



Selected aspects of the habitual diet and levels of intestinal permeability markers of Polish alpinists

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Abstract: The purpose of this study is to investigate selected aspects of habitual diet and levels of intestinal permeability markers in a group of Polish mountaineers. The study included 17 men aged 30.29 ± 5.8 years characterized by long climbing experience (10 ± 5 years) in sport climbing and mountaineering. The study included a quantitative analysis of the alpinists' diet and the determination of zonulin and alpha-1-antitrypsin levels in fecal samples using the ELISA technique. Less than half of the group covered their energy needs in accordance with the recommendations. Most climbers (70.59%) met the carbohydrate requirement, however, the fiber supply was not provided at an adequate level in 23.53% of individuals. All climbers met the requirements for protein and fats. More than one-fifth (23.53%) of them exceeded the normative values for fats, while 5.88% for protein. Mountaineers did not provide adequate amounts of selenium and iodine, as well as vitamins D and K. Fecal zonulin values were elevated in 35.29% of the climbers tested, while fecal alpha-1-antitrypsin values were normal in all subjects. Individuals engaged in climbing and alpine activities are at risk of energy restriction and/or low energy availability. The climbers mostly provided adequate amounts of macro- and micronutrients with their diets. In most of the climbers studied, intestinal permeability was adequate, although elevated zonulin values were observed in some individuals with normal AAT concentrations. These preliminary findings indicate the need for nutritional education and further studies in larger cohorts.

Keywords: intestinal permeability, zonulin, alpha-1-antitrypsin, alpinists, diet, climbing

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INTRODUCTION

Diet is a key component of lifestyle influencing both gut microbiota composition and intestinal permeability. Dietary factors and exposure to psychological stress are among the causes of intestinal disorders [1]. Dietary patterns rich in fiber, polyphenols, and omega-3 fatty acids tend to support barrier integrity by fostering beneficial microbial populations and anti-inflammatory pathways. In contrast, diets high in saturated fats, refined sugars, and food additives have been associated with increased gut permeability, microbial imbalance, and endotoxemia [2]. The habitual diet, therefore, plays a central role in shaping both the microbial environment and the function of the intestinal barrier.

The gut microbiota exerts a critical influence on the preservation of human health through its multifaceted interactions with host physiological processes. This complex microbial ecosystem contributes to essential functions such as nutrient metabolism, modulation of the immune system, and maintenance of intestinal barrier integrity [3]. Alterations in the composition and metabolic activity of the gut microbiota have been associated with changes in nutrient absorption, energy metabolism, inflammatory responses, and recovery - all of which are fundamental to achieving optimal athletic performance and overall well-being [4].

The intestinal epithelium acts as a selective barrier, permitting nutrient absorption while preventing penetration of pathogens and harmful components of the external environment [5]. Disturbances in microbial diversity and function, collectively referred to as dysbiosis, have been implicated in a wide range of health disorders, including gastrointestinal diseases, metabolic syndrome, and systemic inflammation [6]. One of the critical consequences of dysbiosis is an increase in intestinal permeability (IP), increasing the risk of leaky gut syndrome, stimulates the immune system, increasing the risk of developing inflammation and allergic and autoimmune diseases [5]. Intestinal permeability is assessed using zonulin and alpha-1-antitrypsin (AAT), among other tests. Zonulin is a physiological modulator of tight intercellular junctions in the gut, reversibly regulating intestinal barrier permeability [7]. AAT is a protease inhibitor that would be increased during the disruption of the mucosal barrier and increased permeability [8].

In athletes, particularly those exposed to extreme environmental or physiological stress, such as endurance athletes or alpinists, gastrointestinal function is of particular interest. Intense physical activity, heat stress, hypoxia, and energy deficits have all been linked to impaired gut barrier function and elevated intestinal permeability markers [9, 10]. Increased gut permeability in athletes has been associated not only with gastrointestinal symptoms but also with systemic inflammation and impaired recovery, potentially affecting training adaptations and performance outcomes [11]. Despite growing recognition of these associations, data on intestinal permeability markers and their correlation with nutrient intake from the habitual diet among Polish alpinists remain limited.

The purpose of this study is to investigate selected aspects of the habitual diet and the levels of intestinal permeability markers - zonulin and alpha-1-antitrypsin-in a group of Polish mountaineers. The following research hypotheses were put forward: (1) Polish alpinists are at risk of low energy availability, even though their macronutrient intake is within range and (2) intestinal permeability is increased in alpinists. Nutritional knowledge empowers athletes to meet their energy and nutrients requirements [12]. Understanding the interaction between diet and intestinal barrier integrity in this unique population of athletes may offer new insights into optimizing dietary habits and overall health to enhance performance and well-being at high altitudes.

MATERIAL AND METHODS

Study Participants

The study group consisted of 17 male mountaineers from Poland between the ages of 23 and 40 (mean 30.29 ± 5.8 years) with a body weight of 74.96 ± 5.03 kg and a height of 180.47 ± 8.36 cm. A body mass index (BMI) was calculated from the height (cm) and weight (kg) data and was 22.83 ± 2.1 kg/m². The study participants took part in mountain expeditions at least once a year, during which they stayed at an altitude of over 3,000 meters for at least three weeks. Individuals qualified for the study were members of high-mountain clubs affiliated with the Polish Mountaineering Association.

The alpinists participating in the study were characterized by long climbing experience (10 ± 5 years) in sport climbing and mountaineering. The average climbing level of participants, as measured by the IRCRA (The International Rock Climbing Research Association) scale, was 20.69 ± 2.8 , which according to the proposed criteria, places them in the advanced (level 3) category [13]. The climbers spent an average of 8 ± 3 hours per week training.

Before the start of the study, all participants underwent a consultation with a sports medicine physician and were deemed eligible to participate based on the results of the previous standard medical tests, including ECG, and blood and urine analyses. The presence of chronic diseases and age over 45 were disqualifying factors for participation in the project. The Bioethics Committee of the Cracow Regional Medical Chamber approved the research project (67/KBL/OIL/2021), which was conducted in accordance with the Declaration of Helsinki. After learning about the risks and benefits of participating in the study, all climbers gave written informed consent to participate in this study.

Study Design

Nutritional Analysis of Diet

The supply of selected macronutrients was determined by analyzing whole-day rations obtained using the 3-day food diary method. Mountaineers were asked to note all food, meals and supplements consumed and fluids drunk during a standard 3-day period in the 2 weeks prior to the expedition (2 days of sports activity, 1 day of rest). Sports activity days included climbing in a climbing wall, rock or mountain climbing, running and weight training. Climbers were given detailed instructions on how to keep food diaries before leaving for the expedition in order to minimize recording errors. The diaries also included market foods, so the data needed for analysis was read from labels and nutritional tables. For food products such as bars, energy gels and isotonic drinks, among others, climbers were asked to provide the product name, manufacturer and weight of the product.

Data from the diaries were meticulously entered into the Aliant Diet Calculator program (Anmarsoft Marcin Olech, Gdańsk, Poland; version: 85; database: 6.2) to perform a quantitative analysis of the climbers' habitual diet. The program used the "Tables of food composition and nutritional value" [14] as a database, taking into account food recipes and losses due to product processing.

Measurements of energy expenditure were made by the researchers using a monitor recording heart rate Polar M430 (Polar-Electro, Kempele, Finland), which was recorded 24/7 for 3 days (2 days of sports activity, 1 day of rest). Pulsometers consisted of watches (receiver) and straps with transmitters Polar H10 sensor (Polar-Electro, Kempele, Finland), worn on the chest at the level of the gladius process in the anterior midline. The watches were placed on the hand so that they were adjacent to the body just below the wrist bone. Before the measurement began, the required data were entered into

the device for each athlete: gender, age, body weight and height. Before the start of physical activity, the subjects were instructed to put on the transmitter strap, start the physical activity recording option on the device (climbing indoors, outdoors, running or gym), and when the physical activity was over, to turn off the recording by pressing the corresponding button on the watch. All climbers were advised not to remove the devices from their wrists for the duration of the energy expenditure recording being carried out (sleep, daytime activity, physical activity). Energy expenditure was recorded in the Polar Flow app.

Body height of participants was measured using a Seca 217 anthropometer (Seca GmbH & Co., KG, Hamburg, Germany) with an accuracy of 1 mm. The climbers' body weight was determined using InBody 120 body composition analyzer (Inbody Bldg., Seoul, Republic of Korea) in the morning after a standardized meal between 7:45 and 8:30 am.

Intestinal permeability analysis

In order to determine the levels of intestinal permeability markers (alpha-1-antitrypsin and zonulin), a stool sample was collected from the athletes. The athletes were informed about the correct collection of material (faeces) from 3 different locations to standardise the samples. The material was placed in sterile containers and stored in a cool place until delivery to the ALAB laboratory. The correct reference range for the intestinal permeability markers analysed is, for alpha-1-antitrypsin below 27.5 ng/ml and for zonulin below 60 ng/ml. The tests were performed using the ELISA technique and commissioned as an external service by ALAB Laboratories (Warsaw, Poland).

Statistical analysis

The Shapiro-Wilk test was performed indicating non-normality of the distribution. Basic statistical measures in the form median (Me) and quartiles (Q1, Q3) were used to characterize the received results. Correlations between quantitative variables were analyzed using the Spearman-rho correlation coefficient. The significance level was taken as $p \leq 0.05$. The statistical analysis package JASP (University of Amsterdam, Amsterdam, The Netherlands; version: 0.19.3) was used for statistical analyses.

A binary classification approach was used to assess the implementation of nutritional standards. For each nutrient, a value of 1 was assigned if the subject's intake of the nutrient met the recommended standard, and 0 if it did not. Then, based on these data, the percentage of subjects who met the standards for each nutrient was calculated. The analysis was carried out using an Excel spreadsheet (Microsoft Corporation, Washington, DC, USA). The median intake of macronutrients (protein, fat, total carbohydrates), dietary fiber, simple carbohydrates, saturated fatty acids, and micronutrients from 3 days was referred to recommendations for athletes [15–20]. The results were presented in the form of percentages, which made it possible to assess the degree to which the study group implemented the standards.

RESULTS

The median energy demand of the study participants was 2951 (2731; 3318) kcal, and the energy supply was 3031.4 (2470.7; 3702.8) kcal [Table 1]. Dietary supplementation accounted for 1.98% of the total energy supply. More than half of the climbers surveyed (52.94%), maintained a negative energy balance [Figure 1]. The median percentage coverage of energy requirements with diet in the surveyed climbers was 92.64 (85.55; 113.62) %. Most climbers (70.59%) met the carbohydrate requirement; however, the fiber supply was not provided at an adequate level in 23.53% of individuals [Figure 1]. All climbers met the recommended intake levels for protein and fats [Figure 1]. However, 23.53% exceeded the recommended values for fat intake, and 5.88% exceeded those for protein. Less than half of the climbers surveyed met the recommendations for saturated fatty acid intake, and slightly more than a third met the recommendations for dietary

cholesterol intake [Figure 1]. The median energy and nutritional value of the diet obtained from the analysis of the food diaries is shown in Table 1. Compared to standards for athletes [16–20], mountaineers did not provide adequate amounts of selenium and iodine, as well as vitamins D and K. All climbers met the requirements for sodium, phosphorus, vitamins: A, B₁, B₂ and B₁₂ [Figure 2].

Table 1. Energy and nutritional value of the daily food ration of mountain climbers in relation to the recommendations for athletes [16–20]

Nutrients		Diet of alpinists	Nutritional recommendations for athletes
		Me (Q1; Q3)	RDA/AI
Energy and macronutrients, fiber, fatty acids and cholesterol intake	Energy [kcal]	3031.4 (2470.7; 3702.8)	EnExp*
	Protein [%]	15.5 (14.1; 18.4)	15–20 ¹
	Protein [g]	111.8 (104.5; 147.9)	-
	Protein [g/kg bw]	1.53 (1.33; 1.91)	1.2–2.2 ¹
	Plant protein [g]	47.90 (39.5; 56.2)	-
	Animal protein [g]	68.4 (49.8; 81.0)	-
	Carbohydrates [%]	47.6 (43.7; 53.2)	45–65 ²
	Carbohydrates [g]	406.6 (292.6; 446.8)	-
	Carbohydrates [g/kg bw]	5.39 (3.72; 5.67)	6–12 ³
	Simple carbohydrates [g]	98.8 (70.1; 126.2)	-
	Simple carbohydrates [%]	13.34 (10.14; 14.86)	<20 ⁴
	Fiber [g]	36.3 (24.2; 48)	>25 ⁴
	Fats [%]	33.5 (29.6; 36.5)	20–35 ^{2,3,4}
	Fats [g]	105.3 (87.1; 131.4)	-
	Fats [g/kg bw]	1.44 (1.17; 1.52)	0.5–1.5 ¹
	SFA [%]	11.32 (9.7; 12.57)	<10 ³ ; ALAP ⁴
	SFA [g]	36.6 (34.3; 40.4)	ALAP ⁴
	MUFA [g]	32.1 (25.2; 37.9)	-
	PUFA [g]	14.5 (10.4; 19)	ALA 0.5%, LA 4%
	Cholesterol [mg]	403.6 (321.9; 611.6)	<300 ⁵
Micronutrients intake	Sodium [mg] (AI)	2990.3 (2464.3; 3344.4)	1500 – >10,000 ²
	Potassium [mg] (AI)	4488.4 (2882.6; 4780.7)	3500 ⁴ – 4700 ²
	Calcium [mg]	1160.6 (912; 1493.9)	1000 ¹ – 2000 ³
	Phosphorus [mg]	1693 (1600.6; 2023)	700 ¹ – 1500 ²
	Magnesium [mg]	504.7 (432.2; 612.8)	420 ¹
	Iron [mg]	17 (14; 20)	11 ⁴
	Zinc [mg]	13.5 (11.6; 16.1)	11 – 15 ²
	Copper [mg]	2.0 (1.5; 2.5)	0.9 ² – 1.6 ⁴
	Selenium [µg]	13.2 (7.7; 22.7)	50 – 55 ²
	Iodine [µg]	67.2 (45.9; 76.6)	120 – 150 ²
	Vit. A [µg]	1323.2 (1072.7; 2140.5)	900 ¹
	Vit. D [µg]	3.8 (2.7; 7)	15 ^{2,4}
	Vit. E [mg]	16.8 (13.7; 26.5)	15 ^{1,2}
	Vit. C [mg]	247 (179.9; 327.3)	110 ⁴ – 200 ²
	Vit. B ₁ [mg]	1.8 (1.5; 2.1)	1.2 ¹
	Vit. B ₂ [mg]	2.5 (2.3; 2.9)	1.3 ^{1,4}
	Vit. B ₃ [mg]	22.8 (16.6; 29.7)	14–20 ²
	Vit. B ₆ [mg]	3 (1.6; 3.9)	1.5 – 2 ²
	Vit. B ₉ [µg]	494.8 (375.1; 751.4)	400 ^{1,2}
	Vit. B ₁₂ [µg]	5.3 (3.2; 7.5)	2.4 ¹ – 4 ⁴
	Vit. K [µg]	27.8 (11.4; 65.7)	120 ¹ – 700 ²

Abbreviations: *- energy expenditure; 1—[18]; 2—[19]; 3—[17]; 4—[16]; 5—[20]; ME - median; Q1 - first quartile; Q3 - third quartile; RDA - recommended dietary allowance; AI—adequate intake; ALA - α-linolenic acid; ALAP—as low as possible; bw—body weight; LA - linoleic acid; MUFA - monounsaturated fatty acid; PUFA - polyunsaturated fatty acid; SFA - saturated fatty acids

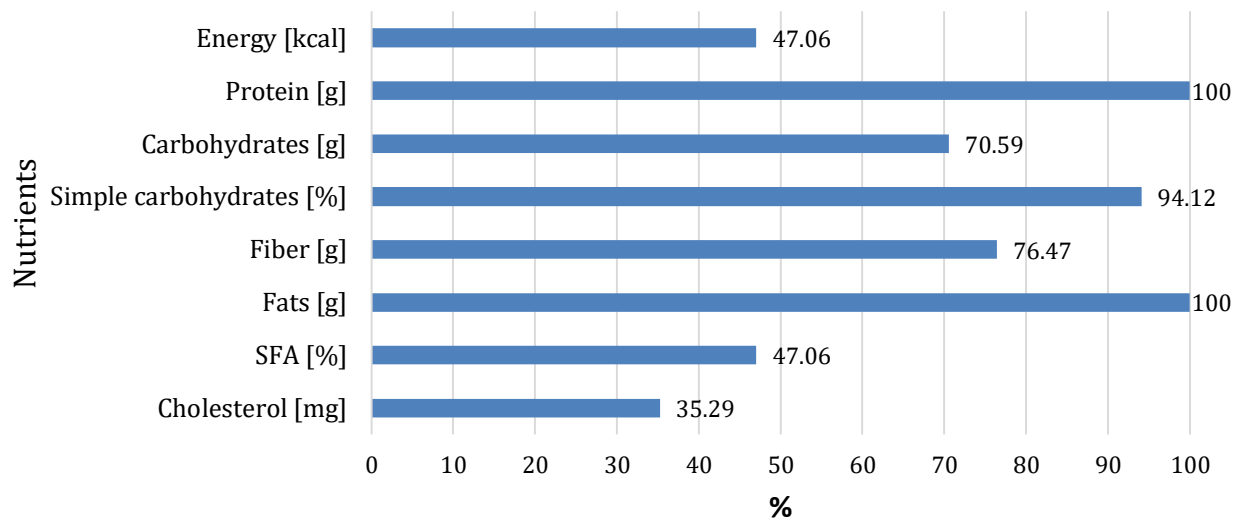


Figure 1. Percentage of alpinists realizing the requirement for energy, macronutrients, fiber, saturated fatty acids (SFA) and cholesterol [%].

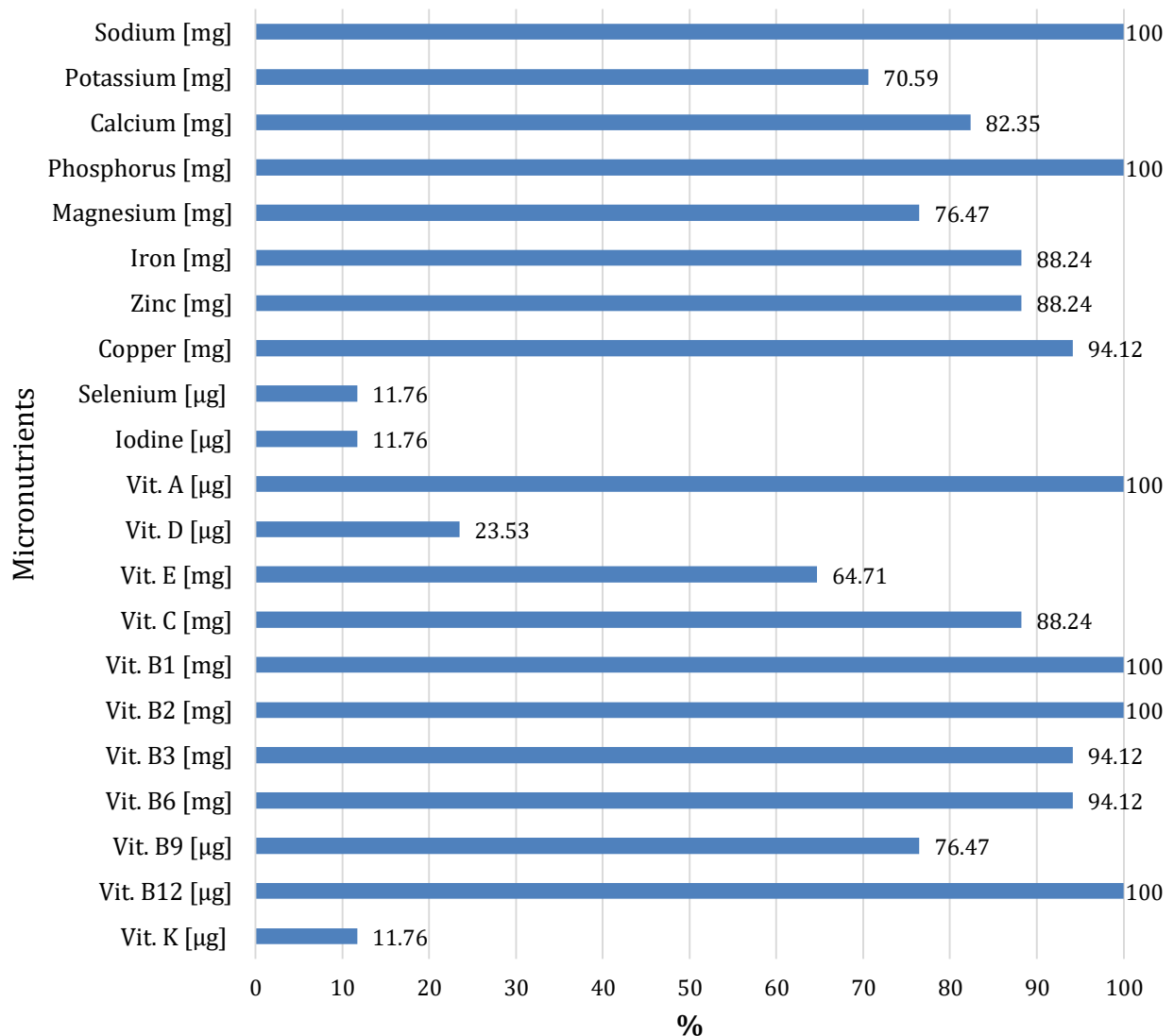


Figure 2. Percentage of alpinists realizing the requirement for specific micronutrients [%].

The median value of fecal zonulin in climbers was 28.40 (14; 193.7) ng/ml, while fecal alpha-1-antitrypsin was 5.95 (4.10; 6.86) ng/ml. Fecal zonulin values were elevated in 35.29% of the climbers tested, while fecal alpha-1-antitrypsin values were normal in all subjects. All correlations between nutrients of the habitual diet of alpinists and levels of intestinal permeability markers among alpinists were not statistically significant [Table 2].

Table 2. Correlations between nutrients of the habitual diet of alpinists and levels of intestinal permeability markers among alpinists

Nutrients	Intestinal permeability markers			
	Zonulin		Alpha-1-antitrypsin	
	S-rho	<i>p</i>	S-rho	<i>p</i>
Energy [kcal]	-0.036	0.920	0.371	0.291
Protein [%]	-0.268	0.454	-0.237	0.510
Protein [g]	-0.600	0.073	0.067	0.865
Plant protein [g]	0.139	0.707	0.079	0.838
Animal protein [g]	-0.152	0.682	-0.127	0.733
Protein [g/kg bw]	-0.539	0.113	0.261	0.470
Carbohydrates [%]	-0.188	0.608	0.212	0.560
Carbohydrates [g]	-0.006	1.000	0.321	0.368
Carbohydrates [g/kg bw]	-0.079	0.838	0.380	0.279
Simple carbohydrates [g]	0.210	0.560	0.140	0.707
Fiber [g]	0.236	0.514	0.224	0.537
Fats [%]	0.333	0.349	-0.236	0.514
Fats [g]	0.152	0.682	-0.055	0.892
Fats [g/kg bw]	0.200	0.584	0.000	1.000
SFA [g]	0.224	0.537	0.079	0.838
MUFA [g]	0.200	0.584	-0.127	0.733
PUFA [g]	0.219	0.544	-0.164	0.657
Cholesterol [mg]	-0.236	0.514	0.030	0.946
Sodium [mg] (AI)	-0.115	0.759	0.067	0.865
Potassium [mg] (AI)	0.042	0.919	0.079	0.838
Calcium [mg]	-0.018	0.973	0.309	0.387
Phosphorus [mg]	-0.042	0.919	-0.176	0.632
Magnesium [mg]	0.055	0.892	-0.127	0.733
Iron [mg]	0.055	0.892	-0.103	0.785
Zinc [mg]	0.127	0.733	-0.248	0.492
Copper [mg]	0.139	0.707	-0.067	0.865
Selenium [µg]	-0.321	0.368	-0.370	0.296
Iodine [µg]	0.176	0.632	-0.576	0.088
Vit. A [µg]	-0.261	0.470	0.176	0.632
Vit. D [µg]	-0.370	0.296	-0.018	0.973
Vit. E [mg]	0.030	0.946	-0.115	0.759
Vit. C [mg]	-0.152	0.682	-0.067	0.865
Vit. B ₁ [mg]	-0.213	0.554	-0.116	0.750
Vit. B ₂ [mg]	-0.486	0.154	-0.012	0.973
Vit. B ₃ [mg]	-0.455	0.191	-0.012	0.973
Vit. B ₆ [mg]	-0.055	0.881	-0.286	0.424
Vit. B ₉ [µg]	-0.079	0.838	-0.115	0.759
Vit. B ₁₂ [µg]	-0.333	0.349	-0.224	0.537
Vit. K [µg]	0.273	0.448	-0.503	0.143

Abbreviations: bw—body weight; MUFA - monounsaturated fatty acid; *p* - statistical significance; PUFA - polyunsaturated fatty acid; S-rho - Spearman's correlation coefficient; SFA - saturated fatty acids

DISCUSSION

Main Findings

The present study evaluated the habitual dietary intake and selected markers of intestinal permeability in a group of Polish alpinists. More than half of the alpinists surveyed maintained a negative energy balance, indicating a notable discrepancy between energy demand and intake, and suggesting a risk of chronic low energy availability (LEA). Low energy intake can result in loss of muscle mass and bone density, as well as an increased risk of fatigue, injury, and illness [21]. The failure to meet energy requirements may exacerbate gut barrier dysfunction [22]. Inadequate energy supply relative to demand can potentially have a negative impact on recovery, immune function and overall performance during training, as well as during planned high-altitude activities [23, 24]. The results of another study involving sport climbers also indicate an inadequate dietary energy intake. Overall, 77.5% of the climbers studied did not meet the predicted energy requirements necessary to maintain a “moderate” level of physical activity [25]. Individuals engaged in climbing and alpine activities are at risk of energy restriction and/or low energy availability, evidenced by suboptimal energy intakes, and a lack of adjustment of energy intake in relation to exercise volume.

Carbohydrate (CHO) intake was adequate in the majority of study participants (70.59%), aligning with the recommendations for athletes engaged in endurance sports [17]. However, dietary fiber intake was insufficient in almost a quarter of respondents. Dietary fiber can significantly influence the composition, diversity, and richness of the microbiome, offering a wide array of substrates for fermentation processes conducted by particular microbial species that possess the requisite enzymatic capabilities to break down these intricate carbohydrates [26]. Low intake of dietary fiber and resistant starch may lead to decreased bowel movements resulting in decreased bowel function, and also decreased the diversity of gut microbiota [27]. Given the role of dietary fiber in supporting gut microbiota diversity and intestinal barrier function, this may have implications for gut health, especially in the context of exposure to hypoxic conditions and physical stress. A proper intestinal barrier is crucial for mountaineers, as hypobaric hypoxia can increase permeability and lead to gastrointestinal distress [28]. Maintaining gut health before expeditions helps prevent symptoms like nausea and diarrhea, ensuring better performance and overall well-being at high altitudes [29].

All mountaineers met the requirements for protein and fats with their diets, with 23.53% of them exceeding the normative values for fats and 5.88% for protein. These findings hold physiological relevance when considering the impact of macronutrient intake on intestinal permeability - a key factor in maintaining gut barrier integrity under environmental and physical stressors. Excessive supply of protein with the diet, though observed in a minority, is less directly implicated in intestinal permeability but may modulate gut microbial activity in ways that affect mucosal health. High protein intake increases the delivery of undigested proteins to the colon, where they undergo microbial fermentation. This process produces potentially harmful metabolites such as ammonia, hydrogen sulfide (H₂S), and phenolic compounds, which have cytotoxic and pro-inflammatory properties. These metabolites can impair epithelial cell respiration, damage DNA, and compromise tight junction integrity, contributing to gut barrier dysfunction [30, 31]. Currently, an increasing body of evidence suggests that the ingestion of excessive dietary lipids can differentially augment intestinal permeability. For instance, dietary lipids influence the expression and localization of tight junctions (TJs), promote a transition towards barrier-compromising hydrophobic bile acids, and additionally provoke oxidative stress and apoptosis in intestinal epithelial cells (IECs) [32]. Another study shows that 6-day ketogenic low-CHO, high-fat dietary intervention combined with a high-fat preexercise meal resulted in greater exercise-associated perturbations to markers of intestinal integrity than a high CHO diet and even low energy availability [33].

The median intake of saturated fatty acids (SFA) with the mountaineers' diet was 11.32%, exceeding the established recommendations. Current guidelines from the European Food Safety Authority advise that SFA intake should be as low as possible [16]. In contrast, the position statement of the Academy of Nutrition and Dietetics, Dietitians of Canada, and the American College of Sports Medicine [17] recommends limiting SFA intake to less than 10% of total energy intake. However, the correlation between SFA intake and fecal zonulin and alpha-1-antitrypsin levels was not statistically significant. The intake of monounsaturated and polyunsaturated fatty acids in the mountaineers' diets slightly exceeded that of saturated fatty acids. Available scientific evidence indicates that a diet with high nutrient density - rich in polyphenols, omega-3 fatty acids, dietary fiber, and probiotics - has a beneficial effect on the composition and function of the intestinal microbiota. In contrast, a diet rich in highly processed foods, saturated fatty acids, and simple sugars is associated with adverse changes in the gut microbiota [34].

Micronutrient analysis revealed dietary inadequacies in selenium, iodine, and vitamins D and K - nutrients essential for immune function, antioxidant defense, and bone health. Emerging evidence suggests that deficiencies in these micronutrients may contribute to increased intestinal permeability, often referred to as "leaky gut", which can lead to systemic inflammation and impaired immune defense - factors particularly detrimental during high-altitude exposure. Vitamin D, beyond its well-established role in calcium homeostasis and immune regulation, has been shown to modulate the expression of tight junction proteins (e.g., occludin and claudins) in intestinal epithelial cells [35]. Its deficiency has been associated with compromised mucosal barrier function and increased translocation of endotoxins into the bloodstream, which may exacerbate systemic inflammation [35]. Given the important role of vitamin D in immune function and its association with an increased risk of anemia when deficient [36], it is essential to monitor serum vitamin D levels before an expedition and adjust supplementation accordingly. Selenium, a crucial cofactor for antioxidant enzymes such as glutathione peroxidase, also plays a protective role in gut health by counteracting oxidative stress-induced epithelial damage [37]. Inadequate selenium intake may impair redox balance and increased epithelial permeability, thereby contributing to greater susceptibility to infections and reduced tolerance to hypoxic stress [38]. Similarly, iodine is a critical micronutrient for thyroid hormone synthesis, which affects metabolic rate and energy balance, but also modulates mucosal immunity and epithelial turnover. Subclinical hypothyroidism, often observed with low iodine intake, can impair gastrointestinal motility and barrier maintenance [39]. Vitamin K has also been implicated in the regulation of inflammation and mucosal immunity. By downregulating expression of cytokines such as TNF- α and IL-6, vitamin K may help mitigate inflammation-induced disruption of tight junction proteins (e.g., occludin, claudin-1, zonula occludens 1), which are critical to the structural integrity of the gut barrier [40]. The observed deficiencies in the habitual diets of the studied alpinists raise concerns about suboptimal gut resilience and systemic immune competence prior to and during expeditions.

Referring to the levels of the analysed intestinal permeability markers, elevated levels of zonulin (28.40 (14; 193.7) ng/ml) were found in the studied group of alpinists, with concentrations exceeding the reference values in approximately 35.29%. The results may indicate a disturbed intestinal microbiota and a potential state of dysbiosis, associated with increased permeability of the intestinal barrier. Similar trends of increased intestinal permeability and elevated levels of relevant markers in athletes have also been found in other studies. Increased intestinal permeability and elevated levels of zonulin have been observed in Polish athletes [41], and increased levels of circulating zonulin (tight junction modulator), occludin (tight junction protein) and LPS translocation have been confirmed in elite Italian soccer players [42]. In a study involving Finnish runners, a significant increase in intestinal permeability (as determined by zonulin and LPS levels) was observed after 90 minutes of intense exercise [43]. Elevated levels of zonulin were also observed in Polish triathletes after completing a race [44]. Changes in

markers of muscle cell damage correlated strongly with changes in zonulin concentration at different stages of the study [44]. The elevated zonulin levels observed in some alpinists may suggest disturbances in the gut microbiota and increased intestinal permeability; however, our analyses did not show significant correlations with nutrient intake. This interpretation should be treated with caution and requires confirmation in studies with larger groups and the use of additional permeability markers.

Another less common marker of intestinal permeability is fecal alpha-1-antitrypsin, the levels of which were normal in the group of alpinists studied. AAT is a protease inhibitor that would be increased during the disruption of the mucosal barrier and increased permeability [8]. It was shown that AAT reduces the production of pro-inflammatory cytokines, inhibits apoptosis and inflammatory reactions [45]. Several studies reported that AAT activity is increased post-exercise [46–48]. Another study reported post-exercise increase of both IL-6 concentration and AAT activity, which may confirm that the release of pro-inflammatory cytokines stimulates the release of acute-phase proteins [49]. There is a lack of studies in athletes in which intestinal permeability was determined using alpha-1-antitrypsin. The anti-inflammatory and immunomodulatory properties of AAT can be used therapeutically to treat pulmonary, infectious diseases and autoimmune diseases [50]. In the studied group, alpha-1-antitrypsin levels remained within the normal range. Possible explanations include insufficient intensity of training stimuli to induce an increase in this marker, or substantial inter-individual variability that may have masked subtle changes. Further studies with larger samples are needed, including additional markers of inflammation and intestinal barrier integrity. Recent research also highlights the need to assess physical activity and fitness in different populations, including non-traditional groups such as esports players. For example, a study assessing physical activity, exercise capacity and fitness level of Polish esports players revealed generally low levels of physical fitness compared with athletic populations [51].

Limitations

The limitations of the study include the limited sample size and lack of a control group and limited statistical power. Moreover, the assessment of intestinal permeability was limited to selected fecal biomarkers, which may not capture the full complexity of gut barrier dynamics. Importantly, other established markers of intestinal integrity, such as lipopolysaccharide (LPS), intestinal fatty acid binding protein (I-FABP), or occludin, were not included.

Another limitation is the lack of detailed validation data regarding the 3-day food diaries, which may be subject to potential reporting errors. Furthermore, possible confounding variables, such as vitamin D supplementation or previous gastrointestinal complaints, were not considered. Future studies should address these limitations, including a more comprehensive set of intestinal permeability markers, validated dietary assessment methods, and the evaluation of additional determinants of gut microbiota as well as psychological aspects.

CONCLUSION

1. Individuals engaged in climbing and alpine activities are at risk of energy restriction and/or low energy availability. The climbers mostly provided adequate amounts of macro- and micronutrients with their diets in exception to selenium, iodine, vitamin D and K.
2. In most of the climbers studied, intestinal permeability was predominantly within the normal range. In some individuals, elevated zonulin values were observed, while AAT concentrations remained within the normal range. Due to the small sample size and the lack of significant correlations with diet, the results should be considered preliminary and require further verification in studies with larger groups.

3. There is a need for nutritional and supplementation education by qualified sports nutritionists to increase awareness among this group of athletes about energy balance and sports nutrition in the preparatory stage before trips to the high mountains.
4. Future research should explore the longitudinal relationship between diet, gut health, and performance at altitude in larger cohorts.

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