Effect of body composition on aerobic capacity of animals submitted to swimming exercise

Efeitos da composição corporal na capacidade aeróbia de animais submetidos ao exercício de natação

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Abstract – The aim of this study was to verify the influence of body composition on the C_{AER} of animals submitted to different types of physical training. Twenty-three Wistar rats were divided into groups according to the training protocol: control (CTLE); aerobic training (TAE); anaerobic training (TAN); and concurrent physical training (TCc). The critical load test (CC) was carried out for determining the anaerobic threshold at the beginning and end of the training period. TAE was composed of swimming exercise with intensity corresponding to 70% of the anaerobic threshold; TAN was composed of water jumps with overload of 50% of the body weight of each animal; and TCc was composed of a combination of aerobic and anaerobic protocols described above. After four weeks of training, the weight of epididymal adipose tissue and soleus, plantar and gastrocnemius muscles was collected and measured. Dispersion analysis was used to draw graphics and analysis of the simple linear regression model was performed to identify the influence of each variable on C_{AER} . Body Fat demonstrated positive influence of 5% on the relationship with the Anaerobic Threshold. Soleus and gastrocnemius muscles showed negative influence of 13.1% and 37.0%, respectively. The plantaris muscle showed no influence on the Anaerobic Threshold. Body composition influenced the C_{AER} of the animals analyzed. Moreover, it was verified that body fat favored floatability, while higher muscle mass impaired it.

Key words: Anaerobic threshold; Rats; Adipose tissue.

Resumo – Objetivou-se verificar a influência da composição corporal na C_{AER} de animais submetidos a diferentes tipos de treino físico. Foram utilizados 23 ratos Wistar, divididos em grupos de acordo com o protocolo de treino: controle (CTLE); treino aeróbio (TAE); treino anaeróbio (TAN); e treino físico concorrente (TCc). Foi realizado o Teste de carga crítica (CC) para a determinação do limiar anaeróbio no início e no final do período de treino. O TAE foi composto por exercício de natação com intensidade correspondente a 70% do Limiar Anaeróbio; e o TAN por meio de saltos aquáticos com sobrecarga de 50% do peso corporal de cada animal; e o TCc pela combinação dos protocolos aeróbio e anaeróbio, descritos anteriormente. Após quatro semanas de treino, foi coletado e mensurado o peso do tecido adiposo epididimal e dos músculos sóleo, plantar e gastrocnêmio. Foi utilizada a análise de dispersão para a elaboração e gráficos e a análise do modelo de regressão linear simples para identificar a interferência de cada uma das variáveis na C_{AER} . A Gordura Corporal demonstrou influência positiva de 5% na relação com o Limiar Anaeróbio. Os músculos sóleo e gastrocnêmio mostraram influência negativa de 13,1% e 37,0%, respectivamente. Enquanto o músculo plantar não mostrou qualquer influência no limiar anaeróbio. A composição corporal influenciou na C_{AER} dos animais analisados. Além disso, notou-se que a gordura corporal favoreceu a flutuabilidade, enquanto a maior massa muscular a prejudicou.

Palavras-chave: Limiar anaeróbio; Ratos; Tecido adiposo.

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INTRODUCTION

The evaluation of the aerobic capacity (C_{AER}) through the maximum steady state blood lactate test (MFEL) is considered the gold standard¹. In order to minimize costs and test interference in the training period, Chimin et al.², have developed a test based on MFEL to estimate C_{AER} called critical workload (CCT). However, when exercise was conducted in the aquatic environment, C_{AER} can be influenced by muscle density and / or body fat accumulation^{3,4}.

Studies by Voltarelli et al.³ corroborate the influence of body composition on physical performance. By analyzing individual C_{AER} in swimming obese female rats, they found that presumably for the same relative exercise intensity, obese animals performed less effort compared to the control group. For determining MFEL in normal and obese rats of both sexes, Araujo et al.⁴ identified MFEL value at 6% of body weight in both obese groups, while in the female control group, this value was around 5% by weight body. Thus, it is suggested that the variation in body composition by increasing body fat may facilitate the floatability capacity of obese rats during swimming exercise.

Another important variable on C_{AER} is the training specificity. While some exercise modalities seem to contribute to performance gain, other protocols may not be effective, especially when performed on the ground or on the liquid medium. Voltarelli et al.³ found that MFEL presented different C_{AER} values when performed on a liquid medium and on a treadmill, suggesting that MFEL is dependent on the type of exercise used.

Thus, the way the exercise protocol is used and body composition seemed to be the variables that affect the C_{AER} values. Therefore, the development of methods aimed at identifying and reducing the influence of this bias is essential for the proper assessment of the physical performance of animals. Thus, the aim of this study was to investigate the influence of body composition and the use of different exercise models performed on the liquid medium on the C_{AER} of Wistar rats.

METHODOLOGICAL PROCEDURES

Animals

In this study, 23 adult male Wistar rats (90 days) with average weight of 411.0 ± 40.7 grams were used, obtained from the Animal Facility of UNESP, Botucatu - SP and kept in the Animal Facility for small rodent of the Department of Physiotherapy of FCT UNESP, Presidente Prudente Campus - SP. Animals were kept in groups of four animals per cage (polyethylene) with controlled ambient temperature ($22 \pm 2^\circ\text{C}$) and brightness (light / dark cycle of twelve hours), with free access to water and food (standard rodent chow). The research was conducted according to rules and ethical principles of animal experimentation after approval by the Ethics Research Committee of FCT- UNESP Presidente Prudente (002/2011).

EXPERIMENTAL GROUPS

Animals were divided into groups according to the training protocol as control (CTLE, $n = 6$); aerobic training (TAE $n = 6$), anaerobic training (TAN $n = 6$); and concurrent physical training (TCc, $n = 5$).

- Control group: animals remained free in their cages, with unrestricted access to water and food.
- Aerobic Training group: composed of swimming sessions lasting 30 minutes, three times a week, in appropriate tanks, divided by PVC cylinders for the individualization of stalls, so that each animal was individually trained. Intensity was equivalent to 70% of the anaerobic threshold (Lan), set by the Critical Workload Test (CC).
- Anaerobic Training Group: composed of four sets of 10 jumps three times per week in a cylindrical PVC container, especially modified for jumps in water (50 cm long) of depth appropriate to the size of animals (38 cm deep). A 1-minute interval was allowed between each series of jumps. The overload used corresponded to 50% of the body weight of each animal adapted in the anterior thorax region through a vest.
- Concurrent Physical Training Group: composed of a combination of both physical training models mentioned above. Exercise sessions were conducted in sequence (aerobic and anaerobic training) without break from one to the other, three times a week, comprising 30 minutes of swimming (load defined at 70% Lan) and four series of 10 jumps, overload of 50% of the body weight of each animal.

Critical load test (CC) and anaerobic threshold determination

The determination of C_{AER} by means of the critical load (CC) was obtained by inducing the exercise in four randomized stimuli and loads corresponding to 7, 9, 11 and 13% of body weight (PC), so that all animals performed four efforts and reached exhaustion between two and ten minutes, as proposed by Hill ⁵ and used by Machado et al.⁶.

The exercise performance time was controlled with the use of a specific timer (TIMEX®, model 85103). Every effort was performed with an interval of 24 hours, according to method proposed by Marangon et al.⁷ and reproduced by Chimin et al.². Then, linear regression was performed between load and the inverse of the time for each effort ($1 / t_{lim}$), from which the equation to check the value corresponding to C_{AER} was generated.

Material collection and preparation

Animals were euthanized by overdose of ketamine hydrochloride and xylazine intraperitoneally association (8), according to the ethical principles in animal research. The epididymal adipose tissue (TecAdp) and the soleus, plantar and gastrocnemius muscles of the right hind limb were collected. Samples were weighed on a precision scale immediately after collection.

Epididymal adipose tissue

The TecAdp analysis was performed by percentage relative to the total weight of the animal using the following formula: $\{[\text{TecAdp (g)} \times 100] / \text{weight (g)}\}$. This procedure was established as a way of correcting the difference in body weight among animals (9).

Statistical analysis

Data obtained were used in the drawn of dispersion graphs involving variables inserted two by two. In each graph, Lowess smoothing curves were plotted (locally weighted scatter plot smooth) to view the trend of points. To identify the influence of muscle weight and fat tissue on the aerobic capacity, as well as the effect of different training protocols on the physical performance of animals, simple linear regression procedures were used, with respective R^2 values. All calculations used the SPSS 17.0 statistical package.

RESULTS

Figure 1 shows the data dispersion according to each of the variables analyzed. Furthermore, it was possible to identify the relationship between values corresponding to variables together.

Figure 1. Influence of body composition on the aerobic capacity [N = 23]. Lines show the average values of each animal. Points identify the real value presented by the animal (no correction by the average). Lan: Anaerobic Threshold. SOLCOR: Soleus Muscle Mass corrected by the animal's weight. FATCOR: Visceral Fat Mass corrected by the weight of each animal. PLANTCOR: Plantaris Muscle Mass corrected by the weight of each animal. GASTCOR: Gastrocnemius Muscle Mass corrected by the weight of each animal.

By observing the relationship between Anaerobic Threshold and Body Fat (Lan vs Corrected Fat) it was observed that the latter has an influence factor of 5% ($R^2 = 5\%$) on C_{AER} . I.e., the larger the animal's fat accumulation, the better its C_{AER} performance in swimming exercises (Table 1).

When observing the influence factor of the soleus muscle on C_{AER} , it was found that this muscle has a negative influence on the density / performance relationship by 13% ($R^2 = 13.1$). Thus, soleus does not seem to be the main muscle acting in the swimming exercise (Table 1). As the plantar muscle, which showed the lowest interference on C_{AER} after isolated verification ($R^2 = 0$). It was also observed that the line is not related to the density values and Lan obtained for each animal after adjusted analysis (Table 1). However, it was found that the negative influence of density of the gastrocnemius muscle on the aerobic performance of animals was higher when compared with other variables (33.7%) (Table 1).

Table 1. Variable anaerobic threshold response as a function of the following predictor variables visceral fat mass, soleus muscle mass, plantaris muscle mass, gastrocnemius muscle mass

Variable	R-sq	Regression line
FATCOR	05.0%	LANCOR = 6.241 - 27.39 FATCOR
SOLCOR	13.1%	LANCOR = 7.222 - 2746 SOLCOR
PLANTCOR	00.0%	LANCOR = 5.571 - 86 PLANTCOR
GASTCOR	37.0%	LANCOR = 12.60 - 1200 GASTCOR

Lan: Anaerobic Threshold. **SOLCOR:** Soleus Muscle Mass corrected by the animal's weight. **FATCOR:** Visceral Fat Mass corrected by the weight of each animal. **PLANTCOR:** Plantaris Muscle Mass corrected by the weight of each animal. **GASTCOR:** Gastrocnemius Muscle Mass corrected by the weight of each animal. R-Sq = R^2 value.

Examining the influence of each treatment on C_{AER} , it was found that the greater the muscle mass, the smaller the Lan value. Probably, increasing muscle mass generated by training models negatively influenced floatability, resulting in decreased performance (Table 2).

Table 2. Variable response anaerobic threshold as function of depending on the predictors variables and grouped by training.

Variable	Group	Regression line	R-sq
FATCOR	Ctle	LANCOR=3.541+9.390*FATCOR	00.4%
	Tae	LANCOR=5.819+14.500*FATCOR	05.0%
	Tan	LANCOR=2.558+127.600*FATCOR	61.0%
	Tcc	LANCOR=5.337+29.040*FATCOR	65.9%
SOLCOR	Ctle	LANCOR=4.239-500*SOLCOR	00.7%
	Tae	LANCOR=7.818-2356*SOLCOR	71.9%
	Tan	LANCOR=11.78-9594*SOLCOR	59.2%
	Tcc	LANCOR=8.677-4738*SOLCOR	22.6%
PLANTCOR	Ctle	LANCOR=1.311+2186*PLANTCOR	20.8%
	Tae	LANCOR=7.110-817*PLANTCOR	06.7%
	Tan	LANCOR=7.346-1465*PLANTCOR	03.5%
	Tcc	LANCOR=7.349-1048*PLANTCOR	23.1%
GASTCOR	Ctle	LANCOR=13.08-1425*GASTCOR	67.7%
	Tae	LANCOR=7.424-208.6*GASTCOR	06.6%
	Tan	LANCOR=11.10-953*GASTCOR	15.6%
	Tcc	LANCOR=11.52-943.1*GASTCOR	29.0%

Lan: Anaerobic Threshold. **SOLCOR:** Soleus Muscle Mass corrected by the animal's weight. **FATCOR:** Visceral Fat Mass corrected by the weight of each animal. **PLANTCOR:** Plantaris Muscle Mass corrected by the weight of each animal. **GASTCOR:** Gastrocnemius Muscle Mass corrected by the weight of each animal. CTLE: control; TAE: aerobic training; TAN: anaerobic training; TCC: concurrent training.

DISCUSSION

The aim of this study was to observe the physical performance of Wistar rats in liquid medium based on body composition. It was found that animals that had greater mass in the Gastrocnemius and Soleus muscles showed lower anaerobic threshold values, and therefore lower C_{AER} . This can be explained by the decreased floatability, making it harmful to the swimming mechanics.

The finding in this study corroborates results reported by Araujo et al.⁴, who when evaluating obese animals, found that they had higher C_{AER} values. In this sense, the inclusion of body composition values when LAN is used as reference for performance is necessary.

It was observed that when evaluating animals with higher fat percentage, they show better performance due to their better floatability. This can lead to false positive results since it overestimates the real anaerobic threshold value, and therefore, C_{AER} .

The effect of different types of exercises was previously described by Voltarelli et al.³. The authors found that C_{AER} can be ergometer dependent. That is, depending on the exercise model used, the animal may have different anaerobic threshold values.

In the present study, exercise models in liquid medium were used with the aim of obtaining the degree of interference of body fat and muscle density. As the swimming exercise suffers interference from animal floatability, lower performance of animals showing higher muscle mass was found, which was specifically attributed to TAN and TCc.

It is likely that the use of a different ergometer could favor animals with greater muscle mass. In this case, a protocol using treadmill could be able to provide better performance, since it influences floatability. Moreover, in the latter case, the favoring factor of body fat in liquid medium could be an aggravating factor in treadmill exercise or other protocol carried out in dry conditions.

According to Araujo et al.⁴, protocols using animal models have gained prominence in the area of exercise physiology. However, there is still lack of physical testing protocols to simulate effort so that it can be adapted or extrapolated to human beings¹⁰. Thus, the accurate determination of C_{AER} becomes substantial for conducting animal studies.

The findings of this study may serve as a parameter for the prescription of exercise intensity in studies using animals in swimming exercises. Identifying the effect of a particular variable on floatability and performance can minimize the favoring of certain animals, particularly if they are submitted to exercise protocols that may result in increased muscle mass or hyper caloric diet, in this case, the increase in body fat percentage.

The study corroborates literature when identifying the influence of body composition on the C_{AER} of Wistar rats submitted to swimming exercise, based on the analysis of muscle mass and visceral fat percentage. However, this study is limited to identifying body composition after euthanasia.

Future studies will contribute to literature based on the development of measurement techniques to identify *in vivo* fat percentage. Thus, prescription can be made before the training period.

CONCLUSION

It was concluded that, when the performance of animals in the aquatic environment was evaluated, the increase in epididymal adipose tissue had a positive influence on the physical performance of animals and the increased muscle mass showed negative influence.

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