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Preliminary study on the effect of sex on skin cooling response during whole body cryostimulation (−110 °C): Modeling and prediction of exposure durations

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ABSTRACT

In order to determine the required duration of whole-body exposure to extreme cold (−110 °C) in males and females for achieving the same cold-induced response, a mathematical model of skin cooling kinetics was developed. This modeling is derived from the implementation of a new experimental cryotherapy protocol to obtain continuous skin temperature maps over time. Each 3-min whole-body cryostimulation session was divided into six incremental sessions of 30 s carried out over six consecutive days. Seventeen young, healthy subjects (8 males aged 22.6 ±3.0 years and 9 females aged 23.7 ±4.7 years) agreed to participate in this study. The smallest sex-related difference in temperature was found in the trunk area (2.93 °C after 3 min) while the greatest temperature drop was found in the lower limbs (5.92 °C after 3 min). The largest temperature variation was observed between the trunk and the lower limbs, and peaked at 2.67 °C in males and 6.99 °C in females. For both sexes, skin cooling kinetics showed a strong transient exponential type decrease followed by linear regression behavior. It appeared that for achieving the same cold-induced response, the required duration of cryostimulation is longer for males. For example, a trunk skin cooling of −12 °C could be achieved in 125s for females vs 170s for males (+36% longer); for the lower limbs, the same skin cooling magnitude could be reached after 87s for females vs 140s for males (+62% longer).

1. Introduction

Whole-body cryostimulation (WBC) is a physiotherapy method where patients are exposed to extreme cold air (generally −110 °C to −140 °C) in minimal clothing, for a relatively short duration (2–4 min) [4,17,27]. The basic medical efficiency of WBC is related to a thermal stress that generates vasoconstriction [9,31] and stimulation of the thermal skin receptors by lowering skin temperature, leading to an analgesic effect by slowing down nerve conduction [1]. WBC is commonly used as a method to relieve pain and inflammation symptoms caused by various diseases, particularly those associated with rheumatic conditions. It is recommended for the treatment of arthritis [19,23] fibromyalgia [3,41] and ankylosing spondylitis [38]. WBC is also used by athletes to relieve muscle soreness after exercise [2,18,37]. Whole-body cryostimulation has also been found to improve sleep quality and insomnia [16,25] as well as depression and anxiety disorders [35].

The basic premise of cryotherapy is that cold exposure activates thermosensitive skin receptors, which would be the main cause for cryostimulation efficiency because it triggers regulatory mechanisms aiming at maintaining a constant central temperature. Resulting changes in skin temperature then become the key parameter on which the cryostimulation process is based. Past studies dedicated to cryostimulation have used data collected post-WBC during the body warming phase (e.g., biological markers, self-reported perceptions) [28]. Studies evaluating the temporal dynamics of skin temperature have been done following a similar procedure [5,8,10,12,33,36]. This was mainly due to the impossibility of documenting skin thermal variations occurring while receiving cryostimulation, inside the chamber. However, the thermal and metabolic response as it appears following cryostimulation is at least partly related to what happened in the course of the cryostimulation session; in particular to the rate of skin cooling. The magnitude of skin tissue cooling is known to determine WBC ability to trigger a meaningful analgesic effect and avoid adverse effects [10]. The

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Abbreviations

ANOVA	Analysis of variance
C_0	Ratio of cooling
RH	Relative humidity
ROI	Region of interest
SFCCE	French Society of Whole-Body Cryotherapy
SD	Standard Deviation
Tr	rectal temperature
Tsk	Skin temperature
WBC	Whole-body cryostimulation

Table 1
Anthropometric data of study participants (Mean ± SD).

	Males (N = 8)		Females (N = 9)		P Value
	Mean	SD	Mean	SD	
HEIGHT **(CM)**	181.5	5.6	167.7	7.2	$^{\beta}$ 0.0005
MASS **(KG)**	74.2	7.5	60.9	4.7	$^{\beta}$ 0.0004
AGE **(YEARS)**	22.6	3.0	23.7	4.7	0.5982
BODY MASS INDEX **(KG/M^2)**	22.6	2.6	21.7	1.4	0.3859
BODY FAT **(%)**	11.2	4.4	24.0	3.7	$^{\beta}$ 0.0001
BODY LEAN MASS **(%)**	83.7	4.2	71.5	3.5	$^{\beta}$ 0.0001
BODY FAT MASS **(KG)**	8.5	4.0	14.6	2,4	$^{\beta}$ 0.0016
LEAN MASS **(KG)**	61.9	5.0	43.5	4.1	$^{\beta}$ 0.0001
BODY SURFACE AREA **(M^2)**	1.9	0.1	1.6	0.1	$^{\beta}$ 0.0004

$^{\beta}$ indicates a significant difference between sexes $P \leq 0.01$.

dynamics of skin temperature during WBC depends on two factors, the thermal gradient between skin and external environment and tissues capacity to attenuate the heat losses from the body's core to the skin [32]. Due to the paucity of data in the literature, it is currently not known whether sex-related differences affect skin thermal cooling during WBC. The reasons for this lack of data are linked to the narrowness of usual cryotherapy chambers (including the larger ones for 4 to 5 people) which does not allow the recommended distances for taking thermal images to be respected [29]. More importantly, the extremely low temperatures are known to damage electronic equipment like thermal cameras. To compensate for the impossibility of obtaining real-time monitoring of skin temperatures, a new experimental protocol had to be developed.

The aim of this paper is to propose a new incremental time-step cooling protocol, making it possible to evaluate the temporal dynamics of skin temperature (via thermal imaging) in males and females while being exposed to extreme cold air. Our methodological innovation consists in collecting temperature data before, during and immediately after the WBC session in order to obtain a more complete picture of skin temperature variations. Experimentally-induced acute cooling responses can be tracked over selected body areas of interest, but also over the whole body surface (excepted head, neck, and areas covered by clothing). On the basis of these experimental data, a mathematical model (statistical regression) can be constructed, that can be used in other studies involving whole-body cold air exposure at −110 °C in order to predict the needed WBC duration to trigger a specific change in skin temperature. Because rectal temperature is considered as the gold standard for core temperature, this measure is also included in our proposed procedure.

2. Materials and methods

2.1. Participants

Seventeen young, healthy subjects (8 males aged 22.6 ±3.0 years and 9 females aged 23.7 ±4.7 years) agreed to participate in this study and constituted the sample selection. Prior to the study and to meet the inclusion criteria, participants were examined by a physician who confirmed that they had no medical contraindications to WBC among those listed by the French Society of Whole-Body Cryotherapy (SFCCE, Paris France). None of the participants had WBC experience prior to the study. A summary of anthropometric characteristics of the participants can be seen in Table 1. Female participants were asked to inform investigators on the stage of their menstrual cycle. Six were in the follicular phase of their menstrual cycle while the other three were in the luteal phase. Given that menstrual cycles are likely to alter core temperature [13], the menstrual cycle changes might not be followed by parallel changes in skin temperature [42].

Participants were also asked to refrain from consuming alcohol, caffeine or food 3 h prior to the cryostimulation session and from vigorous activities during the 6-day whole protocol duration. During cryostimulation sessions, participants were wearing minimal clothing, gloves, socks, shoes, woolen cap to protect ears and surgical mask to avoid any risk of cold burns to the respiratory tract. During cold exposure, they had to walk slowly around the chamber.

2.2. Study design

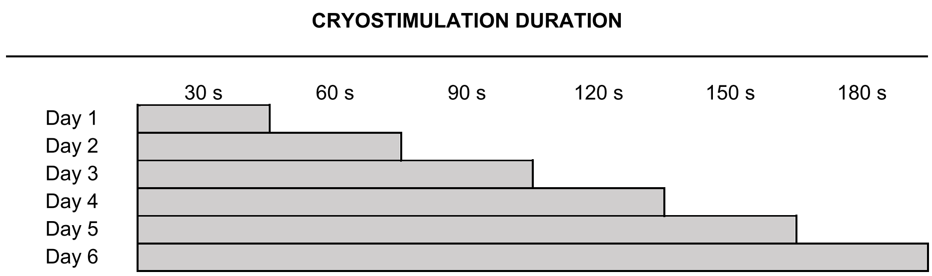
The cooling duration used in this study (−110 °C during 3 min) is typical of those found in practice and reported in the literature [11,12,21,22,37]. In order to obtain continuous information over time on skin cooling kinetics, the innovative protocol developed was based on a decomposition of each 3-min session into six sessions of 30-s, conducted over six consecutive days, according to the timing diagram in Table 2.

To minimize any measurement bias, the sessions were scheduled at the same time each day for each participant. For each session, the skin temperature over the entire front as well as the back of the body was measured, before the session (baseline) and immediately post-WBC. Participants were asked to take their rectal temperature in individual fitting rooms inside the laboratory before the session (baseline) and immediately after the thermal images were obtained.

2.3. Measurements

Body composition of participants was assessed using a dual energy X-ray absorptiometry technology (Horizon DXA system, Hologic, Marborough, USA). For this, all subjects were positioned in the supine position with the arms along the body and then underwent three consecutive full-body scans [20]. Skin temperature measurements took place in a laboratory adjacent to the cryotherapy chamber −110 °C (Mecotec, Germany). Because ambient temperature and humidity may affect the skin temperature, the temperature and relative humidity of the laboratory adjacent to the cryostimulation chamber were initially measured and found to be 21 °C and 35% RH respectively (Sensirion EK-H4, Switzerland). It can be specified that this laboratory temperature of 21 °C is in accordance with the recommendations of Ring and Ammer [34] advocating a controlled environment temperature ranging from 18 °C to 25 °C to avoid any bias in the skin temperature measurement. Rectal temperature was recorded after the participants self-inserted an electronic thermometer (PX-TH519, Tex, Pelimex, Ingwiller, France) in the anal sphincter before and immediately after the cold exposure. Skin temperatures were recorded in orthostatic position using a Variocam HD thermal imaging camera 1024 × 768 (Jenoptik, Germany) in accordance with the standard protocol for infrared imaging in medicine [29]. An emissivity factor of 0.98 for the human skin has been used to get appropriate skin thermographs. The analysis of the thermal images was made possible with the use of post-processing software (IRBIS 3.1 from InfraTec, Germany). Example of skin temperature thermal image is presented in Fig. 1 for a young woman. It is recalled that the thermal analysis concerns both front and back sides of the body even if Fig. 1 presents only front side.

Table 2
Incremental cryostimulation duration per day – Experimental timeline.



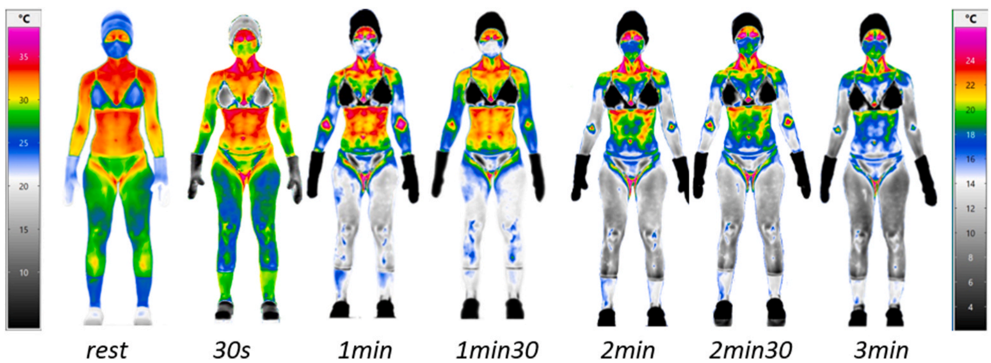


Fig. 1. Evolution in time of skin temperature in the front view of a young female participant – back view thermal images and temperature scales not shown in this figure.

2.4. Data analysis

The choice was made to analyze skin temperatures in three regions of interest (ROI), namely trunk, upper limbs and lower limbs, as shown in Fig. 2. In order not to bias the thermal results, a cutout was made around the underwear, and more specifically around the females's bras, both front and back for evaluating trunk skin temperature.

As this paper focuses on the presentation of a new WBC protocol and inferred modeling, we have deliberately limited the number of regions of interest to three, so as not to significantly increase processing times. The same principle developed in this article can be applied to more localized areas.

The identification of the regions of interest was done by means of a polygonal-type cutout (post-processing software IRBIS 3.1 from Infra-Tec, Germany), the number of segments chosen having to correspond as closely as possible to the exact anatomical shape of the periphery of these identified ROIs. The average skin temperature of each ROI is then a weighted average of the temperatures, quotient for each polygon of the weighted sum of the temperatures by the sum of the pixels. For upper and lower limbs, whatever the sex is, four polygons are necessary (back, front, left, right). For the trunk, two polygons are required for males while for females, between four and eight are necessary, depending on the shape of their bra straps. The average skin temperature of the whole-body surface is then the weighted average of the ROI skin temperatures.

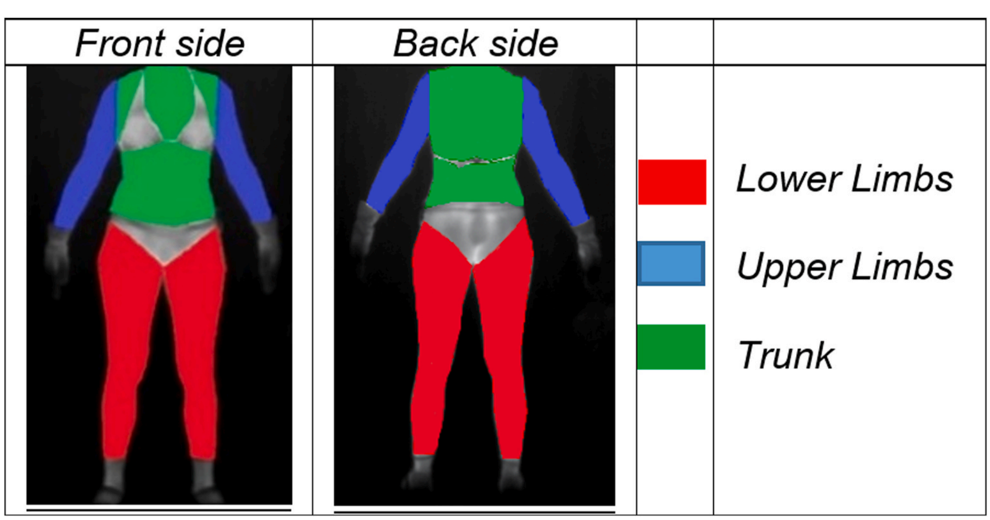


Fig. 2. Choice of the ROIs (underwear excluded).

Obviously, what is called whole body surface is limited to the sum of the ROIs not taking into account the head and neck, and areas covered by clothing.

2.5. Mathematical approach

To predict the duration of whole-body cryostimulation at −110 °C in males and females to achieve the same dose-response effect to cold, a mathematical model of skin cooling kinetics was developed in order to fit experimental data. This modeling can be used to obtain and extrapolate predictive skin temperature at any time by fitting experimental data. Assuming an exponential law during the initial transient [24] from t ∈ [0; 30], followed by a linear one during the second part of the transient from t ∈ [30; β], the time-dependent skin temperature can be expressed as:

$$T_{SK}(t) = \left[Ke^{-(at)^n}\right]_{0\to30} + [At + B]_{30\to\beta} \tag{1}$$

Where K, a, A and B are constants to be analytically determined using both thermal boundary conditions and a C^1- continuous junction between these two functions. The resulting analytical expression of the skin temperature is

$$T_{SK}(t) = \left[T_{sk0}\left(\frac{T_{sk0}}{Tsk_1 + C_0(30-\beta)}\right)^{-\left(\frac{t}{30}\right)^n}\right]_{0\to30} + [C_0 t + (T_{sk1} - C_0\beta)]_{30\to\beta} \tag{2}$$

$$\text{With } \; n = \left(\frac{-30C_0}{[T_{sk1} + C_0(30-\beta)]\,ln\left(\frac{T_{sk0}}{Tsk_1 + C_0(30-\beta)}\right)}\right) \tag{3}$$

Tsk_0, Tsk_1 and C_0 are respectively the baseline skin temperature, the skin temperature at the end of the session and the rate of cooling, and β corresponds to the end of the session.

2.6. Statistical analyses

All data are expressed as means and standard deviations. All statistical analysis were performed using both STATISTICA (data analysis software, version 13.0) and Excel (Microsoft Corporation, Nunes, 2015). Normality of raw data was checked in each group (males and females) using the Shapiro-Wilk and Kolmogorov-Smirnov tests. Five sex-by-time mixed analyses of variance (ANOVA) were performed to investigate changes in time with one between subject's variable (sex: male *vs.* female) and one within-subject variable (moment of measure: 0s, 30s, 60s, 90s, 120s, 150s, 180s) for rectal temperature and skin temperatures (trunk, upper limbs, lower limbs and whole body surface). Statistical significance was set at $p < 0.05$. Associations between body composition variables in Table 1 and changes in skin temperature were analyzed using Pearson's correlation coefficients.

3. Results

3.1. Rectal temperature

For the present study, evolution in time of rectal temperature is shown in Fig. 3 where the dashed line represents the best fit regression linear line. In more detail, rectal temperature was found to vary between 37.04 ± 0.27 °C and 37.19 ± 0.36 °C for males and from 37.39 ± 0.20 °C to 37.47 ± 0.26 °C for females. For this parameter, the ANOVA did not reveal any interaction between sex and time ($F_{(6,\ 90)} = 0.35$, $p = 0.91$). However during the cryostimulation protocol, rectal temperature in females was higher than for males ($F_{(1,\ 15)} = 11.70$, $p = 0.004$). A positive linear correlation between rectal temperature and time was noted, with Pearson's correlation coefficient $r = 0.79$ for males and $r = 0.84$ for

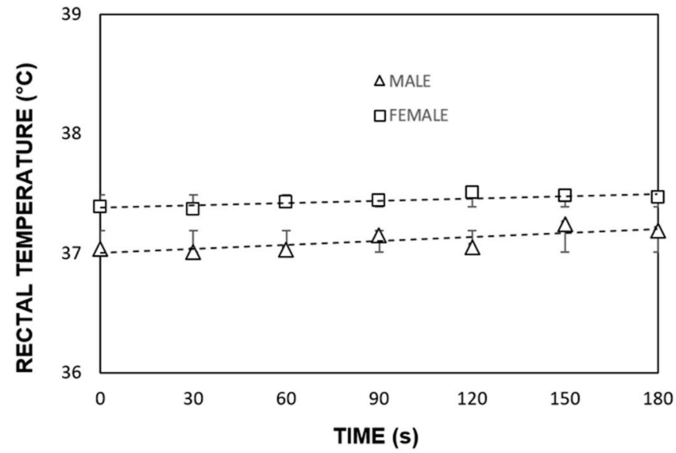


Fig. 3. Time-evolution of the rectal temperature during WBC session at −110 °C.
Error bar indicates SD.

females. Best-fit linear regression lines give a positive thermal gradient of +0.04 °C/min for females and +0.07 °C/min for males during the protocol. For both males and females, bivariate correlation analyses between body fat percentage and rectal temperature at different times of measurement showed a moderate positive relationship ($0.312 < r < 0.669$). Also of interest were a negative relationship of moderate size ($-0.639 < r < -0.442$) between the percentage of lean body mass and rectal temperature at the different moments of measurement. Bivariate correlation analyses between body mass index and rectal temperature at different measurement times showed no relationship.

3.2. Skin temperature of ROI and whole-body surface

Fig. 4 highlights the temporal evolution of skin temperature during the 3 reconstituted minutes that the WBC session lasted at −110 °C, in the same range 10 °C–35 °C. For all ROIs, the dashed curves in Fig. 4 correspond to the best fit mathematical predictive laws that can be deduced (Eq. (2)) where the different parameters in equations (2) and (3) are given in Table 3.

For trunk skin temperature there was a significant interaction effect between sex and time (F(6,90) = 4.66, P < 0.001), a difference between sexes (F(1,15) = 35.74, P < 0.001) and also a difference over time (F(6,90) = 397.65, P < 0.001). For upper limbs skin temperature there was a significant interaction effect between sex and time (F(6,90) = 8.47, P < 0.001), a difference between sexes (F(1,15) = 78.39, P < 0.001) and also a difference over time (F(6,90) = 690.09, P < 0.001). For lower limbs skin temperature there was a significant interaction effect between sex and time (F(6,90) = 10.42, P < 0.001), a difference between sexes (F(1,15) = 168.12, P < 0.001) and also a difference over time (F(6,90) = 565.17, P < 0.001). Females's skin temperature was always lower than that of males throughout the whole cooling in the different ROIs. For both males and females, bivariate correlation analyses between body fat percentage and skin temperature at different measurement times showed a large negative relationship ($-0.799 < r < -0.675$), whereas a large positive relationship ($0.632 < r < 0.820$) was found between the percentage of lean body mass and skin temperature at different times of measurement, as well as an overall large positive relationship ($0.630 < r < 0.821$) between the percentage of body water content and skin temperature. The bivariate correlation analyses between body mass index and skin temperature at the different measurement times did not show any relationship.

Several observations can be drawn from these different temporal patterns. First of all, for participants of both sexes and at whatever body area, the evolution of skin temperatures seems to follow the same

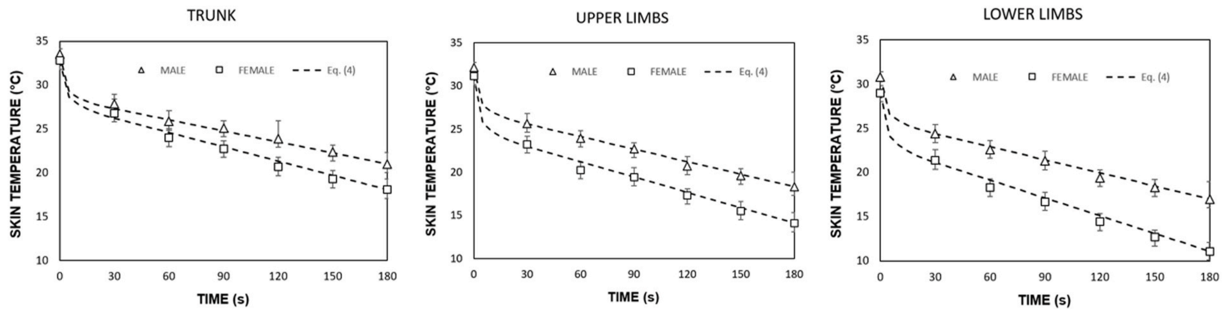


Fig. 4. Time-evolution of the skin temperature for ROI. Error bar indicates SD.

Table 3
Main parameters of the mathematical modeling – ratio of cooling C_0 of the linear phase. Tsk_0, baseline, Tsk_1, skin temperature at the end of the session.

	Trunk	Upper limbs	Lower limbs	Whole body surface
C_0 (°C/s); (SD)				
Females	- 0.0543 (0.0038)	- 0.0592 (0.0025)	- 0.0670 (0.0032)	- 0.0650 (0.0033)
Males	- 0.0419 (0.0026)	- 0.0485 (0.0052)	- 0.0493 (0.0080)	- 0.0467 (0.0053)
Tsk_0 (°C); (SD)				
Females	32.79 (0.53)	31.15 (0.53)	29.00 (0.41)	30.67 (0.44)
Males	33.53 (0.58)	32.11(0.63)	30.75 (0.64)	32.14 (0.63)
Tsk_1 (°C); (SD)				
Females	18.08 (1.26)	14.10 (1.17)	11.09 (1.03)	13.43 (0.75)
Males	21.01 (1.33)	18.34 (1.68)	17.01 (1.93)	18.96 (1.66)

pattern over time: a significant drop in temperature during the first 30 s of the transient, followed by a slower decrease, with a linear trend. This first sudden drop is linked to the phenomenon of massive vasoconstriction coming with the sudden exposure to extreme cold. From 30 s onwards, the skin temperature at all body surface areas are best fit with linear models which attained very high coefficients of determination (i.e., $0.982 < R^2 < 0.994$). The slopes of these lines, denoted C_0, correspond to the ratio of cooling and are given in Table 3. Fig. 5 summarizes the cooling ratio of the second linear phase, for $t > 30s$.

The average whole-body skin temperature is defined as the surface-weighted average of the skin temperatures from the different ROIs. The following relations can be drawn:

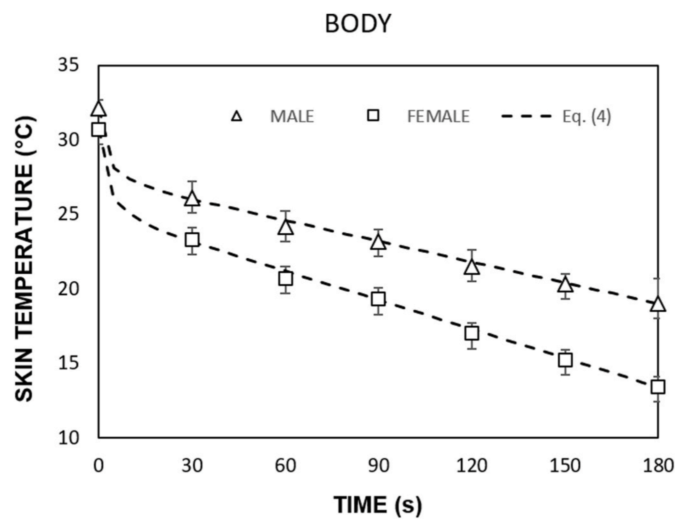


Fig. 6. Temporal pattern of whole-body skin temperature. Error bar indicates SD.

For males, $T_{skin} = 18.7\%\ T_{Upper\ limbs} + 39.0\%\ T_{Lower\ limbs} + 42.3\%\ T_{Trunk}$ (4)

For females, $T_{skin} = 19.0\%\ T_{Upper\ limbs} + 47.9\%\ T_{Lower\ limbs} + 33.1\%\ T_{Trunk}$ (5)

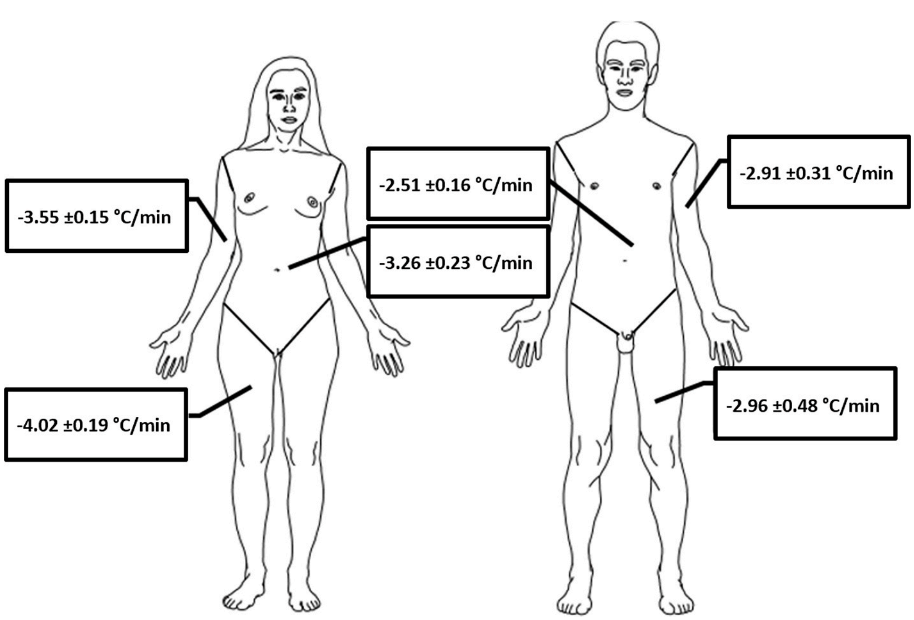


Fig. 5. Thermal gradient C_0 (converted in °C/min) during the linear cooling phase $t > 30s$ – Thermal gradients are built on the average of both front and back views.

Considering the ratio of the surfaces explored, the weight of the skin temperature of the lower limbs (lowest temperatures) will be preponderant in females, whereas it is that of the trunk (highest temperatures) which has priority in males. Fig. 6 highlights the temporal pattern of skin temperature during the 3 min long WBC (−110 °C).

As far as whole-body skin temperature is concerned, there was a significant interaction effect between sex and time ($F_{(6,90)} = 16.68$, $p < 0.001$), a difference between sexes ($F_{(1,15)} = 171.35$, $p < 0.001$) and also a difference over time ($F_{(6,90)} = 870.02$, $p < 0.001$). The best-fitting model of linear nature from 30 s onwards, was also found to have determination coefficients (R^2) close to 1 (0.994 for males and 0.996 for females).

4. Discussion

The purpose of the current study was to investigate the body cooling kinetics of males and females exposed to extreme cold air during a whole-body cryostimulation session. By using mathematical modelling, predictive thermal patterns (for rectal and skin temperatures) could be deducted according to the duration of exposure to cold and the sex of the participants. To the best of our knowledge, this is the first study using such an approach to model the body thermal response to extreme cold exposure.

Some findings in the literature revealed slight discrepancies in the evolution of rectal temperature from pre to post-WBC session. Westerlund et al. [43] indicated that mean rectal temperature did not change during WBC, as confirmed by Zalewski et al. [44] who investigated core temperature (measured from swallowed capsules) and found that this parameter remained unchanged immediately after cryostimulation ($p > 0.05$) compared to baseline. Finally, Cuttel et al. [14] (see Fig. 1 p. 43) and Costello et al. [11] (see Fig. 4 p. 6) even found a slight increased trend for rectal temperature during similar cold air exposure at −110 °C. It should be noted, however, that the latter authors observed a decrease in rectal temperature for immersion in 8 °C cold water. The reason behind these apparently contradicting findings is related to the different modes of heat loss in the latter case (different convective exchange coefficient, absence of radiative transfer in water). At rest, females had slightly higher core temperatures than males, as revealed by Mackowiak et al. [30]. It is known that core temperature usually increases in response to skin cooling [24]. This is in accordance with our observation (see Fig. 3) that regardless of sex, rectal temperature rose slightly during exposure to extreme cold. The distribution of raw data can be satisfactorily fitted with a linear function (coefficient of determination $R^2 = 0.70$ for females and $R^2 = 0.63$ for males) of the type:

$T_R(^\circ C) = 0.07\left(\frac{^\circ C}{min}\right)t(min) + T_{R-Baseline}(^\circ C)$ for males and $T_R(^\circ C) = 0.04\left(\frac{^\circ C}{min}\right)t(min) + T_{R-Baseline}(^\circ C)$ for females.

For both females and males, this finding of a slight increase in rectal temperature is directly related to the initial stimulation of cold receptors in the skin causing constriction of peripheral blood vessels and a redirection of blood flow to the core. Similar trend of a positive rate of rectal temperature $\Delta T_R/\Delta t$ has already been reported in the early adaptation phases of a study involving cold water immersion [39,40]. If exposure to cold was to be prolonged over time, the strong thermal imbalance of the body (heat loss greater than the internal metabolic intake of heat) would lead to body hypothermia with a decrease in core temperature [6].

The skin is the interface between the organism and the external environment which contributes to the dynamic homeostasis of the organism's internal environment, despite changing external conditions. Thermal regulation takes place at the level of the posterior hypothalamus: in the event of exposure to cold, the hypothalamus is stimulated leading to vasoconstriction of the peripheral cutaneous arterioles directing the blood flow towards the deep venous system to form a heat shield protecting the vital organs. This is certainly the reason why the internal temperature rises slightly during this phase of vasoconstriction. Consequently, due to vasoconstriction, the role of thermal insulation played by adipose tissue became preponderant to diminish heat losses. The skin, being more insulated on its body inner side, then becomes more dependent on external conditions [26]. Percentage of BF is known to modulate thermogenic responses to extreme cold [39] and both males and females with lowest BF percentage tended to have higher skin temperature for all measures at all times [7]. In the present study, body fat (BF%) was 24.0 ± 3.7% for females versus 11.2 ± 4.4% for males ($p = 0.0001$). Because fat mass is positively related to tissue insulation [15], and since females have more adipose tissue than males, they have a higher equivalent thermal resistance of 0.037 K/W compared to 0.027 K/W for males in a rest state [32]. The direct consequence is that their skin temperature is much more influenced by environmental conditions, and therefore more abruptly cools down during cold stress compared to males. With regard to body surface areas, it clearly appears from Fig. 4 that for both males and females, the trunk is the body area for which skin temperature fell by the smallest amount. This is probably due to the heat shield effect that it provides by protecting the vital organs, by blood return due to vasoconstriction. It is also in this area of body that the smallest sex-related difference in temperature was found (i.e., 2.93 °C after 3 min of exposure to extreme cold). On the other hand, the lower limbs showed the greatest temperature drop, with a maximum difference between the sexes of 5.92 °C after 3 min. In males, the largest temperature variation was between the trunk and the lower limbs, and peaked at 2.67 °C after 3 min. For females, the largest temperature variation was also between the trunk and the lower limbs, and peaked at 6.99 °C after 3 min. With regard to whole-body skin surface temperature (see Fig. 6), similar findings were reported between males and females, with the largest difference between males and females peaking at 5.53 °C after 3 min.

All these findings suggest that males and females adapt differently to extreme cold air exposure, with females having much more heterogeneous skin temperature variations, depending on body surface areas. This brings into question the current clinical practices in WBC centers which most often use identical cryostimulation protocols for males and females. This issue was previously raised in Cholewka et al. [8]. The amplitude of the cutaneous variation $\Delta T = Tsk - Tsk_0$ can be considered as a key parameter in the evaluation of the effect of sex in cryostimulation. The analytical formulation of the cutaneous thermal behavior (Eq. (2)) makes it possible to easily predict the durations of protocols which make it possible to reach this set point. Table 4 summarizes the different durations numerically calculated in a range of cooling gradient ΔT from −8 °C to −20 °C, for the different body areas considered in our study. It appears that in order to produce the same thermal skin response, protocol durations in males have to be higher than those for females. For instance, to reach a same cooling magnitude of −12 °C in the trunk area, the duration of the protocol must be 36% longer in males compared to females. This difference can reach 54% and even 62% for the upper limbs and lower limbs respectively. It is important to note that these percentages derive from the population of the present study only and must not be applied to any person undergoing a cryostimulation session. Additional studies are necessary to confirm these predictions and propose new guidelines for the duration of cryostimulation sessions. In addition, our analysis revealed that the relative difference (E%) for the duration of protocols between males and females tends to decrease as the desired temperature gradient increases.

The development of a regression model has two major advantages. First, it can allow cryostimulation researchers as well as therapists to easily know the thermal behavior of the skin during cryostimulation sessions at any time. And on the other hand, this analysis can make it possible to predict the duration of protocol, according to the sex and the zones of the body surface considered. To the best of our knowledge, no systematic investigation has yet been performed to identify the intervention dose needed to reach the same skin temperature response in males and females.

Table 4

Analytical evaluation of the protocol durations (s) taking into account sex and body surface areas. Cooling gradients refers to the skin body temperature and E (%) refers to the relative difference for the duration of protocols between males and females. The shaded boxes represent an extrapolation for a cold exposure time of more than 3 min and less than 4 min. The blackened boxes refer to exposure times longer than 4 min, which are mentioned but should not be implemented as they may compromise the health of patients.

Estimation of cryostimulation protocol duration (s)

	Trunk			Upper limbs			Lower limbs		
Cooling gradient	Female	Male	E(%)	Female	Male	E(%)	Female	Male	E(%)
-8°C	55	78	**42**	24	58	**141**	27	59	**120**
-10°C	90	124	**38**	58	99	**72**	57	100	**76**
-12°C	125	170	**36**	91	140	**54**	87	140	**62**
-14°C	160	215	**34**	125	182	**46**	116	181	**55**
-16°C	195	261	-	158	223	**41**	146	222	**51**
-18°C	230	307	-	192	264	-	176	262	-
-20°C	265	352	-	225	305	-	206	303	-

5. Limitation

Some limitations should be taken into consideration when interpreting our findings. First, even though our sample size is roughly similar to what can be found in most published WBC studies it can be said to be fairly "modest" to generalize findings. Consequently, the present study should be considered as a preliminary investigation. Second, directly in relation to the limitation mentioned above, body composition and sex did not appear as explicit input parameters in our regression model. These variables were included in the calculation of C_0 cooling ratios. Further parametric analyses including larger samples should make it possible to include these parameters as explicit input variables in future statistical models. The accuracy of the model is linked, on one hand, to the number of experimental that allowed the construction of the regression laws (a total of 7 measurements points per situation, established every 30 s), and, on the other hand, to the values of the regression coefficients deducted from these curves, which are very close to 1. A final limitation relates to the applicability of our regression model to cold air exposures that would last more than 3 min. In spite of these limitations, we do believe that the initial regression model resulting from our preliminary study can be of great help in defining individualized WBC prescriptions.

6. Conclusion

Due to the extremely low (and potentially damaging to measurements devices) temperatures inside the WBC chambers (−110 °C), a new methodological protocol was developed to investigate the real-time dynamics of skin cooling in healthy male and female participants. The aim of the present study was threefold: (1) providing the scientific and medical communities with pre-, per-, and post-WBC data on skin temperature dynamics at various body areas, (2) defining a regression model that would allow the researchers/practitioners to estimate the average value of skin cooling at the various stages of a typical WBC session (−110 °C), and (3) highlighting sex-based disparities in skin temperature response to cryostimulation.

Major findings can be summarized as follows. Females had slightly higher core temperatures than males during WBC. For both males and females, a positive linear correlation between rectal temperature and time of exposure was found (i.e., $r = 0.79$ for males and $r = 0.84$ for females). Probably due to greater adipose tissue, the magnitude of skin temperature decrease was larger in females, regardless of the body surface area studied (lower limbs, upper limbs, trunk or total surface area). For all sexes and body areas, the dynamics of skin temperature followed two successive transitional regimes: an initial exponential decline period (i.e., steep drop of skin temperature in the first 30 s), followed by a linear-type behavior. Deducted regression laws suggested that to achieve the same dose-response effect, the duration of the WBC protocol must be much longer in males than in females (for example +36% to +62% depending on body area for a same skin cooling magnitude of −12 °C). However these results must be considered as a proof of concept and taken with caution. Anybody using a WBC chamber should not blindly increase the WBC session duration by 36–62% in males compared to females. Protocols should be carried out within exposure times no longer than 4 min at −110 °C not to compromise the health of the patients.

This preliminary study is the first to show that it seems necessary to differentiate in whole body cryostimulation the exposure durations in males and females in order to achieve the same dose-response effect and to propose individualized protocols. Future studies should verify the results of the present study with larger sample sizes and different populations, including for example people with a medical condition.

Ethics statement

No permission from the local ethics committee was obtained, since only biomedical/clinical studies require this approval according to French law. However, the research participants provided written informed consent and the study was conducted in line with the principles of the Helsinki Declaration and its following amendments.

Author contribution statement

GP and BB conceived and designed research. FB and RE conducted experiments. FL contributed statistical tools. GP, FB and BA analyzed data. GP wrote the manuscript. All authors read and approved the manuscript.

Declaration of competing interest

The authors report no conflict of interests regarding the publication of this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.cryobiol.2020.10.014.

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