

CHANGES IN BLOOD PRESSURE WITH COMPENSATORY HEART RATE DECREASE AND IN THE LEVEL OF AEROBIC CAPACITY IN RESPONSE TO REPEATED WHOLE-BODY CRYOSTIMULATION IN NORMOTENSIVE, YOUNG AND PHYSICALLY ACTIVE MEN

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Abstract

Objective: In Poland and all over the world, whole-body cryostimulation is becoming more and more popular in the treatment of different diseases and in sport. However, changes that occur in the human body subjected to cryogenic temperatures are still not completely understood. Therefore, the aim of this study was to evaluate changes in blood circulation and aerobic capacity induced by repeated exposure to whole-body cryostimulation of young and clinically healthy male subjects.

Material and Methods: The study included 25 young men, aged 21 ± 0.9 years, average body weight 74.65 ± 6.98 kg and height 179.5 ± 5.12 cm. The participants were exposed to extremely low temperatures in a cryogenic chamber once a day for 15 days. Each session lasted 3 min at -130°C and was preceded by 30-second, adaptation in a vestibule at -60°C . Blood pressure and heart rate were measured before entering the chamber, immediately after exiting and 10 min later. We also calculated pulse pressure and the mean arterial blood pressure. Before and after the treatment the maximal oxygen uptake was measured. **Results:** Our results showed a significant increase in systolic blood pressure after each cryostimulation (by an average of 19 mmHg) and an increase in diastolic blood pressure only after the first cryostimulation (by 6 mm Hg). The increase in systolic blood pressure was accompanied by a significant decrease in heart rate (by about 7 bpm). No adaptation changes were observed after 15 treatments. There were no changes in aerobic capacity after 15 sessions of WBC, however we observed a significant decrease in RBC and hemoglobin concentration. **Conclusion:** Due to the increase in systolic blood pressure after WBC, this kind of physiotherapy treatment is not recommended for people with advanced or not pharmacologically controlled hypertension.

Key words:

Cryogenic temperature, Cardiovascular response, Aerobic capacity, Hematological indices

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INTRODUCTION

Despite widespread interest in using whole body cryostimulation (WBC) in the treatment of different diseases and in sport, we still do not understand the changes that occur in a body exposed to very low temperatures (-100°C to -160°C). During exposure to cold, normal body temperature is maintained by a complex regulatory system, mainly related to the intensified production and protection of body heat: increased shivering thermogenesis and increased peripheral vasoconstriction. Short cryostimulation, lasting from 1 to 3 min, does not decrease the temperature inside the body, but only reduces the skin temperature. Cutaneous vasoconstriction is maximal below the skin temperature of 31°C and redistributes blood to the core causing increased stroke volumes and changes in heart rate [1]. When the skin tissue temperature drops below 18°C as a result of cooling, the initial reduction in blood flow is followed by a compensatory increase in blood flow. Periodic oscillations in blood flow and skin temperature form an ongoing cycle, hence the name 'hunting reaction' [2].

Literature describes a number of changes occurring in the body after exposure to cold. Usually, the changes concern the endocrine system [3,4,], the immunological system [3–6] lipid profile [7], and hematological values [8–10]. The impact of cryostimulation on the level of physical fitness remains considerably unexplored, both in relation to the components of aerobic and anaerobic capacity. Recent reports [11] suggest a significant increase in anaerobic power and capacity in men.

Cold affects the action of myocardium, especially the sinoatrial node, reducing heart rate (HR) and potentially leading to arrhythmia [12]. In physiotherapeutic practice, it is a standard procedure to test participants before cryostimulation, and before each cryostimulation blood pressure (BP) is measured. Although absolute and relative contraindications to the use of cryotherapy or whole-body cryostimulation do not include unstable blood pressure and hypertension [13], practical observations confirm the occurrence of significant increases in blood pressure in some people, with a simultaneous decrease in heart

rate after cryostimulation. The difficulty with capturing the changes induced by cryostimulation is connected with their short-term nature and the specificity of the procedure which usually involves several people at once and thus the measurements of individual patients are distributed in time. Literature data on the changes in the key cardiovascular indicators in humans exposed to cryogenic temperatures are ambiguous. Westerlund et al. [14], Fricke [15] and Taghawinejad et al. [16] report a significant but short-term increase in systolic and diastolic blood pressure after WBC. Similarly, Komulainen [17] observed a rapid increase in blood pressure in mildly hypertensive subjects exposed to -15°C . Other authors report that the thermal stress (-110°C) does not cause changes in systolic and diastolic blood pressure, but only a decrease in heart rate [18].

In order to determine the safety and real benefits of cryostimulation, we need to examine this problem and find the factual potential increase in blood pressure directly after the whole-body session. In this way, we may prevent undesirable changes in blood pressure, especially in people with unstable blood pressure or mild hypertension.

Therefore, in this study we examined a numerous and homogeneous group of healthy individuals with correct systolic and diastolic blood pressures; measurements were performed in identical time intervals after leaving the cryogenic chamber by each of the participant.

MATERIAL AND METHODS

The study population consisted of 25 healthy men, volunteers, aged 21.3 ± 0.94 years, with normal bodyweight ($\text{BMI} = 23.19 \pm 1.9$), who had never been subjected to any form of cryotherapy or cryostimulation. The subjects were a homogeneous group with regard to age, sex and the level of daily physical activity.

Prior to the experiment, each participant was examined by a physician to test for any contraindications against cryostimulation and maximal physical effort. Basic anthropometric data was collected including: body height, body mass and body mass index (BMI). Blood samples were obtained from an antecubital forearm vein using vacutainer

system tubes (Sarstedt, Germany), after overnight fasting, in the morning before the treatment, and after a 10-min rest in a sitting position, in order to determine blood variables, including number of erythrocytes (RBC), hemoglobin concentration (Hb), hematocrit value (Hct), number of leukocytes (WBC) and thrombocytes (PLT). Blood for the haematological analysis was collected again in the morning on an empty stomach, three days after the series of 15 whole-body cryostimulations.

In the week before cryostimulation, the participants were subjected to tests of maximal physical capacity, establishing their maximal oxygen uptake ($\text{VO}_{2\text{max}}$) using a direct method. In order to estimate $\text{VO}_{2\text{max}}$, a progressive ergocycle test was applied. The exercise was preceded by a 5 min warm-up (25 W). The test was performed on a bicycle ergometer (Monark 839E, Sweden), beginning with a work load of 1 W per kg of fat free mass ($1 \text{ W} \times \text{kg}_{\text{FFM}}^{-1}$) and was increased by $0.5 \text{ W} \times \text{kg}_{\text{FFM}}^{-1}$ every 3 min until volitional exhaustion or to the moment when the participant could not keep the required frequency of revolutions (70 ± 5 rpm). During the exercise, oxygen uptake (VO_2) was measured continuously using an Oxycon gas analyzer (Jaeger, Germany). Heart rate was measured with a Polar sport-tester. Body composition was evaluated with the use of electrical bioimpedance (Bioanalyser 1500 Akern). Three days after the end of cryostimulations the exercise procedures were repeated.

One week after performing the exercise test, the participants were exposed to a fifteen-day-long series of cryostimulation (once a day), at extremely low temperature (-130°C) in a cryogenic chamber. Each cryostimulation session lasted 3 min. The duration of each session and the temperature of the cold chamber were similar to our previous studies [6,7,11,21], and similar to the prevalent practice. Glasses, contact lenses and all jewelry were removed before the participants entered the chamber. Prior to entering the cryogenic chamber, they dried their bodies thoroughly to eliminate the sensation of cold. All the subjects wore shorts and nothing above the waist; they only wore gloves, socks, wooden clogs and head bands to protect the ears. In order to protect the upper airways, their noses and mouths were secured with a surgical mask. Entry to the cryochamber was

preceded by a 30-second adaptation period in the vestibule at the temperature of -60°C . While in the cryochamber, the subjects were advised to slightly move their fingers and legs and to avoid holding their breath.

The cryostimulations took place every day at the same time, between 9 am and 10 am. The participants were non-smokers and were requested not to drink alcohol, coffee and tea or cola drinks 12 h before testing the commenced. They were also advised to maintain the same level of physical activity and their regular diet during the period of tests.

Systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR) were measured on the day of $\text{VO}_{2\text{max}}$ measurement (before the exercise phase), not later than 1 h after light breakfast which was the same for each participant. The measurements of these indicators were performed again on each day of cryostimulation, immediately before and after the exposure, and 10 min after exiting the chamber and resting in a sitting position. The measurements of blood pressure were made in identical time intervals after cryostimulation in case of all participants.

The cuff of the blood pressure monitor was placed around the upper right arm. Blood pressure and heart rate were measured by a clinically validated automatic blood pressure monitor (OMRON).

The mean arterial pressure (MAP) was calculated as:

$$\text{MAP} = \text{DBP} + 1/3 \times \text{pulse pressure} \quad (1)$$

where

$$\text{pulse pressure} = \text{SBP} - \text{DBP}$$

Ethics

Each participant gave a written consent before joining the study, and the Regional Bioethical Committee issued their formal consent, according to the Helsinki Declaration.

Statistics

The obtained data was analyzed statistically. The statistical analysis was performed using the Statistica 6 software. All data is expressed as mean and standard deviation. The normality of distribution of results was estimated using the Shapiro-Wilk tests. In order to determine the significance of

the differences between the series of examinations before and after cryostimulation, one-factor analysis of variance (ANOVA) was applied. When a significant F-value was found, Tukey's post-hoc tests were used to determine the source of significance, which was set at $p \leq 0.05$.

RESULTS

Characteristics of the examined men are presented in Table 1. In case of all participants, the values of all hematological indices were within the clinical and laboratory reference values, both before and after cryostimulation treatment (Table 2). The obtained results of maximal oxygen uptake during a cycloergometer exercise ($VO_{2max} = 55.6 \pm 3.32 \text{ ml} \times \text{kg}^{-1} \times \text{min}^{-1}$), suggest that the participants were highly trained. It is connected with high level of daily physical activity (1.5 h aerobic physical effort, 3 times a week). The determined oxygen uptake per kg of the body weight shows the capacity of the provision and consumption of oxygen by their body tissues. Fifteen sessions of 3-minute-long exposures to cryogenic temperature did not change the level of aerobic capacity (maximal oxygen uptake after treatment $53.3 \pm 4.12 \text{ ml} \times \text{kg}^{-1} \times \text{min}^{-1}$). Similarly, the duration of the exercise did not change significantly ($19.3 \pm 2.21 \text{ min}$ and $18.4 \pm 1.72 \text{ min}$ before and after the treatment), although in both cases we observed a slight decrease. The hematological values show that as a result of repeated daily cryostimulations, the number of

Table 1. Anthropometrical and physiological characteristics as well as hematological indices of the examined men (the values are mean \pm SD, minimum and maximum) (N=25)

Parameter	Mean \pm SD	Min	Max
Age (years)	21.30 \pm 0.94	20.0	24.0
Body height (cm)	179.50 \pm 5.12	170.0	186.0
Body mass (kg)	74.65 \pm 6.98	62.0	89.0
BMI ($\text{kg} \times \text{m}^{-2}$)	23.19 \pm 1.93	19.7	26.1
VO_{2max} ($\text{ml} \times \text{kg}^{-1} \times \text{min}^{-1}$)	55.60 \pm 3.32	51.2	58.4
HR_{max} (bpm)	194.00 \pm 6.50	189.0	201.0

BMI — body mass index; VO_{2max} — maximal oxygen uptake; HR_{max} — maximal heart rate.

Table 2. The results obtained by the participants during the progressive physical effort test and hematological values before and after cryostimulation treatment

Parameter	Time of measurements	Mean \pm SD
VO_{2max} ($\text{ml} \times \text{kg}^{-1} \times \text{min}^{-1}$)	before	55.60 \pm 3.32
	after	53.30 \pm 4.12
Duration of exercise (min)	before	19.30 \pm 2.21
	after	18.40 \pm 1.72
WBC ($10^3/\mu\text{l}$)	before	5.28 \pm 1.75
	after	6.28 \pm 1.75*
RBC ($10^6/\mu\text{l}$)	before	5.24 \pm 0.26
	after	4.90 \pm 0.62*
HGB (g/dl)	before	14.10 \pm 1.50
	after	13.05 \pm 1.08*
HCT (%)	before	43.00 \pm 3.10
	after	42.70 \pm 2.80
PLT ($10^3/\mu\text{l}$)	before	219.30 \pm 36.32
	after	225.90 \pm 54.00

SD — standard deviation.

WBC — white blood cells, RBC — red blood cells, HGB — hemoglobin, HCT — hematocrit, MCV — mean corpuscular volume, MCH — mean corpuscular hemoglobin, MCHC — mean corpuscular hemoglobin concentration, PLT — platelets.

* Significance of differences at $p \leq 0.05$ after cryostimulation vs. before.

white blood cells increased, with a significant decrease in red blood cells and hemoglobin. No change in the number of platelets was observed (Table 2).

All the subjects were healthy and normotensive with normal body mass index $23.19 \pm 1.93 \text{ kg} \times \text{m}^{-2}$ [19]. The mean (mean \pm SD) resting systolic pressure was $127 \pm 12 \text{ mmHg}$, diastolic pressure $78 \pm 10 \text{ mmHg}$, and heart rate $78 \pm 6/\text{min}$.

As a result of whole-body exposure to cryogenic temperatures, we observed changes in the examined physiological cardiovascular indicators on the consecutive days of the research. These results are summarized in Table 3. During the first day of the treatment we observed a marked increase in systolic blood pressure in comparison to the level before cryostimulation from 127 ± 7.5 to $152 \pm 14.2 \text{ mmHg}$ ($p \leq 0.001$), in diastolic pressure from 77 ± 9 to $81 \pm 7 \text{ mmHg}$ ($p \leq 0.05$), which as a consequence led to an increase in the

Table 3. The mean values with standard deviation of systolic blood pressure (SBP), diastolic blood pressure (DBP), heart rate (HR), pulse pressure (PP) and the mean arterial pressure (MPA) before and after cryostimulation and the changes from pre-exposure values (Δ) on the 1st, 5th, 10th and 15th day of the study

Parameter	Time of measurements	Day of cryostimulation			
		1st	5th	10th	15th
SBP (mmHg)	before	127.0±7.5	125.0±11.6	125.0±13.3	126.0±15.5
	after	152.0±14.2***	146.0±18.0***	144.0±14.4**	145.0±11.9**
	10 min after	132.0±12.9	130.0±14.6	138.0±16.3	131.0±12.5
Δ SBP (mmHg)		20.4±13.5	19.0±12.4	18.7±14.3	18.0±6.6
DBP (mmHg)	before	77.0±9.0	78.0±9.0	81.0±13.5	80.0±7.8
	after	81.0±7.0*	80.0±9.5	83.0±10.0	81.0±14.0
	10 min after	76.0±6.7	78.0±7.6	80.0±9.5	84.0±8.4
Δ DBP (mmHg)		6.0±4.3	6.0±4.2	5.0±3.2	5.0±3.5
PP (mmHg)	before	49.0±12.0	47.0±18.0	45.0±18.0	46.0±9.0
	after	67.0±14.0***	73.0±10.0***	74.0±13.0***	72.0±12.0***
	10 min after	50.0±13.0	52.0±18.0	52.0±18.0	47.0±9.0
Δ PP (mmHg)		18.0±2.0	21.0±13.5	23.0±8.6	24.0±8.0
MAP (mmHg)	before	94.0±9.8	99.0±7.1	99.0±13.6	97.0±10.0
	after	103.0±10.4*	104.0±10.1*	106.0±9.7*	105.0±12.0*
	10 min after	97.0±9.2	99.0±7.1	99.0±13.6	105.0±10.0
Δ MAP (mmHg)		9.0±4.3	7.0±4.2	11.0±8.1	9.0±4.2
HR (min ⁻¹)	before	80.0±17.0	82.0±14.0	81.0±9.0	82.0±14.7
	after	70.0±13.0*	77.0±14.8*	70.0±16.1**	77.0±19.4*
	10 min after	81.0±14.0	80.0±11.0	78.0±6.0	79.0±13.0
Δ HR (beat×min ⁻¹)		8.0±4.5	8.0±5.8	7.0±3.4	7.0±5.1

* Significance of differences at $p \leq 0.05$ vs. before.

** Significance of differences at $p \leq 0.01$ vs. before.

*** Significance of differences at $p \leq 0.001$ vs. before.

mean arterial pressure from 94 ± 9.8 to 103 ± 10.4 mmHg ($p \leq 0.05$). At the same time, we observed a compensatory decrease in heart rate from 80 ± 1 to 70 ± 1 bpm, on average by 8 ± 4 bpm ($p \leq 0.01$) immediately after leaving the cryochamber. Therefore, on the first day, the average increase in SBP and DBP was respectively 20 mmHg and 6 mmHg.

The next measurement, made in case of each participant 10 min after cryostimulation, showed a return of HR and BP to the values similar to the initial ones. After the daily application of cryostimulation, on the fifth day of testing, blood pressure and heart rate were re-measured before and after cryotherapy. Just like on the first day, we observed a statistically significant increase in systolic

blood pressure, with values of 125 ± 11 to 146 ± 18 mmHg, and a return to baseline 10 min after the session. This time there was no significant increase in diastolic blood pressure. Heart rate decreased significantly in response to cryogenic temperatures on the discussed fifth day of testing, by 8 ± 5 bpm on average almost identically to the first day of treatment.

During the whole study period, systolic blood pressure invariably increased immediately after cryostimulation and, respectively, heart rate decreased. Similarly to the previous days, the blood pressure returned to the initial value 10 min after the end of cryostimulation. In the following days of stimulation we noticed increases in mean systolic blood pressure which successively reached 19 mmHg

(5th day), 18 mmHg (10th day), and 18 mmHg (15th day). Diastolic blood pressure remained unchanged during the consecutive measurements after cryostimulation. The significant changes ($p \leq 0.001$) in pulse pressure in response to the procedure applied during the experimental period occurred. The mean value of pulse pressure after cryostimulation oscillated from 67 ± 14 on the first day to 72 ± 13 mmHg after the last session, with the mean rising immediately after cryostimulation (ΔPP) from 18 ± 2 to 24 ± 8 mmHg.

DISCUSSION

Understanding the physiological mechanisms occurring during the action of cryogenic temperatures can only be based on research on healthy individuals. The assessment and analysis of the effects can help determine the usefulness of cryotherapy in patients with various diseases and in athletes. Our previous pilot study with 40 healthy men, showed an increase in systolic and diastolic pressure immediately after single whole-body cryostimulation. The next stage of research, presented in this work, was the evaluation of changes in blood pressure and heart rate during the series of 15 daily cryostimulations and observation if these reactions change as a result of adaptation.

It was ascertained that each whole-body cryostimulation resulted in a significant increase in systolic pressure, pulse pressure and mean arterial pressure with a compensatory decrease in respect of heart rate directly after the stimulation, regardless of the day of the series. Interindividual variation range was wide, both in systolic and diastolic blood pressure during the following sessions of cryostimulation. There is some evidence in literature of wider variability in men's hemodynamic response in comparison with women [14]. All the observed changes in the circulatory system subsided after 10 min of resting in a sitting position. The reduction in heart rate by about 10 beats per minute after the exposure to cryogenic temperatures was also observed by other authors [14,17,18,20], although there are also reports of no changes in HR, and even increased HR after a single exposure to cryogenic temperatures and cooling. Koczorowska et al. [20] showed that the reduction

in heart rate occurred only in patients with normal blood pressure, but observed no changes in HR of patients with baseline systolic pressure above 140 mmHg in tests carried out on a group of men and women aged 51 years. Blood pressure seems to react very rapidly to changes in ambient temperature. It is known that whole-body cryostimulation significantly reduces skin temperatures (T_{sk}) [21,22]. The increase of blood pressure and bradycardia in resting subjects obviously is caused reflectorily by the cooling of uncovered body parts, especially facial cooling [23]. A short but strong thermal stimulus — the exposure of the body to temperature of -130°C stimulates the sympathetic nervous system, causes a number of thermoregulatory responses including the metabolic and hormonal responses, in consequence leading to cutaneous veno- and vasoconstriction. The increased tone in the vessels reduces the volume of blood at the periphery and displaces blood into the core producing a rise in the mean arterial pressure, cardiac output and stroke volume with a consequent reduction in cardiac action [14,24,25]. The elevation of peripheral arterial resistance is elicited via β -adrenergic receptor activation through direct action of reduced T_{sk} on blood vessel diameter, and additionally, by an elevated plasma norepinephrine concentration [26]. It is suggested that the β -adrenergic receptors in the heart, blood vessels, adipocytes and muscles participate in mediating the effect of cold on cardiovascular and thermoregulatory responses [27]. Increased activity of the parasympathetic tone due to cold stimulation of the face during WBC could also participate in lowering heart rate [28]. It seems that the reduction of HR could be explained by combined activation of both the components of the autonomic nervous system and via baroreflex [14].

Elevated blood pressure could be a risk factor not only for people with cardiac diseases, but also for healthy individuals who are regularly exposed to cold, due to the greater load on the heart [14,29]. It seems, however, that the observed change in blood pressure and the short duration of these changes are safe for healthy people. It is widely accepted that the constantly increased blood pressure is much more harmful than short-term increases that are also observed during intense physical exercise [14,30,31].

In practice, whole-body cryostimulation is not allowed for patients with cardiac disease and unstable blood pressure. In Finland, cryostimulation is recommended only for people with blood pressure below 160/100 mmHg [14] and in this study we assumed 150/90 mmHg to be the upper limit of blood pressure.

It is postulated that a consequence of the increase in central blood volume is baroreceptor stimulation which in turn can lead to inhibition of vasopressin secretion, inhibition of diuresis intensity, and hypovolemia, as a result of increased urine production (reduced renal water reabsorption) and the relocation of fluid to the interstitium [25].

Changes in Δ SBP, Δ DBP, Δ MAP and Δ PP were not different on the 1st, 5th, 10th and 15th day of the experiment. It can therefore be argued that in this study no adaptation changes occurred in response to the repeated stress associated with cold. A similar lack of adaptation changes in blood pressure was observed by Westerlund [14] in men and women aged 40 ± 12 years, undergoing whole-body cryostimulation three times a week for 3 months, however, adaptation changes in blood pressure were found in winter swimmers who had cold baths 5–6 times a week during the winter season [32].

In therapeutic practice, 10 cryostimulations are usually applied, followed by a 6-month break. It is unknown if a longer series would result in adaptation of the body to cold. There are reports showing increased systolic blood pressure in winter compared to summer in people with mild hypertension. The seasonal variation in blood pressure is greater in older than younger subjects [33,34]. Similarly, some authors describe an adverse effect of chronic cold acting as a risk factor for more severe hypertension, cardiovascular diseases, stroke and myocardial infarction [35–37].

There are no reports on the influence of whole-body cryostimulation on the fitness of athletes and therefore it is difficult to draw any definite conclusions on the usefulness of this type of treatment. It is interesting from the practical point of view how to include best cryostimulation in the training cycle, especially as in this study we found no changes on aerobic capacity evaluated with the maximal oxygen uptake (which was only slightly decreasing). Our results confirm the report of Klimek et al. [11] which examined the

influence of 10 cryostimulation sessions on the aerobic and anaerobic capacity in men and women. The simultaneous decreases in the number of red blood cells and hemoglobin level are not sufficient to indicate a positive effect of the treatment on the oxygen distribution among tissues and organs, especially in the light of consistency between our results and those obtained by Klimek et al.

CONCLUSION

In conclusion, it is worth noting, that a short-term but significant and repeated increase during consecutive days of cryostimulation session observed in systolic blood pressure in normotensive men, could indicate that this physiotherapy treatment is not recommended for people with advanced or not pharmacologically controlled hypertension. Additionally, it seems worth finding if the decrease in the hematological indices is transitional or not and if it depends on the number of sessions.

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REFERENCES

1. Castellani J, Brenner IKM, Rhind SG. *Cold exposure: human immune responses and intracellular cytokine expression*. Med Sci Sports Exerc 2002;34(12):2013–20.
2. Swenson C, Sward L, Karlsson J. *Cryotherapy in sports medicine*. Scand J Med Sports 1996;6:193–200.
3. Ganta CK, Helwig BG, Blecha F, Ganta RR, Cober R, Parimi S, et al. *Hypothermia-enhanced splenic cytokine gene expression is independent of the sympathetic nervous system*. Am J Physiol Regul Integr Comp Physiol 2006;291:558–65.
4. Leppäluoto J, Westerlund T, Huttunen P, Oksa J, Smolander J, Dugué B, et al. *Effects of long-term whole-body cold exposures on plasma concentrations of ACTH, beta-endorphin, cortisol, catecholamines and cytokines in healthy females*. Scand J Clin Lab Invest 2008;68(2):145–53.

5. Banfi G, Melegati G, Barassi A, Dogliotti G, d'Eril GM, Dugué B, et al. *Effects of whole-body cryotherapy on serum mediators of inflammation and serum muscle enzymes in athletes.* J Thermal Biol 2009;34:55–9.
6. Lubkowska A, Szygula Z, Klimek AJ, Torii M. *Do sessions of cryostimulation have influence on white blood cell count, level of IL6 and total oxidative and antioxidative status in healthy men?* Eur J Appl Physiol 2010;109(1):67–72.
7. Lubkowska A, Banfi G, Dołęgowska B, d'Eril GM, Łuczak J, Barassi A. *Changes in lipid profile in response to three different protocols of whole-body cryostimulation treatments.* Cryobiology 2010;61:22–6.
8. Blatteis CM. *Physiology and pathophysiology of temperature regulation.* Singapore-New Jersey-London-Hong Kong: World Scientific; 1998.
9. Stanek A, Cieslar G, Rosmus-Kuczia I, Matyszkiewicz B, Romuk E, Skrzep-Poloczek B, et al. *Influence of whole body cryotherapy on blood morphology parameters in patients with ankylosing spondylitis and in healthy volunteers.* Acta Bio-Opt Inform Med 2006;12(3):207–10.
10. Banfi G, Krajewska M, Melegati G, Patacchini M. *Effects of whole-body cryotherapy on haematological values in athletes.* Br J Sports Med 2008;42:558–9.
11. Klimek AT, Lubkowska A, Szygula Z, Chudecka M, Frączek B. *The influence of the ten sessions of the whole body cryostimulation on aerobic and anaerobic capacity.* Int J Occup Med Environ Health 2010;23(2):181–9. DOI 10.2478/v10001-010-0019-2.
12. Zeman V. *Physical activity in cold environment.* Med Sport 2005;9:225–34.
13. Zagrobelny Z. *Local and whole-body cryotherapy.* Wrocław: Urban & Partner; 2003 [in Polish].
14. Westerlund T, Smolander J, Uusitalo-Koskinen A, Mikkelsen M. *The blood pressure responses to an acute and long-term whole-body cryotherapy (–110°C) in men and women.* J Thermal Biol 2004;29:285–90.
15. Fricke R. *Ganzkörperkältetherapie in einer Kältekammer mit Temperaturen –110°C.* Z Phys Med Baln Med Klim 1989;18:1–10.
16. Taghawinejad M, Birwe G, Fricke R, Hartman R. *Ganzkörperkältetherapie Beeinflussung von Kreislauf — un Stoffwechselfparametern.* Z Phys Med Baln Med Klim 1989;18:23–30.
17. Komulainen S, Oja T, Rintamaki H, Virokannas H, Keinänen – Kiukaanniemi S. *Blood pressure and thermal responses to whole body cold exposure in mildly hypertensive subjects.* J Thermal Biol 2004;29:851–6.
18. Zalewski P, Tafil-Klawe M, Klawe J, Buszko K, Lewandowski A, Panowicz I. *Influence of the whole-body cryotherapy on the hemodynamic parameters in healthy subjects.* Acta Bio-Opt Inform Med 2009;3:209–14.
19. Guidelines Committee. *2003 European Society of Hypertension — European Society of Cardiology guidelines for the management of arterial hypertension.* Eur J Hypert 2003;2:1011–53.
20. Koczorowska M, Skorupska S, Mamcarz A. *The influence of whole-body cryotherapy on chosen hemodynamic parameters.* Folia Cardiologica Excerpta 2007;2(Suppl A):26.
21. Chudecka M, Lubkowska A, Klimek A, Szygula Z. *The impact of systemic cryotherapy on distribution and dynamics of temperature changes within selected parts of the body.* Acta Bio-Opt Inform Med 2008;1(14):103–6.
22. Westerlund T, Oksa J, Smolander M, Mikkelsen M. *Thermal responses during and after whole-body cryotherapy (–110°C).* J Thermal Biol 2003;28:601–8.
23. LeBlanc J, Dulac S, Cote J. *Autonomic nervous system and adaptation to cold in men.* J Appl Physiol 1975;39:181–6.
24. Mouro L, Cluzeau C, Regnard J. *Physiological assessment of gaseous cryotherapy device: thermal effect and changes in cardiovascular autonomic control.* Ann Réadapt Med Phys 2007;50:209–17.
25. Stock JM, Taylor NA, Tipton MJ, Greenleaf JE. *Human physiological responses to cold exposure.* Aviat Space Environ Med 2004;75(5):444–57.
26. Granberg PO, Lennquist S, Low H, Werner S. *Hormonal changes during cold diuresis.* Swed J Defense Med 1971;7:191–202.
27. Simeckova M, Jansky L, Lesna I, Vubiral S, Sramek P. *Role of beta adrenoceptors in metabolic and cardiovascular responses of cold exposed humans.* J Thermal Biol 2000;25:437–42.
28. LeBlanc J, Blais B, Barabe B, Cote J. *Effects of temperature and wind on facial temperature, heart rate and sensation.* J Appl Physiol 1976;40:127–31.
29. Lloyd EL. *The role of cold in ischemic heart disease: a review.* Public Health 1991;105:516–22.

30. Powers S, Howley E. *Exercise Physiology: Theory and Application to Fitness and Performance*. 4th ed. New York: McGraw Hill; 2001.
31. Wilmore JH, Costill DL. *Physiology of Sport and Exercise*. 3rd ed. Champaign, IL: Human Kinetics, 2004.
32. Hirvonen J, Lindeman S, Matti J, Huttunen P. *Plasma catecholamines, serotonin and their metabolites and beta-endorphin of winter swimmers during one winter. Possible correlations to psychological traits*. *Int J Circumpolar Health* 2002;61(4):363–72.
33. Brennan PJ, Greenberg G, Miall WE, Thompson SG. *Seasonal variation in arterial blood pressure*. *Br Med J* 1982;285(2):919–23.
34. Nayha S. *Adjustment of blood pressure date by season*. *Scan J Prim Health Care* 1985;3:99–105.
35. Kim JY, Jung KY, Hong YS, Kim JI, Jang TW, Kim JM. *The relationship between cold exposure and hypertension*. *J Occup Health* 2003;45:300–6.
36. Gyllerup S. *Cold climate and coronary mortality in Sweden*. *Int J Cir Health* 2000;59:160–3.
37. Marchant B, Ranjadayalan K, Stevenson R, Wilkinson P, Timmis AD. *Circadian and seasonal factors in the pathogenesis of acute myocardial infarction: the influence of the environmental temperature*. *Br Heart J* 1993;69:385–7.