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Could human cold adaptation decrease the risk of cardiovascular disease?

I. Kralova Lesna [a,*], J. Rychlikova [b], L. Vavrova [b], S. Vybiral [c]

[a] Laboratory for Atherosclerosis Research, Institute for Clinical and Experimental Medicine, Prague, Czech Republic
[b] Fourth Department of Internal Medicine, First Faculty of Medicine, Charles University and General University Hospital in Prague, Czech Republic
[c] Faculty of Science, Charles University, Czech Republic

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ABSTRACT

The impact of repeated exposure to cold and cold adaptation on human cardiovascular health is not fully understood. The aim of the study was to determine the effect of cold adaptation on cardiovascular risk factors, thyroid hormones and the capacity of humans to reset the damaging effect of oxidative stress. Ten well cold-adapted winter swimmers (CA) and 16 non-adapted controls (CON) were enroled in this experiment to test whether cold adaptation could influence the parameters of lipoprotein metabolism, cholesterol efflux capacity (CEC), homocysteine, thyroid hormones, antioxidant defence markers (reduced glutathione (GSH), glutathione peroxidase 1 (GPX1), glutathione reductase (GR), catalase (CAT) and paraoxonase 1 (PON1)) and oxidative stress markers (concentration of conjugated dienes (CD)). A decreased apolipoprotein B/apolipoprotein A1 (ApoB/ApoA1) ratio was found in the CA group ($p < 0.05$), but other lipoprotein parameters, including CEC, did not differ significantly. Plasma homocysteine was lower in CA subjects in comparison with controls ($p < 0.05$). Higher triiodothyronine (T3) values were observed in the CA compared to the CON ($p < 0.05$) group, but TSH and other thyroid hormones did not differ between both groups. CA subjects had lower activity of GPX1 ($p < 0.05$), lower concentrations of CD ($p < 0.05$) and increased activities of PON1 ($p < 0.001$) compared to CON subjects. A trend for decreased activity of CAT ($p = 0.06$) in CA compared to CON groups was also observed, but GSH levels did not differ significantly. Zn concentration was higher in the CA group than in the CON group ($p < 0.001$). Human cold adaptation can influence oxidative stress markers. Trends towards the improvement of cardiovascular risk factors in cold-adapted subjects also indicate the positive effect of cold adaptation on cardioprotective mechanisms.

1. Introduction

Hospital admissions and deaths due to cardiovascular disease (CVD) complications and stroke reach a marked peak during winter months and drop in summer months, even in moderate climates (Davidkovova et al., 2013; Spencer et al., 1998). Several contributing factors can form a backdrop to these results, i.e. concomitant respiratory diseases, air pollution, reduced immune resistance, flu, etc. However, the biggest single cause of excess mortality in winter is ischaemic heart disease, accounting for almost half of all excess deaths (Keatinge and Donaldson, 1995).

This effect seems to be due to a combination of risk factors of CVD, since cold increases blood pressure and heart rate and causes further changes in blood composition (increased red cell and platelet counts, increased plasma fibrinogen, changes in concentrations of glucose and lipid parameters). Some of these cold-induced changes (Mavri et al., 2001; Neild et al., 1994; Woodhouse et al., 1994) may partially explain increased cardiovascular mortality in winter. The effect of seasonal variation on rheological and haemostatic parameters has been proved even in healthy young subjects (Frohlich et al., 1997).

However, the mechanisms of the effect of cold on cardiac parameters and atherosclerotic plaque disruption are supposed to

Abbreviations: CVD, cardiovascular disease; CA, cold-adapted; CON, control group; CAT, catalase; GPX1, glutathione peroxidase 1; GR, glutathione reductase; GSH, reduced glutathione; CD, conjugated dienes; PON1, paraoxonase 1; CEC, cholesterol efflux; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; UCP, uncoupling proteins; ROS, reactive oxygen species; hsCRP, high sensitivity C-reactive protein; RCT, reverse cholesterol transport; ApoA1, ApoB, apolipoprotein A1, B; NEFA, non-esterified fatty acids; TC, total cholesterol; TG, triglycerides; FFA, fatty acid-free albumin; TSH, thyroid-stimulating hormone; T3, triiodothyronine; T4, thyroxine; fT3, free triiodothyronine; WBC, whole-body cryotherapy

* Correspondence to: Laboratory for Atherosclerosis Research, Institute for Clinical and Experimental Medicine, Videnska 1958/9, Prague 4, 141 28, Czech Republic.
E-mail addresses: ivka@ikem.cz (I. Kralova Lesna), Jana.Rychlikova@vfn.cz (J. Rychlikova), Lucie.Vavrova@vfn.cz (L. Vavrova), stvyb@natur.cuni.cz (S. Vybiral).

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be mainly connected with increased cold stress-related sympathetic activity following the stimulation of cutaneous thermo- and nocireceptors. Sympathetic activity consequently increases heart rate and blood pressure, i.e. increases cardiac output (Lesná et al., 1999) and further affects metabolic parameters by increasing lipolysis and carbohydrate turnover (Vallerand et al., 1999). Although vasoconstriction due to higher sympathetic activity decreases heat loss, additional thermogenesis is required to maintain body temperature. Shivering as well as non-shivering thermogenesis form part of this response, with the former considered the main form of thermogenesis in humans.

During repeated exposure to cold stress, mechanisms of resistance are replaced by mechanisms of adaptation. Repeated exposure to cold induces physiological changes that have been documented in studies in humans, both in the natural environment (Vybiral et al., 2000) and in laboratory conditions (Sramek et al., 2000). For a long time, there were even doubts about whether metabolic adaptation to cold existed in humans. We (Vybiral et al., 2000) have shown that cold adaptation in winter swimmers consists of metabolic, hypothermic and insulative types of cold adaptation, as has also been previously described by Bittel (Bittel, 1992).

The presence of active brown adipose tissue in human adults has been well documented (Virtanen et al., 2009) and even the involvement of brown adipose tissue in processes of human cold adaptation has been described recently (Blondin et al., 2014; van der Lans et al., 2013). Due to the high presence of uncoupling proteins (mainly UCP-1), this organ fulfils an important thermoregulatory function, namely non-shivering thermogenesis. UCP are closely connected to many physiological processes, including their involvement in an antioxidative capacity (Blanc et al., 2003; Nubel and Ricquier, 2006). The supposition that prolonged or repeated cold stress could induce oxidative stress was first verified in animal models (Buzadzic et al., 1992; Spasic et al., 1993); a finding even confirmed in humans (Siems and Brenke, 1992). Oxidative stress is defined as an imbalance between the production of reactive oxygen species (ROS) and their insufficient decomposition by the antioxidant system, resulting in macromolecular damage and disruption of redox signalling and control. Free radicals may induce DNA mutations, structural disorders in proteins and oxidative damage of cell-membrane and plasma lipids (Valko et al., 2007). The antioxidant defence system actively responds to oxidative stress during acute cold-stress conditions, but the effect of repeated cold exposure on the capacity of humans to prevent oxidative stress and damage has not yet been fully established. On the other hand, mild oxidative stress may be protective by its activation of several redox-sensitive transcriptional factors (Surh et al., 2005).

Therefore, we decided to examine levels of lipid parameters as well as different antioxidants and concentrations of lipid peroxidation markers in order to determine whether repeated mild oxidative stress during long-term cold exposure would lead to changes of cardiovascular risk factors and to adaptive changes in the antioxidant system.

The atheroprotective effect of HDL cholesterol and ApoA1 is mainly due to their function in reverse cholesterol transport (RCT), in which process cholesterol efflux plays a critical role. Nevertheless, we (Kralova Lesna et al., 2008) and others (Fadini et al., 2014; Low et al., 2012) have clearly shown that HDL cholesterol and ApoA1 concentrations do not necessarily correspond to the real capacity of RCT. Therefore, we included the direct determination of plasma-induced cholesterol efflux from human macrophages as an RCT measure.

2. Methods

2.1. Subjects

Twenty-six Caucasian male volunteers were included in the study. Ten cold-adapted men, all of whom practised long-term exposure swimming in cold water in natural conditions at least twice a week during the winter, were enroled. The average duration of practise was 5 years (minimum 3 years). The control group consisted of 16 healthy controls (CON) who had never participated in cold adaptation. Inclusion criteria included willingness to participate in the study and moderate physical activity to ensure that the practise of winter swimming was the only difference in lifestyle between the studied groups. The amount of physical activity was determined by a questionnaire. Exclusion criteria for both groups were the following: concurrent antioxidant therapy, supplementation with vitamins and polyunsaturated fatty acids, treatment with hypolipidaemic medications, excessive alcohol consumption (>30 g/day), manifestation of cardiovascular and/or cerebrovascular diseases, type 1 diabetes mellitus, hepatic and/or renal diseases, hypothyroidism as well as recent infections and malignancies. The subjects completed a dietary questionnaire. Food intake was processed using NutriMaster software and the total intake of energy, protein, fat, carbohydrates, vitamins and minerals was evaluated.

The study was carried out in March when adaptation was expected to reach optimal conditions. Informed consent was obtained from all participants. The study protocol, which complied with the Declaration of Helsinki for research on human subjects was approved by the Ethical Committee of the Faculty of Medicine, Charles University, Pilsen.

2.2. Blood samples

Blood samples were collected by venous puncture after a 12-h overnight fast into Vacutainer® tubes. Activities of antioxidant enzymes (with the exception of paraoxonase 1, PON1) and concentrations of reduced glutathione (GSH) were measured in haemolysed erythrocytes, which had been separated from the EDTA plasma and washed three times with saline. Plasma samples for cholesterol efflux measurements were placed immediately in iced water and centrifuged at 3700 rpm at 4 °C for 10 min. All other analyses (including PON1) were performed from serum; samples were stored at −80 °C until the assay. All samples were marked with unique anonymised identification numbers, merging data only after assays had been completed.

2.3. Biochemical and hormonal analysis

Plasma concentrations of total cholesterol (TC) and triglycerides (TG) were measured using enzymatic-colorimetric methods (Boehringer Mannheim, Germany) on a Hitachi Modular analyser (Japan). High-density lipoprotein cholesterol (HDL-C) was determined in the supernatant after precipitation of lipoprotein B with phosphotungstic acid/Mg^{2+} using a kit from the same manufacturer; low-density lipoprotein cholesterol (LDL-C) was calculated according to Friedewald's formula. Concentrations of apolipoproteins (ApoB and ApoA1) were measured with a Laurell rocket electroimmunoassay using standards and specific antibodies (Behringwerke Marburg, Germany) on an Immage analyser (Beckman Coulter, USA). hsCRP was determined by an immunoturbidimetric method using a K-ASSAY CRP kit (Kamiya Biomedical Company, USA; cv=max 7.6%) on a Hitachi Modular analyser (Hitachi Ltd., Japan). Concentrations of TSH and thyroid hormones were determined using an electrogenerated chemiluminescence immunoassay on an Advia Centaur analyser from