

## RELATIONSHIPS BETWEEN THE ENTROPIES OF EEG, HRV, IMMUNOCYTOGRAM AND LEUKOCYTOGRAM

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**Background.** In mathematics the entropy is a measure of uncertainty of a random function; in the theory of information entropy is a measure of uncertainty in a situation, any experience (test) that can have different consequences. The entropy is also a measure of disorder, the degree of chaos present in the system. CE Shannon linked the mathematical dependence of the concept of information and entropy, which characterizes the degree of ordering of the system. This estimate of the amount of information coincides with the estimation of the quantitative measure of elimination of uncertainty of entropy, the degree of organization of the system. It is well known about functional interactions between central and autonomic nervous and immune systems. In the context of this concept, we have prioritized research on the interconnections between the entropies of these systems. **Material and methods.** In basal conditions in 37 men and 14 women with chronic pyelonephritis and cholecystitis in remission as well as without clinical diagnose but with dysfunction of neuro-endocrine-immune complex and metabolism, we recorded twice, before and after balneotherapy at the spa Truskavets', EEG ("NeuroCom Standard") and HRV ("Cardiolab+VSR"). In blood we determined relative content of components (RCC) of Immunocytogram (ICG) (T helper, T cytolytic, B and NK lymphocytes) and Leukocytogram (LCG) (Eosinophils, Stab and Segmentonuclear Neutrophils, Lymphocytes and Monocytes). Than we calculated for each locus of EEG and HRV as well as for ICG and LCG the Entropy (h) of normalized spectral power density (SPD) or RCC using Shannon's formula: **Results.** There was a complete absence of correlation between hHRV and hLCG ( $r=-0,03$ ) as well as hICG ( $r=-0,03$ ), whereas the relationship between hLCG and hICG is significant ( $r=-0,40$ ;

$p < 0,001$ ). Accepting the entropy of EEG HRV as a factor, using the correlation analysis with step-by-step exclusion, we obtain the equations for dependent variables. Canonical correlation between hHRV&EEG, on the one hand, and hLCG&Immunity, on the other hand, is strong:  $R=0,814$ ;  $R^2=0,663$ ;  $\chi^2_{(240)}=296$ ;  $p=0,008$ . **Conclusion.** The entropy of HRV and EEG significantly correlate with the entropy and parameters of immunity, which testifies to their modulating regulatory effects.

**Keywords:** EEG, HRV, Leukocytogram, Immunocytogram, Entropy, Correlations, Women and Man.

## INTRODUCTION

In physics, entropy is a quantity which, in observed phenomena and processes, characterizes the devaluation (scattering) of energy due to the transformation of all its types into heat with a uniform distribution of heat between bodies; in chemistry and thermodynamics it is a measure of the amount of energy in the physical system, which can not be used to perform work; in mathematics it is a measure of uncertainty of a random function; in the theory of information entropy is a measure of uncertainty in a situation, any experience (test) that can have different consequences. The entropy is also a measure of disorder, the degree of chaos present in the system. CE Shannon [27] linked the mathematical dependence of the concept of information and entropy, which characterizes the degree of ordering of the system. This estimate of the amount of information coincides with the estimation of the quantitative measure of elimination of uncertainty of entropy, the degree of organization of the system.

According to PV Biloshytskyi [5,6], the mathematical formula directly indicates the possibility of quantitative change in information to change the ordering of the system, which, in relation to biosystems, can mean a change in quality (stability, workability, health, etc.), and thus indicate the path of purposeful use of bioinformatics in medical practice. Accordingly, the author suggests that instead of the term entropy, we use the term of the **reliability of the functioning of the organism**, which is very impressive to us, as well as his assumption that the dependence of the reliability of the biosystem on information is precisely the elusive **vis vitalis** (life force).

The calculation of entropy is acceptable, in particular, with respect to closed systems of various shaped elements, such as leukocytogram, immune-, spleno- and thymocytogram. Information analysis of cytograms allows us to assess the state of morpho-functional adaptive-protective systems, information about which contained in their cytograms [1,22,29].

Other objects for calculating entropy are the normalized spectral power density of the HRV and EEG [15-17].

It is well known about functional interactions between central and autonomic nervous and immune systems [14-17,23,24,28]. In the context of this concept, we have prioritized research on the interconnections between the entropies of these systems.

## MATERIAL AND METHODS

The object of observation were 37 men and 14 women aged 23-76 years old, who came to the spa Truskavets' (Ukraine) for the treatment of chronic pyelonephritis and cholecystitis in remission as well as without clinical diagnose but with dysfunction of neuro-endocrine-immune complex and metabolism. The survey was conducted twice, before and after weekly balneotherapy.

We recorded electrocardiogram in II lead (hardware-software complex "CardioLab+HRV" produced by "KhAI-MEDICA", Kharkiv, Ukraine) to assess the parameters of heart rate variability (HRV). For further analysis (Frequency Domain Methods) were selected spectral power (SP) bands 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) [2,4,12].

Simultaneously we recorded EEG (hardware-software complex "NeuroCom Standard", KhAI Medica, Kharkiv, Ukraine) monopolar in 16 loci (Fp1, Fp2, F3, F4, F7, F8, C3, C4, T3, T4, P3, P4, T5, T6, O1, O2) by 10-20 international system, with the reference electrodes A and Ref on the tassels of ears. Among the options considered the average EEG amplitude ( $\mu\text{V}$ ), average frequency (Hz), frequency deviation (Hz), index (%), coefficient of asymmetry (%) as well as absolute ( $\mu\text{V}^2/\text{Hz}$ ) and relative (%) spectral power density (SPD) in the standard frequency bands:  $\beta$  (35÷13 Hz),  $\alpha$  (13÷8 Hz),  $\theta$  (8÷4 Hz) and  $\delta$  (4÷0,5 Hz) in all loci, according to the instructions of the device.

In addition, calculated Laterality Index (LI) for SPD each Rhythm using formula [19]:

$$\text{LI, \%} = \Sigma [200 \cdot (\text{Right} - \text{Left}) / (\text{Right} + \text{Left})] / 8$$

We calculated also for HRV and each locus EEG the Entropy (h) of normalized SPD using formula CE Shannon [27]:

$$\text{hHRV} = - [\text{SPDHF} \cdot \log_2 \text{SPDHF} + \text{SPDLF} \cdot \log_2 \text{SPDLF} + \text{SPDVLF} \cdot \log_2 \text{SPDVLF} + \text{SPDULF} \cdot \log_2 \text{SPDULF}] / \log_2 4;$$

$$\text{hEEG} = - [\text{SPD}\alpha \cdot \log_2 \text{SPD}\alpha + \text{SPD}\beta \cdot \log_2 \text{SPD}\beta + \text{SPD}\theta \cdot \log_2 \text{SPD}\theta + \text{SPD}\delta \cdot \log_2 \text{SPD}\delta] / \log_2 4$$

In portion of capillary blood we counted up Leukocytogram (LCG) (Eosinophils, Stub and Segmentonuclear Neutrophils, Lymphocytes and Monocytes) and calculated its Adaptation Index as well as Strain Index by IL Popovych [7,13,21].

The informativeness of these indices was demonstrated by other authors [3,20], as well as its advantage over the Lymphocytes/Segmentonuclear Neutrophils ratio [10,25].

We remind that the algorithm of quantization of the Popovych's indexes is based on the proposed LKh Garkavi et al [9,10] ranges of relative content in the leukocytogram of lymphocytes, which determines the type of General Adaptation Reaction of Organism as well as other components of leukocytogram and total leukocyte levels indicating harmonic or disharmonious character of GARO.

Leukocytogram Lymphocytes level, %	General Adaptation Reaction of Organism	Eosinophiles and Stub Neutrophiles: 1÷6 %; Monocytes: 4÷7 %; Leukocytes: 4÷8 G/l	Eosinophiles and Stub Neutrophiles: <1; >6; Monocytes: <4; >7; Leukocytes: <4; >8 G/l
<21	Stress	1,22	0,02
21÷27	Training	1,46	0,74
28÷33	Quiet Activation	1,95	0,98
34÷43,5	Heightened Activation	1,70	0,50
≥44	Superactivation		0,26

$$\text{Strain Index-1} = [(\text{Eo}/3,5-1)^2 + (\text{SN}/3,5-1)^2 + (\text{Mon}/5,5-1)^2 + (\text{Leu}/6-1)^2] / 4$$

Later, LKh Garkavi et al [11] proposed some other boundaries of ranges, on the basis of which we calculated the second version of the indices, testing of which will debuted in this article.

Leukocyto-gram Lymphocytes level, %	General Adaptation Reaction of Organism	Eosinophiles: 1÷4,5 %; Stub Neutrophiles: 3÷5,5 %; Monocytes: 5÷7 %; Leukocytes: 4÷6 G/l	Eosinophiles: <1; >4,5% Stub Neutrophiles: <3; >5,5; Monocytes: <5; >7; Leukocytes: <4; >6 G/l
<21	Stress	1,22	0,02
21÷27	Training	1,46	0,74
28÷33	Quiet Activation	1,95	0,98
34÷43,5	Heightened Activation	1,70	0,50
≥44	Superactivation		0,26

$$\text{Strain Index-2} = [(Eo/2,75-1)^2 + (SN/4,25-1)^2 + (Mon/6-1)^2 + (Leu/5-1)^2]/4$$

Immune status evaluated on a set of I and II levels recommended by the WHO as described in the manual [18]. 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 (by polyethylene glycol precipitation method).

We calculated also the Entropy (h) of Immunocytogram (ICG) and Leukocytogram (LCG) using classical CE Shannon's formula [27]:

$$h_{ICG} = - [CD4 \cdot \log_2 CD4 + CD8 \cdot \log_2 CD8 + CD22 \cdot \log_2 CD22 + CD16 \cdot \log_2 CD16] / \log_2 4$$

$$h_{LCG} = - [Lymph \cdot \log_2 Lymph + Mon \cdot \log_2 Mon + Eos \cdot \log_2 Eos + SNN \cdot \log_2 SNN + StubN \cdot \log_2 StubN] / \log_2 5$$

Parameters of phagocytic function of neutrophils estimated as described by SD Douglas and PG Quie [8] with moderately modification by MM Kovbasnyuk [16]. 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°C. 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 shaken and placed in thermostat at 37°C 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 as percentage of neutrophils, in which found microbes - Hamburger's Phagocytic Index; intensity as number of microbes absorbed one phagocytes - Microbial Count (MC) or Right's Index; completeness as percentage of dead microbes - Killing Index (KI). Microbial number and index their digestion is determined for each phagocyte and fixed in phagocytic frame.

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

## RESULTS AND DISCUSSION

In the first stage, screening of correlation relationships between the levels of entropy of HRV and loci of EEG, on the one hand, and the entropy of LCG and ICG, as well as immune parameters, was performed on the other.

According to calculations by the formula [23]:

$$|r| = \frac{\exp[2t/(n-1,5)^{0,5}] - 1}{\exp[2t/(n-1,5)^{0,5}] + 1}$$

for a sample of n=102 critical value |r| at p<0,05 (t>2,00) is 0,20, at p<0,02 (t>2,39) is 0,23, at p<0,01 (t>2,66) is 0,26, at p<0,001 (t>3,46) is 0,33.

Based on the results of the screening, a matrix (Table 1) is created, which includes only those parameters that have at least one significant correlation with the parameters of another set. As we see, the locus of F8 and P4, on the one hand, and Phagocytic Index, Stub Neutrophils, Monocytes, CD22<sup>+</sup> B Lymphocytes, IgG, on the other hand, were located outside the matrix.

There was a complete absence of correlation between entropies of HRV and LCG as well as ICG, whereas the relationship between entropies of LCG and ICG is maximal for matrix (r=-0,40; p<0,001).

**Table 1. Matrix of Correlations, left set with right set**

0,05|r|≥0,20; 0,02|r|≥0,23; 0,01|r|≥0,26; 0,001|r|≥0,33

	HRV H	Fp1 H	Fp2 H	F3 H	F4 H	F7 H	T3 H	T4 H	C3 H	C4 H	T5 H	T6 H	P3 H	O1 H	O2 H
LCGH	-,03	-,07	-,12	,19	,09	-,09	-,03	-,03	,12	,03	-,23	-,17	-,22	-,31	-,20
ICGH	-,03	,09	,15	-,16	-,10	,16	-,07	-,04	-,13	-,03	,20	,22	,22	,13	,18
MC Sa	-,12	,21	,24	,23	,23	,18	,14	,08	,13	,27	,10	,16	,02	,13	,09
KI Sa	,28	-,14	-,29	-,09	-,06	-,20	-,23	-,21	-,08	-,09	-,16	-,22	-,19	-,20	-,23
MC Ec	-,27	,25	,23	,28	,27	,18	,13	,10	,18	,20	,10	,17	,11	,16	,00
KI Ec	,27	-,07	-,04	,03	-,09	-,11	-,12	-,05	-,07	-,05	-,08	-,01	-,23	-,12	-,14
Leukoc	-,18	,09	-,00	,18	,19	,02	,03	,01	,09	,07	-,10	-,07	-,11	-,21	-,11
SNN	-,25	-,03	,06	-,25	-,24	,09	-,02	-,01	-,24	-,17	,12	,14	,24	,15	,19
Eosin	-,21	-,10	-,07	,01	-,00	,03	-,04	-,01	,00	,01	-,11	-,02	-,20	-,18	-,15
Lymph	,36	,06	-,05	,21	,24	-,09	,03	,01	,23	,18	-,03	-,08	-,20	-,01	-,15
CD4	-,03	-,02	,01	-,23	-,18	,11	-,10	-,09	-,11	-,07	,10	,13	,17	,01	,08
CD8	,17	,16	,14	,02	,16	,06	,07	,11	,08	,20	,20	,18	,09	,03	,11
Ta	-,13	,07	,17	-,02	-,16	,21	-,08	,01	-,19	-,10	,01	,07	,08	,12	,18
CIC	,04	-,11	-,07	-,17	-,28	-,16	-,06	,00	-,06	-,16	-,06	-,06	-,05	-,14	-,09
IgA	-,10	-,07	,00	-,08	-,20	,12	,02	-,03	-,23	-,16	-,06	,09	-,00	,04	,05
IgM	,09	,14	,14	,13	,04	,11	,08	,09	,25	,17	,12	,11	,10	,17	,11
CD16	-,04	,02	,12	-,26	-,22	,17	-,04	-,04	-,18	-,07	,16	,25	,21	,08	,20
PSI-1	-,18	,00	,07	,05	,10	,04	,07	,10	,12	,09	-,07	-,09	,01	-,23	-,10
PSI-2	-,23	-,03	,04	,03	,07	,04	,07	,09	,10	,08	-,06	-,07	-,07	-,26	-,17
PAI-1	-,04	-,28	-,28	-,12	-,29	-,05	-,01	-,17	-,16	-,17	-,03	-,14	-,01	-,08	-,05
PAI-2	-,21	-,07	-,14	,06	-,11	-,06	,10	-,09	,01	-,07	,04	-,11	,20	-,04	,03

At the second stage, coefficients of the multiplicity correlation of the entropy indices with each other (Tables 2-4) and immunity parameters (Tables 5-19) were calculated on the basis of a regression model with step-by-step exclusion to reach the maximum Adjusted  $R^2$ .

**Table 2. Regression Summary for HRVH**

$R=0,268$ ;  $R^2=0,072$ ; Adjusted  $R^2=0,049$ ;  $F_{(2,8)}=3,2$ ;  $p=0,046$

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(83)}$	p-level
Variable	r		Intercpt	,940	,088	10,7	$10^{-6}$
Fp2H	-0,21	-,169	,109	-,141	,091	-1,55	,126
F3H	-0,21	-,170	,109	-,130	,084	-1,55	,124

**Table 3. Regression Summary for ICGH**

$R=0,346$ ;  $R^2=0,120$ ; Adjusted  $R^2=0,087$ ;  $F_{(3,8)}=3,7$ ;  $p=0,015$

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(82)}$	p-level
Variable	r		Intercpt	,894	,045	20,1	$10^{-6}$
T6H	0,22	,183	,112	,058	,036	1,64	,106
P3H	0,22	,198	,113	,086	,049	1,75	,084
F3H	-0,16	-,228	,106	-,080	,037	-2,14	,035

**Table 4. Regression Summary for LCGH**

$R=0,403$ ;  $R^2=0,162$ ; Adjusted  $R^2=0,142$ ;  $F_{(2,8)}=8,0$ ;  $p=0,0006$

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(83)}$	p-level
Variable	r		Intercpt	,669	,030	22,0	$10^{-6}$
O1H	-0,31	-,360	,102	-,100	,028	-3,53	,001
F3H	0,19	,257	,102	,077	,031	2,51	,014

**Table 5. Regression Summary for Fp1H**

$R=0,362$ ;  $R^2=0,131$ ; Adjusted  $R^2=0,110$ ;  $F_{(2,8)}=6,3$ ;  $p=0,003$

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(83)}$	p-level
Variable	r		Intercpt	,5987	,1416	4,23	$10^{-4}$
Microbian Count vs E. coli	0,25	,226	,103	,0045	,0021	2,20	,031
Popovych's Adaptation Index-1	-0,28	-,263	,103	-,0692	,0271	-2,56	,012

**Table 6. Regression Summary for Fp2H**

$R=0,470$ ;  $R^2=0,220$ ; Adjusted  $R^2=0,182$ ;  $F_{(4,8)}=5,7$ ;  $p<10^{-3}$

		Beta	St. Err. of Beta	B	St. Err. of B	$t_{(81)}$	p-level
Variable	r		Intercpt	,8547	,1842	4,64	$10^{-5}$
Killing Index vs Staphyl. aureus	-0,29	-,287	,100	-,0052	,0018	-2,87	,005
Popovych's Adaptation Index-1	-0,28	-,289	,099	-,0734	,0252	-2,91	,005
Microbian Count vs Staph. aur.	0,24	,160	,100	,0030	,0019	1,60	,114
Immunoglobuline M	0,14	,134	,098	,0705	,0518	1,36	,177

**Table 7. Regression Summary for F3H**R=0,444; R<sup>2</sup>=0,197; Adjusted R<sup>2</sup>=0,157; F<sub>(4,8)</sub>=5,0; p=0,001

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(81)</sub>	p-level
<b>Variable</b>	r		Intercpt	,1954	,2813	,69	,489
Microbian Count vs E. coli	0,28	,314	,101	,0066	,0021	3,11	,003
Entropy of Leukocytogramm	0,19	,180	,102	,5989	,3401	1,76	,082
CD4 <sup>+</sup> T-helper Lymphocytes	-0,23	-,221	,104	-,0046	,0021	-2,13	,036
Circulating Immune Complexes	-0,17	-,131	,102	-,0013	,0010	-1,29	,200

**Table 8. Regression Summary for F4H**R=0,529; R<sup>2</sup>=0,280; Adjusted R<sup>2</sup>=0,215; F<sub>(7,8)</sub>=4,3; p<10<sup>-3</sup>

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(78)</sub>	p-level
<b>Variable</b>	r		Intercpt	,5714	,1915	2,98	,004
Popovych's Adaptation Index-1	-0,29	-,170	,103	-,0491	,0298	-1,65	,103
Circulating Immune Complexes	-0,28	-,197	,098	-,0021	,0011	-2,02	,047
CD16 <sup>+</sup> NK Lymphocytes	-0,22	-,164	,124	-,0048	,0037	-1,32	,189
T Active Lymphocytes	-0,16	-,157	,109	-,0052	,0036	-1,44	,153
Microbian Count vs E. coli	0,27	,235	,101	,0052	,0022	2,33	,022
Leukocytes	0,19	,128	,101	,0212	,0167	1,27	,207
CD8 <sup>+</sup> T-cytolytic Lymphocytes	0,16	,175	,109	,0055	,0035	1,60	,114

**Table 9. Regression Summary for F7H**R=0,333; R<sup>2</sup>=0,111; Adjusted R<sup>2</sup>=0,067; F<sub>(4,8)</sub>=2,5; p=0,047

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(81)</sub>	p-level
<b>Variable</b>	r		Intercpt	,3465	,2121	1,63	,106
T Active Lymphocytes	0,21	,153	,116	,0062	,0048	1,31	,194
Microbian Count vs Staph. aur.	0,18	,157	,106	,0041	,0028	1,48	,142
CD16 <sup>+</sup> NK Lymphocytes	0,17	,128	,117	,0047	,0043	1,10	,275
Circulating Immune Complexes	-0,16	-,180	,106	-,0024	,0014	-1,70	,093

**Table 10. Regression Summary for F8H**R=0,270; R<sup>2</sup>=0,073; Adjusted R<sup>2</sup>=0,051; F<sub>(2,8)</sub>=3,3; p=0,043

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(83)</sub>	p-level
<b>Variable</b>	r		Intercpt	,5860	,2113	2,77	,007
Microbian Count vs E. coli	-0,16	,135	,106	,0039	,0031	1,27	,206
Popovych's Adaptation Index-1	-0,23	-,222	,106	-,0844	,0404	-2,09	,040

**Table 11. Regression Summary for T3H**R=0,233; R<sup>2</sup>=0,054; Adjusted R<sup>2</sup>=0,043; F<sub>(1,8)</sub>=4,8; p=0,031

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(84)</sub>	p-level
<b>Variable</b>	r		Intercpt	1,0430	,1048	9,95	10 <sup>-6</sup>
Killing Index vs Staphyl. aureus	-0,23	-,233	,106	-,0046	,0021	-2,19	,031

**Table 12. Regression Summary for T4H**R=0,282; R<sup>2</sup>=0,080; Adjusted R<sup>2</sup>=0,057; F<sub>(2,8)</sub>=3,6; p=0,032

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(83)</sub>	p-level
Variable	r		Intercpt	1,0583	,1011	10,5	10 <sup>-6</sup>
Killing Index vs Staphyl. aureus	-0,21	-,224	,106	-,0040	,0019	-2,12	,037
Popovych's Adaptation Index-1	-0,17	-,193	,106	-,0473	,0260	-1,82	,072

**Table 13. Regression Summary for C3H**R=0,390; R<sup>2</sup>=0,152; Adjusted R<sup>2</sup>=0,110; F<sub>(4,8)</sub>=3,6; p=0,009

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(81)</sub>	p-level
Variable	r		Intercpt	,7265	,1317	5,52	10 <sup>-6</sup>
Immunoglobuline M	0,25	,210	,106	,0926	,0464	1,99	,049
Microbian Count vs E. coli	0,18	,161	,106	,0026	,0017	1,52	,133
Immunoglobuline A	-0,23	-,175	,109	-,0455	,0283	-1,61	,112
T Active Lymphocytes	-0,19	-,158	,109	-,0039	,0027	-1,45	,152

**Table 14. Regression Summary for C4H**R=0,358; R<sup>2</sup>=0,129; Adjusted R<sup>2</sup>=0,085; F<sub>(4,8)</sub>=3,0; p=0,024

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(81)</sub>	p-level
Variable	r		Intercpt	,5516	,1460	3,78	,0003
Microbian Count vs Staph. aur.	0,27	,244	,104	,0042	,0018	2,34	,022
Immunoglobuline M	0,17	,143	,105	,0686	,0502	1,37	,175
Popovych's Adaptation Index-1	-0,17	-,133	,105	-,0306	,0242	-1,27	,209
Circulating Immune Complexes	-0,16	-,110	,105	-,0009	,0009	-1,05	,298

**Table 15. Regression Summary for T5H**R=0,302; R<sup>2</sup>=0,091; Adjusted R<sup>2</sup>=0,058; F<sub>(3,8)</sub>=2,7; p=0,049

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(82)</sub>	p-level
Variable	r		Intercpt	1,192	,2985	3,99	,0001
Entropy of LCG	-0,23	-,165	,111	-,6284	,4257	-1,48	,144
Killing Index vs Staphyl. aureus	-0,16	-,121	,110	-,0027	,0025	-1,10	,274
CD8 <sup>+</sup> T-cytolytic Lymphocytes	0,20	,173	,107	,0060	,0037	1,61	,111

**Table 16. Regression Summary for T6H**R=0,329; R<sup>2</sup>=0,108; Adjusted R<sup>2</sup>=0,075; F<sub>(3,8)</sub>=3,3; p=0,024

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(82)</sub>	p-level
Variable	r		Intercpt	,6442	,2218	2,90	,005
CD16 <sup>+</sup> NK Lymphocytes	0,25	,218	,108	,0067	,0033	2,02	,047
Microbian Count vs Staph. aur.	0,16	,142	,106	,0032	,0024	1,34	,185
Killing Index vs Staphyl. aureus	-0,22	-,140	,109	-,0030	,0024	-1,28	,204



**Table 17. Regression Summary for P3H**R=0,389; R<sup>2</sup>=0,152; Adjusted R<sup>2</sup>=0,099; F<sub>(5,8)</sub>=2,9; p=0,020

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(80)</sub>	p-level
Variable	r		Intercpt	2,589	1,501	1,72	,089
SN Neutrophils	0,24	-,684	,699	-,0111	,0114	-,98	,331
Killing Index vs E. coli	-0,23	-,246	,123	-,0024	,0012	-2,00	,048
Entropy of LCG	-0,22	-,355	,337	-,9487	,8993	-1,06	,295
Eosinophils	-0,20	-,244	,119	-,0164	,0080	-2,04	,044
Lymphocytes	-0,20	-,661	,545	-,0117	,0096	-1,21	,229

**Table 18. Regression Summary for O1H**R=0,464; R<sup>2</sup>=0,215; Adjusted R<sup>2</sup>=0,156; F<sub>(6,8)</sub>=3,6; p=0,003

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(79)</sub>	p-level
Variable	r		Intercpt	1,645	,3186	5,17	10 <sup>-5</sup>
Entropy of LCG	-0,31	-,427	,157	-1,541	,5677	-2,71	,008
Popovych's Strain Index-2	-0,26	-1,289	,576	-,5618	,2510	-2,24	,028
Popovych's Strain Index-1	-0,23	1,040	,526	,5748	,2908	1,98	,052
Killing Index vs Staphyl. aureus	-0,20	-,151	,107	-,0032	,0023	-1,41	,163
Eosinophils	-0,18	,444	,276	,0404	,0251	1,61	,111
Immunoglobuline M	0,17	,199	,101	,1235	,0630	1,96	,053

**Table 19. Regression Summary for O2H**R=0,321; R<sup>2</sup>=0,103; Adjusted R<sup>2</sup>=0,071; F<sub>(3,8)</sub>=3,2; p=0,027

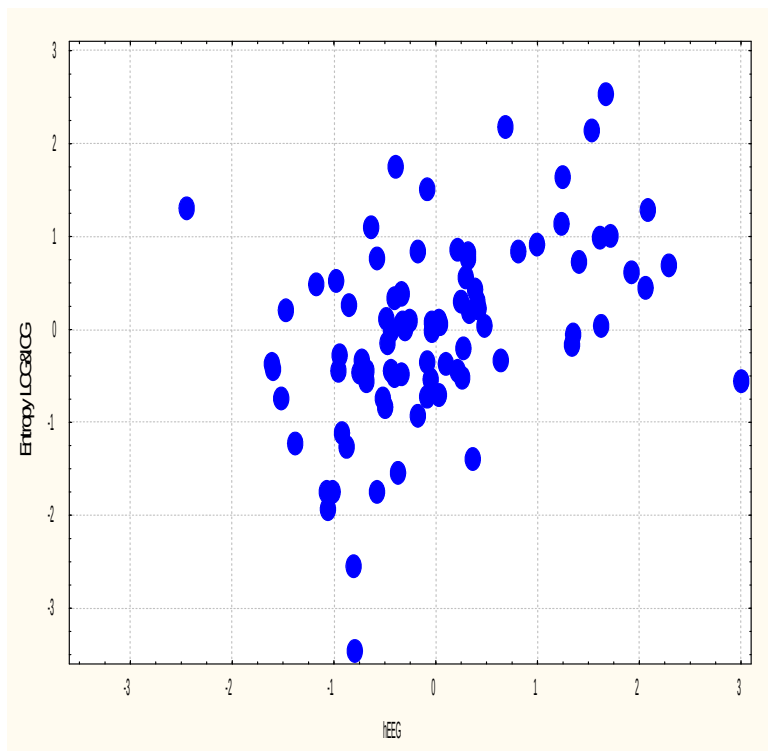
		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(84)</sub>	p-level
Variable	r		Intercpt	,5817	,3440	1,69	,095
Killing Index vs Staphyl. aureus	-0,23	-,233	,104	-,0048	,0022	-2,23	,028
Popovych's Strain Index-2	-0,17	-,130	,105	-,0558	,0448	-1,24	,217
Entropy of ICG	0,18	,141	,105	,4419	,3292	1,34	,183

As we see, the coefficients of the multiplicity correlation, despite the statistical significance, are very moderate, being in the range of 0,233÷0,529.

A similar situation with respect to the influence of entropy of the constellation of EEG loci on the entropy of LCG and ICG is revealed also in the result of canonical correlation analysis (Table 20 and Fig. 1).

**Table 20. Factor Structure Matrix for Entropy of EEG vs Entropy of LCG and ICG**

Right set	R
O1H	<b>-,681</b>
T5H	<b>-,564</b>
P3H	<b>-,563</b>
O2H	<b>-,507</b>
T6H	<b>-,481</b>
F3H	<b>,491</b>
Left set	R
LCGH	<b>,952</b>
ICGH	<b>-,661</b>



R=0,433; R<sup>2</sup>=0,188;  $\chi^2_{(12)}=23$ ; p=0,024;  $\Lambda$  Prime=0,750

**Figure 1. Scatterplot of canonical correlation between Entropy of EEG (X-line) and LCG and ICG (Y-line)**

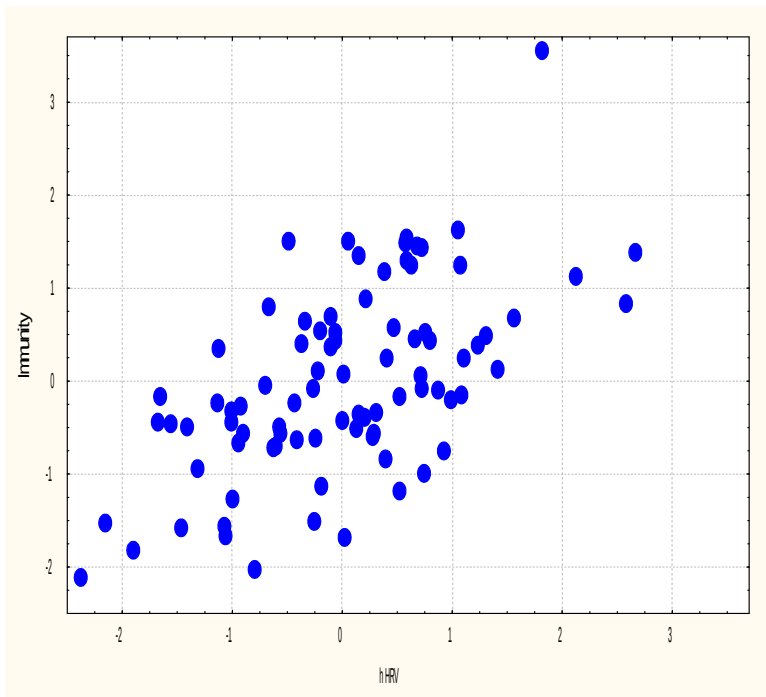
Instead, the maximum determinant influence on the parameters of immunity is the entropy of HRV (Table 20).

This is consistent with the concept [14-17,23,24,28] that the sympathetic and vagal nerves directly affect the immunocytes, more precisely the norepinephrine and acetylcholine released by their terminals, whereas cortical and subcortical neural structures directly regulate the nucleus of the autonomic nervous system (nucleus coeruleus, ambiguus, dorsalis motoris etc).

**Table 21. Regression Summary for HRVH**

R=0,597; R<sup>2</sup>=0,355; Adjusted R<sup>2</sup>=0,278; F<sub>(9,8)</sub>=4,6; p<10<sup>-4</sup>

		Beta	St. Err. of Beta	B	St. Err. of B	t <sub>(76)</sub>	p-level
<b>Variable</b>	r		Intercept	1,2477	,2009	6,21	10 <sup>-6</sup>
Killing Index vs Staphyl. aureus	0,28	,150	,104	,0023	,0016	1,44	,154
CD8 <sup>+</sup> T-cytolytic Lymphocytes	0,17	,221	,102	,0051	,0024	2,16	,034
Microbian Count vs E. coli	-0,27	-,310	,101	-,0050	,0016	-3,07	,003
Segmentonuclear Neutrophiles	-0,25	-,235	,117	-,0037	,0018	-2,02	,047
Popovych's Strain Index-2	-0,23	,635	,437	,1957	,1346	1,45	,150
Popovych's Adaptation Index-2	-0,21	-,190	,104	-,0513	,0282	-1,82	,072
Eosinophiles	-0,21	-,312	,195	-,0201	,0126	-1,60	,114
Leukocytes	-0,18	-,168	,103	-,0204	,0125	-1,63	,106
Popovych's Strain Index-1	-0,18	-,767	,397	-,2999	,1555	-1,93	,057



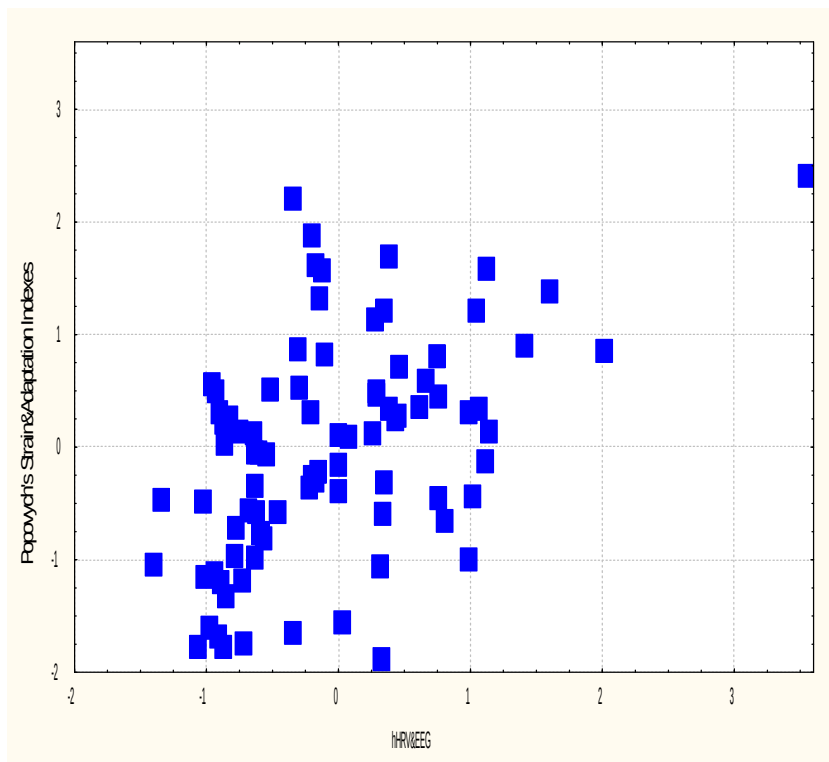
R=0,597; R<sup>2</sup>=0,355;  $\chi^2_{(9)}=35$ ;  $p < 10^{-4}$ ;  $\Lambda$  Prime=0,645

**Figure 2. Scatterplot of canonical correlation between Entropy of HRV (X-line) and Immunity (Y-line)**

A special canonical analysis has shown that the newly proposed modifications of Popovych's Strain and Adaptation Indices are more closely correlated with the entropies of the neural regulatory structures compared to the previous version (Table 22 and Fig. 3).

**Table 22. Factor Structure Matrix for Entropy of HRV and EEG vs Popovych's Strain and Adaptation Indices**

Right set	R
HRVH	-,657
O1H	-,444
FP1H	-,112
FP2H	-,051
P3H	,204
F4H	,018
Left set	R
PSI-2	,739
PSI-1	,709
PAI-2	,510
PAI-1	,100



$R=0,540$ ;  $R^2=0,291$ ;  $\chi^2_{(24)}=54$ ;  $p=0,0005$ ;  $\Lambda$  Prime= $0,514$

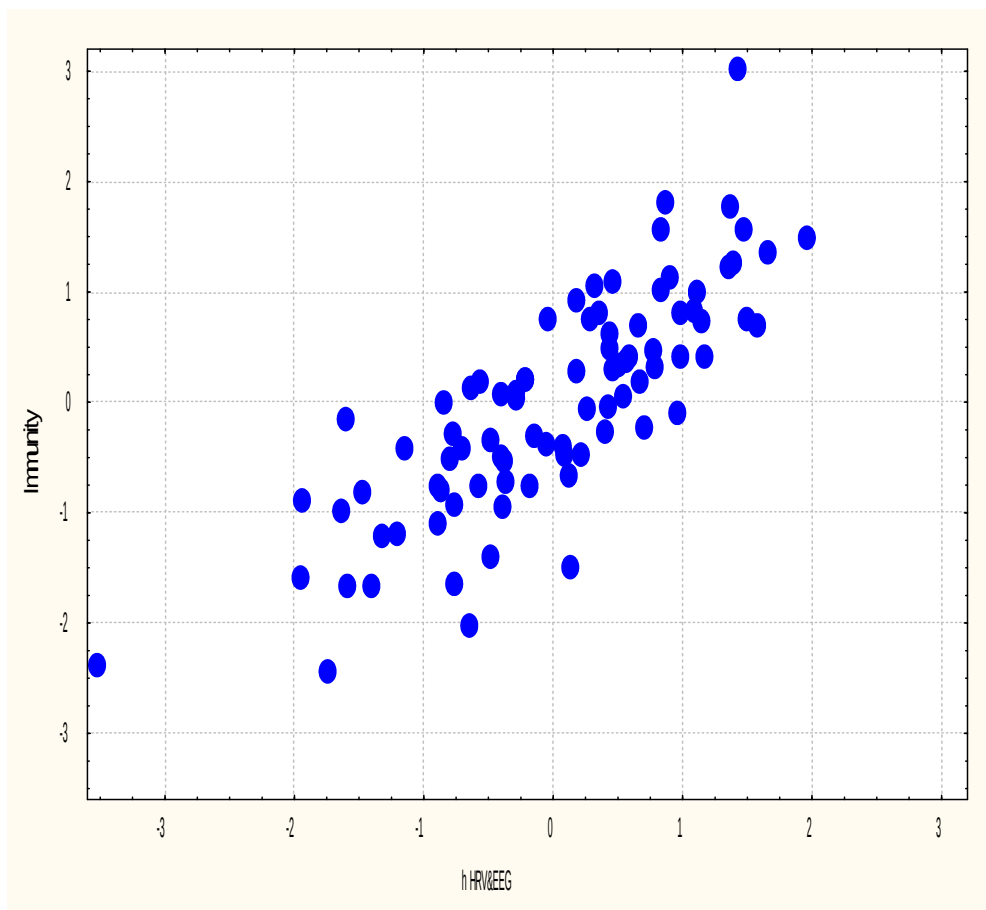
**Figure 3. Scatterplot of canonical correlation between Entropy of HRV and EEG (X-line) and Popovych's Strain and Adaptation Indices (Y-line)**

At the final stage, we investigated the canonical correlation between the entropy of HRV and EEG, on the one hand, and the actual and **informational** parameters of immunity, on the other hand (Table 23 and Fig. 4).

**Table 23. Factor Structure Matrix for Entropy of HRV and EEG vs Immunity in total**

<b>Right set</b>	<b>R</b>
HRVH	,635
C3H	,214
F3H	,102
C4H	,100
F4H	,073
T5H	,046
T6H	,037
P3H	-,290
O2H	-,260
F7H	-,212
FP2H	-,098
O1H	-,037
<b>Left set</b>	<b>R</b>
Lymphocytes in total	,715
Killing Index vs E. coli	,412
Killing Index vs Staph. aureus	,305
Immunoglobuline M	,286
CD8 <sup>+</sup> T-cytolytic Lymphocytes	,141
Circulating Immune Complex	,088
<b>Entropy of LCG</b>	<b>,064</b>

Segmentonucleary Neutrophils	<b>-,535</b>
T-active Lymphocytes	<b>-,420</b>
CD16 <sup>+</sup> NK Lymphocytes	<b>-,345</b>
<b>Popovych's Adaptation Index-2</b>	<b>-,304</b>
Immunoglobuline A	<b>-,283</b>
<b>Popovych's Strain Index-1</b>	<b>-,280</b>
CD4 <sup>+</sup> T-helper Lymphocytes	<b>-,242</b>
<b>Popovych's Strain Index-2</b>	<b>-,226</b>
Leukocytes	<b>-,167</b>
<b>Popovych's Adaptation Index-1</b>	<b>-,124</b>
Microbial Count vs E. coli	<b>-,109</b>
Eosinophils	<b>-,102</b>
Microbial Count vs Staph. aureus	<b>-,029</b>



R=0,814; R<sup>2</sup>=0,663;  $\chi^2_{(240)}=296$ ; p=0,008;  $\Lambda$  Prime=0,013

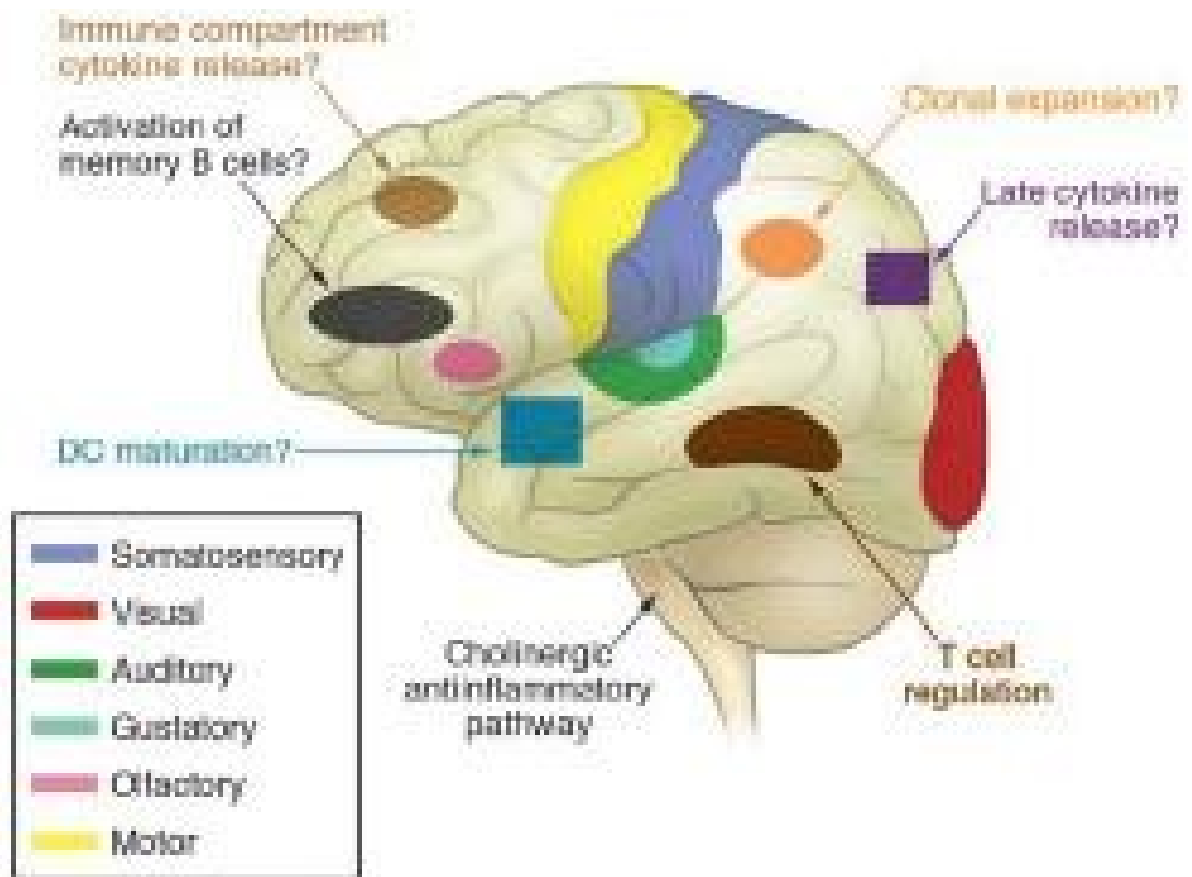
**Figure 4. Scatterplot of canonical correlation between Entropy of HRV and EEG (X-line) and Immunity (Y-line)**

Our data on the significant factor loads on the F3, F4, P3, T5, T6 and Fp2 loci are consistent with the KJ Tracey's [28] scheme of immunological homunculus (Fig. 5) by which the neural structures that are projected onto these loci responsible for the immune compartment cytokines release (F3 and F4), clonal expansion (P3), regulation of T cells (T5 and T6) and activation of memory B cells (Fp2) respectively.

However, we did not find in the factor structure matrix of the anterior temporal locus, on which projected nerve structures responsible for maturation of dendritic cells. Instead, we have reason to supplement the KJ Tracey's scheme with the assumption that the structures

(hippocampus? [26]) that projected on the left central locus are responsible for increasing the content in blood of IgM and total lymphocytes and reducing the content of IgA and segmentonuclear neutrophils. Instead, the structures responsible for increasing the intensity phagocytosis by neutrophils of gram-positive and gram-negative microbes are plotted on the right central locus.

Another addition may be the assumption of localization in the occipital loci of the structures responsible for the entropy of the leukocytogram, its strain, as well as the inhibition of completeness of phagocytosis of *Staph. aureus*, but not *E. coli*. Differences in the neural regulation of these two types of bacteria have been detected by us before [16].



**Figure 5. KJ Tracey's scheme of immunological homunculus [28]**

### 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. Avtandilov GG. Medical Morphometry [in Russian]. Moskva. Meditsina; 1990: 384 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., Malyuchkova RV, Tolstanov OB, Tymochko OB, Hryvna R, Uhryn MR. Comparative estimation of informativeness of leukocytary index of adaptation by Garkavi and by Popovych. *Medical Hydrology and Rehabilitation*. 2013; 11(1): 5-20.
4. Berntson GG, Bigger JT jr, Eckberg DL, Grossman P, Kaufman PG, Malik M, Nagaraja HN, Porges SW, Saul JP, Stone PH, Van der Molen MW. Heart Rate Variability: Origenes, methods, and interpretive caveats. *Psychophysiology*. 1997; 34: 623-648.
5. Biloshytskyi PV. Temperature, Information, Water, Anabiosis, Immortality [in Ukrainian]. *Health and Longevity*. Kyiv; 2007: 46-47.
6. Biloshytskyi PV, Biloshytskyi SP, Klyuchko OM. Conceptual Foundations of Bioinformation [in Ukrainian]. *Cosmos and the Biosphere*. Partenit; 2005: 131-132.
7. Dranovs'kyi AL, Popovych IL. Adaptogenic Balneotherapy on Spa Truskavets' [in Ukrainian]. *Drohobych. Posvit*; 2010: 203 p.
8. Douglas SD, Quie PG. Investigation of Phagocytes in Disease. Churchill; 1981: 110 p.
9. Garkavi LKh, Kvakina EB, Ukolova MA. Adaptation Reactions and Resistance of Organism [in Russian]. Rostov na Donu. Publishing House of Rostov University Press, 3rd ed; 1990: 224 p.
10. Garkavi LKh, Kvakina EB, Kuz'menko TS. Antistressory Reactions and Activating Therapy [in Russian]. Moskva. Imedis; 1998: 654 p.
11. Garkavi LKh, Romasyuk SI, Barantsev FG, Kuz'menko TS, Otkidach SA, Tatkov OV, Barantseva LP. Activation therapy in the complex of the sanatorium-resort stage of rehabilitation of patients with diseases of internal organs [in Russian]. Sochi; 2000: 94 p.
12. Heart Rate Variability. Standards of Measurement, Physiological Interpretation, and Clinical Use. Task Force of ESC and NASPE. *Circulation*. 1996; 93(5): 1043-1065.
13. Kostyuk PG, Popovych IL, Ivassivka SV (editors). Chornobyl', Adaptive and Defensive Systems, Rehabilitation [in Ukrainian]. Kyiv. Computerpress; 2006: 348 p.
14. Kul'chyns'kyi AB, Gozhenko AI, Popovych IL. Interrelations between autonomous and immune systems (review) [in Ukrainian]. *Actual problems of transport medicine*. 2017; 3(49): 7-25.
15. Kul'chyns'kyi AB, Gozhenko AI, Zukow W, Popovych IL. Neuro-immune relationships at patients with chronic pyelonephrite and cholecystite. Communication 3. Correlations between parameters EEG, HRV and Immunogram. *Journal of Education, Health and Sport*. 2017; 7(3): 53-71.
16. Kul'chyns'kyi AB, Kovbasnyuk MM, Korolyshyn TA, Kyjenko VM, Zukow W, Popovych IL. Neuro-immune relationships at patients with chronic pyelonephrite and cholecystite. Communication 2. Correlations between parameters EEG, HRV and Phagocytosis. *Journal of Education, Health and Sport*. 2016; 6(10): 377-401.
17. Kul'chyns'kyi AB, Kyjenko VM, Zukow W, Popovych IL. Causal neuro-immune relationships at patients with chronic pyelonephritis and cholecystitis. Correlations between parameters EEG, HRV and white blood cell count. *Open Medicine*. 2017; 12(1): 201-213.
18. Lapovets' LYe, Lutsyk BD. Handbook of Laboratory Immunology [in Ukrainian]. Lviv. 2002. 173 p.
19. Newberg AB, Alavi A, Baime M, Pourdehnad M, Santanna J, d'Aquili E. The measurement of regional cerebral blood flow during the complex cognitive task of meditation: a preliminary SPECT study. *Psychiatry Research: Neuroimaging Section*. 2001; 106: 113-122.
20. Petsyukh SV, Petsyukh MS, Kovbasnyuk MM, Barylyak LG, Zukow W. Relationships between Popovych's Adaptation Index and parameters of ongoing HRV and EEG in patients with chronic pyelonephrite and cholecystite in remission. *Journal of Education, Health and Sport*. 2016; 6(2): 99-110.
21. Popovych IL (editor). General Adaptation Reactions and Body's Resistance in Liquidators of the Chernobyl Accident [in Ukrainian]. Kyiv. Computerpress; 2000: 117 p.
22. Popovych IL. Information effects of bioactive water Naftyssya in rats: modulation entropic, prevention desynchronizing and limitation of disharmonizing actions water immersion stress for

- information components of neuro-endocrine-immune system and metabolism, which correlates with gastroprotective effect [in Ukrainian]. *Medical Hydrology and Rehabilitation*. 2007; 5(3): 50-70.
23. Popovych IL, Kozyavkina OV, Kozyavkina NV, Korolyshyn TA, Lukovych YuS, Barylyak LG. Correlation between Indices of the Heart Rate Variability and Parameters of Ongoing EEG in Patients Suffering from Chronic Renal Pathology. *Neurophysiology*. 2014; 46(2): 139-148.
  24. Popovych IL, Lukovych YuS, Korolyshyn TA, Barylyak LG, Kovalska LB, Zukow W. Relationship between the parameters heart rate variability and background EEG activity in healthy men. *Journal of Health Sciences*. 2013; 3(4): 217-240.
  25. Radchenko OM. *Adaptation Reactions in Internal Diseases Clinic* [in Ukrainian]. Lviv: League-Press. 2004. 232 p.
  26. Romodanov AP (editor). *Postradiation Encephalopathy. Experimental Researches and Clinical Observations* [in Ukrainian and Russian]. Kyiv. USRI of Neurosurgery; 1993: 224 p.
  27. Shannon CE. *Works on the theory of informatics and cybernetics* [transl. from English to Russian]. Moskwa: Inostrannaya literatura; 1963: 329 p.
  28. Tracey K.J. Physiology and immunology of the cholinergic antiinflammatory pathway. *J Clin Invest*. 2007; 117(2): 289-296.
  29. Yushkovs'ka OG. Using information theory to study adaptive responses in the body athletes [in Ukrainian]. *Medical Rehabilitation, Kurortology, Physiotherapy*. 2001; 1(25): 40-43.