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Original article

Blood levels of hormones, cytokines and leukocyte content versus hypertrophic scar laser treatment outcome

Lyudmila S. Vasilyeva, Maksim V. Kobets, Olga A. Makarova

Irkutsk State Medical University of the Ministry of Healthcare of the Russian Federation

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Abstract: *The goal* was to identify the changes in blood levels of hormones, cytokines, and the number of leukocytes associated with the success of laser treatment of hypertrophic scars.
Material and Methods — The lipid, hormonal, cytokine and leukocyte composition of blood was studied in 15 women with normotrophic scars (Group 1) and 30 women with hypertrophic scars (Group 2). Blood was taken before treatment on days 5-7 of the menstrual cycle, followed by laser treatment. The clinical parameters of scars were assessed before treatment and 3 months after it, and two subgroups were identified: with a successful treatment outcome (2a) and with an unsuccessful outcome (2b). A retrospective analysis of blood composition was performed in each subgroup. The data were processed using the methods of nonparametric statistics. The differences were considered statistically significant at $p<0.05$.
Results — At a successful treatment outcome, the clinical parameters of scars were associated with low estradiol level, high progesterone content and high number of segmented neutrophils. These changes create conditions for scar hypertrophy, but retain the body's capability of responding to the treatment by inflammatory process with normotrophic scarring. At an unsuccessful treatment outcome, the scar hypertrophy was restored under conditions of low blood content of luteinizing hormone, and high levels of growth hormone and transforming growth factor β.
Conclusion — Features of changes in the blood levels of hormones, cytokines, and leukocyte content are associated with the success of laser treatment of hypertrophic scars.

Keywords: hypertrophic scars, systemic factors, hormones, cytokines, number of neutrophils.

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Correspondence to Maksim V. Kobets. Address: 1 Krasnogo Vosstaniya St., Irkutsk 664003, Russia. Phone: +7 9025108405. E-mail: kobetsmv@gmail.com.

Introduction

The choice of an adequate method of hypertrophic scar treatment still causes difficulty due to inability to unambiguously predict its result. Sometimes, to achieve a satisfactory result, the doctor has to use two or three treatment methods. In this regard, the problem of hypertrophic scar treatment remains relevant, and the main task for solving this problem is to reveal the cause of varying efficacy of scar treatment. The answer to this question could be given by the comprehensive knowledge on the etiology and pathogenesis of scar tissue hypertrophy.

By now, the mechanisms of wound healing autoregulation were identified [1], and histochemical and biochemical characteristics of normal and pathological scar tissue were extensively studied. Many molecular mechanisms of hypertrophic scarring have been clarified, and the key role in this regard belongs to mechanotransduction [2] and imbalance of matrix metalloproteases; to expression of cytokines IFN-γ, IL1, IL6, IL10, TNF-α, along with growth factors TGF-β, IGF and other mediators of scar tissue [3, 4].

According to some authors, such studies should be continued to clarify the impact of systemic factors on pathological scarring [5, 6, et al.]. In general terms, systemic factors include immune and hormonal factors; as well as blood cells circulating between blood and tissues and regulating metabolism, vascular and cellular responses to injury, wound fibrosis, and scar remodeling. These authors believe that an assessment of the wound environment in a particular patient (proinflammatory or anti-inflammatory) could provide valuable information for a better understanding of the etiology and pathogenesis of hypertrophic scars. Accordingly, on the hypertrophic scarring model of a wound on a rabbit's ear, it was shown that the balance between proangiogenic and antiangiogenic factors shifts towards an increased level of vascular endothelial growth factor [7] against the background of a low level of endostatin [8] in the blood serum. Data on the blood level of TGF-β1 in hypertrophic scars are scarce. Rorison P. et al. established that the level of this cytokine in the blood of children with burn wounds increased after injury with normotrophic scarring, but did not change with the formation of hypertrophic scars [9]. Bayat A. et al. did not find any differences between the

systemic levels of TGF-β1 in healthy individuals vs. patients with keloid and hypertrophic scars [10]. The role of systemic levels of sex hormones in pathological scarring is discussed mainly in relation to keloids, although there is evidence of a decrease in hypertrophy of postoperative scars by 40% with the use of estrogen receptor blocking tamoxifen [11].

Therefore, the evidence of the effect of systemic factors on hypertrophic scarring is still somewhat limited: it varies and does not give the general picture of what is happening. Nevertheless, the presented literature data demonstrate the interest of researchers not only in the morphofunctional characteristics of pathological scar tissue, but also in changes in the concentrations of systemic factors in keloid and hypertrophic skin scars. The development of this line of research could provide new information about the etiology and pathogenesis of pathological scars and explain the reasons for the different treatment efficacy. In addition, such studies provide an opportunity to identify prognostic biomarkers when choosing an adequate treatment and ways to improve its efficacy, as well as to develop measures to prevent pathological scarring.

The goal of our study was to identify the changes in blood serum levels of hormones, cytokines, and the number of leukocytes associated with the success of laser treatment of hypertrophic scars.

Material and Methods

Study subjects

Our study included 45 women with skin scars, specifically: 15 with normotrophic scars representing Group 1 (control group) and 30 with hypertrophic scars representing Group 2 (study group). A written informed consent was obtained from all subjects. The inclusion criteria were as follows: female, age range 18–35 years, normotrophic and hypertrophic scars (the formation period of 2–5 years), localized in areas with thick slightly stretched skin experiencing significant functional loads (chest, back, upper limbs). The exclusion criteria: presence of keloid scar, diabetes mellitus and other endocrine pathology, malignant tumors, and infectious diseases. The formed groups were comparable in age [28 (24, 30) years and 25 (20, 31) years, p=0.3] and scar formation period [2 (2, 3) years and 2 (2, 4), respectively, p=0.1].

Study design

A controlled, non-randomized retrospective study was conducted in compliance with the standards of Good Clinical Practice and the Declaration of Helsinki principles. After diagnosing the scar type, all study subjects underwent the assessment of scar clinical indicators and blood composition parameters, followed by the laser treatment of hypertrophic scars with its efficacy evaluated later (after three months). All subjects of the study group were retrospectively distributed among two subgroups (with a successful vs. unsuccessful treatment outcome), and blood composition of patients with hypertrophic scars was retrospectively analyzed against the background of the control group. For each patient, the study duration was three months.

Clinical assessment of scars

The Vancouver Scar Scale [12] was used to evaluate five clinical scar indicators (in number of points, *Table* 1). Vascularity of the scar was visually determined (normal color = 0 points, pink = 1 point, red = 2 points, purple = 3 points), as well as scar pigmentation (normal = 0, hypopigmentation = 1, hyperpigmentation = 2). The scar pliability was assessed by palpation (0 – normal = 0, supple = 1, yielding = 2, firm = 3, adherent = 4). Itching was characterized by subjective sensations (none = 0, mild = 1, moderate = 2, severe = 3). The scar height (h_{mm}) was measured in mm using CASTROVIEJO CALIPERS (COMPASS) (with a step of 0.1 mm), followed by the conversion into points (flat = 0, <2 mm = 1 point, 2-5 mm =2 points, >5 mm = 3 points). An integral clinical scar assessment was performed according to the Total Scar Index (TSI), which is the sum of the scores of the scale indicators.

Table 1. Clinical characteristics of hypertrophic scars before and after laser treatment

Clinical indicators (points) / groups	*The control: Group 1 (n=15)*	*Hypertrophic scars (Group 2, n=30)*					
		Subgroup 2a (n=15)			Subgroup 2b (n=15)		
		Before treatment	After treatment	U criterion, p	Before treatment	After treatment	U criterion, p
Vascularity	0 (0,0)	0 (0,0)	1 (0, 1)	p_1=0.005 p_2=0.008	0.5 (0,1)	1 (1, 1)	p_1<0.001 p_2=0.001
Pigmentation	1 (1, 1)	1 (1, 2)	0 (0, 1)	p_1=0.017 p_2<0.001	1 (1, 2)	1 (1, 2)	p_1=0.005 –
Pliability	1 (1, 1)	1.5 (1, 2)	1 (1, 1)	– p_2=0.003	2 (1, 2)	2 (1, 2)	p_1<0.001 –
Height	0 (0, 0)	1 (1, 1)	0.5 (0, 1)	p_1=0.002 p_2=0.002	1 (1, 1)	1,5 (1, 2)	p_1<0.001 p_2=0.021
Itching	0 (0, 0)	0 (0, 0)	0 (0, 0)	– –	0 (0, 1)	1 (0, 1)	p_1<0.001 –
TSI	2 (2, 2)	4 (3, 5)	2 (2, 3)	p_1=0.036 p_2=0.003	5.5 (4, 6)	6.5 (5, 7)	p_1<0.001 p_2=0.029
h_{mm}, mm	0 (0, 0)	1,5 (1, 2)	0.75 (0, 1.5)	p_1<0.001 p_2=0.032	2 (1.5, 2)	2.3 (1.7, 3)	p_1<0.001 p_2=0.048

The results are presented as a median with its lower and upper quartiles – Me (LQ, UQ). p_1-values reflect the statistical significance of differences between the studied parameters of the scar after treatment in the particular subgroup and the control group; p_2-values reflect the statistical significance of differences between the studied parameters of the scar after treatment and before treatment (in the same subgroup). Consistency of grouping = 92.8% (Wilks' lambda = 0.153, p<0.001). TSI, Total Scar Index; h_{mm}, scar height.

Table 2. Examined blood parameters in patients with hypertrophic and normotrophic scars

Parameters/ groups	*Reference values*	*The control: Group 1 (n=15)*	*Hypertrophic scars (Group 2, n=30)*		*U criterion, p<0.05*
			2a (n=15)	2b (n=15)	
Complete blood count					
Segmented neutrophils, $\times 10^9$/L	1.8-6.3	2.9 (2.3, 3.3)	3.8 (3.0, 4.6)	3.0 (2.6, 3.2)	$p_{1\text{-}2a}$=0.024 –
Hormonal profile					
LH, mIU/mL	2.4-12.6	4,7 (3.1, 6.0)	5.0 (4.6, 5.2)	3.2 (2.4, 3.7)	– $p_{1\text{-}2b}$=0.047
GH, ng/mL	0-8	0.8 (0.4, 1.4)	0.8 (0.3, 0.9)	2.7 (1.4, 3.0)	– $p_{1\text{-}2b}$<0.001
FSH, mIU/mL	3.5-12.5	5.1 (3.8, 5.7)	6.3 (6.2, 6.4)	5.0 (4.3, 5.3)	$p_{1\text{-}2a}$=0.002 –
Estradiol, nmol/L	0.01-0.6	0.3 (0.3, 0.5)	0.3 (0.2, 0.3)	0.3 (0.3, 0.4)	$p_{1\text{-}2a}$=0.012 –
Progesterone, nmol/L	1,07-5,27	4.2 (2.6, 5.2)	6.1 (5.9, 6.2)	3.4 (2.3, 4.5)	$p_{1\text{-}2a}$<0.001 –
Testosterone, nmol/L	0.35-2.6	2.2 (1.6, 2.8)	1.5 (1.3, 1.6)	1.4 (1.0, 1.7)	$p_{1\text{-}2a}$=0.016 $p_{1\text{-}2b}$=0.018
Cortisol, nmol/L	185-624	543.8 (433.0, 661.1)	418.8 (367.4, 482.1)	315.4 (240.0, 330.0)	$p_{1\text{-}2a}$=0.031 $p_{1\text{-}2b}$=0.001
Immune profile					
TNF-α, pg/mL	0-6	2.3 (1.5, 2.6)	1.8 (1.8, 2.3)	1.6 (0.01, 1.7)	– $p_{1\text{-}2b}$=0.007
TGF-β_1, ng/mL	0-37.7	30.9 (24.1, 39.1)	34.8 (29.8, 35.0)	45.2 (32.4, 58.6)	– $p_{1\text{-}2b}$=0.043

The results are presented as a median with lower and upper quartiles – Me (LQ, UQ). p-values reflect the statistical significance of differences in the studied parameters of blood between the control group (1) and the studied subgroup (2a or 2b).

Table 3. Results of discriminant analysis of blood parameters in studied subgroups and control group

Groups 1 and 2a *Wilks' lambda: 0.26541, F (.,25) =17.299, p<0.001* *Consistency of grouping 96.7%*			*Groups 1 and 2b* *Wilks' lambda: 0.41033, F (3.26) =12.454, p<0.001* *Consistency of grouping =83.33%*		
Blood parameters	Fisher's F statistic	p-value	Blood parameters	Fisher's F statistic	p-value
Progesterone	24.30	0.001	Cortisol	10.73	0.003
Estradiol	10.84	0.003	TGF-β_1	10.64	0.003
Testosterone	7.03	0.014	GH	6.25	0.019
Segmented neutrophils	5.46	0.028			

The results are presented in the form of F statistic, the value of which is indicative of a degree of participation of blood parameters in the discrimination of subgroups (the larger the F, the more informative the parameter). p-value reflects the statistical significance of the F statistic. Wilks' lambda evaluates the accuracy of discrimination (0 stands for absolutely accurate classification, 1 signifies absolutely erroneous classification).

Blood tests

Blood samples were taken on days 5–7 of the menstrual cycle (follicular phase). Twenty-six parameters were identified in blood and serum to characterize the leukocyte composition, along with the lipid, hormonal, and immune profiles. The units of measurement for each parameter and reference values (for sex hormones: in the follicular phase of the cycle) are presented in *Table* 2. The leukocyte composition was determined on the automated hematology analyzer Mindray BC-5150 (China). The percentage of leukocyte types with differentiation for banded neutrophils and segmented neutrophils was calculated in the blood smear, and then it was converted into absolute number. The concentrations of cholesterol, triglycerides, HDL, LDL, and VLDL in the lipid profile were determined by the dry chemistry method on Reflotron IV express blood analyzer (Reflotron test strips, La Roche, Germany). The method of enzyme-linked immunosorbent assay (ELISA) was employed to assess the hormonal and immune profiles. Using Alcor Bio test systems (St. Petersburg), we determined the concentrations of thyroid-stimulating hormone (TSH), luteinizing hormone (LH), follicle-stimulating hormone (FSH), free thyroxine (free T4), prolactin, growth hormone (GH), cortisol, testosterone, progesterone, and estradiol in the hormonal profile. The concentrations of tumor necrosis factor (TNF-α), interleukins (IL4, IL6, IL10), and transforming growth factor-β (TGF-β) were also assessed in the immune profile (Vector-Best test systems, Novosibirsk).

Treatment method

Laser treatment of hypertrophic scars was performed using Lancet–2 laser system (Russia). Under local infiltration anesthesia with 2% lidocaine solution, the scar tissue was subjected to multiple microperforations with a density of 36 per cm^2 to the depth of the scar height. The perforations were made with a focused laser beam (d=0.5 mm, λ=10.6 nm) in a superpulse mode (50 W/0.05 sec., total absorbed energy 53 J/mm^2). After imposing perforations, the scar surface was treated with 10% aqueous solution of povidone-iodine twice a day for 3-4 days. Healing continued under the eschar and did not require additional medical attention as there were no complications. The scarring process was monitored monthly. After 3 months, the clinical parameters of the scar were assessed using the Vancouver Scar Scale.

Subgroup formation

The change in the clinical parameters of scars after treatment served as the basis for dividing the study group (Group 2) into two subgroups: with a successful outcome of the treatment (subgroup 2a) and unsuccessful outcome (subgroup 2b). The formed subgroups were comparable in terms of their size (15 patients in each), age of patients (p=0.8), and period of scar formation (p=0.8).

In accordance with the study goal, the initial blood parameters (before the treatment) in the formed subgroups with hypertrophic scars were retrospectively analyzed in comparison with the control group.

Statistical data processing

We used Statistica 10.0 software package (StatSoft Inc., USA, 2010) to analyze the data. The sample size was not originally calculated, but the formed subgroups were standardized by number. Consistency of grouping was confirmed by discriminant analysis using the Wilks' lambda statistic. The type of distribution of the variation series was determined using the Shapiro-Wilk test. The results are presented as a median with its lower and upper quartiles Me (LQ; UQ). The parameters of the study groups were compared with the control group using the Mann-Whitney U test, statistical significance was assumed at $p \leq 0.05$. To identify the most informative parameters that distinguish the subgroups from the control group, a pairwise discriminant analysis was performed with stepwise inclusion of variables. The association of indicators was calculated by the method of multiple regression analysis, the correspondence of the regression model to obtained data was considered high at the value of the coefficient of determination, $R^2>0.7$.

Results

The clinical characteristics of hypertrophic scars after laser treatment were compared with the values before treatment (p_2) and against the control group (p_1) (*Table* 1).

All patients exhibited an increase in vascularity (hyperemia) of hypertrophic scars three months after laser treatment. The successful treatment outcome (subgroup 2a) was manifested in scar pliability reduction, as well as in a decrease of the scar height, pigmentation and TSI, and approximation of these indicators to normal values. The unsuccessful treatment outcome foir hypertrophic scars (subgroup 2b) was characterized by the absence of changes in the clinical characteristics of the scar and even an increase in the scar height and TSI. It should be noted that before treatment, hypertrophic scars in subgroups 2a and 2b differed solely in the vascularity degree (p=0.016), scar height (h_{mm}, p=0.021) and TSI (p=0.027), and the values were higher in subgroup 2b.

A retrospective analysis of blood test results, taken before treatment in patients of both subgroups, revealed statistically significant difference from the control group (*Table* 2). Both subgroups were characterized by a reduced concentration of cortisol and testosterone, but alternative trends were identified for other parameters. E.g., in subgroup 2a, a lower concentration of estradiol, and higher levels of progesterone and FSH, as well as higher numbers of segmented neutrophils, were detected. In subgroup 2b, a reduced concentration of LH and TNF-α, along with an increased concentration of GH and TGF-β, were registered. It is worth noting that the differences in mentioned indicators between study subgroups and the control was confirmed statistically (*Table* 2), albeit nearly all of them remained within the reference range of values. Increased beyond the reference range values were established in subgroup 2a solely for the concentration of progesterone (by 10%), and in subgroup 2b for the concentration of TGF-β (by 20%).

Pairwise discriminant analysis revealed a different set of the most informative indicators that distinguished each subgroup with hypertrophic scars from the control group (*Table* 3), and confirmed the consistency of grouping. According to presented data, subgroup 2a differed from the control group in terms of the level of sex hormones and segmented neutrophils in the blood, while the most significant change was the high concentration of progesterone (F statistic = 24.3, p<0.001), exceeding the reference range. Distinctive features of subgroup 2b, equivalent in terms of informativeness (the value of F statistic), were changes in the level of cortisol, TGF-β, and GH.

The method of multiple regression analysis revealed a relationship between clinical indicators of scars before laser treatment (dependent variables TSI and hmm) and some blood parameters (independent variables), which were statistically significantly different from their values in the control group. The method of multiple regression analysis revealed the association between the values of scar clinical indicators before the laser treatment (as dependent variables of TSI and h_{mm}) and some blood parameters (as independent variables), which statistically significantly differed from their values in the control group. Multiple regression equations were obtained for each subgroup:

TSI (2a) = +0.4×Progesterone+0.4×Segmented neutrophils (R^2=0.85, p<0.001),

h_{mm} (2a) = +0.4×Progesterone–3.4×Estradiol (R^2=0.73, p<0.001);

TSI (2b) = +0.8×GH+0.00005×TGF-β (R^2=0.87, p<0.001),

h_{mm} (2b) = +0.4×GH+0.00002×TGF-β–0.1×LH (R^2=0.74, p<0.001).

According to these equations, in subgroup 2a, TSI and h_{mm} directly correlated with the concentration of progesterone. In addition, TSI exhibited a direct dependence on the number of segmented neutrophils, while h_{mm} had an inverse dependence on the estradiol concentration. In subgroup 2b, both dependent indicators (TSI and h_{mm}) displayed a direct correlation with the concentration of GH and TGF-β; besides, h_{mm} had an inverse correlation with LH concentration.

The presented results reflect the association of clinical parameters of hypertrophic scars with the quantitative content of certain hormones, cytokines, and leukocytes in blood, and their combinations are different under successful and unsuccessful outcomes of laser treatment.

Discussion

Accumulated knowledge on inflammatory response of connective tissue indicates that changes in the number of leukocytes, hormone content, and cytokine level in blood could create unfavorable conditions for wound healing, modulate the mechanisms of inflammation autoregulation, and disrupt the process of the intercellular matrix remodeling resulting in pathological scarring.

Based on collected data, unidirectional changes in the hormonal background of both subgroups (more pronounced in subgroup 2b) were detected with hypertrophic scars (*Table* 2). They were represented by reduced levels of cortisol and testosterone. It is known that the effects of cortisol are dose-dependent [13], and its reduced concentration can be achieved by the effects of low doses of this hormone, which are expressed in maintaining (or even stimulating) the processes of proliferation and fibrosis, characteristic of hypertrophic scar tissue. In turn, a reduced concentration of testosterone can significantly weaken its main effects – anti-inflammatory and anabolic [14]. These facts imply that the reduced levels of cortisol and testosterone that we have identified contribute to an increase of proinflammatory effects, which is important in hypertrophic scarring. This assumption is confirmed by the results of discriminant analysis, which demonstrated that the reduced cortisol concentration in subgroup 2b makes a significant contribution (F statistic, *Table* 3) to creating the conditions for scar tissue hypertrophy.

The analysis of the revealed features in the blood composition of the studied subgroups allowed explaining the different results of hypertrophic scar laser treatment. In subgroup 2a, with a successful outcome of treatment, first of all, a high level of progesterone (uncharacteristic of the sexual cycle follicular phase) attracts attention (Table 2). This hormone, according to published sources, has an anti-inflammatory and immunosuppressive effects [15]. In high doses, it stimulates the proliferation of keratinocytes and suppresses the degradation of collagen in the course of scar remodeling via reducing the activity of matrix metalloproteases in fibroblasts [16]. Taking into account these progesterone effects, its high concentration in the blood may be crucial for the formation of hypertrophic scars in patients of subgroup 2a. This conclusion is confirmed by the results of discriminant analysis (F statistic, Table 3) and a direct association of clinical scar indicators in this subgroup with the level of progesterone (multiple regression equations). In addition to progesterone, in subgroup 2a, the reduced concentration of estradiol affected the scar height, which could be manifested, according to the literature, in decelerating wound healing due to the weakening of the production of growth factors by skin cells (TGF-β1 and fibroblast growth factor by fibroblasts; granulocyte-macrophage colony-stimulating factor by keratinocytes; nerve growth factor by macrophages) [17]. Accordingly, it could be assumed that a reduced level of estradiol is also involved in creating conditions beneficial to hypertrophy of scar tissue in the patients of subgroup 2a. The increased number of segmented neutrophils in this subgroup is important for TSI, which may imply a prolongation of the inflammatory leukocyte response that slows down the scar tissue maturation [18]. Thus, there are two possible variants of changes in the body reactivity that affect the processes of scar tissue hypertrophy in patients of subgroup 2a: the preservation of the activity of the leukocyte inflammation link and the consequences of an imbalance of steroid hormones with progesterone predominance and estrogen deficiency. These features create conditions for the emergence and existence of microfoci of destruction, proliferation and fibrosis, characteristic of hypertrophic scar tissue.

On the other hand, an increased number of segmented neutrophils, hypothetically, could be one of the conditions contributing to successful outcome of laser treatment. It is well known that neutrophilic leukocytosis reflects the readiness of the body to carry out an inflammatory leukocyte response, including in new microfoci of inflammation created by the laser. Under these conditions, a timely and adequate leukocyte response (in accordance with the mechanisms of autoregulation of inflammation) is able to restart the mediator cascade in the scar tissue, which can rebuild the inflammatory process according to the normergic type and complete it with normotrophic scarring. The associations between the clinical characteristics of hypertrophic scars, blood composition, and the successful outcome of laser treatment, which we discovered in subgroup 2a, supported our assumption.

Subgroup 2b with unsuccessful treatment outcome is characterized by other changes in blood composition: reduced concentrations of LH and TNF-α, and increased concentrations of GH and TGF-β (*Table* 2). According to published sources, such changes in these indicators are closely interrelated and could, together or separately, contribute to hypertrophy of scar tissue. GH stimulates the synthesis of proteins, including collagen and insulin-like growth factor, which activates fibroblast proliferation [19]. Besides, GH is able to stimulate the production of TGF-β1 via alternative signaling pathways [20], which enhances the synthesis of types I and III collagen by fibroblasts in the scar and activates the transformation of fibroblasts into myofibroblasts. High content of TGF-β1 could cause a reduction in the level of cortisol and the proinflammatory cytokine TNF-α [21].

Discriminant analysis also confirmed these correlations (Table 3), and regression analysis revealed a direct dependence of the clinical characteristics of hypertrophic scars (TSI and h_{mm}) in patients of subgroup 2b on serum concentrations of GH and TGF-β. Consequently, the stimulation of fibroproliferative processes with high doses of GH and TGF-β can be considered a feature of the body reactivity in subgroup 2b. In addition to these factors, scar height (h_{mm}) is inversely related to LH concentration, which may affect the process of scar remodeling. Given that LH activates the production of prostaglandins, eicosanoids, and matrix metalloproteases [16], which increase vascular permeability, proteolysis, and loosening of connective tissue, a decrease in the concentration of this hormone could lead to dehydration and thickening of scar tissue.

According to presented data and the analysis of publications, the unsuccessful outcome of laser treatment in patients of subgroup 2b is probably related to the nature of changes in blood composition. These changes, as explained above, are stabilized due to mutual influence on each other, thereby creating ongoing conditions for pathological scarring. For this reason, after laser treatment, the hypertrophic scar is restored again.

The presented results are limited to female patients, using only one treatment and small sample size; nevertheless, they demonstrate the relationship of scar tissue hypertrophy in women with changes in the number of leukocytes and levels of hormones and cytokines. These changes are not the same in the studied subgroups and may contribute or hinder the success of laser treatment of hypertrophic scars. Further studies to identify the features of systemic factors, associated with efficacy of alternative methods of treating hypertrophic scars, may provide essential information for the development of targeted therapy for this pathology.

Conclusion

We discovered that the outcome of hypertrophic scar laser treatment was associated with the nature of changes in blood composition prior to the treatment. Positive dynamics of scar

clinical parameters was observed in patients with low serum estradiol level, high progesterone level and an increased number of segmented neutrophils. Unsuccessful treatment outcome was observed in case of low serum level of LH, and high levels of GH and TGF-β.

Ethical approval

The protocol of the study was approved by the Ethics Committee at Irkutsk State Medical University of the Russian Federation Ministry of Healthcare (Protocol No. 936/18 of May 24, 2019) and the Medical Commission of the Baikal Center for Multidisciplinary Medicine LLC (Protocol No. 6 of June 6, 2019).

Conflict of interest

The authors declare that they have no conflicts of interest.

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Authors:
Lyudmila S. Vasilyeva – DSc, Professor, Chair of the Department of Histology, Embryology, Cytology, Irkutsk State Medical University, Irkutsk, Russia. http://orcid.org/0000-0003-3495-1560.
Maksim V. Kobets – graduate student, Department of Pathophysiology, Irkutsk State Medical University, Irkutsk, Russia. http://orcid.org/0000-0002-7759-1078.
Olga A. Makarova – PhD, Associate Professor, Department of Histology, Embryology, Cytology, Irkutsk State Medical University, Irkutsk, Russia. http://orcid.org/0000-0002-4093-2502.