gelsk region and the main directions of sanatorium-resort rehabilitation of the consequences of injuries. *Sovremenniye problemy nauki i obrazovania*. 2015; 4. (In Russ.).] http://science-education.ru/ru/article/view?id=20504 (date accessed: 02.03.2020).

2. Соколовская Т.А. Здоровье детей: основные тенденции и возможные пути его сохранения. *Современные проблемы науки и образования*. 2017; 4. [Sokolovskaya T. A. Children's health: main trends and possible ways to preserve it. *Sovremenniye problem nauki i obrazovania*. 2017; 4. (In Russ.).] http://science-education.ru/ru/article/view?id=26572 (date accessed: 02.03.2020).

3. Тырылгин М. А. Проблемы охраны здоровья населения Крайнего Севера (на примере региона Якутия). *Наука*. 2008; 303. [Tyrilgin M. A. Problems of health protection of the population of the Far North (on the example of the region of Yakutia). *Nauka*. 2008; 303. (In Russ.).]

4. Чичахов Д.А., Вербицкая Л.И. Детская смертность в Республике Саха (Якутия). *Дальневосточный медицинский журнал*. 2010; 4: 42-43. [Chichagov D. A., Verbitskaya L. I. Child mortality in the Republic Sakha (Yakutia) Republic. *Dalnevostochniy medicinskiy zhournal*. 2010; 4: 42-43. (In Russ.).] http://www.fesmu.ru/elib/Article.aspx?id=232970

5. Шигаев Н.Н., Кром И.Л., Еругина М.В., Дорогойкин Д.Л. Междисциплинарный анализ социально детерминированных рисков здоровья детского населения. *Современные проблемы науки и образования*. 2016; 2. [Shigaev N. N., Krom I. L., Erugina M. V., Dorogoikin D. L. Interdisciplinary analysis of socially determined risks of children's health. *Sovremenniye problem nauki i obrazovania*. 2016; 2. (In Russ.).] http://science-education.ru/ru/article/view?id=24246 (date accessed: 02.03.2020).

6. Collins S. A., Surmala P., Geraldine O., Greenberg Ch., Williason L. Causes and risk factors for infant mortality in Nunavut, Canada 1999–2011. *Pediatrics*. 2012; 12:190. http://www.biomedcentral.com/1471-2431/12/190

7. Skold P., Axelsson P., Karlsson L., Smith L. Infant mortality of Sami and settlers in Northern Sweden: the era of colonization 1750–1900. *Global Health Action*. 2011; 4: 8441. DOI: 10.3402/gha. v4i0.8441 https://www.researchgate.net/publication/51760521_Infant_mortality_of_Sami_and_settlers_in_Northern_Sweden_the_era_of_colonization_1750-1900

M.I. Mikhaylichenko, K.G. Shapovalov, V.A. Mudrov, O.S. Gruzdeva

THE ROLE OF MATRIX METALLOPROTEINASES AND THEIR INHIBITORS IN PATIENTS WITH LOCAL COLD INJURY

DOI 10.25789/YMJ.2020.70.22

УДК: 616-092.19

The urgency of the local cold injury (LCI) on the territory of the Russian Federation is conditioned by its geographical position, significant specific weight in the structure of the general traumatism, difficulties of etiopathogenetic therapy, disappointing treatment results, which leads to frequent loss of ability to work and permanent disability.

The aim of the study was to identify the dynamics of the level of collagenases of the second subfamily (MMR-2, MMR-9) and their inhibitors (TIMP-1 and TIMP-2) in the blood of those, who sustained local cold injury.

The study included 60 patients with frostbite of the distal segments of the lower extremities. The level of MMR–2, MMR–9, TIMP -1 and TIMP - 2 was studied on the 5th and 30th days after the frostbite using a multiplex blood test with a set of biomedical reagents.

In patients with frostbite, the level of MMR–2 and MMR–9 in the blood serum increases relative to the control group in the early stages of cryodamage, as well as an increase in the index of the markers studied directly proportional to the volume of the affected tissues. A similar dynamics was observed in the study of parameters of matrix metalloproteinase inhibitors. Based on the obtained data and analysis of the linear regression equation, a predictive model was formed that allows us to suspect the level of tissue damage in the early stages of local cold injury with high accuracy.

Keywords: local cold injury, endothelial dysfunction, matrix metalloproteinases, inhibitors of matrix metalloproteinases.

The urgency of the local cold injury (LCI) on the territory of the Russian Federation is conditioned by its geographical position, significant specific weight in the structure of the general traumatism, difficulties of etiopathogenetic therapy, disappointing treatment results [4,7,8]. The frequent loss of ability to work and high percentage of disability indicate the need for further studies of pathogenetic mechanisms of cryodamage. The study and identification of new markers of local cold injury contributes to an earlier diagnosis of the volume of tissue lesions, the creation of qualitatively new approaches to the complex treatment of frostbite of the extremities, as well as successful and fastest rehabilitation of patients [1, 8].

In recent years, scientific works clearly show the deepening of research by domestic and foreign authors on the features of the pathogenesis of cryodamage, early diagnosis of the volume of affected tissues, methods of surgical treatment of deep frostbite of the extremities and rehabilitation of patients [4, 8].

There is no doubt that endothelial dysfunction plays a fundamental role in the pathogenesis of frostbite [7-10]. Endotheliocytes damaged by cold and secondary alterations, as well as cells attracted to the focus of inflammation, increase a large number of biologically active substances, including matrix metalloproteinases (MMR) and their inhibitors (TIMP).

Recent studies have shown that collagenases, as well as their inhibitors, play a significant role in the pathogenesis of connective tissue damage and its fibrosis in various pathologies [2]. Collagenases are Zn2+ and Ca2+ dependent induced endogenous peptidases involved in tissue rearrangement, by the destruction of its organic components, as well as the exchange of proteins of the interstitial matrix [2, 3]. MMR secretion is influenced by cytokines released by endotheliocytes, fibroblasts, macrophages, platelets [3, 10].

Chita State Medical Academy 39a, Gorky street, Chita, 672000, Russian Federation; **MIKHAILICHENKO Maxim I.** – Assistant of department of faculty surgery with the course of urology. Chita state medical academy, Chita, Russian Federation. E-mail – angelo999@yandex.ru. ORCID № 0000-0001-8660-2982. Phone number: +79145222922. **SHAPOVALOV Konstantin G.** – Head of the Department of anesthesiology, intensive care. DM, professor. Chita state medical academy, Chita, Russian Federation. E-mail – shkg26@mail.ru. ORCID № 0000-0002-3485-5176. **MUDROV Viktor A.** – Associate professor of obstetrics and gynecology. Chita state medical academy, Chita, Russian Federation. E-mail – mudrov_viktor@mail.ru, ORCID № 0000-0002-5961-5400. **GRUSDEVA Olga S**. – Assistant of the Department of pathological anatomy. Chita state medical academy, Chita, Russian Federation. E-mail – lolgas18@gmail.com. ORCID № 0000-0002-1738-0850.

Relatively recently, it has been established that MMR can be activated by low pH, hyperthermia and hypothermia, as well as by processes of lipid peroxidation [14]. MMR of the second subfamily play an important role. These include type IV endopeptidases-MMR-2 and MMR-9. This group of matrix metalloproteinases remodels native collagen, changing its conformation, which makes it difficult for cells to migrate to tissues for the implementation of inflammatory processes.

Tissue inhibitors of MMR are proteins that regulate the balance of connective tissue collagen, forming high-affinity complexes with collagenases, thereby leveling the pathological processes provoked by matrix metalloproteinases. The main sites of TIMP expression are the bone tissue and ovaries [8, 9, 16].

Collagenase inhibitors (TIMR-1 and TIMR-2) are little-known and unexplored biomarkers of alterations. Although in medical practice, these biomarkers are used quite successfully in stratifying the risks of undesirable cardiovascular events, arterial hypertension, diabetes mellitus, chronic renal failure and complicated obstetrics [4, 9, 10]. Evaluating the effectiveness of pathogenetically based therapy is extremely relevant and promising for patients with local cold trauma.

It should be noted that the optimal target values in the blood of MMR and TMR, which should be equalized in the course of research, are not yet regulated. At the same time, the scientific justification for determining the target values of markers of collagen balance, which plays an important role in proliferative processes closely related to the course of the wound process, can help control the quality of therapy and allow predicting the course of the wound process in cryodamage.

Purpose of research: to determine the dynamics of the level of collagenases of the second subfamily (MMR-2, MMR-9) and their inhibitors (TIMP-1 and TIMP-2) in the blood of those, who sustained local cold injury.

Research materials and methods. The study included 60 patients (40 men and 20 women), 20 of them with frostbite of the toes (group 1), in the second group of victims, the lesion spread to the pretarsal-molar joint (n=22), group 3 – patients with frostbite of the more proximal parts of the lower limb (n=18). Depending on the volume of the affected tissues, the study was performed on the 5th day after the injury. The average age of patients was 35±7 years. The level of MMR–2, MMR–9, TIMP -1 and TIMP - 2 was measured on the 5th and 30th days after frostbite in the same patients using a multiplex analysis of serum with a set of biomedical reagents. All patients were treated at the regional burn center on the basis of GUZ "City clinical hospital No. 1" in Chita with local cold injury of III-IV degree in the period from 2018 to 2019.

The control group consisted of relatively healthy people aged 30±10 years (n=28).

All patients and volunteers gave their written voluntary informed consent, in accordance with the requirements of the Helsinki Declaration of the world medical Association (ed. 2013)

The exception group included patients with severe somatic pathology: inflammatory processes of various localization, obliterating diseases of the arteries, nerve damage of the extremities, diabetes, hypertension, alimentary exhaustion and obesity, and individual dependence of various Genesis.

Statistical processing of the research results was carried out using the IBM SPSS Statistics Version 25.0 software package. The data obtained were systematized in the form of a median and a confidence interval. Considering the size of the control group of no more than 50 people, the normality of the distribution of features was assessed using the Shapiro-Wilk criterion. The statistical significance of the differences in indicators was assessed by comparing the critical and calculated values of the Kraskel-Wallis criterion with the subsequent determination of the significance level p. For a more precise description of the identified trends used the Mann-Whitney test, which allows to evaluate the performance differences between comparing groups in pairs with application of a Bonferroni correction when assessing the value of R. To assess the relationship between indicators we used the Spearman correlation coefficient (ρ). Based on the obtained values of ρ we revealed the closeness of the connection on a scale of Chedoke and its direction (direct or reverse). Subsequently, we calculated a segregated coefficient of determination, which shows the proportion of the explained dependence. The predictive model is built by linear regression.

The results and discussion. We note an increase in the level of MMR–2 and MMR–9 in serum in patients with frostbite relative to the control group in the early stages of frostbite. The level of MMR-2 decreased to the values of the control group, and the indicators of MMR-9 remained significantly higher than normal later in the period after the injury. This fact pathogenetically justifies a large number of complications in patients with frostbite.

In the late reactive period, the serum level of MMR–2 in patients with cryodamage is 3.4 times higher relative to control (p=0.011), on day 30-the values of MMR-2 do not differ in control indicators (p=0.103) (Table 1). The level of MMR-9 in patients with local cold injury on day 5 is 14.5 times higher than the control indicators (p=0.002), on day 30 – 12.5 times higher relative to the group of healthy volunteers (p<0.001) (Table 1).

Table 1

The content of matrix metalloproteinases and their inhibitors in the blood serum of victims with local cold trauma in different periods of cryodamage

Study group	MMP–2	MMP–9	TIMP–1	TIMP–2
Control group (n = 28)	2.3 (75% ДИ 2.16 – 2.45)	32.00 (75% ДИ 28.55 – 35.37)	734.4 (75% ДИ 733.51 – 770.20)	247.2 (75% ДИ 232.19 – 257.81)
First group (5 day) (n = 30)	7.97 (75% ДИ 7.59 – 8.20) p < 0.001	448.86 (75% ДИ 418.33 – 466.28) p < 0.001	1259.2 (75% ДИ 1230.83 – 1342.35) p < 0.001	555.2 (75% ДИ 528.46 – 578.94) p < 0.001
Second group (30 day) (n = 30)	4.53 (75% ДИ 4.27 – 4.92) p < 0.001 p_1<0.001	354.14 (75% ДИ 343.65 – 375.35) p < 0.001 p_1< 0.001	1481.6 (75% ДИ 1462.76 – 1646.89) p < 0.001 p_1 < 0.001	478.4 (75% ДИ 472.65 – 555.55) p < 0.001 p_1=0.024

p – reliability of the difference in indicators relative to control; p1-reliability of the difference in indicators relative to patients with frostbite in the late reactive period.

In patients with cryodamage of distal stop, the level of MMP–2 was 2.1 times higher relative to the control (p<0.001), in patients with frostbite more proximal segments of the foot – the value of MMP–2 to 2.6 times higher than the benchmarks (p<0.001). The level of MMR-2 in victims with the most severe frostbite is 3.5 times higher than the control values (p<0.001) (Table 2). The level of MMR–9 in victims with frostbite of the toes is 4.9 times higher than the control indicators (p<0.001), and in patients with more severe frostbite – 10 times higher relative to the control group (p<0.001) (Table 2). In patients with group 3 frostbite, the level of MMR-9 increased by 14.5 times relative to the control values (p<0.001) (Table 2).

In the late reactive period in patients with cryodamage, the TIMP–1 level is 1.7 times higher relative to the control (p<0.001), on day 30 – the TIMP–1 value increased 2 times relative to the control (p<0.001) (Table 1). The TIMP-2 Level in patients on day 5 from the moment of cryodamage is 2.3 times higher than the control indicators (p<0.001), on day 30 – 2.2 times higher relative to the control group (p<0.001) (Table 1).

In patients with cryodamage of distal stop, the level of TIMP–1 in the blood 2 times higher relative to the control (p<0.001), in patients with frostbite more proximal segments of the foot – value TIMP–1 is 1.8 times higher than the benchmarks (p<0.001). The level of TIMP-1 in the patients with the most severe frostbite is 1.6 times higher than the control values (p<0.001) (Table 2). The level of TIMP-2 in patients with local cold injury of the toes is 2.1 times higher than the control indicators (p<0.001), in patients with more severe frostbite – 2.2 times higher relative to the control group (p<0.001) (Table 2). In patients with group 3 frostbite, the level of TIMP-2 is 2.3 times higher than the control value (p<0.001) (Table 2).

The data obtained during the study indicate an increase in the content of matrix metalloproteinases and their inhibitors in the blood serum of those, who sustained cold trauma. At the same time, significant dynamics of the studied markers does not occur in the long-term period of cryodamage. Probably, during frostbite, a significant failure in the remodeling of the cytoskeleton, tissue repair, angiogenesis occurs, and there are prerequisites for an atypical, prolonged course of reparative processes.

Previously conducted research in patients with deforming arthrosis, in complicated obstetrics, in patients with CHD, aimed at detecting matrix metalloproteinases and their inhibitors, also revealed a persistent increase in the level of the studied predictors in the serum of patients [2, 5, 10, 11]. Scientists have concluded that metalloproteinases are one of the pathogenetic causes of chronization of any pathology [5, 6]. It is possible that collagenases are the predictor of the adverse course of local cold injury. This is also proved by the large number of adverse consequences of injury. According to a number of authors, complications are present in the majority of patients with local cold trauma [3, 4, 9].

The increased content of matrix metalloproteinase inhibitors probably indicates an attempt on the part of the body to stabilize the emerging catastrophe in the center of alterations. We found a sharp increase in TIMP-1 and TIMP-2 in all periods of cryodamage. Attention is drawn to the fact that there is no correlation of TIMP in patients with the most extensive lesions and the mildest injury limited to the toes. It is also necessary to indicate that the level of TIMP does not decrease for 30 days from the moment of cryodamage. This phenomenon is probably associated with the death of bone tissue, since inhibitors of matrix metalloproteinases are deposited mainly in osteocytes [2, 6, 11, 18].

Thus, there is a high degree of parallelism between the value of matrix metalloproteinases and their inhibitors in blood serum and the level of damage to the distal segments of the lower extremities (Table 3).

According to modern scientific data, with fibrotic tissue disorganization, the MMR index decreases, and the level of their inhibitors on the contrary increases. When collagen is degraded, on the contrary, the content of collagenases increases, and inhibitors decrease [2, 12]. In patients with frostbite, we see an increase in all the studied biomarkers of the matrix metalloproteinase system and their inhibitors, even in the case of a relatively mild cold injury.

Relatively recently, V. A. Tumansky and T. A. Khristenko [6] have shown that MMP and TIMP levels are significantly increased in patients with pancreatic cancer. Given that the complex of MMP and TIMP is formed by the "key-lock" type [5, 6], the authors suggested that the increase in the level of such biologi-

Table 2

The content of matrix metalloproteinases and their inhibitors in the blood serum of victims with local cold trauma depending on the volume of affected tissues

Study group	MMP–2 (ng/ml)	MMP–9 (ng/ml)	TIMP–1 (ng/ml)	TIMP–2 (ng/ml)
Control group (n = 28)	2.3 (75% ДИ 2.16 – 2.45)	32.00 (75% ДИ 28.55 – 35.37)	734.4 (75% ДИ 733.51 – 770.20)	247.2 (75% ДИ 232.19 – 257.81)
First group (n = 20)	4.65 (75% ДИ 4.43 – 4.77) p < 0.001	126.50 (75% ДИ 126.99 – 147.95) p < 0.001	1488.0 (75% ДИ 1420.76 – 1540.74) p < 0.001	513.6 (75% ДИ 486.84 – 529.16) p < 0.001
Second group (n = 22)	6.78 (75% ДИ 6.75 – 7.14) p < 0.001 p_1 < 0.001	314.50 (75% ДИ 315.29 – 326.41) p < 0.001 p_1 < 0.001	1292.8 (75% ДИ 1252.00 – 1352.05) p < 0.001 p_1=0.159	536.8 (75% ДИ 495.50 – 576.50) p < 0.001 p_1=0.778
Third group (n = 18)	7.97 (75% ДИ 7.59 – 8.20) p < 0.001 p_1 < 0.001 p_2 < 0.001	448.86 (75% ДИ 418.33 – 466.28) p < 0.001 p_1 < 0.001 p_2 < 0.001	1259.2 (75% ДИ 1230.83 – 1342.35) p < 0.001 p_1 < 0.001 p_2=0.007	555.2 (75% ДИ 528.46 – 578.94) p < 0.001 p_1=0.013 p_2=0.021

p – reliability of the difference in indicators relative to control; p1-reliability of the difference in indicators relative to 1 group of patients; p2-reliability of the difference in indicators relative to 2 groups of patients.

cal "locks" is a reflection of the antitumor reaction and cells with an attempt to limit the focus of alterations. It is possible that a similar biological phenomenon of cell microenvironment formation is realized in patients with frostbite, aimed at delineating the focus of damaged tissues by rough remodeling of the extracellular matrix and changing the conformation of the cell membrane.

MMR 2 in combination with other collagenases is known to inhibit angiogenesis. This phenomenon is associated with their ability to form antiangiogenic peptides by converting plasminogen into angiostatin, which inhibits proliferation and exacerbates endothelial cell apoptosis [6, 13]. This is another confirmation of the probable biological feasibility of increased MMR expression in patients with frostbite. Inhibition of microcirculation was confirmed by our earlier studies in patients with local cold trauma [3, 8, 9]. Collagenases have been found to have an inhibitory effect on inflammation by processing a significant amount of chemokines, including pro-inflammatory interleukins. Probably, the inhibitory effect of MMR-2 is associated with worsening endothelial dysfunction and the formation of a large number of adverse sequential reactions, forming a kind of protective reaction of the body to increasing perifocal inflammation [3].

It was found that the level of IL-1, IL-8, IL–12 and TNFa in the blood was increased during all periods of cryodamage [9, 10]. However, in the later periods of frostbite, the level of Pro-inflammatory interleukins decreases, but still remains above normal values. However, the level of MMR-9 remains high in patients and in remote periods of local cold injury. Synthesis of these metalloproteinases occurs mainly in macrophages, as well as platelets, which realize their functions in the interstitial [6, 14]. One of the features of MMR–9 is the chemotaxis of fibroblasts into the interstitial space for the implementation of repair processes [12, 16, 17]. On the other hand, MMR-9 contributes to collagen desmoplasia [11, 15]. Remodeling of collagen in the lesion site provokes discoordination of proliferation and, as a result, a prolonged and unfavorable course of the wound process.

Based on the analysis of the linear regression equation, which included the initial indicators (values of matrix metalloproteinases and their inhibitors), the significance of indicators in the structure of the predictive model was determined (Table 4).

Considering the insufficient significance, the TIMP-1 indicator was removed from the structure of the original model. As a result, a pattern was determined by repeated linear regression, which is expressed by the formula: K = 0.165 × MMP-2 + 0.003 × MMP-9 + 0.01 × TIMP-2 – 2.5, the name of the units of measurement of indicators is ng/ml. At values Of K from 1.0 to 4.4, frostbite of the toes is diagnosed, at K from 4.4 to 5.4, the lesion boundary extended to the pre-tarsal-metatarsal joint, more than 5.4 - to the level of the lower third of the lower leg (Table 5).

Taking into account the value of the adjusted coefficient of determination (0.760), we can consider the relationship between the coefficient of K and the level of damage to the distal segments of the lower extremities in local cold trauma to be quite strong.

Consequently, the analysis of the obtained data allows to establish that increased expression of collagenase and their inhibitors reflects the severity and course of the cold injury due to endothelial dysfunction and formation of fibrosis in the lesions and the perifocal. It is possible that an imbalance of the collagenase system can cause structural and functional changes in the extracellular matrix and cellular structures, contributing to their pathological remodeling and fibrosis [3, 7, 9, 10].

Table 3

The actual degree of parallelism between the value of matrix metalloproteinases and their inhibitors and the level of damage to the lower extremities

Parameter	Spearman correlation coefficient	Statistical significance	Relationship strength on a scale of Chedoke
MMP–2	0.797	< 0.001	High straight
MMP–9	0.907	< 0.001	High straight
TIMP–1	- 0.186	0.002	Weak reverse
TIMP–2	0.381	< 0.001	Moderate straight

Table 4

Significance of indicators in the structure of the predictive model

Model	Non-standardized coefficients		Standardized coefficients		
	B	Standard error	Бета	т	Significance in the model structure
Constant	- 0.466	0.181		- 2.575	0.011
MMP–2	0.165	0.017	0.378	9.578	0.0001
MMP–9	0.003	0.0001	0.511	12.970	0.0001
TIMP–1	- 6.809	0.0001	- 0.019	- 0.643	0.520
TIMP–2	0.001	0.0001	0.136	4.170	0.0001

Table 5

Summary for the resulting predictive model

Model	R	R^2	adjusted R^2	The standard error of estimate
K = 0.165 × MMP-2 + 0.003 × MMP-9 + 0.01 × TIMP-2 – 2.5	0.873	0.763	0.760	0.451

Conclusions:

1. It was found that the level of matrix metalloproteinases increases in the blood of patients with local cold injury. The increase in the collagenase index in the blood serum of patients is directly proportional to the severity of the injury.

2. It is revealed that the performance of inhibitors collagenase increased significantly in all periods of cryodamage. Increasing levels of inhibitors collagenase is not directly dependent on the severity of cold injury.

3. The pattern formed on the basis of the studied markers can serve as a predictive criterion for early determination of the severity of cryodamage.

References

1. Васина Л.В., Власов Т.Д., Петрищев Н.Н. Функциональная гетерогенность эндотелия (обзор). *Артериальная гипертензия*. 2017;

(2): 88-102. [Vasina LV, Vlasov TD, Petrishchev NN. Functional heterogeneity of the endothelium (review). *Hypertension.* 2017; (2): 88-102. (In Russ).]

2. Егорова Е.Н., Мазур В.В., Калинкин М.Н., Мазур Е.С. Взаимосвязь эндотоксемии, факторов системного воспаления и компонентов системы матриксных металлопротеиназ – тканевых ингибиторов металлопротеиназ при ХСН. *Сердечная недостаточность*. 2012; 72(4): 233-236. [Egorova E. N., Mazur VV, Kalinkin MN, Mazur ES. Relationship of endotoxemia, factors of systemic inflammation and components of the matrix metalloproteinase system-tissue inhibitors of metalloproteinases in CHF. *Heart failure*. 2012; 72(4): 233 — 236. (In Russ).]

3. Коннов Д.Ю., Малярчиков А.В., Шаповалов К.Г., Коннов В.А. Закономерности изменений параметров микроциркуляции и электрофизиологических показателей сердечного ритма при критической гипотермии. *Забайкальский медицинский вестник*. 2017;4: 17-24. [Konnov DYu, Malyarchikov AV, Shapovalov KG, Konnov VA. Regularities of changes in microcirculation parameters and electrophysiological parameters of heart rate in critical hypothermia. *Zabaikalsky medical journal*. 2017; 4: 17-24. (In Russ).]

4. Николаев В.М., Алексеев Р.З., Федорова С.А. Интенсивность свободнорадикального окисления липидов в организме больных холодовой травмой. *Якутский медицинский журнал*. 2018; 2: 34-38. [Nikolaev VM, Alekseev RZ, Fedorova SA. Intensity of free radical oxidation of lipids in the body of patients with cold trauma. *Yakut medical journal*. 2018; 2: 34-38. (In Russ.)]

5. Соломахина Н.И., Беленков Ю.Н. Прогностическое значение тканевого ингибитора матриксных металлопротеиназ - 1 (TIMP-1) у больных ХСН. *Сердечная недостаточность*. 2010; 61(5): 281-284. [Solomakhina NI., Belenkov YuN. Prognostic value of a tissue inhibitor of matrix metalloproteinases-1 (TIMP-1) in patients with CHF. *Heart failure*. 2010; 61(5): 281-284. (In Russ).]

6. Туманский В. А., Христенко Т. А. Иммуногистохимическое исследование ММП-2, ТИМП-1 при гиперпластических полипах и аденокарциноме желудка кишечного типа. *Морфология*. 2017; 11(3): 21-28. [Tumanskiy VA, Khrystenko TA. Immunohistochemical study of MMP-2, TIMP-1 in hyperplastic polyps and intestinal-type gastric adenocarcinoma. *Morphologia.* 2017;11(3): 21-28. (In Russ).]

7. Целуйко С.С., Заболотских Т.В., Дударикова С.А., Красавина Н.П., Корнеева Л.С. Действие холода на организм. Криопротекторы и средства противоишемической защиты тканей. *Якутский медицинский журнал.* 2018; 2(62): 48-55. [Tseluyko SS, Zabolotskikh TV, Dudnikov SA, Krasavina NP, Korneeva LS. The Effect of cold on the body. Cryoprotectants and means of anti-ischemic protection of fabrics. *Yakut medical journal*. 2018; 2(62): 48-55. (In Russ).]

8. Шаповалов К.Г. Отморожения в практике врача анестезиолога-реаниматолога. *Вестник анестезиологии и реаниматологии.* 2019; 16(1): 63-68. https://doi.org/10.21292/2078-5658-2019-16-1-63-68. [Shapovalov K.G. Frostbites in the practice of an anesthesiologist and emergency physician. *Messenger of anesthesiology and resuscitation.* 2019;16(1): 63-68. https://doi.org/10.21292/2078-5658-2019-16-1-63-68. (In Russ).]

9. Шаповалов К.Г., Томина Е.А., Михайличенко М.И., Иванов В.А., Витковский Ю.А. Содержание цитокинов в крови больных при местной холодовой травме. *Медицинская иммунология.* 2008; 10(1): 89-92. [Shapovalov KG, Tomina EA, Mikhailichenko MI, Ivanov VA, Vitkovsky YA. The content of cytokines in the blood of patients with local cold injury. *Medical immunology*. 2008;10(1): 89-92. (In Russ).]

10. Юлдашева Д.Ю. Роль матриксных металлопротеиназ и их ингибиторов при развитии гиперплазии эндометрия. *Фундаментальные исследования*. 2015; 1(4): 845-847. [Yuldasheva DY. The Role of matrix metalloproteinases and their inhibitors in the development of endometrial hyperplasia. *Fundamental study*. 2015; 1(4): 845-847. (In Russ.) URL: http://fundamental-research.ru/ru/article/view?id=37434 (дата обращения: 17.01.2020)].

11. Blankenberg S., Rupprecht HJ, Poirier O. Plasma concentrations and genetic variation of matrix metalloproteinase-9 and prognosis of patients with cardiovascular disease. *Circulation.* 2003; 107(12): 1579-1585.

12. Gilabert Estelle´s J. Expression of angiogenic factors in endometriosis: relationship to fibrinolytic and metalloproteinase systems. *Human Reproduction.* 2007; 22(8): 2120-2127.

13. Kingma CF, Hofman II, Daanen HM. Relation between finger cold-induced vasodilation and rewarming speed after cold exposure. *Europian Physiology*. 2019; 119(1): 171-180. DOI: 10.1007.

14. Lopez-Rivera E. Matrix metalloproteinase 13 mediates nitric oxide activation of endothelial cell migration. *PNAS*. 2005; 102(10): 3685-3690. DOI: 10.1093/humrep/dem149.

15. Miao MS, Xiang LL, Bai M, Cao LH. Frostbite animal model preparation specification (draft). *Zhongguo Zhong Yao Za Zhi*. 2018; 43(2): 410-414. DOI: 10.19540.

16. Pino M. Association between MMP1 and MMP9 activities and ICAM1 cleavage induced by tumor necrosis factor in stromal cell cultures from eutopic endometria of women with endometriosis. *Reproduction*. 2009; 138 (5): 837-847.

17. Pan Y, Thapa D, Baldissera LJ, Argunhan F, Aubdool AA, Brain SD. Elevance of TRPA1 and TRPM8 channels as vascular sensors of cold in the cutaneous microvasculature. *Pflugers Arch*. 2018; 470(5): 779-786. DOI: 10.1007.

18. Zhang H. Role of Matrix Metalloproteinases and Therapeutic Benefits of Their Inhibition in Spinal Cord Injury. *Neurotherapeutics*. 2011; 8: 206 – 220. DOI: 10.1007/s13311-011-0038-0.

19. Cauchy E, Davis CB, Pasquier M, Meyer EF, Hackett PH. A New Proposal for Management of Severe Frostbite in the Austere Environment. *Wilderness Environ Med*. 2016; 27(1): 92-99. DOI: 10.1016.