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Research Paper

Does hypoxia play a role in the development of sarcopenia in humans? Mechanistic insights from the Caudwell Xtreme Everest Expedition

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ABSTRACT

Objectives: Sarcopenia refers to the involuntary loss of skeletal muscle and is a predictor of physical disability/mortality. Its pathogenesis is poorly understood, although roles for altered hypoxic signaling, oxidative stress, adipokines and inflammatory mediators have been suggested. Sarcopenia also occurs upon exposure to the hypoxia of high altitude. Using data from the Caudwell Xtreme Everest expedition we therefore sought to analyze the extent of hypoxia-induced body composition changes and identify putative pathways associated with fat-free mass (FFM) and fat mass (FM) loss.

Methods: After baseline testing in London (75 m), 24 investigators ascended from Kathmandu (1300 m) to Everest base camp (EBC 5300 m) over 13 days. Fourteen investigators climbed above EBC, eight of whom reached the summit (8848 m). Assessments were conducted at baseline, during ascent and after one, six and eight week(s) of arrival at EBC. Changes in body composition (FM, FFM, total body water, intra- and extracellular water) were measured by bioelectrical impedance. Biomarkers of nitric oxide and oxidative stress were measured together with adipokines, inflammatory, metabolic and vascular markers.

Results: Participants lost a substantial, but variable, amount of body weight (7.3 ± 4.9 kg by expedition end; $p < 0.001$). A progressive loss of both FM and FFM was observed, and after eight weeks, the proportion of FFM loss was 48% greater than FM loss ($p < 0.008$). Changes in protein carbonyls ($p < 0.001$) were associated with a decline in FM whereas 4-hydroxynonenal ($p < 0.001$) and IL-6 ($p < 0.001$) correlated with FFM loss. GLP-1 ($r = -0.45$, $p < 0.001$) and nitrite ($r = -0.29$, $p < 0.001$) concentration changes were associated with FFM loss. In a multivariate model, GLP-1, insulin and nitrite were significant predictors of FFM loss while protein carbonyls were predicted FM loss.

Conclusions: The putative role of GLP-1 and nitrite as mediators of the effects of hypoxia on FFM is an intriguing finding. If confirmed, nutritional and pharmacological interventions targeting these pathways may offer new avenues for prevention and treatment of sarcopenia.

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1. Introduction

Sarcopenia is defined as a significant loss of fat free mass (FFM). This can affect physical function and lead to an increased risk of physical disability and mortality [1]. The pathogenesis of sarcopenia is likely to be multifactorial, and skeletal muscle hypoxia has been proposed as a causal factor for muscle wasting and reduced contractility [2–4].

Weight loss has been widely reported in hypoxic chamber experiments and after sojourns at high altitude, with associations between magnitude of loss and both duration of exposure to hypoxia and level of hypoxemia [5–7]. Current evidence seems to point to an alteration of appetite control and consequent reduction of energy intake as a determinant of the weight loss, whereas energy expenditure (resting and dietary induced thermogenesis) seems to be less affected [8]. More than 60% of the weight lost is typically composed of FFM leading to a decline of muscle contractility and physical performance and, consequently, an individual's capacity to cope with extreme environmental conditions [9].

Changes in appetite hormones [(leptin, ghrelin, glucagon like peptide 1 (GLP-1) and cholecystokinin K (CCK))] have been reported after exposure to hypoxia [10–12]. In particular, the role of leptin and ghrelin as causal factors in the reduction of energy intake has attracted controversy after studies reported opposing associations of these appetite hormones with weight loss [13–17]. Recently, a decreased secretion of post-prandial GLP-1 was observed in healthy men exposed to normobaric hypoxia. [18] Conversely, an in vitro study showed enhanced GLP-1 secretion after hypoxia-inducible factor-1α (HIF-1α) knockdown in human adipocytes [19]. To the best of our knowledge, the role of GLP-1 has never been investigated in humans exposed to prolonged hypobaric hypoxia.

A key role in the onset of these adaptive metabolic responses is played by inflammation and oxidative stress, which are enhanced under conditions of low oxygen availability [7,20–22]. The consecutive disruption of intracellular redox balance and inflammation both contribute to skeletal muscle mobilization [23,24]. Hypoxia disrupts the efficiency of the mitochondrial electron transport chain, inducing leakage of electrons to molecular oxygen and thereby generating reactive oxygen species (ROS) [25]. Interleukin 1 (IL-1), interleukin 6 (IL-6) and C-reactive protein (CRP) are often upregulated during expeditions at high altitude [26], and mechanisms linked to skeletal muscle wasting include activation of HIF-1α and NF-κB catabolic pathways and inhibition of the anabolic mammalian target of rapamycin (mTOR) pathway [27–29]. Nitric oxide (NO) metabolism is also influenced by oxygen availability as demonstrated by raised levels of nitrite and nitrate (markers of NO production) and cyclic guanosine monophosphate (cGMP, a biomarker of NO activity) during acclimatization to high altitude [30].

Therefore, physiological data and plasma samples from the 2007 Caudwell Xtreme Everest (CXE) expedition [31] could provide valuable mechanistic insights into the relationship between hypoxia and FFM loss whereby the extended duration at high altitude serves as a pathogenic model of sarcopenia. We first assess body composition changes (FM and FFM) and fluid shifts [total body water (TBW), intracellular (ICW) and extracellular water (ECW)] that occurred during the expedition. We then evaluate the association between changes in FM and FFM with a comprehensive panel of biomarkers of oxidative stress, inflammation, NO bioavailability, appetite control and intermediary metabolism to identify significant predictors of changes in FM and FFM during prolonged exposure to hypoxia. We hypothesized that changes in FFM would be associated with indices of oxidative stress and inflammation in this group of healthy individuals exposed to hypobaric hypoxia during the CXE study.

2. Methods

The study was approved by the University College of London (UCL) Research Ethics Committee, in accordance with the Declaration of Helsinki. Verbal and written informed consent was obtained from all subjects. The study took place between January and June 2007.

2.1. Subjects

Twenty-four healthy participants (18 male; mean age 34.9 yr; range 19–59 yr) who were investigators on the CXE 2007 research expedition to Mount Everest participated in this study [31]. All participants were sea level natives free of cardiovascular or respiratory disease. Criteria for participation have been described in detail elsewhere [31].

2.2. Study protocol

The design and conduct of this study is described elsewhere [31]. Briefly, participants underwent baseline testing in London (altitude 75 m) before travelling by plane to Kathmandu, Nepal (1300 m). From there they flew to Lukla (2800 m) on expedition day 1 and then trekked to Everest Base Camp (EBC; 5300 m), arriving on expedition day 13. The ascent profile, along with blood hemoglobin levels and oxygen saturations, is detailed in Table S1 of the Online Supplementary Material. Testing was conducted during the ascent in field laboratories at Kathmandu (1300 m; day −3 to 0), Namche (3500 m; day 4–6), Pheriche (4250 m; day 9–10), and at EBC (5300 m; day 15–17, EBC week 1). The participants were divided into two subgroups, Group 1 (base-camp team) who subsequently remained at EBC for the duration of the expedition (n = 10) and Group 2 (climbing team; n = 14) who subsequently ascended above 5300 m, to a maximum of 8848 m on Mount Everest; measurements were repeated in both groups at week 6 and week 8. Ambient temperatures were well controlled during testing [31].

2.3. Dietary intake

Three meals were provided per day along with ample provision of calorific snacks (chocolate, nuts, crisps, malt loaf, cheese, cured meat) ad libitum, guaranteeing access to plenty of carbohydrates and protein to cope with the increased energy cost of the walk /climb; fresh fruit and vegetable intake was low at altitudes above 3500 m. An ample supply of meals and snacks were maintained up to camp 2, beyond this hot food consisted of dehydrated meals whilst snacks remained available.

2.4. Peripheral oxygen saturation

SpO_2 was measured by an independent investigator on the morning of the same day that blood was analyzed, after 10 min of rest, using a pulse oximeter (Onyx 9500, Nonin, USA).

2.5. Anthropometry

Body weight (kg) was measured using mechanical Seca 761 scales (Seca, Birmingham, UK), which were hand-calibrated. An independent investigator recorded body weight to the nearest 0.5 kg with the participant wearing base layers only. Height was measured using a portable stadiometer and recorded to the nearest 0.1 cm.

2.6. Body composition

Bioelectrical impedance measurements were taken after a 10 min period of supine rest as the participant lay on a non-conducting air mattress with their arms away from their torso and their legs separated. Measurements were obtained before meals and after voiding. The