THE CONTENT OF OLIGOMERIC AND TOTAL FRACTIONS OF SOLUBLE CD38 MOLECULES IN BLOOD SERUM OF PATIENTS WITH HYSTEROMYOMA

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© **M.E. Mamaeva,** Head of Gynecology Department of Inpatient Department No.1[1]; Applicant for a Degree, the Department of Oncology, the Faculty of Doctors' Advanced Training[2];
D.V. Novikov, PhD, Leading Research Worker, Scientific Research Institute of Molecular Biology and Regional Ecology[3];
A.V. Alyasova, D.Med.Sc., Professor, the Department of Oncology, the Faculty of Doctors' Advanced Training[2];
G.E. Kopylova, PhD, Associate Professor, the Department of Molecular Biology and Immunology[3];
M.V. Khazov, Chief Doctor of Inpatient Department No.1[1];
A.V. Karaulov, D.Med.Sc., Corresponding Member of of Russian Academy of Medical Science, Head of the Department of Clinical Immunology and Allergology[4];
V.V. Novikov, D.Bio.Sc., Professor, Head of the Department of Molecular Biology and Immunology, Director of Scientific Research Institute of Molecular Biology and Regional Ecology[3]

[1]Privolgzhsky District Medical Center of Federal Medico-Biologic Agency of Russia, Nizhne-Volzhskaya naberezhnaya St., 2, Nizhny Novgorod, Russian Federation, 603005;
[2]Nizhny Novgorod State Medical Academy, Minin and Pozharsky Square, 10/1, Nizhny Novgorod, Russian Federation, 603005;
[3]Lobachevsky State University of Nizhni Novgorod — National Research University, Prospekt Gagarina, 23, Nizhny Novgorod, Russian Federation, 603950;
[4]I.M. Sechenov First Moscow State Medical University, Trubetskaya St., 8, bld. 2, Moscow, Russian Federation, 119991

The aim of the investigation was to study the content of oligomeric fraction and total fraction of soluble CD38 molecules in blood serum of patients with hysteromyomas.

Materials and Methods. 53 patients with hysteromyomas aged 32–58 years were under study. The level of soluble CD38 molecules in blood serum was determined by enzyme immunoassay using polyclonal antibodies and monoclonal antibodies ICO-20 against CD38. The results were stated in conditional units (U/ml).

Results. The development of benign pathology of uterine body was accompanied by simultaneous level growth of oligomeric and total fractions of soluble CD38 molecules. The most significant increase of total fraction level occurred in submucous localization of myomatous nodes. Interstitial-submucous tumors had the highest serum level of oligomeric fraction of CD38 molecules. 4–6 myomatous nodes were accompanied by the highest level of total fraction of CD38 molecules, the content of oligomeric fraction of the molecules under study being normal. By contrast, the patients with one myomatous node were found to have the highest level of soluble oligomeric CD38 molecules, the total fraction level of soluble CD38 molecule being medium.

Key words: soluble CD38 molecules; oligomeric fraction of CD38 molecules; total fraction of CD38 molecules; hysteromyoma.

Hysteromyoma is the most common female benign genital tumor. In the pathogenesis of the disease, among constitutional-hereditary and metabolic disorders, the immune system dysfunction plays a critical role [1]. One of the indicators of immune system dysfunction is imbalance in serum pool of soluble differential molecules participating in immune response regulation and realization [2, 3]. Several tens of soluble differential molecules are known to compose a pool, and among these molecules there are apoptosis mediating soluble molecules Fas (CD95), molecules ICAM-1 (CD54) modulating adherent cell-mediated response, and others [4–9]. The most soluble differential molecules have been found to be able to be in different structural conditions. In particular, they can be monomer or oligomeric, can form various associates having an impact on their functions [3, 10]. So, a soluble CD38 molecule has been shown to be present in blood not only as a monomer protein but also as an oligomer consisting of two or four noncovalently linked identical protein molecules [11, 12]. There have been developed enzyme immunoassays, which enable to determine the content of serum fractions of oligomeric

For contacts: Mamaeva Marina Evgenievna, e-mail: mamaevame@yandex.ru

CD38 molecules, as well as the content of total fraction of CD38 molecules including oligomeric and monomer protein forms [11–13]. Serum content of these fractions in some immune-mediated diseases has been found to be of a monitoring value and change multidirectionally [12, 14, 15]. There is evidence that in hysteromyoma there is tendency for the increase of serum content of soluble CD38 molecules against the growth of a relative number of CD38-positive mononuclear cells of patients' peripheral blood [16].

The aim of the investigation was to carry out a detailed analysis of the content of oligomeric and total fractions of soluble CD38 molecules in blood serum of patients with hysteromyomas.

Materials and Methods. We examined 53 patients with hysteromyomas aged 32–58 years. In all cases the diagnosis was confirmed by postmortem examination of postoperative material. 25 women (47.2%) underwent supracervical uterus amputation, 16 (30.2%) — concervative myomectomy, and 12 (22.6%) — hysterectomy with bilateral oophorectomy. A control group involved 45 women without gynecological pathology, comparable to the patients in age.

The study complies with the declaration of Helsinki (adopted in June, 1964 (Helsinki, Finland) and revised in October, 2000 (Edinburg, Scotland)) and approved by the Ethics Committee of Nizhny Novgorod State Medical Academy. Written informed consent was obtained from all patients.

Blood samples for test were drawn before treatment and 8–10 days after surgery. The level of soluble differential molecules in blood serum was determined by two-site enzyme immunoassay. The content of total fraction of soluble CD38 molecules was determined using polyclonal antibodies against antigens of mononuclear cells of peripheral blood as scaffolding, and monoclonal antibodies ICO-20 horseradish peroxidase-conjugated. The content of oligomeric fraction of soluble CD38 molecules was determined using monoclonal antibodies ICO-20 and monoclonal antibodies ICO-20 horseradish peroxidase-conjugated sorbed in a well-plate for enzyme immunoassay [11, 13]. The findings were calculated at 405 nm wavelength spectrophotometrically using a Multiscan photometer (Finland). The obtained data were stated in conditional units U/ml, and processed using a statistical package program Statistica 6.0.

Results and Discussion. In the control group the content of oligomeric fraction of soluble CD38 (ol. sCD38) molecules was 257.20±14.83 U/ml, total fraction (sCD38) — 203.60±14.54 U/ml. Hysteromyoma development was accompanied by significant ol.sCD38 concentration increase in patients' blood plasma — by 1.8 times compared to the control (463.3±49.0 U/ml). Alongside with that there was the significant increase of sCD38 level — by 2.4 times (496.1±45.40 U/ml). Thus, the development of benign tumors of uterus is accompanied by an increased level of both total and oligomeric fractions of CD38 protein in patients' blood serum. The change of the concentration of soluble CD38 molecules in pericellular space and biological fluids is known [2, 10] to act as a factor influencing the global immunological net state. An increased content of this protein in blood serum of patients resulting from immune system activation limits an immune response (a feedback principle), and, is likely to serve as one of tumor immune escape mechanisms.

ol.sCD38 and sCD38 changes were studied depending on myomatous node localization. In a group of patients with interstitial localization, ol.sCD38 and sCD38 levels in blood serum were significantly higher compared to the healthy subjects — by 1.9 and 2.4 times. In cases with interstitial-subserous localization of tumors, the levels of soluble proteins did not differ from the norm, and ol.sCD38 content appeared to be significantly lower (by 1.6 times) than in the previous group (Table 1). The patients with subserous localization of tumors were

Table 1

Serum content of oligomeric and total fractions of soluble CD38 molecules in patients with myomatous nodes with different localizations, U/ml

Localization of myomatous nodes	Oligomeric fraction	Total fraction
Interstitial (n=9)	498.3±53.6*	497.7±86.9*+
Interstitial-subserous (n=11)	320.3±42.9+€^	347.9±40.1+
Subserous (n=12)	313.5±36.7+€^	351.7±48.2+
Interstitial-submucous (n=10)	563.6±61.0*+	398.5±41.4*+
Submucous (n=11)	499.6±55.8*	937.3±82.2*
Control group (n=45)	257.20±14.83	203.60±14.54

* — the differences are significant compared to the control ($p<0.05$); + — compared to submucous localization of myomatous nodes ($p<0.05$); € — compared to interstitial localization of myomatous nodes ($p<0.05$); ^ — compared to interstitial-submucous localization of myomatous nodes ($p<0.05$).

Table 2

Serum level of oligomeric and total fractions of soluble CD38 molecules in patients with different numbers of myomatous nodes, U/ml

The number of myomatous nodes	Oligomeric fraction	Total fraction
1 node (n=11)	499.2±65.7*+	433.6±36.6*+
2–3 nodes (n=20)	391.4±48.6*	423.5±34.6*+
4–6 nodes (n= 22)	331.1±46.4	648.1±60.4*
Control group (n=45)	257.20±14.83	203.60±14.54

* — the differences are significant compared to the control ($p<0.05$); + — compared to the patients with 4–6 myomatous nodes ($p<0.05$).

Table 3

The effect of surgery type on the level of oligomeric and total fractions of soluble CD38 molecules in patients with hysteromyomas, U/ml

Surgery type	Oligomeric fraction		Total fraction	
	before treatment	after treatment	before treatment	after treatment
CM (n=16)	352.50±44.53	421.12±44.81	339.18±45.14	412.93±44.40
SUA (n=25)	477.80±82.78	467.20±82.87	488.92±64.12	477.0±63.98
HBO (n=12)	580.83±33.67	585.75±43.56	569.58±32.19	575.83±43.91
All surgery types	463.3±49.0	451.73±49.11	496.10±45.40	480.03±58.26
Control group (n=45)	257.20±14.83		203.60±14.54	

Note: CM — conservative myomectomy; SUA — supracervical uterus amputation; HBO — hysterectomy with bilateral oophorectomy; $p<0.05$.

found to have no differences compared to the norm in the content of soluble proteins, though they had significant decrease of ol.sCD38 compared to the patients with interstitial localization of myomatous nodes.

In interstitial-submucous localization of nodes, the ol.sCD38 level significantly differed from the norm — by 2.2 times, the content of sCD38 molecules increased by 1.7 times. In this group the content of ol.sCD38 was significantly higher than in patients with interstitial-subserous and subserous localization of tumors (in both cases — by 1.8 times). Moreover, ol.sCD38 level significantly exceeded the same index in submucous tumors. In patients suffering primarily from submucous tumors, ol.sCD38 content exceeded the norm by 1.9 times ($p<0.05$), sCD38 content — by 4.6 times ($p<0.05$). Serum level of ol.sCD38 in this group of patients was significantly higher than in women with interstitial subserous and subserous tumors (respectively, by 1.6 and 2.7 times). The level of sCD38 molecules in these patients was significantly higher than in all other compared groups (by 1.9–2.7 times).

The findings of the study suggest that submucous localization of myomatous node is accompanied by the more significant increase in total fraction of soluble CD38 molecules against the growth of oligomeric fraction of the protein. In cases with other localizations, the values of ol.sCD38 and sCD38 were increasing more evenly. It should be noted that severe pain syndrome, frequent bleedings, which are manifested moderately in other localizations, prevail in clinical presentation of submucous myomatous nodes. Thus, the revealed characteristics of an immune response in patients with myomas are associated with the formation of benign tumor processes of various localizations in the body of uterus.

We also studied oligomeric and total fractions of soluble CD38 molecules in patients having different numbers of myomatous nodes (Table 2). In patients with 1 myomatous node, ol.sCD38 content in blood serum grew significantly compared to the control: by 1.9 times. In patients with 2–3 tumor nodes the content of ol.sCD38 exceeded the norm by 1.5 times ($p<0.05$). In women with 4–6 myomatous nodes the protein level did not differ from the norm.

sCD38 content in all the groups under study significantly exceeded the norm by 2–3 times. The protein level in patients with 4–6 nodes was higher compared to other groups ($p\leqslant0.05$), it exceeding the serum sCD38 level by 1.4 times in women with 1 myomatous level, and by 1.5 times — in women with 2–3 tumor nodes.

The findings enable to conclude that the increase of disease severity and the probability of complications in women with benign tumors of the uterine body is accompanied by the increase of total fraction of soluble CD38 molecules and growing imbalance in correlative concentrations of sCD38 and ol.sCD38 that potentially is of monitoring value. Similar immune response failures can be a pathogenic element in the development of severer disease forms.

The performed surgery had no significant effect on the content of both oligomeric and total fractions of soluble CD38 molecules in blood serum of patients (Table 3).

Conclusion. Thus, the development of benign pathology of the uterine body is accompanied by simultaneous growth of oligomeric and total fraction levels of soluble CD38 molecules. Submucous myomatous nodes are characterized by the most significant increase of total fraction of soluble CD38 molecules. In turn, interstitial-submucous tumors had the highest serum level of oligomeic fraction of CD38 molecules. 4–6 myomatous nodes were accompanied by high level of total fraction of CD38 molecules, with normal content of oligomeric fraction of the molecules under study. The patients with one myomatous node, on the contrary, were found to have the highest level of soluble oligomeric CD38 molecules with moderate growth of total fraction of CD38 molecules. Any surgery performed has no significant effect on the content of oligomeric fraction and total fraction of soluble CD38 molecules in this group of patients.

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