

A Different Insight into Neuromuscular Performance Evaluation: The Influence of Fatigue in Hamstrings:Quadriceps Ratio

Nöromusküler Performans Değerlendirmesine Farklı Bir Bakış: Yorgunluğun Hamstring:Quadriceps Oranı Üzerine Etkisi

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ÖZ

Bu araştırmanın amacı, maksimum bireysel yorulma intolerans noktasında gerçekleştirilen bir egzersizin ardından ölçülen yorgunluk indeksi ve hamstrings-quadriceps parametrelerinin alternatif bir H/Q_{Yorgunluk} yaklaşımı kullanarak ($T_{Lim}vVO_{2max}$) belirlenmesi ve (H/Q) oranlarını dinlenik koşullarda gerçekleştirilen geleneksel (H/Q_{CR}) yöntemleri ile karşılaştırmaktır. Yorgunluk ve spor modalitesinin elde edilen parametreler üzerinde bir etkisi olup olmadığını belirlemek için farklı sporlar branşlarından 34 erkek sporcu katıldı. Kişilerin maksimum bireysel yorgunluk intolerans noktasını belirlemek amacıyla tüm katılımcılar VO_{2max} , vVO_{2max} ve $T_{Lim}vVO_{2max}$ testlerine tabii tutuldu. Analiz edilen H/Q verileri 180°/s açışal hızda belirlendi. Analiz sonuçlarına göre H/Q⁴⁸⁻⁵⁰ tekrarlarında elde edilen değerler (H/Q_{Sağ}^{48,49,50}: 1.20-1.24 ile H/Q_{CR}: 0.57-0.62; p < 0.05; ve H/Q_{Left}^{48,49,50}: 1.17-1.34 ile H/Q_{CR} 0.53-0.55; p < 0.001) geleneksel yöntemlerden istatistiksel olarak daha yüksek bulundu. Bu sonuçlar doğrultusunda yorucu bir aktivite sonrasında elde edilen H:Q değerleri ve yorgunluk indeksi değerlerinin geleneksel ölçüm yöntemlerine kıyasla daha farklı sonuçlar ortaya çıkardığı ileri sürülebilir.

Anahtar Kelimeler: Egzersiz ölçüm testleri - Oksijen tüketimi - Spor, Yorgunluk indeksi, H/Q

ABSTRACT

The purpose of this investigation was to evaluate the differences among fatigue index and hamstrings-to-quadriceps (H/Q) peak moment ratios subsequent to an exhaustive running ($T_{Lim}vVO_{2max}$) trial performed at maximum individual fatigue intolerance point using an alternative H/Q_{Fatigue} approach rather than other conventional (H/Q_{CR}) methods of determining the hamstrings-to-quadriceps ratio under non-fatigued conditions. Thirty-seven male athletes from different sports participated to determine if there are differences due to the influences of fatigue and sports modality. VO_{2max} , vVO_{2max} , and $T_{Lim}vVO_{2max}$ were measured to determine maximum individual fatigue intolerance point with two preliminary test sessions. H/Q data analyzed were for angular velocities of 180°/s. H/Q_{Fatigue} calculated using the moment developed in repetitions 48-50 was significantly greater than other conventional methods (p < 0.001). Significant differences were apparent among new and conventional methods (H/Q_{Right}^{48,49,50}: 1.20-1.24 vs. H/Q_{CR}: 0.57-0.62; p < 0.05) and (H/Q_{Left}^{48,49,50}: 1.17-1.34 vs. H/Q_{CR}: 0.53-0.55; p < 0.001), respectively. H/Q_{Fatigue} ratios following an exhaustive exercise offer different information compared to conventional methods of determining the hamstrings-to-quadriceps ratio under non-fatigued conditions.

Keywords: Exercise testing - Oxygen consumption - Sports, Fatigue index, H/Q.

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INTRODUCTION

The term “body fatigue”, which in turn provokes a decrease in the ability to continue to maximum performance, refers to a homeostatic disturbance and exercise-induced reduction in maximal voluntary muscle force induced by physical activity (Gandemia, 2001). Muscular fatigue may cause significant muscular asymmetries that could lead athletes to injuries. Therefore, in order to identify muscle imbalances and establish injury prevention programs it is essential to assess the muscular function of elite athletes after muscular fatigue (Scattone-Silva et al., 2012).

Presumably, the hamstring-to-quadriceps-ratio (H/Q) may be the best method to evaluate the muscular injury risk caused by fatigue and muscular imbalances. The hamstrings-to-quadriceps muscle strength ratio calculated using peak moment strength parameters has been widely used as a complementary assessment tool in the evaluation of neuromuscular performance. Despite accumulated fatigue being an important risk factor for both hamstring strain and anterior cruciate ligament injuries in athletes conventional H/Q ratios and fatigue index methods which are the outcome of a test usually carried out using slow velocity, alternating knee extension and flexion focused contractions, are sometimes criticized since prevailing hamstring injuries occur when the knee joint angular velocity is high during the eccentric phase of running (Heiderscheit et al., 2005).

To date, several versions of these tests were developed but one possible problem with current versions of the H/Q ratio test is that conventional H/Q (H/Q_{CR}) tests underestimate other neuromuscular variables which could also influence the antagonist to agonist muscle relationship, such as muscle fatigue in the lower limbs, or muscle activation and are performed under non-fatigued conditions which may lead to imprecise results even though the H/Q ratio was previously demonstrated to be sensitive to experimentally imposed muscular fatigue (Ruas et al., 2019; Cohen, Zhao et al., 2015). Therefore, alternative tests need to be developed, or the H/Q ratio may need to be tested under different muscle contraction conditions to have a greater predictive potential.

Time to exhaustion (T_{Lim}) test protocols are performed at a constant speed or power output and participants are to maintain the test until volitional exhaustion at their vVO_{2max} until they no longer continue the required work rate (Denadai and Denadai, 1998; Wilber and Moffatt, 1992). However, due to the concept of this constant activity a reduction in mechanical efficiency results in an increased local muscular fatigue (Figueiredo et al., 2011).

However, despite the prevalent use of screening tests used in the evaluation of lower extremity injury prevention and rehabilitation, the muscle imbalance, knee joint stability, muscle strength properties and functionality recent data have shown that anterior cruciate ligament (ACL) (re)injury rates have increased in recent years which possibly indicating an ineffectiveness of such tests to predict (re)injury mechanism (Ekstrand et al., 2001; Sanders et al., 2016). It is therefore important to offer alternative H/Q ratios as sensitive screening methods to predict (re)injury occurrence, monitor joint integrity, and identify the relevance of exercise type and intensity in observed H/Q ratios.

Studies to date calculated H/Q using various methods described previously (Weber et al., Pinto, 2010; Thorstensson & Karlsson, 1976; Kawabata et al., 2000). However, despite the relevance of fatigue indexes during H/Q calculation there are wide range of differences among these equations as fatigue index commonly calculated as the percent decrease in the average moment over the last three or five repetitions relative to the percent decrease in the average moment over the first three or five repetitions. Considering the importance of fatigue in the assessment of H/Q ratio Pinto et al. (2018) developed an alternative fatigue index method where the percent decrease in the peak joint moment over the last three repetitions was compared to the maximal joint moment over 30 repetitions at an angular velocity of $300^{\circ}\cdot s^{-1}$ (Pinto et al., 2018). Taking the intra-subject variability of initiation and heterogeneity in maintenance of force production over 30 repetitions

into consideration we tested an alternative method in this study to calculate fatigue index over 50 repetitions at an angular velocity of $180^{\circ}\cdot s^{-1}$ and will be referred to as Equation A, which is a new method proposed in this study. Equation B and C are conventional methods used to calculate fatigue index parameters over a 50 repetition at an angular velocity of $180^{\circ}\cdot s^{-1}$. Thus, we hypothesized that if neuromuscular fatigue in the quadriceps and hamstring muscles during fatigue test affect observed H/Q ratios the H/Q ratios calculated using this new alternative method subsequent to fatigue test would indicate higher H/Q ratios compared to other conventional methods. With this in mind, we aimed to evaluate the effect of fatigue on H/Q ratios subsequent to an exhaustive running (i.e. T_{Lim} at vVO_{2max}) trial performed at maximum individual fatigue intolerance point using an alternative $H/Q_{Fatigue}$ approach rather than other conventional (H/Q_{CR}) methods of determining the hamstrings-to-quadriceps ratio under non-fatigued conditions.

METHODS

Participants: Thirty-seven athletes aged ranged 17 to 33 years (21.41 ± 4.09) volunteered to participate in this study.

Test Procedures: The physical characteristics of participants in each group were presented in Table 1. All participants gave written informed consent prior to participating in the study approved by Mersin University Institutional Review Board in compliance (Protocol number: 156, date of approval: 05.06.2014) with the ethical standards of the Helsinki Declaration. All participants were familiarized to the experimental procedures and informed about equipment. The anthropometric parameters (body fat mass, lean body weight, weight) were assessed using Bioelectrical impedance analysis (Tanita 418-MA Japan) before VO_{2max} and $T_{Lim}vVO_{2max}$ test sessions. Height was measured with a stadiometer in the standing position (Holtain Ltd. U.K.). Athletes performed two tests over a one-week interval with two separate visits to the laboratory. In the first visit, athletes underwent a progressive treadmill test to determine VO_{2max} and vVO_{2max} . In the following session, the $T_{Lim}vVO_{2max}$ test was carried out at a constant speed until volitional exhaustion. Preliminary testing sessions were applied in assessment of maximum fatigue intolerance point of each participant for running time to exhaustion. Heart Rate (HR) was also monitored and recorded throughout VO_{2max} and T_{Lim} test sessions using 12-lead ECG.

Determination of VO_{2max} , vVO_{2max} , and T_{Lim} at vVO_{2max} : VO_{2max} , vVO_{2max} , and $T_{Lim}vVO_{2max}$ were measured in a preliminary test session using a progressive exercise protocol on a treadmill. Oxygen consumption was measured breath by breath through a gas analyzer (CareFusion MasterScreen CPX, ABD) and subsequently averaged over 15-second intervals. Before each test, the automated gas analyzer was calibrated according to the manufacturer's recommendations and using standard gases of known concentration.

All participants maintained standing position on the treadmill and were asked to hold the handrails before initializing device for a test session. Then, the treadmill speed was set to $5\text{ km}\cdot h^{-1}$ (0 % slope) and increased every minute by $1\text{ km}\cdot h^{-1}$. Following this warm-up process, the test was started when the speed reached at $8\text{ km}\cdot h^{-1}$. Throughout the tests, participants received verbal encouragements and they were asked to rate their perceived exertion on Borg's scale at the end of each minute. The test continued until at least two of the following criteria were obtained: a plateau in VO_2 despite an increase in running speed; a respiratory exchange ratio (RER) above 1.1; HR over 90 % of the predicted maximal HR. If the stage of 1 min could not be completed, the velocity of the previous stage was recorded as vVO_{2max} . At the following week, athletes underwent a T_{Lim} test using vVO_{2max} on a treadmill under the same laboratory conditions. Following a 15-min warm-up period at 60% vVO_{2max} , the speed was immediately increased (in less than 20 s) up to vVO_{2max} . Then, the participants were encouraged to run to their volitional exhaustion. The time from when the vVO_{2max} was first attained

until participants' volitional exhaustion was recorded to the nearest second as $T_{Lim} vVO_{2max}$. The test was ended when the participants failed to continue running at the required velocity despite verbal encouragement.

Isokinetic Strength Testing: In assessment of isokinetic peak moments the participants were seated on the Cybex chair in upright position prior to isokinetic test session with the hips flexed at an angle of 90° and using pelvic and thigh straps the hips and thighs of participants were stabilized following treadmill T_{Lim} testing sessions. As part of the familiarization process, the participants were given standard verbal instructions regarding the procedures and then performed a maximum of five repetitions at angular velocities of $60^\circ/s$ to determine isokinetic peak moment strength, and the results were stored for analysis. The participants were instructed to exert effort as hard and as fast as possible for all contractions.

Fatigue Testing Protocol: In order to determine conventional and $H/Q_{Fatigue}$ all participants underwent a fatigue test protocol subsequent to $T_{Lim} vVO_{2max}$ measurements. Participants performed 50 maximal bilateral knee extension and flexion repetitions at an angular velocity of $180^\circ/s$ to induce the participants to fatigue. The participants were asked to perform as quickly as possible and to grasp the handles at the sides of the chair throughout the warm-up and the test. The knee moment of the limbs was measured through a range of motion from 90° (knee flexion) to 0° (full knee extension). Gravity correction was made prior to isokinetic test protocol sessions. The participants underwent the same protocol for both of their legs during all isokinetic testing sessions.

Equations used to calculate the knee extensor and flexor fatigue index are shown below:

Fatigue Index method	Equation
A	$[(Mom_{max} - P_{48,49,50}/Mom_{max}) \times 100]$
B	$[(PM_{1-3} - P_{48-50}/PM_{1-3}) \times 100]$
C	$[(PM_{1-5} - P_{46-50}/PM_{1-5}) \times 100]$

Also, the equations used in the determination of hamstring-to-quadriceps ratio are shown below:

Strength Ratio (abbreviation)	Description	Equation
Fatigue Ratio _(48,49,50)	Fatigue ratio calculated using the 48th, 49th, and 50th repetitions (individual comparisons)	Knee Flexor $Fl_{48,49,50}$ / Knee Extensor $Fl_{48,49,50}$
Mean (48-50) Fatigue Ratio	Mean Fatigue Index value of the last three repetitions	Knee Flexor Fl_{48-50} / Knee Extensor Fl_{48-50}
M-H/Q_{CR}	Maximal H/Q conventional ratio utilizing the maximum joint moment ($Moment_{max}$)	Knee Flexor Mom_{max} / Knee Extensor Mom_{max}
P₍₁₋₃₎ - H/Q_{CR}	Mean Fatigue Index value of the first three repetitions	Knee Flexor PM_{1-3} / Knee Extensor PM_{1-3}
P₍₁₋₅₎ - H/Q_{CR}	Mean Fatigue Index value of the first five repetitions	Knee Flexor PM_{1-5} / Knee Extensor PM_{1-5}
P₍₁₎ - H/Q_{CR}	Joint moment of the first repetition	Knee Flexor PM_1 / Knee Extensor PM_1

Statistical Analysis: Descriptive statistics were used to summarize data. Shapiro Wilk test was applied to test the normal distribution. Since some of the variables were not distributed normally and sample size of the groups are not adequate for parametric test the Kruskal-Wallis H analysis was used to test the statistical significance and the Mann Whitney U test was used to determine any significant difference between branches. A paired sample t-test was used to compare Mom_{max} and joint moments during the time to exhaustion tests subsequent to vVO_{2max} . To test the reproducibility of the data collected subsequent to fatigue test protocol each repetition was compared to the maximum joint moment and the reproducibility was provided using the quotient of individual repetitions (48 to 50) to the maximum joint moment during fatigue test.

Intraclass Coefficient (ICC) and Intraclass Coefficient Confidence Intervals (ICC CI 95%) was determined to represent the proportion of variance in a set of scores that is attributable to the true score variance. The level of statistical significance was set at $p < 0.05$ and $p < 0.001$ for all comparisons and Bonferroni adjustment was applied. To describe differences related to equations, effect sizes were calculated as the difference between means divided by the pooled standard deviation. An effect size ≥ 0.20 and < 0.50 was considered small, ≥ 0.50 and < 0.80 medium, and ≥ 0.80 large using Cohen's criteria. The statistical analysis was performed with SPSS version 20.0 (SPSS Inc., Chicago, IL, USA). GraphPad Software GraphPad Prism 6 was used for graphical expression.

RESULTS

Data of physiological characteristics of participants are shown as mean±standard deviation (Mean±SD) in Table 1.

Table 1: Demographic and physical characteristics of participants for vVO_{2max} and T_{Lim} test sessions. (Mean±SD).

	Cycling n=8	Martial Arts n=10	Soccer n=10	Track and Field n=9
Age (years)	24.75±5.28	20.60 ±3.47	21.30±3.62	19.44±2.46
Height (cm)	177.63±6.65	173.60±5.30	175.00±5.89	174.00±6.46
Weight vVO_{2max} (kg)	70.21±7.37	68.90±10.54	60.57±6.81	64.71±6.26
LBM vVO_{2max} (kg)	64.01±6.53	59.07±7.79	68.22±7.12	56.99±6.31
PFM vVO_{2max} (%)	8.70±5.42	12.92±7.68	10.51±4.16	12.02±2.76
Weight T_{Lim} (kg)	69.53 ±9.25	69.03±10.91	68.22±7.12	64.68±6.16
LBM T_{Lim} (kg)	64.36±5.13	59.73±8.29	61.02±6.61	57.54±6.41
FM T_{Lim} (%)	10.06±6.13	13.78±7.29	10.51±4.16	11.12±2.96

LBM: Lean Body Mass, **PFM:** Percent Fat Mass. Subject characteristics were measured mean Mean±SD.

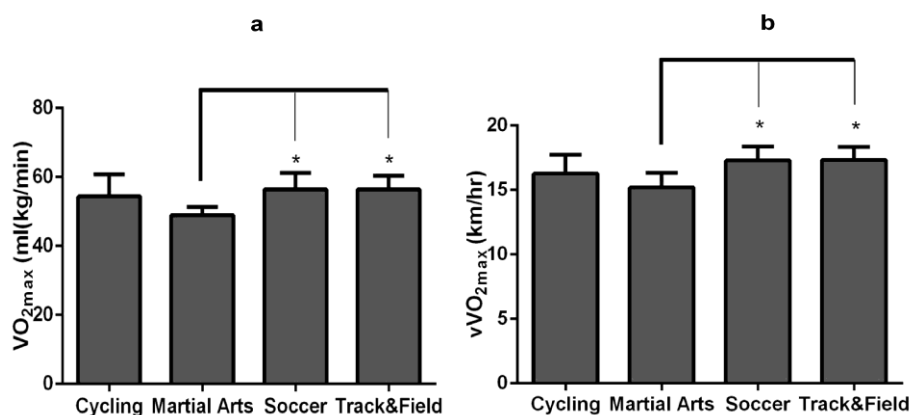
According to statistical analysis no significant differences were found in demographic and anthropometric parameters between groups (height ($\chi^2(3)=2.278$, $p=0.517$), age ($\chi^2(3)= 6.398$, $p=0.094$), body weight ($\chi^2(3)=1.596$, $p=0.660$), LBM ($\chi^2(3)=4.386$, $p=0.223$), $FM_{vVO_{2max}}$ ($\chi^2(3)=3.589$, $p=0.309$), $FM_{T_{Lim}}$ ($\chi^2(3)=0.645$, $p=0.886$), respectively. There was a significant difference in VO_{2max} ($\chi^2(3)=14.153$, $p=0.03$) and vVO_{2max} ($\chi^2(3)=14.287$, $p=0.03$) among groups (Table 2).

Table 2. Comparison of vVO_{2max} , T_{Lim} , VO_{2max} , HR, RE and RER measurements of the groups. Mean rank (Min-Max).

	Cycling n=8	Martial Arts n=10	Soccer n=10	Track and Field n=9
vVO_{2max} (km.h ⁻¹)	16.94 (14-18)	9.50 (13-17)	25.00* (15-18)	24.72* (16-19)
T_{Lim} (seconds)	22.00 (219-800)	17.85 (139-478)	16.05 (160-368)	20.89 (239-492)
VO_{2max} (ml.kg ⁻¹ .min ⁻¹)	19.94 (45.50-63.20)	8.35 (43.70-51.40)	24.05* (50.50-65.30)	24.39* (50.30-61.40)
HR (beat/min)	19.00 (163-197)	22.50 (177-194)	18.85 (175-197)	15.28 (167-192)
RPE	24.19 (13-19)	14.95 (11-17)	22.85 (13-19)	14.61 (7-17)
RER	12.38 (1.07-1.21)	20.55 (1.06-1.24)	23.60 (1.12-1.22)	18.06 (1.10-1.20)

HR: Heart rate, RPE: Rate of Perceived Exertion RER: Respiratory Exchange Ratio *p<0.05

Track and field athletes ($U=4.00$, $Z=-3.35$, $p=0.000$) and soccer players ($U=3.50$, $Z=-3.51$, $p=0.000$) had significantly higher VO_{2max} values than martial arts athletes (Figure 1a). Mann-Whitney U test revealed that vVO_{2max} values of track and field athletes ($U=6.50$, $Z=-3.21$, $p=0.001$) and soccer players ($U=9.50$, $Z=-3.14$, $p=0.001$) were significantly higher than martial arts athletes (Figure 1b).

**Figure 1:** Comparison of (a) VO_{2max} and (b) vVO_{2max} values among groups.

There were no significant differences between groups in T_{Lim} ($\chi^2(3) = 1.745$, $p=0.05$). According to the results of the analysis, there were no significant differences among groups in terms of HR, RPE and RER. However, significant differences were apparent among conventional and new fatigue index methods (Table 3).

Table 3. Comparison of fatigue index parameters using new and conventional equations.

Fatigue Index Extension (Right)			Fatigue Index Flexion (Right)				
Method	Mean±SD (%)	Effect Size (95% CI)	Method	Mean±SD (%)	Effect Size (95% CI)		
A ₄₈ vs. C	41.25±12.60	19.44±13.21	0.81**	A ₄₈ vs. B	38.87±11.40	18.85±16.05	0.72 *
A ₄₉ vs. C	39.26±15.26	19.44±13.21	0.79**	A ₄₉ vs. B	34.68±11.25	18.85±16.05	0.65 *
A ₅₀ vs. C	42.46±13.25	19.44±13.21	0.86**	A ₅₀ vs. B	36.56±10.12	18.85±16.05	0.61 *
A ₄₈ vs. B	41.25±12.60	26.42±11.63	0.60 *	A ₄₈ vs. C	38.87±11.40	19.64±21.18	0.57 *
Fatigue Index Extension (Left)			Fatigue Index Flexion (Left)				
Method	Mean±SD (%)	Effect Size (95% CI)	Method	Mean±SD (%)	Effect Size (95% CI)		
A ₄₈ vs. B	43.54±10.25	20.41±15.58	0.83 **	A ₄₈ vs. B	42.75±18.28	19.41±17.11	0.58 *
A ₄₈ vs. C	43.54±10.25	19.57±14.87	0.86 **	A ₄₈ vs. C	42.75±18.28	20.12±15.06	0.55 *
A ₄₉ vs. C	41.61±13.78	19.57±14.87	0.86* *	A ₅₀ vs. B	35.96±10.33	19.41±17.11	0.59 *
A ₅₀ vs. B	37.53±10.09	20.41±15.58	0.79 **	A ₅₀ vs. C	35.96±10.33	20.12±15.06	0.52 *

Note. *p<0.05, **p<0.001. Subject characteristics were measured as Mean±SD.

Intraclass coefficient (ICC) and Intraclass coefficient confidence intervals (ICC CI 95%) was determined to represent the proportion of variance in a set of scores that is attributable to the true score variance. ICC was found 0.94 (95% CI, 0.74–0.99); 0.90 (95% CI, 0.55–0.98), and 0.74 (95% CI, 0.60–0.94) for knee extensor and 0.97 (95% CI, 0.88–0.99), 0.94 (95% CI, 0.77–0.99), and 0.90 (95% CI, 0.60–0.98) for knee flexor muscles for between the repetitions 48 to 50, respectively (Table 4).

Table 4. Intra-class coefficient (ICC) and intra-class coefficient confidence intervals used to determine the reproducibility between the repetitions 48 to 50.

	Treadmill knee extensors					
	Mean±SD	ICC	ICC CI 95%	CV	SEM	
[(Mom _{max} -P48/Mom _{max})×100]	41.25±12.60	0.94	0.74–0.99	3.88%	3.27	
[(Mom _{max} -P49/Mom _{max})×100]	39.26±15.26	0.90	0.55–0.98	2.14%	3.90	
[(Mom _{max} -P50/Mom _{max})×100]	42.46±13.25	0.74	0.60–0.94	11.77%	2.95	
	Treadmill knee flexors					
	[(Mom _{max} -P48/Mom _{max})×100]	38.87±11.40	0.97	0.88–0.99	2.52%	4.07
	[(Mom _{max} -P49/Mom _{max})×100]	34.68±11.25	0.94	0.77–0.99	0.72%	4.00
[(Mom _{max} -P50/Mom _{max})×100]	36.56±10.12	0.90	0.60–0.98	5.94%	3.31	

Note. CV: The coefficient of variation, SEM: the standard error of the mean. Subject characteristics were measured mean Mean±SD.

Comparison of H/Q_{Fatigue} and H/Q_{CR} parameters: H/Q_{Fatigue} calculated using the moment developed in repetitions 48-50 was significantly greater than other conventional methods. The results of the Kruskal Wallis-H analysis revealed statistically significant differences among new and conventional equations with respect to H/Q absolute fatigue ratios (H/Q_{Right}^{48,49,50}:1.20–1.24 vs. H/Q_{CR}: 0.57–0.62; p<0.05) and (H/Q_{Left}^{48,49,50}:1.17–1.34 vs. H/Q_{CR}: 0.53–0.55; p<0.001) following T_{Lim}VO_{2max} tests, respectively (Figure 3 a,b,c,d).

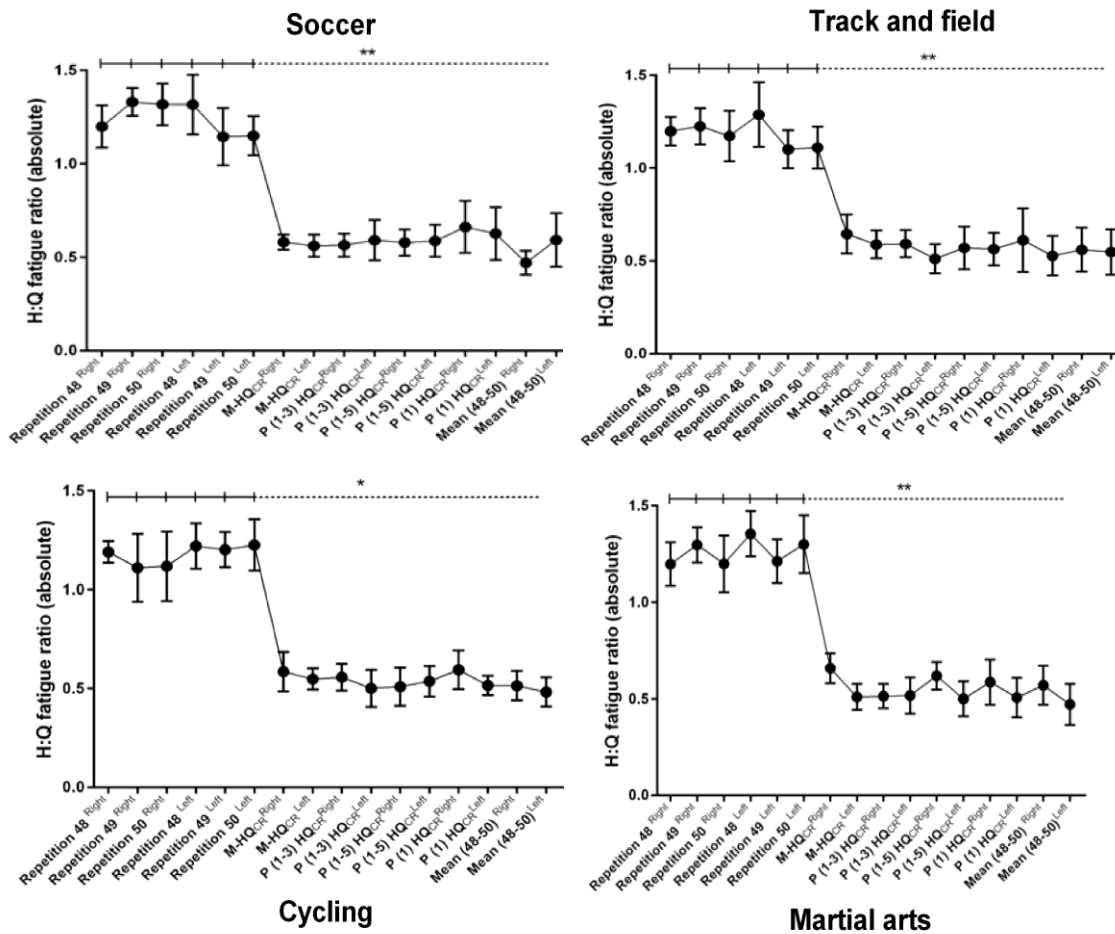


Figure 2 (a). Comparison of hamstring-to-quadriceps ratios of (a) soccer, (b) track and field, (c) cycling and (d) martial arts athletes using new and conventional methods following time to exhaustion test.

Interaction between neuromuscular capacity and fatigue test performance subsequent to treadmill time to exhaustion test: Knee extensor 1st ($Z = -2.092, p = 0.005$) and 2nd ($Z = -2.208, p = 0.041$) repetitions and flexor moment in the 1st ($Z = -2.438, p = 0.031$) and 2nd ($Z = -2.045, p = 0.028$) repetitions were significantly lower than the maximal joint moment subsequent to treadmill time to exhaustion test (extension Mom_{max} , mean from the 5th to the 7th repetitions, and flexion Mom_{max} , mean from the 8th to the 10th repetitions), respectively. Extensor moments decreased by the 8th repetition and remained significantly decreased to the end of the fatigue testing by the 25th repetition while flexor moments decreased by the 14th repetition and remained significantly decreased by the 20nd repetition to the end of the fatigue testing protocol when compared to the Mom_{max} . There was also a heterogeneous intra-subject distribution in the initiation of maximum joint moments during fatigue test subsequent to time to exhaustion test (Table 5).

Table 5. Intra-subject variability in the initiation of maximum joint moment

Frequency distribution	Repetition
39%	8 th repetition
25%	6 th repetition
21%	5 th repetition
9%	3 rd repetition
6%	1 st repetition

Note. The percent of frequency distribution indicates the repetition of which participants reached their maximum joint moment during fatigue test.

DISCUSSION

Running speed related hamstring injuries comprise a great deal of hamstring injuries in athletes and the data related to injury mechanism underlie that the incidence of hamstring injuries occur while the athlete is running at maximum or close to maximum speeds (Askling et al., 2007). Conventional hamstrings-to-quadriceps (H/Q) ratio, which is one of the most common evaluation methods, representing the concentric hamstrings (H_{con}) to quadriceps (Q_{con}) torque ratio (Kellis and Baltzopoulos, 1995).

It was reported that there is a relationship between stride frequency variations and running performance until time to exhaustion at the velocity of VO_{2max} (Boram et al., 2018) and an improved strength in the lower-limb which would require less activation of extensors muscles per stride reducing the actual demand of number of motor units during this constant intensity (Støren et al, 2011). The results of another study showed a significant relationship ($P = 0.024$) between horizontal ground reaction force and the combination of biceps femoris EMG activity during the end of the swing and the knee flexors eccentric peak moment during running at maximum speed. It was also noted that subjects who produced the greatest amount of horizontal force were both able to highly activate their hamstring muscles just before ground contact and present high eccentric hamstring peak moment capability (Morin et al., 2015). Therefore, because great limb velocities prior to foot ground impact occur during sprinting, this swing-stance transition moment is of crucial importance for hamstrings as these muscle group support forces as high as eight times body weight (Sun et al., 2015).

On the other hand, to determine the magnitude of another crucial component among sports injuries Hayes et al. (2004) investigated the effects of fatigue on knee angle at contact and maximal knee flexion during stance in sub-maximal running in which participants increased flexion, a negative correlation was found between local muscle resistance of flexors and extensors of the hip and the kinematic changed in vVO_{2max} (Hayes et al., 2004). Results indicated that less kinematic alteration occurs during the run and athletes with higher local muscle resistance are able to keep their running kinematic stable. Besides, Horita et al. (1999) claimed that athletes must perform a higher muscle workload to be able to provide stretching-shortening cycle at a given running velocity during the propulsion phase resulted in higher fatigue progression (Horita et al., 1999). Consequently, athletes must perform an extra effort during T_{Lim} at vVO_{2max} unlike their regular performance routines and this excessive angular difference during constant loads may decrease fatigue resistance.

When the muscular involvement and biomechanical changes during running into consideration, branches such as soccer and track and field require an improved knee flexor muscle strength since eccentric muscle strength plays an essential role to maintain running performance at which vVO_{2max} intensity (Figure 1b). Hayes et al. (2004) found statistically significant negative correlations between stride length and eccentric knee flexion (KF_{ecc}), ($r = -0.957$) during a run to exhaustion at vVO_{2max} (Hayes et al., 2004). They reported that there is a strong relationship between local muscular endurance of the knee flexors with changes in running mechanics. It has been found in another study that the repeated transient impact of vertical ground reaction force causes an abrupt collision force equal to about 1.5- to 3-fold the body weight during running (Lieberman et al., 2010). Furthermore, Arampatzis et al. (1999) reported an increased mechanical power at the knee joint and muscles to be exposed to a heavier load with the increase in velocity (Arampatzis et al., 1999). Taking such repeated impacts into account it could be speculated that soccer and track and field athletes with higher local muscle resistance may have been fewer changes in kinematic variables during the run and enabled them to cope with lower limb muscle fatigue and prolonged time to exhaustion at their vVO_{2max} (Table 2). This findings may arise a question whether the outcome data used to determine H/Q ratio derived under fatigued conditions present

consistency and reproducibility as power output in the lower extremity muscles especially in quadriceps and hamstring group muscles would decrease proportionally as a result of the fatigue due to the exhaustive exercise such as T_{Lim} at vVO_{2max} . However, Coombs et al., 2005 observed no predictive potential of the test when performed during eccentric actions or at higher joint angular velocities which calls into question the predictive potential of several versions of the test (Coombs and Garbutt, 2002). On the other hand, the results of the study indicates that evaluation of H/Q ratio subsequent to time to exhaustion test offers a different insight into measured data following testing modalities which involves increased activation of knee flexor muscles (Figure 3a,b,c,d). Additionally, as shown in Figure 1(a) and Figure 1(b), soccer and track and field athletes showed greater VO_{2max} and vVO_{2max} and also increased H/Q discrepancy compared to other branches (Figure 3a,b). The significant asymmetries calculated through $H/Q_{Fatigue}$ intervention clearly indicates the heterogeneity of the data when H/Q evaluated under exact fatigue conditions. Based on the results of a research, the percent decline in H/Q_{CR} and fatigue index from repetitions 28-30 were found significant and positive for the knee flexor moments while no significant correlations were observed for the changes in H/Q^{CR} or FI of the knee extensors in repetitions 28 and 30. These findings indicate that knee extensor FI had a weak (or no) effect on the reductions in H/Q_{CR} and the subjects who had a greater percent decline in H/Q_{CR} also tended to demonstrate a greater fatigue in the knee flexors (Pinto et al., 2018). We found in our study that the magnitude and reproducibility of flexor muscles during T_{Lim} at VO_{2max} was dominant even under fatigued conditions when we compared the individual repetitions between 48-50 to maximum joint moment during fatigue test ICC was found 0.97 (95% CI, 0.88–0.99), 0.94 (95% CI, 0.77–0.99), and 0.90 (95% CI, 0.60–0.98) for knee flexor muscles for between the repetitions 48 to 50, respectively (Table 4). Together with this findings it could be asserted that knee flexor fatigue may better explain the reductions in H/Q_{CR} during a fatigue test than knee extensor fatigue. In accordance with this findings, the results of the current study indicates that the evaluation of the neuromuscular determinants such as fatigue index, H/Q ratio following exercise intensities where anaerobic metabolism prevails are also affected by fitness deficiency, and a poor aerobic and anaerobic capacity. These results clearly showed that H/Q ratio provides distinct data regarding actual neuromuscular performance or risk of injury if H/Q is determined after a fatigue test compared to other standard techniques (Table 3). On the other hand, the results of the frequency analysis indicated that all participants reached their maximal moment at different repetition. Only a 6% of the participants reached their maximum joint moment at their first repetition while 9% reached at 3th, 21% at 5th, 25% at 6th and 39% at their 8th repetition (Table 5). It could be thus speculated that the wide range of intra-subject variability of initiation and maintenance of force production during the test which can affect the calculation of fatigue index as well as the changes in joint moment and H/Q ratio during the fatigue test. In terms of a precise evaluation, H/Q ratio and fatigue index rely mainly on the selection of the method to calculate the fatigue index such as fatigue rate, type of the exercise, and intensity. Thus, due to many factors which could possibly affect the observed data are being taken into account during the evaluation of neuromuscular performance this new approach may provide perhaps more precise information than other conventional methods while fatigue is an important factor in the evaluation of isokinetic testing performance, and consequently on the H/Q measurements.

CONCLUSION AND RECOMMENDATIONS

The results of this new proof-of-concept method and traditional equations demonstrated significant overall differences with respect to H/Q ratio and fatigue index under fatigued conditions in the functioning of the exercising leg muscles in the present study. This novel information may provide different information to organize exercise programs and form individual goals, to monitor improvements of athletes and to set training and testing regimes and enable them to control

whether these measurements are indeed helping them to achieve their goals. To this end, it could be speculated that the assessment of H/Q ratio and fatigue index parameters under non-fatigued conditions may not mimic the actual muscular performance of the athletes and leads to an imprecise evaluation of fatigue index and hamstring to quadriceps ratio. With the light of the findings of this study, it is essential to highlight the need of integration of maximum strength training into programs to improve aerobic and anaerobic capacity as participants indicated higher H/Q ratios and increased fatigue indexes a result of fatigue. More studies should be conducted to detect differences among different branches, which can provide a great deal of advantages in comparing physiological parameters and their effects on H/Q ratio and fatigue mechanism.

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