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Lactate Profiling and the Agreement Among Various Lactate Threshold Methods in Professional and Youth Soccer Players

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Abstract: The lactate threshold (LT) and the associated running velocities are important markers used to define physical readiness and prescribe exercise intensity in athletes. This study examined blood LT during maximal cardiopulmonary exercise testing using four methods: visual inspection, log-to-log transformation, the Dmax method, and the 4 mmol/L fixed blood lactate accumulation (FBLA) method. The participants included 96 soccer players, comprising 52 professional (27.37 ± 5.67 years) and 44 elite youth players (16.20 ± 0.8 years). A total of 554 capillary blood lactate samples were analyzed. Bland–Altman and ICC analyses for running velocities, determined using the four LT detection methods, demonstrated poor agreement in both groups. Results indicated that the youth players had significantly ($p < 0.05$) higher $\dot{V}O_2$ max (59.89 ± 5.6 mL·kg^{-1}·min^{-1}) compared to the professional players (56.43 ± 4.81 mL·kg^{-1}·min^{-1}). However, the professional players had significantly better running performance and running economy. A two-way ANOVA revealed a main effect of playing standard, with professional players exhibiting significantly higher 4 mmol/L FBLA LT compared to youth players. A mixed-design ANOVA indicated a significant ($p < 0.01$) interaction, with the youth exhibiting higher lactate accumulation only after completing the 18 Km/h stage. Therefore, youth and professional players should not use the different LT concepts interchangeably. Additionally, the 4 mmol/L FBLA LT method appears to be more robust for youth soccer players.

Keywords: Dmax; football; fixed blood lactate accumulation

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1. Introduction

L-lactate, contrary to some old beliefs, is considered an important energy source for many tissues of the human body, such as the heart [1] and the brain [2]. This is due to the Monocardoxylalate Transporters (MCTs) [3] that allow L-lactate to be absorbed by the cells of various tissues. Additionally, the metabolic process of L-lactate formation from the breakdown of glucose is an essential step in anaerobic metabolism and is mandatory for glycolysis sustainability [4]. L-lactate formation also occurs under aerobic conditions, but not due to mitochondrial hypoxia. In fact, mitochondrial activity is very high during maximal aerobic exercise, and L-lactate accumulation and, presumably, formation are high [5–7]. Under conditions in which the muscle metabolic rate increases, the glycolytic rate also increases, resulting in enhanced L-lactate release and extraction by the active muscles. While L-lactate release by the active muscles increases throughout the exercise phase [8], plasma L-lactate concentrations rise only during high-intensity exercise [2,5]. During continued exercise with gradual increases in intensity, L-lactate will start building up in a curvilinear manner [9]. Soccer demands superior levels of physical fitness [10],

and the aforementioned metabolic processes are favored during high-level professional matches [11]. In addition, player physicality is constantly evolving [12], and varying playing positions impose distinct metabolic demands on players [11]. Thus, particular interest is given to determining the inflection point at which L-lactate starts to build up more rapidly, referred to by many as the lactate threshold (LT) [13].

The particular breakpoint for the commencement of lactate accumulation has been the focus of interest for many researchers, and several methods have been used to determine and define it. Furthermore, exercise intensity at the LT inflection point has become the most important determinant of aerobic fitness [14,15] and is often used by coaches to evaluate athletes' training status or to establish training intensities. Additionally, running velocity at LT (vLT) and running economy (RE) are more sensitive than maximal oxygen consumption ($\dot{V}O_2$ max) in predicting aerobic performance [16] and are also indicative of soccer players' endurance characteristics and their playing standards [17]. The most commonly used methods for detecting LT are visual inspection (VI) [18] and log-to-log transformation [19]. The VI method defines the LT as the workload needed to cause lactate to increase exponentially, whereas the log-to-log transformation technique involves fitting two regression lines of best fit on the curvilinear graph, with the point of intersection denoted as the LT. Another method proposed was Dmax, where the lowest and highest lactate points on the curvilinear graph are connected with a linear line [20]. The longitudinal tangent on the curvilinear path placed at the furthest point between the straight line and the curvilinear curve is considered the LT. Other approaches have proposed a fixed value of lactate, defined as the fixed blood lactate accumulation (FBLA), with the most commonly accepted value being 4 mmol/L [21].

While lactate profiling in the laboratory is a common practice for professional soccer teams, youth players are mainly assessed using field-based tests. Field testing for aerobic power is associated with game performance [22] and has gained acceptance in evaluating aerobic fitness in youth soccer [23]. However, these tests have not been optimized [24] and lack sensitivity for predicting aerobic performance [16]. Moreover, comparisons and assessments of readiness to meet the playing standards of professional soccer players are difficult because lactate profiling is scarce in youth athletes. Thus, the purpose of this study was to profile the lactate levels of elite youth soccer players based on their playing position, compare these profiles to those of professional players, examine differences in LT using the four methods mentioned above, and identify which method is superior in predicting aerobic performance. Since the agreement between the various methods of LT determination is questionable among professional soccer players [25], an additional aim of the study was to examine the agreement among the four LT methods specifically for elite youth players. It is hypothesized that different LT determination methods cannot be used interchangeably in youth soccer players. Additionally, it is hypothesized that professional players will demonstrate superior performance compared to elite youth players, regardless of playing position.

2. Materials and Methods

This cross-sectional study assessed the hypotheses using professional soccer players (pro) from two Division 1 teams in the Eastern Mediterranean who participated in the qualifying rounds of the UEFA European Leagues. Their respective elite youth players (youth) under 17 years of age (U-17) were also included. The testing was performed at the beginning of the preseason period. The testing protocol for the professionals and youth was the same for comparison purposes. The aerobic power of the participants was evaluated using cardiopulmonary exercise testing (CPET) on a motorized treadmill. Lactate sampling was performed simultaneously during gas exchange analysis. This study adheres to the proposed recommendations to ensure valid results [26]. All testing

procedures took place from 8 am to 3 pm on consecutive days for both the youth and the professionals and at an ambient laboratory temperature of 20 °C to 23 °C. The research complied with the relevant national regulations, was conducted in accordance with the Declaration of Helsinki, and was approved by the National Committee of Bioethics (EEBK EP 2022.01.290).

2.1. Participants

A total of 96 soccer players volunteered for the study, comprising 52 professional players (27.37 ± 5.67 years of age) and 44 youth players (16.20 ± 0.8 years of age). Participation was voluntary, and the participants were informed about the benefits and risks of the investigation prior to signing an institutionally approved informed consent form. The respective team boards and parents of U-17 players also signed an informed consent form. The participants were instructed to undergo a retest at the same time as the initial testing to mitigate the potential circadian rhythm effects on the reliability measurements.

2.2. Procedures

Body composition measurements: A wall stadiometer (The Leicester® Height Measure, Tanita®, Tokyo, Japan) was employed to measure the stature of the footballers, and the Tanita scale and body fat analyzer (BC 418 MA, Tanita®,Tokyo, Japan) were used for their mass. Their body composition was analyzed using the 7-site skinfold method [27] with a skinfold caliper (Skyndex®, Greenwood, SC, USA).

Cardiopulmonary exercise testing (CPET): The laboratory temperature was controlled throughout all testing sessions at 20 ± 2 °C, and the relative humidity was approximately 50%. The testing protocol included 3 min running increments with a consistent incline of 1%, which is considered optimal for offsetting the absence of air resistance in the laboratory environment [28]. Specifically, a motorized treadmill (h/p/Cosmos® Quasar med, H-P-Cosmos Sports & Medical GmbH, Nussdorf-Traunstein, Germany) was programmed using the Cosmed Quark® CPET (Rome, Italy) system's software (Omnia Software 2.3, Rome, Italy) to start with a velocity of 8 km/h and speed up 2 km/h every three minutes and 15s. The 15s interval was the dead time that allowed subjects to pause the activity for lactate sampling purposes, thus ensuring that the activity for each increment lasted for three minutes. Breath-by-breath pulmonary gas exchanges were collected using reusable rubber masks (model 7940, Hans Rudolph, Kansas City, MO, USA) and analyzed using the Cosmed Quark® CPET metabolic analyzer (Rome, Italy). The heart rate (HR) was continuously monitored using a Polar H10 chest strap sensor (Polar® Electro Oy, Kempele, Finland). The breath-by-breath data analysis involved filtering the data using 10s averages to smooth them and accurately determine the $\dot{V}O_2$ max . The criterion for exercise cessation was volitional fatigue, and those for the $\dot{V}O_2$ max determination were (a) a plateau in $\dot{V}O_2$ max , (b) an accumulation of more than 8 mmol/L, and (c) a respiratory exchange ratio (RER) greater than 1.10 [29]. The exercise intensity that is appropriate for accurately assessing running economy (RE) is specified at speeds below 85% of the $\dot{V}O_2$ max [30]. Thus, similar to Ziogas et al. (2011) [17], we calculated RE using the average $\dot{V}O_2$ of the last 30 s while running at 12 km/h.

Lactate sampling: Lactate sampling utilized 28G, 1.8 mm side fire single-use safety lancets (Unistik® 3 Comfort, Owen Mumford Ltd., Woodstock, Oxfordshire, UK) to perform finger pricks, preferably on the 3rd and 4th digits according to the methodology described by previous investigators [31]. Blood lactate analysis was conducted using a validated [32] analyzer (Lactate Plus®, Nova Biomedical, Waltham, MA, USA) equipped with a disposable 0.7 μL lactate strip. Before each test, the lactate analyzer was calibrated using Lactate Plus® control solutions to ensure accurate and valid data. In total, 554 capillary blood lactate samples were collected. The blood lactate concentrations during

incremental testing were plotted against the running speed. The validated software "Lactate-E, v2.0" (Galway, Ireland) [33] was used to calculate each player's lactate thresholds using the VI [18], log-to-log transformation [19], Dmax [20], and 4 mmol/L FBLA [21] methods.

2.3. Statistical Analyses

The analysis was performed using IBM® SPSS® Statistics 28 (IBM Corporation, Chicago, IL, USA). The Shapiro–Wilk test was used to evaluate data normality. The independent *t*-test analysis was used to evaluate the differences in the anthropometric variables and performance variables of $\dot{V}O_2$ max, RE, and maximum running time between youth and pro athletes. Levene's test indicated no significant difference; thus, equal variances were assumed. A two-way analysis of variance (ANOVA) was used to evaluate the effects of playing position and playing standards (pro vs. youth) on the various LT methods and their interactions. A mixed-design ANOVA was used to examine the main effects of group (playing standard) on lactate accumulation, the main effect of time (lactate accumulation at each stage of the incremental testing), and the interaction between group and time. Athletes who surpassed the 18 km/h stage or terminated their performance earlier were excluded from this part of the analysis. Thus, only 36 pro and 25 youth athletes completed the 18 km/h stage and were subsequently included in the analysis.

Bland–Altman analysis. The limits of agreement between the different measurement methods of LT for youth and pro athletes were quantified using the Bland–Altman analysis ($M \pm 1.96\ SD$). Scatter plots were prepared by plotting the difference between each of the two LT methods on the *y*-axis and the average of the measurements on the *x*-axis [34]. The three horizontal lines represent the mean difference (bias) and the upper and lower limits of agreement (LOA), which are the mean bias ± (1.96 * SD). Similarly to Cerda-Kohler et al. (2016) [25], and for the same reasons [35], we used less than ± 1 km/h as the criterion for agreement between the LT methods. The agreement was also analyzed using Intraclass Correlation Coefficients (ICCs) with upper and lower 95% confidence intervals. Finally, we performed regression analyses to determine the variation observed in running performance, which was explained by the variation in LT determination methods. For all statistical analyses, significance was accepted at $p < 0.05$.

3. Results

The anthropometric measurements for the youth and pro soccer players are presented in Table 1. Independent *t*-tests demonstrated that the professional players were significantly taller [$t_{(94)} = 2.94$, $p < 0.05$, CI = 1.23–6.39] and heavier [$t_{(94)} = 7.76$, $p < 0.05$, CI = 7.55–12.74] than the youth players. Body composition analysis demonstrated that youth players had a similar body fat percentage (BF%) compared to professional players (Table 1). Furthermore, the analysis demonstrated that the youth players had a significantly higher $\dot{V}O_2$ max (59.89 ± 5.6 $mL \cdot kg^{-1} \cdot min^{-1}$) compared to the professional players (56.43 ± 4.81 $mL \cdot kg^{-1} \cdot min^{-1}$) [$t_{(94)} = -3.24$, $p < 0.05$, CI = −5.55–−1.33]. However, the professional players had significantly better running performance (18:08 ± 1.83 min) than the youth players (17:19 ± 1.86 min) [$t_{(94)} = 2.36$, $p < 0.05$, CI = 0.14–1.64]. Similarly, the professional players had significantly better RE (43.95 ± 3.59 $mL \cdot kg^{-1} \cdot min^{-1}$) than the youth players (46.86 ± 3.33 $mL \cdot kg^{-1} \cdot min^{-1}$) [$t_{(94)} = -2.92$, $p < 0.05$, CI = −4.33–−1.50]. The effect sizes measured by Cohen's *d* were $d = 0.66$ and $d = 0.48$, indicating medium effects for $\dot{V}O_2$ max and running performance, with a large effect for RE at $d = 0.86$. For the same purposes, the η^2 values for multivariate analysis ($\eta^2 \geq 0.01, \geq 0.06, \geq 0.14$) indicated small, medium, and large effects. A two-way ANOVA analysis indicated a main effect of playing standards (pro vs. youth) for the FBLA LT method, with the professional players having significantly higher FBLA LT than the youth (Table 2) [$F(1, 94) = 9.40$, $p < 0.01$, $\eta^2 = 0.10$)].

Table 1. The anthropometric characteristic differences between the pro and youth players.

	Playing Standards	n	M ± SD (km/h)
Age (years)	pro	52	27.37 * ± 5.67
	youth	44	16.20 ± 0.8
Height (cm)	pro	52	179.78 * ± 6.72
	youth	44	175.97 ± 5.84
Body Mass (kg)	pro	52	77.12 * ± 6.54
	youth	44	66.98 ± 6.19
Body Mass Index (BMI)	pro	52	23.83 * ± 1.26
	youth	44	21.59 ± 1.84
Body Fat Percentage (7-site)	pro	52	10.50 ± 3.83
	youth	44	9.24 ± 3.28

Note * = $p < 0.05$.

Table 2. The LT running velocity differences between the pro and youth players for each LT detection method.

Methods for LT Detection	Playing Standards	n	M ± SD (km/h)
Visual Inspection	pro	52	13.51 ± 1.33
	youth	44	13.45 ± 1.78
Log–Log	pro	52	12.32 ± 1.61
	youth	44	12.67 ± 1.83
Dmax	pro	52	13.55 ± 0.89
	youth	44	13.79 ± 1.01
FBLA (4 mmol/l)	pro	52	13.54 * ± 1.13
	youth	44	12.71 ± 1.63

Note * = $p < 0.05$.

The analysis indicated no main effect of playing position or an interaction between playing standards and playing position (Table 3). Furthermore, the analysis indicated no main effect of playing standards (pro vs. youth) and playing position and no interaction for the rest of the LT methods. For this part of the analysis, one youth athlete was removed from the group as coaches utilized the athlete in multiple playing positions.

Table 3. Positional Differences between pro and youth players based on the lactate threshold determination methods.

	Pro				Youth			
Playing Position	**n**	**km/h**	**95% CI**		**n**	**km/h**	**95% CI**	
Visual Inspection								
Defenders	8	14.00 ± 1.26	12.93	15.07	8	13.05 ± 1.04	11.98	14.12
Full Backs	13	13.54 ± 1.42	12.70	14.38	11	13.85 ± 1.58	12.94	14.76
Midfielders	11	13.80 ± 1.29	12.89	14.71	14	14.01 ± 1.85	13.21	14.82
Wingers	7	13.40 ± 0.61	12.26	14.54	6	13.43 ± 1.46	12.20	14.67
Forwards	13	13.00 ± 1.56	12.16	13.84	4	11.70 ± 2.84	10.19	13.21
Log–Log								
Defenders	8	12.10 ± 2.03	10.88	13.32	8	12.86 ± 0.93	11.64	14.08
Full Backs	13	11.95 ± 1.60	10.99	12.90	11	12.19 ± 1.82	11.15	13.23
Midfielders	11	12.83 ± 1.01	11.79	13.87	14	13.27 ± 1.96	12.35	14.19
Wingers	7	12.79 ± 0.71	11.48	14.09	6	11.82 ± 2.40	10.41	13.22
Forwards	13	12.14 ± 2.09	11.18	13.09	4	12.45 ± 2.00	10.73	14.17

DMax								
Defenders	8	13.70 ± 0.82	13.07	14.33	8	13.44 ± 0.98	12.80	14.07
Full Backs	13	13.48 ± 0.72	12.99	13.98	11	13.46 ± 0.67	12.92	14.00
Midfielders	11	14.10 ± 1.24	13.56	14.64	14	13.81 ± 0.85	13.34	14.29
Wingers	7	13.14 ± 0.43	12.47	13.82	6	13.85 ± 1.09	13.12	14.58
Forwards	13	13.28 ± 0.81	12.78	13.77	4	14.68 ± 1.56	13.78	15.57
FBLA-4 mmol/L								
Defenders	8	13.55 ± 0.85	12.55	14.55	8	12.73 ± 1.11	11.73	13.72
Full Backs	13	13.27 ± 1.52	12.49	14.05	11	13.17 ± 1.10	12.32	14.02
Midfielders	11	13.65 ± 1.06	12.80	14.49	14	12.79 ± 2.08	12.03	13.54
Wingers	7	13.81 ± 1.11	12.75	14.88	6	12.17 ± 1.24	11.02	13.32
Forwards	13	13.58 ± 0.99	12.80	14.37	4	12.23 ± 2.75	10.82	13.63

For the mixed-design ANOVA, we used the Greenhouse–Geisser analysis, as Mauchly's test of sphericity was significant. The results indicated an expected significant within-subject effect [$F_{(2, 125)} = 731.98$, $p < 0.01$, $\eta^2 = 0.93$] since, for both groups, it was anticipated that lactate accumulation would increase significantly over time due to increasing increment intensity. However, the results also indicate a significant interaction [$F_{(2, 125)} = 3.25$, $p < 0.01$, $\eta^2 = 0.05$]. Post hoc analysis between the pro and youth group effects indicated a significant difference in lactate accumulation only after completing the last stage of 18 km/h, with the youth participants demonstrating significantly higher values (Figure 1).

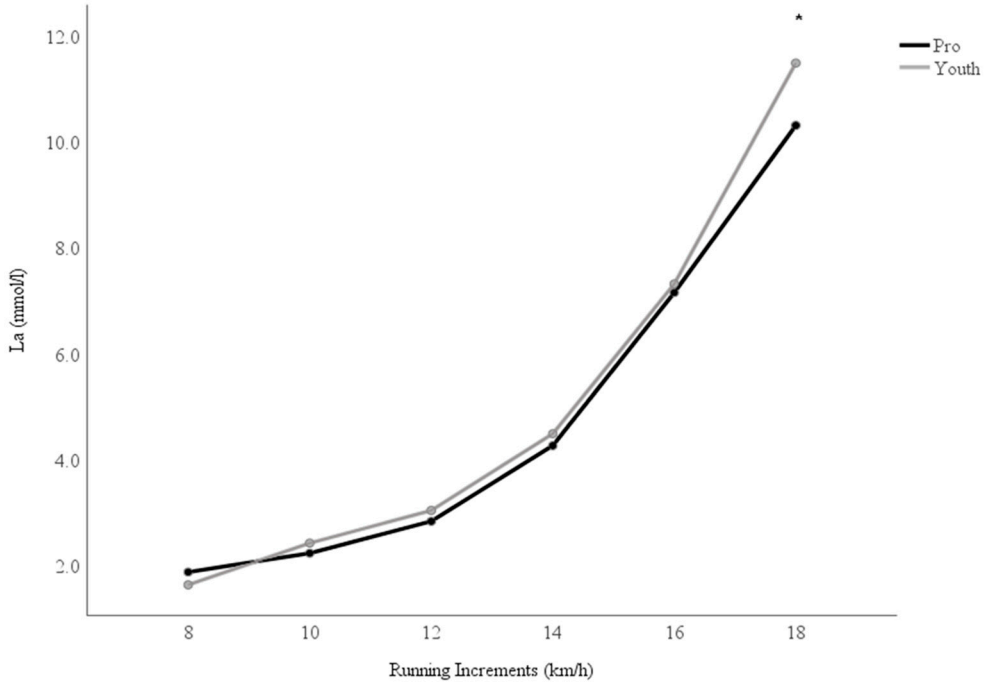
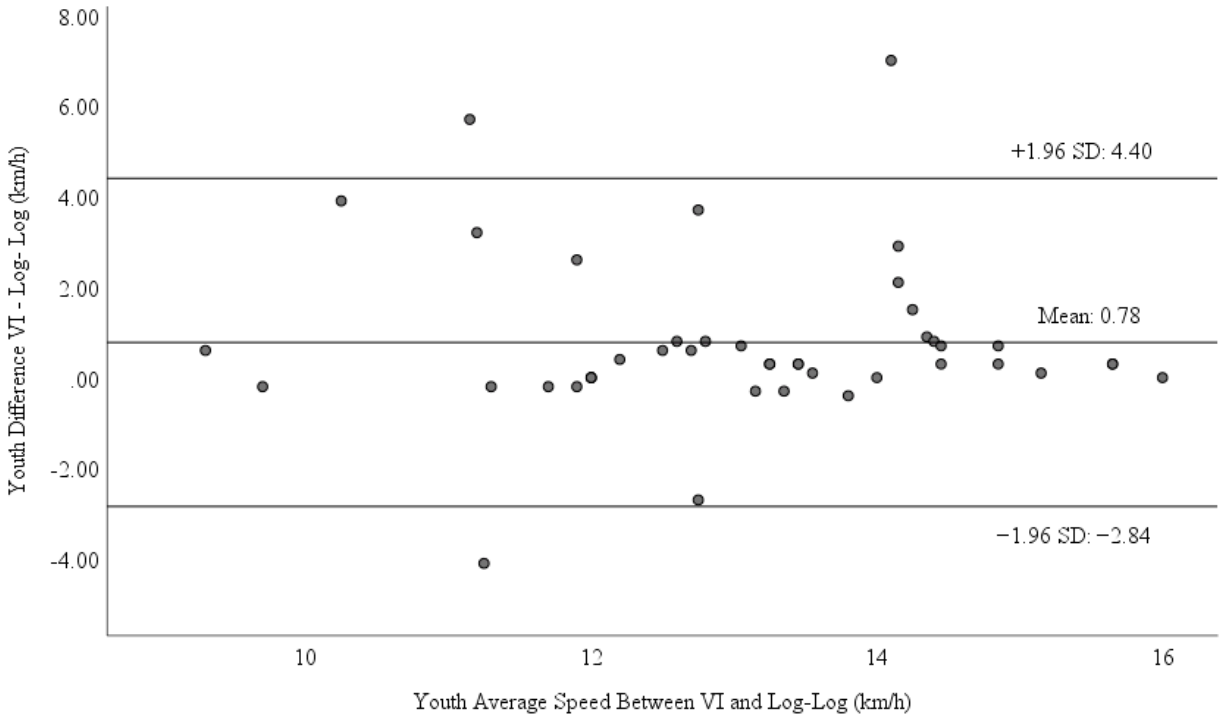
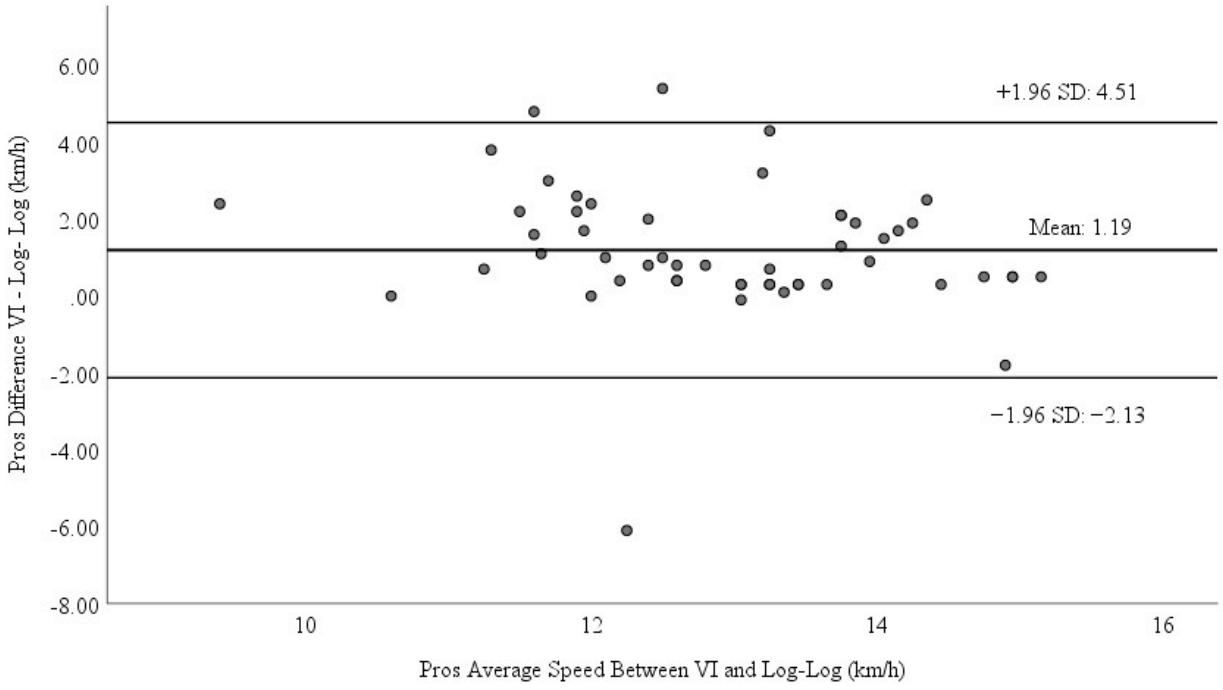


Figure 1. The lactate curve comparison between youth ($n = 25$) and pro ($n = 36$) athletes who completed the 18km/h increment. Note that * = $p < 0.05$.

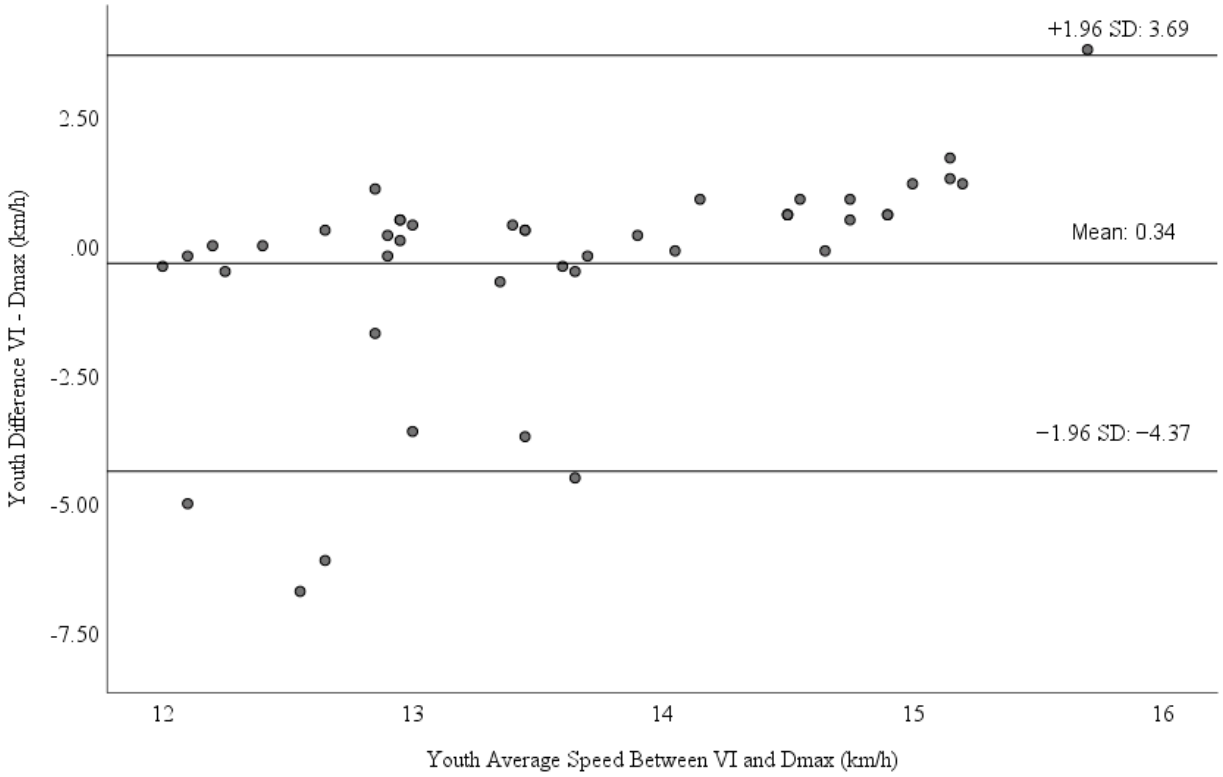
Bland–Altman plots present the limits of agreement between the different measurement methods of LT for youth and pro athletes (Figure 2A–F).


8.00
6.00
4.00
2.00
.00
-2.00
-4.00
Youth Difference VI - Log- Log (km/h)
+1.96 SD: 4.40
Mean: 0.78
−1.96 SD: −2.84
10
12
14
16
Youth Average Speed Between VI and Log-Log (km/h)

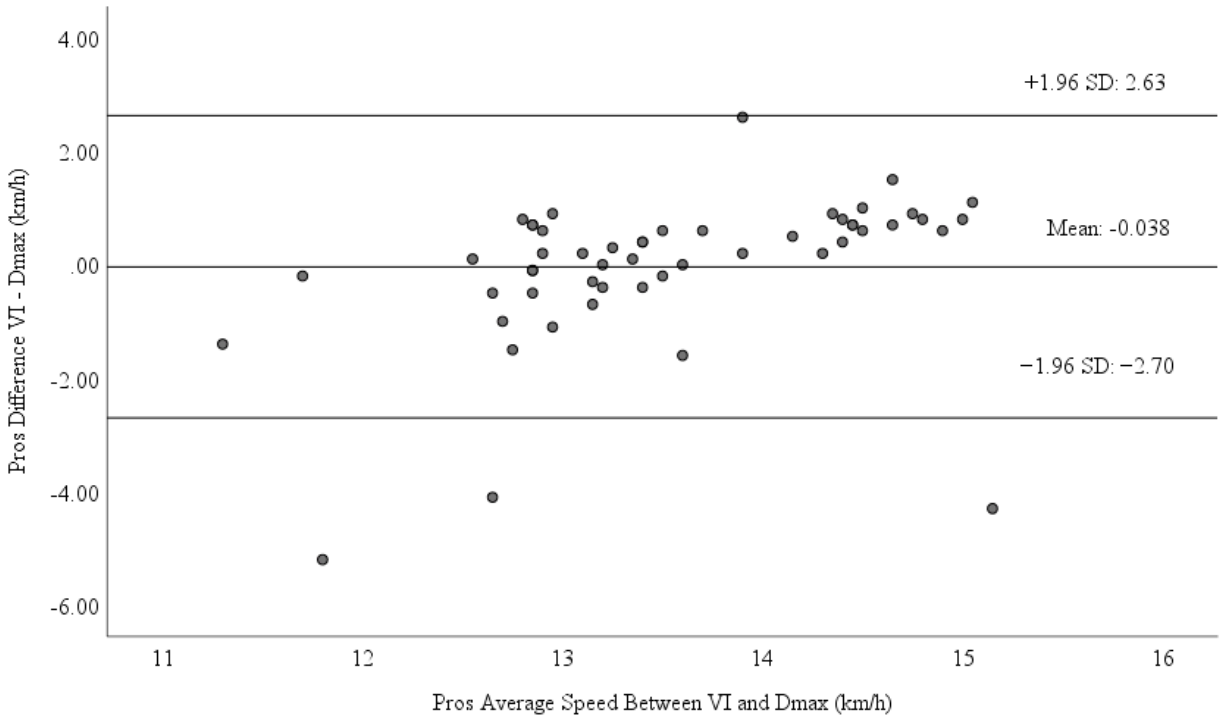
(**A**)


6.00
4.00
2.00
.00
-2.00
-4.00
-6.00
-8.00
Pros Difference VI - Log- Log (km/h)
+1.96 SD: 4.51
Mean: 1.19
−1.96 SD: −2.13
10
12
14
16
Pros Average Speed Between VI and Log-Log (km/h)

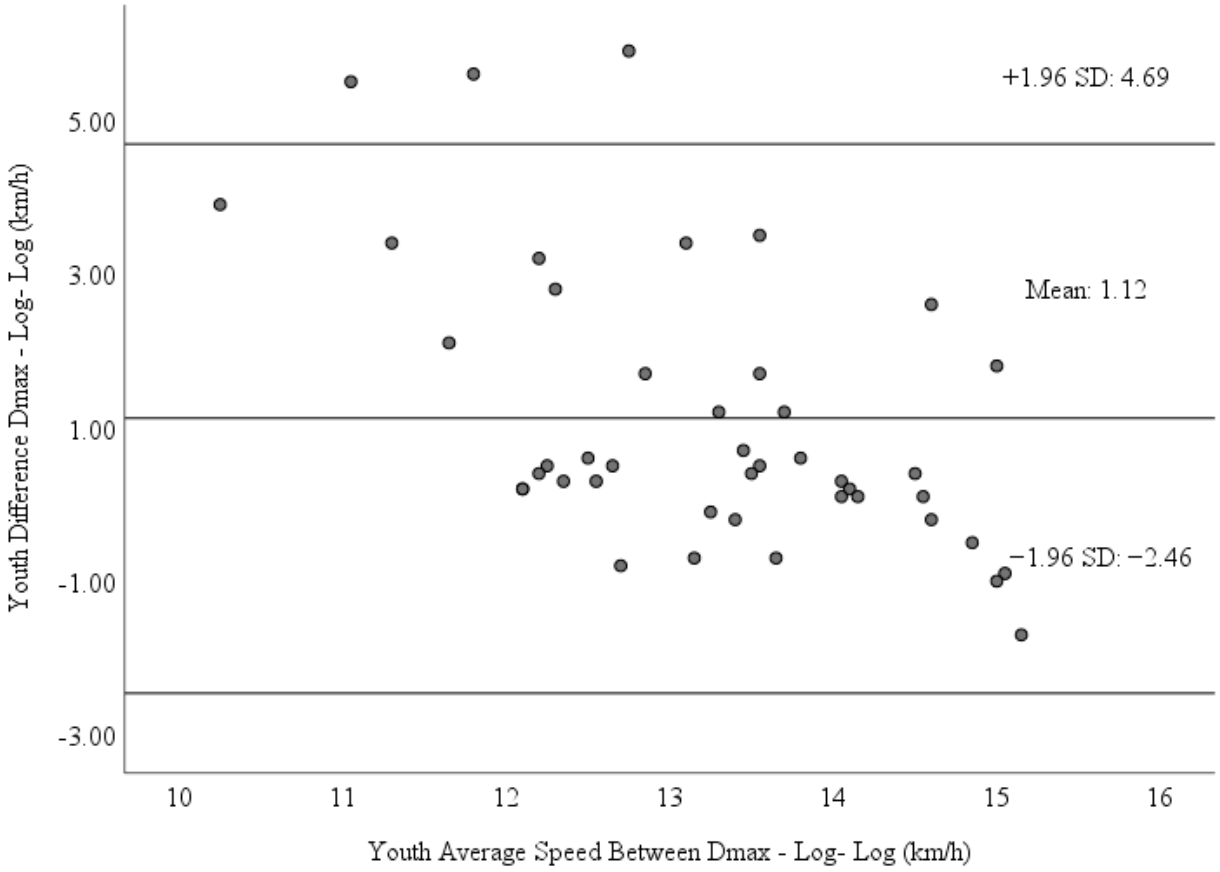
(**B**)


+1.96 SD: 3.69
Mean: 0.34
−1.96 SD: −4.37
2.50
.00
-2.50
-5.00
-7.50
Youth Difference VI - Dmax (km/h)
12
13
14
15
16
Youth Average Speed Between VI and Dmax (km/h)

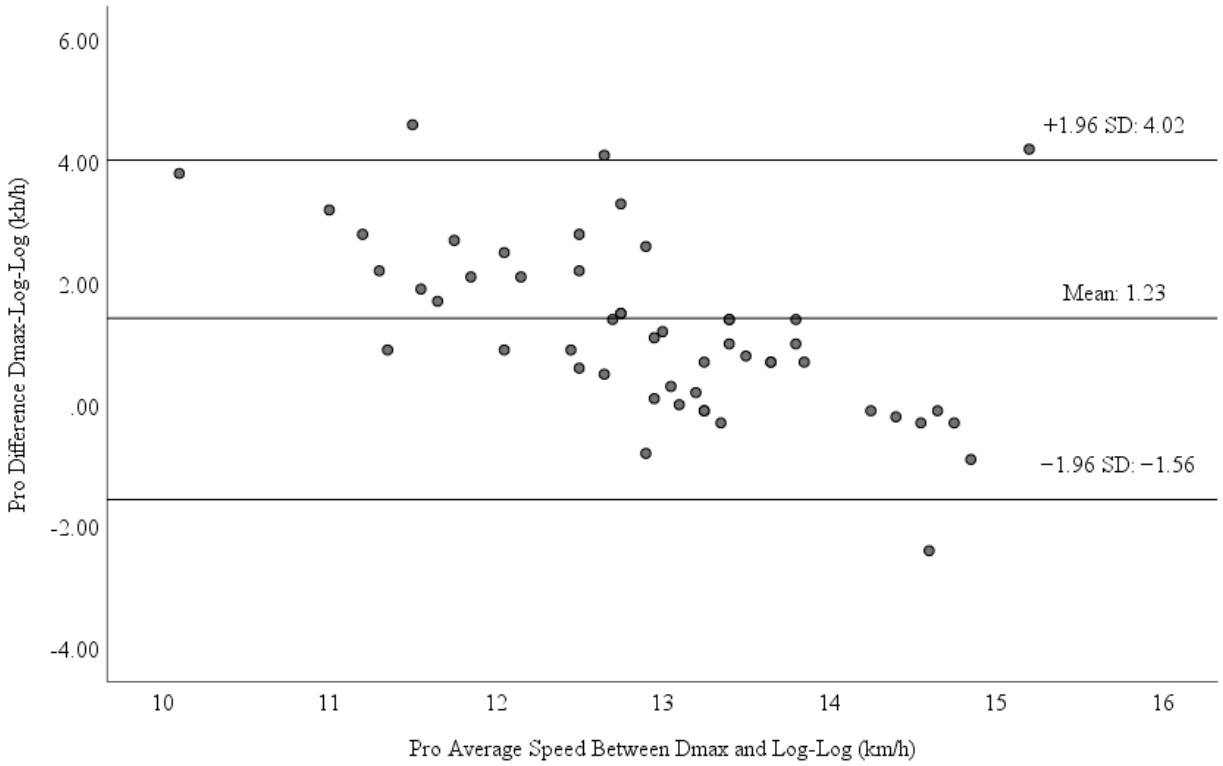
(**C**)


4.00
2.00
.00
-2.00
-4.00
-6.00
+1.96 SD: 2.63
Mean: -0.038
−1.96 SD: −2.70
Pros Difference VI - Dmax (km/h)
11
12
13
14
15
16
Pros Average Speed Between VI and Dmax (km/h)

(**D**)


+1.96 SD: 4.69
Mean: 1.12
−1.96 SD: −2.46
5.00
3.00
1.00
-1.00
-3.00
10
11
12
13
14
15
16
Youth Difference Dmax - Log- Log (km/h)
Youth Average Speed Between Dmax - Log- Log (km/h)

(**E**)



(**F**)

Figure 2. The agreement between VI and Log–Log methods for (**A**) youth and (**B**) pro athletes; VI and Dmax methods for (**C**) youth and (**D**) pro athletes; and Dmax and Log–Log methods for (**E**) youth and (**F**) pro athletes.

Similarly to Cerda-Kohler et al. (2016) [25], an ICC > 0.80 was considered an indicator of agreement between the LT detection methods. Table 4 indicates the significant correlations; however, none were strong enough to meet the agreement criteria.

Table 4. Analysis agreement between the visual inspection (VI), logarithmic (Log–Log) and maximum distance (Dmax) methods in pro and youth soccer players using Intraclass Correlation Coefficients (ICCs).

	LT Speed ICC (95% CI)	
	Youth (*n* = 44)	**Pro (*n* = 52)**
VI vs. Log–Log	0.65 * (0.35–0.81)	0.51 * (0.15–0.72)
VI vs. Dmax	−0.03 (−0.89–0.439)	0.44 * (0.02–0.68)
Dmax vs. Log–Log	0.39 (−0.12–0.67)	0.57 * (0.26–0.76)

Note: * = $p < 0.05$.

The regression analysis and coefficients of determination are presented in Table 5 for both pro and youth soccer players.

Table 5. Predictiveness of running performance on the incremental test based on the various methods of lactate threshold (LT) determination.

	Coefficient of Determination (R^2)	
	Youth (n = 44)	**Pro (n = 52)**
VI	0.35 **	0.48 **
Log–Log	0.17 *	0.04
Dmax	0.01	0.08 *
FBLA (4 mmol)	0.27 **	0.23 **

Note: * $p < 0.05$. ** $p < 0.01$.

4. Discussion

This cross-sectional study aimed to compare the lactate output profile and aerobic fitness of professional soccer players to those of selected elite youth players. Additionally, the study aimed to determine the LT using four methods, examine their agreement, and conclude if LT detection methods can be used interchangeably in youth, as this was shown not to be possible in professional players [25]. The results demonstrated that professional players exhibited superior cardiovascular fitness compared to youth by performing significantly longer during incremental testing. This was also evident from the fact that 69.23% of professional players had a $\dot{V}O_2$ max running velocity (v$\dot{V}O_2$ max) of 18 km/h versus only 56.82% of the youth. Despite the superiority of the professional players in incremental testing, youth players exhibited a significantly higher $\dot{V}O_2$ max. This finding aligns with evidence from other studies, suggesting that other factors might be more sensitive indicators of superior aerobic performance [16,17], such as shifts in lactate accumulation at higher intensities. According to an extensive review by Poole et al. (2021) [36], the mechanism that most likely regulates the relationship between exercise intensity and increases in muscle and blood lactate is the mitochondrial reticulum volume density. It is also well documented that following endurance training; mitochondrial plasticity is enhanced [37]. Thus, in this study, it is possible that professional players had higher adaptations to endurance training before the preseason period. Perhaps this may be due to their strict adherence to the targeted exercise maintenance programs during the transition period or the more competitive nature of their championship compared to youth leagues.

Furthermore, the preseason vLT calculated using the four LT detection methods (Table 2) in this study is in agreement with the literature on both youth [35] and professional players [17,38]. However, the LT thresholds were not significantly different between the professional players and the youth (Table 2), despite the lactate accumulation during each stage being slightly lower for professional players (Figure 1). Only the FBLA (4 mmol/L) LT method yielded a running velocity that was significantly higher in professional players than in youth. Interestingly, the FBLA (4 mmol/l) and VI running velocities were also better performance predictors of incremental testing (Table 5). The use of CLT as a predictor of running performance is contradictory in the literature, as studies have shown inconsistent results regarding the superiority of various LT detection methods. Specifically, some studies demonstrated that the Dmax method is more valid and reliable [39,40] compared to FBLA (4 mmol/l), whereas others demonstrated the opposite [41]. In cyclists, the Dmax intensity threshold is indicative of maximal lactate steady-state (MLSS) intensity [42,43]. However, the reliability of the Dmax method is challenged [44,45], as the accuracy of this method depends on the first and last lactate measurements. Interestingly, our results demonstrated that the final lactate level was significantly higher in the youth group than in the professional group (Figure 1). This is because the youth challenged their anaerobic energy system contribution towards the end of the incremental testing before volitional fatigue. Presumably, youth consider incremental testing as a criterion for their

professional development, aiming to achieve longer running times. This speculation is not supported by Hendry et al. (2018) [46], who demonstrated that elite youth soccer players have higher autonomous motivation, most likely resulting from their engagement in diverse developmental activities rather than being driven by external rewards related to pro status. It could also be due to their lack of experience with the pacing required for incremental testing, unlike the professional players who routinely perform the test throughout their careers. Alternatively, it could simply indicate the superiority of professional players in aerobic fitness, since Chalmers et al. (2015) [47] demonstrated that peak lactate concentrations decrease after endurance training interventions. Thus, the lower value of the last stage in professional players could cause an underestimation of the vLT using the Dmax method [47]. Consecutively, in addition to other disadvantages associated with LT determination, such as the inability of the literature to establish a gold standard for laboratory LT determination [48], Dmax's low reliability [41], the LT underestimations of the log-to-log transformation method, and the experience dependence and subjectivity of the VI method [49], the youth's last value might have influenced the results as outliers could affect these estimation methods that rely on regression analyses [33].

There is limited research on the interchangeability of the various LT detection methods. Specifically, only four other studies have examined the agreement between the various LT detection methods. Davis et al. (2007) [49] examined the agreement between three LT detection methods for cyclists of both sexes. Despite the strong correlations detected, the Bland–Altman analysis indicated that the wattages had a large LOA and thus lacked agreement. Similarly, Hauser et al. (2014) [50], in a larger study using 57 cyclists, indicated poor agreement between the LT concepts used in their study and the MLSS. Furthermore, Carter et al. (2019) [44] examined the agreement of six different LT detection methods in elite cross-country skiers. Their results revealed that these six methods should not be used interchangeably. To our knowledge, only one similar study has examined LT agreement among professional soccer players [25]. Their running velocities at LT, detected with four different methods, were higher than our professional group, but this was expected as they collected the data towards the end of the season, during which the values [35] shifted at higher intensities. Similarly to the literature mentioned above, our results show a failure to meet the criteria for agreement as set for players before [25] and, thus, poor agreement among the LT detection methods investigated in this study (Figure 2A–F and Table 5). Furthermore, there is a paucity of evidence on which method is more accurate in determining vLT in both professional and youth soccer players. Thus, further research is required to determine the most appropriate LT concept, and follow-up validation studies are required to examine its accuracy. A major limitation of LT determination and its practicality is that soccer entails a large interplay of running velocities, many of which are above maximal aerobic speed. Additionally, high-intensity interval training formats are tailored based on the different locomotor profiles (speed, endurance, or hybrid profiles) that characterize soccer players [51]. On the same note, Lyzohub et al. (2021) [52] demonstrated that physical activity alternates constantly between the aerobic and anaerobic systems based on the match's demands. Perhaps those are the reasons for the scarce information on lactate-guided threshold interval training in soccer players, in contrast to the vast body of literature and the ongoing promising approaches in middle- and long-distance runners [53]. Thus, the use of LT determination in soccer is mainly utilized as a determinant of fitness readiness rather than a method for setting training intensities. Further research using global positioning systems (GPSs) during official games would offer a better understanding of how improvements in vLT with training would affect external load and develop training programs to develop all energy systems involved. Additionally, LT determination could be utilized in adjusting the running thresholds set by the various GPS software.

Furthermore, the results of this study demonstrated the importance of lactate profiling in youth for comparative purposes with the professional squad and their lack of agreement. The vLT in soccer is indicative of aerobic status [54], is significantly faster for higher playing standards [17], and is more sensitive to aerobic fitness adaptations of the preseason [55] and in-season periods [35]. Likewise, professional squad practitioners should integrate LT determination as a regular assessment tool for youth to monitor and prescribe endurance sessions. However, LT concepts should never be used interchangeably by both professional and youth soccer players, as they lack agreement. When evaluating the aerobic power of youth for talent identification, lactate profiling during incremental testing and RE should be evaluated, which could be more informative than the widespread field testing usually applied in youth leagues. Practitioners should carefully standardize the termination of incremental testing based on the $\dot{V}O_2$ max criteria and avoid volitional fatigue, especially in youth soccer players, as this will cause an engagement of the anaerobic system beyond what is physiologically required and thus overestimations of the LT.

5. Conclusions and Practical Applications

In conclusion, elite U-17 soccer players exhibit higher $\dot{V}O_2$ max values than professional players, but not necessarily superior aerobic fitness. This study verified that $\dot{V}O_2$ max values alone do not indicate superior aerobic power. For training prescription purposes, practitioners should use the different LT concepts interchangeably in both the youth and professional players, as our results verified the scarce information in the literature that suggested avoiding looking at mean differences among these LT calculation methods and instead using Bland–Altman analyses to verify the agreement. Finally, the velocity of the 4 mmol/L FBLA method might make it the best method to compare professional players to youth, as it is more robust than other methods and a better predictor of professionals' superiority in incremental testing. Nevertheless, caution should be exercised when using this method, as it was shown to be sensitive to dietary modifications [56]. Future verification research should focus on determining which LT method is more suitable for defining training intensities in soccer players and evaluating their effectiveness.

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