Flyunt Viktor R, Flyunt Igor-Severyn S, Fil' Vitalij M, Kovbasnyuk Marta M, Hryvnak Rostyslav F, Popel Svitlana L, Zukow Walery. Relationships between caused by drinking of bioactive water Naftussya changes in urine lithogenicity and neuro-humoral-immune factors in humans with theirs abnormalities. Journal of Education, Health and Sport. 2017;7(3):11-30. eISSN 2391-8306. DOI http://dx.doi.org/10.5281/zenodo.261918
http://ojs.ukw.edu.pl/index.php/johs/article/view/4203

The journal has had 7 points in Ministry of Science and Higher Education parametric evaluation. Part B item 754 (09.12.2016).
754 Journal of Education, Health and Sport eISSN 2391-8306 7

The authors declare that there is no conflict of interests regarding the publication of this paper.
Received: 20.01.2017. Revised 21.01.2017. Accepted: 26.01.2017.

RELATIONSHIPS BETWEEN CAUSED BY DRINKING OF BIOACTIVE WATER NAFTUSSYA CHANGES IN URINE LITHOGENICITY AND NEURO-HUMORAL-IMMUNE FACTORS IN HUMANS WITH THEIRS ABNORMALITIES

Viktor R Flyunt[1,2], Igor-Severyn S Flyunt[3], Vitalij M Fil'[3], Marta M Kovbasnyuk[2], Rostyslav F Hryvnak[4], Svitlana L Popel[4], Walery Zukow[5]

[1]JSC "Dnipro-Beskyd", Truskavets', Ukraine fliunt66@mail.ru
[2]Department of Immunophysiology, OO Bohomolets' Institute of Physiology, Kyïv, Ukraine marta.kovbasnyuk@yandex.ua
[3]Department of Anatomy, Physiology and Valeology, Ivan Franko Pedagogical University, Drohobych, Ukraine iflyunt@bk.ru fillvitalij@rambler.ru
[4]JSC " Truskavets'kurort", Truskavets', Ukraine
[5]Faculty of Physical Education, Health and Tourism, Kazimierz Wielki University, Bydgoszcz, Poland w.zukow@ukw.edu.pl

SUMMARY

Objective. Spa Truskavets' (Ukraine) considered to be indicated for the treatment and metaphylaxis renal stone disease. However, data on the effect of balneotherapy on the parameters of urine Lithogenicity ambiguous. This is due, perhaps, ambiguous influence balneotherapy on the neuroendocrine factors regulating exchange of electrolytes and uric acid. **Aim:** to find out the influence drinking of bioactive water Naftussya on urine lithogenicity and neuro-humoral-immune factors in humans with theirs abnormalities. **Methods.** The object of observation were ten women and ten men aged 33-76 years without clinical diagnose but with dysfunction of neuro-endocrine-immune complex and metabolism. In daily urine and blood we determined the content of electrolytes and nitrogenous metabolites, estimated parameters of immunity, recorded conductivity of acupuncture points and heart rate variability (HRV). After examination volunteers within 7 days used bioactive water Naftussya (250 ml three times a day), then repeated the tests listed. **Results.** Lithogenicity urine Index (Lith) calculated by formula: $Lith=(Uric\ acid \bullet Calcium/Magnesium \bullet Creatinine)^{0,25}$. In 3 people initial normal Lith (0,65÷0,81 units) increased by 0,08÷0,16 units. In 4 people with normal or high Lith its changes were not detected (-0,01÷+0,01). In 12 people from a wide range of primary Lith (0,62÷1,07) it lowered by 0,03÷0,12 un. Even in a man maximum level of Lith (1,45 un.) down to the upper limit of normal (0,84 un.). Found a strong correlation between changes in Lith and a number parameters of HRV, metabolism and immunity. **Conclusion.** The use of bioactive water Naftussya causes ambiguous

changes in Lithogenicity of urine, related to ambiguous changes in HRV, metabolism and immunity.

Keywords: lithogenicity of urine, HRV, phagocytose, immunity, bioactive water Naftussya, spa Truskavets'.

INTRODUCTION

Spa Truskavets' (Ukraine) considered to be indicated for the treatment and metaphylaxis renal stone disease [1,31,37-39,41]. Recently we found that the Ca/Mg ratio of urine as marker its Lithogenicity increased in 29,3% patients of spa Truskavets' with chronic pyelonephrite, some more in 63,2% persons it is on the upper limit of normal or somewhat exceeds it [16]. However it has long been known that balneotherapy on spa Truskavets' did not significantly affect the Ca/Mg ratio of urina (Means±SE: before 1,9±0,2, after 1,8±0,2) [38]. In another observation of 129 patients with oxalate urolithiasis, divided into 4 groups, showed a reduction (100•Ca•Oxalates)/(Mg•Creatinine) ratio (average rate 1,5) in 21 patients by 10,8% (from 6,4 to 5,7), in 83 patients by 16,5% (from 7,0 to 5,9), in 21 patients by 17,3% (from 6,5 to 5,4) and in 4 patients by 25,6% (from 6,1 to 4,55) [13], ie tangible reduction Lithogenicity not been made. Another marker urine Lithogenicity Uric acid/Creatinine ratio different groups of patients respond to balneotherapy ambiguous (0,61±0,09 and 0,43±0,11 in 6; 0,65±0,06 and 0,42±0,03 in 36; 0,65±0,13 and 0,23±0,03 in 15; 0,36±0,11 and 0,63±0,05 in 5 before and after balneotherapy respectively while the normal average is 0,22±0,02) [23]. Recently we found that standardized balneotherapy (drinking of bioactive water Naftussya, application of ozokerite, mineral bathes) not significantly affect $(Ca•Oxalates•Uric\ acid)/(Mg•Creatinine)^{0,2}$ ratio in patients with chronic pyelonephrite and cholecystite, even showed a trend to increase [33,36]. It is detected three types of **immediate** (after 1 h) effects caused by drinking bioactive water Naftussya on Ca/Mg ratio in 41 patients with oxalic urolithiasis: litholytic (in 22%), indefinite (in 12%) and lithogenic (in 66%). The types of effects is caused by exchanges of calciumuria but not magnesiumuria [18]. This is due, perhaps, ambiguous influence balneotherapy on the neuroendocrine factors regulating exchange of electrolytes and uric acid [3,6-8,10-12,14,15,17,19,22,24,25,27,28,30,32,34,35].

In line with the concept of neuro-endocrine-immune complex [29] we have previously analyzed the links $(Ca•Oxalates•Uric\ acid)/(Mg•Creatinine)^{0,2}$ ratio in patients with chronic pyelonephrite and cholecystite with a concentration in urine and plasma electrolytes and nitrogenous metabolites and the parameters Gall-bladder Motility, HRV, EEG and Immunity. Revealed significantly correlation urine Lithogenicity with Uric acid Plasma level and Gall-bladder postprandial Volume, Baevskiy's Stress Index, Sympatho-Vagal Balance Index HRV, δ-rhythm amplitude, α-rhythm frequency and θ-rhythm asymmetry index EEG, blood levels of $CD19^+$ B-Lymphocytes and $CD16^+$ NK-Lymphocytes. Canonical correlation between urine Lithogenicity and Neuro-Humoral-Immune factors is very strong: R=0,97 [21].

It is known that stress increases urine level of lithogenic factors [4] and decreases urine level of litholytic factors [5]. On the other hand, bioactive water Naftussya has stresslimiting effects, but not all patients [3,28,30]. Based on the above, we set a goal: to find out the influence drinking of bioactive water Naftussya on urine lithogenicity and neuro-humoral-immune factors in humans with theirs abnormalities.

MATERIAL AND RESEARCH METHODS

The object of observation were ten women and ten men aged 33-76 years without clinical diagnose but with dysfunction of neuro-endocrine-immune complex and metabolism, characteristic for premorbid (intermediate between health and illness) state. Echoscopy kidney (echocamera "Radmir") stones not found. First volunteers collected daily urine, in which we determined the content of electrolytes and nitrogenous metabolites, and then on an empty stomach in the morning they took samples of capillary and venous blood for biochemical, hormonal and immune tests, recorded conductivity of acupuncture points and heart rate variability (HRV). Among electrolytes estimated calcium (by the reaction with arsenazo III), magnesium (by the reaction with colgamite), phosphate (phosphate molibdate method), chloride (mercury rodanide method), sodium and potassium (flame photometry method). Uric acid estimated by uricase method, creatinine by Popper's method as described in the handbook [20]. Also determined plasma concentration of total cholesterol (direct method by reaction Zlatkis-Zach) and its distribution as part of α-lipoprotein (applied enzymatic method Hiller as described in the handbook [20]) as well as cortisol (by ELISA). Use analyzers "Pointe-180" ("Scientific", USA), "Reflotron" ("Boehringer Mannheim", BRD), "Tecan" (Oesterreich) and flame spectrophotometer.

Electroskin conductivity recorded in follow points of acupuncture: Pg(ND), TR(X) and MC(AVL) Right and Left (used complex "Medissa").

To assess the parameters of HRV we recorded during 7 min electrocardiogram in II lead (by hardware-software complex "CardioLab+HRV" produced "KhAI-Medica", Kharkiv, Ukraine). For further analysis the following parameters HRV were selected. Temporal parameters (Time Domain Methods): the standart deviation of all NN intervals (SDNN), the square root of the mean of the sum of the squares of differences between adjacent NN intervals (RMSSD), the percent of interval differences of successive NN intervals greater then 50 ms (pNN_{50}), triangulary index (HRV TI); heart rate (HR), moda (Mo), the amplitude of moda (AMo), variational sweep (MxDMn). Spectral parameters (Frequency Domain Methods): spectral power density (SPD) components of HRV: high-frequency (HF, range 0,4÷0,15 Hz), low-frequency (LF, range 0,15÷0,04 Hz), very low-frequency (VLF, range 0,04÷0,015 Hz) and ultra low-frequency (ULF, range 0,015÷0,003 Hz). Expectant as classical indexes: LF/HF, LFnu=100%•LF/(LF+HF) and Centralization Index (CI=(VLF+LF)/HF), Baevskiy's Stress Index (BSI=AMo/2•Mo•MxDMn) as well as Baevskiy's Activity Regulatory Systems Index (BARS) [2].

Parameters of phagocytic function of neutrophils estimated as described by SD Douglas and PG Quie [9] with our (MM Kovbasnyuk) moderately modification. To do this, 5 drops of blood immediately after collection, made in glass centrifuge tubes with 2 ml of 4% solution of sodium citrate. Blood samples were stored in a refrigerator at a temperature of 4^0C. Further samples were centrifuged (5000 rev/min for 5 min). The supernatant was removed with the help of the Pasteur's pipette. We used a fraction of leukocytes with traces of erythrocytes. The objects of phagocytosis served daily cultures of Staphylococcus aureus (ATCC N 25423 F49) as typical specimen for Gram-positive Bacteria and Escherichia coli (O55 K59) as typical representative of Gram-negative Bacteria. Both cultures obtained from Laboratory of Hydro-Geological Regime-Operational Station JSC "Truskavets'kurort". To prepare the suspension microbes did wipes with relevant shoals sterile saline, immersed tubes in boiling water for 3 seconds, cooled to room temperature. Integrity microbes controlled with the aid of a microscope. To do this, drop the suspension of microbes applied to skimmed substantive piece of glass, fixed in alcohol lamp

flame. Ready preparations stained by Papenheim, microscoped during immersion, lense h90, eyepiece x10. The test samples were prepared as follows. In Vidal's plastic tubes made in the following order of 0,05 mL of heparin, 0,05 mL of sterile saline, 0,1 mL suspension of leukocytes, 0,05 mL suspension of microbial bodies. Samples shaked and placed in thermostat at 37^0C for 30 min, shaking them with every 10 mins. Then, to stop phagocytosis, the sample was cooled under running water for 10 min. In further samples are centrifuged (5000 rev/min, for 5 min), the supernatant removed with the help of the Pasteur's pipette. From the suspension of leukocytes (with traces of red blood cells) prepared strokes, dried in air at room temperature and stained by Papenheim. Microscoped during immersion lens h90, x10 eyepiece. Take into account the following parameters of phagocytosis: activity (percentage of neutrophils, in which found microbes - Hamburger's Phagocytic Index PhI), intensity (number of microbes absorbed one phagocytes - Microbial Count MC or Right's Index) and completeness (percentage of dead microbes - Killing Index KI). Bactericidal capacity (BC) of neutrophils calculated by formula:

BC (10^9 Bact/L)=Leukocytes (10^9/L)•Neutrophils (%)•PhI (%)•MC (Bact/Phag)•KI (%)

Immune status evaluated on a set of I and II levels recommended by the WHO. For phenotyping subpopulations of lymphocytes used the methods of rosette formation with sheep erythrocytes on which adsorbed monoclonal antibodies against receptors CD3, CD4, CD8, CD22 and CD16 from company "Granum" (Kharkiv) with visualization under light microscope with immersion system. Subpopulation of T cells with receptors high affinity determined by test of "active" rosette formation. The state of humoral immunity judged by the concentration in serum of immunoglobulins classes G, A, M (ELISA, analyser "Immunochem", USA) and circulating immune complexes (with polyethylene glycol precipitation method) as described in the handbook [26]. After examination volunteers within 7 days used bioactive water Naftussya (250 mL three times a day), then repeated the tests listed. Normal values for surveyed contingent (including age and gender) were obtained from the database of Truskavets' Scientific School of Balneology.

Results processed by methods of correlation and canonical analyses, using the software package "Statistica 5.5".

RESULTS AND DISCUSSION

Lithogenicity urine Index (Lith) we calculated by modified formula [40]:

$$Lith=(\text{Uric acid}\bullet\text{Calcium}/\text{Magnesium}\bullet\text{Creatinine})^{0,25}.$$

If we take as the norm range 0,65÷0,81 (Mean±2SE), it is possible to state the following. The first survey lithogenicity was within normal limits in 13 individuals, increased in 6 and one person appeared to be reduced. After a week use of bioactive water Naftussya persons with high lithogenicity remained unchanged, while lowered lithogenicity has been found in 4 and normal in 10 (Fig. 1).

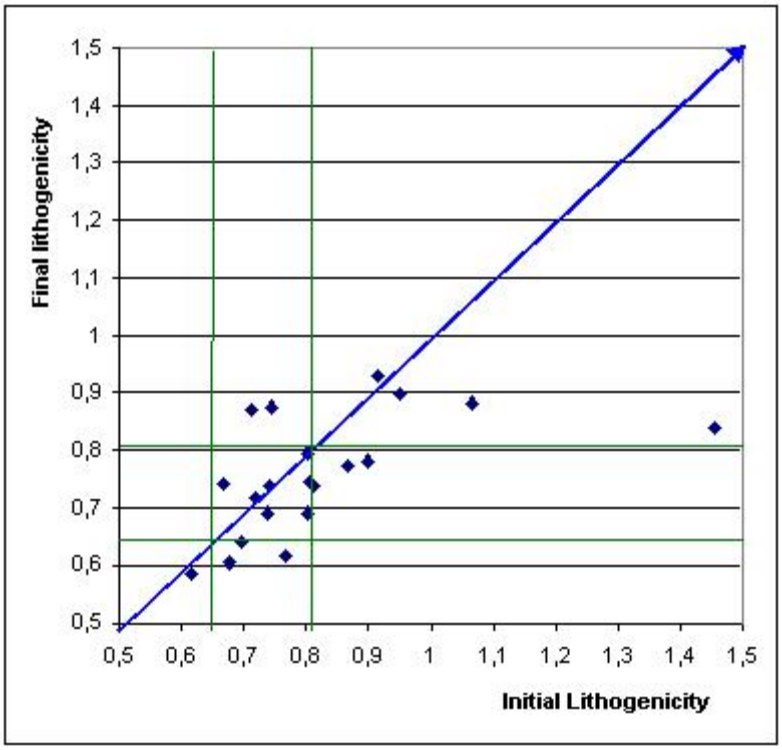


Fig. 1. Individual lithogenicity urine before (axis X) and after (axis Y) a course of drinking bioactive water Naftussya. Green lines show the limits of norm.

Analysis of individual changes in lithogenicity shows that normal levels varied in two ways, while elevated levels declined (Fig. 2).

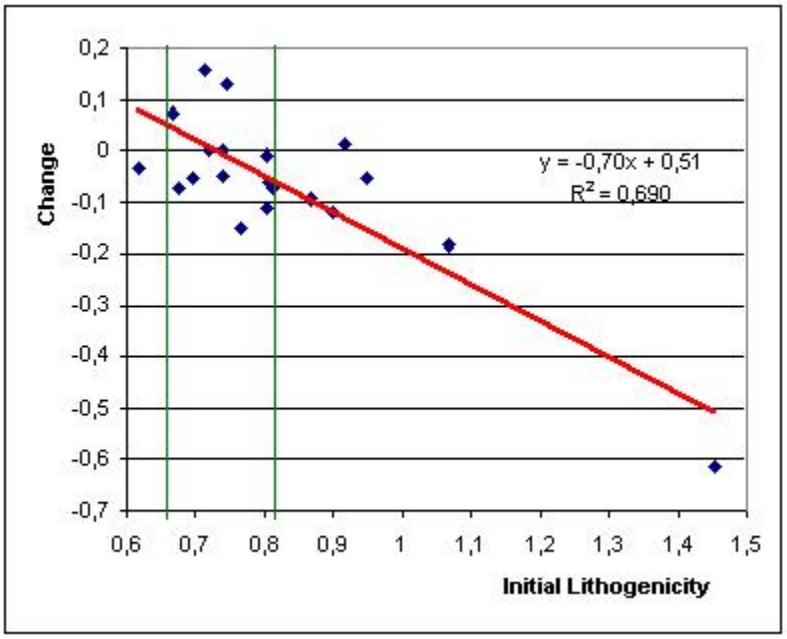


Fig. 2. Individual initial lithogenicity urine (axis X) and their changes after a course of drinking bioactive water Naftussya (axis Y). Green lines show the normal limits.

For convenience, the following analysis was formed three groups (Table 1). In 3 individuals initial normal lithogenicity increased by 0,08÷0,16 units. In 4 people with normal or high lithogenicity its changes were not detected (-0,01÷+0,01). In 12 individuals from a wide range of primary lithogenicity (0,62÷1,07 units) it lowered by 0,03÷0,12 units. Even in a man maximum level of lithogenicity (1,45 units) down to the upper limit of normal (0,84 units), that change was 0,61 units.

Table 1. Changes in urine concentration lithogenic and litholytic substances in various variants change in lithogenicity

Changes in Lithogenicity	n	Level	Lithogenicity Index	Uric Acid, mM/L	Calcium, mM/L	Magnesium, mM/L	Creatinine, mM/L
Decrease	13	Initial	0,86±0,06	3,14±0,71	2,42±0,23*	2,34±0,25*	5,68±0,35*
		Final	0,73±0,03	1,92±0,15	2,39±0,20*	2,62±0,27	6,35±0,38
		Change	-0,13±0,04#	-1,22±0,64	-0,02±0,28	+0,27±0,34	+0,67±0,36
Missing	4	Initial	0,79±0,04	2,68±0,35	2,33±0,20*	2,46±0,28	6,45±0,67
		Final	0,79±0,05	2,13±0,19	2,05±0,18*	1,98±0,38	5,93±0,55
		Change	0,00±0,00	-0,55±0,44	-0,28±0,32	-0,47±0,28	-0,53±0,90
Increase	3	Initial	0,71±0,02	2,73±0,44	2,43±0,34	3,53±0,49	7,33±0,27
		Final	0,83±0,04	1,87±0,24	2,90±0,36	2,95±0,83	4,20±0,76*
		Change	+0,12±0,04#	-0,87±0,41	+0,47±0,59	-0,59±1,07	-3,13±0,72#
	30	Norm	0,73±0,04	2,14±0,10	3,13±0,12	2,93±0,14	7,86±0,43

Notes: 1. Given the average values and their standard errors.
2. Significant difference from the norm marked with *.
3. Significant changes marked #.

As you can see, Lithogenicity decline was due to reduction in the concentration of Uric Acid but not Calcium and increased concentration of Creatinine but not Magnesium. In the absence of changes in Lithogenicity none of its components has not changed significantly. Instead Lithogenicity increase was due to a decrease Creatinine greater extent than the decline Uric Acid. Correlation analysis somewhat corrects visual impression from the data in Table 1. It was found that changes in Lithogenicity strongly correlate directly with changes in the concentration of Uric Acid, significantly inversely with changes in Creatinine and moderately inversely with changes in Magnesium (Table 2), while correlation with changes in Calcium missing (r=0,03).

Table 2. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in Urine Concentration
R=0,955; R^2=0,911; Adjusted R^2=0,895; $F_{(3,2)}$=54,7; $p<10^{-5}$; Std. Error of estimate: 0,050

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(16)}$	p-level
Urine Conc.	r		Intercpt	-,010	,013	-,78	,446
Uric Acid	0,72	,723	,075	,059	,006	9,62	10^{-6}
Creatinine	-0,56	-,546	,076	-,044	,006	-7,15	10^{-5}
Magnesium	-0,40	-,206	,077	-,026	,010	-2,69	,016

For the other controlled electrolytes found significant reduction in concentrations of phosphates as at decreasing and at increasing Lithogenicity, as well as increase daily urine output in the latter case (Table 3).

Table 3. Changes in daily diurese and urine concentration electrolithes in various variants change in lithogenicity

Changes in Lithogenicity	n	Level	Sodium, mM/L	Potassium, mM/L	Chloride, mM/L	Phosphates, mM/L	Diurese, L/24 h
Decrease	13	Initial	120±2*	38,5±2,5*	119±3	12,2±2,0*	1,43±0,21
		Final	129±6*	39,7±1,3*	135±11	10,5±1,6*	1,49±0,09
		Change	+9±7	+1,2±2,3	+16±13	-1,7±0,8#	+0,06±0,20
Missing	4	Initial	117±2*	38,8±4,2	112±3	9,8±2,3*	1,39±0,15
		Final	125±9*	40,1±2,7	127±15	10,0±1,7*	1,81±0,31
		Change	+8±8	+1,3±2,6	+15±15	+0,2±1,5	+0,43±0,27
Increase	3	Initial	122±5*	44,0±5,1	116±6	16,1±2,5	1,02±0,24
		Final	121±8*	39,7±1,9	119±16	13,3±3,0	1,28±0,23
		Change	-1±11	-4,3±3,6	+3±17	-2,8±0,5#	+0,27±0,12#
	30	Norm	161±6	46,4±2,3	120±4	18,0±1,0	1,40±0,07

However, correlation analysis revealed, that changes in Lithogenicity significantly inversely correlate with changes in the concentration of Potassium (r=-0,55), Sodium (r=-0,53) and Chloride (r=-0,44). In the final multiple regression model with stepwise exception came in only Sodium (Table 4).

Table 4. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in Urine Concentration
R=0,964; R^2=0,929; Adjusted R^2=0,911; $F_{(4,2)}$=49,3; $p<10^{-5}$; Std. Error of estimate: 0,046

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(15)}$	p-level
Urine Conc.	r		Intercpt	-,010	,012	-,80	,435
Uric Acid	0,72	,850	,095	,069	,008	8,97	10^{-6}
Creatinine	-0,56	-,600	,075	-,049	,006	-7,95	10^{-5}
Magnesium	-0,40	-,197	,071	-,025	,009	-2,78	,014
Sodium	-0,53	,190	,097	,001	,001	1,96	,068

Among plasma electrolytes and nitrogenous metabolites in one direction with Lithogenicity varied concentration of Calcium (r=0,48), Sodium (r=0,30), Chloride (r=0,30) and Creatinine (r=0,28). In the final multiple regression model with stepwise exception came in only two variables changes in which determine changes in Lithogenicity on 30,5% (Table 5 and Fig. 3).

Table 5. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in Plasma Concentration
R=0,615; R^2=0,378; Adjusted R^2=0,305; $F_{(2,2)}$=5,2; p=0,018; Std. Error of estimate: 0,129

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(17)}$	p-level
Plasma Conc.	r		Intercpt	-,0307	,0308	-1,00	,332
Calcium	0,48	,556	,195	,7981	,2793	2,86	,011
Creatinine	0,28	,384	,195	,0056	,0028	1,97	,065

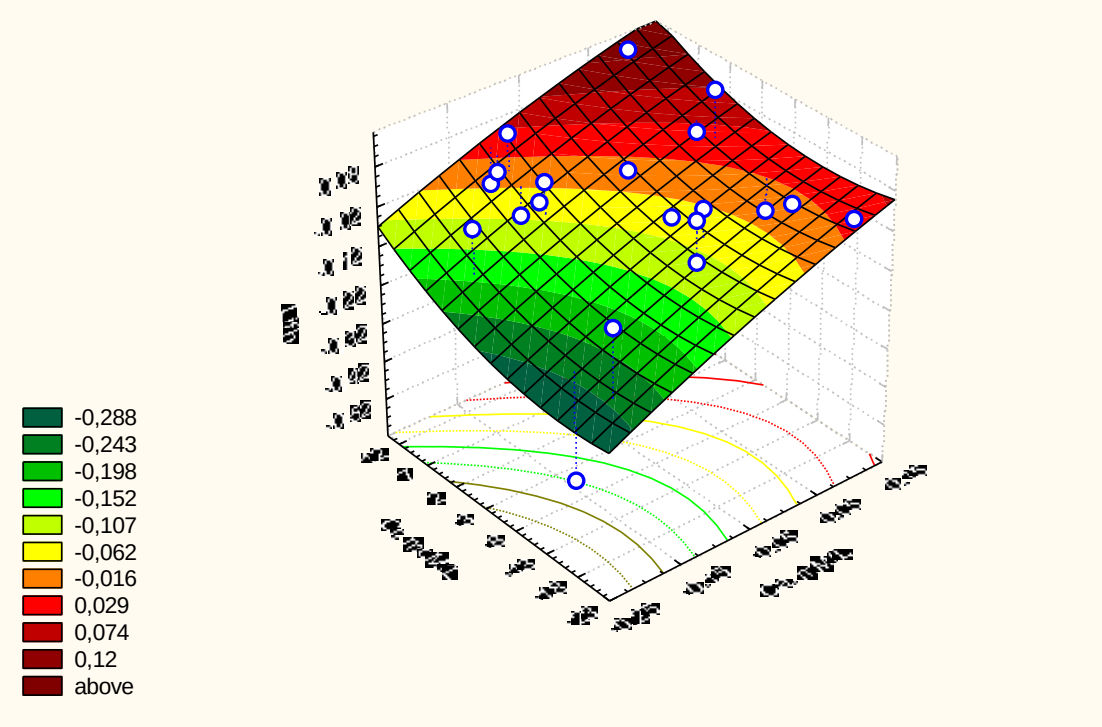


Fig. 3. Dependence changes in urine Lithogenicity (axis Y) from changes in plasma levels of calcium (axis X) and creatinine (axis Y)

Based on obtained data expected hormonal activities: mineralocorticoid MCA=$(Ku/Nau)^{0,5}$ and parathyrine PTA=$(Cap/Pp)^{0,5}$, based on the classical principles and guidelines IL Popovych [28]. Changes in this hormonal markers and cortisol determine the change in Lithogenicity only 24% (Table 6 and 7).

Table 6. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in Hormones
R=0,599; R^2=0,359; Adjusted R^2=0,239; $F_{(3,2)}$=2,99; p=0,062; Std. Error of estimate: 0,135

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(16)}$	p-level
Hormones	r		Intercpt	-,0895	,0315	-2,84	,012
Cortisol	0,29	,314	,206	,0002	,0001	1,52	,147
PTA	0,39	,386	,208	,7655	,4119	1,86	,082
MCA	-0,41	-,287	,207	-,7061	,5089	-1,39	,184

Table 7. Changes in humoral factors connected with change in Lithogenicity

Changes in Lithogenicity	n	Level	Creatinine Plasma, μM/L	Calcium Plasma, mM/L	PTA as $(Cap/Pp)^{0,5}$	Cortisol Plasma, nM/L	MCA as $(Ku/Nau)^{0,5}$
Decrease	13	Initial	77±2*	2,25±0,06	1,49±0,04	703±107*	0,56±0,02
		Final	74±2	2,18±0,04*	1,49±0,04	718±73*	0,56±0,01
		Change	-2±3	-0,06±0,03#	0,00±0,02	+15±57	0,00±0,02
Missing	4	Initial	82±3*	2,15±0,06*	1,34±0,07	603±32*	0,57±0,03
		Final	75±3	2,18±0,05*	1,36±0,04	546±88	0,57±0,02
		Change	-7±6	+0,03±0,05	+0,05±0,03	-57±117	-0,01±0,02
Increase	3	Initial	81±5*	2,13±0,09*	1,33±0,03	620±75*	0,60±0,03
		Final	88±4*	2,17±0,03*	1,43±0,03	837±131*	0,57±0,03
		Change	+7±4	+0,03±0,07	+0,09±0,01#	+217±162	-0,02±0,00#
	30	Norm	69±3	2,30±0,03	1,38±0,04	405±23	0,54±0,03

Among changes in electroskin conductivity in recorded points of acupuncture coefficients correlation with changes in Lithogenicity ranked as follows: Pg(ND) Right: 0,43; TR(X) Left: 0,40; Pg(ND) Left: 0,33; MC(AVL) Right: 0,31; TR(X) Right: 0,29. In the multiple regression model with stepwise exception came in only two first variables characterizing the state of the nervous and endocrine systems, changes in which determine changes in Lithogenicity on 13,5% only (Table 8).

Table 8. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in Conductivity in Acupuncture Points
R=0,475; R^2=0,226; Adjusted R^2=0,135: $F_{(2,2)}$=2,48; p=0,114; Std. Error of estimate: 0,144

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(17)}$	p-level
AP	r		Intercpt	-,087	,036	-2,45	,025
Pg(ND) Right	0,43	,307	,252	,010	,008	1,22	,241
TR(X) Left	0,40	,234	,252	,009	,010	,93	,366

Among changes in spectral parameters HRV coefficients correlation with changes in Lithogenicity ranked as follows: VLF: 0,85 (Fig. 4); ULF: 0,61; LF: 0,51; HF: 0,49.

Among changes in temporal parameters HRV coefficients correlation with changes in Lithogenicity ranked as follows: SDNN: 0,79 (Fig. 5); RMSSD: 0,69; pNN_{50}: 0,66.

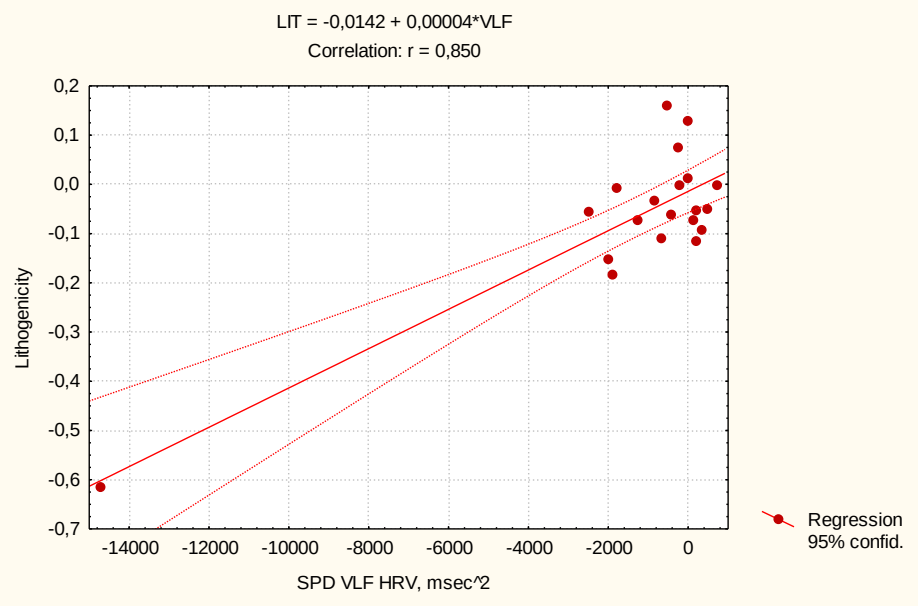


Fig. 4. Relationship between changes in Spectral Power Density VLF HRV and urine Lithogenicity

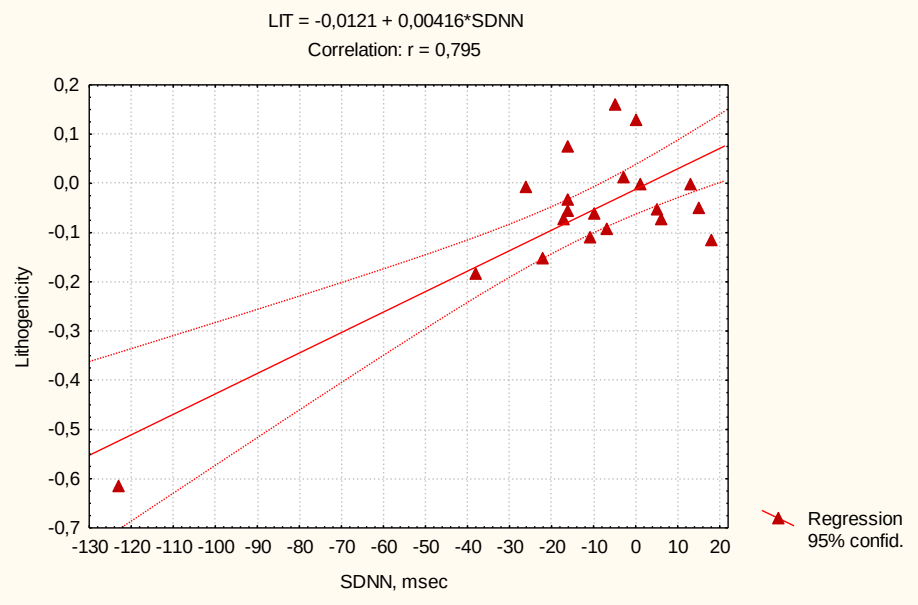


Fig. 5. Relationship between changes in SDNN HRV and urine Lithogenicity

Among changes in Baevskiy's parameters HRV coefficients correlation with changes in Lithogenicity ranked as follows: Heart Rate: 0,70; Moda: -0,60; AMo: -0,52; Activity Regulatory Systems Index: 0,52; Stress Index: 0,33; MxDMn: -0,29.

Changes in three variables HRV determine changes in Lithogenicity on 80% (Table 9).

Table 9. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in HRV parameters
R=0,911; R^2=0,831; Adjusted R^2=0,799; $F_{(3,2)}$=26,2; $p<10^{-5}$; Std. Error of estimate: 0,069

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(16)}$	p-level
HRV	r		Intercpt	-,0048	,0183	-,26	,795
VLF	0,85	,615	,237	,0000	,0000	2,59	,020
Heart Rate	0,70	2,220	,785	,0176	,0062	2,83	,012
Moda	-0,60	2,083	,667	,0018	,0006	3,12	,007

Changes in HRV, AP and metabolic parameters included in multiple regression model determine changes in Lithogenicity on 84,5% (Table 10,11, Fig. 6).

Table 10. Changes in neural factors connected with change in Lithogenicity

Changes in Lithogenicity	n	Level	Heart Rate, beats/min	Moda HRV, msec	SPD VLF HRV, $msec^2$	ESC TR(X) Left, units	Ch HDLP, mM/L
Decrease	13	Initial	72,2±6,6	854±66	2776±1130	61,9±1,9	1,63±0,14
		Final	70,3±2,5	842±35	1003±164*	62,1±1,0*	1,50±0,11
		Change	-1,9±6,8	-12±63	-1773±1114	+0,2±1,3	-0,13±0,06#
Missing	4	Initial	76,2±6,1	788±77	1403±609	66,0±1,8*	1,10±0,13*
		Final	73,7±6,3	825±83	1070±86*	65,2±1,4*	1,29±0,09
		Change	-2,5±0,9#	+37±13#	-333±530	-0,8±0,6	+0,19±0,14
Increase	3	Initial	71,0±2,0	817±17	872±303	63,3±3,3	1,41±0,22
		Final	68,7±3,5	867±44	608±159*	65,3±3,9	1,31±0,17
		Change	-2,3±1,8	+50±29	-264±146	+2,0±1,0#	-0,11±0,10
	30	Norm	68,8±0,2	871±2	1353±52	58,0±0,6	1,49±0,11

Table 11. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in HRV, AP and metabolic parameters
R=0,941; R^2=0,886; Adjusted R^2=0,845; $F_{(5,1)}$=21,7; $\chi^2_{(5)}$=33,6; $p<10^{-5}$; Std. Err.: 0,062

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(14)}$	p-level
Variables	r		Intercpt	-,01689	,01673	-1,01	,330
VLF HRV	0,85	,411	,226	,00002	,00001	1,82	,091
Heart Rate	0,70	3,267	,808	,02596	,00642	4,04	,001
Moda HRV	-0,60	2,933	,676	,00248	,00057	4,34	,001
Sodium Urine	-0,53	,179	,128	,00123	,00088	1,39	,185
TR(X) Left	0,40	,245	,106	,00971	,00422	2,30	,037

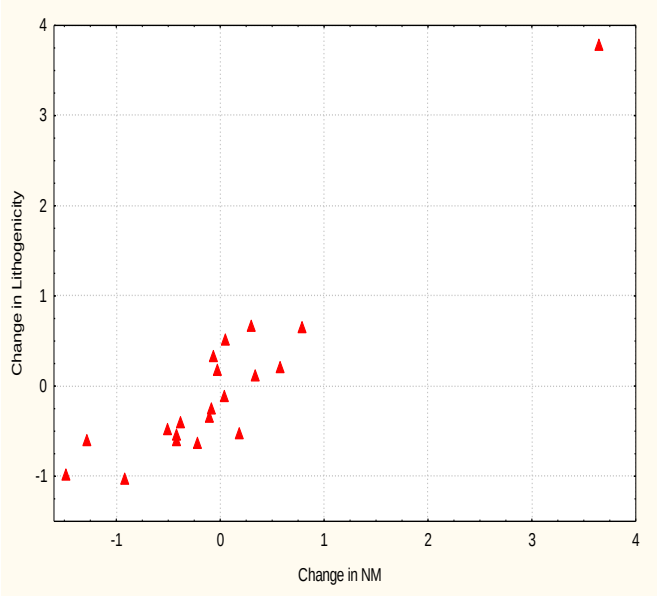


Fig. 6. Canonical correlation between changes in Neural and Metabolic factors (axis X) and urine Lithogenicity (axis Y)

We now turn to the analysis of parameters of immunity. Revealed (Table 12) that examined cohort characterized by low completeness of phagocytosis of E. coli by neutrophils in combination with high intensity and activity phagocytosis, so bactericidal capacity (BC) of neutrophils is in the majority in the lower zone rules.

It is noteworthy that the BC rises for all variants of changes in Lithogenicity urine, and to the maximum extent in the case of increase, which is very beneficial in terms of prevention of pyelonephritis agent is often just E. coli [31,37,39].

Table 12. Changes in parameters of phagocytosis of E. coli by neutrophils in various variants change in Lithogenicity

Changes in Lithogenicity	n	Level	Killing Index vs E. coli, %	Microbial Count for E. coli	Phagocytic Index vs E. coli, %	BC vs E. coli, 10^9 Bact/L
Decrease	13	Initial Final Change	39,4±1,8* 41,8±2,0* +2,4±2,7	65,4±2,2* 66,3±1,3* +1,0±2,8	99,7±0,1* 99,8±0,1* +0,1±0,2	86±6 98±5 +12±5#
Missing	4	Initial Final Change	35,4±3,3* 43,1±2,6* +7,7±2,2#	67,8±2,6* 73,7±4,6* +6,5±3,7	99,3±0,2* 99,8±0,2* +0,5±0,3	86±7 112±1* +26±7#
Increase	3	Initial Final Change	40,4±2,7* 46,7±2,0* +6,3±3,9	61,0±4,7 68,2±2,5* +7,2±7,2	99,0±0,6 99,7±0,3* +0,7±0,3#	70±6* 106±7 +36±4#
	30	Norm	62,0±1,0	54,7±1,1	98,3±0,1	95±2

Concerning changes in parameters of phagocytosis of Staph. aureus situation is less clear (Table 13). The same table shows the data on Popovych's LeukocytaryStrain Index, which shows a downward trend in cases both reduce Lithogenicity and its stability and prone to increases in cases of its increase.

Table 13. Changes in parameters of phagocytosis of Staph. aureus by neutrophils in various variants change in lithogenicity

Changes in Lithogenicity	n	Level	Killing Index vs Staph. aureus, %	Microbial Count for Staph. aur.	Phagocytic Index vs Staph. aur., %	BC vs Staph. aur., 10^9 Bact/L	Popovych's Leukocytary Strain Index
Decrease	13	Initial Final Change	43,5±1,4* 46,4±1,1* +2,9±1,5	58,6±1,1* 63,5±2,2 +4,9±2,3#	99,3±0,2* 99,6±0,1* +0,3±0,2	85±5* 103±4 +18±6#	0,27±0,12 0,13±0,04 -0,14±0,10
Missing	4	Initial Final Change	43,8±1,6* 42,3±1,3* -1,5±2,6	62,3±3,5 69,8±3,0* +7,5±1,3#	98,5±0,3 100±0,0* +1,5±0,3#	97±5 106±3 +9±6	0,16±0,05 0,09±0,03 -0,08±0,05
Increase	3	Initial Final Change	43,0±5,5* 50,0±2,9* +7,0±6,9	62,0±4,1 62,2±4,2 +0,2±0,4	98,7±0,7 99,3±0,3* +0,7±0,9	77±14* 103±10 +26±22	0,12±0,02 0,22±0,11 +0,10±0,10
	30	Norm	58,9±0,8	61,6±1,0	98,3±0,2	102±2	0,09±0,02

Concerning changes in parameters of cellular immunity found that the decrease Lithogenicity accompanied by an increase in blood both T-killers and Natural Killers as well as "active" subpopulation of T-Lymphocytes. The latest increases also in cases increasing Lithogenicity (Table 14).

Table 14. Changes in parameters of cellular immunity in various variants change in lithogenicity

Changes in Lithogenicity	n	Level	$CD4^+$ T-Lymph., %	$CD8^+$ T-Lymph., %	$CD16^+$ NK-Lymph., %	"Active" T-Lymph., %
Decrease	13	Initial Final Change	36,7±1,5 37,6±1,6 +0,9±1,1	23,1±1,0 25,3±1,6 +2,2±1,0#	19,1±1,2 21,4±1,3* +2,3±1,0#	29,8±1,2 32,2±0,9 +2,4±0,9#
Missing	4	Initial Final Change	39,5±2,2 39,0±2,5 -0,5±2,1	24,2±0,7 25,2±2,3 +1,0±1,7	22,0±2,2* 22,8±2,2* +0,8±2,2	32,3±2,9 32,0±1,2 -0,3±2,2
Increase	3	Initial Final Change	33,3±2,0* 35,7±0,7* +2,3±1,4	20,3±0,3* 19,7±0,9* -0,7±0,7	14,7±2,6 14,8±0,7* +0,1±2,9	28,0±3,8 31,3±3,0 +3,3±0,9#
	30	Norm	39,5±0,6	23,5±0,6	17,0±0,5	30,0±0,9

Table 15. Changes in parameters of humoral immunity in various variants change in lithogenicity

Changes in Lithogenicity	n	Level	$CD22^+$ B-Lymph., %	IgM, g/L	IgG, g/L	IgA, g/L	CIC, units
Decrease	13	Initial Final Change	23,6±0,9* 24,2±1,0* +0,6±1,4	2,25±0,03* 2,10±0,04* -0,15±0,05#	16,5±1,0* 15,8±1,2* -0,7±0,6	2,25±0,03* 2,10±0,04* -0,15±0,05#	31±4* 43±4 +12±4#
Missing	4	Initial Final Change	22,2±1,9 25,0±3,5 +2,8±3,9	2,14±0,07* 2,07±0,07* -0,07±0,08	14,4±0,5* 14,8±0,8* +0,4±0,7	2,14±0,07* 2,07±0,07* -0,07±0,08	38±8 43±6 +5±5
Increase	3	Initial Final Change	20,0±2,1 19,0±3,5 -1,0±2,6	2,05±0,25* 2,03±0,27* -0,03±0,05	14,5±1,2 14,8±0,3* +0,3±0,9	2,05±0,25 2,03±0,27 -0,03±0,05	30±6* 35±2* +5±6
	30	Norm	20,0±0,6	1,15±0,05	12,8±0,5	1,88±0,06	45±3

Concerning changes in parameters of humoral immunity found that the decrease Lithogenicity accompanied by an decrease in blood elevated levels Igg M and A in combination with increase low levels of CIC (Table 15).

Described immune responses we interpret as favorable as changes in the intensity of phagocytosis Staph. aureus (Fig. 7).

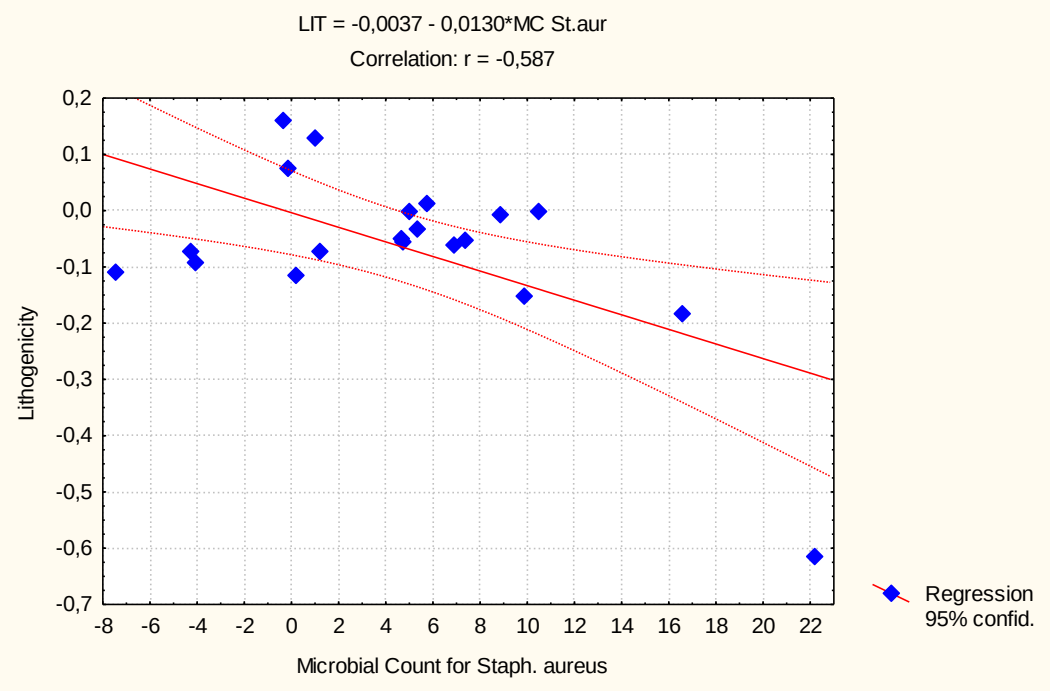


Fig. 7. Relationship between changes in Microbial Count for Staph. auteus and urine Lithogenicity

In the multiple regression model with stepwise exception came in only 5 immune variables changes in which determine changes in Lithogenicity on 44% (Table 16, Fig. 8).

Table 16. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in immune parameters
R=0,764; R^2=0,584; Adjusted R^2=0,436; $F_{(5,1)}$=3,93; $\chi^2_{(5)}$=13,6; p=0,019; Std. Error: 0,116

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(14)}$	p-level
Immune par	r		Intercpt	-,0855	,0642	-1,33	,204
MC St. aur.	-0,59	-,441	,221	-,0097	,0049	-2,00	,066
PhI E. coli	0,27	,326	,176	,0791	,0428	1,85	,086
BC E. coli	0,27	,219	,186	,0020	,0017	1,17	,260
Strain Index	0,29	,260	,174	,1368	,0916	1,49	,158
T-L active	0,41	,256	,220	,0116	,0100	1,16	,264

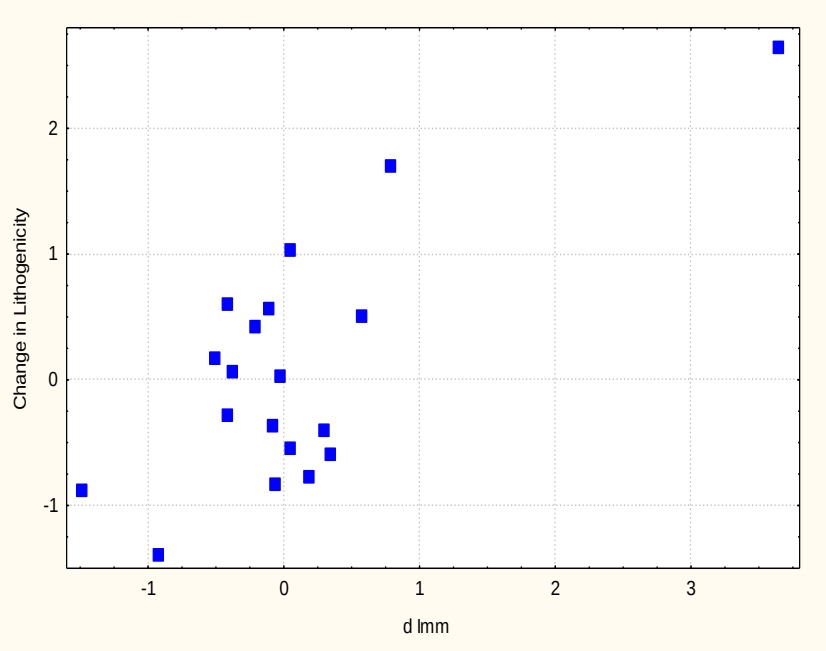


Fig. 8. Canonical correlation between changes in Immune factors (axis X) and urine Lithogenicity (axis Y)

In the **final** multiple regression model with stepwise exception came in 4 neuro-endocrine, 5 immune and one metabolic variables changes in which determine changes in Lithogenicity on 94% (Table 17, Fig. 9).

Table 17. Regression Summary for Dependent Variable: change in Lithogenicity; Independent Variables: change in neural, metabolic and immune parameters
R=0,980; R^2=0,961; Adjusted R^2=0,939; $F_{(7,1)}$=42,6; $\chi^2_{(7)}$=47,2; $p<10^{-7}$; Std. Error: 0,038

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(12)}$	p-level
Variables	r		Intercpt	-,08808	,01822	-4,83	,0004
VLF HRV	0,85	,375	,149	,00002	,00001	2,52	,0270
Heart Rate	0,70	3,210	,547	,02551	,00435	5,86	,0001
TR(X) Left	0,40	,232	,068	,00921	,00268	3,44	,0049
T-Lymph active	0,41	,193	,074	,00876	,00335	2,61	,0226
BC E. coli	0,27	,269	,062	,00242	,00056	4,33	,0010
Moda HRV	-0,60	2,816	,463	,00238	,00039	6,08	,0001
Sodium Urine	-0,53	,311	,088	,00214	,00060	3,54	,0041

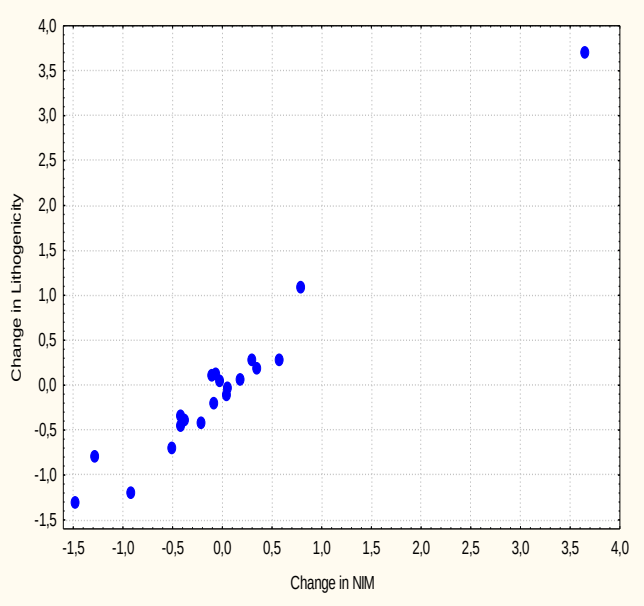


Fig. 9. Canonical correlation between changes in Neural, Metabolic and Immune factors (axis X) and urine Lithogenicity (axis Y)

At the final stage analyze the relationships between changes in Neural, Metabolic and Immune factors, on the one hand, and in components of Lithogenicity urine, on the other.

Based on the factor loadings, the maximum proportion variance of changes in the litogenicity explain changes in the concentration of uric acid, while the contribution of calcium worthless (Table 18).

Table 18. Canonical Analysis Summary. Factor Structure of left set

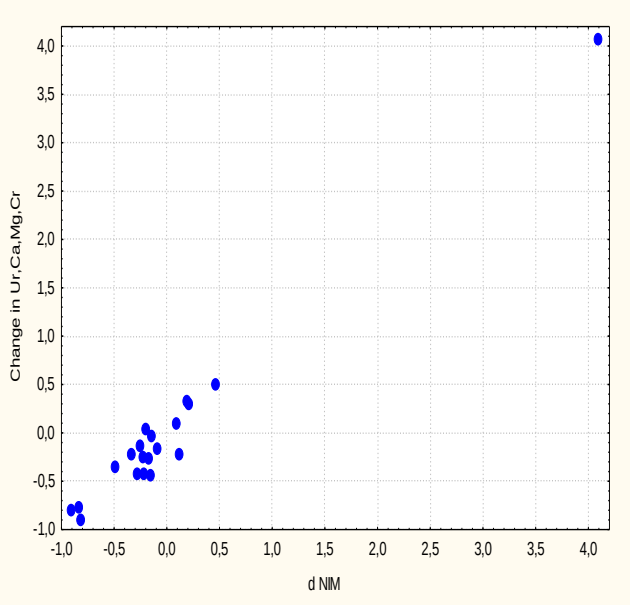
Change in Urine Concentration	Root 1
Uric Acid	**-,853**
Creatinine	**,412**
Magnesium	**,331**
Calcium	,014

Table 19. Canonical Analysis Summary. Factor Structure of right set

Changes in Neural, Metabolic and Immune factors	Root 1
SPD VLF HRV	**-,938**
Heart Rate	**-,843**
HDLP Cholesterol Plasma	**-,565**
Active T-Lymphocytes	**-,453**
Phagocytic Index for E. coli	**-,408**
Conductivity in AP TR(X) Left	**-,360**
Bacterocidal Capacity for E. coli	**-,166**
Moda HRV	**,755**
Sodium Urine Concentration	**,653**
Potassium Urine Concentration	**,626**
Microbial Count for Staph. aureus	**,598**
Bacterocidal Capacity for Staph. aureus	**,506**

On the other hand, the maximum load on the neuro-immune-metabolic canonical root gives SPD VLF band HRV. It is speculated that VLF band HRV associated with oscillation blood levels of renin (0,04 Hz) and epinephrine (0,025 Hz), reflects thermoregulatory cycles, cerebral ergotropic and metabolotropic outflows, activation of cerebral sympathetic-adrenal system, sympathetic activity [cit by: 2]. All variables can be roughly classified as **lithogenic** and **litholytic**.

Canonical correlation between changes in Neural, Metabolic and Immune factors and urine concentration of Lithogenicity factors is very strong (Fig. 10).



R=0,989; R^2=0,978; $\chi^2_{(48)}$=73; p=0,011

Fig. 10. Canonical correlation between changes in Neural, Metabolic and Immune factors (axis X) and urine concentration of Lithogenicity factors (axis Y)

We [36] have previously shown that reducing urine Lithogenicity accompanied by a decrease urine concentration Chloride and Potassium, as well as Baevskiy's Activity Regulatory Systems Index, Asymmetry δ-, θ- and α-rhythms of background EEG and α-rhythm normalized SPD in right loci Fp2, F8, T6 and O2, while HRV markers of vagal tone (RMSSD, pNN_{50}, TI) also Frequency Deviation θ- and β-rhythms, Amplitude β- and δ-rhythms, δ-rhythm PSD in loci Fp2, F8, T3, T5, O1, O2, β-rhythm PSD in loci Fp1, F4 and θ-rhythm PSD in loci T3 and F7 rising.

A limited amount of articles makes us complete its correlation matrices, but postpone the discussion in the following article.

Table 20. Canonical Analysis Summary. Correlations left set with right set (p<0,05; p<0,01; p<0,001)

Varia-bles	VLF HRV	Mo HRV	HR	ESC TR(X) L	K Urine	Na Urine	HDLP Chol	BC Staph	BC Esch	MC Staph	MC Esch	T-L act
Urates	**,89**	**-,77**	**,83**	,12	**-,70**	**-,65**	**,62**	**-,51**	-,09	**-,45**	**,33**	**,49**
Creatin	-,28	,07	-,15	**-,53**	-,09	,25	-,16	,24	**-,39**	**,37**	**-,43**	,08
Mg	-,16	,17	-,20	-,09	**,40**	,09	,01	,09	-,16	,07	,12	-,17
Ca	-,04	,11	-,10	,22	**,42**	,12	-,13	,16	,25	-,18	,12	-,12

Table 21. Canonical Analysis Summary. Correlations within right set

Variables	Chol α-LP	Moda HRV	HR	VLF HRV	TR(X) Left	Ku	Nau	MC Staph	MC Esch	BC Staph	BC Esch
Chol α-LP	1,00										
Moda	**-,44**	1,00									
HR	**,50**	**-,98**	1,00								
VLF HRV	**,61**	**-,81**	**,87**	1,00							
TR(X) L	,16	**-,36**	**,33**	**,40**	1,00						
Ku	-,30	**,52**	**-,56**	**-,56**	,03	1,00					
Nau	**-,42**	**,58**	**-,64**	**-,63**	-,20	**,56**	1,00				
MC Staph	-,22	**,58**	**-,56**	**-,64**	**-,70**	**,35**	**,38**	1,00			
MC Esch	,29	-,24	**,31**	**,43**	,27	-,22	**-,45**	-,20	1,00		
BC Staph	**-,45**	**,65**	**-,64**	**-,62**	-,37	**,41**	**,49**	,30	**-,59**	1,00	
BC Esch	,06	,15	-,09	-,05	-,16	-,07	-,13	-,19	-,02	,28	1,00
T-L act	,02	**-,46**	**,44**	**,48**	**,38**	**-,54**	**-,47**	**-,55**	-,00	-,21	-,16

Table 22. Canonical Analysis Summary. Correlations within left set

Variables	Creatinine	Uric Acid	Calcium
Creatinine	1,00		
Uric Acid	,04	1,00	
Calcium	**-,33**	-,26	1,00
Magnesium	,20	-,12	**,57**

ACKNOWLEDGMENT

We express our sincere gratitude to administration JSC "Truskavets'kurort" for help in carrying out metabolic and immune tests and recording HRV.

ACCORDANCE TO ETHICS STANDARDS

Tests in patients are conducted in accordance with positions of Helsinki Declaration 1975, revised and complemented in 2002, and directive of National Committee on ethics of scientific researches. During realization of tests from all participants the informed consent is got and used all measures for providing of anonymity of participants.

For all authors any conflict of interests is absent.

REFERENCES

1. Alyekseyev OI, Shymonko IT, Orlov OB. Treatment and Rehabilitation on the Spa Truskavets' and Skhidnytsia [in Russian]. Kyiv: Zdorovya, 1994. 176 p.
2. Baevskiy RM, Ivanov GG. Heart Rate Variability: theoretical aspects and possibilities of clinical application [in Russian]. Ultrazvukovaya i funktsionalnaya diagnostika. 2001; 3: 106-127.
3. Barylyak LG, Kruhliy YuZ, Zukow W, Yanchiy OR, Popovych IL. Indicators, distinctive for women with different ovarian status and different responses streslimiting effect of bioactive water Naftussya spa Truskavets'. Journal of Education, Health and Sport. 2015; 5(3): 247-258.
4. Brundig P, Berg W, Schneider HJ. Stress und Harnsteinbildungsrisiko. I. Der Einfluss von Stress auf lithogene Harnsubstanzen. Urol. Int. 1981; 36(4): 199-207.
5. Brundig P, Berg W, Schneider HJ. Stress und Harnsteinbildungsrisiko. II. Der Einfluss von Stress auf litholytische Harnsubstanzen. Urol. Int. 1981; 36(4): 265-273.
6. Chebanenko OI, Chebanenko LO, Popovych IL. Multivariate Balneoeffects of Factors of Spa Truskavets' and

Forecasting [in Ukrainian]. Kyiv: UNESCO-SOCIO. 2012. 496 p.
7. Chebanenko OI, Flyunt IS, Popovych IL, Balanovs'kyi VP, Lakhin PV. Water Naftussya and Water-Salt Exchange [in Ukrainian]. Kyiv: Naukova dumka. 1997. 141 p.
8. Chebanenko OI, Popovych IL, Chebanenko LO. Adaptogenic Essence of Balneophytoterapy [in Ukrainian]. Kyiv: UNESCO-SOCIO. 2013. 380 p.
9. Douglas SD, Quie PG. Investigation of Phagocytes in Disease. Churchil. 1981. 110 p.
10. Dranovs'kyi AL, Babelyuk VYe, Popovych AI**,** Krayevyi VO, Flyunt VR**.** Emergency effects of bioactive water Naftussya on level testosteronemia and related changes of some physiological parameters in healthy men [in Ukrainian]. Health males. 2013; 4(47): 161-163.
11. Flyunt IS. Features exchange electrolytes in patients with urolithiasis with stones of different composition and influence on him balneotherapy on spa Truskavets' [in Ukrainian]. Medical rehabilitation, kurortology, physiotherapy. 1999; 3(suppl): 85.
12. Flyunt IS, Chebanenko OI. Parathyrinelike and calcitoninelike term effects of bioactive water Naftussya [in Ukrainian]. Ukrainian Balneologycal Journal. 2001; 2: 52-56.
13. Flyunt IS, Fajda OI, Flyunt VR et al. Natural classification effects balneotherapy on spa Truskavets' in patients with oxalate urolithiasis. Communication 1: Level lithogenic and litholytic substances [in Ukrainian]. Medical Hydrology and Rehabilitation. 2004; 2(4): 53-56.
14. Flyunt IS, Fajda OI, Flyunt VR et al. Natural classification effects balneotherapy on spa Truskavets' in patients with oxalate urolithiasis. Communication 2: Concentration in urine and plasma osmotic active substances and the acidity of urine [in Ukrainian]. Medical Hydrology and Rehabilitation. 2005; 3(1): 74-77.
15. Flyunt IS, Velychko LM, Flyunt VR et al. Natural classification effects balneotherapy on spa Truskavets' in patients with oxalate urolithiasis. Communication 3: Urinary excretion of various solid substances [in Ukrainian]. Medical Hydrology and Rehabilitation. 2005; 3(2): 14-19.
16. Flyunt VR, Flyunt IS, Barylyak LG**,** Piskov GG, Yanchiy OR. Links of urine Ca/Mg coefficient of Lithogenicity of some parameters of electrolyte exchange in patients spa Truskavets' [in Ukrainian]. Medical Hydrology and Rehabilitation. 2013; 11(3): 43-49.
17. Flyunt VR, Flyunt IS, Popovych IL. Neuro-hormonal mechanism of action of bioactive water Naftussya on urine lithogenicity in rats [in Ukrainian]. Achievements of Clinical and Experimental Medicine. 2013; 2(19): 288.
18. Flyunt VR, Koval'chuk GYa, Flyunt IS, Tymochko OB, Velychko LM, Shats'ka OM, Lips'ka AM. The immediate effects of bioactive water Naftussya on urinal lithogenicy and some parameters of electrolithic exchange in patients with oxalic urolithiasis [in Ukrainian]. Medical Hydrology and Rehabilitation. 2007; 5(4): 30-33.
19. Flyunt IS, Nishcheta IV. Effect of balneotherapy on spa Truskavets' on water-electrolyte metabolism in patients with urolithiasis [in Ukrainian]. In: Health resources Carpathians and adjacent regions. Chernivtsi: BSMA, 1999. 64-66.
20. Goryachkovskiy AM. Clinical Biochemistry [in Russian]. Odesa: Astroprint. 1998. 608 p.
21. Hrinchenko BV, Snihur OV, Flyunt VR, Zukow W. Relationships between urina lithogenicity and neuro-humoral immune factors in patients with chronic pyelonephrite and cholecystite. Journal of Education, Health and Sport. 2015; 5(5): 44-51.
22. Ivassivka SV, Popovych IL, Aksentiychuk BI, Bilas VR. Nature of Balneofactors Water Naftussya and the Essence of its Therapeutic and Preventive action [in Ukrainian]. Truskavets': Truskavets'kurort. 1999. 125 p.
23. Ivassivka SV, Popovych IL, Aksentiychuk BI, Flyunt IS. Physiological Activity of Uric Acid and its Role in the mechanism of action of Water Naftussya [in Ukrainian]. Kyiv: Computerpress. 2004. 163 p.
24. Kostyuk PG, Popovych IL, Ivassivka SV (editors). Chornobyl', Adaptive and Defensive systems, Rehabilitation [in Ukrainian]. Kyiv: Computerpress. 2006. 348 p.
25. Kozyavkina OV, Kozyavkina NV, Gozhenko OA, Gozhenko AI, Barylyak LG, Popovych IL. Bioactive Water Naftussya and Neuroendocrine-Immune Complex [in Ukrainian]. Kyiv: UNESCO-SOCIO. 2015. 349 p.
26. Lapovets' LYe, Lutsyk BD. Handbook of Laboratory Immunology [in Ukrainian]. L'viv. 2002. 173 p.
27. Nishcheta YuM, Flyunt IS, Nishcheta IV, Stetsenko IM. Effect of rehabilitation at the spa Truskavets' the exchange of uric acid in the victims of the Chornobyl' accident [in Ukrainian]. In: Medical rehabilitation of victims of the Chernobyl disaster. Truskavets'. 1996. 39-40.
28. Popovych IL. Stresslimiting Adaptogene Mechanism of Biological and Curative Activity of Water Naftussya [in Ukrainian]. Kyiv: Computerpress. 2011. 300 p.
29. Popovych IL. The concept of neuro-endocrine-immune complex (Review) [in Russian]. Medical Hydrology and Rehabilitation. 2009; 7(3): 9-18.

30. Popovych IL, Barylyak LG. Influence of course using of bioactive water Naftussya on stress level at women with endocrine and gynecological pathology [in Ukrainian]. Medical Hydrology and Rehabilitation. 2009; 7(3): 100-118.
31. Popovych IL, Flyunt IS, Alyeksyeyev OI, Barylyak LG, Bilas VR. Sanogenetic Principles of Rehabilitation on Spa Truskavets' Urological Patients Chernobyl Cohort [in Ukrainian]. Kyiv: Computerpress. 2003. 192 p.
32. Popovych IL, Popovych AI. Personality traits in the reaction of the autonomic nervous system per application ozokerite and the possibility of its prediction [in Russian]. In: Actual problems of biophysical medicine. Kyiv. 2012. 114-115.
33. Popovych IL, Shulyak OV, Flyunt VR, Petsyukh SV. Influence balneotherapy on spa Truskavets' with takig Canephron® N on urina lithogenicity and its metabolic and neuroendocrine-immune accompaniments in patients with chronic pyelonephrite and cholecystite [in Ukrainian]. Medical Hydrology and Rehabilitation. 2015; 13(4): 101.
34. Popovych IL, Shymonko IT, Flyunt IS, Stremenyuk YeS, Bilas VR. Effects of balneotherapy on the exchange of electrolytes in patients with urolithiasis [in Ukrainian]. In: Problems and prospects of further development of sanatorium and resort business. Truskavets'. 1991. 138-140.
35. Popovych IL, Vis'tak (Markevych) HI, Humega MD, Ruzhylo SV. Vegetotropic Effects of Bioactive Water Naftussya and their Neuroendocrine-Immune, Metabolic and Hemodynamic Accompaniments [in Ukrainian]. Kyiv: UNESCO-SOCIO. 2014. 163 p.
36. Shulyak OV, Flyunt IS, Flyunt VR, Fil' VM, Hrinchenko BV, Snihur OV, Zukow W, Yanchiy OR, Popovych IL. Influence balneotherapy on spa Truskavets' with using Canephron® N on urina lithogenicity and its metabolic and neuroendocrine-immune accompanyments in patients with chronic pyelonephrite and cholecystite. Journal of Education, Health and Sport. 2015; 5(11): 453-465.
37. Skorobogatov MO. Spa treatment of urolithiasis [in Ukrainian]. Kyiv: Zdorovya. 1991. 64 p.
38. Stetsenko HI, Beyda PA, Perchenko VP, Loyik IV, Flyunt IS, Yatsyuk VM. Restoration of the digestive and urinary excretion systems of victims of the Chernobyl disaster [in Ukrainian]. In: Resort Rehabilitation Victims of Chornobylian Accident. Kyiv: Zdorovya. 1999. 65-79.
39. Tiktinskiy OL, Aleksandrov VP. Urolithic Disease [in Russian]. SPb: Piter, 2000. 384 p.
40. Tiselius HS. A biochemical basis for grouping of patients with urolithiasis. Europ Urol. 1978; 4: 241-249.
41. Yessypenko BYe. Physiological effects of mineral water Naftussya [in Russian]. Kyiv: Naukova dumka, 1981. 216 p.