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Some hemodynamic and metabolic predictors and accompaniments of the two-stage cycle ergometric test

O. I. MEL'NYK <https://orcid.org/0000-0001-7928-4760>

Stepan Gzhyskyi National University of Veterinary Medicine and Biotechnologies,
 Lviv, Ukraine

E-mail: omelnyk7@gmail.com

Summary. Introduction and aim. Ergometric testing of physical working capacity (PWC) has a long tradition in occupational and sports medicine, as well as in medical rehabilitation and recreation. In previous experimental and clinical-physiological studies of the Truskavetsian Scientific School of Balneology the influence of the spa's balneological agents on the PWC of both healthy rats and different categories of patients of both sexes was analyzed, as well as its relationship with the parameters of the neuro-endocrine-immune complex, metabolism, hemostasis and biophotonics. Among the critical remarks of the reviewers, the most significant was the small sample size and gender imbalance. This study initiates a new project «Predictors and accompaniments of the two-stage bicycle ergometric test», the implementation of which begins with the analysis of hemodynamics and metabolic parameters.

Material and methods. We examined 63 women (aged 29÷76 years) and 56 men (aged 25÷70 years) with chronic gastroenterological and urological pathology in remission before and after a 1–3-week course of balneotherapy at the Truskavets' spa. The main subject of the study was a two-stage bicycle ergometry test. At the same time, blood pressure, blood flow in both forearms and its response to occlusion, as well as lipid, electrolyte and nitrogen profiles of plasma/serum were recorded.

Results. A wide range of PWC₁₅₀ levels (Mean ± SD) was found, grouped into 4 clusters. The most numerous (180 cases out of 238) was a normal test (2.66 ± 0.54 W/kg; Z-score: 0.00 ± 0.54). In 21 cases, a reduced PWC₁₅₀ level was found (1.53 ± 0.32 W/kg; Z: -1.52 ± 0.46), and in 31 cases, an increased one (3.43 ± 0.45 W/kg; Z: 1.35 ± 0.28). At the same time, in 6 cases an abnormally high PWC₁₅₀ level was registered (4.45 ± 0.73 W/kg; Z: 3.32 ± 0.83). Discriminant analysis revealed 22 variables (4 rheographic, 4 electrolytic, 2 uricemic, 4 hemodynamic, 7 lipids, and the metabolic syndrome index), in the aggregate of which all four PWC₁₅₀ clusters significantly differ from each other (classification accuracy 96.6 %). A regression model was created with 6 variables (basal blood flow in both forearms, serum Ca, erythrocyte K, plasma TG/ α -LP index, and sex index), which explain 79.9 % of the variance of PWC₁₅₀ and allow predicting its level with an error of 0.33 W/kg.

Conclusion. In patients of the Truskavets' spa, the level of PWC_{150} is in a wide range – from low to abnormally high, and is naturally associated with deviations in a number of hemodynamic and metabolic parameters caused by chronic diseases, as well as sex. Targeted influence on the identified predictors will probably make it possible to increase the efficiency of recovery or further increase PWC_{150} .

Key words: two-stage cycle ergometric test, hemodynamics, metabolism, relationships, chronic urological and gastroenterological pathology, the Truskavets' spa.

Деякі гемодинамічні і метаболічні предиктори та супутники двоступеневого велоергометричного тесту

О. І. МЕЛЬНИК

*Львівський національний університет ветеринарної медицини та біотехнологій
імені С. З. Гжицького, Львів, Україна*

E-mail: omelnyk7@gmail.com

Резюме. Вступ та мета. Ергометричне тестування фізичної працездатності має давні традиції в медицині праці і спорту, а також у медичній реабілітації та рекреації. У попередніх експериментальних і клініко-фізіологічних дослідженнях Трускавецької наукової школи бальнеології був проаналізований вплив бальнеочинників курорту на фізичну працездатність, а також її зв'язки з параметрами нейро-ендокринно-імунного комплексу, метаболізму, гемостазу і біофотоніки. Водночас ці дослідження мали суттєві недоліки – як невеликий обсяг вибірки і гендерний дисбаланс. Наше дослідження започатковує новий проект «Предиктори і супутники двоступеневого велоергометричного тесту», реалізація якого починається з аналізу параметрів гемодинаміки і метаболізму.

Матеріал і методи. Ми обстежили 63 жінок (віком $29 \div 76$ років) та 56 чоловіків (віком $25 \div 70$ років) із хронічною гастроентерологічною та урологічною патологією в стадії ремісії до та після 1–3-тижневого курсу бальнеотерапії на курорті Трускавець. Основним предметом дослідження був двоступеневий велоергометричний тест. Одночасно реєстрували артеріальний тиск, кровоплин в обох передпліччях та його реакцію на окклюзію, а також ліпідний, електролітний та азотистий профілі плазми / сироватки.

Результати. Виявлено широкий діапазон рівнів PWC_{150} (Mean $\pm$ SD), згрупованих у 4 кластери. Найчисленнішим (180 випадків із 238) виявився нормальний тест ($2,66 \pm 0,54$ W/kg; Z-score: $0,00 \pm 0,54$). У 21 випадку констатовано знижений рівень PWC_{150} ($1,53 \pm 0,32$ W/kg; Z: $-1,52 \pm 0,46$), а у 31 випадку – підвищений ($3,43 \pm 0,45$ W/kg; Z: $1,35 \pm 0,28$). Аномально високий рівень PWC_{150} ($4,45 \pm 0,73$ W/kg; Z: $3,32 \pm 0,83$) зареєстровано у 6 випадках. Дискримінантний аналіз виявив 22 змінні (4 реографічні, 4 електролітні, 2 урикемічні, 4 гемодинамічні, 7 ліпідні, а також індекс метаболічного синдрому), за сукупністю яких всі чотири кластери PWC_{150} значуще відрізняються один від одного (коректність класифікації 96,6 %). Створено регресійну модель із 6 змінних (базальний кровоплин в обох передпліччях, Са сироватки, К еритроцитів, TG/ α -LP індекс плазми та секс індекс), які пояснюють 79,9 % дисперсії PWC_{150} і дозволяють передбачити її рівень з похибкою 0,33 W/kg.

Висновки. У пацієнтів курорту Трускавець рівень PWC_{150} знаходиться в широкому діапазоні – від зниженого до аномально підвищеного, і закономірно пов'язаний з відхиленнями низки параметрів гемодинаміки та

метаболізму, зумовленими хронічними захворюваннями, а також статтю. Цілеспрямований вплив на виявлені предиктори, мабуть, уможливить підвищення ефективності відновлення чи дальшого зростання PWC_{150} .

Ключові слова: двоступеневий велоергометричний тест, гемодинаміка, метаболізм, взаємозв'язки, хронічна урологічна та гастроентерологічна патологія, курорт Трускавець.

Introduction. Ergometric physical working capacity (PWC) testing has a long tradition in occupational medicine for assessing whether a sufficiently high level of physical performance for coping with the daily work requirements is given; in sports medicine to assess the effectiveness of the training process and recovery methods; in rehabilitation medicine and recreation [1–7]. PWC can be tested maximally or submaximally, using performance indicators like $VO_2\max$ [8] or the mechanical power [9, 10]. In the case of submaximal PWC testing measuring the mechanical power, the achieved power at a given heart rate serves as performance indicator. There are age- and sex-specific norm values [11] that can be used to judge whether differences or changes are within the normal range or can be considered significant.

Our group has previously demonstrated the role of innate muscular endurance in reactions to acute stress of neuro-endocrine-immune complex in rats [12–14]. Physical performance and stress resistance of the body are considered one of the main objects of influence of adaptogens and are used for quantitative assessment of adaptogenic activity [15]. The stress-limiting effect of bioactive Naftussya water as the main healing factor of the Truskavets' spa is known [16, 17]. Data on the influence of balneotherapy at the Truskavets' spa on the muscular performance of patients (adults and children), assessed by велоergometric or step tests, are ambiguous [6, 7, 18, 19]. In previous experimental and clinical-physiological studies of the Truskavetsian Scientific School of Balneology the influence of the spa's balneological agents on the PWC of both healthy rats and different categories of patients of both sexes was analyzed, as well as its relationship with the parameters of the neuro-endocrine-immune complex, metabolism, hemostasis and biophotonics [20–23]. Among the critical remarks of the reviewers, the most significant was the small sample size and gender imbalance. This study initiates a new project «Predictors and accompaniments of the two-stage bicycle ergometric test», the implementation of which begins with the analysis of hemodynamics and metabolic parameters.

Material and methods. Participants. We examined 63 women (aged 29–76 years) and 56 men (aged 25–70 years) with chronic gastroenterological and urological pathology in remission before and after a 1–3-week course of balneotherapy at the Truskavets' Spa. Inclusion criteria: chronic cholecystitis, gastritis, enterocolitis, pyelonephritis, and cystitis in remission, no exacerbation for at least 3 months before balneotherapy. Exclusion criteria: acute urinary or digestive tract infection, decompensated concomitant diseases of the cardiovascular system, diabetes mellitus, oncological diseases. All patients signed an informed consent to participate in the study. The study was approved by the local ethics committee in accordance with the principles of the Declaration of Helsinki.

Procedure / Test protocol / Skill test trial / Measure / Instruments. Systolic and diastolic BP simultaneously with HR was measured by tonometer «Omron M4-1» (Netherlands) with the following calculation Kerdő's Vegetative Index [24, 25].

Next, without removing the cuff, blood flow was measured in both forearms by rheovasography («Georg V 1.0», «Medtech», Kharkiv, Ukraine) under basal conditions. After that, following the classical method of *Sagach VF et al.* [26], blood flow occlusion was induced in the right («active») forearm by compressing the shoulder for 30 seconds with air pressure in the cuff, which was 30 mmHg higher than the just measured systolic BP. This maneuver allows us to assess

the state of endothelial function by the blood flow response to occlusion in the form of its increase in percentage – flow-mediated dilation (FMD) [27, 28]. It has been demonstrated [7] that it is more correct to use the logarithm of the ratio of the levels of reactive and basal blood flow: $FMD = \lg (BF_{\text{reactive}}/BF_{\text{basal}})$.

Following the concept of the functional-metabolic continuum [29], functional tests were combined with biochemical tests.

Lipoprotein profile of plasma was estimated by: triglycerides (certain meta-periodate method); total cholesterol (direct method after the classic reaction by Zlatkis-Zack) and content of him in composition of α -lipoproteins (HDLP) (by the Hiller [30] enzyme method after precipitation of non-lipoproteins); pre- β -lipoproteins (VLDLP) (expected by the level of triglycerides as ratio $TG/2,1834$ [31]; β -lipoproteins (LDLP) (expected by a difference between a total cholesterol and cholesterol in composition α - and pre- β -lipoproteins).

Two versions of Atherogenity Index (AI) were calculated: Dobiášová & Frolich as $\lg (TG/\alpha\text{-LP})$ [32, 33] as well as previously widely used Klimov's AIP as ratio non- α -LP/ α -LP [34].

On sodium and potassium exchange judged by their levels in the serum and erythrocytes (flame photometry method) as well as activity of Na,K-ATPase of erythrocyte ghosts, determined by the increase of Pi in the supernatant of the incubation medium [35]. Additional determined serum levels of calcium (by reaction with arsenase III) and magnesium (by reaction with colgamite) as well as activity of Ca-ATPase and Mg-ATPase of erythrocyte ghosts. At last, determined serum levels of nitric metabolites: creatinine (by Jaffe's color reaction by Popper's method), urea (urease method by reaction with phenolhypochlorite), and uric acid (uricase method) as well as glucose (glucose-oxidase method) [36].

An analyzers «Reflotron» (BRD) and «Pointe-180» (USA) and corresponding sets of reagents as well as flame photometer «CΦ-47» was used for biochemical studies.

The reference values are taken from the manuals [36, 37] instructions as well as database of the Truskavetsian Scientific School of Balneology.

The next day, physical working capacity (PWC_{150}) was assessed. A bicycle ergometer «Tunturi» (Finland) was used. The power of the first load was 0.5 W/kg, the second load (after 3 min) 1.5 W/kg at a pedaling frequency of 60–75 rpm. This corresponded to the recommendations for ergometer testing in occupational medicine [38–40]. Before and after two loads we recorded HR and BP by tonometer «Omron M4-I» and calculated submaximal PWC_{150} with the mechanical power in Watt per kilogram body weight (W/kg) as indicator of cardiorespiratory fitness [39].

In addition, each patient underwent Stange's and Guenchi's breath-holding test, but after preliminary analysis it turned out that they do not reflect the state of the cardiorespiratory system [41], therefore they were not considered in this article.

Data collection and analysis / Statistical analysis. Statistical processing was performed using a software package «Microsoft Excell» and «Statistica 6.4 StatSoft Inc» (Tulsa, OK, USA).

Results and discussion. Given the known sexual dimorphism of PWC_{150} , its individual raw values (V) were transformed into Z-scores using the formula: $Z = (V - R)/SD$, where R is the reference value for men (3.04 W/kg) and women (2.26 W/kg), SD is the standard deviation (0.61 and 0.57, respectively).

A wide range of PWC_{150} levels (Mean $\pm$ SD) was found, grouped into 4 clusters. The most numerous (180 cases out of 238) was a normal test (2.66 ± 0.54 W/kg; Z-score: 0.00 ± 0.54). In 21 cases, a reduced PWC_{150} level was found (1.53 ± 0.32 W/kg; Z: -1.52 ± 0.46), and in 31 cases, an increased one (3.43 ± 0.45 W/kg; Z: 1.35 ± 0.28). At the same time, in 6 cases an abnormally high PWC_{150} level was registered (4.45 ± 0.73 W/kg; Z: 3.32 ± 0.83).

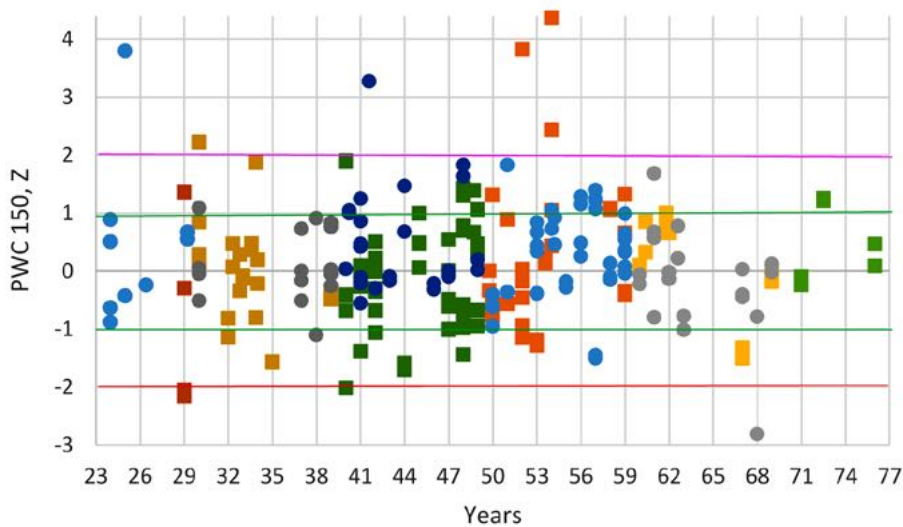


Fig. 1. Scatterplot of sex-normalized PWC₁₅₀ levels. Men are circles, women are squares

Since the reference values of the components of the lipid and nitrogen spectrum are characterized not only by gender, but also by age differences, their row levels were also transformed into Z-scores. At the same time, the remaining registered variables were unified, which made it possible to correctly compare them, as well as calculate two variants of the metabolic syndrome index (MSI).

$$\text{MSI-4} = (\text{TGz} + \alpha\text{-LPz} + \text{BPSz} + \text{BPDz})/4;$$
$$\text{MSI-5} = (\text{TGz} + \alpha\text{-LPz} + \text{BPSz} + \text{BPDz} + \text{GlucoseZ})/5.$$

In order to identify variables, in the aggregate of which all four PWC₁₅₀ clusters significantly differ from each other, discriminant analysis was performed (forward stepwise method [42]).

In the discriminant model (Tables 1 and 2), the program included, in addition to the normalized PWC₁₅₀ by definition, 22 variables: 4 rheographic, 4 hemodynamic, 4 electrolytic, 2 uricemic, 7 lipids, and the metabolic syndrome index-5.

Table 1

Discriminant Function Analysis Summary for Variables, their actual levels (Mean ± SE) as well as Reference levels and Coefficients of Variability

Step 23, N of vars in model: 23; Grouping: 4 grps; Wilks' Λ: 0,091;
approx. F₍₇₀₎ = 11,3; p < 10⁻⁶

Variables currently in the model	Clusters of PWC ₁₅₀ level (n)				Parameters of Wilks' Statistics					
	Pes- simal (21)	Nor- mal (180)	Opti- mal (31)	Ano- mal (6)	Wilks' Λ	Partial Λ	F- remove (3,21)	p- level	Tole- rancy	Refe- rence (SD)
PWC _{150s} Z	-1,52 0,10	0,00 0,04	1,35 0,05	3,32 0,34	0,189	0,481	76,17	10 ⁻⁶	0,511	0 2
Flow-Mediated Di- lation in passive FA	-0,03 0,032	0,104 0,015	0,068 0,025	-0,02 0,015	0,096	0,946	4,005	0,008	0,398	0,085 0,191
Basal BF in passive forearm, mL/min	52,5 5,2	75,3 1,9	99,3 6,0	143 9,5	0,096	0,953	3,497	0,016	0,115	78 30
Basal BF in active forearm, mL/min	53,7 7,2	95,2 3,3	133 16	137 14	0,094	0,969	2,295	0,079	0,257	97 54

Reactive BF in active FA, mL/min	51,7 6,1	105 3,3	138 11	171 10	0,094	0,973	1,995	0,116	0,117	107 51
Kerdö VI after load 1,5 W/kg, %	41,4 1,9	35,1 0,5	28,7 1,1	28,4 4,5	0,102	0,891	8,657	10 ⁻⁴	0,341	34,6 7,4
BP systolic after load 0,5 W/kg, mmHg	134 4,1	128 0,7	128 2,1	119,4 ,7	0,095	0,960	2,914	0,035	0,293	129 11
BP systolic after load 1,5 W/kg, mmHg	152 6,0	143 1,1	144 3,1	123 6,1	0,092	0,988	0,889	0,448	0,247	143 17
BP systolic basal, mmHg	123 2,7	132 1,4	128 2,8	117 3,4	0,093	0,980	1,423	0,237	0,283	124,5 15,2
Magnesium Serum, mM/L	0,74 0,01	0,82 0,01	0,94 0,03	0,79 0,03	0,104	0,872	10,35	10 ⁻⁵	0,487	0,83 0,11
Calcium Serum, mM/L	2,15 0,07	2,34 0,02	2,86 0,08	3,70 0,44	0,110	0,830	14,51	10 ⁻⁶	0,235	2,43 0,45
Sodium Serum, mM/L	135 3,6	148 1,0	166 3,9	189 15	0,094	0,966	2,489	0,061	0,227	150 18
Mg-ATPase Erythro- c. ghost, M _p /L•h	1,04 0,05	0,87 0,01	0,70 0,05	0,64 0,09	0,092	0,990	0,718	0,542	0,254	0,86 0,19
Uricemia, Z	0,47 0,57	0,27 0,14	-0,76 0,29	-1,65 0,52	0,097	0,942	4,389	0,005	0,029	0 2
Uricemia, μM/L	302 22	303 6	260 13	224 21	0,096	0,947	3,969	0,009	0,029	293 42
TG/α-LP, Z	1,37 0,54	-0,12 0,05	-0,13 0,06	-0,39 0,05	0,096	0,952	3,531	0,016	0,137	0 2
Nonα-Lipoproteins, units	61,0 4,1	50,2 1,0	55,1 2,8	48,6 7,0	0,099	0,922	6,010	0,001	0,199	51,7 14,6
Cholesterol total, mM/L	4,50 0,19	5,28 0,08	5,34 0,17	5,24 0,41	0,097	0,936	4,815	0,003	0,113	5,40 1,04
α-Lipoproteins Cholesterol, Z	-0,72 0,23	0,07 0,08	0,31 0,18	0,58 0,19	0,095	0,961	2,890	0,036	0,074	0 2
Triglycerides, mM/L	2,62 0,35	1,51 0,07	1,69 0,10	1,32 0,14	0,093	0,980	1,427	0,236	0,086	1,14 0,67
IgTG/α-LP as Dobi- ášová&Frolich AGI	0,34 0,08	-0,03 0,02	0,04 0,03	-0,10 0,04	0,094	0,968	2,333	0,075	0,137	-0,09 0,13
Nonα-LP /α-LP, Z	1,21 0,61	0,14 0,12	-0,32 0,21	-0,82 0,45	0,094	0,970	2,187	0,091	0,074	0 2
Metabolic Syndrome Index-5, Z	0,77 0,23	0,30 0,05	0,36 0,12	-0,39 0,18	0,094	0,972	2,026	0,111	0,158	0 2

Table 2

Summary of Stepwise Analysis Variables, ranked by criterion Λ

Variables currently in the model	F to enter	p-level	Λ	F-value	p-level
PWC ₁₅₀ , Z	226	10-6	0,2567	226	10 ⁻⁶
Magnesium Serum, mM/L	11,4	10-6	0,2239	86,5	10 ⁻⁶
Calcium Serum, mM/L	11,1	10-6	0,1959	59,9	10 ⁻⁶
IgTG/α-LP as Dobiášová&Frolich AGI	7,11	10-4	0,1793	46,6	10 ⁻⁶
Kerdö VI after load 1,5 W/kg, %	5,26	0,002	0,1678	38,5	10 ⁻⁶
Mg-ATPase Erythrocyte ghost, MP/L•h	4,31	0,006	0,1589	33,0	10 ⁻⁶
Reactive BF in active forearm, mL/min	4,37	0,005	0,1502	29,2	10 ⁻⁶

BP systolic after load 0,5 W/kg, mmHg	3,03	0,030	0,1444	26,0	10 ⁻⁶
BP systolic basal, mmHg	3,02	0,031	0,1389	23,6	10 ⁻⁶
Metabolic Syndrome Index-5, Z	3,18	0,025	0,1332	21,8	10 ⁻⁶
TG/ α -LP, Z	2,82	0,040	0,1284	20,2	10 ⁻⁶
Non α -LP / α -LP, Z	1,95	0,122	0,1251	18,7	10 ⁻⁶
BP systolic after load 1,5 W/kg, mmHg	1,70	0,167	0,1223	17,4	10 ⁻⁶
Uricemia, Z	1,89	0,131	0,1192	16,4	10 ⁻⁶
Uricemia, μ M/L	2,15	0,095	0,1158	15,5	10 ⁻⁶
Sodium Serum, mM/L	2,16	0,094	0,1125	14,7	10 ⁻⁶
Flow Mediated Dilation in passive FA	1,58	0,195	0,1101	14,0	10 ⁻⁶
Basal BF in passive forearm, mL/min	2,05	0,109	0,1071	13,4	10 ⁻⁶
Basal BF in active forearm, mL/min	2,29	0,079	0,1038	12,9	10 ⁻⁶
Non α -Lipoproteins, units	1,66	0,177	0,1014	12,3	10 ⁻⁶
Cholesterol total, mM/L	3,76	0,012	0,0964	12,1	10 ⁻⁶
α -Lipoproteins Cholesterol, Z	2,65	0,050	0,0929	11,7	10 ⁻⁶
Triglycerides, mM/L	1,43	0,236	0,0910	11,3	10 ⁻⁶

Interestingly, raw PWC₁₅₀, along with a number of other variables, despite their obvious distinguishing features, turned out to be outside the model as carriers of duplicate discriminant information (Table 3).

Table 3

Summary of the analysis of discriminant functions for variables currently not in the model

Variables	Clusters of PWC ₁₅₀ level (n)				Parameters of Wilks' Statistics					
	Pes-simal (21)	Nor-mal (180)	Opti-mal (31)	Ano-mal (6)	Wilks' Λ	Partial Λ	F to enter	p-level	Tole-rancy	Refe-rence (SD)
PWC ₁₅₀ , W/kg	1,53 0,07	2,66 0,04	3,43 0,08	4,45 0,30	0,090	0,989	0,784	0,504	0,138	2,62 0,59
Reactive BF in pas-sive FA, mL/min	50,9 5,9	101 3,6	126 14	137 11	0,091	0,999	0,088	0,966	0,047	101 54
Flow-Mediated Di-lation in active FA	0,00 0,04	0,04 0,01	0,07 0,04	0,105 0,03	0,090	0,990	0,660	0,570	0,510	0,044 0,172
BP diastolic after load 0,5 W/kg	84,6 2,4	80,8 0,4	81,3 1,3	75,8 1,7	0,090	0,988	0,857	0,464	0,211	81,1 6,1
BP diastolic after load 1,5 W/kg	85,4 2,0	82,4 0,5	82,7 1,3	75,2 1,6	0,090	0,986	0,994	0,396	0,173	82,6 6,9
BP diastolic basal, mmHg	80,0 2,3	81,2 0,7	83,0 2,2	71,5 3,8	0,091	0,997	0,199	0,897	0,159	79,0 6,6
Kerdö VI basal, %	-24,2 7,0	-16,0 1,7	-16,1 4,8	-1,8 7,0	0,091	0,995	0,373	0,772	0,633	-16 24
Sodium Erythrocy-tes, mM/L	16,9 0,7	18,9 0,2	20,8 0,9	22,6 2,4	0,090	0,991	0,674	0,569	0,481	19,1 3,5
Ca-ATPase Eryth-roc. ghost, M _p /L•h	1,07 0,08	1,15 0,03	1,27 0,06	1,16 0,17	0,090	0,994	0,453	0,715	0,812	1,16 0,34
Na,K-ATPase Eryt-hroc. ghost, M _p /L•h	0,96 0,06	1,06 0,02	1,20 0,06	0,86 0,12	0,090	0,991	0,665	0,574	0,520	1,06 0,31
Triglycerides, Z	2,78 0,77	0,59 0,10	0,80 0,15	0,33 0,20	0,091	0,999	0,079	0,971	0,035	0 2

α -Lipoproteins Cholesterol, mM/L	1,12 0,10	1,41 0,03	1,50 0,07	1,62 0,09	0,091	0,996	0,268	0,848	0,019	1,39 0,41
β -Lipoproteins Cholesterol, Z	-1,98 0,33	-0,35 0,12	-0,29 0,44	-0,65 0,56	0,090	0,994	0,460	0,777	0,600	0 2
β -Lipoproteins Cholesterol, mM/L	2,18 0,23	3,18 0,07	3,07 0,11	3,02 0,36	0,090	0,999	0,199	0,848	0,053	3,50 0,67
Cholesterol total, Z	-0,84 0,17	-0,10 0,07	-0,12 0,14	-0,13 0,38	0,091	0,999	0,042	0,988	0,061	0 2
Non α -LP / α -LP as Klimov Atherogen I	3,53 0,35	2,99 0,08	2,72 0,15	2,28 0,26	0,091	0,998	0,152	0,928	0,053	2,91 0,36
Metabolic Syndrome Index-4, Z	0,88 0,28	0,34 0,05	0,33 0,12	-0,47 0,18	0,091	0,995	0,347	0,791	0,031	0 2
Sex Index (M=1; W=2)	1,81 0,09	1,49 0,04	1,52 0,09	1,67 0,22	0,090	0,992	0,593	0,620	0,224	1,53 0,50

The identifying information contained in the 23 discriminant variables is condensed into three roots. The major root contains 78,0 % of discriminatory opportunities ($r^* = 0,888$; Wilks' $\Lambda = 0,091$; $\chi^2_{(69)} = 536$; $p < 10^{-6}$), second root 14,3 % ($r^* = 0,638$; Wilks' $\Lambda = 0,432$; $\chi^2_{(44)} = 187$; $p < 10^{-6}$), and third root 7,7 % ($r^* = 0,520$; Wilks' $\Lambda = 0,729$; $\chi^2_{(21)} = 71$; $p < 10^{-6}$).

Raw coefficients and constants for discriminant Variables (Table 4) allow us to calculate individual values of discriminant roots and visualize them in the information field (Fig. 2).

Table 4

Standardized and raw coefficients and constants for Variables

Coefficients Variables	Standardized			Raw		
	Root 1	Root 2	Root 3	Root 1	Root 2	Root 3
PWC ₁₅₀ , Z	1,134	-0,025	0,039	2,280	-0,049	0,079
Magnesium Serum, mM/L	0,190	0,113	-0,918	2,011	1,198	-9,716
Calcium Serum, mM/L	-0,416	-1,201	-0,061	-1,189	-3,437	-0,174
lgTG/ α -LP as Dobiášová&Frolich AGI	0,489	-0,274	-0,230	1,595	-0,894	-0,750
Kerdö VI after load 1,5 W/kg, %	-0,213	-0,765	0,412	-0,031	-0,113	0,061
Mg-ATPase Erythrocyte ghost, MP/L·h	0,073	-0,295	-0,009	0,428	-1,718	-0,055
Reactive BF in active FA, mL/min	-0,113	0,733	-0,135	-0,002	0,016	-0,003
BP systolic after load 0,5 W/kg, mmHg	-0,251	-0,408	0,255	-0,023	-0,037	0,023
BP systolic basal, mmHg	0,098	-0,005	0,479	0,006	-0,0003	0,027
Metabolic Syndrome Index-5, Z	-0,088	0,428	-0,595	-0,130	0,630	-0,876
TG/ α -LP, Z	-0,270	-0,831	0,180	-0,056	-0,171	0,037
Non α -LP / α -LP, Z	0,649	0,421	-0,045	0,385	0,250	-0,027
BP systolic after load 1,5 W/kg, mmHg	-0,023	-0,231	-0,322	-0,001	-0,014	-0,019
Uricemia, Z	1,211	1,254	-0,888	0,638	0,661	-0,468
Uricemia, μ M/L	-1,211	-1,031	0,992	-0,015	-0,013	0,013
Sodium Serum, mM/L	0,364	0,289	0,206	0,023	0,018	0,013
Flow Mediated Dilation in passive FA	0,194	-0,139	0,599	1,031	-0,739	3,190
Basal BF in passive forearm, mL/min	0,102	-0,571	0,997	0,004	-0,022	0,038
Basal BF in active forearm, mL/min	-0,086	0,238	-0,587	-0,0017	0,0047	-0,0115

Nona-Lipoproteins, units	0,476	-0,637	-0,432	0,033	-0,044	-0,030
Cholesterol total, mM/L	-0,755	0,285	0,544	-0,763	0,288	0,550
α-Lipoproteins Cholesterol, Z	0,760	-0,207	-0,474	0,745	-0,203	-0,464
Triglycerides, mM/L	-0,410	0,438	0,274	-0,444	0,474	0,296
		Constants		8,104	20,29	-6,300
		Eigenvalues		3,747	0,688	0,371
		Cumulative Proportions		0,780	0,923	1

Table 5 shows the correlation coefficients of Z-scores of discriminant variables with canonical discriminant roots and the cluster centroids of roots. Following the algorithm of our group, the table also included extra-model variables.

Table 5

Correlations Variables-Canonical Roots, Means of Roots and Z-scores of Variables

Variables	Correlations Variables-Roots			Pessimal (21)	Normal (180)	Optimal (31)	Anomal (6)
	R 1	R 2	R 3				
Root 1 (78,0%)				-3,81	-0,30	2,98	6,84
PWC ₁₅₀	0,88	-0,05	0,13	-1,52 ± 0,10	0,00 ± 0,04	1,35 ± 0,05	3,32 ± 0,34
Calcium Serum	0,39	-0,34	-0,01	-0,62 ± 0,16	-0,19 ± 0,05	0,96 ± 0,17	2,83 ± 0,97
Sodium Serum	0,32	-0,12	-0,03	-0,82 ± 0,19	-0,12 ± 0,05	0,87 ± 0,21	2,12 ± 0,80
Basal BF in passive FA	0,30	-0,09	0,08	-0,85 ± 0,17	-0,09 ± 0,06	0,71 ± 0,20	2,17 ± 0,32
Reactive BF in active FA	0,25	0,12	0,02	-1,08 ± 0,12	-0,02 ± 0,07	0,63 ± 0,22	1,26 ± 0,19
Reactive BF in passiv FA				-0,93 ± 0,11	0,00 ± 0,07	0,47 ± 0,26	0,67 ± 0,21
Basal BF in active FA	0,19	0,09	-0,12	-0,81 ± 0,13	-0,04 ± 0,06	0,65 ± 0,29	0,73 ± 0,26
FMD in active FA				-0,25 ± 0,21	0,00 ± 0,07	0,13 ± 0,22	0,35 ± 0,16
α-LP Cholesterol	0,12	0,14	0,05	-0,72 ± 0,23	0,07 ± 0,08	0,31 ± 0,18	0,58 ± 0,19
Sodium Erythrocyte				-0,64 ± 0,19	-0,06 ± 0,06	0,47 ± 0,25	0,99 ± 0,68
Kerdö VI basal				-0,33 ± 0,29	0,02 ± 0,07	0,01 ± 0,20	0,61 ± 0,29
Mg-ATPase	-0,26	-0,06	0,14	0,96 ± 0,27	0,07 ± 0,05	-0,85 ± 0,25	-1,17 ± 0,47
Kerdö VI after load 1,5	-0,23	-0,10	0,17	0,91 ± 0,26	0,06 ± 0,06	-0,18 ± 0,15	-0,84 ± 0,60
TG/α-LP	-0,16	-0,38	-0,18	1,37 ± 0,54	-0,12 ± 0,05	-0,13 ± 0,06	-0,39 ± 0,05
IgTG/α-LP	-0,10	-0,30	-0,25	1,02 ± 0,26	-0,12 ± 0,07	0,09 ± 0,10	-0,33 ± 0,13
Nona-LP /α-LP	-0,11	-0,09	-0,03	1,21 ± 0,61	0,14 ± 0,12	-0,32 ± 0,21	-0,82 ± 0,45
Triglycerides	-0,10	-0,30	-0,24	2,78 ± 0,77	0,59 ± 0,10	0,80 ± 0,15	0,33 ± 0,20
Met Syndrome Index-5	-0,10	-0,07	-0,26	0,77 ± 0,23	0,30 ± 0,05	0,36 ± 0,12	-0,39 ± 0,19
Met Syndrome Index-4				0,88 ± 0,28	0,34 ± 0,05	0,33 ± 0,12	-0,47 ± 0,18
Nona-Lipoproteins	-0,04	-0,20	-0,25	0,64 ± 0,28	-0,11 ± 0,07	0,23 ± 0,19	-0,21 ± 0,48
Uricemia	-0,11	0,10	0,07	0,47 ± 0,57	0,27 ± 0,14	-0,76 ± 0,29	-1,65 ± 0,52
BP systolic after load 1,5	-0,10	-0,01	-0,26	0,50 ± 0,35	-0,02 ± 0,06	0,04 ± 0,18	-1,17 ± 0,36
BP systolic after load 0,5	-0,09	-0,04	-0,19	0,49 ± 0,37	-0,03 ± 0,06	-0,01 ± 0,19	-0,87 ± 0,42
BP diastol after load 0,5				0,57 ± 0,38	-0,06 ± 0,06	0,04 ± 0,22	-0,86 ± 0,28
BP diastolic afte load 1,5				0,41 ± 0,29	-0,02 ± 0,07	0,02 ± 0,18	-1,08 ± 0,24
Root 2 (14,3%)	R 1	R 2	R 3	-1,99	0,37	-0,21	-2,93
BP systolic basal	-0,02	0,24	0,03	-0,12 ± 0,18	0,51 ± 0,09	0,23 ± 0,18	-0,48 ± 0,22
FMD in passive FA	0,01	0,26	0,05	-0,58 ± 0,17	0,10 ± 0,08	0,07 ± 0,03	0,08 ± 0,02
Cholesterol total	0,08	0,21	0,05	-0,84 ± 0,00	-0,10 ± 0,07	-0,12 ± 0,14	-0,13 ± 0,38
Sex Index				0,56 ± 0,18	-0,07 ± 0,07	-0,03 ± 0,18	0,27 ± 0,42
Root 3 (7,7%)	R 1	R 2	R 3	-0,43	0,19	-1,24	2,20
Magnesium Serum	0,19	0,21	-0,50	-0,83 ± 0,11	-0,07 ± 0,05	0,97 ± 0,27	-0,34 ± 0,24
BP diastolic basal				0,15 ± 0,36	0,34 ± 0,11	0,61 ± 0,33	-1,14 ± 0,58
Na,K-ATPase Eryt. ghost				-0,31 ± 0,17	-0,01 ± 0,07	0,46 ± 0,20	-0,63 ± 0,40
Ca-ATPase Erythr. ghost				-0,27 ± 0,24	-0,02 ± 0,07	0,34 ± 0,18	0,01 ± 0,49
Potassium Erythrocyte				-0,68 ± 0,12	0,04 ± 0,07	0,23 ± 0,24	-0,01 ± 0,75

As we can see (Fig. 2), the clusters are quite clearly delimited along the axis of the first root.

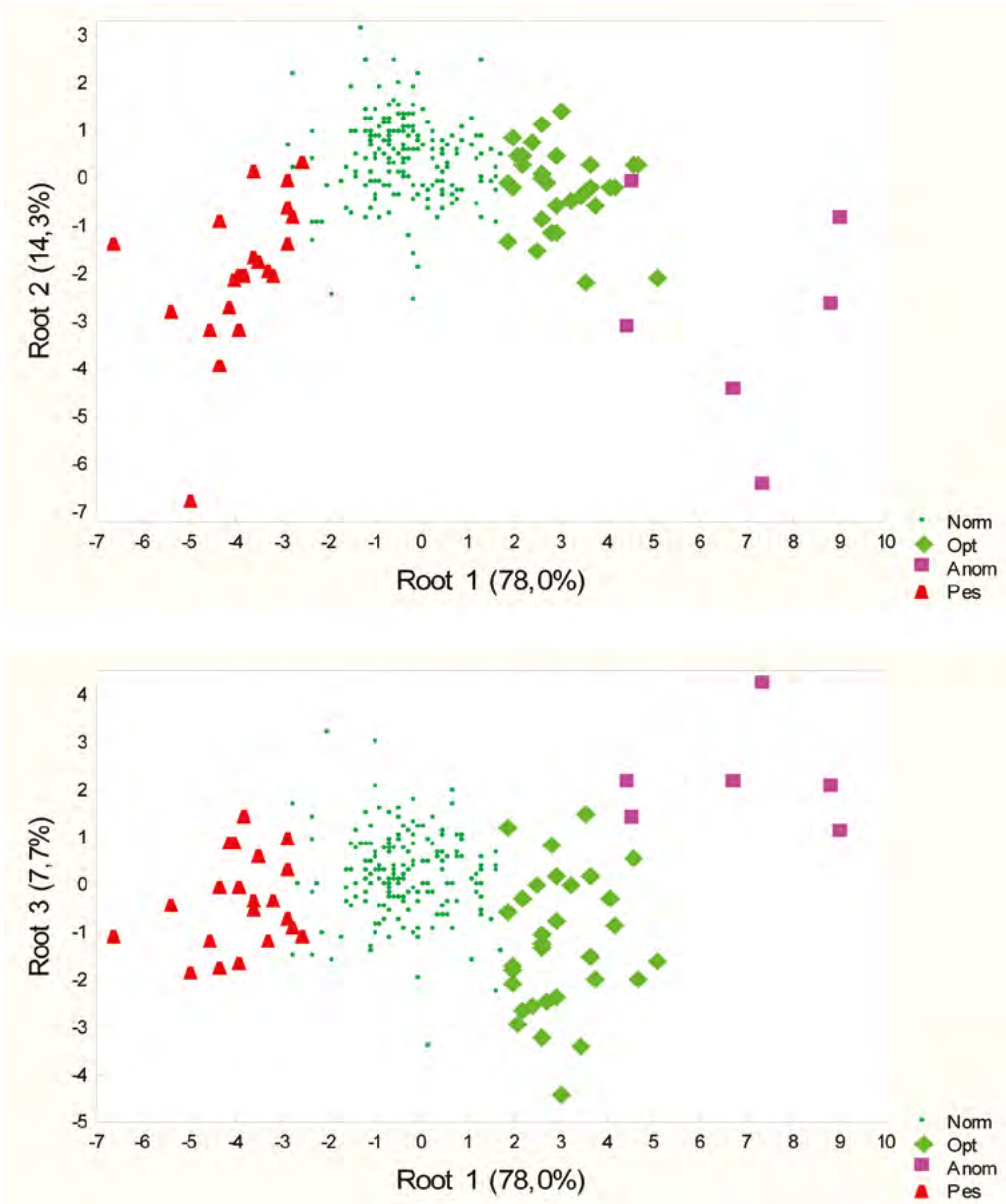


Fig. 2. Scattering of individual values of the discriminant roots, which contain condensed information about hemodynamic and metabolic predictors and accompaniment of PWC_{150} level

Patients with a normal PWC_{150} level are further demarcated from others along the second root axis, and with an optimal PWC_{150} level – along the third root axis.

As a result, the demarcation of the four clusters in the information field of the three roots turned out to be very clear, which is documented by the calculation of Mahalanobis distances (Table 6).

Table 6

Squared Mahalanobis Distances between Clusters and **F-values**
(df = 23,2; for all p < 10⁻⁶)

Clusters of PWC ₁₅₀	Normal (180)	Optimal (31)	Anomal (6)	Pessimal (21)
Normal (180)	0	13,1	65,8	18,3
Optimal (31)	13,6	0	34,1	50,0
Anomal (6)	15,1	6,75	0	121,3
Pessimal (21)	13,6	24,6	22,3	0

Another consequence of discriminant analysis is the possibility to retrospectively identify each level (cluster) of PWC₁₅₀ in each patient. This goal of discriminant analysis is realized with the help of coefficients and constants of classification functions (Table 7).

Table 7

Coefficients and Constants for Classification Functions for Clusters of PWC₁₅₀

Clusters of PWC ₁₅₀	Normal (180)	Optimal (31)	Anomal (6)	Pessimal (21)
Variables currently in the model	p = ,756	p = ,130	p = ,025	p = ,088
PWC ₁₅₀ , Z	−10,08	−2,697	6,513	−18,03
Magnesium Serum, mM/L	150,3	170,0	141,1	146,3
Calcium Serum, mM/L	81,86	80,22	84,38	94,27
IgTG/α-LP as Dobiášová&Frolich AGI	−65,79	−58,98	−52,97	−68,83
Kerdö VI after load 1,5 W/kg, %	7,553	7,429	7,823	7,893
Mg-ATPase Erythrocyte ghost, MP/L•h	278,5	281,0	287,2	281,1
Reactive BF in active FA, mL/min	−0,373	−0,386	−0,449	−0,401
BP systolic after load 0,5 W/kg, mmHg	2,865	2,778	2,871	3,020
BP systolic basal, mmHg	0,579	0,558	0,673	0,543
Metabolic Syndrome Index-5, Z	−34,38	−33,92	−39,15	−34,87
TG/α-LP, Z	7,284	7,149	7,526	7,860
Nona-LP /α-LP, Z	−37,68	−36,52	−35,81	−39,61
BP systolic after load 1,5 W/kg, mmHg	0,458	0,489	0,455	0,508
Uricemia, Z	−83,27	−80,90	−81,84	−86,79
Uricemia, μM/L	2,029	1,969	1,988	2,106
Sodium Serum, mM/L	1,937	1,984	2,069	1,803
Flow-Mediated Dilation in passive FA	59,35	58,61	75,56	55,50
Basal BF in passive forearm, mL/min	0,827	0,798	1,004	0,841
Basal BF in active forearm, mL/min	−0,367	−0,359	−0,418	−0,365
Nona-Lipoproteins, units	0,179	0,357	0,503	0,186
Cholesterol total, mM/L	47,82	44,37	42,53	49,49
α-Lipoproteins Cholesterol, Z	−63,15	−59,93	−58,10	−65,01
Triglycerides, mM/L	18,91	16,75	14,77	19,17
Constants	−1243	−1226	−1298	−1327

Точність класифікації становить 96,6 % (Table 8).

Table 8

Classification matrix. Rows: Observed classification;
Columns: Predicted classification

Clusters of PWC ₁₅₀	Percent correct	Normal p = ,756	Optimal p = ,130	Anomal p = ,025	Pessimial p = ,088
Normal (180)	98,3	177	2	0	1
Optimal (31)	96,8	1	30	0	0
Anomal (6)	83,3	0	1	5	0
Pessimial (21)	85,7	3	0	0	18
Total	96,6	181	33	5	19

Therefore, the quantitative-qualitative level of PWC₁₅₀ is determined by a set of functional and metabolic factors, which fits perfectly into the concept of the functional-metabolic continuum of Gozhenko AI [29].
At the final stage, the possibility of predicting not only the qualitative level of PWC₁₅₀, but also the quantitative one was clarified. First, a matrix of significant correlations was created through screening (Table 9).

Table 9

Classification matrix for predictors of PWC₁₅₀

Variable	r
Calcium Serum, mM/L	0,606
Basal BF in passive FA, mL/min	0,577
Reactive BF in active FA, mL/min	0,527
Sodium Serum, mM/L	0,509
Chloride Serum, mM/L	0,500
Sodium Erythrocyte, mM/L	0,492
Basal BF in active FA, mL/min	0,469
Reactive BF in passive FA, mL/min	0,449
Magnesium Serum, mM/L	0,395
Potassium Erythrocyte, mM/L	0,288
β-Lipoproteins Cholesterol, mM/L	0,254
α-Lipoproteins Cholesterol, Z	0,253
β-Lipoproteins Cholesterol, Z	0,229
α-Lipoproteins Cholesterol, mM/L	0,186
Sex Index (M = 1; W = 2)	-0,624
Mg-ATPase Erythrocyte ghost, MP/L·h	-0,439
TG/α-LP, Z	-0,338
TG/α-LP	-0,329
Triglycerides, Z	-0,323
Triglycerides, mM/L	-0,285
lg TG/α-LP	-0,285
Metabolic Syndrome Index-5, Z	-0,240
Metabolic Syndrome Index-4, Z	-0,225

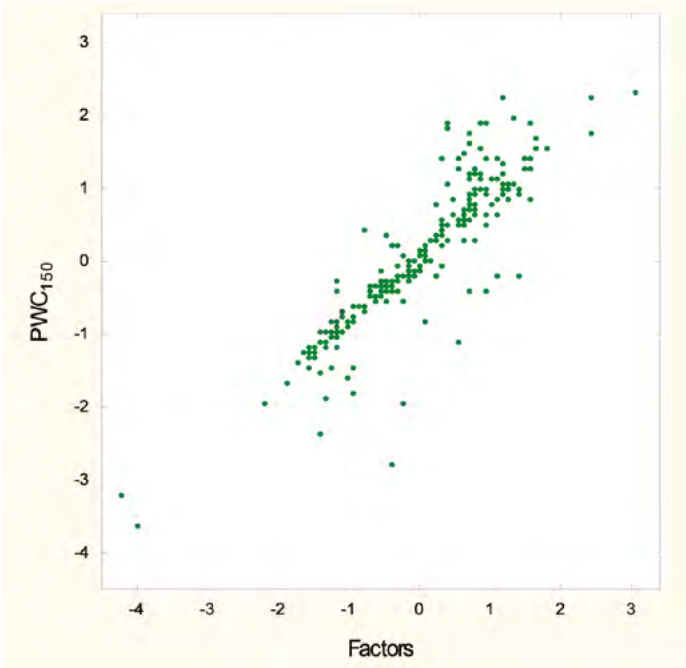
A regression model was then built by stepwise elimination until the maximum Adjusted R² level was reached (Table 10).

Table 10

Regression Summary for absolute level of PWC₁₅₀ R = 0.894; R² = 0.799;
Adjusted R² = 0.794; F_(6,2) = 153; p < 10⁻⁶; SE of estimate = 0.33 W/kg

N = 238		Beta	St. Err. of Beta	B	St. Err. of B	t ₍₂₃₁₎	p-level
Variables	r	Intercpt		1,916	0,163	11,8	10 ⁻⁶
Calcium Serum, mM/L	0,606	0,461	0,042	0,756	0,068	11,1	10 ⁻⁶
Basal BF in passive forearm, mL/min	0,577	0,240	0,050	0,0058	0,0012	4,79	10 ⁻⁵
Basal BF in active forearm, mL/min	0,469	-0,165	0,053	-0,0022	0,0007	-3,11	0,002
Potassium Erythrocyte, mM/L	0,228	0,077	0,040	0,0020	0,0010	1,94	0,053
Sex (M = 1; W = 2)	-0,624	-0,578	0,031	-0,8462	0,0450	-18,8	10 ⁻⁶
TG/α-LP ratio	-0,329	-0,231	0,030	-0,1206	0,0157	-7,66	10 ⁻⁶

As a result, it was found that the upregulating actotropic factors are basal blood flow in the forearms (probably in all limbs), calcemia and erythrocyte kalihistia (probably also in myocytes). In contrast, the downregulating actotropic factors are female sex and plasma atherogenicity. Interestingly, the model included the TG/α-LP ratio, but not its logarithm. Taken together, these factors explain 79.9 % of the variability of PWC₁₅₀ (Fig. 3) and allow predicting its level with an error of 0.33 W/kg.



R = 0,894; R² = 0,799; $\chi^2_{(6)} = 368$; p < 10⁻⁶; Λ Prime = 0,206

Fig. 3. Scatterplot of canonical correlation between hemodynamic and metabolic factors (X-line) and PWC₁₅₀ (Y-line)

Conclusion. In patients of the Truskavets’ spa, the level of PWC₁₅₀ is in a wide range – from low to abnormally high, and is naturally associated with deviations in a number of hemodynamic and metabolic parameters caused by chronic diseases, as well as sex. Targeted influence on the identified predictors will probably make it possible to increase the efficiency of recovery or further increase PWC₁₅₀.

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