European Journal of Sport Science
The Official Journal of the European College of Sport Science

Altitude-induced effects on muscular metabolic stress and hypertrophy-related factors after a resistance training session

Belen Feriche[1]; Brad J. Schoenfeld[2]; Juan Bonitch-Gongora[1].; Blanca de la Fuente[3]; Filipa Almeida[1]; Javier Argüelles[3]; Cristina Benavente[1]; Paulino Padial[1]

[1]Department of Physical Education and Sport, Faculty of Sport Sciences, University of Granada, Spain.
[2] Department of Health Sciences, CUNY Lehman College, Bronx, New York
[3]High performance Center of Sierra Nevada, Spanish Sport Council, Spain.

FUNDER INFORMATION
This work was supported by the Ministry of Education, Culture and Sport of Spain under Grant DEP2015-64350-P, MINECO/FEDER and PGC2018-097388-B-I00 MCI/AEI/FEDER, UE

ABSTRACT

This study examined the acute effects of exposure to moderate altitude on factors associated with muscular adaptations following whole-body hypertrophy-oriented resistance training (R_T) sessions. Thirteen resistance-trained males completed both counterbalanced standard hypertrophic R_T sessions (3 sets x 10RM, 2 min rest) at moderate-altitude (H; 2320 m asl) and under normoxic conditions (N; <700 m asl). Participants rested 72h between training sessions. Before and after the exercise session, blood samples were obtained for determination of metabolites and ions (lactate, inorganic phosphate, liquid carbon dioxide and calcium) and hormones (testosterone and growth hormone). Session-related performance and perception of effort (s-RPE) were also monitored. Results showed no meaningful differences in performance or s-RPE (8.5±1.4 vs 8.6±0.8 respectively for N and H; p=0.603). All blood variables displayed statistically significant changes throughout the recovery period compared to basal levels ($p<0.05$), except for the testosterone. However, no altitude effect was observed in maximal blood lactate, calcium or anabolic hormones ($p>0.05$). The reduction observed in the liquid carbon dioxide concentration in H (21.11±1.46 vs 16.19±1.61 $mmol \cdot l^{-1}$) seems compatible with an increase in buffering capacity. Compared to N, inorganic phosphate displayed lower recovery values after the R_T in H (2.89±0.64 vs 2.23±0.60 $mg \cdot dl^{-1}$; p=0.007). The results of this study do not support an accentuated effect of acute moderate terrestrial hypoxia on metabolic and hormonal factors linked to muscle growth during hypertrophic resistance training.

INTRODUCTION

The influence of hypoxia on the development of strength and muscle mass is an area of current interest (Friedmann et al., 2003; Kon et al., 2010; Kurobe et al., 2015; Manimmanakorn, Hamlin, Ross, Taylor, & Manimmanakorn, 2013; Manimmanakorn, Manimmanakorn, et al., 2013; Nishimura et al., 2010). Exercise under hypoxic conditions is characterized by a greater desaturation of muscular oxygen, which in turn leads to an increase in anaerobic metabolism and therefore to a potential increase in the production of metabolites (Kon et al., 2010; Kon, Ikeda, Homma, & Suzuki, 2012; Kurobe et al., 2015; Schoenfeld, 2013; Scott, Slattery, Sculley, & Dascombe, 2014). Hypothetically, this metabolic condition supports a benefit of training in a hypoxic environment to enhance the hypertrophic response. In particular, increases in blood lactate (Filopoulos, Cormack, & Whyte, 2017; Kon et al., 2010, 2012; Ramos-Campo, Rubio-Arias, et al., 2017; Scott, Slattery, Sculley, Hodson, & Dascombe, 2015; Scott, Slattery, Sculley, Lockhart, & Dascombe, 2017), growth hormone (GH) (Filopoulos et al., 2017; Kon et al., 2010, 2012; Yan, Lai, Yi, Wang, & Hu, 2016) and circulating ions (Na^+, Cl^- & H^+) (Ramos-Campo, Rubio-Arias, et al., 2017) have been reported after R_T exercise at acute moderate and severe hypoxia. However, other anabolic agents, such us testosterone, have not displayed consistent responses under this environmental condition (Kon et al., 2010, 2012; Yan et al., 2016). Furthermore, additional high-threshold motor units could be recruited due to the heightened anaerobic metabolism in hypoxic conditions (Schoenfeld, 2013; Scott et al., 2017; Scott, Slattery, & Dascombe, 2014). Hence, if the number of muscle fibers stimulated during a training session increase, the training load can increase, and conceivably a larger portion of the muscle could respond and adapt to the training stress (Scott et al., 2015).

Despite the potential effect of the hypoxic training for these purposes, there currently is no consensus in the literature as to the effects of hypertrophic resistance training (R_T) in hypoxia compared to the same protocol performed in normoxia (Ramos-Campo, Scott, Alcaraz, & Rubio-Arias, 2017). Differences in methodologies between studies (populations, training protocols, muscles worked, program duration, and severity of hypoxia), limit the ability to draw strong conclusions from the available literature (Feriche, García-Ramos, Morales-Artacho, & Padial, 2017; Ramos-Campo, Scott, et al., 2017; Scott, Slattery, & Dascombe, 2015). Moreover, to date, studies on the topic have exclusively employed simulated hypoxia. No studies have examined whether terrestrial altitude shows similar results, since it has been suggested that *"altitude"* is a more severe environmental condition that leads to different physiological adaptations (Millet, Faiss, & Pialoux, 2012).

Therefore, the aim of this study was to compare the acute response of a hypertrophy-oriented R_T session under normoxic conditions and after the ascent to a moderate altitude on exercise-induced stress variables associated with muscular gains (metabolites, ions and anabolic hormones). We hypothesized that the exposure to altitude would produce an increased stress response compared to the normoxia condition and thus enhance acute factors associated with muscle hypertrophy.

MATERIALS AND METHODS

Experimental Approach to the problem

In a counter-balanced order, participants performed two standard hypertrophic R_T sessions, one at moderate terrestrial altitude (H; 2320 m asl) and the other under normoxic conditions (N; <700 m asl). Participants were randomly assigned to perform the initial training session in H or N. We compared the session's performance,

perception of effort, acute metabolic stress response, and related factors obtained under both environmental conditions. One week before the first R_T session, subjects engaged in a preparatory session to determine the load associated with their 10 repetition maximum (RM) for each exercise. Afterwards, participants attended the laboratory two days before the start of the study under fasting conditions for baseline anthropometric and resting blood sample testing. All preliminary laboratory assessments were performed under normoxic conditions and participants were requested to abstain physical activity and alcohol intake, and to maintain their customary sleep and diet habits for 48 h before evaluations. To ensure standardized nutritional intake for performance of the R_T sessions, subjects fasted after midnight the day before a training session and were provided with a meal replacement (610 calories) 1.5 hours prior to the start of the warm-up. Both testing sessions were conducted in the morning at the same time of day and under the following environmental conditions: ~22º C and ~60% humidity, or ~22º C and ~28% humidity, respectively, for N and H conditions. The hypoxic environmental condition was evaluated by assessing the arterial oxygen saturation (SaO_2) before the start of the warm-up.

Subjects

Thirteen male volunteers (age: 22.31 ± 2.59 years; height: 178.31 ± 4.96 cm; body mass: 76.92 ± 9.17 kg) participated in the study. Subjects had no existing cardiorespiratory or musculoskeletal disorders, were free from consumption of anabolic steroids or any other legal or illegal agents known to increase muscle size for the previous year, and had not been exposed to more than 3-4 consecutive days of altitudes above 1500 m at least two months before study. All participants were considered experienced lifters, defined as consistently lifting weights a minimum of 3 times per

week for at least 12 months. This study was approved by the Local University Research Ethics Committee and conducted in accordance with the Helsinki Declaration. Informed written consent was obtained from all participants prior to beginning the study.

Hypertrophic Resistance Training session

The R_T protocol comprised six exercises per session targeting the major muscle groups of the body in the following order: flat barbell press, barbell military press, wide grip lat pulldown, seated cable row, barbell back squat, and machine leg press. These exercises were chosen based on their common inclusion in bodybuilding- and strength-type R_T programs. A standardized warm up of 15 min (cardiovascular activity, joint mobility & stretching and 2 sets of 10 repetitions [the first with 20 kg and the second at 50% 1RM estimated from the 10RM preliminary test, 2 min rest] of bench press, seated cable row and back squat) was completed at the beginning of each session. Participants lived at a low altitude to ensure that responses were specific to acute hypoxia exposure. Participants rested 72h between training sessions and were instructed to refrain from performing any additional exercise for the duration of the study.

The routines for each session comprised 3 sets of 10 RM per exercise. Sets were carried out to the point of momentary concentric muscular failure (inability to perform another concentric repetition while maintaining proper form). Cadence of repetitions was carried out in a controlled fashion, with a concentric action of approximately one second and an eccentric action of approximately two seconds. Subjects were afforded 2 minutes rest between sets. The load was adjusted for each exercise as needed on successive sets to ensure that subjects achieved failure in the target repetition range (8-12 RM). Barbell exercises were performed with calibrated equipment (Eleiko) and used

for comparison between conditions. All routines were directly supervised by the research team to ensure proper performance of the respective routines.

Absolute training load by exercise (kg), repetitions to failure and ratings of perceived exertion (s-RPE) were monitored during and after each training session respectively. s-RPE was obtained by showing a graphical scale to participants 30 min after completing the training session (Foster et al., 2001). Total volume-load was calculated as the sum of the load lifted x the repetitions of each set and exercise. Immediately before each session, SaO_2 was measured in duplicate using a pulse oximeter (Wristox 3100; Nonin, Plymouth, MN, USA). Participants displayed a mean SaO_2 value of 94.2 ± 1.3 and 98.0 ± 1.5% in H and N respectively ($P<0.001$). Height (Seca 202, Seca Ltd., Hamburg, Germany) and body mass (Tanita BC 418 segmental, Tokyo, Japan) were assessed before the start of the first session.

Blood Measurements

Before and after the training sessions, venous blood samples were taken for determination of metabolites/ions (lactate, inorganic phosphate, liquid carbon dioxide and calcium) and hormones (testosterone and growth hormone). The basal condition was established in N conditions from a blood analysis collected after 48h of rest, two days prior to the first training session. Blood extractions after the RT sessions were performed at the same altitude condition that the corresponding session. All blood samples were extracted by specialized staff.

Immediately following each training session, the antecubital vein of the arm was canalized via a catheter. The catheter remained permeable by using physiological saline. Between 0.5 and 5 ml of blood was extracted in minutes 3, 5, 10, 15 and 30 post exercise in quantities according to the variables to be determined in each case. An

amount of 2 ml of blood before each extraction was discarded to avoid dilution of the sample. In all cases, blood samples were kept at cold conditions and centrifuged in the following 4 hours during 10 min at 3000 rpm. Finally, 500µl aliquots were stored at -70°C until use.

Blood was processed according to the following methodology: 1) minutes 3, 5, 10, 15 and 30, 10 µl of blood were reserved for the determination of the blood lactate concentration [Lac] and analyzed by a photometric procedure (Dr. Lange, LP 20 plus, Berlin, Germany); 2) Ions and hormones were measured at minutes 5, 10, 15 and 30 after exercise; for this, 3 ml of blood was collected in tubes without additives. Determinations were performed in a COBAS C-311 and E-411 System (Roche, Basel, Switzerland). To minimize bicarbonate loss, specimens were kept tightly capped.

Statistical Analyses

Descriptive statistics are presented as mean ± standard deviation (SD). Normal distributions of the data were confirmed using a Shapiro-Wilk test. A two-factor ANOVA with repeated measures was used to assess the effect of time during the recovery (within-participant factor with 4 levels [minutes 5, 10, 15 and 30] and the environmental condition (within-participant factor with 2 levels [H *vs.* N]) on the ions and hormone variables (i.e. inorganic phosphate, liquid CO_2, calcium, testosterone and growth hormone). When the sphericity assumption in ANOVAs was violated (Mauchly's test), a Greenhouse-Geisser correction was employed. Post-hoc tests were performed by means of Bonferroni procedures when appropriate for multiple comparisons. Generalized Eta-Squared measures of effect size and thresholds (<0.04 [small], 0.04>ES<0.36 [medium] and >0.36 [large]) were calculated along with ANOVA effects. Differences with respect to the basal conditions of all blood variables

and between condition comparisons for maximal blood lactate concentration, RPE and physical performance were analyzed through paired samples t-tests. Standardized differences (i.e. Cohen's *d* effect sizes) were calculated as the mean altitude-normoxia change divided by the pooled standard deviations on all dependent variables to quantify the magnitude of the change. Threshold classifications of >0.2 [small], >0.6 [moderate], >1.2 [large] and very large [>2] were used to categorize the magnitude of effects.

All analyses were performed using the software package SPSS (version 24.0: SPSS, Inc., Chicago, IL, USA). Effects were considered significant at $p \leq 0.05$.

RESULTS

During both training sessions, all participants reached muscular failure between 8 to 12 repetitions. Mean group training load, number of repetitions, and total volume-load accumulated during the 3 sets of the 3 free barbell exercises in both training conditions are displayed in Table 1. Slight differences in the load or in repetitions to failure were observed due to the load adjustment on successive sets to ensure maintenance of the target repetition range. Small to moderate mean variations of less than 1 repetition per set were observed between conditions in the flat barbell press and the barbell back squat exercises, while the absolute load displaced showed a non-significant mean increase from 0.24 to 1.54 kg respectively. A trivial increase in the total volume-loads was observed in N condition in the flat barbell press and back squat (p=0.23; 95%IC [111.4; 9.8]; ES=0.18 and p=0.043; 95%IC [390.3; 7.3]; ES=0.50 respectively). H and N training sessions displayed similar effort perception 30 minutes after the end of the training (8.5 ±1.4 vs 8.6±0.8 respectively for N and H; p=0.603; 95%IC [-1.00; 0.5]; ES=0.14). The lactate recovery curve was similar in N and H conditions (Fig 1) and no differences in maximal blood lactate concentration were

observed (16.08 ± 3.73 vs 15.52 ± 2.66 mMol·l^{-1} in N and H; p= 0.322; 95%IC [-1.73; 0.62]; ES=0.17).

[Insert Table 1 about here]

[Insert Figure 1 about here]

Figure 2 displays the results of calcium (Ca_2^+), liquid carbon dioxide (CO_2-L) and inorganic phosphate (Pi) values under basal conditions and the comparison of the recovery curve between altitude conditions. Significant increases in Ca_2^+ (P<0.001) and reductions in Pi and CO_2-L were observed after the R_T sessions versus the basal condition in H (p<0.001) and N (p<0.05) conditions. ANOVA showed a main effect of *time* in all these variables due to a change in the values each five minutes throughout the recovery period (p<0.05). The Pi and CO_2-L displayed an *environmental condition* main effect (F = 12.22, p = 0.007, η^2_G = 0.58 and F = 14.42, p = 0.004, η^2_G = 0.59) due to the lower values across the recovery time in the hypoxia condition (2.89 ± 0.64 vs 2.23 ± 0.60 mg·dl^{-1}; 21.11 ± 1.46 vs 16.19 ± 1.61 mmol·l^{-1}, for N and H respectively). No *recovery time* × *environmental condition* effect was observed in ions (F = 0.689, p = 0.515, η^2_G = 0.06; F = 2.03, p = 0.133, η^2_G = 0.18; F = 0.711, p = 0.553, η^2_G = 0.06 respectively for Ca_2^+, Pi and CO_2-L).

[Insert Figure 2 about here]

No differences between the basal condition and the minute 5 of recovery were observed in testosterone. However, there was a main effect of *time* (F = 14.37, p = 0.001, η^2_G = 0.59) due to a progressive reduction in testosterone throughout the recovery. This behavior was similar in both altitude conditions (F = 2.50, p=0.145, η^2_G

= 0.20) (Fig 3). Growth hormone (GH) increased by more than 82 fold after the training session compared to basal values. No recovery *time* or *environmental condition* main effect was observed (F = 0.38, p=0.603, η^2_G = 0.04; F = 1.02, p=0.339, η^2_G = 0.10 respectively). No interaction *recovery time* × *environmental condition* effect was observed in any of the studied hormones (F = 2.03, p= 0.131, η^2_G = 0.17; F = 0.39, p= 0.654, η^2_G = 0.04 respectively for testosterone and GH) (Fig 3).

[Insert Figure 3 about here]

DISCUSSION

The aim of this study was to investigate the effect of acute moderate altitude on metabolic stress and associated responses induced by a standard whole-body hypertrophy-oriented R_T session. Contrary to what we hypothesized, the results did not display differences in RPE, maximal blood lactate, calcium nor anabolic hormones (testosterone and GH) responses when a similar R_T (3 sets x 10RM, 2 min rest) was performed under both environmental conditions. Even post-exercise, Pi displayed lower values in H when compared to N. Thus, the results of this study do not support the theory that R_T combined with acute terrestrial hypoxia enhances factors linked to muscle growth.

An increase in markers of performance during R_T at moderate altitude could be expected based on the possibility of recruiting additional high-threshold motor units via an augmented anaerobic metabolism in hypoxic conditions (Schoenfeld, 2013; Scott, Slattery, & Dascombe, 2015; Scott et al., 2017; Scott et al., 2014). In the present study, although some differences were detected in the training load and in the number of repetitions to failure between N and H, the mean absolute values amounted to ~1

repetition less in H or ~ 1.6 kg more in N in bench press and back squat exercises. However, despite that interpretation of training load magnitude must jointly consider the modulation of the load and the number of repetitions to failure accumulated, trivial reductions in the total volume-load were attained in the flat bench press and the back squat in H conditions. These results reflect the mean reduction of 0.36 and 0.95 repetitions per set in H and the consistent small but nonsignificant corresponding increase in the workload (between 0.2 [~ 0.5%] and 1.5 kg [~ 2%] respectively for each exercise). In addition, all participants reached muscular failure between 8 to 12 repetitions and maintained a constant cadence, which allows direct comparison of the training response in both conditions (see Table 1).

Consistent with previous studies that employed a hypertrophy-oriented training routine (6-12 RM; 8-10 RPE) (Helms, Cronin, Storey, & Zourdos, 2016), a mean s-RPE value of ~ 8.5 was observed in this study. No differences in s-RPE were detected between altitude conditions. This result is consistent with those of Scott et al. 2015 (Scott et al., 2015), who reported no changes in RPE between moderate (FiO_2 =16%) and severe (FiO_2 =13%) hypoxia compared to N after a traditional heavy R_T session. Conversely, significant increases in RPE between high hypoxia (FiO_2= 13%) with respect the moderate (FiO_2= 16%) and N conditions have been observed after a high-intensity resistance circuit (Ramos-Campo, Rubio-Arias, et al., 2017) or several all-out sets of continuous jumps (Álvarez-Herms et al., 2016). The contradictory RPE response after R_T under hypoxic conditions could be explained by the differences in the way that RPE was collected (immediately after the training bout *vs*. after each set *vs*. 30 min after the end of the training bout [s-RPE]), the exercise protocols compared (jumping *vs.* traditional *vs.* high-intensity resistant circuit) and/or the implicit differences in the loads used (body weight *vs*. moderate load *vs.* heavy load).

Despite the strong influence of oxygen availability on phospho-creatine (PCr) resynthesis, some studies hypothesize that recovery times longer than 90 s could compromise hypoxic effects and thus mitigate the potential anabolic impact of metabolic stress (Ho, Huang, Chien, Chen, & Liu, 2014; Kurobe et al., 2015; Scott et al., 2017). Therefore, the somewhat longer inter-set rest periods employed in this study (2 min) could have favored PCr re-phosphorylation (McMahon & Jenkins, 2002; Yquel, Arsac, Thiaudiere, Canioni, & Manier, 2002) and also the degree to which metabolic byproducts are removed from the muscle before the ensuing set (Scott et al., 2015). This is compatible with our findings of an absence of change in blood lactate and circulating Ca_2^+ between conditions. Some studies have reported similar blood lactate results at moderate hypoxia after a high intensity R_T circuit (Ramos-Campo, Rubio-Arias, et al., 2017), high load traditional R_T (Scott et al., 2018), explosive strength session (Álvarez-Herms et al., 2016) or high-intensity intermittent exercise (Feriche et al., 2007). Contrarily, others found higher maximal lactate levels after low to high intensity R_T at moderate (Scott et al., 2015, 2017) and severe hypoxia R_T (Filopoulos et al., 2017; Kon et al., 2010, 2012; Ramos-Campo, Rubio-Arias, et al., 2017). The participants training status and R_T protocol design (i.e. muscles involved, number of exercises, intensity and volume) could affect the magnitude of this response, since our results reported maximal blood lactate above that reached in all the aforementioned studies (~ 16 *vs.* 5 to 8 $mMol \cdot l^{-1}$). However, the lower CO_2-L levels detected in H, also shown in other studies (Ramos-Campo, Rubio-Arias, et al., 2017), clearly indicate a higher muscle buffering response at this condition, which could reduce the net blood lactate measured in H. Moreover, lactate enters the bloodstream via concentration gradient differences and by the use of carbon monoxide transporters (Bonen, 2001; Halestrap & Meredith, 2004) reducing the effectiveness of both mechanisms as lactate concentration increases.

Accordingly, it is possible that lactate levels may have been higher in H, but the greater buffering activity and the slow lactate release from muscle in H may have limited the ability to differentiate an increase in glycolytic pathway in H (Kawada, 2005), explaining the similar maximal lactate values observed in both sessions. This hypothesis requires further study.

As noted above, we observed an unexpected reduction in Pi after the R_T in H (~20% minor in H along the 30 min of recovery) compared to N. The maximal aerobic capacity of single muscles is more limited by peripheral factors than central ones during ATP production under no O_2 limitations (McMahon & Jenkins, 2002). Thus, throughout the initial hours of exposure to moderate hypoxia, there is an increase in ventilation, submaximal heart rate and cardiac output (Hahn & Gore, 2001), which act as compensatory mechanisms to increase muscle buffering capacity, O_2 availably and PCr restauration rate (Ramos-Campo, Rubio-Arias, et al., 2017). Moreover, the shift to the right of the oxyhemoglobin curve (O_2-Hb) during exercise in H (Bohr Effect, the increase in 2,3-bisphosphoglycerate and pH elevation), improves the oxygen release in active muscles (Gerbino, Ward, & Whipp, 1996). Taken together, the acute improvements in cardiac function and the O_2-Hb curve, combined with the non-predominant aerobic pathway during the R_T methodology assessed, could favor recovery between sets, accelerating the regeneration of ATP and PCr in acute H. In accordance with the Pi results, the Ca_2^+ levels did not change after the training session (Glaister, 2005). Ramos-Campos et al. also found similar serum Ca_2^+ during R_T at normoxia, moderate or severe acute hypoxia (Ramos-Campo, Rubio-Arias, et al., 2017). Further research is needed to establish the role of the oxygen availability on ATP and PCr resynthesis during intervening rest periods.

The results obtained did not reflect a significant altitude effect on testosterone and GH after the training sessions. The GH value, in particular, seems to be mediated by blood lactate and H^+ levels (Kon et al., 2010; Scott et al., 2015). Indeed, increases in blood lactate and GH after low to moderate R_T sessions have been detected in other studies under acute hypoxic conditions (Filopoulos et al., 2017; Kon et al., 2010, 2012; Yan et al., 2016). Our results are consistent with this finding, since similar elevations in lactate and GH levels were found in both cases. In regard to testosterone, several studies have reported increases in this hormone after a hypertrophy-oriented R_T session (Ahtiainen, Kraemer, & Häkkinen, 2003; Smilios, Pilianidis, Karamouzis, & Tokmakidis, 2003). Consistent with our findings, other researches have failed to detect differences in post-exercise testosterone secretion under N and H conditions (Kon et al., 2010, 2012; Kurobe et al., 2015; Yan et al., 2016), finding the maximal value occurs immediately after the R_T session (Yan et al., 2016). A relationship between testosterone response and metabolic stress has not been well-established (Durand et al., 2003), and other factors related to the training protocol (i.e. muscle mass implicated, intensity or volume) could be more influential (Ahtiainen et al., 2003). It therefore can be speculated that the acute serum testosterone response to R_T may not be significantly affected by hypoxia (Kon et al., 2012; Yan et al., 2016). We would note that although chronic anabolic hormonal levels have been considered by some as key factors for skeletal muscle development, the role of the acute post-exercise endocrine response on muscle growth remains dubious (Fink, Schoenfeld, & Nakazato, 2018). Additional research is needed to fully elucidate the role of these hormones on muscular adaptations.

Our study had some limitations that need to be addressed. For one, we only monitored 30 min of post-exercise recovery. Therefore, our insights into markers are limited to this period; it remains possible that results may have diverged at later time

points. For another, we cannot rule out the possibility of a placebo effect since it was not feasible to employ a single- or double-blind design at terrestrial altitude. However, this seems unlikely given that participants were not aware nor informed about the expected effects of altitude on performance.

In conclusion, the exposure to acute moderate terrestrial hypoxia during a whole-body hypertrophy-oriented R_T session does not appear to differentially affect the metabolic stress response compared to R_T in normoxia. The acute response observed in this study therefore cannot necessarily be extrapolated to longer periods of exposure, raising the possibility that a more stressful response to the same exercise occurs after the initial acclimatization phase. Future studies should seek to identify the mechanisms involved in the hypertrophic response and clarify the effect of prolonged continuous or intermittent exposure to moderate altitude combined with longitudinal R_T programs on muscular adaptations.

ACKNOWLEDGMENTS

The authors thank all the participants who volunteered for this investigation.

DECLARATION OF INTEREST STATEMENT

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

REFERENCES

Ahtiainen, J., Kraemer, W. J., & Häkkinen, K. (2003). Acute Hormonal and Neuromuscular Responses and Recovery to Forced vs. Maximum Repetitions Multiple Resistance Exercises Musculoskeletal Complaints and Repetition Number View project. *Article in International Journal of Sports Medicine*. https://doi.org/10.1055/s-2003-41171

Álvarez-Herms, J., Julià-Sánchez, S., Gatterer, H., Blank, C., Corbi, F., Pagès, T., … Viscor, G. (2016). Anaerobic training in hypoxia: A new approach to stimulate the

rating of effort perception. *Physiology & Behavior*, *163*, 37–42. https://doi.org/10.1016/J.PHYSBEH.2016.04.035

Bonen, A. (2001). The expression of lactate transporters (MCT1 and MCT4) in heart and muscle. *European Journal of Applied Physiology*, *86*(1), 6–11. https://doi.org/10.1007/s004210100516

Bosco, C., Colli, R., Bonomi, R., Von Duvillard, S. P., & Viru, A. (2000). *Monitoring strength training: neuromuscular and hormonal profile*. *Med. Sci. Sports Exerc* (Vol. 32). Retrieved from http://www.msse.org

Durand, R. J., Castracane, V. D., Hollander, D. B., Tryniecki, J. L., Bamman, M. M., O'Neal, S., … Kraemer, R. R. (2003). Hormonal responses from concentric and eccentric muscle contractions. *Medicine and Science in Sports and Exercise*, *35*(6), 937–943. https://doi.org/10.1249/01.MSS.0000069522.38141.0B

Feriche, B., Delgado, M., Calderón, C., Lisbona, O., Chirosa, I. J., Miranda, M. T., … Álvarez, J. (2007). The effect of acute moderate hypoxia on accumulated oxygen deficit during intermittent exercise in nonacclimatized men. *Journal of Strength and Conditioning Research*, *21*(2). https://doi.org/10.1519/R-19095.1

Feriche, B., García-Ramos, A., Morales-Artacho, A. J., & Padial, P. (2017). Resistance Training Using Different Hypoxic Training Strategies: a Basis for Hypertrophy and Muscle Power Development. *Sports Medicine - Open*, *3*(1). https://doi.org/10.1186/s40798-017-0078-z

Filopoulos, D., Cormack, S. J., & Whyte, D. G. (2017). Normobaric hypoxia increases the growth hormone response to maximal resistance exercise in trained men. *European Journal of Sport Science*, *17*(7), 821–829. https://doi.org/10.1080/17461391.2017.1317834

Fink, J., Schoenfeld, B. J., & Nakazato, K. (2018). The role of hormones in muscle

hypertrophy. *The Physician and Sportsmedicine*, *46*(1), 129–134. https://doi.org/10.1080/00913847.2018.1406778

Foster, C., Florhaug, J. A., Franklin, J., Gottschall, L., Hrovatin, L. A., Parker, S., … Dodge, C. (2001). A new approach to monitoring exercise training. *Journal of Strength and Conditioning Research*, *15*(1), 109–115. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/11708692

Friedmann, B., Kinscherf, R., Borisch, S., Richter, G., Bartsch, P., & Billeter, R. (2003). Effects of low-resistance/high-repetition strength training in hypoxia on muscle structure and gene expression. *Pflugers Archiv : European Journal of Physiology*, *446*(6), 742–751. article. https://doi.org/10.1007/s00424-003-1133-9

Gerbino, A., Ward, S. A., & Whipp, B. J. (1996). Effects of prior exercise on pulmonary gas-exchange kinetics during high-intensity exercise in humans. *Journal of Applied Physiology (Bethesda, Md. : 1985)*, *80*(1), 99–107. https://doi.org/10.1152/jappl.1996.80.1.99

Glaister, M. (2005). Multiple Sprint Work. *Sports Medicine*, *35*(9), 757–777. https://doi.org/10.2165/00007256-200535090-00003

Hahn, A. G., & Gore, C. J. (2001). The Effect of Altitude on Cycling Performance. *Sports Medicine*, *31*(7), 533–557. https://doi.org/10.2165/00007256-200131070-00008

Halestrap, A. P., & Meredith, D. (2004). The SLC16 gene family from monocarboxylate transporters (MCTs) to aromatic amino acid transporters and beyond. *Pflugers Archiv European Journal of Physiology*, *447*(5), 619–628. https://doi.org/10.1007/s00424-003-1067-2

Helms, E. R., Cronin, J., Storey, A., & Zourdos, M. C. (2016). Application of the Repetitions in Reserve-Based Rating of Perceived Exertion Scale for Resistance

Training. *Strength and Conditioning Journal* , *38*(4), 42–49. https://doi.org/doi: 10.1519/SSC.0000000000000218

Ho, J.-Y., Huang, T.-Y., Chien, Y.-C., Chen, Y.-C., & Liu, S.-Y. (2014). Effects of acute exposure to mild simulated hypoxia on hormonal responses to low-intensity resistance exercise in untrained men. *Research in Sports Medicine*, *22*(3), 240–252. https://doi.org/10.1080/15438627.2014.915834

Ho, J., Kuo, T. Y., Liu, K. L., Dong, X. Y., & Tung, K. (2014). Combining normobaric hypoxia with short-term resistance training has no additive beneficial effect on muscular performance and body composition. *Journal of Strength and Conditioning Research*, *28*(4), 935–941. article. https://doi.org/10.1519/JSC.0000000000000289

Kawada, S. (2005). What phenomena do occur in blood flow-restricted muscle? *International Journal of KAATSU Training Research*, *1*(2), 37–44. https://doi.org/10.3806/ijktr.1.37

Kon, M., Ikeda, T., Homma, T., Akimoto, T., Suzuki, Y., & Kawahara, T. (2010). Effects of acute hypoxia on metabolic and hormonal responses to resistance exercise. *Medicine and Science in Sports and Exercise*, *42*(7), 1279–1285. https://doi.org/10.1249/MSS.0b013e3181ce61a5

Kon, M., Ikeda, T., Homma, T., & Suzuki, Y. (2012). Effects of Low-Intensity Resistance Exercise Under Acute Systemic Hypoxia on Hormonal Responses. *Journal of Strength and Conditioning Research*, *26*(3), 611–617. https://doi.org/10.1519/JSC.0b013e3182281c69

Kurobe, K., Huang, Z., Nishiwaki, M., Yamamoto, M., Kanehisa, H., & Ogita, F. (2015). Effects of resistance training under hypoxic conditions on muscle hypertrophy and strength. *Clinical Physiology and Functional Imaging*, *35*(3),

197–202. https://doi.org/10.1111/cpf.12147

Manimmanakorn, A., Hamlin, M. J., Ross, J. J., Taylor, R., & Manimmanakorn, N. (2013). Effects of low-load resistance training combined with blood flow restriction or hypoxia on muscle function and performance in netball athletes. *Journal of Science and Medicine in Sport / Sports Medicine Australia*, *16*(4), 337–342. https://doi.org/10.1016/j.jsams.2012.08.009

Manimmanakorn, A., Manimmanakorn, N., Taylor, R., Draper, N., Billaut, F., Shearman, J. P. J. P., & Hamlin, M. J. M. J. (2013). Effects of resistance training combined with vascular occlusion or hypoxia on neuromuscular function in athletes. *European Journal of Applied Physiology*, *113*(7), 1767–1774. https://doi.org/10.1007/s00421-013-2605-z

McMahon, S., & Jenkins, D. (2002). Factors Affecting the Rate of Phosphocreatine Resynthesis Following Intense Exercise. *Sports Medicine*, *32*(12), 761–784. https://doi.org/10.2165/00007256-200232120-00002

Millet, G. P., Faiss, R., & Pialoux, V. (2012). Point: Counterpoint: Hypobaric hypoxia induces/does not induce different responses from normobaric hypoxia. *Journal of Applied Physiology*, *112*(10), 1783–1784. https://doi.org/10.1152/japplphysiol.00067.2012

Nishimura, A., Sugita, M., Kato, K., Fukuda, A., Sudo, A., & Uchida, A. (2010). Hypoxia increases muscle hypertrophy induced by resistance training. *International Journal of Sports Physiology and Performance*, *5*(4), 497–508. article.

Ramos-Campo, D. J. D. J., Rubio-Arias, J. A. J. A., Dufour, S., Chung, L., Ávila-Gandía, V., & Alcaraz, P. E. P. E. (2017). Biochemical responses and physical performance during high-intensity resistance circuit training in hypoxia and

normoxia. *European Journal of Applied Physiology*, *117*(4), 809–818. https://doi.org/10.1007/s00421-017-3571-7

Ramos-Campo, D.J., Rubio-Arias, J. A., Dufour, S., Chung, L., Ávila-Gandía, V., & Alcaraz, P. E. (2017). Biochemical responses and physical performance during high-intensity resistance circuit training in hypoxia and normoxia. *European Journal of Applied Physiology*, *117*(4), 809–818. https://doi.org/10.1007/s00421-017-3571-7

Ramos-Campo, Domingo J., Rubio-Arias, J. Á., Freitas, T. T., Camacho, A., Jiménez-Diaz, J. F., & Alcaraz, P. E. (2017). Acute Physiological and Performance Responses to High-Intensity Resistance Circuit Training in Hypoxic and Normoxic Conditions. *Journal of Strength and Conditioning Research*, *31*(4), 1040–1047. https://doi.org/10.1519/JSC.0000000000001572

Ramos-Campo, Domingo J., Scott, B. R., Alcaraz, P. E., & Rubio-Arias, J. A. (2017). The efficacy of resistance training in hypoxia to enhance strength and muscle growth: A systematic review and meta-analysis. *European Journal of Sport Science*, 1–12. https://doi.org/10.1080/17461391.2017.1388850

Schoenfeld, B. J. (2013). Potential Mechanisms for a Role of Metabolic Stress in Hypertrophic Adaptations to Resistance Training. *Sports Medicine*, *43*(3), 179–194. https://doi.org/10.1007/s40279-013-0017-1

Scott, B.R., Slattery, K. M., & Dascombe, B. J. (2015). Intermittent hypoxic resistance training: Is metabolic stress the key moderator? *Medical Hypotheses*, *84*(2), 145–149. https://doi.org/10.1016/j.mehy.2014.12.001

Scott, Brendan R., Slattery, K. M., Sculley, D. V., & Dascombe, B. J. (2014). Hypoxia and Resistance Exercise: A Comparison of Localized and Systemic Methods. *Sports Medicine*, *44*(8), 1037–1054. https://doi.org/10.1007/s40279-014-0177-7

Scott, Brendan R., Slattery, K. M., Sculley, D. V., Hodson, J. A., & Dascombe, B. J. (2015). Physical Performance During High-Intensity Resistance Exercise in Normoxic and Hypoxic Conditions. *Journal of Strength and Conditioning Research*, *29*(3), 807–815. https://doi.org/10.1519/JSC.0000000000000680

Scott, Brendan R., Slattery, K. M., Sculley, D. V., Lockhart, C., & Dascombe, B. J. (2017). Acute Physiological Responses to Moderate-Load Resistance Exercise in Hypoxia. *Journal of Strength and Conditioning Research*, *31*(7), 1973–1981. https://doi.org/10.1519/JSC.0000000000001649

Scott, Brendan R., Slattery, K. M., Sculley, D. V., Smith, S. M., Peiffer, J. J., & Dascombe, B. J. (2018). Acute physiological and perceptual responses to high-load resistance exercise in hypoxia. *Clinical Physiology and Functional Imaging*, *38*(4), 595–602. https://doi.org/10.1111/cpf.12451

Scott, B. R., Slattery, K. M., & Dascombe, B. J. (2014). Intermittent hypoxic resistance training: does it provide added benefit? *Frontiers in Physiology*, *5*, 397. https://doi.org/10.3389/fphys.2014.00397

Smilios, I., Pilianidis, T., Karamouzis, M., & Tokmakidis, S. P. (2003). Hormonal Responses after Various Resistance Exercise Protocols. *Med. Sci. Sports Exerc*, *35*(4), 644–654. https://doi.org/10.1249/01.MSS.0000058366.04460.5F

West, D. W. D., & Phillips, S. M. (2010). Anabolic Processes in Human Skeletal Muscle: Restoring the Identities of Growth Hormone and Testosterone. *The Physician and Sportsmedicine*, *38*(3), 97–104. https://doi.org/10.3810/psm.2010.10.1814

Yan, B., Lai, X., Yi, L., Wang, Y., & Hu, Y. (2016). Effects of Five-Week Resistance Training in Hypoxia on Hormones and Muscle Strength. *Journal of Strength and Conditioning Research*, *30*(1), 184–193.

https://doi.org/10.1519/JSC.0000000000001056

Yquel, R. J., Arsac, L. M., Thiaudiere, E., Canioni, P., & Manier, G. (2002). Effect of creatine supplementation on phosphocreatine resynthesis, inorganic phosphate accumulation and pH during intermittent maximal exercise. *Journal of Sports Sciences*, *20*(5), 427–437. https://doi.org/10.1080/026404102317366681

Table 1. Training load and number of repetitions accumulated during the three training sets in both training conditions

	Training Load (kg)					Rep x set					Total Volume-Load (Kg)	
	N	**H**	**Δ (Kg)**	**ES**	**P [95% CI]**	**N**	**H**	**Δ (rep)**	**ES**	**P [95% CI]**	**N**	**H**
Flat barbell press	66.3 ±11.4	66.5 ±11.4	0.2	0.02	0.557 [-0.64;1.12]	10.1 ±1.0	9.7 ±0.9	0.4	0.38	0.009 [-0.6; -0.11]	1997.4 ±322.2	1936.9 ±332.8*
Barbell military press	36.0 ±4.6	35.0 ±4.0	-1.0	-0.24	0.125 [-2.38;0.33]	8.6 ±0.6	8.5 ±0.5	-0.1	-0.14	0.661 [-0.45;0.29]	922.2 ±123.2	891.8 ±125.3
Barbell back squat	74.3 ±12.7	75.9 ±15.0	1.5	0.11	0.219 [-1.78; -0.11]	11.0 ±0.7	10.1 ±1.2	0.9	0.99	0.029 [-1.78;-0.11]	2460.8 ±463.6	2261.9 ±331.4*

N: normoxic condition; H: altitude condition; 10RM: 10 repetition maximum; Rep x set: mean repetitions completed per set; Change: mean percentage of change between conditions in the load or repetition sets; Δ: absolute mean group difference between conditions in the load or repetitions sets; ES: effect size [calculated as (H mean-N mean) ÷ (pooled standard deviation)]; P: p value; 95% IC: 95% interval of confidence. * $p<0.05$: Significant difference between N and H conditions in the Total Volumen-Load variable

FIGURE CAPTIONS

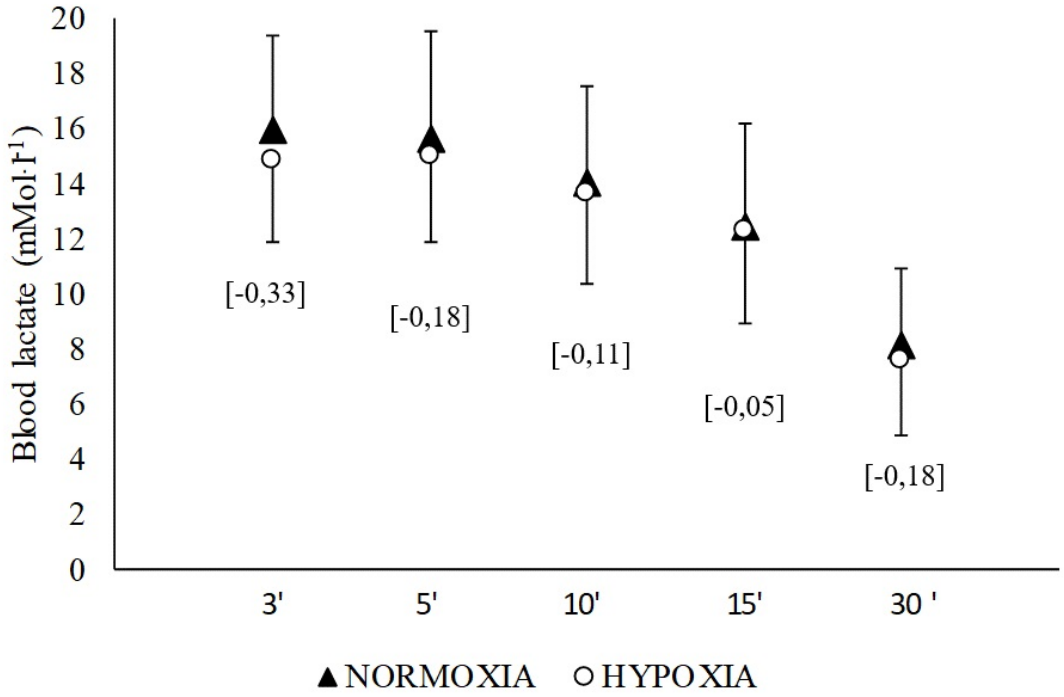

Fig 1. Blood lactate concentration throughout the recovery period in normoxia and hypoxia condition. No significant differences between conditions; [ES]: effect size calculated as (H mean-N mean) ÷ (pooled standard deviation).

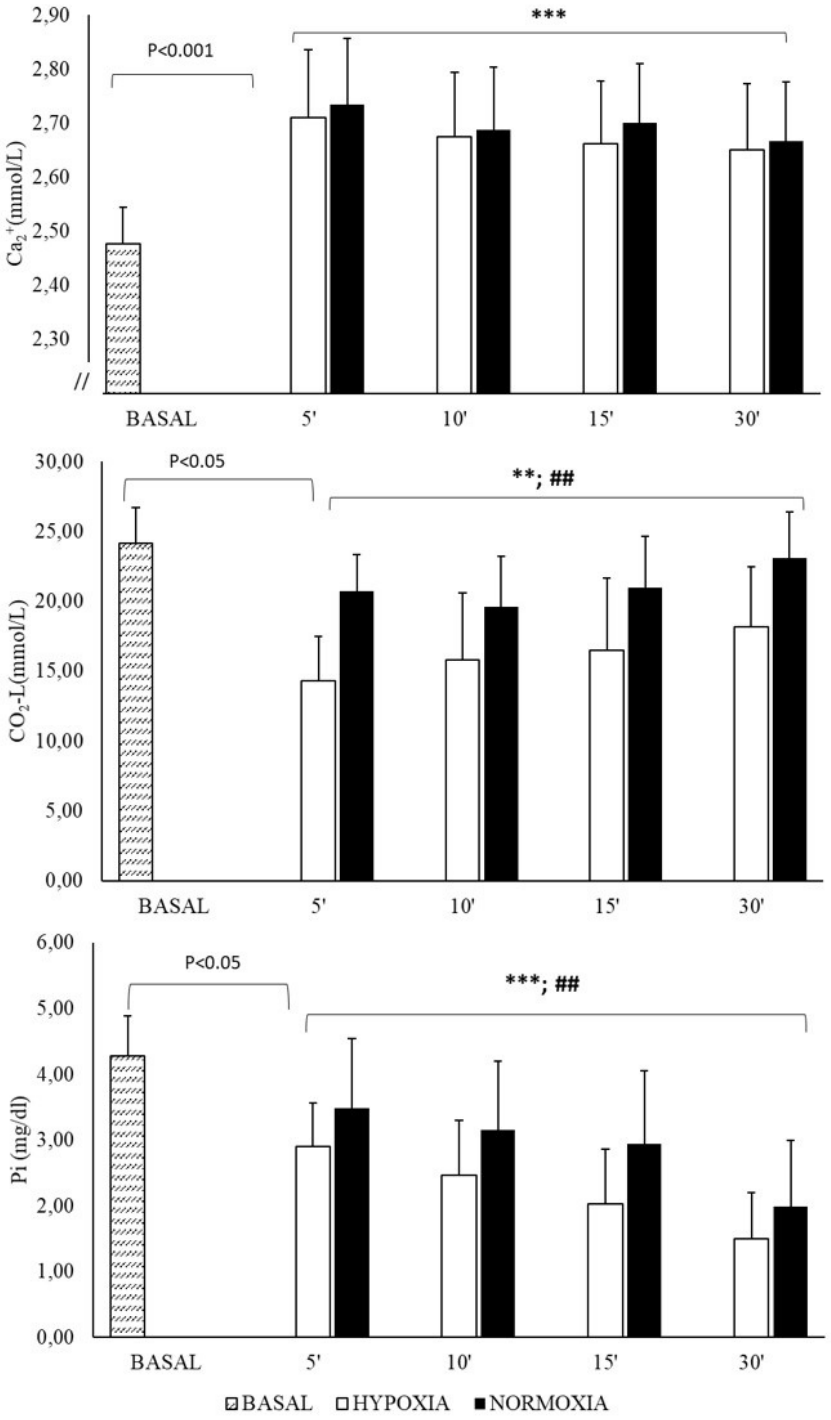

Fig 2. Comparison of calcium (Ca_2^+), liquid carbon dioxide (CO_2-L) and inorganic phosphorus (Pi) values in basal and between altitude condition through the recovery period. * time effect (* p<0.05; **p<0.01; ***p<0.001; # altitude effect (# p<0.05; ## p<0.01; ### p<0.001; ¥ interaction time x altitude condition (¥ p<0.05; ¥¥ p<0.01; ¥¥¥ p<0.001

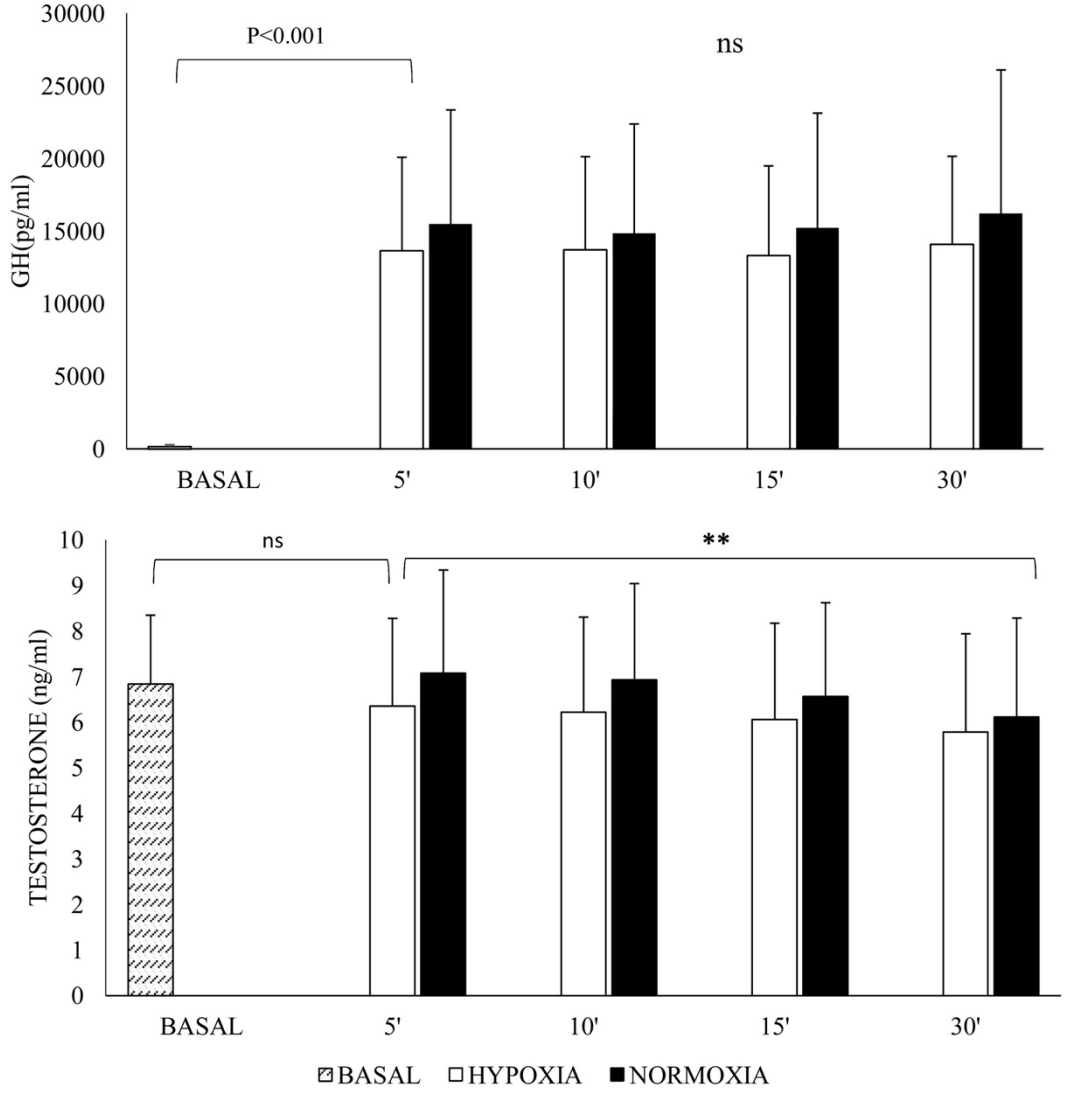

Fig 3. Comparison of testosterone and hormone of growth (GH) values in basal and between altitude condition through the recovery period. * time effect (* p<0.05; **p<0.01; ***p<0.001; # altitude effect (# p<0.05; ## p<0.01; ### p<0.001; ¥ interaction time x altitude condition (¥ p<0.05; ¥¥ p<0.01; ¥¥¥ p<0.001