PHYSIOLOGICAL AND HEMATOLOGICAL ADAPTATIONS TO ENDURANCE TRAINING IN ELITE MIDDLE AND LONG-DISTANCE RUNNERS: IMPLICATIONS FOR PERFORMANCE OPTIMIZATION

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Abstract

Endurance training (EnT) is a critical component for elite middle and long-distance runners, necessitating a comprehensive examination of the hematological and physiological adaptations that occur throughout a training cycle. This study investigated the effects of EnT on various physiological and hematological factors that influence running performance. A well-structured training program with dynamic intensities and intervals was developed. A quasi-experimental approach was used to conduct pre- and post-tests. The key findings revealed substantial improvements, particularly in erythrocyte volume and hemoglobin (Hgb) mass. Specifically, the RBC count increased significantly from a pre-training mean of 5.18 (±0.40) million/L to 5.45 (±0.39) million/L post-training (t = 5.23, P = 0.001). Furthermore, maximal oxygen consumption (VO_{2max}) demonstrated a marked increase from 71.9 (±1.27) to 78.4 (±2.02) ml/kg/min (t = 16.24), indicating enhanced aerobic capacity. In addition to these improvements, resting heart rate (RHR) exhibited a significant reduction, decreasing from 57 (±3) to 53 (±4) beats per minute (bpm) (t = 4.48, P = 0.001). Collectively, these physiological enhancements accounted for an impressive explanatory power of 86.9% for performance outcomes, particularly evident in the 5000m time trial (TT). These results underscore the importance of EnT programs in optimizing performance for elite runners, highlighting the need of individualized training regimens that promote the physiological adaptations essential for maximizing athletic potential.

Keywords: Endurance training, Hematological adaptations, physiological adaptations, Oxygen consumption, high-intensity training, Red blood cells

INTRODUCTION

Endurance training (ET) is a fundamental component in the development of athletes across diverse sports disciplines, significantly contributing to enhancements in athletic physiology and performance (Mujika, Bourdillon, Zelenkova, Vergnoux, & Millet, 2024a). Attaining elite status as an middle and long-distance athlete (MLDA) necessitates extensive and specialized training, often involving countless hours of dedication and effort (Aagaard & Andersen, 2010; Steiner, Maier, & Wehrlin, 2019). The pathway to success in this field requires an unwavering commitment to hard training regimens, which typically encompass the completion of vast distances annually and the systematic improvement of both physical and technical skills (Joyner & Coyle, 2008). Central to this process is the incorporation of advanced training methodologies and the integration of strength and muscle development techniques focusing on optimizing overall athletic performance (Paquette et al., 2017).

EnT encompasses a multitude of factors that influence athletic performance and general well-being (MacInnis & Gibala, 2017). While the physical dimensions of training are frequently emphasized, the impact of EnT on hematological adaptations (HemA) is equally critical (Tilahun Muche et al., 2021). Endurance MLDA often experience significant alterations in HemA, including RBC counts, Hgb levels, and Htc, all of which play a vital role in performance and overall well-being (Mujika et al., 2024; Schmidt and Prommer, 2008; Shivalingaiah et al., 2015). Recent evidence underscores the importance of high-intensity interval training (HIIT) in modulating these HemA (Koç, Özen, Abanoz, & Pulur, 2018; Stankovic, Djordjevic, Trajkovic, & Milanovic, 2023). By integrating HIIT with traditional EnT methods, both elite and recreational athletes can optimize their athletic outcomes (Nybo et al., 2010; Ramírez-Campillo et al., 2014).

A comprehensive understanding of the effects of various training modalities on HemA is essential for refining training programs and maximizing MLDA potential (Schmidt & Prommer, 2008, 2008). The relationship between ET, HEMA, and athletic performance highlights the necessity for continued research to explain the mechanisms driving these physiological changes (Mancera-Soto, Chamorro-Acosta, Ramos-Caballero, Torrella, & Cristancho-Mejía, 2022). This knowledge is crucial not only for enhancing athletic performance but also for ensuring the long-term well-being of endurance MLDA

Ethiopia, research has highlighted the effectiveness of tapering strategies, such as HIIT low-volume and HIIT moderate-volume training, in improving RBC count, Hgb concentration, and Htcs (Htc) (Mujika et al., 2024). Altitude training, which enhances oxygen-carrying capacity (Mujika, Bourdillon, Zelenkova, Vergnoux, & Millet, 2024b), further contributes to improved endurance performance (Boullosa et al., 2020). Additionally, EnT fosters chronic adaptations in thyroid hormone metabolism and other biochemical markers, aiding MLDA in managing competitive stress (Haugen, Sandbakk, Seiler, & Tønnessen, 2022).

The dynamic interplay between ET, HA, and athletic performance underscores the necessity for personalized training strategies that maximize physiological adaptations while safeguarding long-term health (Brisebois, Rigby, & Nichols, 2018). A holistic approach ensures that MLDA can achieve their full potential and excel in their respective disciplines (Haugen et al., 2022). This study aims to investigate the effects of EnT on the physiological and HemA influencing running performance among selected male elite MLDA.

METHODS

Study Design

A quasi-experimental research design was employed to examine the effects of EnT protocols on physiological and HemA influencing running performance among elite male Ethiopian endurance MLDA. The study spanned six weeks and utilized systematic data collection methods to ensure reliability and validity.

Participants and Sampling

Twenty-one elite male MLDA specializing in mid-distance (1,500m and 3,000m) and long-distance (5,000m and 10,000m) events were purposefully selected based on their representation in international competitions and their willingness to participate in the study. MLDA with health conditions that could potentially affect study outcomes were excluded from participation.

Ethical Compliance

Prior to participation, all MLDA were thoroughly briefed on the study's objectives and procedures. Informed consent was obtained, and measures to ensure confidentiality and data protection were strictly implemented. The experiments complied with the current laws of the country, Ethiopia in which they were performed.

(Birhanu, Ambelu, Berhanu, Tesfaye, & Woldemichael, 2017; MacInnis & Gibala, 2017).

Physiological adaptations induced by EnT span several domains, including cardiovascular, respiratory, muscular, and metabolic systems (Mancera-Soto et al., 2022; Montero et al., 2017). These adaptations significantly enhance performance and consistency (Alcaraz-Ibañez & Rodríguez-Pérez, 2018; Barnes & Kilding, 2015). For instance, increased cardiac output, driven by elevated stroke volume and left ventricular hypertrophy, enhances the heart's pumping efficiency (Birhanu et al., 2017). Elite endurance MLDA often exhibit bradycardia, with RHR below 60 beats per minute, alongside high maximal cardiac output, facilitating superior oxygen delivery and utilization during exertion (MacInnis & Gibala, 2017). Similarly, respiratory adaptations improve pulmonary diffusion capacity and VO_{2max}, which are critical for middle- and long-distance events (Kenney, Wilmore, & Costill, 2022).

Skeletal muscle adaptations resulting from EnT include increased mitochondrial density, enhanced capillarization, and shifts in muscle fiber composition toward oxidative profiles, thereby enabling sustained aerobic metabolism (Alcaraz-Ibañez & Rodríguez-Pérez, 2018). Enhanced oxidative enzyme activity and glycogen storage further extend endurance by delaying the onset of fatigue (Wutzler, Yu, Schrumpf, & Zaehle, 2022). Improvements in neuromuscular coordination reduce energy wastage, while metabolic adaptations facilitate better lipid utilization and lower blood lactate levels during submaximal exercise (Bayati, Farzad, Gharakhanlou, & Agha-Alinejad, 2011). Additionally, hormonal shifts, such as increased insulin sensitivity and modulated catecholamine release, further optimize energy utilization and recovery (Hughes, Ellefsen, & Baar, 2018; Paquette et al., 2017).

Research has emphasized the influence of training load, specificity, age, and baseline fitness on the HemA induced by EnT (MacInnis & Gibala, 2017). Key improvements associated with EnT include increased Hgb mass, RBC volume, and white blood cell (WBC) count. However, chronic EnT can also lead to exercise-induced anemia, characterized by reduced Hgb concentration despite increases in total Hgb mass, underscoring the necessity for balanced training protocols (Birhanu et al., 2017; Prommer, Sottas, Schoch, Schumacher, & Schmidt, 2008). Coaches must design training programs that prioritize athlete health and sustainable performance (Nybo et al., 2010).

EnT has been shown to be essential for enhancing cardiovascular fitness, muscular strength, and mental resilience, benefiting MLDA across various disciplines (Jafer, Mondal, & Abdulkedir, 2019). In physiological adaptations, and performance metrics were compared using a one-way repeated measures ANOVA. To test for differences between modalities, two-way repeated measures ANOVA was utilized. Paired t-tests were conducted to compare $test_0$ and $test_1$ values within each group. Principal Component Analysis (PCA) was conducted to evaluate the significant differences between $test_0$ and $test_1$. Regression analysis was employed to evaluate the association of various factors with performance. All analyses were performed using R software (R Core Team, 2022), version 4.2.3.

RESULTS

The EnT program resulted in significant improvements across several HA, as detailed in Table 1. The RBC count increased from a $test_0$ training mean of 5.18 (±0.40) million/µL to 5.45 (±0.39) million/µL $test_1$ (t = 5.23, P < 0.001), demonstrating a strong correlation between $testest_0$ and $test_1$ values (r = 0.81, P < 0.01). Notably, RBC counts exhibited a consistent increase across all weight categories, with a mean change of 5.97%.

The mass Hgb also rose significantly, from 15.1 (±1.1) g/dL to 16.0 (±1.1) g/dL (t = 4.92, P < 0.01). Heavier MLDA recorded higher mean changes (5.97%). Htc levels demonstrated a highly significant increase, rising from 42.5% (±3.12) to 46.9% (±3.24) (t = 9.86, P < 0.001). Similar to Hgb, Htc levels increased across all weight groups (mean change = 10.45%), with heavier MLDA showing higher values (Figure 1). In contrast, the mean WBC count exhibited a non-significant increase, from 7.5 (±1.9) to 7.7 (±1.77) (t = -1.58, P > 0.05), although a robust correlation was observed between $test_0$ and $test_1$ WBC values (r = 0.99, P < 0.01) (Table 1).

RHR decreased significantly from 57 bpm (±3) to 53 bpm (±4) $test_1$ (t = 4.48, P < 0.001). Additionally, VO_{2max} increased markedly, from 71.9 (±1.27) ml/kg/min to 78.4 (±2.02) ml/kg/min (t = 16.24, P < 0.001). VO_{2max} values improved across all weight categories, with heavier individuals generally exhibiting slightly higher VO_{2max} values (mean change = 8.61%, Figure 1).

Data Collection Protocols

Blood samples were collected at baseline (pre-test, $test_0$) and after the six-week training intervention (post-test, $test_1$). Participants adhered to a 48-hour period of rest from intense activity and a 12-hour fasting regimen before sampling. Blood samples (10 ml) were drawn from the antecubital vein and analyzed in a hematology laboratory to assess Hgb mass (g/dL), Htc mass (%), WBC count (thousand/µL), and RBC count (million/µL).

Key physiological parameters, including indirect VO_{2max} (ml/kg/min), 400m speed test (ST), and RHR, were measured at both $test_0$ and $test_1$ stages. VO_{2max} data were collected using Garmin devices during event-specific time trials to ensure accuracy. Athletic performance was assessed through standardized time trials (s) conducted on a 400m track, and MLDA provided qualitative feedback on perceived performance improvements during the intervention.

Training Protocol

A structured training regimen with progressive intensity was implemented over the six-week period. Each session commenced with a zero-minute warm-up, followed by 15 minutes of dynamic stretching and mobilization exercises. The main training segment involved continuous running at 70–85% of VO_{2max}, performed as a single 60-minute session, two 30-minute sessions, or three 20-minute intervals at a consistent pace. Training also included a 3km cool-down and 10 minutes of stretching. Weekly training volume began at 80% of the MLDA' baseline and increased incrementally by 10% each week until reaching a maximum of 120 km. This progressive and systematic approach aimed to optimize endurance adaptations while minimizing the risk of overtraining or injury.

Data Analysis

Statistical tools, such as t-test, ANOVA, regression analysis were employed to analyze changes in HA, physiological adaptations, and performance metrics over time at a 95% confidence level. Variation in HA,

Although the scatter plot in Figure 3b illustrates the regression model of $test_1$ VO_{2max} values, the model did not fit the data well (-0.097). Regression analyses identified predictors of performance improvements. Model 1, which included RHR, ST times, and D_{12min}, explained 58.9% of the variance in $test_1$ training performance (R^2 = 0.59, P = 0.001). The addition of endurance-specific metrics in Model 2 improved the explanatory power to 86.9% (R^2 = 0.87, P < 0.001).

The D_{12min} improved significantly, increasing from 3723 meters (±57) to 4009 meters (±90) (t = 17.86, P < 0.001). Similarly, the TT improved from 14:00 minutes (±7 seconds) to 13:53 minutes (±8 seconds) (t = 5.49, P < 0.001) (Figure 2). Conversely, the ST exhibited a non-significant decrease from 58 seconds (±2) to 57 seconds (±3). A strong correlation was observed between improvements in VO_{2max} and D_{12min} (r = 0.92) (Figure 3a).

variance in the data, while five components explained 76%. The inclusion of eight components captured over 90% of the dataset's variability

The inclusion of HemA in Model 3 yielded marginal gains, achieving an R^2 of 0.87 (P = 0.001).

Principal component analysis (PCA) revealed that the first three components accounted for 59% of the

Table 1: Summary of statistical comparison for HemA and Performance parameter at *$test_0$* and *$test_1$* (EnT intervention**)**

Parameter	**Test**	**Max**	**Min**	**Mean ± SD**	**Variance**	**Mean Change ± SD**	***t-test***	***r***
RBC (million/µL)	t_0	5.84	4.55	5.18 ± 0.40	0.16	-0.28 ± 0.24	5.23**	0.81**
	t_1	6.12	4.81	5.45 ± 0.39	0.15			
WBC (thousand/µL)	t_0	10.6	4.5	7.5 ± 1.9	3.5	-0.1 ± 0.30	-1.58	0.99**
	t_1	9.3	5.92	7.7 ± 1.77	0.15			
Hgb (g/dL)	t_0	17	12.6	15.1 ± 1.1	1.3	-0.88 ± 0.82	4.92**	0.73**
	t_1	17.5	13.8	16.0 ± 1.1	1.2			
Htc(%)	t_0	49	37	42.5 ± 3.12	9.76	-4.4 ± 0.02	9.86**	0.80**
	t_1	53	42	46.9 ± 3.24	10.5			
RHR (bpm)	t_0	60	52	57 ± 3	9	3.8 ± 3.89	4.48**	0.38
	t_1	60	48	53 ± 4	15			
D_{12min} (m)	t_0	3810	3620	3723 ± 57	3201	-29 ± 73.52	17.86**	0.58**
	t_1	4130	3800	4009 ± 90	8146			
VO_{2max} (ml/kg/min)	t_0	73.9	69.6	71.9 ± 1.27	1.6	-6.4 ± 1.7	16.24**	0.57*
	t_1	81	73.7	78.4 ± 2.02	4.06			
Speed test (s)	t_0	61	54	58 ± 2	5	1.19 ± 3.34	1.63	0.12
	t_1	62	52	57 ± 3	8			
TT (min)	t_0	14:13	13:47	14:00 ± 0:07	0:44	0:07 ± 0:05	5.49**	0.71**
	t_1	14:10	13:44	13:53 ± 0:08	1:04			
Race Time (min)	t_0	13:46	12:57	13:18 ± 0:14	3:16	0:02 ± 0:06	1.43	0.89**
	t_1	13:42	12:50	13:16 ± 0:12	1:22			

Notes: t = pre-post tests, *r* = correlation coefficient, SD = Standard deviation. Statistical Significance: $p < 0.05$ = *, $p < 0.01$ = **.

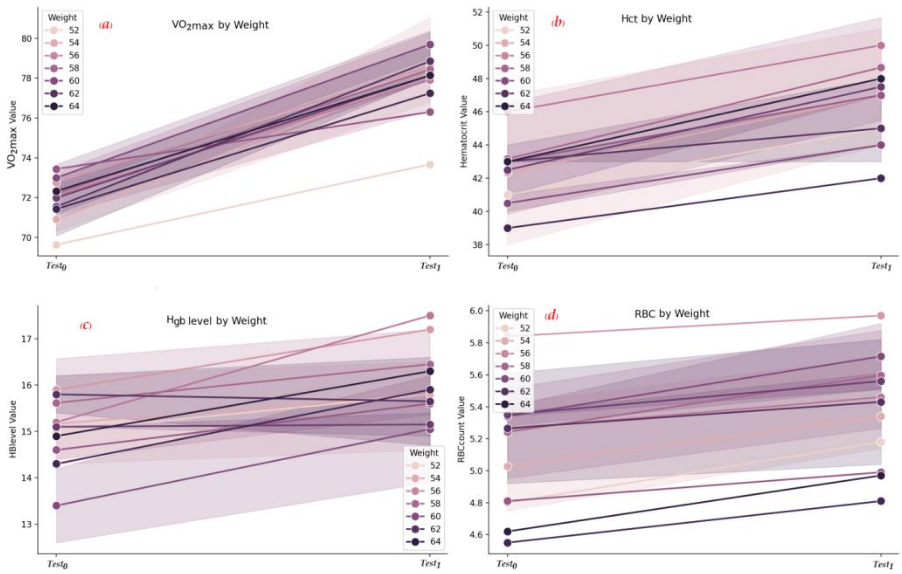

Figure 1. Line chart to show the *test₀*- *test₁* changes for key parameters across different subject weights; "lines highlight the trends in" VO_2 max" (a), "Hct" (b), "Hgb" (c), and "RBC" (d) by Weight at $test_0$ and $test_1$ intervals.

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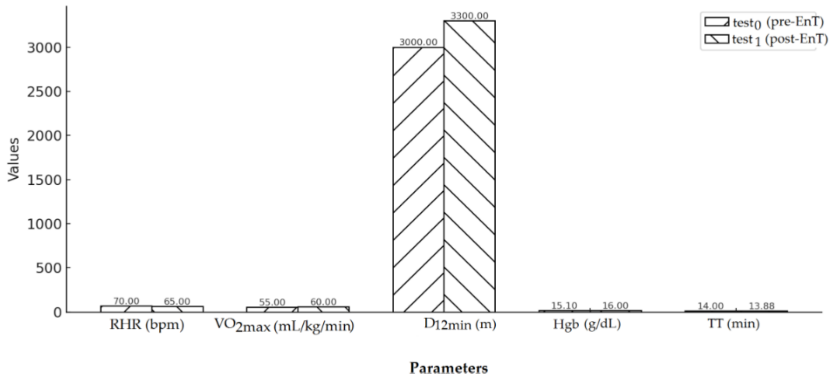

Figure 2. Graph to show analysis result of performance over all improvement from $test_0$ to $test_1$ (intervention)

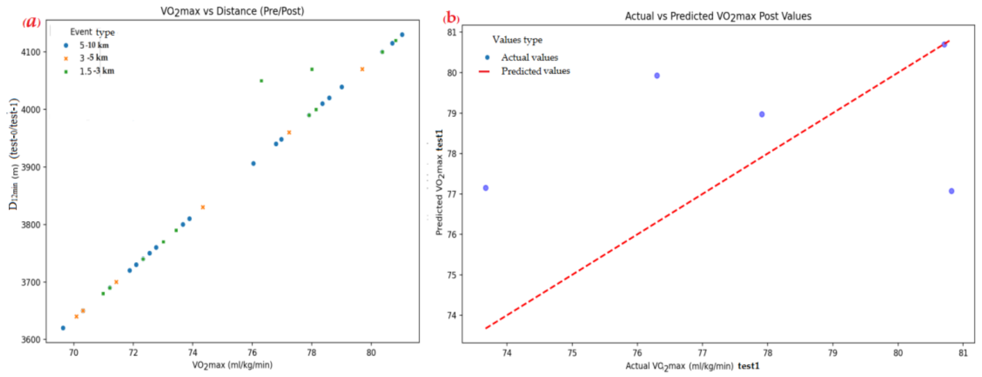

Figure 3. (a).VO_{2max} trend chart to explain VO_{2max} by D_{12min} correlation (r = 0.92) across event; and (b) actual with predicted VO_{2max} scatter plot to show actual and predicted VO_{2max} $test_1$ values with regression line in red color. Each blue dot represents an individual data point comparing actual and predicted values. The red dashed line is the line of equality, where predicted values match actual values exactly.

first principal component (PC1). RHR negatively loaded on PC2, while subject weight exhibited a

Key contributors to variance included $test_0$ VO_{2max} and D_{12min}, which exhibited strong loadings on the

Physiological Adaptations of Endurance Training

The results regarding RHR and VO_{2max} reflect improved cardiovascular efficiency, enhanced aerobic capacity, and increased autonomic control over heart function, alongside augmented endurance potential. The significant increase in VO_{2max} and the reduction in RHR are consistent with expected outcomes of EnT (Prommer et al., 2008; Steiner et al., 2019). However, the extent of VO_{2max} improvement observed in this study is particularly noteworthy, potentially attributable to the unique physiological characteristics of Ethiopian MLDA (Tilahun Muche et al., 2021). The reduction in RHR indicates enhanced cardiovascular efficiency, while the increase in VO_{2max} signifies improved oxygen utilization during HIIT efforts (Mancera-Soto et al., 2022). Research involving European and North American MLDA reports lower baseline VO_{2max} levels and smaller increments $test_1$, even under rigorous training conditions. This discrepancy may be linked to genetic factors, as studies by Larsen have highlighted that East African MLDA, particularly those of Ethiopian and Kenyan descent, possess unique physiological traits, such as a higher percentage of slow-twitch muscle fibers and an efficient running economy, which may predispose them to superior endurance performance (Larsen, 2003).

The physiological improvements observed in this study, including enhanced VO_{2max} and improved running performance, are consistent with adaptations described by Moxnes and Hausken (2012). These studies highlight that EnT leads to notable gains in cardiovascular efficiency and muscular adaptations, collectively enhancing overall performance (Figure 4).

Athletic Performance Enhancement

The findings from the ST suggest that while overall endurance has improved, enhancements in speed may require a more specialized training approach. The lack of significant improvement in ST indicates that the training program may need to incorporate more speed-focused elements to enhance short-distance performance (Stankovic et al., 2023). This highlights the importance of a balanced training approach that addresses both endurance and speed. Moreover, the study suggests that performance metrics related to endurance are paramount in predicting race outcomes, indicating a potential need for targeted speed training. This result aligns with findings from Chen et al. (2023) and Mujika et al., (2024), who reported that altitude training and HIIT improve both HemA and performance metrics (Stankovic et al., 2023). This suggests that the positive loading. Age demonstrated a notable negative loading on PC3. These findings underscore the significant roles of aerobic capacity and demographic factors in performance outcomes

DISCUSSION

Post-Endurance Training Hematological Adaptations

The findings indicate that EnT effectively enhanced erythropoiesis among elite male MLDA, consistent with previous studies that reported increased RBC counts as an adaptation to elevated oxygen demands during exercise (Sheykhlouvand et al., 2018; Tilahun Muche et al., 2021). The observed increases in RBC count, Hgb mass, and Htc align with existing literature indicating that EnT enhances the oxygen-carrying capacity of the blood (Sheykhlouvand et al., 2018). The modest change in RBC count suggests that while MLDA maintained overall health, there was no significant alteration in immune response attributable to prolonged EnT (Mujika et al., 2024b). Furthermore, the minimal variability in WBC count, coupled with a strong correlation between $test_0$ and $test_1$ WBC measurements, supports the notion that WBC counts are less sensitive to EnT adaptations compared to RBC-related parameters (Billat, 2001; Mancera-Soto et al., 2022). Variations in the magnitude of these HemA can be attributed to factors such as altitude and training intensity (Mujika et al., 2024b). This study underscores the significant impact of EnT on physiological adaptations, particularly focusing on HemA and their implications for athletic performance.

The increase in Hgb mass supports the hypothesis that EnT improves the oxygen transport capacity of MLDA. The observed increment in Htc highlights training-induced adaptations that enhance blood viscosity and oxygen delivery mechanisms. The increase in Hgb and RBC volume corroborates previous research emphasizing the role of EnT in promoting erythropoiesis (Montero et al., 2017). Comparative studies emphasize that increased erythropoiesis contributes to improved oxygen delivery, which is crucial for sustaining prolonged exercise (Montero et al., 2017). Our findings also align with those of Corsetti et al. (2012), who reported similar adaptations in elite cyclists. The significant increases in Hgb mass and erythrocyte volume observed in our study suggest that EnT induces substantial HemA that enhance aerobic capacity. This is further supported by (Steiner et al. (2019), who noted that EnT promotes erythropoiesis and optimizes oxygen transport.

observed improvements in performance are not solely attributable to increased RBC mass but also to enhanced cardiovascular and muscular efficiency (Carroll, 2018). The improved results in D_{12min} further highlight the effectiveness of the training regimen in significantly enhancing endurance capacity.

The improvement in TT performance indicates a favorable impact of EnT on overall race performance (Figure 3). The observed enhancements in TT align with findings from other studies on elite EnT (Paquette et al., 2017).

The study also highlights the importance of HIIT in maximizing endurance performance. demonstrated that HIIT can lead to significant improvements in both aerobic and anaerobic capacities, which are crucial for endurance MLDA (Boullosa et al., 2020). Our findings support these conclusions, indicating that incorporating HIIT into training regimens can yield substantial performance benefits.

PRACTICAL ASPECTS

The results contribute to the growing body of literature on the physiological adaptations induced by ET, particularly in the context of elite endurance MLDA. The insights gained from this research not only enhance our understanding of the mechanisms underlying athletic performance but also provide practical implications for coaches and MLDA seeking to optimize training strategies. Future research should continue to explore the interplay between various training modalities and performance outcomes, with a focus on personalized approaches that maximize both endurance and speed while ensuring the long-term health and well-being of MLDA

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