patients with ulcerative colitis and patients with Crohn's disease. The post-treatment levels of sICAM-l, D-lactate and WBC were significantly lower than before treatment (sICAM-l 206.57 ± 79.21 *vs* 146.21 ± 64.43, $P = 0.000$), (D-lactate 1.46 ± 0.94 *vs* 0.52 ± 0.32, $P = 0.000$) and (WBC 7.24 ± 0.2.33 *vs* 5.21 ± 3.21, $P = 0.000$).

CONCLUSION: sICAM-1, D-lactate and DAO are closely related to the specific conditions of IBD, and thus could be used as a major diagnostic index.

Key words: Inflammatory bowel diseases; Intercellular adhesion molecule-1; D-lactate; Diamine oxidase

Peer reviewers: Bret Lashner, MD, Professor of Medicine, Department of Gastroenterology, Cleveland Clinic/A30, 9500 Euclid Avenue, Cleveland, OH 44195, United States; Atsushi Mizoguchi, Assistant Professor, Experimental Pathology, Massachusetts General Hospital, Boston, MA 02114, United States

Song WB, Lv YH, Zhang ZS, Li YN, Xiao LP, Yu XP, Wang YY, Ji HL, Ma L. Soluble intercellular adhesion molecule-1, D-lactate and diamine oxidase in patients with inflammatory bowel disease. *World J Gastroenterol* 2009; 15(31): 3916-3919 Available from: URL: http://www.wjgnet.com/1007-9327/15/3916.asp DOI: http://dx.doi.org/10.3748/wjg.15.3916

INTRODUCTION

The barrier function of the intestinal mucosa is of vital importance in inflammatory bowel diseases (IBD). Analysis of this function provides an important basis in the diagnosis of intestinal mucosal barrier dysfunction. Therefore, the importance of a timely, correct assessment of intestinal mucosal barrier function cannot be overestimated in judging the patient's disease state, estimating the prognosis and determining a comprehensive treatment program. However, direct observation of the intestinal barrier function involves many difficulties, so observation mostly needs to be made indirectly.

D-lactate, a metabolic end product of gastrointestinal bacteria, can be produced by many of these bacteria. As mammals do not have an enzyme system capable of its decomposition, D-lactate will enter the blood when the intestinal barrier function is damaged, and as they do

not have D-lactate dehydrogenase, a rise in the level of D-lactate can be detected when the permeability of the intestinal mucosa is increased. Therefore, examination of peripheral blood can reveal the degree of damage of the intestinal mucosa and the change in its permeability[1]. Diamine oxidase (DAO) is a highly active intracellular enzyme in the cytoplasm of the upper chorial cells of the intestinal mucosa. In cases where the intestinal mucosal epithelial cells and barrier function are damaged, the release of DAO is increased, and DAO enters the extracellular space, lymphatic vessels and bloodstream, thus increasing the level of DAO in the plasma. As the activity of DAO is stable, its concentration in the blood can reflect the damage and restoration of the intestinal cavity[2].

Overactivation of leukocytes is an important pathological process of IBD. The stagnation and infiltrative exosmosis of leukocytes depend on the expression and function of the intracellular adhesion molecules (ICAMs) of leukocytes and endotheliocytes at the inflammatory location.

ICAMs are a type of glycoprotein synthesized by cells and assembled on the cell surface or secreted to the cell epimatrix, and can promote the adhesion between cells or between cells and the epimatrix. The role of ICAM-1, belonging to the immunoglobulin superfamily, in the development of IBD, has been receiving more and more attention in recent years. The ICAM-1 of normal tissues is usually expressed at low levels in vascular endothelial cells, and in mononuclear macrophagocytes in the intestinal mucosal lamina propria and lymph. In the intestinal tissues of patients with IBD, the expression and distribution of ICAM-1 is significantly increased and is closely related to the degree of inflammation of the tissues[3]. The adhesive molecules in vascular endothelial cells, leukocytes or other cells can be swallowed into the cells, or peel off into the blood circulation, becoming soluble intercellular adhesion molecule-1 (sICAM-1). The post-translation product of the mRNA of some cells, are possibly not expressed on the cellular surface, but are directly secreted into the blood and is thus another important source of sICAM-1[4,5]. The increase in sICAM-1 in the serum is a marker of the damage or activation of endotheliocytes. Therefore, the level of the sICAM-1 in the serum is a significant index in the detection of some diseases[6,7].

In this research, sICAM-1, D-lactate and DAO of 69 patients with IBD were measured quantitatively with the objective of assessing their changes and clinical significance.

MATERIALS AND METHODS

Materials

Subjects: Test group including 69 patients, aged 18-60 years, with IBD hospitalized in our department [41 cases of ulcerative colitis (UC), 27 males and 13 females; 19 cases of Crohn's disease (CD), 11 males and eight females]. All patients were diagnosed by enteroscopy and were given the standard treatment set by the National Symposium on IBD, 2000 for 15 d before reassessment. Thirty healthy blood donors are used as control group.

Reagents and apparatus: Human serum sICAM-1 enzyme-linked immunosorbent assay (ELISA) kit (purchased from Boehringer Mannheim, Germany); D-lactate standard solution and D-lactic acid dehydrogenase, O-dianisidine, cadaverine dihydrochloride, horseradish peroxidase and DAO standard solution (all purchased from Sigma). The following analytical reagents were prepared in the laboratory: methotrexate injection (purchased from Zhejiang Wanma Pharmaceutical Ltd.); sulfasalazine tablets (purchased from Shanghai Sanwei Pharmaceutical Ltd.); superoxide dismutase and myeloperoxidase kit (purchased from Nanjing Jiancheng Biological Institute).

Major analysis apparatus: 721 spectrophotometer (Shanghai Sophisticated Scientific Instruments Ltd.).

Methods

A 3 mL sample of venous blood was collected from each subject of the test and control groups. After the sample was injected into dry test tubes and the serum was separated centrifugally, the serum was stored at -20℃ for examination.

The level of sICAM-1 in the serum was detected with an ELISA, the plasma level of D-lactate was determined, after the plasma was deproteinized with perchloric acid, using enzyme-coupled UV-spectrophotometry[8]; the serum DAO was determined by spectrophotometry as by Luk *et al*[2]; the number of white blood cells (WBC) was detected routinely.

Statistical analysis

All the data are presented as mean ± SD. Statistical methods used included the *t*-test, analysis of variance and linear correlation analysis. Statistical software SPSS (version 10.0) was employed for data analysis.

RESULTS

Comparison between the test group and the control group

As is shown in Table 1, the levels of sICAM-1 (206.57 ± 79.21 *vs* 107.25 ± 52.41), D-lactate (1.46 ± 0.94 *vs* 0.82 ± 0.47), DAO (9.91 ± 5.64 *vs* 2.04 ± 0.95), and WBC (7.24 ± 2.33 *vs* 4.82±1.46) in IBD patients were significantly higher than those of the control group ($P < 0.01$).

Comparison of WBC, sICAM-1, DAO and D-lactate between the 49 UC cases of UC and 19 CD cases

As is shown in Table 2, no distinct difference was demonstrated in the levels of all four substances in the UC group and CD group ($P > 0.05$). Analysis of the levels of WBC, sICAM-1, DAO and D-lactate of the test group demonstrated that sICAM-1 was related to the levels of DAO, with the correlation coefficient being 0.321 and $P < 0.01$, and also to the level of D-lactate, with the correlation coefficient being 0.412, and $P < 0.01$.

Table 1 Comparison of the values of WBC, serum sICAM-1, DAO and plasma D-lactate between the test IBD group and control group (mean ± SD)

Group	*n*	WBC ($\times 10^9$/L)	sICAM-1 (ng/mL)	DAO (U/mL)	D-lactate (μg/mL)
Test	69	7.24 ± 2.33	206.57 ± 79.21[a]	9.91 ± 5.64[a]	1.46 ± 0.94[a]
Control	30	4.82 ± 1.46	107.25 ± 52.41	2.04 ± 0.95	0.82 ± 0.47

[a]$P < 0.01$ *vs* control group. sICAM-1: Soluble intercellular adhesion molecule-1; DAO: Diamine oxidase; WBC: White blood cells; IBD: Inflammatory bowel disease.

Table 2 Comparison of the values of the 4 substances in the ulcerative colitis (UC) group and Crohn's disease (CD) group (mean ± SD)

Group	*n*	WBC ($\times 10^9$/L)	sICAM-1 (ng/mL)	DAO (U/mL)	D-lactate (μg/mL)
UC	49	7.40 ± 2.61	212.94 ± 69.89[a]	8.65 ± 3.54[a]	6.35 ± 2.35[a]
CD	20	7.29 ± 2.25	208.31 ± 51.05[b]	8.58 ± 2.49[b]	6.32 ± 2.23[b]
Control	30	4.82 ± 1.46	107.25 ± 52.41	2.04 ± 1.35	0.82 ± 0.17

[a]$P < 0.01$ *vs* control group, [b]$P < 0.01$ *vs* control group.

Table 3 Comparison of WBC, sICAM-1, DAO and D-lactate of the IBD group before and after treatment (mean ± SD)

IBD	WBC ($\times 10^9$/L)	sICAM-1 (ng/mL)	DAO (U/mL)	D-lactate (μg/mL)
Pre-treatment	7.24 ± 2.33	206.57 ± 79.21	9.91 ± 5.64	1.46 ± 0.94
Post-treatment	5.21 ± 3.21	146.21 ± 64.43[a]	6.42 ± 2.18[a]	0.52 ± 0.32[a]

[a]$P < 0.01$ *vs* pre-treatment.

Comparison of WBC, sICAM-1, DAO and D-lactate of the test group before and after treatment

The levels of WBC, sICAM-1, DAO and D-lactate of the test group were examined after the subjects were treated and the IBD was shown to be improved substantially by clinical symptoms and enteroscopy. As is shown in Table 3, the post-treatment levels of sICAM-1 (206.57 ± 79.21 *vs* 146.21 ± 64.43), D-lactate (1.46 ± 0.94 *vs* 0.52 ± 0.32), DAO (9.91 ± 5.64 *vs* 6.42 ± 2.18) and WBC (7.24 ± 2.33 *vs* 5.21 ± 3.21) were significantly lower than before treatment ($P < 0.01$).

DISCUSSION

In case of an acute attack of IBD, the structure and function of the intestinal mucosa of the patient can be seriously damaged, resulting in intestinal barrier dysfunction and thus release of intestinal bacteria, and may even induce multi-organ failure thus endangering life.

Presently, whether IBD is active is comprehensively judged primarily from clinical symptoms, erythrocyte sedimentation rate, C-reactive protein, endoscopic observations and pathology. This method can, in most cases, determine the activity of IBD quite accurately. However, for a small number of patients, those with the disease in the early stage and those with recurrence of the disease, the biochemical features are not conspicuous enough and endoscopic examination fails to display the acute activity of IBD, thus missing the opportunity of treatment because of the lack of a timely diagnosis; therefore, working out a set of monitoring indexes which are more sensitive is of substantial importance to clinical work.

The permeability of the intestinal mucosa has been used for the assessment of the development and prognosis of some diseases, such as CD, UC, *etc*. The prospective research by Tibble *et al*[9] showed that 12 mo's duration of the alteration in intestinal mucosal permeability worsens the disease, with the sensitivity and specificity being 84% and 61%, respectively. In addition, this index can also be used in prediction of the recurrence of the disease. The multi-parameter regression analysis of 47 cases of intensive care patients[9,10] demonstrated that the intestinal mucosal permeability is the only index which can predict the development of multi-organ function failure syndrome in critical patients.

Injury is a process of inflammation, during which the adhesion of leukocytes and the damage to intestinal endotheliocytes will increase the level of DAO and D-lactate. In this study, the serum DAO and D-lactate of the 69 patients with IBD before treatment were higher than those of the control group ($P < 0.01$), but no distinct difference was observed between the UC group and the CD group. The levels of DAO and D-lactate after treatment saw a marked decline. This offers proof to the fact that the levels of DAO and D-lactate, which reflect the changes in the disease condition, increase with activity of IBD but decline when the damage to the intestinal mucosa is restored after treatment.

Sans *et al*[11] found that sICAM-1 increased in the inflammatory intestinal mucosa of patients with IBD and was related to the concentration of sICAM-1 in the blood. The research by Mack *et al*[12] revealed that when

the expression of endotheliocyte ICAM-1 was lowered, the interaction between endotheliocytes and leukocytes weakened, thus alleviating the inflammatory activity. Taniguchi *et al*[13] relieved the symptoms of colitis induced by dextran sulphate sodium by adopting preventive treatment using anti-ICAM-1 monoclonal antibody.

In our research, it was found that the content of DAO and D-lactate of IBD patients were markedly higher than those of the control group, showing that damage to a certain degree existed in the intestinal mucosa of IBD patients in the acute stage ($P < 0.01$), and the permeability increased. In addition, the post-treatment content of both DAO and D-lactate decreased, indicating that the permeability had somewhat improved.

It was also demonstrated in the research that the IBD patients' leukocyte number and sICAM-1 level before treatment were markedly higher than those of the control group ($P < 0.01$) but markedly declined after treatment ($P < 0.01$). This testified to the fact that the sICAM-1 level was obviously related to the condition of the disease. The research on anti-intracellular adhesion molecule monoclonal antibody in the treatment of IBD also pointed to the role of sICAM-1 in IBD.

To sum up, the tissue damage caused by the adhesion between leukocytes and endotheliocytes is one of the common pathologies in IBD produced by bacteria and cytokines. As high expression of sICAM-1 is the important molecular basis for the increase of the adhesion between leukocytes and endotheliocytes, blocking this adhesion has become an effective preventive and therapeutic method; therefore, sICAM-1, DAO and D-lactate in the serum can be utilized as important monitoring indexes in the treatment of IBD, and sICAM-1 may be a promising molecular indicator of the activity of IBD.

COMMENTS

Background

The barrier function of the intestinal mucosa is of vital importance in inflammatory bowel diseases (IBD). Detection of this function provides an important basis in the diagnosis of intestinal mucosal barrier dysfunction.

Research frontiers

The importance of a timely correct assessment of the intestinal mucosal barrier function cannot be overestimated in judging the patient's disease state, estimating the prognosis and determining a comprehensive treatment program. However, direct observation of the intestinal barrier function involves many difficulties, so the observation mostly needs to be made indirectly.

Innovations and breakthroughs

Presently, whether IBD is active is comprehensively judged primarily from clinical symptoms, erythrocyte sedimentation rate, C-reactive protein, endoscopic observations and pathology. This method can, in most cases, determine the activity of IBD quite accurately. However, for a small number of patients, those with the disease in the early stage and those with recurrence of the disease, the biochemical features are not conspicuous enough and endoscopic examination will fail to display the acute activity of IBD, thus missing the opportunity of treatment because of the lack of a timely diagnosis; therefore, determination of a set of monitoring indexes which are more sensitive is of substantial importance to clinical work.

Applications

Serum soluble intercellular adhesion molecule-1 (sICAM-1), plasma D-lactate and diamine oxidase can be used as very important diagnostic indices, and sICAM-1 may be a useful molecular marker of the activity of IBD.

Peer review

In this manuscript, Song *et al* report that serum levels of sICAM-1, D-lactate, and diamine oxidase are increased in IBD patients. Although the sample numbers tested seem to be relatively low, I feel that this manuscript contains potentially attractive data that would be of benefit to both clinical and basic scientific fields of IBD.

REFERENCES

1 **Murray MJ**, Barbose JJ, Cobb CF. Serum D(-)-lactate levels as a predictor of acute intestinal ischemia in a rat model. *J Surg Res* 1993; **54**: 507-509

2 **Luk GD**, Bayless TM, Baylin SB. Plasma postheparin diamine oxidase. Sensitive provocative test for quantitating length of acute intestinal mucosal injury in the rat. *J Clin Invest* 1983; **71**: 1308-1315

3 **Cheon JH**, Kim JS, Kim JM, Kim N, Jung HC, Song IS. Plant sterol guggulsterone inhibits nuclear factor-kappaB signaling in intestinal epithelial cells by blocking IkappaB kinase and ameliorates acute murine colitis. *Inflamm Bowel Dis* 2006; **12**: 1152-1161

4 **Madri JA**, Graesser D. Cell migration in the immune system: the evolving inter-related roles of adhesion molecules and proteinases. *Dev Immunol* 2000; **7**: 103-116

5 **Stanimirovic D**, Satoh K. Inflammatory mediators of cerebral endothelium: a role in ischemic brain inflammation. *Brain Pathol* 2000; **10**: 113-126

6 **Theien BE**, Vanderlugt CL, Eagar TN, Nickerson-Nutter C, Nazareno R, Kuchroo VK, Miller SD. Discordant effects of anti-VLA-4 treatment before and after onset of relapsing experimental autoimmune encephalomyelitis. *J Clin Invest* 2001; **107**: 995-1006

7 **Engelhardt B**, Wolburg-Buchholz K, Wolburg H. Involvement of the choroid plexus in central nervous system inflammation. *Microsc Res Tech* 2001; **52**: 112-129

8 **Günel E**, Cağlayan O, Cağlayan F. Serum D-lactate levels as a predictor of intestinal ischemia-reperfusion injury. *Pediatr Surg Int* 1998; **14**: 59-61

9 **Tibble JA**, Sigthorsson G, Bridger S, Fagerhol MK, Bjarnason I. Surrogate markers of intestinal inflammation are predictive of relapse in patients with inflammatory bowel disease. *Gastroenterology* 2000; **119**: 15-22

10 **Suenaert P**, Bulteel V, Den Hond E, Geypens B, Monsuur F, Luypaerts A, Ghoos Y, Rutgeerts P. In vivo influence of nicotine on human basal and NSAID-induced gut barrier function. *Scand J Gastroenterol* 2003; **38**: 399-408

11 **Sans M**, Panés J, Ardite E, Elizalde JI, Arce Y, Elena M, Palacín A, Fernández-Checa JC, Anderson DC, Lobb R, Piqué JM. VCAM-1 and ICAM-1 mediate leukocyte-endothelial cell adhesion in rat experimental colitis. *Gastroenterology* 1999; **116**: 874-883

12 **Mack WJ**, Mocco J, Hoh DJ, Huang J, Choudhri TF, Kreiter KT, Lozier A, Opperman M, Poisik A, Yorgason J, Solomon RA, Mayer SA, Connolly ES. Outcome prediction with serum intercellular adhesion molecule-1 levels after aneurysmal subarachnoid hemorrhage. *J Neurosurg* 2002; **96**: 71-75

13 **Taniguchi T**, Tsukada H, Nakamura H, Kodama M, Fukuda K, Saito T, Miyasaka M, Seino Y. Effects of the anti-ICAM-1 monoclonal antibody on dextran sodium sulphate-induced colitis in rats. *J Gastroenterol Hepatol* 1998; **13**: 945-949

S- Editor Tian L **L- Editor** Cant MR **E- Editor** Zheng XM