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## Peculiarities of modulating effects of "Myroslava" and "Khrystyna" mineral waters on neuroendocrine-immune complex and metabolism in patients of Truskavets' spa

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### Abstract

**Background.** Earlier we found effects on the neuroendocrine-immune complex and metabolism, which are common to the mineral waters "Myroslava" and "Khrystyna", instead different from the effects of Naftussya water. The purpose of this article is to find the specific effects of these waters. **Materials and Methods.** The object of clinical-physiological observation were 22 men aged 23-70 years, who underwent rehabilitation treatment of chronic cholecystitis and pyelonephritis in remission in the Truskavets' spa. The examination was performed twice, before and after a 7-10-day course of balneotherapy. Half of the patients received bioactive water Naftussya and sulfate-chloride sodium-magnesium mineral water "Myroslava" (M 5 g/L), and the other half - Naftussya and "Khrystyna" (M 10 g/L). The subject of the study were the parameters of the neuroendocrine-immune complex, microbiota, cholekinetics and metabolism. **Results.** Screening revealed specific changes in 37 parameters grouped into 5 patterns. The first pattern combines 11 parameters that decrease under the influence of "Myroslava" water (Myr), while increase under the influence of "Khrystyna" water (Khr). For the other 7 parameters of the second pattern, Myr acts similarly, while Khr is ineffective. Both mineral waters have a stimulating effect on 8 parameters, while Myr is inferior to Khr. Myr has a upregulating effect on 9 parameters of the fourth pattern, while Khr has a downregulating effect. Finally, sodium excretion and leukocyturia are reduced under the influence of both waters, but to a greater extent under the influence of Khr. **Conclusion.** The newly created sulfate-chloride sodium-magnesium drinking mineral waters of Truskavets' spa

have both common and specific favorable modulating neuroendocrine, metabolic and immune effects on patients with chronic cholecystitis and pyelonephritis.

**Keywords:** sulfate-chloride sodium-magnesium drinking mineral waters, Truskavets' spa, EEG, HRV, hormones, metabolism, immunity.

## INTRODUCTION

Earlier we found effects on the neuroendocrine-immune complex and metabolism, which are common to the mineral waters "Myroslava" and "Khrystyna", instead different from the effects of Naftussya water [6,12,13]. The purpose of this article is to find the specific effects of these waters.

## MATERIALS AND METHODS

The object of clinical-physiological observation were 22 men aged 23-70 years, who underwent rehabilitation treatment in the Truskavets' spa of chronic cholecystitis and pyelonephritis in remission with of neuroendocrine-immune complex dysfunction. The examination was performed twice, before and after a 7-10-day course of balneotherapy. Half of the patients received bioactive water Naftussya (3 mL/kg one hour before meals three times a day) and sulfate-chloride sodium-magnesium mineral water "Myroslava" (in half an hour in the same dose), and the other half - Naftussya and "Khrystyna" waters [12].

The day before, daily urine was collected, in which was determined the concentration of electrolytes and nitric metabolites [12] as well as bacteriuria, erythrocyturia and leukocyturia. The same metabolic parameters were determined in plasma as well as glucose and lipids [12].

We determined content in plasma major hormones of adaptation: Cortisol, Testosterone, Calcitonin and Triiodothyronine as well as Parathyroid activity evaluated by coefficient  $(\text{Cap}\cdot\text{Pu}/\text{Cau}\cdot\text{Pp})^{0,25}$  [12].

The state of the autonomous regulation we estimated by the method heart rate variability (HRV) [3,8,12]. Calculated the entropy (h) of the relative spectral powers (SP) of the HRV bands [7] as well as Kerdö's Vegetative Index [4]. We recorded simultaneously with HRV EEG monopolar in 16 loci [13]. In addition, calculated Laterality Index and Shannon's entropy for PSD each Rhythm [7].

Immune status evaluated as described in previous articles [7,12].

In portion of capillary blood we counted up Leukocytogram and calculated the Entropy of Leukocytogram as well as Popovych's Strain and Adaptation Indicis [7].

On the tone and motility of gall-bladder judged by its volume on an empty stomach in the morning and after 5, 15 and 30 min after ingestion cholekinetic. To quantify cholekinetics, the area between the cholecystovolumogram and the basal line was calculated [12].

The condition of microbiota is evaluated on the results of sowing of feces.

Normal (reference) values of variables are taken from the database of the Truskavetsian School of Balneology.

For statistical analysis used the software package "Statistica 64".

## RESULTS AND DISCUSSION

The effects of balneotherapy were assessed by direct differences between the final and initial Z-scores of the recorded parameters.

Screening revealed specific changes in 37 parameters grouped into 5 patterns (Figs 1 and 2).

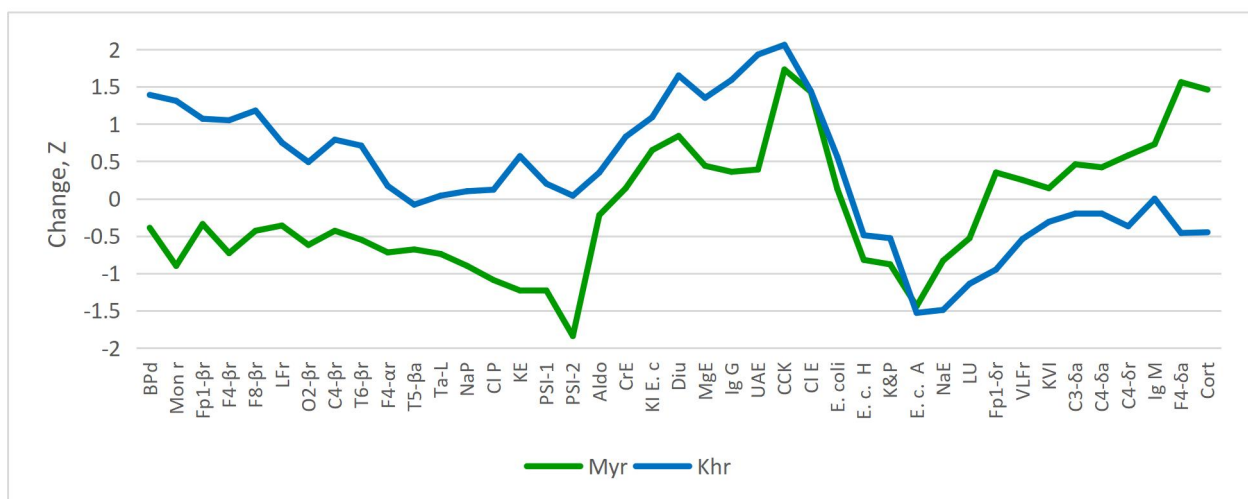
The first pattern combines 11 parameters that decrease under the influence of "Myroslava" water, while increase under the influence of "Khrystyna" water. In particular, it is power spectral density (PSD) of beta rhythm in loci Fp1, F4, F8, C4, T6 and O2 as well as LF band

of HRV; diastolic but not systolic blood pressure; plasma Aldosterone, Potassium excretion and relative level blood Monocytes.

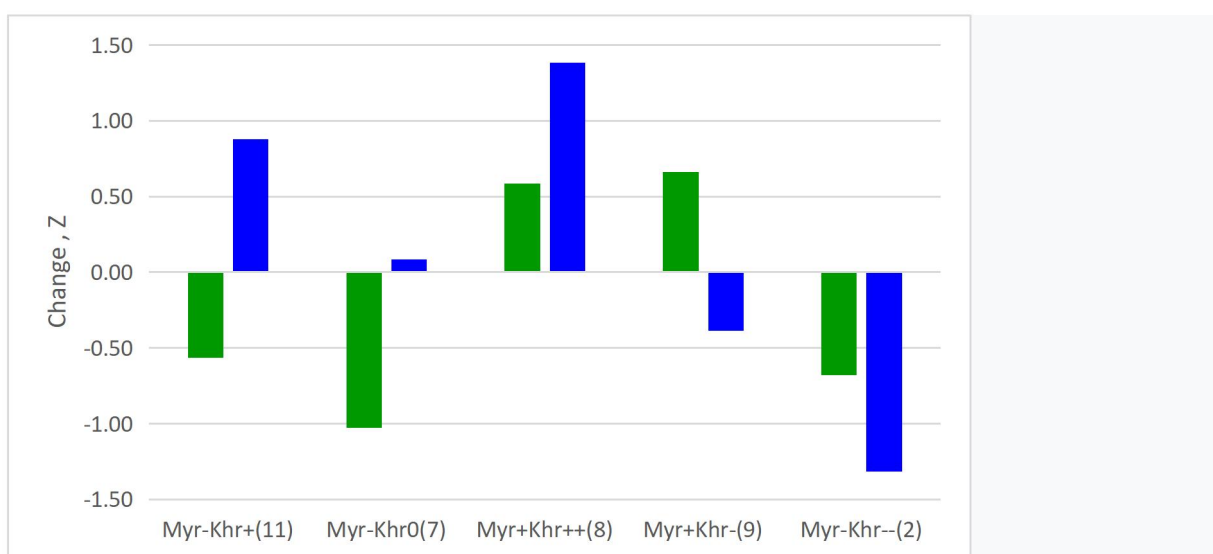
For the other 7 parameters of the second pattern (PSD T5-β and F4-α; plasma Sodium and Chloride; relative level blood T active Lymphocytes as well as both variants Popovych's leukocytary Strain Index) "Myroslava" acts similarly, while "Khrystyna" is ineffective.

Both mineral waters have a stimulating effect on 8 parameters, while "Myroslava" is inferior to "Khrystyna". In particular, it is Diuresis and urinary excretion of Creatinine, Uric acid and Magnesium; serum IgG level, completeness phagocytosis E. coli by blood Neutrophils; content in the feces of E. coli as well as Cholecystokinetics index. It should be noted that the content in the feces of E. coli strains with hemolytic and weakened enzymatic ability, as well as Klebsiela&Proteus is reduced to the same extent.

"Myroslava" water has a upregulating effect on 9 parameters of the fourth pattern, while "Khrystyna" water has a downregulating effect. In particular, it is PSD of delta rhythm in loci Fp1, F4, C3 and C4 as well as VLF band of HRV and Kerdoe's Vegetative Index; plasma Cortisol and serum IgM levels.



**Fig. 1. Profiles of specific effects of "Myroslava" and "Khrystyna" mineral waters**



**Fig. 2. Patterns of specific balneoeffects (Mean±SD) of mineral waters. In parentheses - the number of parameters**

Finally, Sodium excretion and Leukocyturia are reduced under the influence of both waters, but to a greater extent under the influence of “Khrystyna”.

However, based on the discriminant analysis [14], only 12 parameters were included in the model (Tables 1 and 2), while the rest were outside the model (Tables 3-5), apparently, as carriers of redundant/duplicate information.

**Table 1. Summary of the analysis of discriminant functions in relation to the changes in neuroendocrine, metabolic and immune parameters**

Step 12, N of vars in model: 12; Grouping: 2 grps; Wilks'  $\Lambda$ : 0,078; appr.  $F_{(13)}=8,8$ ;  $p=0,0014$

Variables currently in the model	Groups (n) and Means $\pm$ SE			Parameters of Wilks' Statistics					
	Before therapy (22)	Effect by Myros-lava (11)	Effect by Khrystyna (11)	Wilks' $\Lambda$	Partial $\Lambda$	F-remove (1,9)	p-level	Tolerance	Norm Cv
<b>F8-<math>\beta</math> PSD, %</b>	31,0 +0,11	-8,6 -0,43	+23,7 +1,18	0,296	0,265	25,01	0,001	0,116	28,7 0,702
<b>Monocytes, %</b>	6,8 +0,77	-0,9 -0,90	+1,3 +1,31	0,213	0,369	15,40	0,003	0,096	6,0 0166
<b>Uric acid Excretion, mM/24 h</b>	3,90 +1,19	+0,29 +0,39	+1,45 +1,93	0,259	0,303	20,75	0,001	0,164	3,00 0,250
<b>Corticosterone, nM/L</b>	363 -0,06	+163 +1,46	-51 -0,45	0,159	0,492	9,30	0,014	0,180	370 0,303
<b>Sodium Excretion, mM/24 h</b>	276 +3,74	-27 -0,83	-48 -1,49	0,146	0,538	7,72	0,021	0,283	154 0,211
<b>Escherichia coli feces, IgCFU/g</b>	8,33 -0,86	+0,05 +0,13	+0,22 +0,56	0,087	0,905	0,94	0,357	0,388	8,66 0,045
<b>C3-<math>\delta</math> PSD, <math>\mu V^2/Hz</math></b>	87 -0,26	+90 +0,46	-0,40 -0,20	0,083	0,950	0,48	0,507	0,243	139 1,423
<b>Creatinine Excretion, mM/24 h</b>	6,84 -1,26	+0,47 +0,14	+2,73 +0,86	0,144	0,543	7,56	0,022	0,273	11,0 0,300
<b>Killing Index vs E. coli, %</b>	48,3 -1,41	+6,3 +0,65	+10,6 +1,09	0,101	0,776	2,59	0,142	0,274	62,0 0,156
<b>VLF HRV, %</b>	46,8 -0,51	+3,4 +0,25	-8,2 -0,54	0,141	0,558	7,13	0,026	0,184	53,9 0,277
<b>Ig G Serum, g/L</b>	13,49 +0,28	+0,95 +0,36	+4,18 +1,59	0,113	0,697	3,92	0,079	0,318	12,75 0,206
<b>T5-<math>\beta</math> PSD, <math>\mu V^2/Hz</math></b>	94 +0,19	-53 -0,68	-6 -0,08	0,098	0,802	2,22	0,170	0,132	79 0,995

Notes. In the first column, the top row is the average actual value, the bottom row is the average Z-value. The second and third columns show the average direct differences between the final and initial values. In the last column, the top row is the average reference value, the bottom row is Cv or *SD*.

**Table 2. Summary of step-by-step analysis of discriminant variables ranked by criterion  $\Lambda$**

Variables currently in the model	F to enter	p-level	$\Lambda$	F-value	p-level
<b>F8-<math>\beta</math> PSD, %</b>	5,54	0,029	0,783	5,54	0,029
<b>Monocytes, %</b>	8,23	0,010	0,547	7,88	0,003
<b>Uric acid Excretion, mM/24 h</b>	5,69	0,028	0,415	8,45	0,001
<b>Corticosterone, nM/L</b>	3,21	0,091	0,349	7,92	0,001
<b>Sodium Excretion, mM/24 h</b>	4,97	0,040	0,266	8,81	0,0004
<b>Escherichia coli feces, lgCFU/g</b>	3,11	0,098	0,221	8,83	0,0003
<b>C3-<math>\delta</math> PSD, <math>\mu V^2/Hz</math></b>	1,76	0,205	0,196	8,20	0,001
<b>Creatinine Excretion, mM/24 h</b>	1,53	0,239	0,175	7,64	0,001
<b>Killing Index vs E. coli, %</b>	1,96	0,187	0,151	7,51	0,001
<b>VLF HRV, %</b>	1,86	0,200	0,129	7,43	0,001
<b>Ig G Serum, g/L</b>	3,19	0,104	0,098	8,39	0,001
<b>T5-<math>\beta</math> PSD, <math>\mu V^2/Hz</math></b>	2,22	0,170	0,078	8,82	0,001

**Table 3. Neuro-Endocrine Variables currently not in the model**

Variables	Groups (n) and Means $\pm$ SE			Parameters of Wilks' Statistics					Norm Cv/ $\sigma$
	Before therapy (22)	Effect of Myros-lava (11)	Effect of Khrystyna (11)	Wilks' $\Lambda$	Partial $\Lambda$	F to enter	p-level	Tolerance	
<b>LF HRV, %</b>	33,5 +0,92	-3,3 -0,26	+6,9 +0,75	0,076	0,969	0,25	0,629	0,055	26,1 0,312
<b>Fp1-<math>\beta</math> PSD, %</b>	30,7 +0,19	-4,7 -0,34	+14,8 +1,07	0,078	0,997	0,02	0,886	0,297	28,1 0,492
<b>Fp1-<math>\delta</math> PSD, %</b>	25,0 -0,04	+6,8 +0,35	-18,5 -0,95	0,078	0,995	0,04	0,847	0,258	25,9 0,748
<b>F4-<math>\beta</math> PSD, %</b>	28,5 +0,30	-9,8 -0,73	+14,0 +1,05	0,078	1,000	0,00	0,960	0,162	24,5 0,544
<b>F4-<math>\alpha</math> PSD, %</b>	38,4 +0,31	-13,4 -0,72	+3,2 +0,17	0,071	0,910	0,79	0,399	0,308	32,7 0,564
<b>F4-<math>\delta</math> PSD, <math>\mu V^2/Hz</math></b>	101 -0,23	+369 +1,56	-100 -0,46	0,078	1,000	0,00	0,970	0,146	155 1,521
<b>C4-<math>\delta</math> PSD, %</b>	20,2 -0,52	+10,7 +0,58	-6,9 -0,37	0,078	0,998	0,02	0,900	0,104	29,9 0,617
<b>C4-<math>\delta</math> PSD, <math>\mu V^2/Hz</math></b>	82 -0,24	+87 +0,42	-42 -0,20	0,075	0,954	0,39	0,551	0,114	131 1,593
<b>T6-<math>\beta</math> PSD, %</b>	32,0 +0,10	-10,6 -0,55	+13,9 +0,71	0,078	0,997	0,03	0,872	0,078	30,1 0,646
<b>C4-<math>\beta</math> PSD, %</b>	26,2 -0,01	-5,6 -0,43	+10,3 +0,79	0,077	0,980	0,16	0,698	0,275	26,3 0,493
<b>O2-<math>\beta</math> PSD, %</b>	24,5 +0,07	-9,5 -0,62	+7,5 +0,49	0,077	0,977	0,18	0,679	0,379	23,4 0,652
<b>Aldosterone, pM/L</b>	227 -0,25	-10 -0,22	+15 +0,35	0,078	0,996	0,03	0,863	0,439	238 0,187
<b>Kerdoe Vegetative Index, units</b>	-18 +0,28	+3 +0,14	-6 -0,31	0,078	0,998	0,02	0,900	0,104	-23 20
<b>Blood Pressure Diastolic, mmHg</b>	83,7 -0,18	-1,8 -0,39	+6,3 +1,38	0,072	0,918	0,71	0,423	0,371	84,5 0,054

**Table 4. Metabolic Variables currently not in the model**

Variables	Groups (n) and Means±SE			Parameters of Wilks' Statistics					Norm Cv/σ
	Before therapy (22)	Effect of Myros-lava (11)	Effect of Khrystyna (11)	Wilks' Λ	Partial Λ	F to enter	p-level	Tolerance	
Diuresis, L/24 h	2,19 +2,07	+0,32 +0,84	+0,63 +1,65	0,076	0,969	0,25	0,628	0,214	1,40 0,274
Potassium Excretion, mM/24 h	88 +1,31	-21 -1,23	+10 +0,57	0,074	0,947	0,45	0,522	0,423	65 0,269
Magnesium Excretion, mM/24 h	5,04 +0,89	+0,46 +0,44	+1,42 +1,35	0,078	0,998	0,02	0,900	0,104	4,10 0,256
Chloride Excretion, mM/24 h	217 +1,73	+41 +1,43	+41 +1,44	0,078	1,000	0,00	0,960	0,162	167,5 0,172
Chloride Plasma, mM/L	102,9 +0,42	-3,6 -1,09	+0,4 +0,12	0,071	0,910	0,79	0,399	0,308	101,5 0,032
Sodium Plasma, mM/L	144,3 -0,15	-4,5 -0,90	+0,5 +0,10	0,072	0,918	0,71	0,423	0,371	145,0 0,034
Cholecystokinetic Index, units	519 -1,28	+142 +1,73	+168 +2,06	0,078	1,000	0,00	0,960	0,162	624 0,131

**Table 5. Immune and Microbiota Variables currently not in the model**

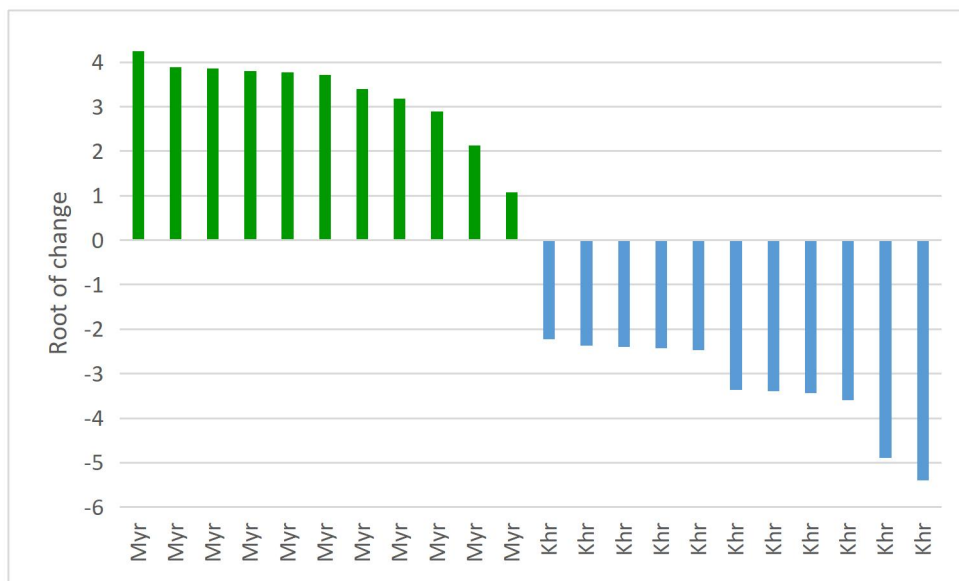
Variables	Groups (n) and Means±SE			Parameters of Wilks' Statistics					Norm Cv/σ
	Before therapy (22)	Effect of Myros-lava (11)	Effect of Khrystyna (11)	Wilks' Λ	Partial Λ	F to enter	p-level	Tolerance	
CD3 <sup>+</sup> active T-Lymphocytes, %	27,9 -0,43	-3,7 -0,74	+0,2 +0,04	0,075	0,952	0,40	0,544	0,627	30,0 0,167
Ig M Serum, g/L	1,35 +0,71	+0,20 +0,73	0,00 0,00	0,076	0,969	0,25	0,628	0,214	1,15 0,239
Popovych Strain Index-1	0,162 +1,18	-0,068 -1,23	+0,011 +0,20	0,074	0,947	0,45	0,522	0,423	0,097 0,559
Popovych Strain Index-2	0,206 +2,43	-0,101 -1,84	+0,002 +0,04	0,074	0,947	0,45	0,522	0,423	0,072 0,762
E. coli hemolytica feces, %	20 0,82	-20 -0,82	-12 -0,49	0,074	0,945	0,46	0,515	0,382	0 25
E. coli attenuated feces, %	58,1 +2,34	-25,0 -1,44	-26,5 -1,53	0,078	0,998	0,02	0,900	0,104	17,4 1,000
Klebsiela&Proteus feces, %	9,7 +0,88	-9,6 -0,88	-5,9 -0,53	0,071	0,904	0,85	0,384	0,140	0 11
Leukocyturia, IgLeu/L	3,29 +0,58	-0,26 -0,53	-0,57 -1,14	0,078	0,995	0,04	0,847	0,258	3,00 0,070

Calculation of individual values of the discriminant root by raw coefficients and constant (Table 6) allows you to visualize the balneoeffects of mineral waters for each patient (Fig. 3).

**Table 6. Standardized and raw coefficients and constants for discriminant variables**

Coefficients	Standardized	Raw
<b>Variables</b>	Root 1	Root 1
<b>F8-β PSD, %</b>	-2,618	-0,128
<b>Monocytes, %</b>	-2,677	-1,091
<b>Uric acid Excretion, mM/24 h</b>	-2,151	-1,643
<b>Corticosterone, nM/L</b>	-1,751	-4,038
<b>Sodium Excretion, mM/24 h</b>	-1,331	-0,012
<b>Escherichia coli feces, lgCFU/g</b>	0,515	1,845
<b>C3-δ PSD, μV<sup>2</sup>/Hz</b>	-0,474	-0,004
<b>Creatinine Excretion, mM/24 h</b>	1,347	0,361
<b>Killing Index vs E. coli, %</b>	-0,941	-0,072
<b>VLF HRV, %</b>	1,616	0,129
<b>Ig G Serum, g/L</b>	-1,018	-0,189
<b>T5-β PSD, μV<sup>2</sup>/Hz</b>	1,273	0,031
	<b>Constant</b>	3,992
	<b>Eigenvalue</b>	11,75
<b>Squared Mahalanobis Distance=43; F=8,8; p=0,0014</b>		
<b>Canonical R=0,960; Wilks' Λ=0,0784; χ<sup>2</sup>(12)=37; p=0,0004</b>		

Opposite root values of patients receiving different mineral waters reflect (Table 7), first, an increase under the influence of “Myroslava” water versus decrease under the influence of “Khrystyna” water in initially normal Cortisol levels and electrical activity of delta-rhythm-generating neurons projected at locus C3 (probably the left hippocampus) [27]) and moderately reduced relative power of VLF components of HRV. As noted Shaffer F & Ginsberg JP [28], there is uncertainty regarding the physiological mechanisms responsible for activity within the VLF (0,04÷0,0033 Hz) band. The heart’s intrinsic nervous system appears to contribute to the VLF rhythm and the sympathetic nervous system influences the amplitude and frequency of its oscillations. VLF power may also be generated by physical activity, thermoregulatory, renin–angiotensin, and endothelial influences on the heart. Vagal activity may contribute to VLF power since parasympathetic blockade almost completely abolishes it. In contrast, sympathetic blockade does not affect VLF power. The VLF rhythm appears to be generated by the stimulation of afferent sensory neurons in the heart. This, in turn, activates various levels of the feedback and feed-forward loops in the heart’s intrinsic cardiac nervous system, as well as between the heart, the extrinsic cardiac ganglia, and spinal column. This experimental evidence suggests that the heart intrinsically generates the VLF rhythm and efferent sympathetic nervous system activity due to physical activity and stress responses modulates its amplitude and frequency.



**Fig. 3. Individual values of the discriminant root of balneoeffects**

**Table 7. Correlations between variables and root and Z-scores of changes in variables**

Variables	R	Effect of Myroslava	Effect of Khrystyna
VLF HRVr	0,142	+0,25	-0,54
C3- $\delta$ PSDa	0,114	+0,46	-0,20
Corticosterone	0,015	+1,46	-0,45
Sodium Excretion	0,031	-0,83	-1,49
F8- $\beta$ PSDr	-0,153	-0,43	+1,18
T5- $\beta$ PSDa	-0,112	-0,68	-0,08
Monocytes	-0,137	-1,80	+2,62
Uric acid Excretion	-0,135	+0,39	+1,93
Creatinine Excretion	-0,093	+0,14	+0,83
Ig G Serum	-0,092	+0,36	+1,59
Killing Index vs E. coli	-0,050	+0,65	+1,09
Escherichia coli feces	-0,078	+0,13	+0,56

Second, “Myroslava” water reduces increased Sodium excretion to a lesser extent than “Khrystyna” water.

Third, “Myroslava” water inhibits the initially normal activity of beta-rhythm-generating neurons projected at the F8 and T5 loci, whereas “Khrystyna” water is excitatory or neutral, respectively. It is more likely that the locus F8 record the activity of right anterior cingulate cortex and the locus T5 projected left caudal anterior cingulate cortex. These cortical structures affect the activity of the vagal and sympathetic nuclei [29,30]. In the same opposite way, mineral waters affect the increased relative level of blood monocytes.

Fourth, “Khrystyna” water significantly increases the initially reduced bactericidal activity of neutrophils against E. coli and its content in feces, as well as Creatinine excretion. In addition, there is an increase in normal IgG levels and further increase in Hyperuricosuria. On the other hand, “Myroslava” water affects these parameters to a much lesser extent.

The visual impression of a very clear delineation of the effects of both mineral waters is documented by the calculation of Mahalanobis Distance (Table 6), as well as 100% accuracy of classification based on the coefficients and constants given in Table. 8.



**Table 8. Coefficients and constants of classification functions**

Clusters	Effect of Myroslava	Effect of Khrystyna
Variables	p=,500	p=,500
<b>F8-β PSD, %</b>	0,120	0,957
<b>Monocytes, %</b>	0,365	7,497
<b>Uric acid Excretion, mM/24 h</b>	2,013	12,76
<b>Corticosterone, nM/L</b>	2,181	28,58
<b>Sodium Excretion, mM/24 h</b>	-0,002	0,079
<b>Escherichia coli feces, lgCFU/g</b>	-5,650	-17,71
<b>C3-δ PSD, μV<sup>2</sup>/Hz</b>	0,004	0,031
<b>Creatinine Excretion, mM/24 h</b>	-0,430	-2,793
<b>Killing Index vs E. coli, %</b>	0,116	0,587
<b>VLF HRV, %</b>	-0,191	-1,037
<b>Ig G Serum, g/L</b>	0,354	1,588
<b>T5-β PSD, μV<sup>2</sup>/Hz</b>	-0,087	-0,291
<b>Constants</b>	-2,75	-28,8

It should be noted that the direction of balneal effects is not fully consistent with the previously proposed concept about ambivalence-equilibratory character of influence of curative water Naftussya on organism of human [2]. Indeed, along with the increase in decreased and decrease in increased parameters, other variants of effects were found, usually physiologically favorable, except for the increase in diastolic pressure under the influence of "Khrystyna" water.

To determine the mechanism of the detected effects of mineral waters, the discriminant parameters were divided into two sets. The first set consisted of neuroendocrine parameters as causal, and the second set - parameters of immunity, metabolism, microbiota, cholekinetics and diastolic pressure, which, apparently, are subject to regulatory effects of the former (Table 9).

**Table 9. Matrix of correlations between changes in Neuro-Endocrine and others parameters**

Variables	KI EC	Mon	Pop SI-1	CD3 TaL	Ig G	Ig M	Cr Ex	UA Ex	Na Ex	CCK Ind	Mg Ex	BP d	Ig LU	Ig EC
LF r	<b>-0,42</b>	<b>0,43</b>	0,41	0,05	0,03	0,12	0,41	0,14	-0,34	0,40	-0,15	0,12	<b>0,43</b>	-0,34
VLF r	-0,02	-0,34	-0,34	-0,23	0,14	-0,03	-0,29	-0,08	0,35	<b>-0,46</b>	0,28	-0,07	-0,14	-0,15
T5-β a	-0,27	0,08	0,18	-0,07	-0,08	-0,22	0,14	0,36	-0,39	<b>0,63</b>	0,03	0,18	0,35	-0,31
C3-δ a	-0,20	-0,04	-0,17	-0,03	-0,07	0,36	-0,22	-0,29	0,25	-0,53	<b>-0,49</b>	-0,18	0,03	0,04
Cortisol	0,02	<b>-0,59</b>	-0,34	-0,23	-0,15	-0,04	0,01	-0,04	-0,20	0,35	-0,17	-0,16	-0,00	-0,06
F8-β r	0,12	-0,12	0,17	-0,06	-0,23	<b>-0,45</b>	0,11	0,26	<b>-0,44</b>	0,36	0,28	0,32	-0,04	0,01
Aldost	-0,04	0,00	0,03	-0,12	0,07	-0,02	0,35	0,03	-0,15	-0,04	-0,05	0,03	0,15	-0,09

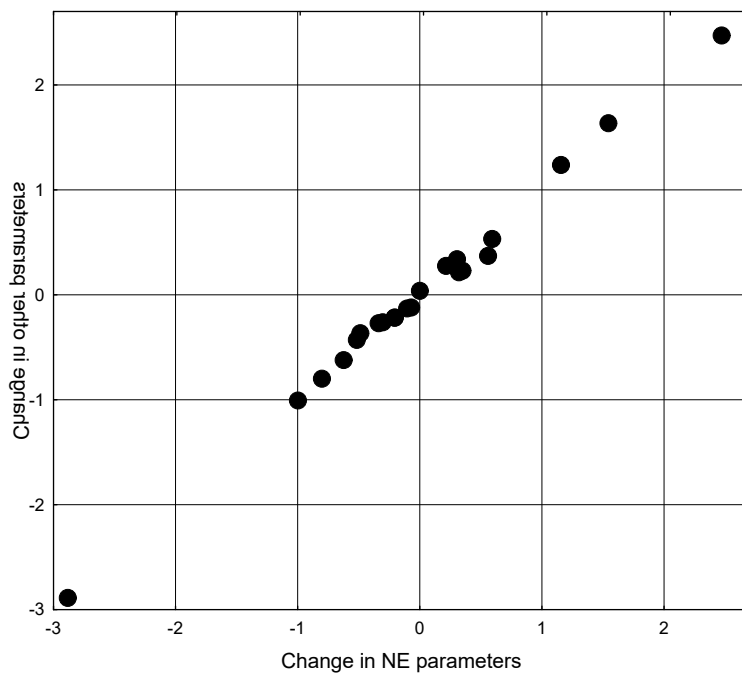
The canonical correlation between the two sets of parameters was then analyzed. Two pairs of canonical roots are distinguished.

The factor structure of the first neuroendocrine root (Table 10) is formed by: LF component of HRV (reflects both sympathetic and vagal effects), VLF band and beta-rhythm-generating nerve structures (their physiological essence is described above), as well as cortisol and delta-rhythm-generating nerve structures projected at locus C3 (probably the left hippocampus [27]).

Changes in the mentioned neuro-endocrine factors determine the changes in the parameters of the subordinate set by 99,5% (Fig. 4).

**Table 10. Factor load on first pair of canonical roots**

Neuro-Endocrine Variables	R 1
LF, %	-0,823
T5 $\beta$ , PSD, $\mu\text{V}^2/\text{Hz}$	-0,536
VLF, %	0,559
Cortisol	0,489
C3- $\delta$ PSD, $\mu\text{V}^2/\text{Hz}$	0,175
Other Variables	R 1
Monocytes, %	-0,643
Popovych's Strain Index-1	-0,469
Leukocyturia, IgLeu/L	-0,435
Creatinine Excretion, mM/24 h	-0,354
<b>Cholecystokinetic Index, units</b>	-0,298
Ig G Serum, g/L	-0,211
Uric acid Excretion, mM/24 h	-0,200
Blood Pressure Diastolic, mmHg	-0,126
Ig M Serum, g/L	-0,124
CD3 <sup>+</sup> active T-Lymphocytes, %	-0,121
Killing Index vs E. coli, %	0,421
Escherichia coli feces, IgCFU/g	0,375
Sodium Excretion, mM/24 h	0,141



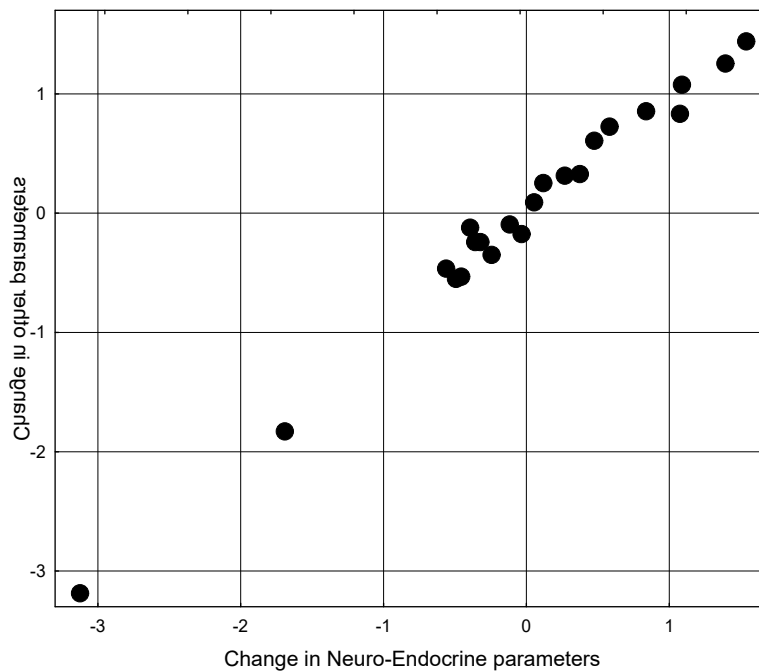
$R=0,997$ ;  $R^2=0,995$ ;  $\chi^2_{(91)}=155$ ;  $p<10^{-4}$ ;  $\Lambda \text{ Prime}<10^{-6}$

**Fig. 4. Scatterplot of canonical correlation between change in Neuro-Endocrine (X-line) and other (Y-line) parameters. First pair of Roots**

The factor structure of the neuroendocrine root of the second pair (Table 11) is supplemented by two parameters (F8- $\beta$  PSD and aldosterone). Their changes determine changes in the parameters of cholekinetics, metabolism, microbiota and immunity by 98,6% (Fig. 5).

**Table 11. Factor load on second pair of canonical roots**

Neuro-Endocrine Variables	R 2
<b>F8-β PSD, %</b>	<b>0,710</b>
<b>T5 β, PSD, μV<sup>2</sup>/Hz</b>	<b>0,674</b>
<b>Cortisol</b>	<b>0,669</b>
<b>LF, %</b>	<b>0,307</b>
<b>Aldosterone</b>	<b>0,125</b>
<b>C3-δ PSD, μV<sup>2</sup>/Hz</b>	<b>-0,574</b>
<b>VLF, %</b>	<b>-0,189</b>
Other Variables	R 2
<b>Cholecystokinetic Index, units</b>	<b>0,615</b>
<b>Uric acid Excretion, mM/24 h</b>	<b>0,216</b>
<b>Leukocyturia, lgLeu/L</b>	<b>0,163</b>
<b>Blood Pressure Diastolic, mmHg</b>	<b>0,130</b>
<b>Creatinine Excretion, mM/24 h</b>	<b>0,103</b>
<b>Sodium Excretion, mM/24 h</b>	<b>-0,466</b>
<b>Monocytes, %</b>	<b>-0,330</b>
<b>Escherichia coli feces, lgCFU/g</b>	<b>-0,272</b>
<b>Ig M Serum, g/L</b>	<b>-0,238</b>
<b>Ig G Serum, g/L</b>	<b>-0,209</b>
<b>CD3<sup>+</sup> active T-Lymphocytes, %</b>	<b>-0,197</b>
<b>Killing Index vs E. coli, %</b>	<b>-0,182</b>



**R=0,993; R<sup>2</sup>=0,986;  $\chi^2_{(72)}=100$ ; p=0,017;  $\Lambda$  Prime<10<sup>-4</sup>**

**Fig. 5. Scatterplot of canonical correlation between change in Neuro-Endocrine (X-line) and other (Y-line) parameters. Second pair of Roots**

## CONCLUSION

Thus, the mineral waters "Myroslava" and "Khrystyna" have both similar [12,13] and different effects on the body. It is important that the differences are manifested not only in the severity of changes in the registered parameters, but also in their direction. Since the chemical composition of both mineral waters is qualitatively identical, the difference in physiological effects is obviously due to their total mineralization (5 g/L and 10 g/L, respectively). In addition, it should be borne in mind that our patients used this water in combination with biologically active water Naftussya. However, the specificity of physiological effects was confirmed in an experiment on rats [1,9,10,11,24].

Based on the data of our previous studies [5,15-23,25,26] and presented in this article, we hypothesize that the primary effect of mineral waters is the modulation of the structures of the autonomic and central nervous and endocrine systems, which, in turn, have regulatory modulating effects on the immune system, microbiota, metabolism, cholekinetics, blood pressure and, apparently, others, not yet registered body parameters.

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## ACCORDANCE TO ETHICS STANDARDS

Tests in patients are carried out 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.

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