Natural Resources for Human Health

eISSN: 2583-1194

Original Research

Received 09 May 2026
Revised 20 June 2026
Accepted 01 July 2026

Natural Resources for Human Health 2026, Vol. 6 (11s): 1543–1553

KEYWORDS:
Lactate Threshold-guided training; Endurance performance; Lactate threshold; Distance runners; Physiological adaptations.

Lactate Threshold-Guided Training for Physiological Adaptations and Endurance Performance in Competitive Distance Runners

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ABSTRACT: The lactate threshold is the maximum exercise intensity at which lactate production exceeds its clearance, causing lactate to accumulate in the bloodstream. This threshold is well recognized as a key determinant of endurance performance in long-distance runners. The main goal of this study was to evaluate the impact of six months of lactate threshold-based training on physiological variables and endurance performance in competitive long-distance runners. One hundred (57 males, 43 females) registered competitive long-distance runners were randomly assigned to either a control group (mean age 24.16 ± 3.06 years) or an EG (mean age 24.52 ± 3.26 years). One participant withdrew from the CG during the follow-up period; therefore, 99 subjects were included in the six-month outcome analysis. Measurements were obtained before and after six months of training. All training sessions included a 10-minute warm-up and cool-down period at the beginning and end of each session. Following six months of training, statistically significant differences were observed in the EG compared with the CG for the following outcome variables: Lactate Threshold Stage (3.78 vs. 3.15), Mean Heart Rate at LT (160.11 vs. 157.07 bpm), 5000 m Running Performance (18:09.7 vs. 18:59.0 min), and Running Pace (4.588 vs. 4.389 m/s), with all variables demonstrating significance at $p < 0.005$. Improvements in endurance performance and physiological adaptations following lactate threshold (LT)-guided training were demonstrated in distance runners after six months. The findings of the study suggest that LT-guided training can improve endurance-related outcomes and should be used with competitive distance runners as an effective method for achieving these results. Future research should explore other training protocols and intervention durations that may influence endurance performance across different athletic populations.

1. INTRODUCTION

In the late 19th century when there were organized competitions for outdoor track and field, there was a focus on physiological research into athletic performance. This pursuit of better athletic performances at local, regional, national, and international levels has fostered a substantial volume of research into the mechanisms that modify human physiology during exercise [1]. Within the athletic environment, the competitive distance runner must excel not only in endurance through physical activity, but also in psychological ability (i.e., mental toughness), effective pacing strategies, and sustaining a high level of energy metabolism.

The distances for distance-running events vary from 5000-meter races to the marathon (42.195 km). Distance-running events are primarily endurance activities that require runners to perform prolonged exercise while delaying physical fatigue [2]. As training methods designed specifically to improve performance in endurance events attract increased interest, studies based on lactate threshold (LT) suggest that physiologic changes resulting from this type of training can positively affect endurance through increased running efficiency and reduced fatigue during prolonged physical activity [3].

If less than adequate oxygen is available through aerobic pathways to support the required movements when performing an activity (i.e., through aerobic metabolism), the body will rely more heavily upon anaerobic sources to generate energy for movement. Prolonged reliance on anaerobic energy sources uses glucose more quickly than aerobic pathways and therefore increases lactate production. While lactate is frequently described as simply a waste product produced as a result of anaerobic metabolism, it also has significance as an indicator of energy-source utilization and therefore relates to the type and duration of exercise bouts, including marathon running; lactate can also be used as an additional energy source during periods of high-intensity exercise [4].

With increasing exercise intensity, lactate production may exceed the body's ability to clear or utilize it, resulting in its accumulation in the bloodstream. During high-intensity exercise, anaerobic metabolism provides energy at a faster rate than aerobic metabolism. Increased glycolytic activity is associated by the collection of hydrogen ions, leading to decreased muscle pH. Severe muscle acidosis can impair the muscle's ability to produce force and power and thereby contribute to fatigue, although exercise-induced fatigue is multifactorial [5].

Eventually, during incremental exercise, lactate production may exceed the rate at which it can be cleared from the blood. This is referred to as the LT, which is the point at which lactate production exceeds lactate clearance from the bloodstream, and it serves as an estimate of one's endurance performance capacity [6]. A significant reduction in blood pH while exercising can contribute to fatigue and performance decline [5]. Lactate-threshold training programs are designed to allow individuals to sustain higher levels of intensity during physical activity by increasing their ability to produce, utilize, and clear lactate from the body [7, 3].

An individual's LT is a measure of how physical demands during exercise correlate with how well the body has adapted to those demands. This has made LT testing useful for coaches and exercise scientists in evaluating performance and designing training programs. Lactate measures are also used by strength and conditioning trainers to help determine an athlete's training readiness and to develop appropriate endurance training programs [8]. The maximum lactate steady state (MLSS) is a term that defines the highest exercise intensity at which an athlete can maintain a stable blood lactate level while performing continuous endurance exercise [9].

LT, Maximal Oxygen Uptake (VO_2max), and Running Economy as Influencers of Endurance Performance. The physiological components that influence the success of an athlete competing in an endurance-based sport are extensive. Three components are known as the "big three" and include VO_2max, LT and running economy. Of the three components, LT is often seen as an important predictor of an athlete's endurance capabilities because it provides a quantitative measure of how well the athlete can continue to perform at higher intensities for a prolonged period while participating in an exercise bout [10,11]. Many researchers have proposed different definitions of what constitutes LT and various models for determining where the LT occurs. Although there are multiple definitions and models of LT, the literature indicates that the application and interpretation of these definitions/models vary according to the testing protocols, characteristics of the athlete, and athletic context [6,12,13].

This may partially explain the lack of available long-term data on the effect of structured training programs incorporating the LT on the physiological adaptations of competitive distance runners. Therefore, further research is needed to develop optimal endurance training strategies.

The association between endurance training and the LT has been widely studied, but due to limited research on the physiological adaptations resulting from long-term training programs for competitive distance runners based on the LT. A six-month training period is sufficient to demonstrate significant changes in both physiological characteristics and performance-related outcomes. The LT is particularly important when evaluating training effects because it is widely recognized as a reliable measure of endurance fitness and a marker of training adaptation. There is a growing body of research reporting that LT and metabolic transition points are significant predictors of endurance performance across various athletic populations [13].

As different physiological variables can be evaluated, the LT appears to be particularly sensitive to endurance training and may provide important information regarding endurance-running performance [14,11]. Recent findings have resulted in a shift in the understanding of exercise metabolism. Lactate is now recognized not only as a major fuel source for muscle activity but also as an important contributor to energy production during aerobic metabolism, with substantial amounts of lactate being produced during aerobic exercise. Through oxidative metabolism, lactate can provide energy to muscles and is produced continuously regardless of the type of exercise being performed (i.e., anaerobic or aerobic) [4]. Inefficiencies in lactate removal, greater recruitment of fast-twitch muscle fibers (Type II), and disturbances in the balance between oxidative metabolism via the mitochondria and anaerobic glycolytic metabolism can contribute to impaired endurance performance [14]. In addition, hypoxia or a reduction in blood supply can further limit exercise performance by reducing oxygen availability at the tissue level, thereby impairing endurance performance [15].

The knowledge gained from understanding the effects of continuous LT-guided training should contribute to a better understanding of how to optimize endurance training programs and improve competitive performance in distance runners.

2. MATERIALS AND METHODS

2.1 Study Design and Participants

The prespecified primary outcome was 5000-m completion time at baseline and six months. Secondary outcomes were lactate-threshold stage and mean heart rate at LT at the same time points. Running pace, derived from completion time, was not treated as an independent outcome, $d = 0.53$. An a priori sample-size calculation was performed using G*Power for a one-tailed independent-samples comparison, with the prespecified Cohen's effect size, 80% power, α = 0.05, and 1:1 allocation. The required sample was 90 (45 per group); allowing approximately 10% attrition, 100 participants were recruited. Participants were recruited from 15 March to 20 April 2025 and began the intervention on staggered dates between 5 May and 13 August 2025. Baseline and follow-up assessments were conducted immediately before and after each participant's six-month programme. The overall study period ended on 13 February 2026.

A total of 100 endurance-trained competitive distance runners were enrolled in this randomized controlled study, with 50 participants assigned to the experimental group (EG) and 50 to the control group (CG). Prior to the intervention, baseline demographic and physiological characteristics, including age, height (cm), body weight (kg), body mass index (BMI), weekly training volume (practice hours), and resting respiratory rate were recorded for all participants (Table 1). Baseline assessments were conducted to determine the comparability of the two groups and to establish pre-intervention equivalence for subsequent outcome comparisons.

Participants were eligible for inclusion if they were male or female competitive distance runners aged 18–30 years, had a normal BMI (18.5–24.9 kg/m²), completed a minimum of 2 hours of training per day, and had participated in 5000-m running events at the district level. Participants were also required to follow the dietary requirements prescribed by the dietician.

Table 1: Baseline Demographic and Physiological Measures

	Group	n	Mean	S.D.	t	P value
Age (yrs)	Experimental	50	24.52	3.261	0.570	0.570
	Control	50	24.16	3.062		
Height (cms)	Experimental	50	172.48	3.325	0.116	0.908
	Control	50	172.38	5.138		
Weight (kgs)	Experimental	50	68.24	3.569	0.387	0.700
	Control	50	67.94	4.118		
BMI	Experimental	50	22.45	1.148	0.629	0.531
	Control	50	22.30	1.233		
Resting RR (per min)	Experimental	50	15.16	2.120	0.161	0.872
	Control	50	15.09	2.204		
Practice Hours (hr/day)	Experimental	50	2.23	0.471	1.559	0.122
	Control	50	2.42	0.726		

*p < 0.05 (Significant)

Participants were excluded if they had a recent musculoskeletal injury, trauma or surgery; a neurological condition affecting functional activity; pregnancy; hypertension, diabetes mellitus, cardiovascular disease, or other conditions requiring long-term medication; or if they used ergogenic aids.

Figure 1 shows the age distribution of male and female subjects involved in the study. Subjects were recruited from training facilities in Navi Mumbai. Before enrolment, all participants received information about the study and provided written informed consent in accordance with ethical research guidelines.

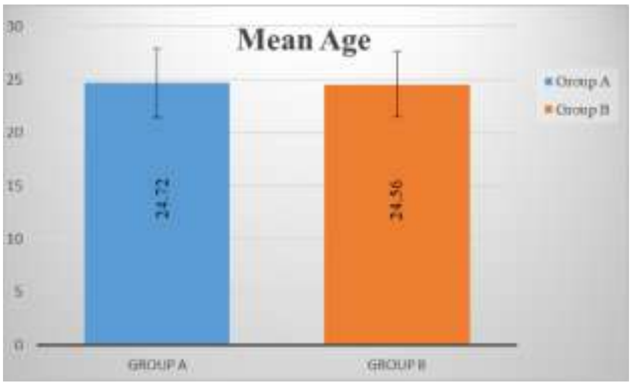

Figure 1: Mean age Comparison between groups

2.2 Randomization, Allocation Concealment, and Masking

Participants were allocated 1:1 using an unblocked, unstratified computer-generated sequence created with randomizer software by an assistant. An assistant prepared sequentially numbered, opaque, sealed envelopes. The researcher enrolled participants and opened the envelopes after baseline assessment. Due to the nature of the intervention, participants and training personnel could not be blinded. However, outcome assessors were blinded, and participants were instructed not to reveal their allocation. The statistical analyst used coded group labels and remained blinded until completion of the primary analysis.

2.3 Lactate-Threshold Assessment

LT was assessed on an AFTON computerized treadmill using the Bruce Protocol [16], with the stage-wise speed and grade presented in Table 2. After each stage, one fingertip capillary sample was collected during a 60-second interval and analysed using a StatStrip Xpress 2 lactate analyser (Nova Biomedical, USA). The first stage at which lactate reached ≥4 mmol/L was recorded as the LT stage [17,18], although this criterion may not represent each runner's maximal lactate steady state. Mean heart rate was monitored using a Polar H9 sensor.

The analyser was used according to the manufacturer's instructions, with strip expiry and storage checked. A researcher collected one sample per stage using a standardized procedure; duplicate testing and formal intra- or inter-rater reliability analyses were not performed. Nutritional intake was monitored using a dietician-supervised plan [19].

Table 2: Stage-wise Lactate Threshold Assessment Using the Bruce Protocol with a 3-Minute Duration per Stage[17]

Stage	Speed (mph/kmph)	Grade (Inclination)
1	1.7/2.7	10 %
2	2.5/4.0	12 %
3	3.4/5.5	14 %
4	4.2/6.8	16 %
5	5.0/8.0	18 %
6	5.5/8.9	20 %
7	6.0/9.7	22 %

Interpretation: A 60-second recovery interval was provided between successive exercise stages to allow for blood sample collection and mean heart rate measurement. Blood lactate level was assessed at the end of each stage, and the LT was considered to have been reached when the blood lactate level was equal to or greater than 36 mg/dL (4 mmol/L).

2.4 Training Intervention and Control Condition

After baseline LT assessment, the EG completed a six-month individualized programme based on LT-guided training principles [20]. Participants attended one supervised session weekly: during months 1–3 at their LT heart rate (±5 bpm), and during months 4–6 at 5% above that heart rate.

Each session comprised a 10-minute warm-up, an individualized workout, and a 10-minute cool-down. Workload was adjusted to individual capacity, and heart rate was monitored using Polar H9. A researcher administered all sessions using standardized procedures. Attendance and heart-rate responses were recorded, but no prospective adherence threshold or detailed fidelity measures were specified.

The CG continued its usual endurance training without receiving lactate-threshold-guided training. Detailed frequency, duration, and intensity data for the CG's usual training were not systematically recorded. Outcome assessments were repeated after six months using the same baseline procedures.

One CG participant discontinued for personal reasons before the six-month assessment. All 100 randomized participants remained in the intention-to-treat analysis, with available baseline data retained for this participant. A complete-case sensitivity analysis included 99 participants (EG = 50; CG = 49). Figure 2 presents participant flow.

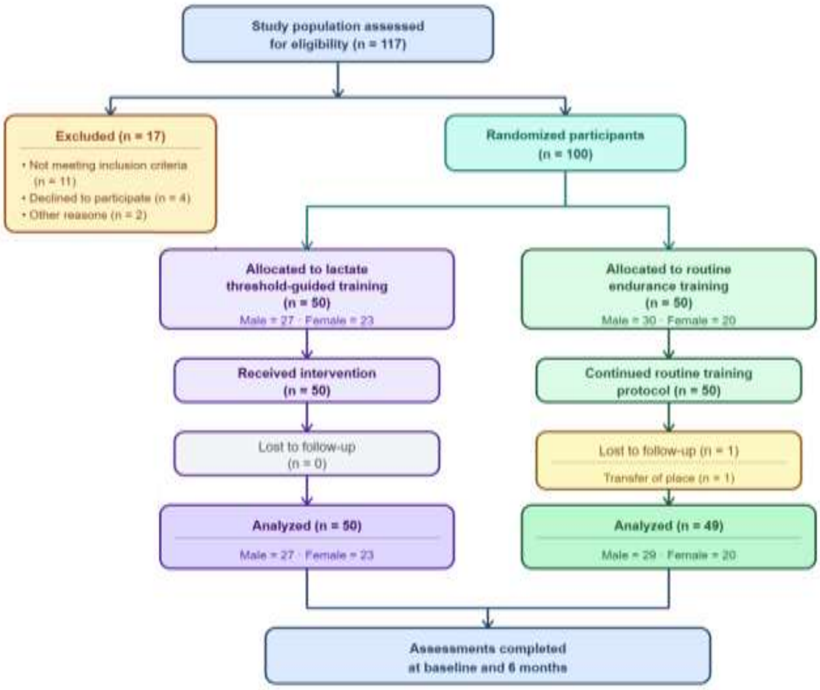

Figure 2: Study Enrollment and Allocation Flow

2.5 Statistical Analysis

Analyses were performed using IBM SPSS Statistics Version 26. The primary estimand was the between-group difference in change in 5000-m completion time from baseline to six months; corresponding changes were estimated for secondary outcomes. Data are reported as mean ± standard deviation (SD).

All outcomes, including LT stage, were analysed using mixed repeated-measures ANOVA, with group as the between-subject factor and time as the within-subject factor. The group-by-time interaction tested between-group differences in change.

Sphericity was not applicable with two time points. Complete-case sensitivity analysis assessed the effect of the missing follow-up. Statistical significance was set at $p < 0.05$, with effect sizes also considered to assess the magnitude of treatment effects.

3. RESULTS AND DISCUSSION

3.1 Baseline Characteristics and Study Outcomes

All 100 randomized participants were included in the primary analysis according to their assigned groups. Six-month outcome data were available for 50 participants in the EG and 49 participants in the CG. One control participant had missing follow-up data. The complete-case sensitivity analysis produced conclusions consistent with those of the primary analysis.

Baseline evaluation showed no statistically significant differences between the EG and the CG with respect to demographic characteristics, physiological measures, and performance-related variables ($p > 0.05$), confirming that the two groups were comparable before commencement of the intervention. At the six-month follow-up assessment, complete data were available for 99 participants, as one individual from the CG withdrew from the study during the intervention period (experimental = 50; control = 49).

Following completion of the training program, participants in the EG exhibited greater improvements in LT stage, mean heart rate, 5000 m running performance, and running pace compared with the CG. The average time required to complete the 5000 m run decreased by 1 minute and 23 seconds in the EG, whereas the corresponding improvement in the CG was approximately 35 seconds. Furthermore, the mean running velocity increased from 4.26 m/s before to 4.588 m/s at six months post-training, and the second measure of performance improved from 2.85 to 3.78. For further detail see Tables 3 and 4.

Table 3: Baseline Assessment of Study Parameters

Outcome	Group	N	Mean	S. D.	t	P value
LT Stage/Level (1/2/3/4/5/6/7)	Experimental	50	2.85	0.43	0.528	0.599
	Control	50	2.89	0.32		
Mean HR	Experimental	50	154.49	3.48	0.033	0.974
	Control	50	154.51	2.56		
Time Taken to 5000m(in secs)	Experimental	50	1172.67	74.03	0.085	0.932
	Control	50	1173.91	71.65		
Time Taken to 5000 m (hh:mm:ss.000)	Experimental	50	19:32.7	01:14.0	0.085	0.932
	Control	50	19:33.9	01:11.7		
Pace (m/s)	Experimental	50	4.26	0.27	0.189	0.851
	Control	50	4.27	0.26		

Table 4: Assessment of Study Variables After 6 Months

Outcome	Group	N	Mean	S. D.	t	P value
LT Stage/Level (1/2/3/4/5/6/7)	Experimental	50	3.78	0.371	8.356	0.001
	Control	49	3.15	0.379		
Mean HR	Experimental	50	160.11	2.471	5.192	0.001
	Control	49	157.07	3.289		
	Experimental	50	1089.68	46.654	6.042	0.001

Time Taken to 5000m(in secs)	Control	49	1139.04	33.724		
Time Taken to 5000 m (hh:mm:ss.000)	Experimental	50	18:09.7	00:46.7	6.042	0.001
	Control	49	18:59.0	00:33.7		
Pace (m/s)	Experimental	50	4.588	0.1963	6.669	0.001
	Control	49	4.389	0.1504		

3.2 Mixed Repeated-Measures Analysis

The results of the mixed repeated-measures ANOVA revealed that all outcome variables were significantly affected by the group-by-time interaction (Table 5). Effect sizes were large for LT stage ($\eta^2p = 0.263$), mean heart rate ($\eta^2p = 0.320$), completion time for the 5000 m distance ($\eta^2p = 0.160$), and running pace ($\eta^2p = 0.201$), indicating substantial physiological and endurance performance adaptations following six months of LT-guided training. Furthermore, the findings revealed that a considerable proportion of the variance in endurance performance outcomes could be attributed to the training program, as reflected by the partial eta squared values.

Table 5. Mixed Repeated Measures ANOVA Results

Outcome	**F(1,97)**	**p value**	**η^2p**
LT Stage	34.53	0.001	0.263
Mean HR	45.66	0.001	0.320
Time Taken to 5000 m	18.48	0.001	0.160
Pace	24.39	0.001	0.201

3.4 Discussion

The findings showed that six months of LT-guided training produced considerable improvements in physiological and performance variables among competitive distance runners. The EG showed favourable changes in lactate-threshold stage, running pace, mean heart rate, and 5000-m performance, indicating improvements in the physiological and performance outcomes measured in this study. These findings support previous studies demonstrating that training based on LT leads to improved performance and endurance capacity [11, 20]. In addition, several studies support the use of LT when prescribing exercise for endurance adaptations and evaluating the performance of long-distance runners [20].

The observed improvements in the EG occurred over a period of six months, which is noteworthy considering the limited research currently available on this type of training. The improvements in the measured outcomes may be related to physiological mechanisms commonly associated with endurance training, including more efficient lactate transport and clearance, increased peripheral circulation, and greater mitochondrial density within skeletal muscle tissue. However, because these mechanisms and running economy were not directly measured, they represent possible explanations rather than confirmed adaptations. Increased metabolic efficiency and the ability to delay the accumulation of fatigue-related metabolites may allow a higher running intensity to be maintained before rapid lactate accumulation occurs. Improved buffering capacity may also regulate hydrogen-ion concentration in the muscles, reduce the effects of exercise-induced acidosis, and facilitate endurance performance [5,13]. These possible mechanisms were not directly assessed in the present investigation.

Exercise performed around or above the LT has been identified as an important component of endurance training because of its close relationship with distance-running performance. The greater the percentage of an individual's VO_2max that can be sustained for an extended period, the longer the onset of metabolic acidosis may be delayed and the greater the ability to maintain performance during high-intensity exercise. Therefore, LT has been identified as a practical measure of endurance capacity when compared with VO_2max alone [4,13]. Improvements in the regulation, transport, and utilization of lactate have been proposed as possible mechanisms through which lactate-threshold training may improve anaerobic and aerobic performance [21]. However, VO_2max and lactate transport and utilization were not directly measured in this study.

Endurance exercise may increase capillary density, blood volume, and skeletal-muscle mitochondrial characteristics, supporting oxygen delivery and energy production. Similar adaptations have been reported after concurrent strength and endurance training [22], but these mechanisms were not measured here. The findings agree with studies reporting improved endurance outcomes and supporting LT as an indicator of running performance [23,24]. Proposed mechanisms include mitochondrial biogenesis, oxidative-enzyme activity, lactate clearance, and stroke volume [25,26]; these remain hypothetical explanations.

Physiology plays an important role in athletic performance. Factors contributing to athletic performance include muscle-fibre characteristics, VO_2max, running economy, and the ability to sustain a high percentage of VO_2max during competition [27]. Many studies have demonstrated a strong relationship between VO_2max utilization and performance in distance events ranging from 5-km races to longer-distance competitions [28,29]. Another adaptation associated with endurance training is an improved ability of skeletal muscle to utilize and remove lactate from the bloodstream. Improvements in the body's ability to transport and utilize lactate may enhance its capacity to produce energy and sustain performance at higher exercise intensities [30]. Previous research has associated lower blood lactate concentrations at a given workload with increased mitochondrial volume, mitochondrial density, and oxidative-enzyme activity [23]. Nevertheless, VO_2max, muscle-fibre characteristics, mitochondrial adaptations, oxidative-enzyme activity, and lactate utilization were not measured in this study.

Increased energy production through the oxidative energy system and decreased reliance on anaerobic energy production may reduce blood lactate concentrations during exercise at the same relative intensity. Physiological adaptations reported in previous endurance-training research may provide possible explanations for the performance improvements observed in this study; however, these mechanisms were not directly assessed. The six-month duration of the intervention allowed changes in lactate-threshold stage and performance outcomes to be evaluated over an extended training period. However, because the study did not directly compare programmes of different durations, it cannot establish whether six months of training provides greater benefits than interventions lasting less than three months. The findings nevertheless support lactate-threshold-guided training as a potential method for improving endurance-related performance.

Distance runners place considerable physical demands on their cardiovascular and muscular systems. Athletes who compete in long-distance events commonly demonstrate physiological adaptations that may improve oxygen delivery and capillary density [31]. Such adaptations may improve cardiorespiratory fitness and endurance [32, 33]. Enhanced muscular blood flow, aerobic energy production, and recruitment of fatigue-resistant fibres may also support long-distance performance, although these mechanisms were not assessed.

The findings of this study support previous investigations that reported improvements in running pace and lactate-threshold-related measures following an endurance-training intervention. Blood lactate measurement has also been identified as a useful method for determining the exercise intensity at which blood lactate begins to accumulate and for predicting endurance performance [18]. Previous research has associated endurance training with muscle-fibre and oxidative adaptations that may contribute to endurance capacity [24]. Mitochondrial-enzyme activity, including succinate dehydrogenase activity, has been associated with aerobic performance and energy-production efficiency. Increased capillary density has also been associated with improved oxygen delivery and substrate utilization within working muscles. However, muscle-fibre composition, mitochondrial-enzyme activity, and capillary density were not assessed in this study.

Previous research suggests that these physiological adaptations may contribute to running economy and long-distance endurance performance. Running economy may be influenced by several factors, including stride mechanics, ground-contact characteristics, energy expenditure, and fatigue resistance [34]. Running economy is a key determinant of distance-running performance and is defined as the energy expended while maintaining a specified running speed. From a biomechanical perspective, running economy may be improved through efficient movement patterns that minimize unnecessary energy expenditure while optimizing the magnitude, direction, and timing of forces applied to the ground. Previous research has associated endurance training with adaptations in skeletal-muscle antioxidant-enzyme activity that may contribute to fatigue resistance and endurance performance [35, 36]. However, running economy, running biomechanics, muscle-fibre characteristics, and enzyme activity were not measured in this study.

High-intensity exercise may improve aerobic fitness and mitochondrial adaptation, while greater oxidative capacity can enhance submaximal endurance performance [37–39]. Training volume may also be associated with lower lactate and greater fat oxidation at comparable workloads [40], although these mechanisms were not evaluated. Participant and trainer blinding was infeasible; intervention fidelity and CG training exposure were not systematically documented; and outcomes may have been influenced by effort and testing conditions. One follow-up heart-rate value was missing. Because participants were competitive runners from a single Navi Mumbai setting, generalizability is limited. Although both male and female runners were included, sex-specific responses to the intervention were not separately analysed. Differences in training history and external training load may also have influenced the outcomes. Future studies should replicate these findings across different running distances, performance levels, and athletic populations.

4. CONCLUSION

The results demonstrate that the six-month LTG Training Plan improved race performance in competitive distance runners compared with pre-training performance across several important physiological and performance variables, including 5000 m completion time, average running pace, average heart rate, and LT-related physiological adaptations. The findings of the present study reinforce the use of LT as a basis for prescribing exercise in endurance training programmes to improve performance. The results suggest the importance of developing an individualised training intensity profile for each athlete according to their unique physiological characteristics to maximise the benefits of training. Future research should seek to determine what effect individualised training programmes based on the use of LT are on multiple athlete populations and levels of competition and the possible long-term effects on physiological, psychological, and performance-related variables.

ACKNOWLEDGEMENTS

We thank all the participants enrolled in the study.

AUTHOR CONTRIBUTIONS

J. Ezhil Mathi: Research design, Data collection, Data analysis, Manuscript writing, Project development, Data interpretation, Data management, Quality control.

Ramesh Chandra Patra: Research design, Data collection, Data analysis, Blood biochemical measurements, Data interpretation, Manuscript revision, Project development, Data management, Quality control.

Kanav Khera: Blood biochemical measurements, Data collection, Project development, Data management, Quality control.

All authors: Manuscript editing and approval of the final version.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICS STATEMENT

This study was approved by the Institutional Research Committee of Dr. G. D. Pol Foundation's YMT College of Physiotherapy (Ethics Approval No. YMT/BPTh/165/25; approval date: 7 March 2025). Written informed consent was obtained from all participants before their participation in the study. All procedures were performed in accordance with the Declaration of Helsinki and other relevant ethical guidelines.

DATA AVAILABILITY STATEMENT

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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