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Abstract. Research Objective: To discuss exercise performance and biochemical reactions of different load weight-bearing running of rats. Research Methods: 24 male 3-month-old SD rats were randomly divided into 3 groups (n = 8), namely, a none weight-bearing running group, a 35% maximum load weight-bearing running group, a 75% maximum load weight-bearing running group. After weight-bearing running training for 6 weeks, the maximum weight-bearing capacity, the blood lactic acid value and serum CK value of rats were tested and compared. Research Results: 30m/min speed running and 45m/min speed running respectively enabled the lactic acid value to generate first and second inflection points; weight-bearing running increased the serum CK-M value with the increase of bearing weight and load, and the blood lactic acid value and the serum CK value were significantly increased. Conclusion: 30m/min is the anaerobic threshold intensity of rat running; maximum load capacity of rats is about 65% of sole weight; weight-bearing running with appropriate load can not damage muscle fibers and can effectively mobilize Ⅱ type muscle fiber, 75% maximum load weight-bearing running can mobilize Ⅱ type muscle fiber to be fully raised, but can cause muscle fiber damage.

Keywords: weight-bearing running; exercise performance; quantification

1 Research Objective

Rat resistance training is internationally and widely adopted in biological adaptation mechanism of researching on strength training, the load running training still has many problems in experiments as an effective method of resistance training. Since weight-bearing running training comprises two exercise load modes of weight-bearing and running, the load effect of running is mainly to improve the functions of respiratory and circulatory system, it is also still effective in improving muscle endurance simultaneously, and weight-bearing training has the main value of developing muscle maximum strength and power endurance. In weight-bearing training, the size of load strength may have different influences on muscle functions

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and even different types of muscle fibers. Therefore, When training plans are formulated in rat weight-bearing run experiments, the quality and quantification features of running and gravity load must be made clear, simultaneously, the boundary among maximum weight-bearing ability, aerobic exercise and anaerobic exercise of rats should be comprehended. The experiment aims at discussing the exercise performance and biochemical reactions of running with different loads and bearing weights of 3-month-old rats, thereby providing quantitative basis of exercise load control for rat weight-bearing running experiments, and also providing reference for rat related exercise experiment in the future.

2 Materials and Methods

2.1 Experiment Object

24 clean male 3-month-old SD rats were provided by Animal Center of Jiangxi Medical College and were fed in Animal Room of Animal Center of Jiangxi Medical College. Breeding conditions comprised: free water and food feeding, natural lighting, room temperature of 18DEG C to 22 DEG C, humidity of 40% to 45%; normal feedstuff feeding, and the feedstuff was provided by Animal Center of Jiangxi Medical College.

2.2 Experimental Grouping

24 rats were randomly divided into 3 groups: a none weight-bearing running group, a 35% maximum load weight-bearing running group, a 75% maximum load weight-bearing running group with eight rats in each group.

2.3 Instruments and Reagent

(1) ZH-PT-type Animal Experiment Treadmill produced by Anhui Huaibei Zhenghua Bioinstrumentation Equipment Co, Ltd; (2) YSIl500 SPORT Blood Lactic Acid Analyzer produced by United States Gimcheon Instruments Inc; (3) TU-1810 UV-Vis Spectrophotometer produced by Beijing Persee Instrument Co., Ltd; (4) MP200A Type Electronic Balance produced by Shanghai Second Balance Instrument Factory;(5) Creatine Kinase Kit produced by Beijing Biosino Bio-technology and Science Inc; (6) Blood Lactic Acid Test Kit produced by Nanjing Jiancheng Biological Technology Co, Ltd.

2.4 Experimental Methods

(1) Rat maximum load capacity test: heavy objects were applied on waists and backs of rats (iron half-cylinder bodies with the diameters of 2cm and 0.6cm with the length of 3cm, the weights were 80 grams and 10 grams respectively), the weight was progressively increased, and the increase grades were 80 grams, 40 grams and 10 grams respectively. The weight which enabled rats to be incapable of walking was defined as the maximum load intensity in rats.

(2) Determination of anaerobic threshold in rats: the treadmill was inclined for 0 degree. 8 rats were made to run for 3min at the speed of 25m/min, tail blood was sampled at the moment's notice, 1min, 1.5min, 2min, 2.5min and 3min before and after running for blood lactic acid analysis, thereby measuring the time of rat muscle lactic acid and blood lactic acid to reach equilibrium. 8 rats were made to warm up for 2min at the speed of 10m/min, then, the rats were made to run without stop for 2min under gradual increasing load intensity of 25m/min, 30m/min, 35m/min, 40m/min, 45m/min, 50m/min, 55m/min and 60m/min, and the tail blood was immediately sampled at the end of each grade load respectively.

(3)Comparison of blood lactic acid values after weight-bearing and non weight-bearing running of rats: rats were respectively made to bear no weight, and to bear 75% maximum load bearing weight and 35% maximum load bearing weigh (n = 8) and were made to run for 2min at the speed of 25m/min, and blood was sampled in 1.5min after running for testing blood lactic acid values. Blood lactic acid tests adopted YSI1500 SPORT lactic acid analyzer, and testing process was operated according to kit instructions.

(4) Comparison of serum CK and CK-MB values of rats after exercise under different loads and bearing weights: rats were respectively made to bear no weight, and to bear 75% maximum load bearing weight and 35% maximum load bearing weigh (n = 8) and were made to run for 2min at the speed of 15m/min, the tail blood was sampled immediately after running for CK and CK-MB test. Test methods adopted electro-optic colorimetry and could be finished on TU-1810 UV-Vis spectrophotometer. Testing process was operated according to kit instructions.

2.5 Statistical Analysis

Measurement information and data were represented with mean ± standard deviation, data statistics was processed through adopting SPSS15.0 statistical software, and statistical methods adopted LSD multiple comparison method for analysis.

3 Results and Discussion

3.1 Experimental Results

(1) Maximum load capacity of rats. In the incremental load test, it could be obtained from table 1 that the even maximum load capacity of rats was 65% of sole weight through computing according to rat weight percentage.

Table 1. Rat Weight and Average Maximum Load Capacity (n = 8)

Group	Average BodyWeight (g）	Maximum Load Capacity (g)
None weight-bearing group	209±15.12	136.18±47.2
35% maximum load weight-bearing running group	235±9.13	154.18±10.3
75% maximum load weight-bearing running group	198±13.25	128.18±30.4

(2) Testing Result of Rat Anaerobic Threshold . The time for muscle lactic acid and blood lactic acid to achieve balance in the experimental test was 1.5 minutes. In order to carry out rat weight-bearing running training, changes of blood lactic acid of rats in the process of running with different intensities should be firstly comprehended, thereby effectively controlling aerobic and anaerobic components in rat weight-bearing running training. In the continuous run process, with the increase in the intensity of running, anaerobic components of energy metabolism was gradually increased, showing an increase in muscle lactic acid production. Gravity load also changed energy metabolism forms in the muscle work process. In order to distinguish causes of generating lactic acid in rat weight-bearing running training, the effect direction of load intensity of running should be comprehended, thereby determining the influence of loading on muscle energy metabolism in rats. The experiment results showed that the blood lactic acid of rats was gradually increased with the increase of load intensity, the inflection point appeared in the speed point of 30m/min. When the speed was increased to 45m/min, the second inflection point of blood lactic acid increase appeared. The first inflection point appeared between running speed of 30m/min and 35m/min, blood lactic acid was increased by 1.46mmol / L and increased very significantly. The second inflection point appeared between the running speed of 45m/min and 50m/min, blood lactic acid was increased by 1.80 mmol / L, and the increase extend was further enlarged. In Table 2, Treadmill the first inflection point 30m/min speed of blood lactic acid changes in rat increasing load running could be defined as rat anaerobic threshold speed.

Table2. Blood Lactic Acid Values during Rat Increasing Load Running (n = 8)

Speed（m/s）	25	30	35	40
Blood Lactic Aci（mmol/L）	3.15±0.37	3.63±0.49	5.09±0.38	5.32±0.41
Speed（m/s）	45	50	55	60
Blood Lactic Acid（mmol/L）	5.79±0.64	7.59±0.33	8.17±0.69	8.58±0.47

(3) Blood Lactic Acid Value of Rats after Running with Different Loads and Bearing Weights and after Non-weight Bearing Running. In Table 3, the after-running blood lactic acid highest value was correspondingly increased with the increase of bearing weight and load of rats after 75% maximum load weight-bearing group (big load group), 35% maximum load weight-bearing group (small load group) and none weight-bearing group running. The small load weight-bearing group was averagely increased by 3.63mmol / L compared with none weight-bearing group. The big load weight-bearing group was averagely increased by 4.12mmol/L compared with small load weight-bearing group. It was indicated that running with different loads and bearing weights had extremely prominent influence on blood lactic acid values.

Table 3. Blood Lactic Acid Value of Rats after Running with Different Loads and Bearing Weights (n = 8)

Group	Blood Lactic AcidValue（mmol/L）	P Value
Big load group	11.09±2.09	* P< 0.05
Small load group	6.97±1.12	** P< 0.01
None weight-bearing group	3.34±0.43	*** P< 0.05

* Big load group was compared with the small load group; ** big load group was compared with the none weight-bearing group; *** small load group was compared with none weight-bearing group.

(4) Rat serum CK and CK-MB Value Testing Results of Running with Different Loads and Bearing Weights. From Table 4, it could be seen that with the increase in weight-bearing load, CK values were increased significantly, while the prominence level of differences among different groups was also increased. CK-MB was also increased, but the increase was not obvious, especially the changes between small weight-bearing group and the big weight-bearing group were not obvious, and there was no significant difference.

Table 4. Rat Serum CK and CK-MB Values (n = 8) of Running with Different Loads and Bearing Weights

Group	CK（U/L）	CK-MB（U/L）	P Value（CK）	P Value（CK-MB）
None weight-bearing group	150.65±18.39	27.89±35.78	*P< 0.05	*P< 0.05
Small load group	301.03±28.56	66.01±18.52	**P<0.01	**P> 0.05
Big load group	690.05±87.05	63.29±31.77		

* Different prominent levels between none weight-bearing group and the small load group; ** Different prominent levels between small load group and big load group.

3.2 Analysis and Discussion

(l) The average maximum load capacity of rats was about 65% of their sole weights, the determination standard treated the load that rats could essentially stand up for support and could simply walk as limit load, this movement mode did not always reflect the overall features of rat muscle function. In addition, the rat loading mode used in this study was to apply heavy objects on waists and backs of rats, load was dispersedly acted on rat's limbs and trunk, this load capacity embodied rat systemic comprehensive endurance. The results of this experiment only provided reference basis for load control of weight-bearing run training of 3-month-old rats, its change rules of comprehensive weight-bearing capacity with age needed further study.

(2) When rat lactic acid threshold speed was 30m/min and the speed achieved 45m/min, the increase in blood lactic acid generated the second inflection point. In this study, twice blood lactic acid inflection points in rats were results of twice changes of anaerobic metabolism process, the first inflection point marked that the anaerobic glycolysis functional systems began to be greatly employed, temporary equilibrium between energy consumption and supply occurred immediately after employ, with further increase of exercise intensity, the second inflection point occurred, which marked that the potential of body anaerobic glycolysis power supply system functions was further developed. Scientists often treated the time between the appearance of the second inflection point to the end of exercise as important indexes for measuring tolerance fatigue ability of athletes, and the second inflection point was used to control intensity of anaerobic capacity training of athletes.

Two inflection points obtained in this study were only inflection points of blood lactic acid curves, and had certain correspondence with two inflection points of exercise physiology states in human physiology, the appearance mechanisms of two inflection points may have certain same mechanisms in twice changes of physiological function states obtained in studies in human physiology, and it was concrete performance of body function capacity change on blood lactic acid value in rat exercise. The blood lactic acid change form of the experiment can provide reference data of exercise intensity control for future rat exercise experiments.

(3) The blood lactic acid highest value was increased with the increase of bearing weight and load after running at the speed of 25m/min under no bearing weight, small load bearing weight and big load bearing weight, and as the increase of load intensity, the increase extend of blood lactic acid had increasing trend. This study showed that when rats run under the speed of 25m/min, the rat body energy metabolism was basically in aerobic metabolism, after running, the blood lactic acid was little increased, thereby, the increase of blood lactic acid after bearing weight could be considered to be caused by changes in muscle working status. The blood lactic acid in the exercise process was mainly generated by anaerobic glycolysis in muscles, the contents and activities of amber mitochondrial malate dehydrogenase in different types of muscle fiber were different, the contents and activities in II-type fiber were higher than that of I type fiber, thereby it can be inferred that when the II type fiber was more used, the blood lactic acid contents after exercise was increased considerably. It could be further inferred that 75% load running can effectively mobilize II type muscle fiber recruitment.

In weight-bearing training, the effect directions of different load intensities were always important tissues of sports training study, according to features of different sports games, athletes need to develop different muscle strength qualities and develop different types of muscle fibers; athletes of speed and strength type need to develop II type muscle fibers, the neurons for dominating II type muscle fiber is large, excitation threshold is high, only high-intensity resistance load can effectively enable II type muscle fiber to be fully collected for participating in contraction. In this study, the blood lactic acid results of different load weight-bearing running trainings support this view. Namely, large weight-bearing load running more fully uses II type muscle fiber, the main form of II-type muscle fiber energy metabolism is glycolysis, and thus increase extent of blood lactic acid after running is higher than that of big load weight-bearing group and none weight-bearing group.

(4)75% maximum load weight-bearing running could result in a certain degree of damage in muscle fibers, while 35% maximum load weight-bearing running could not cause injury. Two kinds of maximum load weight-bearing running could not cause obvious damage of myocardial fibers.

In experimental studies in mammals, CK-M and CK-B in serum CK were frequently applied, CK-M and CK-B 22 were combined to form CK-MM, CK-MB and CK-BB, CK-MM was usually a skeleton muscle source, CK-MB was a cardiac source, whereas CK-BB was a brain source. In the studies, the activity of three isozymes was often used to compare with total CK activity to determine the skeletal muscle, heart muscles, or brain injury and its extent.

CK was commonly found in muscle cells, muscle fiber micro-injury may lead to increased permeability of fibrous membrane, and thereby CK could be very vulnerable to enter into the blood through the muscle cell membrane, so that CK was significantly increased. Field of sports medicine treated increased activity of CK and its isoenzymes as a sensitive indicator of muscle damage judgments, but in animal experiments, quantitative criteria to judge these indicators were still deficient. Su Quansheng [1] studied and found that when rat CK and CK-MM were above 409 U / L, the injury symptoms of muscle fibers were more obvious. Cui Yupeng [2] studied and found that when rat CK was up to 600U / L, the skeletal muscle damage and CK were significantly correlated. The results of this study were compared, it could be determined that the rat CK value reached 690U / L after weight-bearing running of 75% the maximum load, it was shown that the weight-bearing running under load intensity may cause certain damage to muscle fibers. However, CK value was 301U / L after weight-bearing running of 35% maximum load, it was shown that the load and weight-bearing running could not cause significant damage in skeletal muscle.

4 Summary

Through different load weight-bearing running experiments of 3-month-old SD rats on 0 degree slope treadmill, it is showed that: 30m/min speed running is the rat anaerobic threshold intensity; the rat maximum load capacity is about 65% of their sole weight. Load can prominently increase blood lactic acid of rats under aerobic running intensity, the weight-bearing running with appropriate load can not damage muscle fibers and can effectively mobilize Ⅱ type muscle fiber; 75% maximum load weight-bearing running makes II muscle fiber to be more fully raised, but may cause minor damage to the muscle fibers.

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