

HEMOGLOBIN ANALYSIS AFTER OVERLOAD TRAINING IN ATHLETES



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ANÁLISE DE HEMOGLOBINA APÓS TREINAMENTO DE SOBRECARGA EM ATLETAS

ANÁLISIS DE LA HEMOGLOBINA TRAS EL ENTRENAMIENTO DE SOBRECARGA EN ATLETAS

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ABSTRACT

Introduction: The cardiovascular system provides athletes with the proper conditions for blood circulation, ensuring the stability and normal metabolism of the body's internal environment during exercise. **Objective:** Investigate the effect of overload training on the hemoglobin of male taekwondo athletes. **Methods:** Twenty-one male taekwondo athletes (level 2 or higher) were selected and trained for four weeks, five days per week, with an initial load intensity of 60% of the maximum heart rate and a weekly intensity increase of 10%. Before training and on every weekend during training, hemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), reticulocyte count (Ret) were checked, RBC volume distribution width (RDW), hemoglobin content distribution width (HDW), mean reticulocyte volume (MCVr), mean reticulocyte hemoglobin concentration (CHCMr), serum iron (Fe) and ferritin (Fer). **Results:** After four weeks of increasing load training, athletes showed a progressive and significant decrease in Hb ($P < 0.01$), manifested as exercise-induced hypohemoglobin, and MCV, MCH, MCHC, CHCMr, HDW, and serum ferritin were significantly or extremely decreased ($P < 0.05$, $P < 0.01$); RDW increased significantly ($P < 0.05$); Changes in Ret and serum iron showed no statistical significance ($P > 0.05$). Correlation analysis found that hemoglobin concentration had the highest correlation with MCHC and CHCMr. **Conclusion:** Four-week incremental load training can induce exercise-induced hypohemoglobin in male taekwondo athletes, its changes being most correlated with MCHC and CHCMr, but without significant correlation with serum Fe, RDW, HDW, and MCV. **Level of Evidence: Therapeutic Studies - Investigation of Outcomes.**

Keywords: Athletes; Hemoglobins; Reticulocyte Count; Exercise.

RESUMO

Introdução: O sistema cardiovascular fornece aos atletas as condições propícias para a circulação sanguínea, garantindo a estabilidade e o metabolismo normal do ambiente interno corporal durante o exercício. **Objetivo:** Investigar o efeito do treinamento em sobrecarga sobre a hemoglobina dos atletas masculinos de Taekwondo. **Métodos:** Vinte e um atletas masculinos de taekwondo (nível 2 ou superior) foram selecionados e treinados durante 4 semanas, 5 dias por semana, com uma intensidade de carga inicial de 60% da frequência cardíaca máxima e um aumento de intensidade semanal de 10%. Antes do treinamento e em todos os fins de semana durante o treinamento, foi verificada a hemoglobina (Hb), volume corpuscular médio (MCV), hemoglobina corpuscular média (MCH), concentração corpuscular média de hemoglobina (MCHC), contagem de reticulócitos (Ret), largura de distribuição do volume de hemácias (RDW), largura de distribuição do conteúdo de hemoglobina (HDW), volume médio de reticulócitos (MCVr), concentração média de hemoglobina reticulócitos (CHCMr), ferro sérico (Fe) e ferritina (Fer). **Resultados:** Após 4 semanas em treinamento de carga crescente, os atletas mostraram uma diminuição progressiva e significativa em Hb ($P < 0,01$), manifestada como hipohemoglobina induzida por exercício, e MCV, MCH, MCHC, CHCMr, HDW, e ferritina sérica foram significativamente ou extremamente diminuídos ($P < 0,05$, $P < 0,01$); RDW aumentou significativamente ($P < 0,05$); Alterações de Ret e ferro sérico não apresentaram significância estatística ($P > 0,05$). A análise de correlação constatou que a concentração de hemoglobina teve a maior correlação com MCHC e CHCMr. **Conclusão:** O treinamento de carga incremental de quatro semanas pode induzir hipohemoglobina induzida por exercício em atletas taekwondo masculinos, sendo suas alterações mais correlacionadas com MCHC e CHCMr, mas sem correlação significativa com Fe sérico, RDW, HDW e MCV. **Nível de evidência: Estudos Terapêuticos - Investigação dos Resultados.**

Descritores: Atletas; Hemoglobinas; Contagem de Reticulócitos; Exercício Físico.

RESUMEN

Introducción: El sistema cardiovascular proporciona a los deportistas las condiciones propicias para la circulación sanguínea, garantizando la estabilidad y el metabolismo normal del medio corporal interno durante el ejercicio. **Objetivo:** Investigar el efecto del entrenamiento de sobrecarga en la hemoglobina de los atletas masculinos de taekwondo. **Métodos:** Se seleccionaron 21 atletas masculinos de taekwondo (de nivel 2 o superior) y se entrenaron durante 4 semanas, 5 días a la semana, con una intensidad de carga inicial del 60% de la frecuencia cardíaca máxima y un aumento semanal de la intensidad del 10%. Antes del entrenamiento y cada fin de semana durante el mismo, se comprobó la hemoglobina (Hb), el volumen corpuscular medio (MCV), la hemoglobina corpuscular media



(HCM), la concentración de hemoglobina corpuscular media (MCH) y el recuento de reticulocitos (Ret), Anchura de distribución del volumen de glóbulos rojos (RDW), anchura de distribución del contenido de hemoglobina (HDW), volumen reticulocitario medio (MCVr), concentración media de hemoglobina reticulocitaria (CHCMr), hierro sérico (Fe) y ferritina (Fer). Resultados: Después de 4 semanas de entrenamiento con carga creciente, los atletas mostraron una disminución progresiva y significativa de la Hb ($P < 0,01$), que se manifestó como hipohemoglobina inducida por el ejercicio, y el MCV, el MCH, el MCHC, el CHCMr, el HDW y la ferritina sérica disminuyeron de forma significativa o extrema ($P < 0,01$). 05, $P < 0,01$); el ADE aumentó significativamente ($P < 0,05$); los cambios en el Ret y el hierro sérico no mostraron significación estadística ($P > 0,05$). El análisis de correlación encontró que la concentración de hemoglobina tenía la mayor correlación con MCHC y CHCMr. Conclusión: El entrenamiento de carga incremental de cuatro semanas puede inducir hipohemoglobina inducida por el ejercicio en atletas masculinos de taekwondo, siendo sus cambios los más correlacionados con MCHC y CHCMr, pero sin correlación significativa con el Fe sérico, RDW, HDW y MCV. **Nivel de evidencia: Estudios terapéuticos - Investigación de resultados.**

Descriptores: Atletas; Hemoglobinas; Recuento de Reticulocitos; Ejercicio Físico.

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INTRODUCTION

Taekwondo is an official Olympic competition in the combat category. It has the characteristics of fierce confrontation and fast transition between attack and defense. The energy supply method of athletes in the competition is characterized by alternating aerobic and anaerobic metabolism. Oxygen metabolism also requires athletes to have a better level of aerobic metabolism in the stalemate stage. During the exercise of Taekwondo, the cardiovascular system provides the athletes with the power and pipeline for blood circulation, which ensures the relative stability and normal metabolism of the body's internal environment during exercise.¹⁻³

The pathogenesis of cardiovascular events during exercise is often related to the underlying cardiac disease and the effects of exercise on the heart. According to the World Health Organization, cardiovascular disease has become the leading cause of death in middle- and high-income countries. Exercise can effectively reduce the risk of cardiovascular disease, but vigorous exercise may also increase a series of cardiovascular risks, such as acute sudden cardiac death. Exercise causes a series of physiological changes, such as hyperthermia, dehydration, acidosis, electrolyte disturbance, etc., which may induce ventricular arrhythmias, which in turn lead to exercise-induced sudden death. Studies have shown that professional athletes are twice as likely to experience acute cardiovascular events when participating in sports as non-professional athletes. Exercise-induced sudden death is the most concerned and the most serious type of sports accident among exercise-induced cardiovascular events.^{4,5}

In recent decades, more and more studies have been conducted on the physical and physiological characteristics of taekwondo athletes, which have their own unique physical and physiological characteristics. However, there is no experimental study on the evaluation of the hemoglobin of Taekwondo athletes in high-intensity exercise. This study evaluates the hemoglobin of Taekwondo athletes in the combat category and finds out that the sports-type hemoglobin of Taekwondo athletes is reduced as soon as possible, which can not only effectively reduce the probability of danger in the process of special sports, but also comprehensively evaluate the effect of long-term training in Taekwondo on athletes. The adaptive changes brought about by physiology maximize the benefits of exercise training while avoiding risk factors. In addition, the main technique of taekwondo is only for the movements of legs, and there is a similar routine "quality" in practice, so through the effective evaluation of the hemoglobin of taekwondo athletes and the beneficial adaptation changes, it can provide suggestions for the normal people and the people who have suffered from low dynamic hemoglobin.^{6,7}

OBJECTS AND METHODS

Research objects

Twenty-one male taekwondo athletes (level 2 or above) were selected, aged 21-25 years, with a height of 175.40 ± 5.12 cm and a weight of 77.22 ± 13.07 kg. Before the experiment, the subjects had undergone strict health checks and were in good health. The subjects read the informed consent form before the experiment to understand the whole experimental situation, and voluntarily signed the informed consent form to conduct the experiment.

The authors state that the research was conducted in accordance with the principles embodied in the Declaration of Helsinki and in accordance with local statutory requirements.

The authors state that the participants have given written informed consent to participate in the study.

The authors confirming that consent was given for publication by all participants

Research methods

The experiment lasted for 4 weeks. In the first week, 60% HRmax (HRmax = $220 - \text{age}$) was used as the initial training intensity, and thereafter the intensity was increased by 10% HRmax every week. During the training process, the subjects were all equipped with a Polar heart rate monitor to monitor the heart rate to control the intensity. The training is carried out in the Taekwondo training hall in the gymnasium of our city. The training time is from 6:00 to 7:30 in the morning from Monday to Friday. The 4-week incremental load training schedule is shown in Table 1.

Observation indicators and methods

Before the experiment and during the training, every Saturday morning, the subjects took venous blood on an empty stomach, anticoagulated with EDTA, and tested the hemoglobin concentration (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin content (MCH), mean corpuscular hemoglobin concentration (MCHC), reticulocyte count (Ret), red blood cell volume distribution width (RDW), hemoglobin content distribution width (HDW), mean reticulocyte volume (MCVr),

Table 1. 4-Week incremental load-intensity training schedule.

	Exercise intensity	Exercise time (min/times)	Exercise times	Intermittent time (min)
Week 1	60%HRmax	30	2	5
Week 2	70%HRmax	20	3	5
Week 3	80%HRmax	12	4	5
Week 4	90%HRmax	8	6	5

mean reticulocyte hemoglobin concentration (CHCMr). Part of the blood was centrifuged to separate serum to test serum iron (Fe) and ferritin (Fer). Jinan Xisumekang Medical Electronics Co., Ltd. provided kits. Hitachi 7020 automatic biochemical analyzer and Ferene method were used to test serum iron. Bechman Coulter Access Immunoassay system and its companion kits test for ferritin.

Statistical analysis

All data were analyzed by SPSS 24.0, and the experimental data were expressed as mean ± standard deviation. Paired sample t test, Pearson correlation analysis and cluster analysis between indicators were carried out. The significance level was $P < 0.05$, and the extremely significant level was $P < 0.05$. 0.01.

RESULTS

Changes in indicators related to hemoglobin and red blood cells

Table 2 shows that during the 4-week incremental load training, the hemoglobin concentration of the subjects decreased week by week, and there was a significant difference between each week and before exercise ($P < 0.01$). At the end of the 4th week, 86.7% of the subjects had hemoglobin levels. The decrease was more than 10%, indicating that the incremental load training in this experiment could induce exercise-induced hypohemoglobin in subjects.⁸

The MCV of the subjects was significantly higher than that before exercise at the 1st and 2nd weekends (both $P < 0.01$), and then decreased to the pre-exercise level; < 0.05 , $P < 0.01$); MCHC decreased significantly, then increased, and then decreased, and the MCHC was significantly lower than that before exercise during the whole experimental period ($P < 0.01$); RDW showed an increasing trend, and it increased significantly at the 4th weekend, and the differences were statistically significant compared with those before exercise, the 1st weekend, and the 2nd weekend ($P < 0.01$); HDW showed a decreasing trend, and the difference was statistically significant compared with that before exercise ($P < 0.05$); In the experiment, Fer decreased significantly ($P < 0.05$), and reached the lowest point at the end of the 4th week; Fe and Ret changed in the whole training process, but there was no statistical significance ($P > 0.05$); CHCMr decreased significantly ($P < 0.05$) during the 4-week training period.

Correlation analysis and cluster analysis

It can be seen from Table 3 that MCH, MCHC, Ret, Fer, MCVr and CHCMr all have a certain correlation with hemoglobin, among which MCHC and CHCMr have the largest correlation coefficient with hemoglobin, reaching a moderate correlation ($P < 0.01$), while serum Fe, CHCMr and hemoglobin have the largest correlation coefficient. RDW, HDW and MCV had no significant correlation with hemoglobin, meanwhile, HDW

had certain correlation with MCH, MCHC, Ret, MCVr ($P < 0.01$), and MCV also had certain correlation with MCH and MCVr ($P < 0.01$); There was no significant correlation between serum Fe, RDW and hemoglobin concentration and other related indexes.^{9,10}

Using the dendrogram of the average connection (between groups) to readjust the distance clustering and merging, the analysis results show that if the 10 red blood cell-related indicators are divided into 3 categories, the first category is MCHC, CHCMr, MCH and Fer; the second category is MCHC, CHCMr, MCH and Fer; The categories are RDW, serum iron and HDW; the third category is MCV, MCVr and RETIC.

DISCUSSION

Overtraining can lead to lower hemoglobin in male taekwondo athletes

The occurrence of exercise-induced hypohemoglobin in athletes is a common problem in sports training. Therefore, it has been a research hotspot for many years to monitor the athlete's hemoglobin and erythrocyte-related indexes and find out the cause of exercise-induced hypohemoglobin. At present, the research on exercise-induced anemia is mostly about the mechanism of exercise-induced anemia, and most of them are limited to animal experiments. Even human experiments are mostly horizontal investigations on athletes, while there are few studies on the changes of red blood cell-related indicators in male taekwondo athletes during the development and development of exercise-induced anemia. In recent years, relevant animal experimental studies have also begun in China. In this experiment, increasing load training was used. During the whole increasing exercise load, the hemoglobin of male taekwondo athletes showed a continuous decrease, and 18 of the 21 subjects had exercise-induced hypohemoglobin, indicating that increasing load training can easily cause the subjects to exercise Low hemoglobin is an effective way to form exercise-induced low hemoglobin. The reason may be that male taekwondo athletes

Table 3. Correlation analysis between hemoglobin and red blood cell parameters during 4-week incremental load training.

	MCV	MCH	MCHC	Ret	RDW	HDW	Fe	Fer	MCVr	CHCMr
Hb	0.091	0.184**	0.399**	0.270**	-0.083	0.143	0.131	0.269**	-0.220*	0.554**
MCV		0.920**	-0.162	0.210*	-0.064	-0.558**	0.126	0.071	0.802**	0.143
MCH			0.131	-0.099	-0.030	-0.466**	0.122	0.174	0.705**	0.340**
MCHC				0.355**	-0.076	0.355**	0.073	0.335**	-0.396**	0.575**
Ret					0.212*	0.492**	-0.105	-0.107	-0.201*	0.180*
RDW						0.065	-0.116	-0.136	0.050	-0.171
HDW							-0.181*	0.120	0.479**	0.083
Fe								0.054	0.065	0.083
Fer									-0.251**	0.491**
MCVr										-0.281**

Table 2. Changes of hemoglobin and erythrocyte-related indexes of subjects during 4-week incremental load training (n=21).

	Before exercise	Weekend 1	Weekend 2	Weekend 3	Weekend 4
Hb (g/L)	137.60±11.80	129.70±9.10 [#]	126.40±9.20 [#]	124.40±8.30 ^{###}	119.50±7.70 ^{###&}
MCV (fl)	79.95±6.45	81.12±6.63 [#]	80.93±6.48 [#]	79.73±6.18 ^{##&}	79.37±5.89 ^{###&}
MCH (pg)	28.01±2.72	27.86±2.58	27.68±2.56 [#]	27.61±2.59 [#]	27.68±2.45 [#]
MCHC (g/L)	349.90±9.10	343.10±8.60 [#]	341.70±9.10 [#]	346.00±10.20 ^{###&}	344.10±7.70 [#]
Ret (x10 ⁹ /L)	56.19±12.94	55.96±16.59	56.15±12.63	61.31±19.31	54.79±10.76
RDW (%)	12.91±0.54	12.98±0.59	13.05±0.68	13.07±0.82	13.25±0.77 ^{###&}
HDW (%)	2.58±0.30	2.53±0.29 [#]	2.55±0.28 [#]	2.52±0.26 [†]	2.53±0.30 [#]
Fe (μmol/L)	12.10±5.88	10.20±5.09	13.20±8.01	9.10±4.36	11.50±6.38
Fer (μg/L)	59.87±21.51	44.57±4.94 [#]	42.03±12.86 [#]	38.02±15.63 ^{##&}	27.74±11.17 ^{###&}
MCVr (fl)	90.73±5.23	96.84±4.44 [#]	96.99±4.28 [#]	96.09±4.10 [#]	96.88±4.66 [#]
CHCMr (g/L)	38.28±7.30	33.73±1.64 [#]	33.44±1.72 [#]	32.99±1.68 [#]	33.17±1.70 [#]

Note: [#] $P < 0.01$: Compared with before exercise; ^{**} $P < 0.01$: compared with the week 1; [†] $P < 0.05$, [&] $P < 0.01$: Compared with the week 2.

not only bear a heavy load of exercise in the process of increasing load training, but also have insufficient nutrient reserves caused by reducing food intake in order to maintain a good state, and are more prone to exercise-induced low hemoglobin.^{11,12}

Analysis of the changes of erythrocyte-related indexes in the process of exercise-induced hypohemoglobin

The indicators usually used to monitor the changes of hemoglobin during heavy exercise mainly include red blood cell morphological characteristic indicators, reticulocyte-related indicators and iron metabolism-related indicators. The changes of erythrocyte morphological indexes MCV, MCH and MCHC can reflect the aging and hemolysis of erythrocytes caused by exercise training. When there is obvious hemolysis *in vivo*, the increase of free hemoglobin will lead to the increase of MCH and MCHC; When red blood cells age, shrink and volume decreases, and when red blood cells age and oxidative damage, MCHC will also increase, and internal viscosity will increase. The results of this experimental study showed that in the early stage of increasing load exercise in male subjects, the shape of red blood cells changed to large cells and hypochromic red blood cells, and later to small cells and hypochromic red blood cells, and the reduction of cell volume was more obvious than that of red pigment, suggesting that the ability of red blood cells to carry oxygen decreases during the process of increasing load exercise. The reason may be that the reduction of VitB12 is the main reason in the early stage of exercise. In the later stage of exercise, due to the low reserve of basic substances in male athletes and insufficient iron supply in the hematopoietic system, the body's red blood cells divide and heme synthesis is affected. RDW and HDW are objective indicators reflecting the volume of peripheral red blood cells and hemoglobin content, and can be used as observation indicators for the diagnosis of early iron-deficiency anemia and the effect of iron-deficiency treatment. When iron deficiency and reticulocytosis are excluded, RDW is abnormally elevated. Athletes are often prompted to rupture red blood cells. Comparing the erythrocyte-related indicators in the results of this study, it can be found that the decrease in hemoglobin concentration is most likely due to the increase in the production of free radicals caused by incremental load exercise, which increases the fragility of erythrocytes, and the erythrocytes are easily destroyed and dissolved.^{13,14}

Serum iron, ferritin and reticulocyte parameters are closely related to the regenerative capacity of red blood cells. MCVr and CHCMr reflect the relationship between the rate of cell proliferation and differentiation and the rate of hemoglobin production in the process of proliferation and differentiation of erythroid cells in the bone marrow. When the volume of reticulocytes increases and the average reticulocyte hemoglobin concentration decreases, it indicates that the reticulocytes in the bone marrow are defective in factors that promote hemoglobin synthesis, such as bone marrow iron deficiency, or the differentiation capacity of reticulocytes is limited, such as Folic acid, vitamin B12 deficiency, etc. Combined with the results of this study, CHCMr and serum ferritin were significantly decreased without significant changes in serum iron, indicating that during the process of increasing load training, male subjects were not deficient in serum iron, but there was a decrease in iron storage, from the first week of exercise. There are factors that affect hemoglobin production and inhibit the proliferation and differentiation of reticulocytes from the beginning.^{14,15}

Correlation analysis between hemoglobin and erythrocyte parameters in the course of exercise-induced hypohemoglobinemia

There are few reports on the relationship between hemoglobin reduction and erythrocyte parameters in the process of hypokinesia, and most of them explore the relationship between anemia and erythrocyte parameters CHCMr, HDW and MCVr. Ashenden et al. found that CHCMr is an early and sensitive indicator of erythropoiesis in the occurrence of iron-deficiency anemia in male athletes, and iron supplementation can significantly increase CHCMr; Rafi et al. believed that CHCMr is more sensitive and accurate than traditional detection indicators such as transferrin saturation and serum iron in monitoring iron deficiency during hemodialysis in renal disease; HDW is also widely used in clinical disease diagnosis to rapidly diagnose the development degree of sickle cell anemia; MCVr is often used as a sensitive indicator in the diagnosis of iron deficiency anemia. The results of this experiment also support the previous research results. In the process of exercise-induced hypohemoglobin in male taekwondo athletes, the correlation analysis between hemoglobin and red blood cell parameters shows that MCHC and CHCMr have the greatest correlation with hemoglobin concentration, reaching a moderate correlation, while serum Fe, RDW, HDW and MCV are closely related to hemoglobin. There was no obvious correlation, and there was also a certain correlation between HDW and MCH, MCHC, Ret, and MCVr ($P < 0.01$). Among them, CHCMr is related to a number of red blood cell parameters, and from the perspective of cluster analysis, it is grouped with MCHC, MCH and ferritin, so it can reflect the state of iron deficiency and erythropoiesis; Cluster analysis found that HDW was grouped with RDW and serum iron, but HDW was significantly correlated with several red blood cell parameters, indicating that HDW had more advantages than RDW and serum iron in reflecting the process of hypokinetic hemoglobin.¹³⁻¹⁵

CONCLUSION

Induction of exercise-induced hemoglobinemia in male taekwondo athletes during 4-week incremental load training. In the process of hypokinetic hemoglobin, red blood cell related indexes MCV, Ret, RDW, Fe did not change significantly, while MCH, MCHC, HDW, Fer, MCVr and CHCMr showed progressive and significant changes. Correlation analysis showed that the hemoglobin concentration had the greatest correlation with MCHC and CHCMr, while there was no significant correlation with serum Fe, RDW, HDW and MCV. This paper studies and analyzes the change characteristics of red blood cells and their related indicators during the process of low cylinder hemoglobin in Chinese male taekwondo athletes during incremental load training, and provides a basis for taking effective measures to prevent exercise-induced hemoglobin reduction.

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