Original Article
Effects of low-dose acetazolamide on exercise performance in simulated altitude

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Abstract: Preventive effects of acetazolamide (ACZ) on acute mountain sickness (AMS) are well established but effects on exercise performance at high altitude or in hypoxia have been less considered and are still inconsistent. We hypothesized that low-dose ACZ would not impair exercise performance at simulated high altitude. Thus, the aim of this study was to evaluate the interaction between low-dose ACZ and exercise performance in normobaric hypoxia. Sixteen subjects (8 males and 8 females) were randomly assigned either to receive low-dose ACZ (3×125 mg in 36 hours) or placebo. Incremental cycle spiro-ergometry was performed before and after drug treatment in normobaric hypoxia (inspired fraction of oxygen, FiO_2 = 13.5%; equivalent to about 4000 m). Whereas maximal power output and submaximal exercise responses did not change differently from pre- to post-treatment between ACZ and placebo, absolute and relative VO_{2max} values and maximal oxygen pulse were slightly decreased in hypoxia after ACZ pre-treatment. ANOVA results suggest that aerobic capacity in males might be more affected by ACZ pre-treatment than in females. In conclusion, the presented findings may be of practical importance, possibly more meaningful for female mountaineers, because low-dose ACZ (125 mg bd) was shown to prevent AMS development with similar effectiveness as higher doses. This means that low-dose ACZ would prevent both, AMS development and a pronounced reduction in exercise performance.

Keywords: Acetazolamide, hypoxia, exercise performance, sex

Introduction

A gradual ascent to high altitude represents the most promising strategy for optimal acclimatization [1]. However, in certain cases, i.e. inevitable rapid ascent or considerable susceptibility to develop high altitude illness, acetazolamide (ACZ) is the drug of choice for prevention [2, 3]. The ACZ dose used for prevention has been discussed inconsistently ranging between 250 mg and 1 g per day [2, 4-6]. For instance, Kayser and colleagues demonstrated that rising daily doses from 250 to 500 and 750 mg resulted in increasing reduction of acute mountain sickness (AMS) by 45, 50 and 55% [7]. ACZ acts via inhibition of carbonic anhydrase (CA), an enzyme located particularly in erythrocytes, pulmonary tissues, the brain and the kidneys [3]. The CA inhibition in the proximal renal tubuli induces diuresis by depressing the proximal tubular bicarbonate reabsorption and the distal tubular H^+ secretion resulting in metabolic acidosis, hyperventilation and improved arterial partial pressure of oxygen [3, 8-11]. Whereas the preventive effects of ACZ on AMS are well established, effects on exercise performance at high altitude or in hypoxia have been less considered and are still conflicting. Some studies report impaired performance with ACZ [12-16], and others found no or even beneficial effects [17-20]. It seems likely that higher ACZ doses are more effective in preventing AMS but cause more adverse effects likely including impairment of exercise performance. However, even low ACZ doses (125 mg bd) can effectively prevent AMS [3], but data regarding its effects on exercise performance are lacking. We hypothesized that low-dose ACZ would not impair exercise performance when acutely exposed to simulated high altitude. Thus, the aim of this study was to evaluate the interaction between low-dose ACZ and exercise performance in normobaric hypoxia.

Table 1. Physiological measurements pre- and post-drug intake in normoxia

	ACZ group (n = 8)		CG (n = 8)		ANOVA time x treatment	ANOVA* time x treatment x gender
	Pre	Post	Pre	Post		
Body mass and resting cardiorespiratory parameters						
Body mass [kg]	66.6±10.9	66.0±11.0	67.9±10.8	67.8±10.5	n.s.	n.s.
HR_{rest} [b/min]	77.9±13.0	77.4±11.9	66.4±13.9	72.5±11.6	n.s.	n.s.
$BPsys_{Rest}$ [mmHg]	137.6±15.4	130.6±14.9	141.9±16.9	129.5±19.8	n.s.	n.s.
$BPdia_{Rest}$ [mmHg]	83.1±11.2	83.1±6.1	86.8±9.1	81.9±8.9	n.s.	n.s.
SpO_2 [%]	97.6±0.7	97.5±0.9	97.5±0.5	97.4±0.7	n.s.	n.s.
Haematological parameters						
Erythrocytes [10^6/μl]	5.2±1.2	4.9±0.5	4.7±0.5	4.7±0.4	n.s.	n.s.
Leukocytes [10^3/μl]	7.6±2.7	7.4±2.5	5.3±1.2	5.6±1.3	n.s.	n.s.
Hematocrit [%]	46.2±7.3	48.2±4.7	46.6±4.6	45.1±4.8	.08	n.s.
Haemoglobin [g/dl]	14.6±1.2	14.9±1.7	14.0±1.1	13.7±1.1	n.s.	n.s.
Pulmonary function testing						
FEVC [l]	5.3±1.0	5.5±0.9	5.3±1.0	5.5±1.0	n.s.	n.s.
FEV1 [l]	4.3±0.7	4.5±0.7	4.2±0.8	4.3±0.8	n.s.	n.s.

Data are mean values ± SD. *P*-values in italics indicate a trend toward significance (< 0.1 but > 0.05). *mixed design ANOVA has been used. ACZ, acetazolamide; CG, control group; HR, heart rate; BPsys, BPdia, systolic and diastolic blood pressure; SpO_2, oxygen saturation; FEVC, forced expiratory vital capacity; FEV1, forced expiratory volume in the 1st sec.

Methods

Subjects

Sixteen voluntary young and healthy subjects (8 men and 8 women; age 26.7±2.3 years) not acclimatized to altitude above 1000 m were recruited. Exclusion criteria were any acute or chronic illness or known susceptibility to severe AMS. Both, purely sedentary subjects or competitive athletes have been excluded. The study has been approved by the local Ethics Committee. All subjects provided written informed consent prior to their participation.

Study design and protocol

This randomized controlled double-blind study was conducted at the Department of Sport Science, Medical Section, of the University of Innsbruck, Austria. After gender stratification subjects were randomly assigned either to receive low-dose ACZ (ACZ group; n = 8; 4 males and 4 females) or the control group receiving placebo (CG; n = 8; 4 males and 4 females). After clinical routine examination followed by a 30 min rest, cardiopulmonary exercise testing (pre) was performed in normobaric hypoxia (simulated altitude of about 4000 m; inspired fraction of oxygen, FiO_2 = 13.5%). About 6 hours later participants started with ACZ intake (3×125 mg within 36 hours) and the con-

trol group took placebos (identical in appearance) during the same time period. 48 hours after the first exercise test the same test was repeated in simulated altitude (post). Subjects were advised not to perform intense physical activity at least 2 days before the first exercise test as well as during ACZ and placebo treatment, not to have a heavy meal at least 2 hours before exercise testing but were free to drink ad libitum.

Baseline characteristics

Physical routine examination and the recording of medical history, physical activity, smoking and dietary habits were followed by the assessment of anthropometric data (body mass, height), resting heart rate, systemic blood pressure, ECG, oxygen saturation (SpO_2), lung function (FEVC, FEV1), red (haemoglobin, haematocrit) and white blood cell counts (**Table 1**).

Exercise testing

Participants performed a graded exercise test to exhaustion on a cycle ergometer (Ergoselect 100, Ergoline®, Bitz, Germany). The starting work load was set at 50 watts and was increased by 50 watts every 3 minutes. During the test, heart rate (HR) was registered continuously whereas blood pressure, oxygen saturation measured by finger pulse oximetry (SpO_2)

Table 2. Maximal and submaximal exercise responses pre- and post-drug intake in hypoxia

	ACZ group (n = 8)		CG (n = 8)		ANOVA time x treatment	ANOVA* time x treatment x gender
	Pre	Post	Pre	Post		
Maximal responses						
P_{max} [watts]	225.0±46.3	209.4±37.6	240.6±62.6	218.8±63.7	n.s.	n.s.
P_{rel} [watts/kg]	3.4±0.3	3.2±0.4	3.5±0.5	3.2±0.6	n.s.	n.s.
VO_{2max} [ml/min]	2,587±647	2,395±530	2,619±629	2,647±720	.031	< 0.01
VO_{2max} [ml/min/kg]	38.5±4.7	36.1±3.9	38.2±4.2	38.6±6.1	.047	0.02
SpO_2 [%] at VO_{2max}	70.0±5.5	71.0±5.9	69.0±7.4	69.4±5.1	n.s.	n.s.
HR_{max} [b/min]	181±7.1	179.6±8.3	178.3±11.1	178.6±13.6	n.s.	n.s.
O_2Pulse_{max} [ml/beat]	14.3±3.6	13.4±3.0	14.7±3.3	14.8±4.0	.045	0.02
V_{Emax} [l/min]	117.9±36.0	120.4±29.9	129.8±38.5	128.5±41.6	n.s.	n.s.
V_E/VO_2 at VO_{2max}	43.3±4.2	48.2±5.0	47.0±5.5	46.0±6.4	.06	.08
La_{max} [mmol/l]	8.9±3.1	8.2±2.1	7.9±3.1	8.2±3.7	n.s.	n.s.
$BPsys_{max}$ [mmHg]	224.1±10.2	214.3±11.8	217.9±9.6	217.3±20.4	n.s.	n.s
$BPdia_{max}$ [mmHg]	86.6±8.6	78.3±10.6	87.8±7.7	79.4±10.9	n.s.	n.s.
RPE (Borg 0-20)	19 (18.3-19.8)	19.1 (19-19.8)	19.1 (19-19.8)	19 (18.3-20)	n.s.	n.s.
Submaximal responses						
VO_2 at VT [ml/min]	1,894±488	1,696±465	2,010±592	1,946±665	n.s.	n.s.
V_E at VT [l/min]	65.9±19.2	64.5±19.9	69.0±22.3	63.5±20.4	n.s.	n.s.
HR at VT [b/min]	157.4±14.5	149.0±14.8	151.5±11.5	148.1±14.4	n.s.	n.s.
O_2Pulse at VT [ml/beat]	12.2±3.4	11.4±3.0	13.2±3.5	13.1±4.2	n.s.	n.s.

Data are mean values ± SD. *P*-values in italics indicate a trend toward significance (< 0.1 but > 0.05). *mixed design ANOVA has been used. ACZ, acetazolamide; CG, control group; P, power; VO_2, oxygen uptake; SpO_2, oxygen saturation; HR, heart rate; V_E, minute ventilation; La, blood lactate; BPsys, BPdia, systolic and diastolic blood pressure; RPE, rating of perceived exertion; VT, ventilatory threshold.

and rating of perceived exertion (RPE, Borg) were determined at the end of each stage. Gas exchange variables, including the rate of ventilation (VE), oxygen uptake (VO_2) and carbon dioxide output (VCO_2) were recorded breath by breath with a portable gas analyser (Oxycon Mobile; CareFusion, Germany). The test was considered maximal when 3 of the following criteria were fulfilled: VO_2 plateau at peak exercise, respiratory exchange ratio (VCO_2/VO_2) > 1.10, peak HR reaching at least 90% of the theoretical maximal HR predicted for age and gender, an indication of maximal exhaustion by the participant. Maximal power output (P_{max}) was defined as the last completed work rate plus the fraction of time spent in the final uncompleted work rate multiplied by 50 watts. VO_{2max} was calculated as the mean of all VO_2 values measured within the last 30 s of the test. The ventilator threshold (VT) was defined as the point of a nonlinear increase in VE in combination with an increasing equivalent for oxygen (VE/VO_2) without a concomitant increase in the ventilatory equivalent of CO_2 (VE/VCO_2). Furthermore, at the end of every stage blood lactate concentration ([La]) was measured (Biosen 5040, EKF, Germany) from capillary blood samples drawn from the earlobe.

Statistics

Descriptive statistics are presented as means ± standard deviations (SD). After testing normality mixed-design ANOVA (time x treatment x sex) was used to evaluate treatment and sex effects/interactions. Main outcome measures (P_{max}, VO_{2max}, maximal oxygen pulse) are reported separately for males and females of both groups and paired t-tests were used for the evaluation of pre-post changes within groups. Significance was accepted at P < 0.05. Data analyses were conducted with the use of the SPSS statistical software package.

Results

Study participants completed all measurements. Altitude exposure in the hypoxic chamber was well tolerated by the participants except for one subject who collapsed after the ergometer cycle test in the hypoxic chamber for a few seconds but without any further adverse consequences. No side effects including paresthesias and/or altered taste have been reported during or post ACZ intake.

Changes of body mass, resting cardiopulmonary and haematological parameters from pre-

Table 3. Sec-specific changes of maximal performance parameters from pre- to post-treatment

	ACZ group				CG			
	Males (n = 4)		Females (n = 4)		Males (n = 4)		Females (n = 4)	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post
P_{max} [watts]	262.5±25.0	237.5±25.0	187.5±25.0	181.3±23.9	293.8±31.5	275.0±20.4	187.5±25.0	162.5±25.0
P_{rel} [watts/kg]	3.5±0.3	3.1±0.4	3.3±0.4	3.2±0.4	3.8±0.4	3.6±0.3	3.2±0.3	2.8±0.6
VO_{2max} [ml/min]	3,113.3±358.0	2,814.3±265.0*	2,061.5±332.4	1,975.8±340.1	3,157.8±302.2	3,261.5±388.9	2,079.5±237.2	2,032.8±225.9
VO_{2max} [ml/min/kg]	40.9±3.6	37.2±3.1*	36.0±4.7	35.1±4.8	40.8±3.8	42.5±4.5	35.6±3.2	34.8±5.3
O_2Pulse_{max} [ml/beat]	17.2±1.7	15.8±0.8	11.5±2.3	10.9±2.2	17.5±1.9	18.0±3.0	11.9±1.1	11.6±0.6

Data are mean values ± SD. ACZ, acetazolamide; CG, control group; P, power; VO_2, oxygen uptake. *indicates significant differences between pre and post; changes from pre to post did not differ between sexes of the ACZ group.
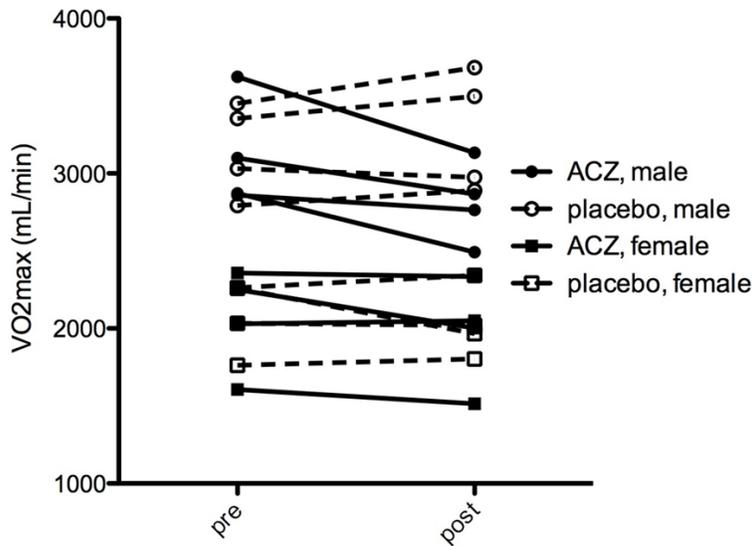
Figure 1. Individual changes of maximal oxygen uptake (VO_{2max}) in males and females from pre- to post-treatment with acetazolamide (ACZ) or placebo.

to post-treatment with ACZ or placebo did not differ between groups (**Table 1**). Only haematocrit tended to increase after ACZ compared to placebo without differences between sexes. Absolute and relative VO_{2max} values and maximal oxygen pulse decreased and VE/VO_2 tended to increase in hypoxia after ACZ pre-treatment compared to placebo (**Table 2**). Maximal power output and submaximal exercise responses did not change differently from pre- to post-treatment between ACZ and placebo. ANOVA results suggest a significant sex-effect of ACZ treatment on VO_{2max} (and maximal oxygen pulse) (**Table 2**). Thus, maximal performance data are reported separately for males and females of both groups (**Table 3**). Significant within-group changes from pre to post were only found for absolute and relative VO_{2max} values in males of the ACZ group however, changes from pre to post did not reach significance between sexes (P = 0.8); (**Table 3**; **Figure 1**). No treatment-dependent sex differences were observed for submaximal exercise performance.

Discussion

The main findings of the present study are that pre-treatment with low-dose (3×125 mg within 36 hours) ACZ slightly affected maximal aerobic capacity but not maximal power output or submaximal exercise responses. To our knowledge, this is the first study evaluating low-dose

ACZ effects on exercise performance in hypoxia.

It has been suggested that ACZ doses of 7-12 mg/kg as used for instance in the study by Garske et al. may inhibit CA in red blood cells and tissue [13] likely resulting in inefficient CO_2 transport during exercise [11-13]. In contrast, participants of the present study showed slightly reduced aerobic capacity at an ACZ dose even below 5 mg/kg. This observation seems to be particularly true for males since females had relatively higher ACZ doses due to their lower body mass. Studies demonstrating compromised exercise performance in acute hypoxia mostly included only male subjects [13-16] or a large proportion of males [12] therefore, at least partly supporting our findings. These studies however, used higher ACZ doses. The tendency of a larger VO_{2max} reduction in males may be simply due to their higher baseline VO_{2max} and/or because they are more sensitive to the performance limiting effect of ACZ than females. Differences in ACZ sensitivity are also supported by the tendency of increased VE/VO_2 in men with ACZ probably linked to more distinct peripheral and/or central chemoreceptor stimulation [18]. On the other hand however, sex-dependent performance differences could also result from benefits of ACZ intake in women. Karnik and colleagues suggested that women in comparison to men have a significantly increased cerebral vasodilatory capacity after ACZ [21]. This might help to maintain cerebral oxygenation and aerobic exercise performance [22] thereby compensating for negative ACZ effects, e.g. arising from the impairment of metabolic processes involved in muscle contraction [18]. Furthermore, decreased plasma volume and maximal stroke volume resulting from enhanced diuresis might also contribute to the reduced VO_{2max} and maximal oxygen pulse observed in the present study. However, this mechanism is rather unlikely because ACZ treatment did not cause sex-dependent different changes in body mass or haematocrit. Although there was a small

decrease in the aerobic capacity after low-dose ACZ, maximal power and submaximal exercise responses remained unaffected. Thus, the presented findings may be of practical importance, possibly more meaningful for female mountaineers, because low-dose ACZ (125 mg bd, a dose less than 5 mg/kg) was found to prevent AMS development with similar effectiveness as 375 mg bd, in males and females as well [4]. This means that low-dose ACZ would prevent both, AMS development and a pronounced reduction in exercise performance. Since submaximal exercise and maximal power output were not affected by low-dose ACZ, especially mountaineers and athletes susceptible to AMS should benefit from ACZ pre-treatment.

Limitations

Of course, the small size of sub-samples may represent an important limitation when interpreting potential sex-differences. However, with regard to the effects of ACZ versus placebo we are convinced of a sufficient study power (based on data provided by Garske et al. [13]). Nevertheless, these findings may only be valid for the specific conditions of the present study (young and healthy subjects; graded exercise in acute hypoxia equivalent to about 4000 m).

In conclusion, low-dose ACZ (125 mg bd) did not affect maximal power output and submaximal exercise performance but compromised slightly maximal aerobic exercise capacity during acute exposure to normobaric hypoxia. Further studies are necessary to evaluate whether women are actually less affected by ACZ pre-treatment than men.

Disclosure of conflict of interest

None.

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