Title: Blood volumes following pre-season heat versus altitude: a case study of Australian Footballers

Submission type: Brief report – case study

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Running head: Comparison of pre-season heat vs altitude

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Abstract word count: 249

Text only word count: 1,738

Number of figures and tables: 3

This manuscript has been read and approved by all the listed co-authors and the authors declare no potential conflicts of interest.
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ABSTRACT

Purpose: There is debate as to which environmental intervention produces most benefit for team sport athletes, particularly comparing heat and altitude. This quasi-experimental study aimed to compare blood volume (BV) responses to heat and altitude training camps in Australian Footballers. Methods: The BV of seven professional Australian Footballers (91.8±10.5 kg, 191.8±10.1 cm) was measured throughout three consecutive spring/summer pre-seasons. During each pre-season, players participated in altitude (year 1 and year 2) and heat (year 3) environmental training camps. Year 1 and year 2 altitude camps were in November/December in the USA, while the year 3 heat camp was in February/March in Australia after a full exposure to summer heat. BV, red cell volume (RCV) and plasma volume (PV) were measured at least three times during each pre-season. Results: RCV increased substantially following altitude in both year 1 (d=0.67) and year 2 (d=1.03), before returning to baseline four weeks post-altitude. Immediately following altitude, concurrent decreases in PV were observed during year 1 (d=-0.40) and year 2 (d=-0.98). With spring/summer training in year 3, BV and PV were substantially higher in January than temporally matched post-altitude measurements during year 1 (BV d=-0.93, PV d=-1.07) and year 2 (BV d=-1.99, PV d=-2.25), with year 3 total BV, RCV and PV not changing further despite the 6-day heat intervention. Conclusions: We found greater BV after training throughout spring/summer conditions, compared with interrupting spring/summer exposure to train at altitude in the cold, with no additional benefits observed from a heat camp following spring/summer training.

Keywords: hypoxia, environmental, plasma volume, red cell volume
INTRODUCTION

Many high-intensity, intermittent team sports undertake prolonged pre-season training, which are important for developing physical capacities essential for competitive success. Environmental stimuli such as altitude\(^1\) or heat\(^2\) have been applied during these periods, in an attempt to enhance players’ physiology and performance. Indeed, improvements in team-sport athletes’ running performance has been shown following heat,\(^2\) altitude\(^1\) and a combination of heat and hypoxic exposures.\(^3\)

High-intensity training leads to increases in plasma volume (PV)\(^4\) which occur within 48 hours of the first training bout and may be further augmented by hot environments.\(^5\) Red cell volume (RCV) is less influenced by training alone, but may be augmented by living/training at altitude for more than two weeks.\(^6\) While these environmental interventions (i.e. altitude/hypoxic or hot environments) offer haematological\(^6\) and non-haematological benefits,\(^7\) changes in PV (heat\(^4\)) and RCV (altitude/hypoxia\(^6\)) are thought to be the primary adaptations contributing to improved exercise capacity following such interventions. The time course of these haematological responses is also important to consider as PV adaptations can dissipate in as short as 10 days with detraining and RCV usually normalizes by four weeks of returning to sea-level.\(^8\) However, some evidence suggests that environmentally induced increases in both PV and RCV may be maintained for up to four weeks in team sport athletes continuing with pre-season training programs.\(^3\)

There has recently been debate as to which environmental interventions produce the most beneficial changes in physiology and performance, particularly for team sports.\(^5\) However, there is a paucity of data comparing physiological and/or performance responses to different environmental interventions in team sport athletes. Therefore, the aim of this case study was to compare hematological responses to altitude and heat interventions in elite Australian Football (AF) players.
METHODS

Forty-three AF players were tracked over three pre-seasons (November-March 2011-2014). Twenty-five players were on the clubs list for all three seasons and individuals were eliminated if data were missing from any measurements (see Figure 1) due to not participating in all camps/training because of injury (n=8) or team planning (n=8), with two players unavailable on specific testing days. This resulted in a final cohort of seven players (mean±SD: 91.8±10.5 kg, 191.8±10.1 cm). All subjects provided written informed consent and this study was approved by the Human Research Ethics Committee at Australian Catholic University. Results from year 1 and year 2 have previously been reported.

The primary training location in every year was Melbourne, Australia, with the following addition of off-site training camps:

- **Year 1** – 21-day ‘altitude’ camp, Flagstaff, Arizona, USA
- **Year 2** – 19-day ‘altitude’ camp, Park City, Utah, USA
- **Year 3** – 6-day ‘heat’ camp, Gold Coast, Queensland, Australia

Timeline of hematological measures:

- **Year 1** – 15th November, 16th December, 10th January
- **Year 2** – 27th November, 5th December, 19th December, 16th January
- **Year 3** – 14th January, 24th February, 7th March

Blood volume (BV), PV and RCV were assessed via venous and capillary blood samples in combination with carbon monoxide (CO) rebreathing to calculate total haemoglobin mass (Hb mass). Preceding venous blood-sample collection, subjects’ were seated for 15 minutes. The same hemoximeter (OSM3, Radiometer, Copenhagen, Denmark) was used for year 1 and 2 measurements, while a different hemoximeter (ABL80, Radiometer, Copenhagen, Denmark) was used during year 3.

INSERT FIGURE 1 ABOUT HERE
Venous blood samples collected in Melbourne (i.e. all samples excluding year 2 measures at altitude) were analyzed using a Sysmex XE-5000 Automated Hematology Analyzer (Roche Diagnostics, Castle Hill, Australia) at St Vincent’s Hospital Pathology (Fitzroy, Australia). Altitude samples (i.e. during year 2 camp) were analyzed using a Sysmex XT-4000i Automated Hematology Analyzer (Sysmex, Lincolnshire, USA) at Park City Medical Centre (Park City, USA). Mean corpuscular haemoglobin concentration (MCHC), RCV, BV and PV were calculated as follows:

\[
MCHC = \frac{\text{venous haematocrit (\%)} \times \text{venous haemoglobin concentration (g/dl)}}{100}
\]

\[
\text{RCV (mL)} = \frac{(\text{Hb mass (g)})}{(\text{MCHC})} \times 100
\]

\[
\text{BV (mL)} = \frac{(\text{Hb mass (g)})}{\text{haemoglobin concentration (g/dL)}} / 0.91
\]

\[
\text{PV (mL)} = \text{BV (mL)} - \text{RCV (mL)}
\]

Our typical error for these measures were calculated using those data without any intervention; that is, January and February data from year 3 on all players with duplicate measures who participated that year (n=20). The typical errors for MCHC, RCV, BV, PV and Hb mass were 2.0, 3.9, 3.2, 3.8 and 2.8 %, respectively.

Weather data were recalled from the Australian Government Bureau of Meteorology [Melbourne (37.81°S, 144.97°E), Gold Coast (27.94°S, 153.43°E)] and National Oceanic and Atmospheric Administration [Flagstaff, Arizona (35.15°N, 111.68°W), Park City, Utah (40.62°N, 111.53°W)] databases.

**Statistical analysis**

A magnitude-based approach\textsuperscript{10} was used to detect effects of practical importance. Changes were assessed relative to the smallest worthwhile change (SWC), set to a small effect size (d=0.2 x the between-participant SD) and reported as mean change/difference ±90% confidence limits. Changes were deemed ‘substantial’ if there was >75% likelihood of the difference exceeding the SWC.
RESULTS

During year 1 (d=0.67) and year 2 (d=1.03) there was a substantial increase in RCV following altitude, that returned to baseline in January (see Figure 2). Post-altitude RCV during year 1 (d=0.74) and year 2 (d=1.57) was substantially higher than RCV in January (d=0.74) of year 3.

In year 1, PV was highest during November, before substantially decreasing in December (d=-0.40) and January (d=-0.74). During year 2, PV decreased from November to December (d=0.98) and January, (d=0.47). All year 3 PVs were substantially higher than December and January values during year 1 (but not November) and all year 2 measurements.

Year 1 BV substantially decreased from November to January (d=-0.59). In year 2, BV increased from November to December (d=0.81), returning to baseline in January. All year 3 BV measurements were substantially higher than January BV during year 1 and year 2.
**DISCUSSION**

Our main finding is that total BV and PV after 8 weeks of warm-weather (spring/summer), pre-season training was higher than the preceding two years, when warm-weather training was interrupted for 3 weeks to complete altitude training camps in cold conditions. These findings are particularly meaningful as they are taken from real world data, thereby producing ecological validity and relevance to practitioners. Immediately post-altitude in *years 1 and 2*, BVs were similar to *year 3* values, due to altitude-induced increases in RCV. But 4 weeks after altitude, RCV returned to baseline without a compensatory increase in PV. This erythropoietic timeline following altitude exposure is consistent with previous results in team sport athletes.⁴

Plasma volume increases following high intensity training⁴ and environmental heat may further augment PV expansion.⁲ During January in *year 1 and 2*, PVs were relatively low considering the athletes’ training status (i.e. 2-3 months of pre-season training) and the warm spring/summer conditions of Melbourne. In contrast, January PV in *year 3* was higher, which was likely affected by warm training environments throughout November-December and suggests that these athletes had capacity to increase PV which was not realized in *year 1 and 2*. During *years 1 and 2*, athletes experienced regular temperatures below freezing while in the northern hemisphere winter. As previous longitudinal data shows that PV is suppressed during colder months,¹¹ this cold exposure may have contributed to lower PV.

In addition to cold environments, positive heat adaptations can be blunted when heat training interventions are combined with living in hypoxia.¹² McCleave et al showed heat-induced increases in PV and BV do not occur when the same heat exposure is combined with living in hypoxia (13 h·d⁻¹, FiO₂ = 14.4%).¹² Following stays at altitude, a complex hormonal cascade initiates active red blood cell destruction, a process known as neocytolysis.¹³ This hormonal cascade is stimulated by lower erythropoietin levels, and may also influence PV. In this case study, relatively low PVs during *year 1 and 2* persisted at least one month after returning to sea-level, despite hot (∼28 ± 6 °C) living/training conditions during this time. As RCV normalizes during the same period, our work showed lower BVs (driven by low PV) following winter altitude training camps, when compared with exclusively training in the Australian summer. While specific detail of training content is not available, this should not be overlooked when interpreting these data as training prescription can also mediate BV responses. Indeed, the overall periodization varied slightly during *year 3* in this case study, as eliminating the altitude camp, and associated international travel, allowed for increased training frequency (and therefore overall volume) during November/December.
Year 3 also describes selected heat adaptations during an AF pre-season. Initial year 3 measurements were taken after eight weeks of warm-weather pre-season training in Melbourne and show higher PV and BV than almost all year 1 and 2 measurements. Initial year 3 BVs and PVs were higher than those previously reported after a ‘successful’ heat training intervention in professional AF (PV–57.0 vs 54.6 ml/kg; BV–90.9 vs 86.1 ml/kg) and there were no additional increases following the 6-day ‘heat’ camp undertaken during March. This suggests that players had maximized PV adaptations prior to the first measurement in year 3 and could be approaching a physiological ‘ceiling’ over this length training cycle (i.e. 3-4 months). These data suggest that warm temperatures in the Melbourne [home base for 9/18 Australian Football League (AFL) teams] summer are sufficient to maximize heat-related physiological adaptations.

Limitations

While this work adds to the understanding of physiological responses to environmental stimuli, it must be acknowledged that performance data are not included, and these observations do not constitute a causal link to performance changes. Detailed information on training content is also not available, which may impact performance and physiological variables. The order of interventions should also be considered as all subjects undertook the heat intervention in year 3, following two years of additional training (including altitude camps). While this work does not represent a controlled experimental study, these real world data with repeat measures in the same elite team sport athletes across multiple years has a high degree of ecological validity.

Conclusion

More optimal BV profiles (i.e. higher PV and BV) are evident in AF players after completing a full pre-season training in warm environmental conditions, compared with the inclusion of cold, altitude camps within the corresponding period. There appears to be no additional haematological benefit for southern-based (e.g. Melbourne) AF teams traveling north to complete ‘heat’ training camps during the pre-season. This may inform planning of pre-season training camps/locations, as physiological benefits are often cited as a driving factor in these decisions.
Training in the heat should be chosen over cold, altitude environments, if the goal is to optimize athletes’ haematological profile.

Heat training adaptations are evident in Melbourne during pre-season, spring/summer months. Therefore, AF teams (and other Australian winter team sports) may not achieve additional benefits traveling to warmer climates during this period.
The authors would like to thank the players and staff at Collingwood Football Club for their support of this research project. No additional external funding was received to complete this work and the authors declare no conflicts of interest.


Table 1. Melbourne and training camp temperatures during years 1-3 during for each month, separated into total days in that month and training days only. Training camp locations; year 1 Flagstaff (elevation ~2100 m), USA, year 2 Park City (elevation ~2000 m), USA and year 3 Gold Coast (elevation < 20 m), Australia. Values are mean temperature (°C) ± standard deviation, with number of days spent in each condition represented in brackets. No temperatures are shown after the last day of data collection in each year and ‘rest period’ days (see Figure 1) are not included.

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^ Substantially higher than year 1 * substantially higher than year 2 # substantially higher than year 3.

NA=not applicable.
FIGURE LEGENDS

Figure 1. Timeline of BV measurements over years 1-3.

Figure 2. Relative A) Red Cell Volume, B) Plasma Volume, and C) Total Blood Volume during years 1-3 (data is for same seven players during each year). Year listed in brackets next to testing date. Substantially higher than all other measurements during that year; substantially lower than all year 3 measurements for that variable. Individual data points defined by symbol shape.
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