

RESEARCH ARTICLE

Detoxification by Medicinal Leeches (*Hirudo medicinalis*)**Authors**

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Abstract

The success of body detoxification by medicinal leeches (*Hirudo medicinalis*) depends on the rich bioactive composition of the medicinal leeches salivas. There is a total of 150 identified bioactive substances that have detoxifying and regenerative effects on the human body. However, the human organism has its own regulatory mechanisms for removing toxic products from the body: detoxification. The aim of our explorative study was to advance these mechanisms and demonstrate the efficiency of the bioactive substances of leech saliva to support the regulation of selected blood parameter levels, i.e. lower cholesterol, triglycerides and glucose levels, and to ensure the prevention of diseases caused by the fluctuation of the levels of these blood components.

The participants in our study numbered 17. None of them had presented any significant medicinal complications from or contraindications to leech therapy. They participated in a total of seven leech procedures. After the therapy, blood tests for the patients were carried out to compare the blood counts before therapy and after. Using the results, we evaluated the outcomes of the detoxification therapy by using statistical analysis in the R program, version 3.0.0.

Detoxication therapy using medicinal leech (*Hirudo medicinalis*) had no significant effect on the level of blood components provided to us, but its effect was observed in the subjective health improvements reported by patients.

Keywords: detoxification, *Hirudo medicinalis*, bioactive substances, blood count

Introduction

The term **detox** refers to the process of alternative treatment of addicted patients [13]. A healthy organism can deal with the accumulated waste substances by its own regulatory mechanisms. If these mechanisms do not work well, some blood parameters, such as glucose and fat, including cholesterol and triglycerides, may increase. Hyperlipidemia has almost no symptoms but its presence poses an increased risk of cardiovascular disease. Elevated blood glucose levels are also a major and common problem. Recurrent hyperglycaemia increases the risk of developing diabetes mellitus, but also heart attack, stroke, kidney, eye and lower limb damage [5, 6, 9, 10]. All these risks can be combined into one and cause so-called metabolic syndrome [7].

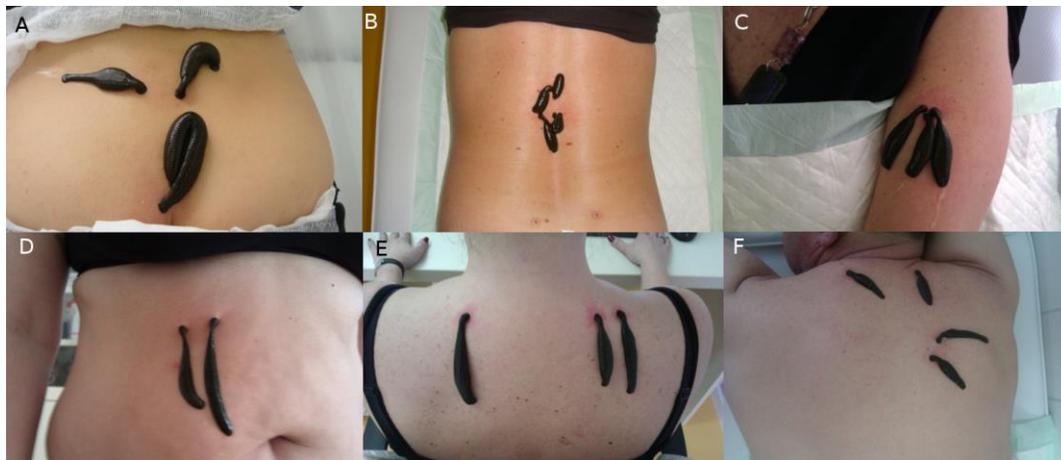
In our study, a selected group of patients underwent detoxification therapy with a medical leech (*Hirudo medicinalis*). Several dozen bioactive substances that have a positive effect on fluctuating levels of blood parameters can be found in the salivary glands of leeches. At the same time, following therapy, there is an overall improvement in health: normal temperature is restored, blood pressure is lowered, sugar levels are stabilized, cholesterol is lowered,

metabolism is improved, and the activity of the immune, hormonal and nervous systems is increased. These and many other positive improvements in the body occur due to the mentioned complex of bioactive substances in the salivary glands of leeches which have antibacterial, bacteriostatic, microcirculatory and, especially, detoxifying effects, having a beneficial effect on the whole organism. After therapy, there are subjective changes and an improvement in the health of the organism. Because of these substances and enzymes, the action of leeches on the body may be considered complex.

Detoxifying organs

The body is equipped with self-regulating mechanisms that can withstand the effects of the external environment, caused by bacteria, viruses or excessive physical activity, all of which are part of our daily lives. Through urine, stool, sweating and breathing, the body discharges a substantial amount of waste every day. Therefore, it is very important to encourage our natural detoxification mechanisms, including the actions of the liver, lungs, skin, kidneys, intestines and the lymphatic system (Fig. 1), to function optimally [16].

Figure 1. Detoxification zones: A) coccyx and cruciate zone B) kidney zone C) heart zone (left hand) D) liver zone E) trapeze zone E) right scapular zone



Bioactive composition of leech saliva

The salivary glands of the medical leech include a whole complex of bioactive substances (see Table 1), which have a beneficial and detoxifying effect on the whole

organism. When sucking blood, the leeches simultaneously discharge a complex of bioactive substances into the patient's body within 20 - 45 minutes.

Table 1: Bioactive composition of the salivary glands of leeches and their detoxifying effect on the organism [14].

Bioactive substance	Effect
Hirudin, Hirustazin	Anticoagulant
Destabilase	Antiischemic
Bdelin	Antibacterial
Hyaluronidase	Antithrombotic
Apyrase	Immunostimulatory
Kinin	Analgesic
Eglin	Anti-inflammatory
Collagen	Anti-swelling
Triglyceride	Detoxifying
Lipase and esterase	Microcirculatory
Lipolytic enzyme	Analgetic

It should be noted that hirudin, triglyceride, lipase, esterase and destabilase, are those components most involved in the detoxification of the body. They are involved in regulating blood pressure, stabilizing the level of sugar, cholesterol and triglycerides in the blood, increasing the activity of the immune system and contributing to the overall regeneration of the body.

Hirudin is an important component of the salivary glands of leeches. It is a substance with an extremely strong anticoagulant effect and belongs to the first

biological molecules, the structure and function of which we have known about for several decades [11]. The anticoagulant properties of leech saliva were discovered in the late 19th century by Haycraft [8]. In 1950, Fritz Markwardt of Germany isolated a protein from the saliva of *Hirudo medicinalis*, which he named hirudin, after the Latin generic name of the medical leech *Hirudo medicinalis* [3, 4, 12]. Due to its affinity for thrombin, hirudin inhibits almost all the physiological functions of thrombin and ensures blood clotting.

No adverse side effects, including platelet effects, have been observed with its application. Hirudin mixes with the victim's blood during food intake and, when swallowed, is kept in a liquid state in the stomach so that the leech retains its full mobility while digesting a large volume of food intake. No other molecule has such a high specificity for thrombin [18].

Destabilase has glycosidase activity and is an enzyme characterized by a strong anticoagulant effect on blood clotting, acting to break down clotted blood [1, 19]. It also contains a unique compound, prostaglandin, which significantly regulates blood vessels, the digestive tract and also regulates blood sugar levels. The most mysterious property of prostaglandin is the self-regulation of blood pressure: under its effects, high pressure decreases and low pressure increases.

Esterase and lipase are involved in the breakdown of fats and lower cholesterol [2] and also have a detoxifying effect.

The goal

Monitored parameters

As already mentioned, the salivary glands of leeches successfully regulate the levels of various blood components, such as glucose, cholesterol and triglycerides, and contribute to the overall renewal of cells, tissues and organs in the body and contribute to the overall revitalization of the body. In this study, we look to confirm these claims.

Triglycerides and cholesterol

Cholesterol is a fat-soluble steroidal animal alcohol. It is the body's own substance which is formed not only in the liver but also in the skin, adrenal cortex, intestines and other organs. The daily synthesis fully covering the body's need is about 1 g; about 0.3 g of cholesterol is ingested by food. Cholesterol is involved in the construction of cell membranes, is a precursor of steroid hormones and vitamin D3. A large amount of cholesterol is metabolized by the liver into

bile acids which, together, provide the emulsifying properties of bile. In addition to bile acids, cholesterol produces neutral sterols and both types of compounds are excreted in the feces [16, 17]. There are two types of cholesterol in the blood that differ in the lipoproteins that carry it:

- LDL, or harmful cholesterol, is associated with low-density lipoproteins and promotes arteriosclerosis.

- HDL, or good cholesterol, is associated with high-density lipoproteins and protects against arteriosclerosis.

The expression **blood cholesterol level** describes the total amount of cholesterol, the sum of both species [15, 16].

Cholesterol is needed in the body and, as such, should not be considered a toxic substance. The body is able to produce enough cholesterol for its own needs without being dependent on consumption from external sources. Cholesterol is harmful only because it settles on the walls of the arteries and causes arteriosclerosis. Exceeding a certain level in the blood increases the risk of arteriosclerosis and heart attack. Cholesterol is needed, but only in an amount that does not cause arteriosclerosis [15]. Esterase and lipase contribute to its reduction.

Triglycerides, along with cholesterol and phospholipids, are part of lipoproteins. Their increased level contributes to arteriosclerosis and, subsequently, to a heart attack or stroke. Triglycerides chemically consist of glycerin and fatty acids. They are found in all fats and are also a major component of oils [15, 16].

Glucose

Glucose is one of the carbohydrates, which are essential nutrients in the body. It is the most important simple blood sugar. It enters the body through food from the digestive system by converting simple or complex sugars. After absorption from the small intestine, it is taken up by liver cells

(80%), muscles, adipose and brain tissue. It is used as a direct source of energy or is stored in the form of glycogen and fatty acids. Glucose is the primary substrate for the central nervous system (CNS), erythrocytes, the immune system and for the supply of oxaloacetate to the Krebs cycle. Glucose is chemically a monosaccharide or simple sugar that is able to enter the bloodstream directly, without digestive processes [15].

Material and methods

The set of patients

We enrolled 17 patients in the study (11 women and 6 men), none of whom had health complications from or contraindications to hirudotherapy, including insect bite allergy, anemia, hemophilia, diabetes, cancer, poorly healing wounds, pregnancy, AIDS [14]. Prior to the detoxification treatment, they underwent blood tests which revealed increased or decreased levels of certain parameters that

could be regulated by leeches. These were mainly elevated levels of glucose, cholesterol and triglycerides.

The study was approved by the ethics committee of the University Hospital of the Merciful Brothers in Bratislava (Reference number 116/2015).

Hirudotherapy - treatment with medical leech (Hirudo medicinalis)

We used pure certified leeches *Hirudo medicinalis* (Linné 1758) and *Hirudo verbana* (Carena 1820) for the treatment. We applied the leeches to precisely determined parts of the body (see Table 2) and, after being attached to the patient's body, they sucked blood, usually for 20-45 minutes, until they excreted the required amount of bioactive substances into the body. We used each leech once only. Before the application itself, we disinfected the leeches for 2 - 5 minutes in a blue rock solution to get rid of impurities from the body surface.

Table. 2. Detoxification table – detoxification sites of application

Application order	Place of attachment	Number of leeches	Distance between applications
1.	liver (under the rib arch)	3	4-7 days
2.	coccyx (cartilage, cruciate ligaments)	3	4-7 days
3.	kidneys	2-3 each	4-7 days
4.	left hand (chest - heart area)	3	4-7 days
5.	behind the ears	2 on each	4-7 days
6.	liver + sacrum	2	4-7 days
7.	at the right scapula, next to Th3	4	4-7 days

Detoxification sites of application

1. Application in the area of the liver, under the rib arch - application is beneficial for the liver, its function, cleansing, cleansing of the body, total detox. However, it also affects digestion. It is preferred for allergies, skin problems, rashes. 3 medical leeches are added. 2. Application in the area of the coccyx, cartilage, cruciate ligaments - this place is considered universal and very

effective for the whole pelvis and lower limbs. It is preferred for problems with varicose veins or ischemia. It is mainly used when it is not possible to apply it directly to the lower limbs. 3 medical leeches are added. 3. Kidney application - this application is the trigger for strong cleansing processes. It also affects the entire urinary tract and liver. It is the basis of cleansing, helps with all eczema and allergies. 2-3 medical leeches are added

to each kidney. 4. Application in the area of the left hand, biceps and triceps, or on the chest - the area of the heart. At this point there are minimal reactions. It is very suitable for test application of medical leeches, also if we need to quickly get bioactive substances into the body. It is a direct path to the heart and from there the substances spread very quickly throughout the body. 3 medical leeches are added. 5. Applying in the area behind the ears, behind each ear - this place is a highly effective place for the whole head, eyes, ears, cavities, brain, middle ear, migraines, blood vessels of the head. This place is often used for problems with high blood pressure. Attached 2 medical leeches for each ear. 6. Application in the area of the liver and sacral triangle - this triangle has similar properties as the coccyx. Both sites are closest to the rectum and hemorrhoids. Separately, this area is used for the pelvis, solving female and male problems, diseases of the uterus, ovaries, bladder, prostate, fibroids, endometriosis. 2 medical leeches are added. 7. Application in the area of the right scapula, between the sacral area and the scapula and between the ribs Th3 and Th4 - this application is the final point of the detoxification treatment. It is a great place for everything related to the lungs, allergies, asthma, shortness of breath, etc. It has a direct effect on the properties of the blood. It is also called first aid for the lungs. 3 leeches are added.

Statistical analysis

Due to the exploratory nature of this study and lack of prior data on expected effect size, no formal calculation of sample size was done prior to the study. Sample size was guided by practical considerations and duration of recruitment. We prepared the patients for therapy - blood sampling and consultation within two weeks and hirudotherapy alone lasted two months. Statistical analysis was performed in statistical program R, version 3.0.0 (R Core Team, 2013). Prior to analysis, data were

checked for normality and homogeneity of variance using the Shapiro-Wilk and Bartlett assays. The difference in selected blood parameters before and after processing the leeches was evaluated using a paired t-test.

Results

The monitored blood parameters of red and white blood cell count, hemoglobin level, hematocrit, MCV, MCH, MCHC, platelet count, percentage and absolute numbers of different white blood cell types, glucose, cholesterol and triglyceride levels were checked after completion of the detoxification therapy and showed only minor changes that were likely to have no clinical significance (see Table 3). We recorded a slight decrease in the number of red blood cells (on average $-0.05 \times 10^{12} / l$), hemoglobin levels ($-2.41 \text{ g} / l$), as well as a slightly reduced hematocrit (-0.006). These changes were probably related to the prolonged bleeding at the application site of the leeches. Interestingly, changes in these blood parameters were two to four-fold more pronounced in women than in men. Nevertheless, they were not statistically significant. The platelet count decreased by $16.06 \times 10^9 / l$ after the detoxification cycle and this decrease was statistically significant ($P = 0.0183$). This may be related to blood loss, but also to increased platelet consumption when the bleeding stopped after the leech application. Equally significant was the decrease in the percentage of mixophils (-1.41% ; $P = 0.00718$) and the increase in the percentage of neutrophils ($+ 3.62\%$; $P = 0.0473$) in the white blood cells, which may signal the effect of leeches on the immune system. From a clinical point of view, however, these are negligible changes. Blood glucose levels remained virtually unchanged after the therapy (decrease of $-0.006 \text{ mmol} / l$; $P = 0.9417$). We even saw a slight increase in the amount of fat in the blood (total cholesterol: $+0.15 \text{ mmol} / l$; $P = 0.2118$; triglycerides: $+0.017 \text{ mmol} / l$; $P = 0.8498$),

which was contrary to our expectations. Subjective improvements in patients' health played a very important role in assessing the degree of detoxification of the body. They

reported the relieving of fatigue and drowsiness, improved skin condition, and adjusted metabolism and digestion.

Table 3. Results of statistical analysis after „detoxification therapy“

Blood parameter (unit)	Reference interval	Before treatment	After treatment	Difference (95% CI)	P (paired t-test)
WBC (x10 ⁹ /l)	4-10	6.96 (6.16-7.81)	7.21 (6.49-7.97)	+0.05 (-0.55 to +0.75) ♀: +0.0002 (-0.63 to +0.94) ♂: +0.08 (-1.43 to 1.59)	0.8836 ♀: 0.9996 ♂: 0.8927
RBC (x10 ¹² /l)	3.8-4.9 ♀ 4.3-5.7 ♂	4.51 (4.35-4.65) ♀ 4.95 (4.37-5.53) ♂	4.41 (4.20-4.60) ♀ 4.94 (4.43-5.45) ♂	-0.05 (-0.15 to +0.03) ♀: -0.08 (-0.22 to +0.03) ♂: -0.002 (-0.17 to +0.15)	0.2044 ♀: 0.149 ♂: 0.9781
HGB (g/l)	120-160 ♀ 135-175 ♂	137.82 (132.26-143.38) ♀ 146.17 (135.17-157.16) ♂	134.73 (126.21-143.25) ♀ 145.00 (134.38-155.62) ♂	-2.41 (-6.35 to +1.53) ♀: -3.09 (-8.68 to +2.49) ♂: -1.17 (-8.33 to +5.99)	0.2125 ♀: 0.2457 ♂: 0.6928
HCT	0.35-0.47 ♀ 0.40-0.50 ♂	0.411 (0.398-0.424) ♀ 0.437 (0.410-0.464) ♂	0.402 (0.384-0.418) ♀ 0.435 (0.416-0.453) ♂	-0.006 (-0.014 to +0.002) ♀: -0.008 (-0.021 to +0.003) ♂: -0.001 (-0.016 to +0.012)	0.1602 ♀: 0.1378 ♂: 0.8732
MCV (fl)	82-98	90.53 (87.53 to 98.64)	90.92 (88.07 to 93.61)	+0.22 (-0.18 to +0.62) ♀: +0.14 (-0.46 to +0.76) ♂: +0.37 (-0.17 to +0.90)	0.2546 ♀: 0.6069 ♂: 0.1377
MCH (pg)	28.0-34.0	30.75 (29.96 to 31.42)	30.97 (30.07 to 31.74)	-0.04 (-0.55 to +0.46) ♀: +0.03 (-0.59 to +0.64) ♂: -0.17 (-1.40 to +1.07)	0.8648 ♀: 0.9232 ♂: 0.7422

MCHC (g/l)	320-360	336 (329.91 to 342.09)	334.13 (327.52 to 340.74)	-1.87 (-7.57 to +3.83) ♀: -0.27 (-6.82 to +6.27) ♂: -4.8 (-19.14 to +9.54)	0.4963 ♀: 0.9279 ♂: 0.429
9 PLT (×10 ⁹ /l)	150-400	235.45 (211.45 to 265.61)	220.00 (196.78 to 249.43)	-16.06 (-29.01 to -3.10) ♀: -17.73 (-38.48 to +3.02) ♂: -13.00(-23.80 to -2.20)	0.0183 ♀: 0.08612 ♂: 0.0271
LYM (%)	25-40	35.56 (32.74 to 38.39)	33.35 (29.68 to 37.02)	-2.21 (-5.57 do +1.15) ♀: -3.04 (-7.77 to +1.68) ♂: -0.68 (-6.76 to +5.39)	0.1821 ♀: 0.1814 ♂: 0.7842
MXD (%)	2-16.5	10.05 (8.25 to 11.85)	8.64 (7.05 to 10.23)	-1.41 (-2.38 to -0.44)	0.07187
NEUT (%)	47-70	54.38 (51.27 to 57.50)	58.01 (53.87 to 62.15)	+3.62 (+0.05 to +7.20) ♀: +4.72 (-0.31 do +9.94) ♂: +1.62 (-4.61 do +7.85)	0.0473 ♀: 0.0630 ♂: 0.5343
LYMabs (×10 ⁹ /l)	0.8-4.0	2.47 (2.17 to 2.77)	2.36 (2.13 to 2.59)	-0.11 (-0.33 to +0.11) ♀: -0.15 (-0.42 to +0.13) ♂: -0.05 (-0.59 to +0.49)	0.3013 ♀: 0.2593 ♂: 0.8207
MXDabs (×10 ⁹ /l)	0.18-1.09	0.70 (0.57 to 0.83)	0.60 (0.50 to 0.70)	-0.10 (-0.20 to +0.01)	0.07526
NEUTabs (×10 ⁹ /l)	2.0-7.0	3.88 (3.26 to 4.50)	4.32 (3.63 to 5.01)	+0.20 (-0.31 to +0.83) ♀: +0.35 (-0.31 to +1.23) ♂: -0.05 (-0.84 to +1.41)	0.4558 ♀: 0.3087 ♂: 0.9087
S-GLU (mmol/l)	4.0-5.5	5.21 (5.04 to 5.44)	5.32 (5.07 to 5.57)	-0.006 (-0.18 + to 0.18)	0.9417 ♀: .3941 ♂: 0.2878

				♀: +0.09 (-0.13 to +0.35) ♂: -0.16 (-0.49 to +0.18)	
S-KREA (umol/l)	44-80 ♀ 62-106 ♂	68.48 (61.18-75.78) ♀ 91.87 (80.49-103.25) ♂	67.75 (61.81-73.70) ♀ 88.98 (76.38-101.59) ♂	-1.49 (-4.74 to +1.77) ♀: -0.73 (-4.64 to +3.18) ♂: -2.88 (-10.80 to +5.04)	0.3469 ♀: 0.6872 ♂: 0.3924
S-KM (umol/l)	140-390 ♀ 200-420 ♂	276.21 (229.56-322.86) ♀ 372.57 (316.18-428.96) ♂	292.74 (257.91-327.57) ♀ 370.24 (300.50-439.98) ♂	+14.95 (-4.63 to +32.44) ♀: +21.87 (-2.78 to +43.40) ♂: -2.32 (-40.92 to +36.27)	0.1199 ♀: 0.0740 ♂: 0.883
S-CHOL (mmol/l)	3.5-5.0	4.77 (4.33 to 5.20)	4.91 (4.50 to 5.33)	+0.15 (-0.09 to +0.39) ♀: +0.10 (-0.23 to +0.43) ♂: +0.23 (-0.24 to 0.70)	0.2118 ♀: 0.5057 ♂: 0.2652
S-TAG (mmol/l)	0.45-1.70			+0.017403 (-0.163617 to +0.228581)	0.8498 1 pc less

Abbreviations

W: women
M: men
WBC: Leukocytes
MXD: Number of mean leukocytes, number of mixed cells
RBC: Erythrocytes
NEUT: Relative neutrophil count
HGB: Hemoglobin
LYMabs: Absolute lymphocyte count
HCT: Hematocrit
MXDabs: Medium leukocyte ratio, percentage of mixed cells

MCV: Mean erythrocyte volume
NEUTabs: Absolute neutrophil count
MCH: Medium concentration
S-GLU: Glucose hemoglobin in erythrocytes
S-KREA: Creatinine
MCHC: Medium color concentration hemoglobin
PLT: Platelets
S-KM: Uric acid
LYM: Relative lymphocyte count
S-CHOL: Cholesterol

Discussion

The goal of various forms of detox therapies is to resolve chronic ailments and to stimulate the metabolism and the immune system. These humoral treatments draw from centuries old traditions. Renowned ancient healers such as Hippocrates and Galen applied these therapies with conviction. To date, many scientific studies have shed light on the effect mechanisms of leeches. Although more than 100 particular proteins with different molecular masses are observed in leech secretions, only a few have been identified that have a major active role. Following a leech bite, it has to establish a sucking pathway (extracellular matrix degradation); inhibit adhesion, aggregation, and coagulation (inhibition of platelet functions, and anticoagulant effect); increase blood flow; protect itself (antimicrobial activity); and avoid detection (analgesic and anti-inflammatory effects). Medical leeches do not only treat a diseased organ. It is a living, very complex, original, non-specific stimulator, acting on the whole human body. In particular, it acts on the metabolic system in cells, capillaries and lymphatic vessels. The result of treatment with medical leeches - is an increase in immunobiological resistance and restoration of disturbed balance in the body. Examples are the normalization of blood pressure, sugar levels and thus the overall improvement in health. Eliminates other bacteria in the body, inflammatory processes. Reflexion on the body is also important, which leads to adequate reactions of the autonomic and central nervous systems. The main role of bioactive substances from leeches is the sterilization and refining of blood. During its development, the leech had to adapt to the environment and the conditions for survival, so it became a living factory for the production of bioactive substances. The effect of the leech is evident on the blood sample within 30 minutes after application of the leech. We see a clear difference in blood

before and after applying leeches to the body. After the application of leeches, red blood cells are visible, beautifully separated from each other and thus they can better perform their function in the body. Even in case of healthy person, it is possible the therapy with medical leeches (a detoxification treatment).

Conclusion

No similar type of study has yet been carried out. Our goal was to confirm or refute the goals we set. Based on our hypotheses, detoxification therapy using medical leeches (*Hirudo medicinalis*) was supposed to contribute to the regulation of blood fat and glucose levels. These hypotheses were not confirmed in all patients from an objective assessment of their health status based on the results of the blood tests. According to the statistical processing of the blood test results before and after therapy, detoxification therapy did not have a significant effect on the regulation of established levels of blood components. Our claims were not confirmed. This may have been because we applied only one cycle of therapy.

Leeches in detoxification therapy, with their bioactive substances, contribute to improved blood flow to the tissues and the supply of tissues with oxygen and nutrients, to an improvement in heart function due to the mechanical stimulation of the blood flow, and to stimulation of the immune system, which confirms a decrease in the percentage of mixophiles and the percentage in white blood cells. They have bactericidal, anti-inflammatory and anti-edematous effects on the body. Overall, medical leeches have a detoxifying effect. The observed patients reported mainly subjective improvements in health immediately after therapy, but their condition also improved in the following weeks. Based on our experiment, we can still state that detoxification using medical leeches acts using its bioactive substances, which when

sucking blood into the body is beneficial to the health of the body.

Each leech therapy was associated with certain complications and side effects. Patients complained mainly of heavy bleeding from certain parts of the body, especially around the liver, kidneys and lower back, they reported itching a few hours after the treatment and swelling or bruising around the wound area a day or two after the treatment. Some patients experienced fatigue on the day of therapy, which could be due to blood loss and insufficient fluid replenishment.

Detoxification therapy is recommended to be performed twice a year to restore all systems and functions in the body.

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