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## CONTENT


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# EFFECTS OF ANTI-FATIGUE FUNCTION OF SPINE GRAPE AND CERASUS HUMILIS NORMAL JUICE ON PHYSICAL FITNESS OF STUDENTS AND DEVELOPMENT OF SPORTS

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**ABSTRACT**
Adolescent students can gain energy and nutritional substances and recover from fatigue by drinking sports beverage after sports. Taking spine grape and cerasus humilis as raw materials, we developed a kind of anti-fatigue sports beverage with special flavor according to national formulation requirements for sports beverage. Besides, anti-fatigue activity function of the sports beverage was evaluated by carrying out animal experiment. The beverage made from spine grape and cerasus humilis is proved to be a natural sports beverage with good anti-fatigue function and can effectively strengthen physical performance, relieve fatigue, help remove free radicals brought by high-strength or exhaustive exercise, prevent early generation of fatigue, increase human tolerance, improve overall athletic ability and promote the development of sports field.

## 1. Introduction

Comprehensive development of adolescent students has long been a hot topic concerned worldwide (Kokko et al., 2006; Hu et al., 2015; Akiyama et al., 2013). Having a healthy body is the basic requirement put forward by Central Committee of the Chinese Communist Party and the State Council for adolescent students from the perspectives of Chinese talent cultivation and sustainable development strategy, which is not only the premise for serving the country and people but also the sign of vigorous vitality of Chinese nation (Sun, 2013; P-Mar-Apr, 2006). Physical fitness is an important measuring standard for physique conditions. Good physical fitness is an important condition for physical education and sports. Athletic ability of teenagers which can be reflected directly by physical fitness is an important sign of physique conditions. Overall, good physical fitness during adolescence is an important condition for healthy life in future (Wang et al., 2015; Zheng et al., 2013; Feng et al., 2014). Due to the demand of sportsmen and body builders, functional sports beverage develops (Dimitrovski et al., 2016). Internal environment will change, i.e., human body will be tired, lose water and have excessive free radicals when sports load of human body exceeds certain value. Therefore, it is of great significance to supplement exogenous substrates to relieve fatigue and help rapid recovery of body (Spierer et al., 2013; Galemore, 2011; Kinnear and Kock, 2011); moreover, it provides a good environment for expansion of sports beverage market. This study focuses on two natural fruits, i.e., spine grape (Lai et al., 2014) and cerasus humilis (Chang et al., 2011), which contain rich vitamins, amino acid, anthocyanin, resveratrol and microelement. Spine grape and cerasus humilis normal juice can relieve fatigue, remove residual free radicals and accelerate recovery. Functional sports beverage made

from spine grape and cerasus humilis is useful for improving athletic ability of adolescent students and promote the development of sports field.

## 2. Materials and methods

### 2.1. Experimental materials

Sports beverage made from spine grape and cerasus humilis, Gatorade sports beverage and normal saline were taken as research objects. One hundred healthy Kunming mice (50 males and 50 females), weighed (18 ± 2) g, were selected. Experimental instruments included superoxide dismutase (SOD) detection kit, malondialdehyde (MDA) detection kit, blood urea nitrogen (BUN) detection kit, blood lactic acid (LAC) detection kit, liver glycogen detection kit, visible spectrophotometer, TDL-40B low-temperature high-speed centrifuge, FA/JA electron balance, tank for loaded swimming (70 × 50 × 40 cm), lead sheath, etc.

### 2.2. Experimental method

***Raising and grouping of experimental animals***

Before exhaust experiment, the experimental mice were raised for a period of time; and in that period, the mice were quarantined. During quarantine, the mice ate and drank freely at temperature of 20 ~ 30 °C. One week later, all qualified mice experienced two-day adaptive swimming in a tank with water deep of 30 cm and temperature of 25 ± 2 °C. After swimming training, mice which grew normally and were able to swim were selected and divided into three groups, i.e., group one, group two and group three (control group), 10 in each group. Mice in group one received gavage of sports beverage made from spine grape and cerasus humilis; mice in group two received gavage of Gatorade sports beverage; mice in group three received gavage of normal saline. The gavage was performed for 20 days, once a day, with a daily dose of 0.1ml/10g (beverage quantity/weight).

***Safety inspection***

During 20 days, food and water intake, defecation, growth, behavior and toxic signs of mice in three groups were observed. After experiment, whether liver, heart, lung, kidney, stomach, jejunum, spleen, thymus gland and ovary/testis of test mice had injury or pathological changes were checked.

***Determination of growth speed of mice***

Weight of experimental mice would change before and after experiment; hence tracking and record were necessary in order to calculate the average net weight gain.

***Loaded swimming experiment***

Gavage lasted for 20 days. Thirty minutes after the last gavage, the mouse was loaded with lead sheath whose weight was 5% that of mouse on the tail and put in a 30 cm-deep swimming tank with water temperature of 25 ± 2 °C. The experiment ended when mice ran out of energy, i.e., mice failed to float or show righting reflex after sinking into water for 10 s. That period of time was regarded as duration of loaded swimming.

***Determination of viscera index of mice***

Weight of the mice was measured after sports. Then the mice were killed and the organs were taken out for weighing. Viscera index was calculated (viscera index was expressed by the ratio of weight of organs (mg) and weight (g)). The experimental operation strictly followed the instruction on blood lactic acid detection kit.

***Determination of LAC content***

The mice were taken out of water after running out of energy; then blood was immediately extracted from eye socket for preparation of serum. Then the content of blood lactic acid was detected using blood lactic acid detection kit and blood lactic acid instrument. The experimental operation strictly followed the instruction on the blood lactic acid kit.

***Determination of hepatic glicogen content***

Organs were taken out after exhaustive mice were killed. The organs were washed by normal saline and dried by filter paper. 100 mg of liver was weighed precisely using electronic analytical balance. The content of hepatic glucogen was detected using hepatic glucogen kit. The experimental operation strictly followed the instruction on the hepatic glucogen kit.

***Determination of superoxide dismutase (SOD) content***

Mice were taken out from water after running out of energy; then blood was immediately extracted from eye socket. Serum was prepared and then the content of SOD in serum was detected. The experimental operation strictly followed the instruction on the SOD detection kit.

***Determination of MDA content***

Mice were taken out from water after running out of energy; then blood was immediately extracted from eye socket. After preparation of serum, the content of MDA in serum was detected using visible spectrophotometer. The experimental operation strictly followed the instruction on the MDA detection kit.

***Determination of BUN content***

Mice were taken out from water after running out of energy; then blood was immediately extracted from eye socket. After 3 hour-standing, the blood was centrifuged at 3000 r/min at low temperature for 15 min. Finally 1μl of serum was obtained. BUN detection kit was used to detect the content of serum BUN. The experimental operation strictly followed the instruction on the BUN detection kit.

### 2.3. Statistical analysis

Statistical Package for Social Sciences (SPSS) was used for statistical analysis. Data were expressed as mean ± SD and processed by independent sample t test. Difference was considered statistically significant if $p < 0.05$.

## 3. Results and discussions

### 3.1. Analysis of safety of sports beverage made from spine grape and cerasus humilis

During 20 days, mice in the three groups behaved normal in aspects of eating, water drinking and defecation and grew well; besides, no obvious changes or intoxicating phenomenon were observed. At the end of the experiment, organs of the experimental mice were examined; and no significant pathological changes were observed, suggesting no damage was induced by the test sample. Therefore, we consider sports beverage made from spine grape and cerasus humilis is safe and nontoxic.

### 3.2. Changes of growing speed of mice under the effect of sports beverage made from spine grape and cerasus humilis

Results of weight and net weight increase of mice in the three groups before and after experiment are shown in Table 1. Average net weight increase of mice that were gavaged with sports beverage made from spine grape and cerasus humilis for 20 days was 4.823 g and average net weight increase of mice that were gavaged with Gatorade sports beverage was 4.941 g, higher than that of the mice in the control group; but there was no significant difference. Therefore, sports beverage made from spine grape and cerasus humilis was considered to have no effects on growth of body.

### 3.3.Changes of duration of exhaustive swimming under the effects of sports beverage made from spine grape and cerasus humilis

The effect of anti-fatigue drugs can be evaluated through loaded swimming experiment; exercise tolerance directly affects duration of swimming and thus reflects fatigue degree after sports (Ting-Jun and Yi-Qing, 2012).

**Table 1.** Changes of growing speed of mice under the effect of sports beverage made from spine grape and cerasus humilis (mean ± SD, n = 10)

| Group | Weight before experiment (g) | Weight after experiment (g) | Average net weight increase (g) |
|---|---|---|---|
| Group one | 18.781±2.649 | 22.997±1.008 | 4.982±1.718 |
| Group two | 17.963±2.481 | 22.756±1.067 | 4.818±1.766 |
| Group three (control group) | 19.276±1.957 | 23.845±0.738 | 4.988±2.001 |

Table 2 shows duration of loaded swimming of mice gavaged with different things. We found 20-day gavaging of sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage can significantly extend duration of loaded swimming; compared to the control group, there were significant differences. But duration of loaded swimming of two sports beverage groups had no significant difference. Thus sports beverage made from spine grape and cerasus humilis has certain anti-fatigue effect.

**Table 2.** Changes of duration of loaded swimming under the effect of sports beverage made from spine grape and cerasus humilis (mean ± SD, n = 10)

| Group | Duration of loaded swimming (min) |
|---|---|
| Group one | 26.458±2.741 |
| Group two | 29.836±3.391 |
| Group three (control group) | 17.569±2.756 |

### 3.4. Changes of visceral index of mice under the effects of sports beverage made from spine grape and cerasus humilis

Ratio of visceral index to weight of mice in each group at the end of experiment is shown in Table 3. The ratio of liver, kidney and spleen to weight of mice gavaged with sports beverage made from spine grape and cerasus humilis and mice gavaged with Gatorade sports beverage had no significant difference. However, the ratio of thymus gland to weight of mice gavaged with sports beverage was significantly different from that of mice in the control group.

### 3.5. Changes of biochemical indexes of mice after exhaustive swimming under the effect of sports beverage made from spine grape and cerasus humilis

***Content of LAC***

In the beginning of sports, glycolysis reaction provides energy for muscle and moreover a large amount of LAC generates. When sports end, glycolysis stops and LAC reduces, which leads to the decrease of LAC content and recovery of body. It can be seen that, lowering output of LAC and improving removal speed of LAC is effective in resisting fatigue. The content of LAC becomes higher as LAC produced during sports will rapidly penetrate into blood; the phenomenon continues until LAC in muscle and blood reaches a balance. Content of LAC of mice after exhaustive swimming is shown in Table 4. It can be seen from Table 4 that, the content of LAC of mice gavaged with sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage was 7.147 ± 0.502 mmol/ml and 6.566 ± 0.418 mmol/ml, much lower than the control group; there were significant differences. However, there was no significant difference of content of LAC between mice gavaged with sports beverage made from spine grape and cerasus humilis and mice gavaged with Gatorade sports beverage. Therefore, it can be concluded that, sports beverage made from spine grape and cerasus humilis has strong effect in removing LAC and relieving fatigue.

**Table 3.** Changes of visceral index of mice under the effect of sports beverage made from spine grape and cerasus humilis (mean ± SD, n = 10)

| Group | Liver (mg/10g) | Spleen (mg/10g) | Kidney (mg/10g) | Thymus gland (mg/10g) |
|---|---|---|---|---|
| Group one | 6.522±0.398 | 0.739±0.071 | 6.852±0.398 | 0.354±0.006 |
| Group two | 6.587±0.408 | 0.728±0.047 | 7.065±0.189 | 0.352±0.008 |
| Group three (normal group) | 6.392±0.465 | 0.757±0.052 | 6.557±0.485 | 0.313±0.001 |

***Content of hepatic glycogen***

Hepatic glycogen can provide energy for contraction of muscle fiber and keep normal level of glucose in blood. The availability of hepatic glycogen is the key factor influencing the beginning of muscle fatigue (Eduardo et al., 2009). The content of hepatic glycogen of mice after exhaustive swimming is shown in Table 4. Both of the content of hepatic glycogen of mice gavaged with sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage had significant difference with that of the control group; but the difference between the two groups was not remarkable. Thus it can be concluded that, the two kinds of sports beverage can provide more hepatic glycogens and lower consumption of hepatic glycogen, thus to improve physical fitness, tolerance and fatigue resistance.

***Content of SOD***

SOD is the only enzyme whose substrate is oxygen free radical and an important enzyme which is effective in removing hydrogen peroxide. As shown in Table 4, the content of SOD of mice gavaged with sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage significantly increased, both of which showed a significant difference from the control group; but the content of SOD of mice gavaged with sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage had no remarkable difference. The action mechanism may be that, functional sports beverage contains substances that can activate activity of antioxidant enzyme or induce synthesis of antioxidant enzyme.

***Content of MDA***

MDA can be used for measuring metabolism of free radicals. MDA as the representative product of lipid peroxide can reflect the level of free radicals objectively. The change direction of activity of SOD is opposite to change direction of MDA. Research results are shown in Table 4. Compared to the control group, the content of MDA of mice gavaged with sports beverage made from spine grape and cerasus humilis and mice gavaged with Gatorade sports beverage had remarkable decline; however, the content of MDA of mice gavaged with sports beverage had no significant difference. Therefore, it can be concluded that, sports beverage made from spine grape and cerasus humilis is effective in improving antioxidant ability and inhibiting lipid peroxidation.

***Content of BUN***

Catabolism of nitrogen substance can be directly reflected by the content of BUN and it is also the evaluation index for sports load that can be tolerated in special conditions. Table 4 shows the changes of content of BUN of mice after loaded swimming. It can be seen from Table 4 that, BUN content of mice gavaged with sports beverage and mice gavaged with normal saline had no significant difference ($p > 0.05$).

When sports time was lower than 30 min, there was no involvement of protein during energy supply; as BUN is the product of protein metabolism, there are no remarkable changes of BUN. That might also be the reason why the regulatory effects of the sports beverages on BUN are not remarkable.

**Table 4.** Effects of sports beverage made from spine grape and cerasus humilis on biochemical indexes of mice after exhaustive swimming (mean ± SD, n = 10)

| Group | LAC (mmol/ml) | Hepatic glycogen (mg/g) | SOD (u/ml) | MDA (nmol/ml) | BUN (mmol/ml) |
|---|---|---|---|---|---|
| Group one | 7.147±0.502 | 2.321±0.276 | 129.685±11.321 | 2.681±0.316 | 22.985±2.256 |
| Group two | 6.566±0.418 | 2.348±0.296 | 137.521±6.758 | 2.411±0.359 | 25.174±1.285 |
| Group three (control group) | 10.078±0.832 | 1.859±0.187 | 114.556±5.875 | 4.215±0.348 | 24.295±1.927 |

In this study, we gavaged mice that weighed 18 g with normal saline, sports beverage made from spine grape and cerasus humilis and Gatorade sports beverage for 20 days, then observed the growth condition of mice and tested the effects of sports beverage on weight and visceral index of mice. Research results showed that, food intake and water drinking conditions of mice were normal; no mice died; there was no significant difference of weight before and after experiment; no pathological changes and abnormality were observed in various organs after anatomy; visceral index of liver, spleen and kidney of mice gavaged fluctuated within normal scope, and the difference between mice gavaged with sports beverage and normal saline had no statistical significance. Therefore, it can be concluded that, sports beverage made from spine grape and cerasus humilis has no adverse effects on growth and development of animals and it is safe and non-toxic.

Wild spine grape which contains natural antioxidative substance such as procyanidine and resveratrol is the main component of sports beverage made from spine grape and cerasus humilis. It has been found that, resveratrol has good immunomodulatory effect (Basini et al., 2010; Delmas et al., 2013). Besides, a study (Liu and Huang, 2006) suggested that long-term intensive training could result in apoptosis of lymphocyte through consuming nutritional substances, changing neuro-endocrine function and attacking membrane of immune cells and thus lead to decline of immune function. In the test, thymus index of the mice gavaged with sports beverage was much higher than that of the control mice, which was because of polyphenols contained in the sports containing spine grape.

SOD, the defense system of human body, can eliminate free radicals to keep its dynamic balance (Tina et al., 2009; Cai and Wang, 2014). In the experiment, two kinds of sports beverage both led to significant increase of SOD, suggesting functional sports beverage was able to eliminate free radicals and strengthen antioxidant capacity. MDA as the final product of metabolism of lipid peroxidation cam indirectly reflect the severity of injury of cells induced by oxygen free radical (Valente et al., 2011). Besides, the functional sports beverages resulted in significant decline of content of MDA, which further suggested the ability of eliminating free radicals of functional sports beverage. Hence it can be concluded that, the sports beverage can improve antioxidant capacity and thus enhance athletic ability of mice.

Besides, it has been pointed out that, procyanidine in red grape wine can eliminate free radicals, resist oxidation and prevent cardiovascular disease and atherosclerotic plaque (Gutha et al., 2010). Therefore, it can be concluded that, sports beverage made from spine grape and cerasus humilis has functions of eliminating free radicals and resisting oxidation, which is because of the high content of procyanidine in the sports beverage made from spine grape and cerasus humilis.

Sports beverage made from spine grape and cerasus humilis as a natural sports beverage with effective anti-fatigue function can effectively enhance body functions, relieve fatigue, help eliminate free radicals induced by high-strength or exhaustive sports, improve

tolerance and enhance overall athletic ability; hence it can fully satisfy the need of teenagers during sports. The development of sports beverage made from spine grape and cerasus humilis extends the market of sports beverage.

## 4. Conclusions

Wild spine grape and cerasus humilis are the raw materials of the developed sports beverage. The application of two fruits in development of functional sports beverage can not only stimulate appetite of people with full flavor, but also can promote the supplement of body fluid of teenagers during sports and satisfy their need of nutrition. We break the conventional idea of supplementing athletes with sugar and electrolyte only and make use of the high content of resveratrol and procyanidine to delay sense of fatigue and enhance sports performance. Besides, in view of the high safety and functionality, the sports beverage is expected to be promising in the market of functional sports beverage.

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# EFFECT OF NANO SWEET POTATO RESIDUE CELLULOSE ON BLOOD GLUCOSE LEVEL OF ATHLETES

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**ABSTRACT**

This study aimed to investigate the regulatory mechanism of nano sweet potato residue cellulose in lowering blood glucose and its effect on blood glucose level of athletes based on the research of the antidiabetic effect and molecular mechanism of nano sweet potato residue cellulose. Diabetes models were built to explore the effect of nano sweet potato residue cellulose in lowering blood glucose level of diabetic rats as well as its functional mechanism. In addition to reducing fasting blood glucose level of diabetic rats, nano sweet potato residue cellulose also had a regulatory effect on blood lipid of diabetic rats; it was conducive to easing body's blood sugar regulation mechanism and improving glucose tolerance. Through the restraint effect on glucose, nano sweet potato residue cellulose can reduce the intestinal absorption of glucose and inhibit postprandial hyperglycemia; with relatively high viscosity values, it can increase intestinal peristalsis and hinder the diffusion of glucose and lipids; by reducing intestinal absorption of glucose and lipid, it can increase athletes' blood glucose excretion, which is a synergistic effect of lowering blood glucose.

## 1. Introduction

Sweet potatoes are rich in starch, dietary fiber, glycoprotein, minerals and vitamins which make for balanced and high nutritional value (Ma and Mu, 2016; Bernstein, et al., 2016). As we know, with the improvement of living standards and changes in diet, a lot of "civilized diseases", such as diabetes, hypertension and obesity, have become more frequent, especially among the people whose dietary fiber intake is insufficient (Altug et al., 2009). Therefore, dietary fiber (DF) is of great significance to human health (Zamot, et al., 2015). Since the body structure of rats is similar to that of human, we used rats as models to study the effect of nano sweet potato residue cellulose in lowering blood glucose, so as to analyze its impact on athletes' blood glucose level.

Previous studies showed that nano sweet potato residue cellulose could significantly inhibit the activity of amylase and pancrelipase (Zhu et al., 2015; Ji et al., 2015). Another study indicated that the decrease in activity of amylase and pancrelipase could reduce the hydrolysis of starch and lipids, while high viscosity values helped to delay and reduce gastrointestinal absorption of glucose and lipid (Zhang et al., 2015). It was reported that dietary fiber could not only elevate serum insulin level, but also reduce and balance postprandial blood glucose level to avoid violent fluctuation of blood glucose level (Marquard et al., 2016). In 2014, Wu et al. performed a vitro experiment which showed that DF inhibited the effect of a-amylase on starch and prolonged enzymolysis time, thus slowing glucose release (Yunshan et al., 2014). Gui et al. (2014) found that barley

husk played a role in regulating metabolism by reducing the activity of amylase, lipase and chymotrypsin in human small intestine (Gui et al., 2014). This study analyzed the possible mechanism of nano sweet potato residue cellulose lowering blood glucose as well as its effect of on blood glucose level of athletes.

## 2. Materials and methods

### 2.1. Experimental materials and instruments

Experimental materials included sweet potato residues (Chew and Ong, 2016), sweet potato residue cellulose, nano sweet potato residue cellulose, microcrystalline sweet potato residue cellulose and healthy male rats.

Experimental instruments included electronic balance, numerical control ultrasonic cleaner, desk-type high speed centrifuge, ultrapure water machine, ultra low temperature freezer, blood glucose tester, refrigerated microcentrifuge, gel imaging system (Lu, et al., 2013) and slicing machine.

Experimental reagents included sodium hydroxide, corn starch, anhydrous calcium carbonate, potassium sulfate, potassium citrate, ether, citric acid, potassium chloride, petroleum ether (Peng et al., 2011), glucose, serum low-density lipoprotein kit, rat serum insulin kit and total protein extraction kit.

### 2.2. Experimental methods

#### *2.2.1. Model building*

Forty male rats (each weighting about 190 g) were selected and separately fed in stainless steel cages at room temperature (25 °C). After one-week feeding, they were divided into five groups randomly according to their weights. One group was selected as the blank group and fed with basic diet, while the remaining four groups of rats were fed with only water for 12 h; citric acid-sodium citrate buffer solution was used for the preparation of streptozocin (STZ) solution (2mg/mL) which was injected into each fasting rat (with a dosage of 60mg/kg). After injection, all rats were free to eat and drink. After three days, blood was drawn from rats' tails for determination of fasting blood glucose. The rats whose blood glucose values were greater than 11.1mmol/L were confirmed of having diabetics; the ones that did not meet the standard were given another injection of STZ solution.

The model diabetic rats were randomly divided into four groups: model control group (MC group), group of ordinary sweet potato residue cellulose (OC group), group of microcrystalline sweet potato residue cellulose (MCC group) and group of nano sweet potato residue cellulose (CNC group). Feed formulation for each group is shown in Table 1. During the experiment, rats in each group were free to drink and eat, with their daily feed intake and weight recorded; fasting blood glucose values were measured every two weeks; the test cycle lasted for 4 weeks.

#### *2.2.2. Determination of fasting blood glucose*

Respectively, in the first week, the second week and the fourth week, the rats fasted for 12 hours before their blood was drawn from the tail and dropped on the reaction end of blood glucose test strip; then the rats' fasting blood glucose values were determined by a glucometer (Kim et al., 2010).

#### *2.2.3. Determination of glucose tolerance*

After fasting for 12 hours, glucose solution was injected into rats' stomachs, and their blood glucose values were measured at 0 min, 30 min, 60 min and 120 min; area under the curve (AUC) was calculated according to the measured results, and sugar tolerance level was compared.

#### *2.2.4.Determination of glycosylated serum protein*

Serum glucose can have nonenzymatic glycation reaction with N-terminal amino of albumin and other serum proteins, forming macromolecule ketone amine structure (Unwin et al., 2010).

**Table 1.** Feed formulation for the experiment

| Ingredients | CON | Diabetes group | | | |
|---|---|---|---|---|---|
| | | MC | OC | MCC | CNC |
| Corn starch | 550 | 550 | 450 | 450 | 450 |
| Casein | 200 | 200 | 200 | 200 | 200 |
| Soybean oil | 100 | 100 | 100 | 100 | 100 |
| Cane sugar | 100 | 100 | 100 | 100 | 100 |
| Mixed minerals | 35 | 35 | 35 | 35 | 35 |
| Mixed vitamin | 10 | 10 | 10 | 10 | 10 |
| L-cystine | 3 | 3 | 3 | 3 | 3 |
| Choline chloride | 2.5 | 2.5 | 2.5 | 2.5 | 2.5 |
| Sweet potato residue cellulose | - | - | 100 | - | - |
| Microcrystalline sweet potato residue cellulose | - | - | - | 100 | - |
| Nano sweet potato residue cellulose | - | - | - | - | 100 |

#### *2.2.5. Determination of serum lipid*

Serum total cholesterol (TC), triglyceride (TG), high density lipoprotein (HDL-C) and low density lipoprotein (LDL-C) were detected with the kit and determined with a full automatic biochemical analyzer.

#### *2.2.6. Determination of serum insulin*

Specimens, standard and detection antibody were added into coated microtiter for incubation and thoroughly washed. Substrate tetramethylbenzidine (TMB) was used for coloration; catalyzed by peroxidase, TMB turned blue and eventually turned yellow under the action of acid. Insulin was positively correlated with the shade of color. Absorbance at the wavelength of 450 nm was measured with Elisa and sample concentrations were calculated.

### 2.3. Data analysis method

The test data were compiled by Excel and then analyzed by statistical software SPSS 17.0. The results were shown in the form of mean ± standard deviation. When p was less than 0.05, there was significant difference.

## 3. Results and discussions

### 3.1 Changes in feed intake of diabetic rats affected by nano sweet potato residue cellulose

As shown in Figure 1, in the four groups of diabetic rats (except the blank group), feed intake increased significantly ($p < 0.05$); meanwhile, feed intake also decreased with the decrease of cellulose particle size, yet the decrease range was insignificant ($p < 0.05$). Therefore, we assumed that the differences in particle size of sweet potato residue cellulose could not affect feed intake of rats (Keiko, et al., 2007).

### 3.2 Changes of fasting blood glucose in diabetic rats affected by nano sweet potato residue cellulose

As shown in Figure 2, compared to the rats (in blank group) whose fasting blood glucose values were within the normal range, the rats in other groups had higher blood glucose level (higher than 11mmol/L). In the second week, except the rats in blank group, the rats in other groups had decreased blood glucose level due to the difference in particle size of sweet potato residue cellulose; in CNC group, blood glucose values of diabetic rats decreased significantly ($p < 0.05$).

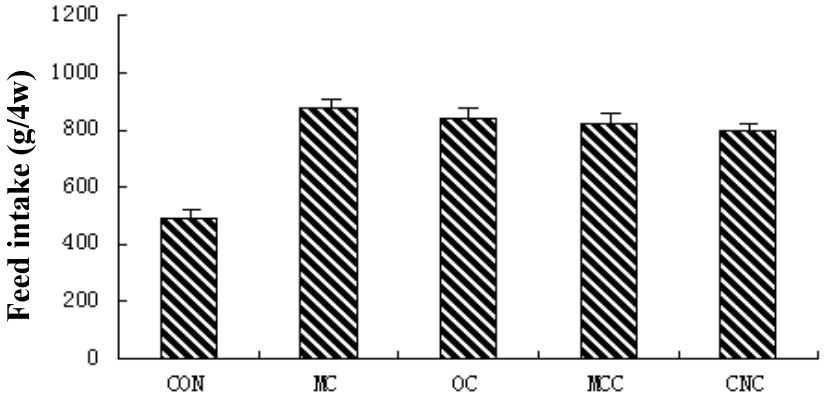


**Figure 1.** Changes in feed intake of diabetic rats affected by nano sweet potato residue cellulose
Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose. Experiment data were shown in the form of mean ± standard deviation (n = 8).

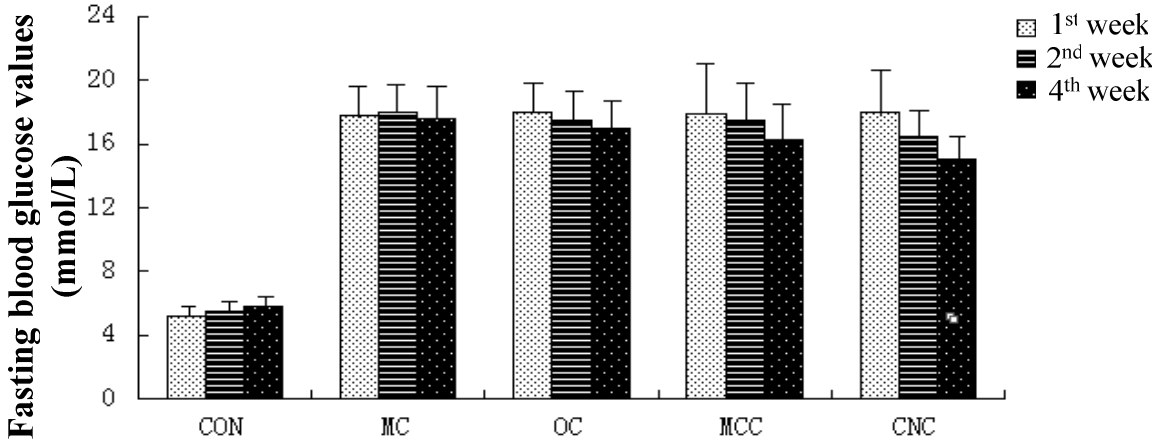


**Figure 2.** Changes of fasting blood glucose in diabetic rats affected by nano sweet potato residue cellulose
Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose. Experiment data were shown in the form of mean ± standard deviation (n = 8).

In the fourth week, except blank group and control group, blood glucose values declined significantly in the other groups ($p < 0.05$). Therefore, it was considered that nano sweet potato residue cellulose could inhibit the increase of fasting blood glucose in rats.

### 3.3. Changes of glucose tolerance in diabetic rats affected by nano sweet potato residue cellulose (Defronzo, et al., 2011)

According to Figure 3, in the first minute, blood glucose values of rats in diabetic groups were higher than 11mmol/L, while blood glucose values of the rats in blank group were 5.68mmol/L. Thirty minutes after intragastric infusion of glucose (of certain concentration), blood glucose level increased significantly in each group; 60 min after the infusion, there was a decline trend; 120 min after the infusion, blood glucose level of the rats in blank group decreased to 6.12 mmo1/L. When a large amount of glucose was consumed all at once, under the effect of intracorporal regulation mechanism, intestinal tract could absorb almost all the glucose, resulting in rapid increase of blood glucose. Accordingly, it was considered that sweet potato residue cellulose helped to ease glucose regulation mechanism in the body and improve glucose tolerance.

### 3.4. Changes of glycosylated serum protein in diabetic rats affected by nano sweet potato residue cellulose

According to Figure 4, except in blank group, glycated serum protein content of the experimental diabetic rats in other groups increased significantly ($p < 0.05$). In comparison with model control group, glycated serum protein content of the rats in MCC group decreased by 0.2mmol/L, while glycated serum protein content of the rats in CNC group decreased by 0.28mmol/L. It can be seen from the data that glycated serum protein content decreased more significantly in CNC group than in MCC group ($p < 0.05$), based on which we considered that the effect on glycosylated serum protein content was enhanced with the decline in particle size of sweet potato residue cellulose (Yoshitomo, et al., 2003).

### 3.5 Changes of serum lipids in diabetic rats influenced by nano sweet potato residue cellulose

As can be seen from Table 2, in comparison with blank group, the concentrations of TC, TG and LDL-C were obviously higher in serum of the rats in other groups ($p < 0.05$), while the concentration of HDL-C reduced significantly in serum of the rats in other groups in comparison with blank control group ($p < 0.05$). Except blank control group, TC, TG and LDL-C concentrations decreased in the remaining groups since the rats were fed with sweet potato residue cellulose (of different particle sizes). Among the experimental rats, there was no obvious decrease in OC group, while a significant downward trend was found in CNC group ($p < 0.05$). Thus, it was considered that lipids of diabetic rats could be regulated by nano sweet potato residue cellulose to some extent (Teng, et al., 2012).

### 3.6 Changes of serum insulin in diabetic rats affected by nano sweet potato residue cellulose

According to Figure 5, serum insulin was in a significant decreasing trend in the rats of model control group, in comparison with other groups ($p < 0.05$). After the rats were fed with different particle size of sweet potato residue cellulose, serum insulin began to rise again in each group (except blank control group and model control group). The increasing trend was especially significant in CNC group ($p < 0.05$).

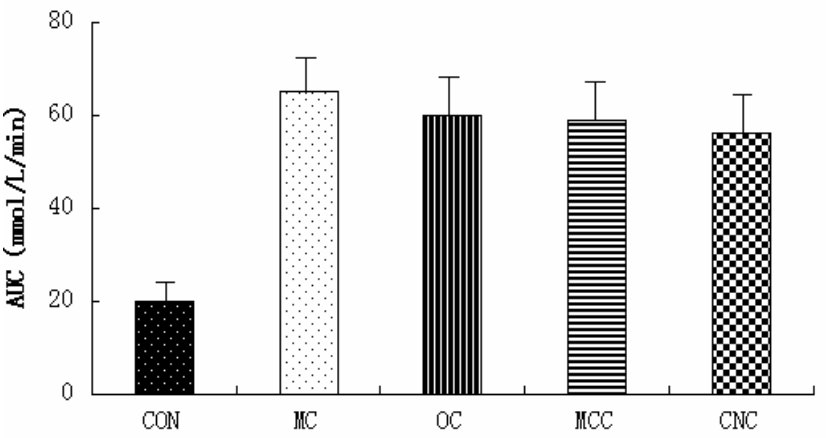


**Figure 3.** Changes of glucose tolerance in diabetic rats affected by nano sweet potato residue cellulose
Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose.

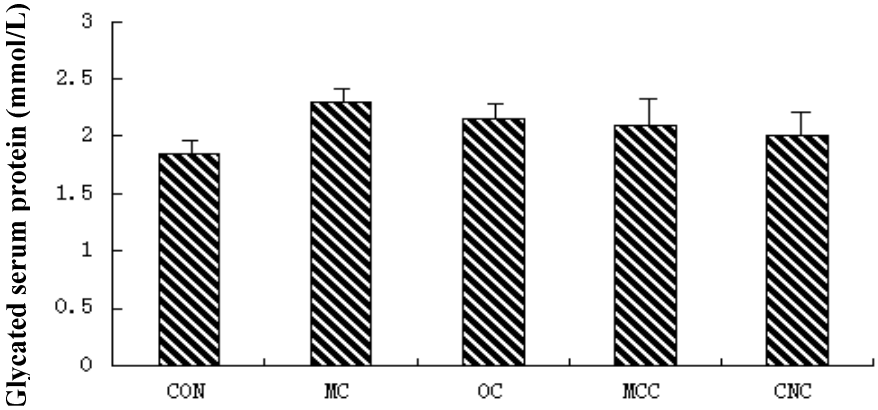


**Figure 4.** Changes of glycosylated serum protein in diabetic rats affected by nano sweet potato residue cellulose
Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose. Experiment data were shown in the form of mean ± standard deviation (n = 8).

**Table 2.** Changes of serum lipids in diabetic rats influenced by nano sweet potato residue cellulose

| Indexes (mmol/L) | Blank group | Diabetic group | | | |
|---|---|---|---|---|---|
| | CON | MC | OC | MCC | CNC |
| TC | 1.3±0.18 | 1.5±0.12 | 1.4±0.08 | 1.4±0.09 | 1.4±0.07 |
| TG | 0.3±0.05 | 0.4±0.05 | 0.4±0.08 | 0.4±0.05 | 0.4±0.04 |
| HDL-C | 0.6±0.07 | 0.4±0.05 | 0.4±0.07 | 0.4±0.08 | 0.5±0.11 |
| LDL-C | 0.1±0.02 | 0.2±0.05 | 0.2±0.03 | 0.2±0.03 | 0.2±0.03 |

Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose. Experiment data were shown in the form of mean ± standard deviation (n = 8).

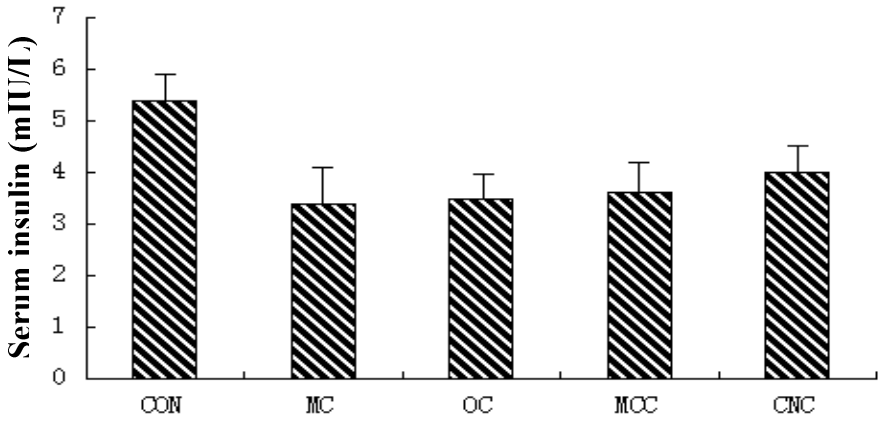


**Figure 5.** Changes of serum insulin in diabetic rats affected by nano sweet potato residue cellulose
Note: CON refers to blank group; MC refers to model control group, OC refers to the group of ordinary sweet potato residue cellulose; MCC refers to the group of microcrystalline sweet potato residue cellulose; CNC refers to the group of nano sweet potato residue cellulose.

## 4. Conclusions

Good eating habits and nutritional balance are of great significance to the prevention of diabetes (Mokhtari, et al., 2013). Dietary fiber can keep the balance of postprandial blood glucose and contribute to regulating blood glucose level for athletes (Wu et al., 2015; Li et al., 2015).

In this study, based on diabetic models of rats, we explored the effect of nano sweet potato residue cellulose on lowering blood glucose level of diabetic rats (Khanra et al., 2015). The results showed that the effect of sweet potato residue cellulose on regulating blood glucose was more significant with the decrease of particle size. The content of serum insulin and hepatic glycogen increased significantly in CNC group ($p < 0.05$). Nano sweet potato residue cellulose not only helped to decrease fasting blood glucose, glycosylated serum protein content, serum TC, TG and LDL-L content ($p < 0.05$), but also led to a decrease in liver fat concentration, content of TC and TG, which indicated that nano sweet potato residue cellulose had favorable effects on improving pancreatic tissue morphology as well as reducing blood glucose (Kovac, et al., 2015; Cheng, et al., 2010).

In summary, nano sweet potato residue cellulose can reduce fasting blood glucose and improve glucose tolerance mainly by slowing down the body's absorption of glucose; moreover, it increases insulin secretion and the body's sensitivity to insulin; in addition, it promotes the synthesis of hepatic glycogen and regulates the balance of glucolipid metabolism in liver (Zheng et al., 2015). Athletes can take nano sweet potato residue cellulose to lower blood glucose level effectively.

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# CONSTRUCTION AND IMPLEMENTATION OF QUALITY SAFETY TRACEABILITY SYSTEM OF FRESH AGRICULTURAL PRODUCTS— TAKING BEEF SUPPLY AS AN EXAMPLE

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**ABSTRACT**

In this study, taking beef supply as an example, we applied information technology to the construction and implementation of a quality safety (QS) traceability system of fresh agricultural products, under the guidance of the related experience in guaranteeing QS of agricultural products. In view of the relevant practical experience, we revealed the existing problems of the QS traceability system for fresh agricultural products. Then, we proposed the modeling method of Petri net, followed by establishment of traceability model of feeding process based on Petri net; moreover, we completed the modeling of traceability system. In addition, we established the database and traceability platform for quality safety information of beef supply; using information technology and database technology, we completed the construction and implementation of the quality safety traceability system of fresh agricultural products.

## 1.Introduction

Recently, the occurrence of food quality safety incidents, such as the outbreak of mad cow disease and application of melamine, has caused people's fear on food quality safety. Under the circumstance, consumers request more severe regulation of food quality safety (Amadou, 2014). With extensive attention from government departments and the public, the problems have promoted the researches of traceability systems for food quality safety (Zhen-Huan et al., 2011; Wu et al., 2013; Bazant et al., 2014). Food safety issues not only bring damage to consumers' health and social economy, but also restrict the sustainable development of food industry; consequently, economic development and social stability are affected (Jarvis et al., 2016). In China, agricultural exports account for a large proportion of the export products. However, in recent years, the agricultural products exported from China have been repeatedly investigated, detained and prohibited because of quality safety problems, which has affected the international competitiveness of Chinese agricultural products (Chen et al., 2011). Therefore, the current research emphasis is laid on developing high-yield and high-quality agriculture as well as establishing an agricultural product traceability and regulation system (Zanella and Milhorance, 2016; Mahmood et al., 2015). In terms of livestock and poultry products, pilot sites for traceability application of beef products were established in Beijing; in Shanxi province, the whole-course tracking system for beef supply was established; in Shenzhen city, the quality traceability system for the production process of beef products was established. The whole traceability system includes three basic

elements: individual mobile registration, information transmission system as well as the individual identification and database of agricultural products. Therefore, it is of important practical significance to establish a traceability system which covers the key links (production, processing, transportation, storage and marketing) of agricultural products, to ensure their quality safety.

In this study, we established a quality safety traceability system which could be applied to the whole-process tracking and supervision of production, processing, storage, transport and marketing of fresh agricultural products. The system was conducive to promoting the information transparency of each key link in the supply chain (Villeneuve, 2014), improving the quality assurance of agricultural products, increasing market competitiveness and achieving traceability of quality safety; furthermore, it could further promote the development of agricultural industrialization.

## 2. Analysis and construction of traceability system

### 2.1. Analysis on the performance requirements of traceability system

The performance requirements of quality safety traceability system for fresh agricultural products included: instantaneity and accuracy of data acquisition, reliability of data transmission, consistency of data storage, friendly interface, qualified operation time, reliability of safety performance. At each stage of system development, the authentication and authorization needed to be considered. The system was mainly targeted at consumers, managers and key point enterprise users. Therefore, it was necessary to authorize different users appropriately, so that they were able to use specific functions of the system to complete different tasks (Pang et al., 2016).

### 2.2. Establishment of traceability system model

#### *2.2.1. Feeding model based on Petri net*

Traceability covered two aspects: firstly, when purchasing fresh food, consumers could use barcode technology to inquire and browse the historical information related to the supply links of fresh products; secondly, when quality safety issues of fresh agricultural products occurred, the relevant departments could use bar code labels to trace down the problems. The model is shown in Figure 1.

Production was a key link in guaranteeing the value and quality of fresh agricultural products (Holgersson et al., 2016). In this paper, beef cattle breeding was taken as an example and traced. In the production process, each fresh agricultural product was assigned with a barcode label to certify its identity and record its basic information and key information such as feeding and drug use in the production process. Petri net model of the raising process (from introducing to fattening) of qualified feeder cattle is shown in Figure 2.

Through the analysis on the transition of feeder cattle fattening model, the transition mapping table was obtained, as shown in Table 1.

#### *2.2.2. Traceability information model of the storage and transportation link*

In the storage and transportation link, the data information that needed to be collected mainly included: the basic information of the staff responsible for QS of the fresh agricultural products, transport equipment and route, QS information of the products during transport. The flow diagram is shown in Figure 3.

#### *2.2.3. Traceability information model of marketing link*

Taking the consumption of fresh agricultural products as an example, the application of barcode in the marketing link was illustrated. The flow diagram is shown in Figure 4.

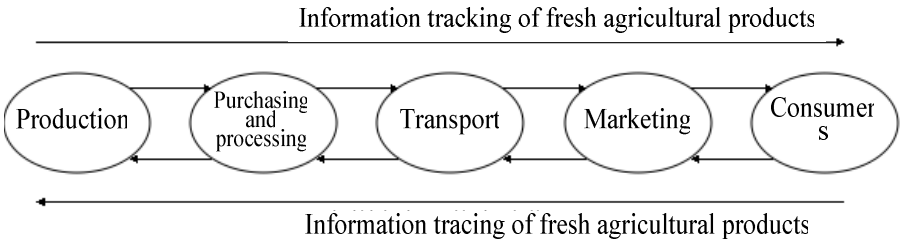


**Figure 1.** Model of information tracking and tracing of fresh agricultural products

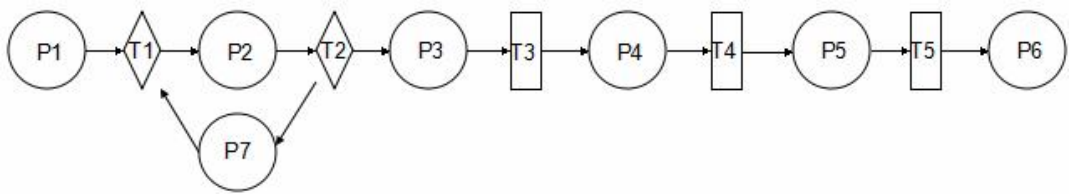


**Figure 2.** Petri net model of the raising process of feeder cattle

**Table 1.** Petri net transition mapping table of feeder cattle

| Transition | Meaning | Place | Meaning | Role |
|---|---|---|---|---|
| T1 | Managers introduced feeder cattle | P1 | Feeder cattle to be introduced | Managers |
| T2 | The feeder cattle were quarantined | P2 | Feeder cattle introduced from pasture | Quarantine officers |
| T3 | After examination and approval, the cattle were switched to another group | P3 | Qualified feeder cattle | Quarantine officers |
| T4 | The feeder cattle were fed | P4 | The cattle to be fattened (in the new group) | Feeders |
| T5 | Managers registered the cattle for marketing | P5 | Fattened cattle | Feeders |
| -- | -- | P6 | The total fattened cattle | Managers |
| -- | -- | P7 | Unqualified feeder cattle | Quarantine officers |

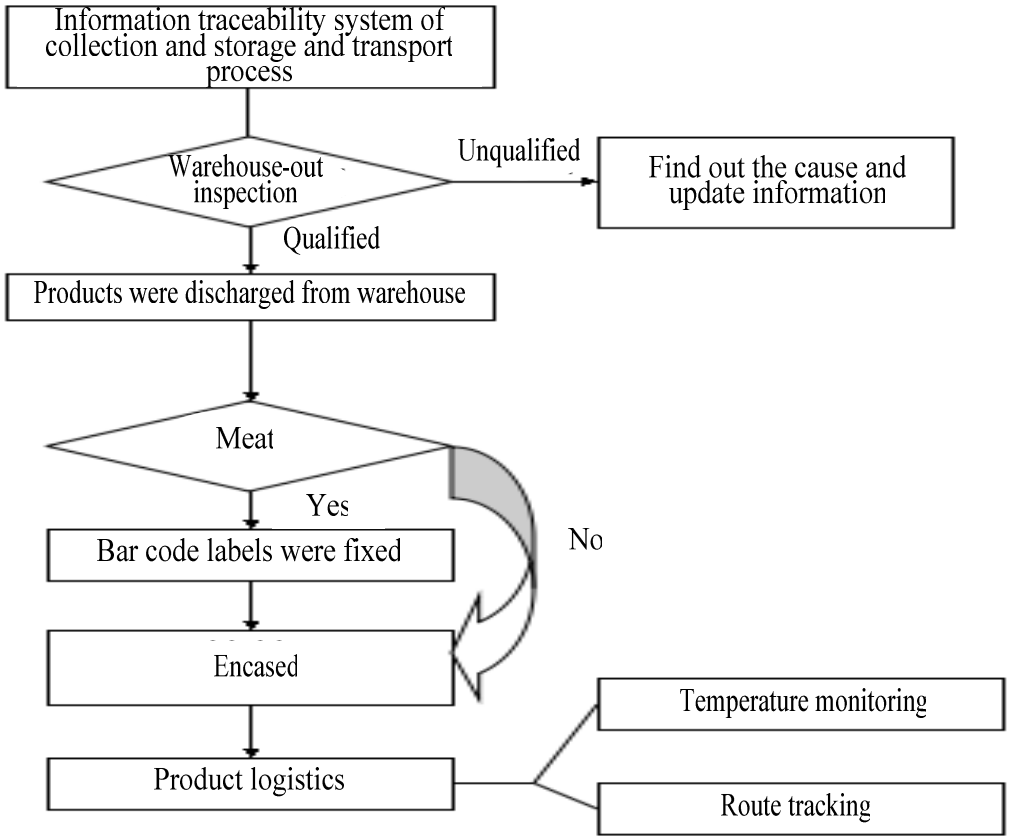


**Figure 3.** Flow diagram of storage and transport link of fresh agricultural products

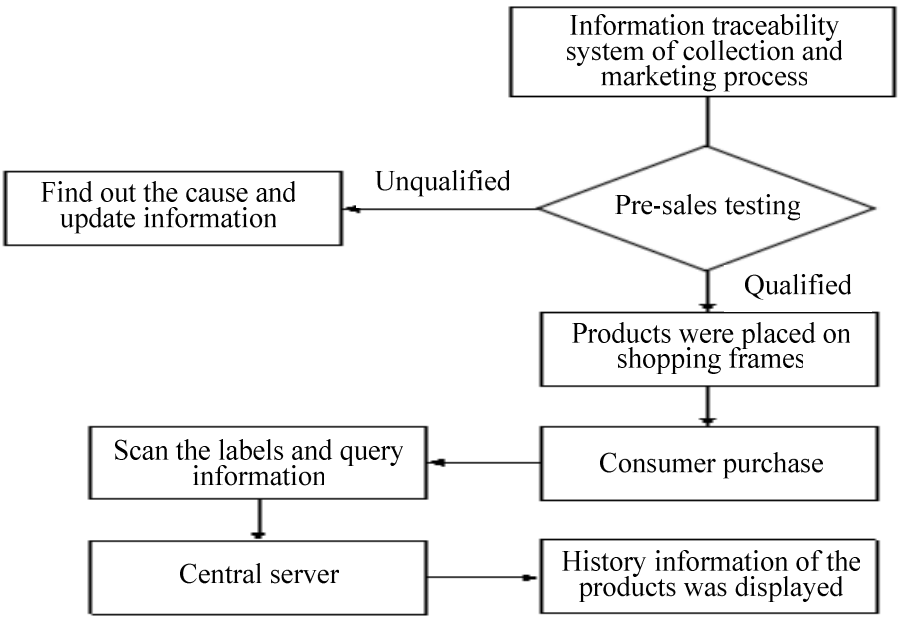


**Figure 4.** Flow diagram of the marketing link of fresh agricultural products based on bar code

As can be seen from the flow chart, if consumers bought the fresh agricultural products, they could use bar code labels as the medium and retrieve the historical information of the products through the central server, which allowed the consumers to have a better understanding of the QS information of the products and make rational choices in purchasing.

## 2.3. Construction of quality safety traceability system for fresh agricultural products

### *2.3.1. Construction steps of the system*

First, the core database system of the fresh agricultural products was designed. Next, the quality safety evaluation system of fresh agricultural products was established. Then, the origin traceability system of fresh agricultural products was established, followed by the establishment of thematic information mapping system. At last, the traceability system platform for product supply chain safety was established (Samarasinghe et al., 2009).

### *2.3.2. Functional design of traceability system*

The traceability system of fresh agricultural products consisted of information collection, query and marketing. Information collection module included user management, producing area management, acquisition and processing management, logistics management. Information query included consumer query and regulation of management departments. Marketing information module included inventory management, marketing management and early warning. The information of information collection module could be read and transferred to traceability information database automatically with radio frequency identification technology.

### *2.3.3.Software architecture design of traceability system*

The traceability system of fresh agricultural products mainly included user interface layer, exterior layer, business layer, data access layer and database. User interface layer was used for displaying the fresh product information that met the requirements of users; exterior layer acted as the interface of traceability system to isolate business logic; business layer functioned by establishing business components according to management objects and calling the defined services to construct corresponding applications; data access layer was the data interface for business layer of traceability system to enter and access the central database. Database was used for storing the information of key links (such as the breeding process of beef cattle, slaughter of beef cattle, processing of beef, beef storage and marketing), thus to provide reliable information on quality safety of products for the access to terminal applications (Pereira et al., 2015; Jacob et al., 2010).

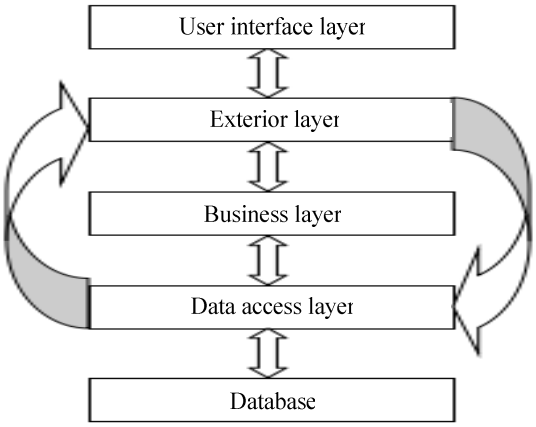


**Figure 5.** Overall software architecture of traceability system

## 2.4.Construction and implementation of quality safety traceability system of fresh agricultural products

### *2.4.1. Development of production subsystem*

Supervision departments and consumers could inquire detailed information of production link of fresh agricultural products via the system terminal. After harvest, fresh agricultural products had to be examined and proved qualified by relevant departments before they flew into the next link. The framework of production subsystem is shown

in Figure 6. In terms of the production process of beef cattle, the information that needed to be recorded included farm information, breeding mode, beef cattle varieties, source of calves, fodder, cowshed disinfection, epidemic situation, quarantine level and slaughter dates (Bene et al., 2010; Mummed et al., 2013).

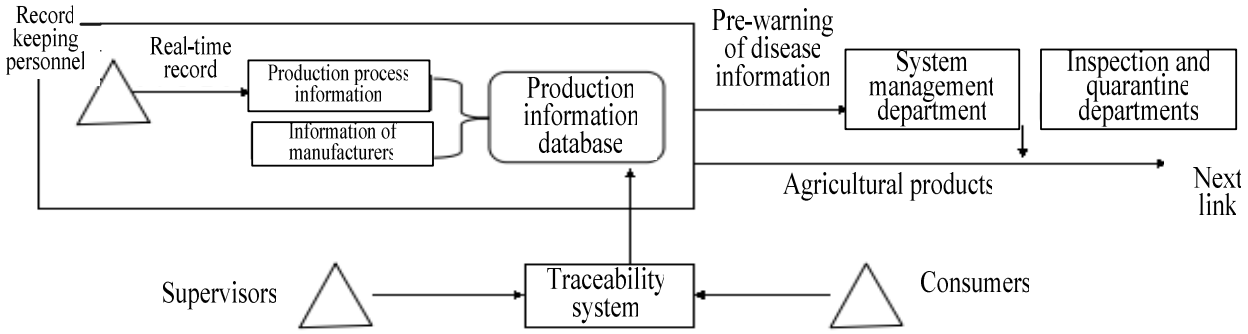


**Figure 6.** Framework of production subsystem

### *2.4.2. Development of processing subsystem*

The processing subsystem of fresh agricultural products required real-time recording of detailed information of fresh agricultural products in the process of collection and processing; and then, the information was timely input to the information database of processing link, so that regulation departments and consumers could inquire the detailed information of processing link. The frame diagram is shown in Figure 7.

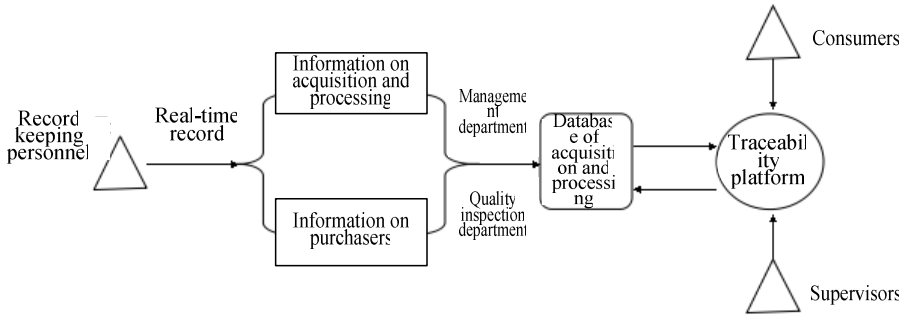


**Figure 7.** Architecture of processing subsystem

Beef processing involved pre-slaughter, slaughter, acid drainage, joint separation, packaging and storage.

Controlled objects in the processing chain included logistics and information flow: logistics referred to the one-way flow from pre-slaughter to storage, while information flow was a two-way flow, as shown in Figure 8.

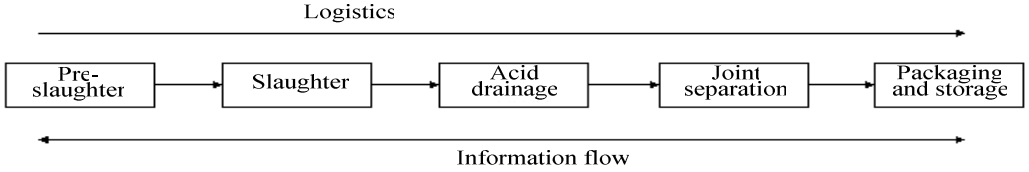


**Figure 8:** Logistics and information flow in beef processing chain

### *2.4.3. Implementation of traceability system*

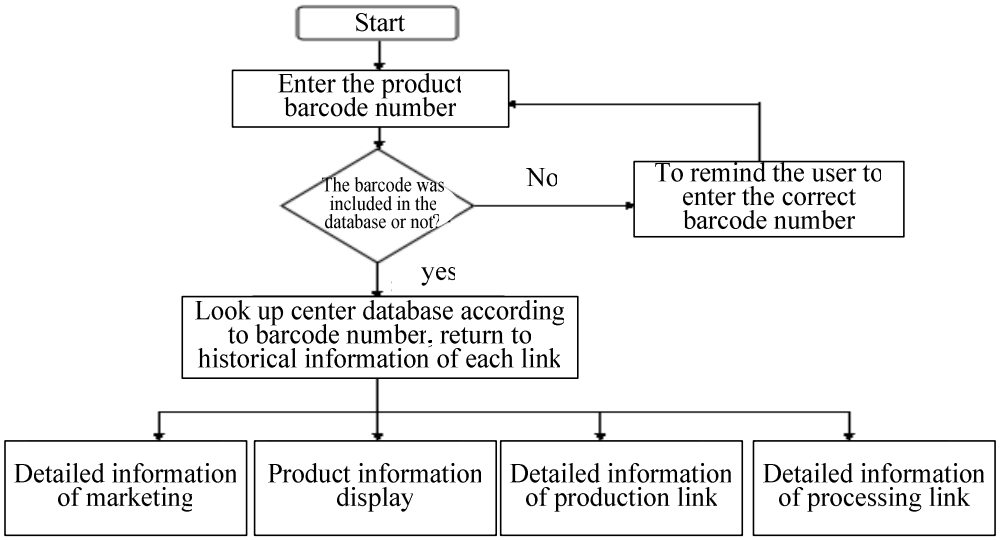


**Figure 9.** Module function flow chart of the traceability system

### *2.4.4. Implementation of product information query interface*

When users input effective barcodes in the tracing interface, they could obtain corresponding history information of the barcodes. The query interface mainly displayed the product information on breeding, processing and marketing, including bar codes, product names, origin, prices, grades, security level, expiration date, manufacturers, slaughter processors, vendors, product number and marketing situation. In addition, the interface also displayed associated controls of the information on production, processing, marketing, system management and security pre-warning. By clicking on the corresponding associated controls, users could view the corresponding historical information (Karni et al., 2015).

### *2.4.5. Implementation of information query interface of production process*

By clicking on the associated control of production information, users could enter the production link information query interface which would display the historical information of the products in production process, including production unit, address, contact information, breeders, breeding objects, drug use, barcode numbers and examinations on the breeding objects (Hao et al., 2016).

### *2.4.6. Implementation of query interface of marketing link*

In the product information interface, by clicking on the associated control of marketing information, users could enter marketing link query interface which displayed the information of vendors, marketing corporation, address, contact information, product names, product codes, marketing status and product inventory. With quality safety traceability system of fresh agricultural products, users could inquire the detailed historical information of the products in the process of production, processing and marketing by inputting the product barcodes, which improved information transparency of the products and guaranteed the rights to know of consumers (Dewhirs et al., 2015).

## 3. Conclusions

Firstly, the performance requirements of traceability system were studied; then, the modeling theory of Petri net (applied in the modeling of traceability system) was analyzed (Sara et al., 2015). Furthermore, the customer demand for quality safety traceability system of fresh agricultural products was analyzed. Based on Petri net modeling method, this study implemented the design of information models for the key links (feeding, storage and transportation, marketing), and performed modeling analysis on the whole traceability system (Peyraud et al., 2011). On the basis of requirement analysis, system model and the overall goal of the traceability system, this study designed the technical route, functional module and software system architecture (Corazza et al., 2016). Finally, design and development of the traceability system were performed, including the development of production subsystem and processing subsystem. Via successful connection with the database, display interfaces for production information, query information and marketing information of fresh agricultural products were obtained (Wang et al., 206; García-Sandoval et al., 2016).

In summary, construction and implementation of the quality safety traceability system for fresh agricultural products not only improved the transparency of QS information of fresh agricultural products, but also enhanced information sharing and authenticity effectively (Chiang and He, 2010). With the help of quality safety traceability system, enterprises, supervision departments, sales departments and consumers could inquire the information of fresh agricultural products by entering barcode numbers, thus to safeguard their rights and interests.

## 4. Acknowledgements

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# REMISSION EFFECT OF SOY ISOFLAVONE ON SPORT FATIGUE AND ITS ACTION MECHANISM

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**ABSTRACT**

As the internal structure of mice is the most similar to that of human body, we used mice instead of humans for the experimental test and observed the anti-fatigue effect of soy isoflavone on the mice during swimming. Then, we further confirmed the remission effect of soy isoflavone on sports fatigue, which provided a solid theoretical basis for its application in sports. We selected the mice as the research objects and divided them into three groups. Through the swimming experiment, we observed the influences of soy isoflavone on hepatic glycogen content, muscle glycogen content, hepatic malondialdehyde and serum urea and the activity of serum lactic acid dehydrogenase in mice. Under the regulation effect of soy isoflavone, protein consumption of the mice reduced in the process of swimming, while fat increased, thus to provide energy for the body, relieve the sports fatigue and improve the exercise tolerance of the body. In addition, supplement of soy isoflavone could also improve the function of skeletal muscle mitochondria of the mice and reduce the generation of oxidative injury products; consequently, soy isoflavone was beneficial to easing the oxidative stress damage during sports. In conclusion, with a favorable regulating effect on the energy metabolism of mice during movement, soy isoflavone can effectively relieve physical fatigue.

## 1. Introduction

As our pace of life is speeding up, fatigue is becoming more and more common. For the people who play sports regularly, fatigue can cause a decline in their athletic ability as well as in their resistance. If body fatigue is not eliminated timely, it is likely that over-training syndromes and chronic fatigue syndromes will come up, which can decrease immunity of human body and affect body health. Therefore, research on how to relieve sport-induced fatigue is of great significance (Lombardi et al., 2009; Maes, 2011)

Widely and abundantly distributed in China, soybeans contain rich nutrients and bioactive substances, including soy isoflavone. With the ability to improve body's immunity, soy isoflavone has a variety of medical and health care effects, such as resisting oxidation, reducing blood pressure and regulating body's immune functions. One of the features of sports fatigue is that the body produces apparent oxidative stress, consequently, antioxidant ability of the body decreases and oxidative stress injury appears. In this case, soy isoflavone can play its favorable anti-fatigue role by inhibiting the oxidation (Rietjens et al., 2013; Akaza, 2012; Chang et al., 2016).

In recent years, a lot of experts and scholars analyzed and studied the application of soy isoflavone in resisting fatigue. In 2009, Cavallini et al. (Cavallini et al., 2009) pointed out that the mechanism of fatigue might be associated with the free radicals in human body; they further found that soy isoflavone could remove excess free radicals and increase

the activity of antioxidant enzymes, thus playing the role of oxidation resistance. In 2012, Hunerberg et al. (Flachowsky et al., 2011) found that soy isoflavone could extend exercise time and enhance the vitality of super oxide dismutase in human livers, which indicated its anti-fatigue function. In 2015, Gleason et al. (Gleason et al., 2015) revealed that soy isoflavone was similar with the excretive estrogen of human body in structure; it could improve the mineral content in bones and reduce the risk of osteoporosis.

Taking mice as the research objects, this paper explores the remission effect of soy isoflavone on physical fatigue of mice and its action mechanism, which provides a solid theoretical basis for the application of soy isoflavone in health food and medical domain.

## 2. Materials and methods

### 2.1. Experiment subjects

A total of 30 male mice, each of which weighted about 20 g, were selected. All the experimental procedures were performed according to the national health guidelines for management and application of experimental animals. In addition, the experiment received the approval of the National Ethics Committee.

### 2.2. Experimental groups

The 30 male mice were averagely divided into three experiment groups— normal control group, low-dose soy isoflavone group and high-dose soy isoflavone group. All the three groups of mice were free to drink water and eat for 15 days. Their weights and food intake were recorded every day. During feeding, the mice took 10-minute adaptive swimming for 3 times. After 15 days of feeding, all the experimental mice took loaded swimming (under 2% of their weights) in a glass swimming box (36 °C) for 60 minutes, and the data were collected according to their conditions at the 30th minute and the 60th minute of the swimming. After swimming, the mice were wiped dry; then, their heads were cut off to collect their blood, and the serum was isolated; moreover, their livers and skeletal muscles of hind legs were separated. Then, the materials were frozen in a refrigerator (-20 °C) for later use.

### 2.3. Experimental instruments and materials

The experimental instruments included: a microplate reader, a high-speed refrigerated centrifuge, a glass swimming box, a precise torque balance, a spectrophotometer and a vortex mixer.

The experiment materials included: soy isoflavone, blood glucose, serum urea, free fatty acid, lactic dehydrogenase and calcium lactate.

### 2.4. Determination of biochemical indicators

#### *2.4.1. Determination of blood glucose*

First, 10 μl of serum and 1 ml of working liquid were added into a test tube; they were mixed evenly with a vortex mixer. Then, the mixed liquid was kept under 37 °C for heat preservation for 15 min; reagent blank was adjusted to zero; blood glucose was determined by 505 nm colorimetric determination.

#### *2.4.2 Determination of serum urea*

(1) First, 20 μl of serum was added into a measurement tube; 20 μl of standard liquid was added into a standard tube; 20 μl of double distilled water was added into a blank tube. Respectively, 250 μl of buffer enzyme liquid was added into each tube and mixed with the former solutions by shaking up.

(2) Then, 1 ml of phenolic color developing agent and 1 ml of alkaline sodium hypochlorite were added into the three tubes in turn. Then, they were shaken up again and placed in 37 °C water bath for 10 to 15 minutes.

(3) After water bath, the test tubes were taken out and placed at the wavelength of 640 nm for colorimetric assay so as to determine serum urea.

#### *2.4.3. Determination of liver glycogen content and muscle glycogen content*

(1) The mice livers and muscles were washed with NaCL solution twice and wiped clean with filter paper. Respectively, 1 g of

liver tissue and 1 g of muscle tissue were accurately weighed and added into a test tube.

(2) Then, 3 ml of KOH solution (30%) was added into the tube which was then treated with 25-minute boiling water bath; then, the test tube was taken out and cooled off at room temperature.

(3) After cooling off, the liquid was transferred to a flask (50 ml) to reach a constant volume; then, the liquid was made into glycogen extract.

(4) The glycogen extract was placed in boiling water bath for 10 minutes; then it was taken out and cooled off. The blank tube was adjusted to zero point, and the absorbance was measured at the wavelength of 620 nm to determine liver glycogen content and muscle glycogen content.

#### *2.4.4. Determination of blood lactic acid*

(1) First, 20 ml of whole blood was drawn into the bottom of a test tube; 0.5 ml of NaF solution and 1.5 ml of protein precipitating agent were added into the tube; then, after being shaken up, the mixed liquid was centrifuged at 3000 r/min for 15 minutes. After centrifugation, the supernate was extracted.

(2) Then, 0.5 ml of mixed liquid (precipitant-NaF) was added into a blank tube; 0.5 ml of lactic acid standard application liquid was into a standard tube; 0.5 ml of supernate was added into a sample tube of fluid. Then, 0.1 ml of CuSO4 (4% ) and 3 ml of concentrated sulfuric acid were added into each tube which was fully shaken up and heated in boiling water bath for 5 minutes; then, they were taken out and cooled off at room temperature.

(3) Respectively, 0.1 ml of para-hydroxydiphenyl (1.5%) was added into each test tube which was shaken up again and placed in water bath (30 °C) for half an hour. Then, the tubes were taken out and heated in boiling water bath for 100 s; afterwards, the tubes were taken out and cooled off. At last, the absorbance was measured at the wavelength of 560 nm to determine the content of blood lactic acid.

#### *2.4.5. Determination of hepatic malondialdehyde content*

(1) First, 1 ml of 0.9 NaCL was added into the blank tube; 1 ml of homogenate was added into the sample tube; respectively, 2 ml of TCA-TBA-HCL was added into standard tube, blank tube and sample tube; then, the tubes were plugged and shaken up.

(2) The test tubes were placed in boiling water bath for 15 min; then, they were taken out and cooled off; afterwards, they were centrifuged at the speed of 4000 r/min for 15 min.

(3) Supernate was extracted from the test tubes. The absorbance was measured at the wavelength of 553 nm and the content of liver malondialdehyde was determined.

### 2.5. Data collection

Respectively, with relaxation edit pulse sequence and diffusion edit pulse sequence, the data of serum samples were collected and used for observation of micromolecule metabolites and lipid metabolites in the serum.

### 2.6. Statistical analysis

Statistical software SPSS17.0 was used for statistical analysis in the study. Experimental data were expressed in the form of mean ± standard deviation. Variance analysis was applied to between-group test. Least significant difference (LSD) method was applied to the comparison of every two items; $p < 0.05$ means there is statistical significance; when p is greater than 0.05, there is no statistical significance.

## 3. Results and discussions

### 3.1. Changes of mice weights

During the experiment, body weight growth and feeding conditions of the mice in each group were satisfactory. Body weight changes of the mice during experiment are shown in Figure 1.

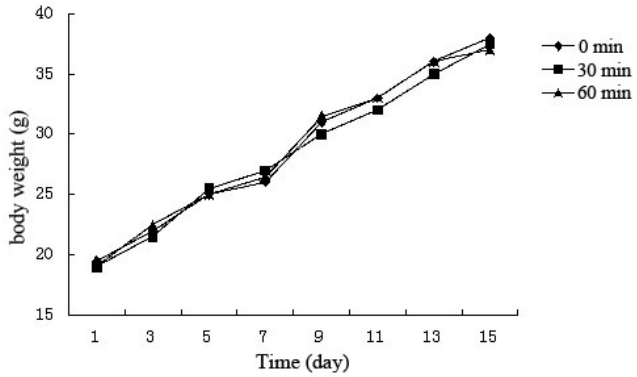


**Fig. 1** Line chart of the change in body weights of three groups of mice during experiment

According to Figure 1, the increase in body weights of the mice was not significant after swimming (in comparison with their weights before swimming), which indicated that the growth of the mice was not affected, with quantitative supply of soy isoflavone.

### 3.2. Influence of soy isoflavone on serum urea of mice

**Table 1.** Influence of soy isoflavone on serum urea of mice

| Experimental groups | Serum urea (mmol/L) |
|---|---|
| Normal control group | 8.5±1.3 |
| Low-dose soy isoflavone group | 6.8±0.9 |
| High-dose soy isoflavone group | 6.5±1.1 |

As can be seen from Table 1, either in low-dose soy isoflavone group or high-dose soy isoflavone group, the serum urea was lower than that of normal control group ($p < 0.05$); in addition, the serum urea of mice in low-dose group was close to that in high-dose group of soy isoflavone. The difference was of statistical significance.

### 3.3. Influence of soy isoflavone on liver glycogen content and muscle glycogen content of mice

In comparison with normal control group, liver glycogen content and muscle glycogen content decreased obviously in the other two experimental groups after swimming. By the 30th minute of the swimming, the decrease range of liver glycogen and muscle glycogen content in the two experimental groups was the greatest; afterwards, the content kept decreasing in a flattening trend, basically remaining unchanged as the level of the 30th minute (as shown in Figure 2 and Figure 3). The differences were of statistical significance.

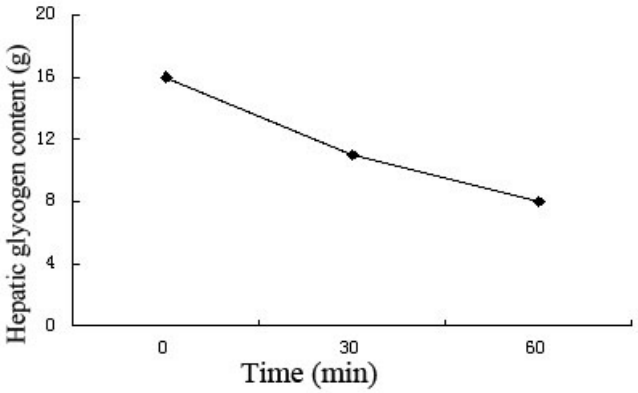


**Fig. 2** Influence of soy isoflavone on hepatic glycogen content in mice

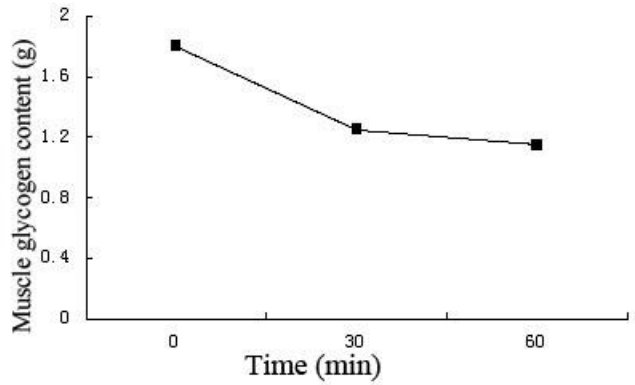


**Fig. 3** Influence of soy isoflavone on muscle glycogen content in mice

### 3.4 Influence of soy isoflavone on blood lactic acid content of mice

**Table 2.** Effects of soy isoflavone on blood lactic acid content of mice

| Groups | Blood lactic acid (mg/L) | | |
|---|---|---|---|
| | Before swimming | After 30-min swimming | After 60-min swimming |
| Normal control group | 313±122 | 302±96 | 296±85 |
| Low-dose soy isoflavone group | 258±71 | 201±72 | 185±67 |
| High-dose soy isoflavone group | 217±125 | 170±60 | 156±51 |

According to Table 2, in comparison with the situation before swimming, blood lactic acid content of the mice in high-dose and lose-dose groups of soy isoflavone was obviously lower than that of the mice in normal control group. As the swimming time increased, the content of blood lactic acid in mice was decreasing, and content of blood lactic acid of the mice in high-dose soy isoflavone group was much lower than that of the normal control group and low-dose group of soy isoflavone, which indicated that a certain amount of soy isoflavone had an influence on blood lactic acid of mice, and the difference was of statistical significance ($p < 0.05$).

### 3.5. Influence of soy isoflavone on hepatic malondialdehyde content of mice

**Table 3.** Soy isoflavone effects on mice liver malondialdehyde (MDA) content

| Groups | Hepatic MDA content (nmol/g) |
|---|---|
| Normal control group | 3.8±0.8 |
| Low-dose soy isoflavone group | 3.2±1.0 |
| High-dose soy isoflavone group | 2.8±1.1 |

According to Table 3, it can be observed that the liver MDA content of mice in low-dose group and high-dose group of soy isoflavone was lower than that of normal control group ($p < 0.05$). With the increase of the dose of soy isoflavone, the content of MDA in liver subsequently decreased faster, which was of statistical significance. The results indicated that the supply of a certain dose of soy isoflavone had an important influence on hepatic MDA content of mice.

Being a kind of complex physiological and chemical change in the body, fatigue can indicate a temporary decline in the original operation ability of the body; moreover, it is a previous sign of injury status that the body has developed into. On account that excessive fatigue has an impact on the physical and mental health of humans, how to relieve sports fatigue with proper remedy remains a problem for us to solve (Liao et al., 2012; Shi et al., 2012).

In this study, with the supply of different dosage of soy isoflavone on mice, we researched and analyzed the change law of serum metabolism and its influence by the 30th minute and the 60th minute of the loaded swimming of the mice (Han et al., 2016; Wang et al., 2013). Through the experiment, we observed the effects of soy isoflavone on the body weights, serum urea, muscle glycogen, liver glycogen, blood lactic acid and hepatic MDA of the mice (Ozden et al., 2013; Wang et al., 2016; Upadhaya et al., 2016). We found that a proper dose of soy isoflavone could delay the exhaustion time of the mice in loaded swimming and enhance their sports endurance. Liver glycogen content and muscle glycogen content are two important indexes reflecting the fatigue degree; therefore, we also observed the changes of the two indexes. The supply of soy isoflavone could slow down the digestion of liver glycogen and muscle glycogen of the mice after swimming, thus to provide more sufficient energy for the movement of the body, which was contributive to easing fatigue (Wang et al., 2016). The activity of serum lactic acid dehydrogenase of mice was obviously higher in high-dose group of soy isoflavone than that of the control group, which showed that soy

isoflavone could enhance the activity of blood lactic acid to some extent; consequently, it was beneficial to the decrease of fatigue and recovery of vitality of the body (Wiest, 2015; Banovic et al., 2010). Liver MDA content of the mice in low-dose soy isoflavone group was lower than that of control group, and the difference was more obvious between the high-dose group and control group. Accordingly, we speculated that with the application of soy isoflavone, the clearing of free radicals in the experiment subjects could be accelerated, thus to relieve fatigue (Yazdanbakhsh et al., 2015; Camblong et al., 2012).

To sum up, a proper dose of soy isoflavone has a relieving effect on sport-induced fatigue to some degree. The study on the action mechanism of soy isoflavone provides a persuasive theoretical basis for the remission of physical fatigue as well as a scientific basis for its application in the fields of health food and medicine.

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# STATUS OF ENGLISH TRANSLATION OF CHINESE DISH NAME AND ADAPTABILITY ANALYSIS

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**ABSTRACT**
This study aimed to discuss English translation of Chinese dish name in cross-cultural perspective by listing the most characteristic Chinese dishes and introducing language and cultural characteristics of Chinese dish name. First, background that causes the difference of Chinese and western food culture as well as the content of difference were discussed, followed by diet concept, social cultural implication, category and structure of raw materials and naming features of dish. Then based on the status of study on translation of diet name and the existing problems, necessity of translation standardization of dish name as well as principles, methods and contents were analyzed. Moreover, we discussed over English translation of Chinese dish name in intercultural communication context and under adaptability theory. At last, we pointed out the limitation of these English translation method, put forward the importance of transliteration and advised translators to show subjectivity.

## 1.Introduction

Language and culture are interdependent and integrated. As the important carrier of culture, language can reflect the difference of geographical environment, history, custom and concept of English countries and China. Language can not only reflect wonderful history and culture, but also express different living approaches and thinking pattern. Chinese language has been existed for a extremely long time. With the expansion of Chinese territory and development of trade in Han and Tang dynasty, Chinese characters were spread to other countries and produced certain effects on East Asia and Southeast Asia (Junun, 2013). In the interview for the original editor of Language and Characters Newspaper, Du Yongdao, Dingyangyang said that, Chinese Word Sea Dictionary published by Zhonghua Book Company and Chinese Friendship Press in 1994 including 85, 568 characters is a dictionary collecting the most characters (Yangyang, 2013). Compared to millions of English words, Chinese characters are of relatively small amount, but the meaning and culture implied is rich and distinctive. Chinese dish name is the most example. Chinese dish name is neat and orderly in form and distinctive in content, though it is simple; thus it is worth being concerned and studied.

American communication scholars Potter and Samovar believe that, the best explanation for intercultural communications is different recognition of social objects caused by culture difference (Manqiong, 2007). Moreover, Nida once said that, translation is communication (Lili, 2008). Translation of Chinese dish name not only involves simple conversion of Chinese and English words, but also is cross-cultural communication. Low efficiency of spread of Chinese catering industry oversea caused by low translation quality severely weakens international competitiveness of Chinese catering industry, and insufficient

understanding of ultimate objective of catering spread of translators goes against adjustment and optimization of national economical industry structure, which greatly blocks overall comprehensive development of economy; oversea spread of Chinese diet culture has not influence the world as expected (Xin, 2010; Xin, 2009; Xin, 2012). A key factor that determines the success of development of catering enterprise oversea is whether the concept insisted by the enterprise can be popular among foreign public which depends on the way expressing information of dish and foreign cultural emotional appeal by authentic target language.

Thus this study attempted to explore principles, method and strategy of translating Chinese dish name as well as status, adaptability and importance of translation standardization in intercultural perspective.

## 2. Difference of food culture between China and western countries

Chinese food culture is well-known, and about two thousand dished are developed from folk flavor (Xiaowen and Yifan, 2014). Western dish emphasizes nutrition of food and focus less on color, smell and form of dish. In cooking process, western dish lays emphasis on keeping nutritional component of raw materials; therefore, western catering is considered to be rational compared to Chinese dish which stresses sensory stimulation. On the contrary, sensuous Chinese catering excessively pursues for delicacy and beautiful form of dish and even ignores original color and flavor and nutritional component of raw materials. In Chinese food culture, eating is endowed rich cultural connotation by Chinese people and implies deep social significance that reflects national cultural psychology and recognizes the world, while western people consider eating as a behavior that ensures the normal operation of body. Raw materials of Chinese dish are diversified and not contraindicated, while some materials in western world are contraindicated due to humanity and history factor.

Difference of naming of Chinese and western dishes include the following points. First, Chinese dish is usually named considering rhyme and name structure matching as well as significance; naming of western dishes focuses on the complete expression of dish information, i.e., reality. As to food processing, Chinese dish pay great attention to diversity of matching of main ingredients and auxiliary materials and means of processing raw materials such as cutting, chopping, splitting, scraping as well as scaling, peeling, shelling and triturating. As to cooking method, Chinese dish emphasizes use of fire and dosage of seasoning, while western dish pay less attention on that. Main course, especially specialty (meat dish), is usually put on the most striking position of most menus used in Chinese restaurant, followed by stir-fry dish, cold dish, soups, snacks, drinks. Western dish menu usually put appetizers first, followed by soups, salads and entrees. Drinks are listed separately. As China is in line with the world, design of many Chinese menus gradually refers to western menu.

## 3. Standard of Chinese dish name transl ation in intercultural communication

### 3.1 Status of research of Chinese dish name translation

Nonstandard translation of Chinese dish name is common in translation activities in intercultural communication. Dish is often translated into different names as there is no reference to be used. Name of dish may be widely divergent even in different restaurants in the same area. Method of translation is also diversified, including transliteration, free translation and innovated translation. If not to be controlled, unified translated name will damage the image of Chinese culinary culture. Translated names that are used under different contexts are significantly different in word and structure now, which is hard for foreign people to know the dish translated into different names under different context.

### 3.2 Problems existing in name translation standard and the necessity

Appropriateness of translation of Chinese dish name remains to be enhanced. Blind translation should be avoided as it is bound to cause misunderstanding for translated names. The severest problems of intercultural communication of Chinese dish name is the disordered use of translated names and a unified and standardized translation method and term use principle has not formed. Translators with different theoretical level, practical experience and different view on translation objective will translate dish name in different ways, thus cause disordered translated names. Next we will analyze a piece of words from a paper On the Translation of Featured Dishes in Kangba (Rong, 2010) "dish name composed of cooking method and raw materials can be translated using literal translation, i.e., translating corresponding cooking method or cutting technique, then taking main materials as center word, and finally connect juice with preposition in or with"; with such a formula, most Chinese dish names can be translated, such as Kaoruzhu (roast suckling pig), kaoji (roast chicken), kaoquanyang (roast mutton), xunniurou (smoked beef), …, ganchaoniurousi (stir-fried yak shred), ganbianjiangdou (stir-fried cow pea), yangcongnouroupian (fried sliced beef with onion), hongshaoniurou (braised beef with brown sauce), ganbanniushe (Ox tongue in chili sauce), qingjiaoniurousi (stir-fried shredded beef with green pepper), …". On the premise of using the formula, the author translated the cooking method "kao" in the first three dishes into "roast". Here, the "roast" is not a verb but an adjective referring to being made by roasting. However, cooking method of the latter dishes "xun", "chao" and "shao" is translated into "smoked", "fried" and "braised", past participle of three verbs. By doing this, pre-and-post consistency of translation cannot be kept under one translation principle. Such translation formula is not beneficial to establishment of translation principle of Chinese dish name, let alone the establishment of translation standard for Chinese dish name.

### 3.3 Principle, method and content of translation standard

This study suggests to achieve standardization and unification of Chinese dish name translation with transliteration in intercultural communication of dish name. English name of every dish followed by several words of explanation and annotation is bound to lower ordering speed and affect economical benefits. Therefore, it is necessary to add explanation after dish name that emerges in the first time; and for those dish names that has been used for several years, explanation is not required. For example, like pot sticker and chow mein, spring rolls, Dan Dan noodles, Wonton, Beijing roast duck, general tso's chicken or governor Tso's chicken have been adopted and popular in Chinese restaurants over the world. Thus it is unnecessary to add explanation after dish name.

Transplanting word symbol or voice of source language into translated text breaks through the barrier of word symbol and lead readers to recognize and understand in the language and cultural environment of source language (Xin, 2010). Dissimilation value of transliteration only exists in language level. In the perspective of language, transliterated word is alienated; while in the perspective of culture, cultural meanings attached on terms mostly lose (Zhipei and Yunxiang, 2013). He Wu (Wu, 2010) once detailedly discussed over translation of Chinese dish name, especially foreignization and domesticating strategy and emphasized concerning about the relationship between dish names, culture and translation strategy, but he has not make an analysis of status of English translation of Chinese dish name as well as a detailed exploration of Chinese dish names of different types. The most appropriate words used to express special flavor of dishes, i.e., taste, smell and color should be found out from target language. For example, "cui" has corresponding words "crisp" and "crispy" in English and use of these

two words are usually mixed as they can be adjective and noun. Structure paradigm of Chinese dish translated name refers to the components that standard Chinese dish translated name should be included. To spread Chinese language and cultural characteristics more effectively, both realistic and impressionistic translated dish name can adopt the standard structure of "transliteration+ annotation/explanation". Explanatory translation can make western people understand the specific cultural imagery characteristics in Chinese language.

## 4. Adaptability theory

### 4.1. Adaptability of transliteration of impressionistic dish name

Yuan Na proposed multiple translation methods for Chinese dish names and detailedly discussed naming pattern and word formation means of English translation of Chinese dish name. She believed that, realistic dish names are mostly word group consisting of a modifier and the word it modifies and centers on noun, which is hard to operate (Na, 2012). The reason why impressionistic Chinese dish name is interesting is that, those names sounds elegant and contains a large amount of Chinese literary quotations, historical events, idioms, folk adage and proverbs and rhetorical device. When transliteration cannot give target audiences with enough information, it would be well to freely translate the dish name referred by impressionistic words with realistic translation. Transliterated words that have been entered English and accepted by English readers is few, though phonetic Chinese alphabet and English letters both belong to latin letter. Transliteration can maintain Chinese cultural elements implied in dish name to the largest extent indeed and arise readers' curiosity; however, transliterated words that have not been accepted by target audiences, such as Lvdagunr, Aiwowo and Long Kudou emerging without any explanation will confuse readers and make them unwilling to spend much energy and time on understanding Chinese dishes and Chinese culture contained in the names and even produce psychological resistance.

### 4.2 Adaptability of compound Chinese dish name in intercultural communicative translation

Compound dish name refers to dish names composes of main materials as well as metaphor, metonymy and exaggeration words for modifying auxiliary material, cooking method, shape and texture or those containing people's name and place name. It contains humanity of impressionistic dish name as well as actual connotation of realistic dish name. Besides several dish names that can be freely translated, others require to be added with explanation to make people understand information and culture therein. That also can be seen in western menu, for example, "Buffalo Shrimp - Crisp, golden brown fried shrimp tossed in a mild or hot Buffalo shrimp" and "Mediterranean Shrimp Pasta - broiled shrimp are served over tender linguini pasta in a basil pesto cream sauce, Topped with fresh Mediterranean tomato bruschetta" (Yingfang, 2012). It can be noted that, the way elaborating translating dish name in western dish, i.e., free translation of place name + main ingredient + explanation" is useful for translation of Chinese dish in intercultural communication. For instance, "Beijing kaoya" can be translated into "Beijing Duck---Beijing famous food, pulp delicate, flavor mellow & fat but not greasy", "Yangzhou Chaofan" into "Yangzhou Fried Rice or Yangzhou Chaofan - Yeung Chow Egg-fried Rice, healthy & tasty" using this principle. Dish name named by people's name can also be translated like that, for example, "dongporou" can be translated into "Dongpo Pork - Inspired by Su Dongpo, the greatest poet & calligrapher in Song Dynasty". Dish name expressed in such form is concise and comprehensive, highlighting regional characteristics and informing people with the cooking method, raw materials, eating method and quality of dishes.

## 5. Conclusions

Chinese dish as a means for delivering information and culture plays an important role in external exchange. Translation of Chinese dish name plays a unique role in catering culture guided by words. Translation of Chinese dish should use proper method considering the characteristics of dish name and eating environment and pay attention to cross-cultural awareness. Accurate and authentic Chinese dish name translation can not only express the cooking method and characteristics of dishes but also promote Chinese catering culture to go onto the world stage. However, the study has a limitation, i.e., cross-cultural awareness cultivation in Chinese dish name translation still remains in theory and idea, and whether it can produce better effect requires to be further analyzed in practice.

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# ENGLISH REPORT ON SIMULATION OF CANNED LIQUID FOOD STERILIZATION PROCESS

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**ABSTRACT**
Canned industry is a traditional export industry of China, and canned food is also an important export product of food industry at present. To acquire the best technological parameters of hot working, method of doing a large amount of experimental verification not only consumes much time and effort, but also requires enormous investment. Based on COMSOL Multi-physics field software and other novel research methods, this study simulated the sterilization process of canned liquid food and explored the influence of different viscosities on canned liquid food sterilization process. Results revealed that viscosity had great effects on temperature and speed during canned liquid food sterilization process, and greater viscosity tended to show a slower overall heating and smaller natural convective effect. Slowing heating zone (SHZ) caused by natural convection kept moving in the can, basically at 10~30% height. Besides, fatality rate in different positions rose as viscosity increased in the process of sterilization.

## 1.Introduction

Heat sterilization of canned food as a traditional processing technique is widely used due to its effectiveness and convenience (Farid and Abdul Ghani, 2004). Heated margin of canned liquid food near the wall of can makes the buoyancy decrease and move upward to produce natural convection during the heating process (Mermelstein, 1997), which makes temperature and flow speed of liquid inside can become quite complicated in the process of sterilization. Hence, it is extremely essential to simulate heat sterilization process numerically with the help of computer, thereby acquiring distribution rules of speed and temperature of liquid inside can.

Computer simulation refers to establish a mathematical model according to characteristics and requirements of system and solve the mathematical model on the computer to obtain information close to actual system. Its superiorities lie in improving economy efficiency, speedability and exhaustivity of test, shortening the progress of amplifying achievements of small test as large-scale industrial production and overcoming weaknesses during physical simulation, such as unchangeable input variables, difficulties in calculation and big error, which are convenient for studying stability and sensitivity of system as well as dynamic performance and control scheme, and meanwhile, looking for optimal plan (Anand Paul et al., 2011).

In recent years, computer simulation has been widely applied in canned food sterilization process. Simulating canned food sterilization process is able to predict changes of temperature distribution, flow speed, slowing heating zone (SHZ) and microorganism (Pedro et al., 2010; Pedro et al., 2010; Pedro and Marcelo, 2010; Feiruh et al., 2010; Selin et al., 2010; Hu, 2009). Taking water and carboxyl methyl cellulose (CMC) solution with different concentrations as

experimental materials, this study drawing support from COMSOL Multi-physics software simulates distributions of temperature and speed as well as microbial fatality rate with various viscosities and verifies simulation results through temperature experiment.

## 2. Materials and methods

### 2.1. Materials and instruments

Materials included purified water (Wahaha Company, China); carboxyl methyl cellulose (CMC); chemically pure (Sinopharm Chemical Reagent Co., Ltd, China); metal can (model: 307×113, Jinri Food Co., Ltd, Ningbo, China). Instruments were automatic autoclaves sterilizer (model: G154DWS, Zealway Instrument Inc., Xiamen, China); thermal characteristics analyzer (model: KD2 Pro, High Technology Co., Ltd, Beijing, China); Data Trace RF wireless real-time temperature sensor (Mesa Laboratories Inc., USA); manual can seamer (model: YJ-C200, Easy Jet Automation Equipment, Zhangjiagang, China); rotational viscometer (model: BROOKFIELD DV-+pro, Labthink Technology Instrument Co., Ltd, Shanghai, China); densimeter (model: YL, Yilian Control Temperature Apparatus Factory, Shanghai, China); electric drill (model: J1Z-BLT-65, Bailite Electric Appliance Co., Ltd, Shanghai, China).

### 2.2. Experimental methods

*(1)Measurement of basic heat transfer coefficients*

Density and viscosity of CMC solution (1% and 2.5%) under different temperatures were measured with densimeter and rotational viscometer respectively, and then KD2 Pro thermal characteristics analyzer was used in detecting thermal conductivity and atmospheric heat capacity value at room temperature. Basic heat transfer coefficients of water were provided by COMSOL Multi-physics software.

*(2)Thermal penetration test*

Purified water plus CMC solution (1% and 2.5%) at room temperature were poured into metal can that was punched in advance, Data Trace temperature probe was stretched to the geometric center of can and the probe was marked and taken down, then put back after can was sealed (Figure 1). Both of metal can and one temperature sensor were put into automatic autoclaves sterilizer and sterilized for 30 min at 121 °C, and then the probe was removed to read temperatures of sterilizer as well as central point inside can.

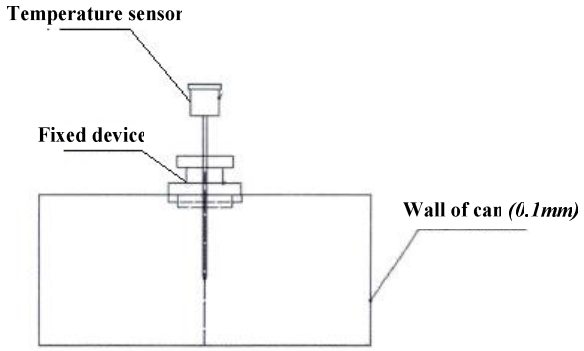



**Figure 1.** Temperature measuring device

*(3)Numerical simulation*

Temperature of sterilizer measured by wireless temperature sensor was led into COMSOL Multi-physics software by interpolation function for simulating heat transfer process using non-isothermal flow module (Adrian, 1993).

#### *2.2.1. Model assumption*

To simplify problems, the following hypotheses were made: liquid inside can was uniform and symmetrical; 3d cylindrical system was transferred into 2d axisymmetric processing; the influence of wall of can and probe on heat transfer was ignored; liquid had no slip on the inner wall of can; thermal conductivity and atmospheric heat capacity of CMC solution as constant values did not change with temperature; temperature on the outer wall of liquid was always equal to sterilizer's.

#### *2.2.2. Control equation*

Basic equation set of convective heat transfer was made up of heat exchange

differential equation, every direction momentum equation, equation of continuity and energy equation. As to simplified 2d convective heat transfer, equations are as follows (Abdul et al., 1999):

Equation of continuity:

$$\frac{1}{r}\frac{\partial}{\partial r}(r\rho v)+\frac{\partial}{\partial r}(\rho u)=0 \quad (1)$$

Where r refers to radial direction; z is axial position; u expresses axial velocity of fluid; v refers to radial velocity of fluid; ρ expresses fluid density.

Energy conservation equation:

$$\frac{\partial T}{\partial t}+v\frac{\partial T}{\partial r}+\mu\frac{\partial T}{\partial z}=\frac{k}{\rho C_p}\left[\frac{1}{r}\frac{\partial}{\partial r}(r\frac{\partial T}{\partial r})+\frac{\partial^2 T}{\partial z^2}\right] \quad (2)$$

Herein, T expresses temperature; t refers to time; k stands for heat conductivity coefficient; Cp is heat capacity at constant pressure.

Momentum conservation equation:

Momentum conservation equation in the Z direction:

$$\rho(\frac{\partial u}{\partial t}+v\frac{\partial u}{\partial r}+u\frac{\partial u}{\partial z})=-\frac{\partial p}{\partial z}+\mu\left[\frac{1}{r}\frac{\partial}{\partial r}(r\frac{\partial u}{\partial r})+\frac{\partial^2 u}{\partial z^2}\right]+\rho g \quad (3)$$

Where p refers to static pressure; μ stands for viscosity; g is gravitational acceleration.

Momentum conservation equation in the r direction:

$$\rho(\frac{\partial v}{\partial t}+v\frac{\partial u}{\partial r}+u\frac{\partial v}{\partial z})=\frac{\partial p}{\partial r}+\mu\left[\frac{\partial}{\partial r}(\frac{1}{r}\frac{\partial}{\partial r}(rv))+\frac{\partial^2 v}{\partial z^2}\right] \quad (4)$$

### *2.2.3 Boundary conditions*

Boundary conditions applied in model were shown below:

$T_w = T_{ret}$, u = 0 and v = 0 when r = R and $0 \le z \le H$ (5)

$T_w = T_{ret}$, u = 0 and v = 0 when z = 0 or z = H and $0 \le r \le R$ (6)

$\frac{\partial T}{\partial z}$ = 0 and v = 0 when r = 0 and $0 \le z \le H$

(7)$T_{in} = T_i$, u = 0 and v = 0 when $0 \le r \le R$ and $0 \le z \le H$ (8)

Herein, T w refers to temperature of liquid external border; Tret expresses temperature of sterilizer; R is radius of metal can; H stands for height of metal can; T in is temperature of liquid inside can; $T_i$ expresses initial temperature.

### *2.2.4. Grid division*

COMSOL Multi-physics grid generator was used in dividing the whole area into standard triangular grid.

## 3. Results and discussions

### 3.1.Analysis of basic heat transfer parameters

Water and CMC solution (1% and 2.5%) were applied in modeling liquid food to explore canned food heat sterilization process under various viscosities as CMC widely used in canned food performs important functions in thickening, emulsifying, holding water and suspending.

Thermal conductivity values of CMC solution (1% and 2.5%) at room temperature were measured with KD2 Pro thermal characteristics analyzer for 5 times (Table 1), showing no obvious significance. Average value 0.576 W/ (m × K) was taken as simulation parameter, and atmospheric heat capacity under the same condition was measured to be 4100J/ (kg × K).

**Table 1.** Thermal conductivity values of CMC solution (1% and 2.5%)

| CMC solubility | Thermal conductivity | | | | | Average value |
|---|---|---|---|---|---|---|
| 1% | 0.578 | 0.576 | 0.572 | 0.574 | 0.57 | 0.577 |
| 2.5% | 0.572 | 0.583 | 0.534 | 0.575 | 0.607 | 0.575 |

Figures 2 and 3 display change trend of density and viscosity of CMC solution in different temperatures, and Table 2 shows it fitting parameter values. Density fluctuation well meeting the linear equation was found, which was in line with the relationship that

density changed with temperature put forward by Adrian (Nelson et al., 2010). Besides, viscosity changing with temperature fitted quadratic function.

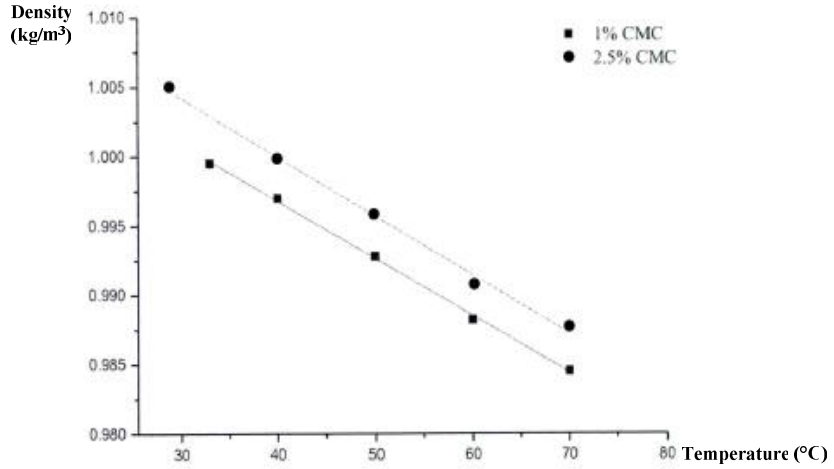



**Figure 2.** Changes of density of CMC solution (1% and 2.5%) with temperature

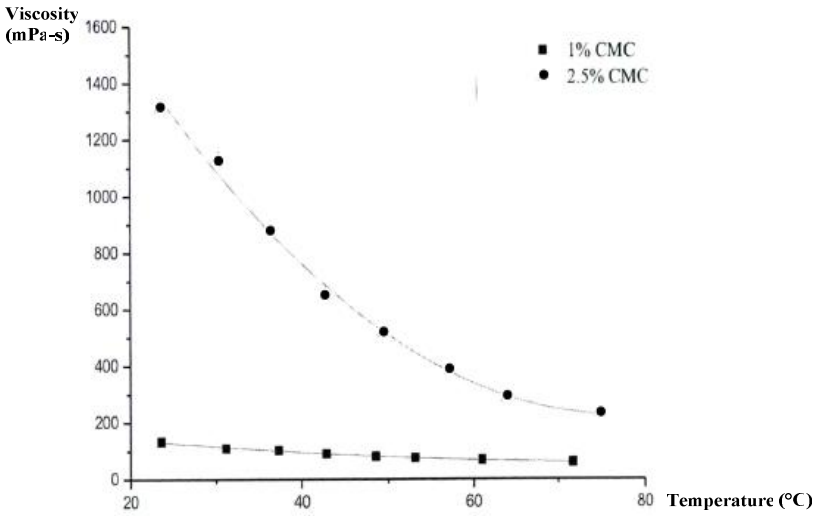



**Figure 3.** Changes of viscosity of CMC solution (1% and 2.5%) with temperature

**Table 2.** Fitting parameters of density, viscosity and temperature change

| Material | Density | Viscosity |
|---|---|---|
| 1%CMC | Y=-0.0004123x+1.012 $R^2$=0.997 | Y=0.0235x2-3.7126x+205.76 $R^2$=0.993 |
| 2.5%CMC | Y=-0.0000425x+1.0126 $R^2$=0.994 | Y=0.3963x2-60.792x+2561 R2=0.994 |

### 3.2. Comparison of model and experiment

It could be seen from Figure 4 that heating rate of water was higher than 1% CMC solution, and 1% CMC solution exceeded 2.5% CMC solution in heating rate, which might be caused by slow flow of liquid in the can with the increase of viscosity. However, in general, all the three heated up quickly and had significant convective heat transfer character.

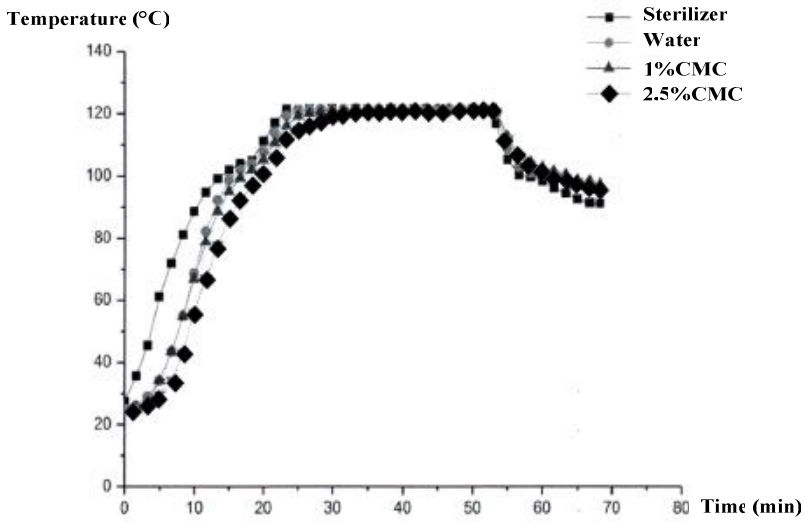



**Figure 4.** Temperature variation of water and CMC solution (1% and 2.5%) during sterilization

Figures 5, 6 and 7 display contrastive analysis of water and CMC solution (1% and 2.5%) in simulation and experiment respectively. For purified water, central point of model below 100 °C heated faster than experimental value, while both of them had a good fitting degree over 100 °C. It was possible that the probe interfered in the flow of liquid inside can below 100 °C, while liquid flowed intensively and the probe interfered slightly over 100 °C. As to CMC solution (1% and 2.5%), model heated up fast followed by slow comparing experimental value, which might be caused by inaccurate viscosity fitting formula, and a certain difference existed in temperature measurement range (20 °C ~ 80 °C and 121 °C) of viscosity used in this study. To improve the accuracy of model, measurement of physical parameters was required to increase to 80 °C, and even above 100 °C. In addition, heating curve of thermal convection model was not smooth enough, and jumpy, as neither grid of model nor time step were fine and dense enough. However, a finer grid and denser time step would increase calculation time and consume computing resources greatly, which is one of the shortcomings of finite element thermal convection model.

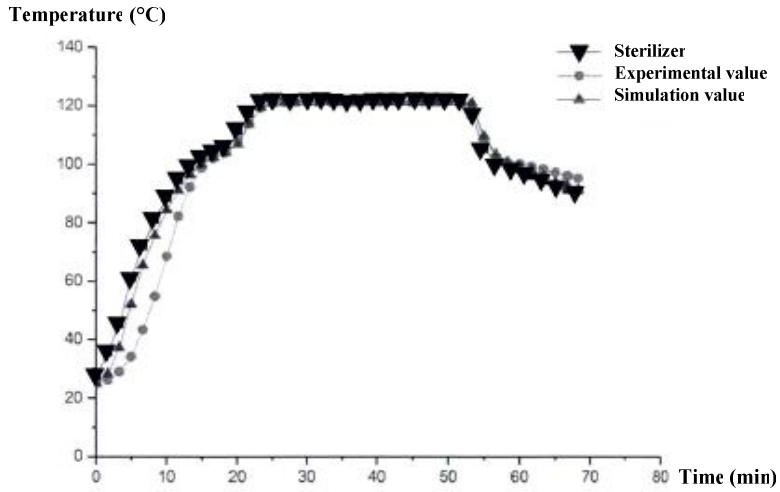



**Figure 5.** Temperature variations of purified water in simulation and experiment

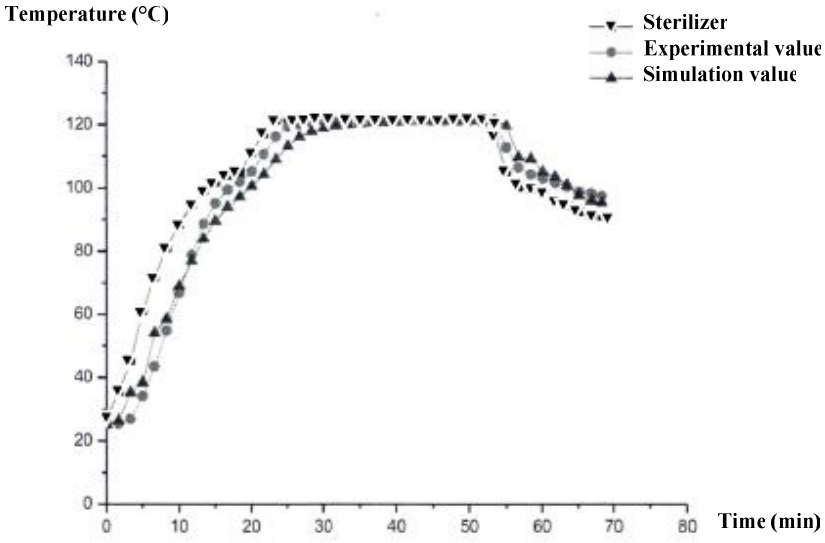



**Figure 6.** Temperature variations of 1% CMC solution in simulation and experiment

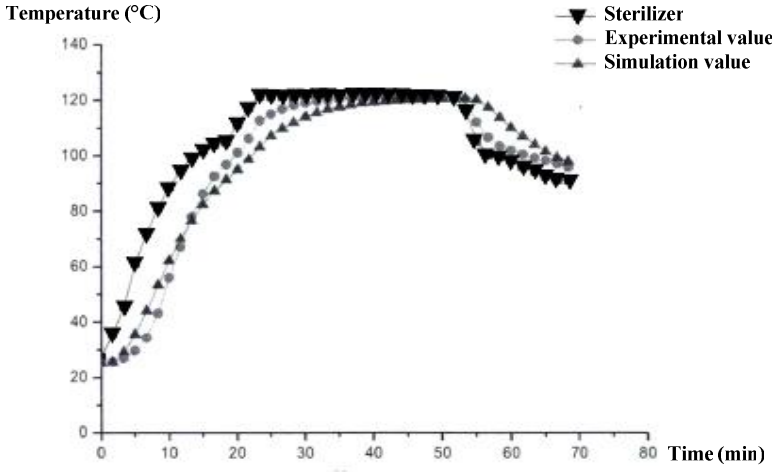


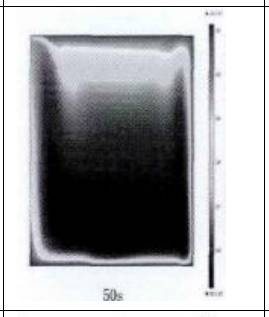
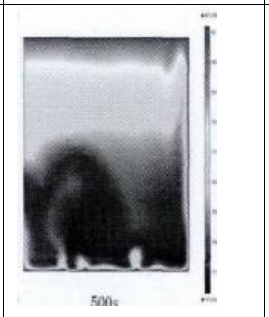
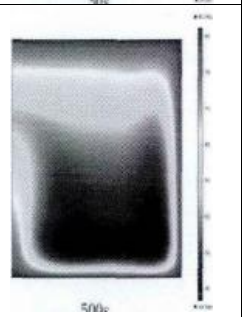
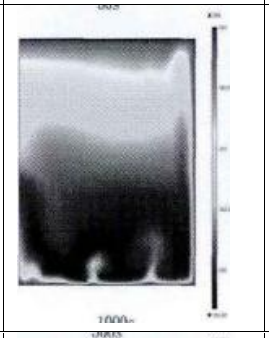
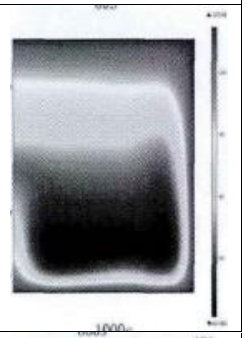
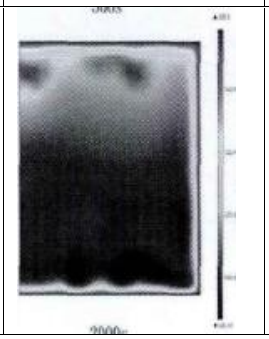
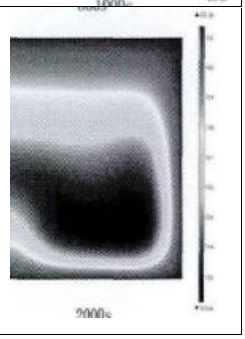

**Figure 7.** Temperature variations of 2.5% CMC solution in simulation and experiment

### 3.3. Temperature distribution

According to Figures 5, 6 and 7, model was basically identical with experimental value, so temperature distribution of each point inside the can in different times could be predicted with the help of the model. It could be seen from temperature distribution of water and 2.5% CMC solution inside can in 50 s, 500 s, 1000 s and 2000 s in Table 3 that natural convection inside pure water with relatively small viscosity had a strong influence on heat transfer when heating to 50 s, while its heat transfer of 2.5% CMC solution with large viscosity was closer to heat conduction. However, temperature went up constantly, viscosity was reduced and the influence of heat convection increased gradually as thermal sterilization proceeded. The position of SHZ inside can was found to be changeable, and was not in the geometric center of the can. It moved downward gradually. When heating to 2000 s, temperature difference of pure water inside the can was very small, while an obvious SHZ still existed in 2.5% CMC solution and was at 10% ~ 30% high, which was in line with research results proposed by Dimou (Datta and Arthur, 1988), Datta and Tekeira (Abdul Ghani

and Farid, 2007), together with Ghani et al (Abdul et al., 1999). Nevertheless, Ghani and Farid (Abdul Ghani and Farid, 2006) discovered SHZ in 30% ~ 35% high through simulating thermal sterilization of canned pineapple. It was likely to be caused by pineapple slice inside can which interfered in the migration of SHZ.

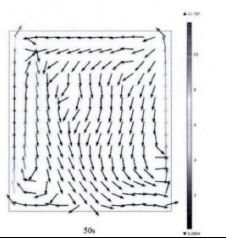
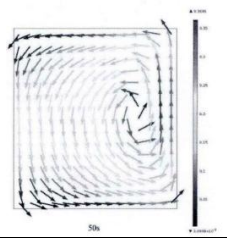
**Table 3.** Temperature distribution of water and 2.5% CMC solution inside can in 50 s, 500 s, 1000 s and 2000 s

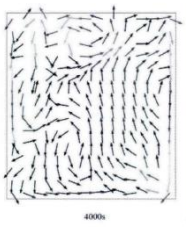
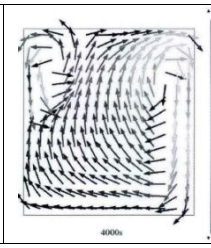
| | **Water** | **2.5%CMC** |
|---|---|---|
| 50s |  |  |
| 500s |  |  |
| 1000 s |  |  |
| 2000 s |  |  |

### 3.4. Velocity distribution

To better reflect liquid flow inside can in heating and cooling section during the sterilization, Table 4 displays velocity field distribution of water and 1% CMC solution in 50 s and 4000 s. It was observed that liquid near the wall of can flowed upward when heating to 50 s, and liquid near the can intermediate shaft flowed downward; when heating to 4000 s, liquid close to the can wall and can intermediate shaft flowed downward and upward respectively. During the sterilization process, water flowed strongly, while CMC solution was gentler, which was induced by increased temperature, expanded thermal volume and decreased density as liquid near wall edge was heated first. At this moment, the liquid away from wall edge had not been heated yet and was in a low temperature condition due to heat transfer time difference. Heated liquid near the wall edge was floated upward due to buoyancy which was brought by density changes, and moved to the axis direction as a result of rebound effect from the upper wall when getting to the top. However, the heavier liquid went down and touched the ground because of relatively low temperature inside can, thus generating thermal cycle (Nelson *et al.*, 2010). In the cooling stage, liquid close to can wall cooled first and density increased; and partial liquid started to flow upward once temperature went up. In 50 s and 4000 s, maximum flow speeds of water were 6.36 mm/s and 5.05mm/s respectively, and 0.394mm/s and 1.124mm/s for 1% CMC solution which were apparently smaller than water's.

**Table 4.** Velocity distribution of water and 1% CMC solution inside can in 50 s and 4000 s

| | **Water** | **1%CMC** |
|---|---|---|
| 50s |  |  |

| | | |
|---|---|---|
| 4000 s |  |  |

### 3.5. Changes of fatality rate

Referring to clostridium botulinum killing, Table 4 shows maximum and minimum fatality rates inside can after water and CMC solution (1% and 2.5%) finished sterilization. A great difference was found in minimum fatality rates of three, which was possibly because water had small viscosity, but flowed fast and transferred heat evenly; while higher concentration of CMC solution tended to show a greater viscosity and slower flow, thus leading to lagging heat transfer.

**Table 5.** Fatality rates of water and CMC solution (1% and 2.5%)

| Fatality rate (min) Tref = 121 °C, Z = 10 °C | Materials | | |
|---|---|---|---|
| | Water | 1%CMC | 2.5%CMC |
| $F_{max}$ | 35.13 | 35.05 | 34.73 |
| $F_{min}$ | 32.58 | 22.64 | 15.07 |

Note: Tref expresses reference temperature; Z is temperature that is required to be changed when sterilization time changes 10 times.

### 4. Conclusions

Based on COMSOL Multi-physics, this thesis sets up a simple 2d heat transfer model for simulating actual heat transfer process, and distributions of temperature, speed and microbial fatality rate in the whole heat transfer process are simulated using this model. Temperature and speed change sharply with the influence of viscosity during the sterilization process of canned liquid food, and greater viscosity tends to show a slower heating and smaller natural convection effect. During the sterilization process, fatality rate of canned liquid food in different positions rises as viscosity increases, which indicates that liquid food with higher viscosity is likely to induce insufficient sterilization in some positions in the static sterilization process. However, some excessively sterilized positions suggest that it is necessary to carry out rotational sterilization on food with high viscosity.

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# PROTECTIVE EFFECT OF *MOMORDICA GROSVENORI* LEAF EXTRACTIVE ON IMMUNE SYSTEM OF BODY WITH EXERCISE-INDUCED EXHAUSTION AND ITS MECHANISM

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*Immune organ.*

**ABSTRACT**

This study discussed over the efficacy of *Momordica grosvenori* leaf extractive flavones on the immune system of body through analyzing the effects of flavones on the immune system of body with exercise-induced exhaustion, aiming to provide certain scientific basis and theoretical basis for the development and utilization of economic value and healthcare value of *Momordica grosvenori*. Fifty rats were taken as the research subjects and evenly divided into five groups, 10 in each group. Each rat was gavaged with 200 mg/kg *Momordica grosvenori* leaf extractive flavones. Then the rats were trained for three weeks. The effects of *Momordica grosvenori* leaf extractive flavones on the immune system of rats with exercise-induced exhaustion were observed. It was found that, *Momordica grosvenori* leaf extractive flavones could effectively improve the duration of exhaustive exercise, increase the number of peripheral white blood cells, and increase the content of plasma indexes and it could also protect the immune system of body. *Momordica grosvenori* leaf extractive flavones is effective in improving fatigue resistance capacity and accelerating body recovery and moreover can promote the growth and development of immune organs and strengthen body immune functions.

## 1. Introduction

*Momordica grosvenori* is a special plant with important application value and nutritional value in Guangxi and has long been extensively applied in medicines, drinks and seasonings (Chang-Bao et al., 2012). Someone studied the chemical components of Momordica grosvenori leaf recently and found it contained rich flavonoid materials, especially kaempferol and quercetin. In vitro experiment suggested that, Momordica grosvenori extractive flavonoids have strong antioxidant activity and they are effective natural antioxidants which can remove free radicals, improve blood circulation, reduce cholesterol, inhibit the effusion of inflammatory bio-enzyme, promote the healing of wound and relieve pain.

Many people think that physical activity is sure to strengthen immunity (Miyazawa et al., 2015; Murakami et al., 2013). However, the opinion is of divergence because exercise only is not enough to improve the functions of immune system. Many experts and scholars hold that, exercise with different strengths can produce different effects on the immune system of body (Vlachopoulos et al., 2015; Wanner et al., 2014). For example, the high-strength and long-term exercise can inhibit the immune

functions of human body, induce the inhibition of exercise related immune functions, and increase the risks of virus infection. Therefore, it is of great significance to explore which substance can protect immune system. A report suggested that 10-day gavage of Momordica grosvenori leaf extractive flavones to rats significantly increased the proportion of a - Naphthyl Acetate Esterase (ANAE) positive cells and the proportion of rosettes formed in splenocyte, without affecting PPMNP, indicating Momordica grosvenori leaf extractive flavones could improve specific cellular immunity and humoral immunity on the condition of not affecting the non-specific immune function of body, which further revealed the regulatory and protective effects of Momordica grosvenori leaf extractive flavones on body immunity (Di et al., 2011; Dong-Lian et al., 2011; MO et al., 2013). The purpose of this study aimed at excavating and exploring the healthcare function and medical value of Momordica grosvenori leaf extractive flavones, especially the effect on exercise-induced immunity. Flavones extracted from Momordica grosvenori leaves were acted on rats with exercise-induced exhaustion. The experimental analysis proved the regulatory and protective effect of Momordica grosvenori leaf extractive flavones on the immune functions of body, providing a powerful theoretical and factual basis for its value development.

## 2. Materials and methods

### 2.1. Research subjects

Fifty healthy and clean male rats, aged 5 weeks and weighed 170 g, were selected. After adaptive feeding, five rats which were not suitable for treadmill exercise were excluded and classified into sedentary group. The remaining 45 rats were randomly and evenly divided into five groups, i.e., 5 for sedentary group, together with the five rats excluded, 10 for repeated exhaustive exercise control group, 10 for once exhaustive exercise control group, 10 for drug intervention combined with repeated exhaustive exercise group and 10 for drug intervention and once exhaustive exercise group. A model of the effect of Momordica grosvenori leaf extractive flavones on immune system of rats after exhaustive exercise was established; experimental treadmill for animals was used as the mode of exercise. All animal experimental operations were made according to the requirements of experimental animal management committee, reviewed and approved by the animal ethics committee and verified by pathologists.

### 2.2. The establishment of animal exercise model

(1) Except the sedentary group which was treated by conventional feeding but did not take adaptive training, the other four groups experienced three-day adaptive treadmill training;

(2) After three-day adaptive treadmill training, rats in the repeated exhaustive exercise control group and the drug intervention combined with repeated exhaustive exercise group experienced three-week exhaustive treadmill training additionally;

(3) Rats in the repeated exhaustive exercise control group and the drug intervention combined with repeated exhaustive exercise group experienced 6 times of exhaustive treadmill running in one week; at the $7^{th}$ day, training stopped and blood was collected from the rats.

(4) One day before dissection, once exhaustive treadmill exercise proceeded in the once exhaustive exercise control group and the drug intervention combined with once exhaustive exercise group; but before exercise, all rats were weighed;

(5) The determination criteria for whether rats exhausted included indifferent expression, dull reaction, running in a supine position, and temporary disappearance of righting reflex

### 2.3. The selection of observation indexes and test indexes

The duration of exhaustive swimming, plasma superoxide dismutase (SOD) activity,

and the content of methane dicarboxylic aldehyde (MDA) were detected and recorded.

### 2.4. Acquisition and processing of blood specimens

(1) Blood was collected from the exhausted rats using decollation; 4 ml of blood was taken and added into a centrifuge tube loaded with heparin;
(2) Then the blood was added into a large-volume low-speed refrigerated centrifuge and centrifuged at 3250 r/min for 13 min;
(3) The plasma was transferred into tubes respectively and stored in a cryogenic refrigerator as a preparation for the detection of plasma indexes and white blood cells.

### 2.5. The source of Momordica grosvenori leaf extractive and dose detection

#### *2.5.1. The source of Momordica grosvenori leaf extractive*

300 kg of Momordica grosvenori leaves were weighed and put into an extraction pot. After the addition of 3000 L of water, reflux extraction was performed for two hours, followed by cooling and filtration. The above procedures were repeated for four times.

The filter liquor flew through large pore resin absorption column in a proper speed; then it was washed by distilled water till colorless liquid flew out; then it was eluted by 75% ethyl alcohol.

Ethyl alcohol was recycled from the condensed eluent till solid-containing content became 20%; 10 kg of Momordica grosvenori leaf extractive flavones crude products were obtained after spray drying.

The crude products were mixed with 75% ethyl alcohol for reflux extraction. Filtration was repeated for three times. Ethanol solution was merged and condensed. Then ethyl alcohol was recycled till the solid-containing content became 30%. Finally, spray drying was performed. At the end of the experiment, 5 kg of Momordica grosvenori leaf extractive flavones competitive products was obtained.

#### *2.5.2. The determination of the dose of Momordica grosvenori leaf extractive flavones*

A certain quantity of Momordica grosvenori leaf extractive flavones was dissolved in a certain amount of water and then gavaged to rats in the drug intervention combined with once exhaustive exercise group and the drug intervention combined with repeated exhaustive exercise group (Genyk et al., 2016; Zehetner et al., 2015). Rats in the sedentary group, once exhaustive exercise control group and repeated exhaustive exercise control group were gavaged with the same dose of normal saline. The gavage was performed at noon 12'o clock. Half an hour after gavage, the rats did exhaustive treadmill training. The gavage was performed once each day, for three weeks.

### 2.6. Statistical method

Data were statistically processed using SPSS version 18.0. The measured data were expressed as mean ± standard deviation (SD). The comparison between groups was statistically processed using analysis of variance. The difference between groups was processed by t test. $p < 0.05$ meant difference had statistical significance.

## 3. Results and discussions

### 3.1. The change of duration of exhaustive exercise of rats under the effect of Momordica grosvenori leaf extractive flavones

Every experimental step proceeded according to the dose of gavage drug and exercise experimental scheme. The comparison between four groups suggested the effects of grosvenori leaf extractive flavones on the duration of exhaustive exercise of rats, as shown in Table 1. It could be seen from table 1 that, under the same training condition and feeding condition, the duration of exhaustive exercise of the drug intervention combined with exhaustive exercise groups was much longer than that of the exhaustive exercise control groups.

**Table 1.** The change of duration of exhaustive exercise of rats under the effect of *Momordica grosvenori* leaf extractive flavones

| Group | Duration of exercise (min) |
|---|---|
| Once exhaustive exercise control group | 31.42±18.13 |
| Drug intervention combined with once exhaustive exercise group | 49.25±17.37 |
| Repeated exhaustive exercise control group | 43.33±22.15 |
| Drug intervention combined with repeated exhaustive exercise group | 61.31±20.27 |

### 3.2. The change of plasma indexes of rats under the effect of different doses of Momordica grosvenori leaf extractive flavones

As shown in Table 2, the content of MDA of the once exhaustive exercise control group, drug intervention combined with once exhaustive exercise group, repeated exhaustive exercise control group and drug intervention combined with repeated exhaustive exercise group was significantly/extremely significantly lower than that of the sedentary group, but the differences between those groups were not statistically significant. The SOD activity of the drug intervention combined with once exhaustive exercise group, repeated exhaustive exercise control group, drug intervention combined with repeated exhaustive exercise group was much higher than that of the sedentary group ($p < 0.05$), and the SOD activity of rats taking drugs was not significantly different ($p > 0.05$).

**Table 2.** The change of rat plasma indexes under the effect of different doses of Momordica grosvenori leaf extractive flavones

| Group | Sedentary group | Once exhaustive exercise control group | Drug intervention combined with once exhaustive exercise group | Repeated exhaustive exercise control group | Drug intervention combined with repeated exhaustive exercise group |
|---|---|---|---|---|---|
| MDA | 3.84±0.94 | 2.42±0.44 | 1.87±0.63 | 1.67±0.43 | 2.95±1.21 |
| SOD | 150.93±8.55 | 144.49±14.53 | 171.78±11.19 | 171.72±8.26 | 167.23±8.55 |

### 3.3. The change of number of peripheral white blood cells of rats in groups under the effect of Momordica grosvenori leaf extractive flavones

The number of peripheral white blood cells was calculated through experiment. The effects of Momordica grosvenori leaf extractive flavones on rats in groups are shown in Table 3. Compared to the drug intervention combined with exhaustive exercise group, the number of peripheral white blood cells of the exhaustive exercise control group was lower, i.e., the data of the drug intervention combined with once exhaustive exercise group and the drug intervention combined with repeated exhaustive exercise group were much higher than the once exhaustive exercise control group and the repeated exhaustive exercise control group ($p < 0.05$); compared to the sedentary group, the number of white blood cells of the peripheral white blood cells was higher, and the difference was statistically significant ($p < 0.05$).

**Table 3.** The change of white blood cells in peripheral blood of rats in groups under the effect of *Momordica grosvenori* leaf extractive flavones

| Group | The number of white blood cells ($1\times10^9$) | | | |
|---|---|---|---|---|
| | 0th day | 8th day | 16th day | 24th day |
| Sedentary group | 8.45±0.56 | 8.77±0.27 | 8.11±0.64 | 9.19 ±0.75 |
| Once exhaustive exercise control group | 8.27±0.53 | 8.73±0.55 | 8.85±0.29 | 9.54±0.37 |
| Drug intervention combined with once exhaustive exercise group | 8. 76±0. 43 | 8. 85±0. 61 | 9. 26±0. 43 | 9. 59±0. 29 |
| Repeated exhaustive exercise control group | 8.27±0.69 | 8.87±0.64 | 9.25±0.44 | 9.33±0.46 |
| Drug intervention combined with repeated exhaustive exercise group | 8. 27±0.93 | 9. 19±0. 52 | 9. 38±0. 27 | 9. 35±0. 26 |

*Momordica grosvenori* is one of the special fruits in China and its extractive has special healthcare function and high nutritional value; hence it plays an important role in medicine and healthcare. Why Momordica grosvenori is famous in China and oversea is in a direct correlation to its special healthcare function and high nutritional quality (Mohamed et al., 2012; Yoon et al., 2013; Wu et al., 2013; Allen et al., 2016). For this reason, it is also called east magical fruit in China. We all know that, high-strength or long-term exercise may damage normal physiological balance and even result in the failure of self repair (Quinn et al., 2012; Damasio and Damasio, 2015; Zainol et al., 2012), thereby greatly affecting the health condition of body. Therefore, one hot research topic is the discovery of nutritional substance which can prevent tissue cells from damage, improve physiological tissue of various systems, relieve fatigue during body activity (Segizbaeva et al., 2013; Ferraresi et al., 2015; Leal et al., 2010), protect the health of body, and enhance exercise performance.

As to the change of the number of peripheral white blood cells after exercise, most of experimental results suggested that, the number of white blood cells increased after exercise, which resulted in the isolation of white blood cells from endothelial cells of blood vessel wall (Ronco et al., 2011; Icardo et al., 2012; Jagadeesha et al., 2011; Nugent et al., 2012) and the release of white blood cells from liver and spleen into blood. The results of this study demonstrated the above opinion. The number of peripheral white blood cells of the two exhaustive exercise control groups was higher than that of the sedentary group; the number of peripheral white blood cells of the drug intervention combined with exhaustive exercise group was higher than that of the two corresponding exhaustive exercise group respectively, and the difference was statistically significant. Thus it can be inferred that, long-time administration of Momordica grosvenori leaf extractive flavones or exhaustive exercise can increase the number of peripheral white blood cells, but the long-time administration of certain dose of Momordica grosvenori leaf extractive flavones is more effective in increasing the number of peripheral white blood cells.

## 4. Conclusions

This study explored the effects of Momordica grosvenori leaf extractive flavones on immune system functions of rats by establishing a rat exhaustive exercise model and detecting relevant immune organ and cell indexes through experiment and found Momordica grosvenori had sound effect in strengthening the immune functions of rats. But due to the limitations of resources and conditions, there were some defects in the experimental and analysis process. We will further supplement and perfect the results in future experiments.

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# EFFECTS OF SOLID EXERCISE DRINKS ON BODY FLUID EQUILIBRIUM OF SPORTSMEN ENGAGING IN ENDURANCE EVENTS

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**ABSTRACT**

This study aimed at investigating the effects of solid exercise drinks with different components on the body fluid equilibrium of sportsmen engaging in endurance events and providing a more accurate, high-efficient and economic reference basis for the selection of exercise drinks in competition. Twenty male sportsmen engaging in endurance events were selected as research subjects. The effects of solid exercise drinks which contained low molecular sugar (L-CHO), high molecular sugar (H-CHO) and sugar + protein (CHO-Pro) and placebo (blank control) on sportsmen were investigated. Research results demonstrated that, the weight of sportsmen in different groups significantly decreased after physical activities ($p < 0.05$); plasma osmotic pressure and total protein were much higher after exercise compared to before exercise ($p < 0.05$), and the difference between sportsmen taking different fluids had no statistical significance; urine osmotic pressure had no obvious change after exercise, but the urine osmotic pressure of H-CHO group was much higher than that of the placebo blank group ($p < 0.05$); plasma sodium and chlorine showed no remarkable changes after exercise, and the differences between sportsmen taking different fluids had no statistical significance; serum potassium in all groups demonstrated an obvious increase after exercise compared to during exercise ($p < 0.05$), and there was no obvious difference between groups; urine sodium and chlorine showed no significant differences after exercise compared to before exercise, and urine chlorine of the H-CHO group was much lower than that of the placebo blank group ($p < 0.05$); urine potassium after exercise was much lower than that before exercise ($p < 0.05$), and the difference between different groups had no statistical significance; the level of lactic acid after exercise was significantly higher than that in static state ($p < 0.05$), and the difference between different groups had no statistical significance; pH value had no remarkable change; the duration of exercise in groups had no remarkable difference. Thus it can be concluded that, low molecular sugar drinks can effectively maintain the metabolic balance of water and electrolyte and sports drinks containing low molecular sugar, high molecular sugar and sugar + protein have similar performance in regulating acid-base balance and promoting exercise performance.

## 1. Introduction

Energy consumption, water loss and the increase of core temperature during exercise all can result in the decline of exercise capacity (Lafata et al., 2014). Sports drink as a kind of sports supplement can rapidly and efficiently help human body maintain or recover to an ideal state and supplement the energy consumed during exercise (Buxton et al., 2012). Due to the different energy supply methods and body fluid metabolism characteristics (Zhou et al., 2013), components of different sports drinks differ greatly. A large number of studies in this aspect have been carried out to

comprehensively and systematically compare and analyze the effects of different concentrations of electrolyte, different kinds of sugar and special components on the physicochemical level of human body (Attarzadeh et al., 2013).

Alexander et al. (Kratz et al., 2005) found that, among 140 marathon runners who did exhaustive exercise, 25% of the runners showed hypernatremia, 12% of them showed increased plasma osmotic pressure, 9% of them developed hyponatremia, and 16% of them had decreased osmotic pressure. Gonzalez-Alonso (1998) pointed out that, the ability for sportsmen to fulfill skilled movement decreased by 7 ~ 8% in dehydration but non-heat exhaustion state. Some researchers (Tam and Noakes, 2013; Seo et al., 2014] proposed that, exercise capacity was inversely proportional to temperature in dehydration state. Another study (Firsov et al., 2012) suggested that, there was a significant loss of water and electrolyte, but no obvious change of osmotic pressure, after normal people did one-hour exercise with strength of 60% maximum oxygen uptake; a loss of water which was 5% of the weight of body could result in a decrease of 20% ~ 30% of skeletal muscle function; a loss of 1 L of water could result in 8 times more heart rate every one minute, a decrease of 1 L of cardiac output and an increase of 0.3 °C of core temperature. The increase of core body temperature can further increase heart rate and pulmonary ventilation volume (Hayashi et al., 2011). Further loss of water can induce symptoms such as fatigue and heat exhaustion (Moreno et al., 2013). Perspiration rate is in direct proportion to exercise strength (Kounalakis et al., 2010).

All in all, sports drinks can supplement nutritional substances lost during exercise (Millar et al., 2014) and it is consistent with the physical characteristics of sportsmen and manual labor population and the targeted nutritional demand (Sibthorpe et al., 2011). This study first explored the composition proportion, ionic concentration and osmotic pressure of solid sports drinks, then performed exercise tests on 20 male sportsmen who engaged in endurance events and took solid sports drinks in different composition proportion, and finally detected various blood and urine indexes of the sportsmen.

## 2. Test materials and subjects

### 2.1. Preparation before test

Twenty male sportsmen who engaged in endurance events were selected as research subjects and all of them had normal index values. Three different kinds of solid sports drinks were selected from the list of centralized purchasing nourishment for national teams; besides, placebo was taken as a negative control. The three kinds of solid sports drinks contained low molecular sugar, high molecular sugar and sugar + protein respectively. The placebo was made from multiple kinds of sweetening agents dominated by aspartame. The sugar content of the placebo depended on the sugar content of sports drinks. The taste and flavor of the placebo and sports drinks should be kept consistent as far as possible. The drinks and placebo were mixed with pure water to prepare water solution with sugar content of 8%, following the principle of equal sugar content. The three kinds of sports drinks and sweetening agent were all in apple flavor. The main components of the drink water solution systems are shown in Table 1.

**Table 1.** Main components of different sports drinks

| Drink | L-CHO | H-CHO | CHO-Pro | Placebo |
|---|---|---|---|---|
| Types of sugar and protein | Low molecular sugar | High molecular sugar | Sugar + whey protein | / |
| Molecular weight of sugar | <1500 | 500000~750000 | <1500 | / |
| Sugar content (%) | 8 | 8 | 8 | 8 |

| | | | | |
|---|---|---|---|---|
| Protein content (%) | 0 | 0 | 2.68 | 0 |
| Other | $Na^+$, $K^+$, VitC, nicotinic acid, inositol, etc. | $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, etc. | $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, VitC, VitE, etc. | Asparta me, etc. |

The ion concentrations of the three kinds of sports drinks containing different components and placebo were detected using a fully automatic biochemical analyser; the osmotic pressure was detected using a freezing point osmotic pressure detector. The components are shown in Table 2.

**Table 2.** Components of the placebo and drinks

| Different components and ions | $Na^+$ (mmol/L) | $K^+$ (mmol/L) | $Cl^-$ (mmol/L) | Osmotic pressure (mOsm/L) |
|---|---|---|---|---|
| L-CHO | 21.6 | 2.25 | 14.5 | 98.32 |
| H-CHO | 13.60 | 2.35 | 6.6 | 52.34 |
| CHO-Pro | 4.66 | 10.76 | 15.1 | 420.35 |
| Placebo | 9.1 | 0.75 | 2.6 | 17.68 |

The 20 subjects aged from 19 to 27 years old. All of them did not take drugs that can affect sugar and lipid metabolism such as β receptor blocker, drugs that can affect liver and hepatic functions such as diuretic and antitubercular agent, β-lactam antibiotics and quinolones three months before test. One day before test, the subjects were asked to relieve the bowels after waking up in the morning and then their weight and body composition were detected. The whole test was performed in the conditions of constant temperature, humidity and pressure (average 25.06 ± 0.65 °C, 56.95 ± 5.30 % and 1004.13 ± 5.15 mbar). During test, the oxygen intake and diet were assigned to each sportsman uniformly. The exercise scheme designed by Nybo et al. (2009) was improved by combining cycle ergometer with progressively increased load to test the maximum oxygen intake (de et al., 2009).

## 2.2. Formal exercise test

This study adopted Cortex Metalyzer II-R for gas metabolism analysis (Hillman et al., 2012). The curve equation of oxygen intake and exercise load as well as the load corresponding to 70% of the maximum oxygen intake was determined through trend line. The subjects were tested by means of riding cycle ergometer under the condition of the maximum oxygen intake. According to crossover design, the test process was divided into four stages to reduce individual variation. Every stage was composed of exercise test (one day) and washout period (seven days). Fluids given to each sportsman every day were the same. The diet and exercise of sportsmen were monitored and recorded; besides, the sportsmen took standard diet every day to expel the influence of other variables except drinks. Two days before the first stage of exercise test, factors that can affect test indexes should be avoided. The last three stages of test should be kept the same with the first stage.

## 2.3. Blood and urine test indexes

### *2.3.1. The preparation of blood samples*

Firstly, 2 ml of venous blood was taken and transferred into an Ethylene Diamine Tetraacetic Acid (EDTA) anticoagulation tube. Blood glucose and blood lactic acid were detected after standing. Then 4 ml of whole blood was taken and put into a vacuum aseptic tube containing blood clotting catalyst. After 30-min standing, it was centrifuged at 3000 r/min for 15 min. Plasma and serum were

obtained after centrifugation and they were stored at – 20 °C for the detection of various indexes.

### *2.3.2. The preparation of urine samples*

Containers from the same batch were used to collect urine before and after exercise for the detection of the osmotic pressure of urine. The rest were stored in – 20 °C for the content detection of $Na^+$, $K^+$ and $C^-$.

### *2.3.3. Blood and urine test indexes*

Osmotic pressure: the osmotic pressure of the plasma and urine were tested using a freezing point osmotic pressure meter and the sample was added using a micropipettor. The average value of measured results was taken as the final result.

Blood lactic acid: 20 μl of the prepared venous whole blood was mixed with 40 μl of membrane rupture diluents. It was shaken constantly till the membranes completely ruptured. Then the content of lactic acid in the whole blood was detected using YSI1500 lactid acid detector and lactate dehydrogenase method.

Total protein: 1 mL of the prepared plasma was taken and the total protein content of the plasma was detected using a fully automatic biochemical analyser and colorimetric method.

Na+, K+ and Cl-: The ion concentrations of Na+, K+ and Cl- in the blood and urine samples were detected and the average values were regarded as the final results.

## 2.4. Statistical method

Data were statistically analyzed using SPSS ver. 17.0 and expressed as mean ± standard error (SE). Difference between groups was analyzed using group F test. Difference was considered as statistically significant if $p < 0.05$.

## 3. Results and discussions

## 3.1. Changes of water balance

### *3.1.1. Changes of weights of the sportsmen after the supplement of drinks*

The changes of weights of the sportsmen are shown in the Figure 1.

Figures 1 and 2 demonstrate that, the weights of the sportsmen significantly decreased after exercise; the sports drinks containing different components had no obvious influence on their weights ($p < 0.05$); the weight difference between the L-CHO group and the H-CHO group had no statistical significance ($p < 0.05$), so did the CHO-Pro group and the placebo group. It indicated that, the components of the three kinds of sports drinks produced no effect on perspiration rate compared to the placebo and moreover the weight loss percentage of sportsmen taking solid sports drink containing low molecular sugar was the smallest.

### *3.1.2. Changes of osmotic pressure after the supplement of drinks*

Before the supplement of drinks, the plasma osmotic pressure between groups had no statistically significant difference ($p > 0.05$). The changes of plasma osmotic in pressure are shown in Figure 3. Figure 3 demonstrates that, the plasma osmotic pressure at the three time points had no remarkable difference; compared to the static state, the plasma osmotic pressure during exercise had an obvious increasing tendency ($p < 0.05$); the plasma osmotic pressure of the four groups had no obvious difference ($p > 0.05$).

Before the supplement of drinks, the osmotic pressure of urine had no significant difference. The change of urine osmotic pressure is shown in Figure 4. Figure 4 demonstrates that, urine osmotic pressure had no significant change after exercise compared to before exercise; urine osmotic pressure of the four groups had no obvious difference; but urine osmotic pressure of the H-CHO group and placebo group had remarkable difference ($p < 0.05$, suggesting that urine ion concentration of the H-CHO group was much higher than that of the L-CHO group, which might be correlated to the weight change difference between L-CHO group and H-CHO group. There might be a correlation between weight, urine osmotic pressure and perspiration rate.

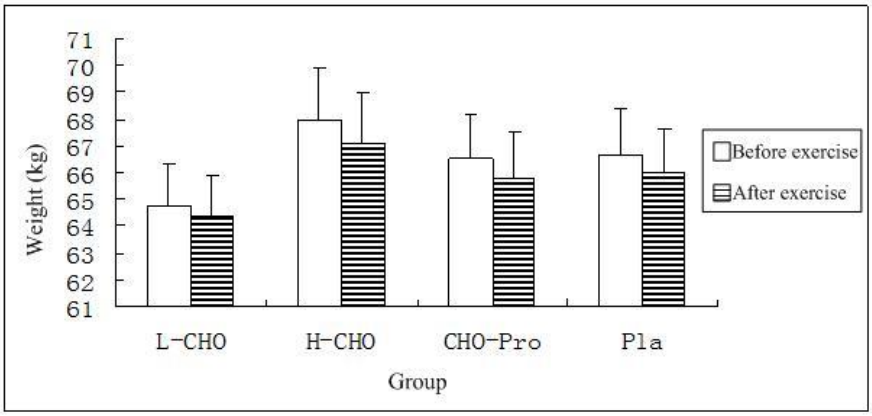


**Figure 1.** Effects of sports drinks containing different components on weight

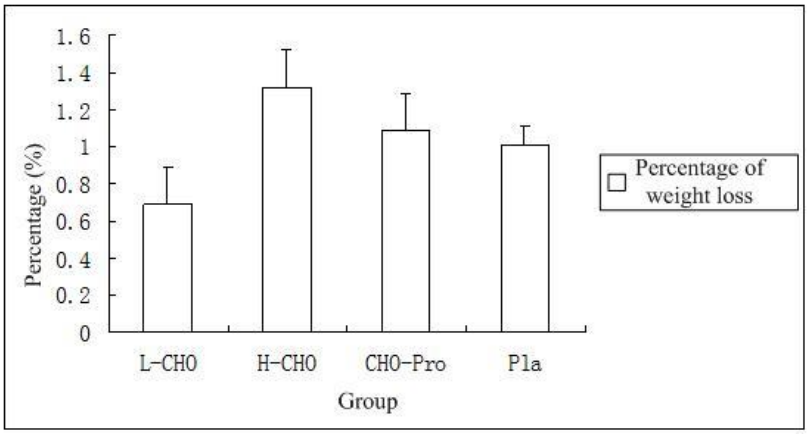


**Figure 2.** Weight loss percentages of sportsmen after exercise

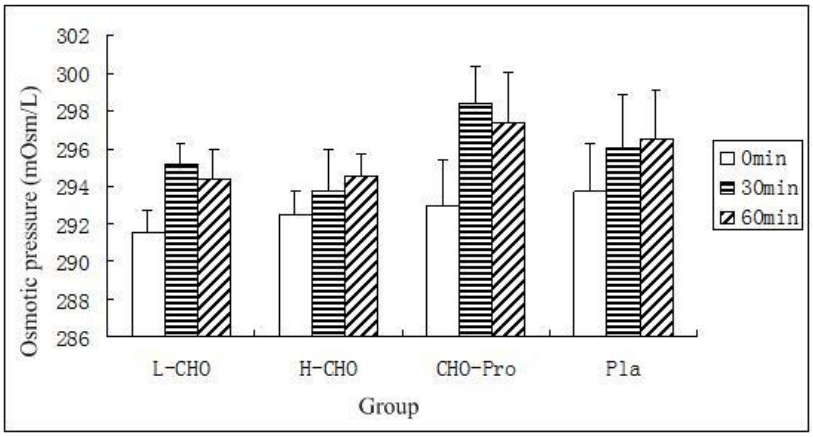


**Figure 3.** Effects of sports drinks containing different components on plasma osmotic pressure

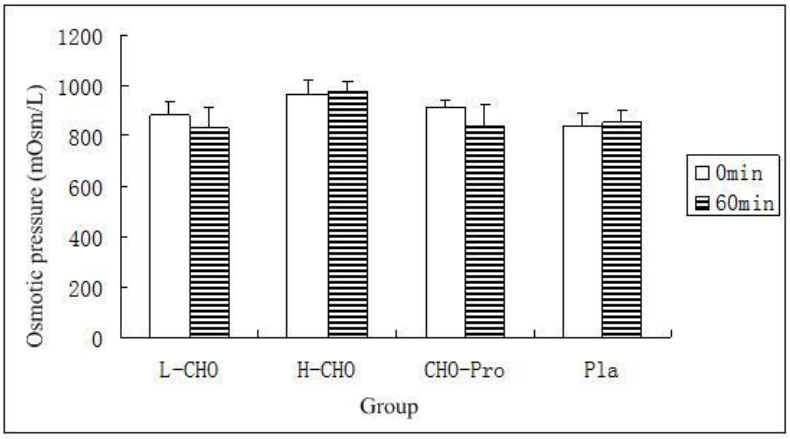


**Figure 4.** Effects of sports drinks containing different components on urine osmotic pressure

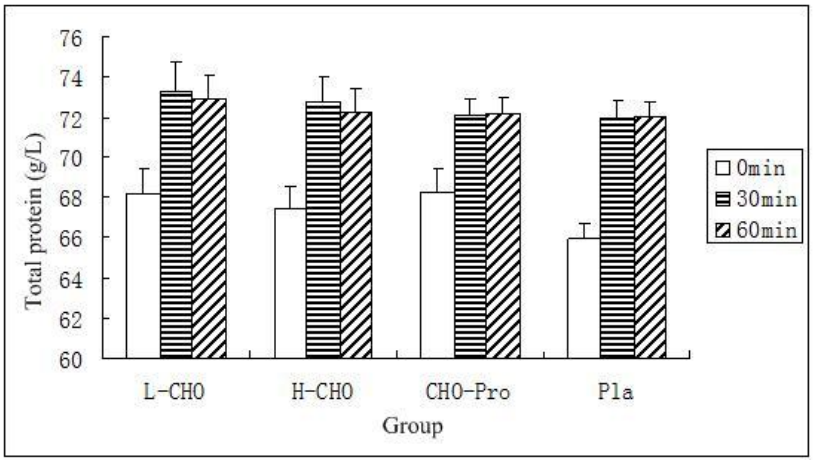


**Figure 5.** Effects of sports drinks containing different components on total protein in plasma

### *3.1.3. Change of total protein after the supplement of drinks*

The total protein in plasma of sportsmen had no statistically significant difference before the supplement of drinks. The change of total protein in plasma is shown in Figure 5. Figure 5 demonstrates that, the duration of exercise had an obvious effect on total protein in plasma ($p < 0.05$); compared to before exercise, total protein in plasma had an obvious increase after exercise ($p < 0.05$); the effects on total protein in different groups had no significant difference.

## 3.2. Change of electrolyte balance

### *3.2.1. Change of sodion in body fluid after the supplement of drinks*

The content of sodium in serum of sportsmen had no statistically significant difference before the supplement of drinks. The change of sodium content is shown in Figure 6.

It could be known from figure 6 that, the content of serum sodium of sportsmen had no obvious change after exercise; the content of serum sodium of the four groups had no obvious difference.

Before the supplement of drinks, the content of sodium in urine had no statistically significant difference. The change of urine

sodium is shown in Figure 7. Figure 7 demonstrated that, the content of urine sodium of sportsmen had no obvious change after exercise; the content of urine sodium of the four groups had no remarkable difference. It indicated that, the content of sodium in three kinds of drinks had certain effect on the concentration of serum sodium. The control group also contained sodium and moreover the concentration was higher than that of the CHO-Pro group; but the low osmotic pressure of the drink in the control group might dilute serum sodium, thereby resulting in the decrease of serum sodium concentration.

### *3.2.2. Change of potassium ion in body fluid after the supplement of drinks*

There was no statistically significant difference in the content of serum potassium before the supplement of drinks. The change of blood potassium is shown in Figure 8. Figure 8 demonstrates that, the content of blood potassium of sportsmen increased significantly after exercise ($p < 0.05$); the content of blood potassium in the late stage of exercise significantly increased compared to the middle stage of exercise ($p < 0.05$); the content of blood potassium of the four groups had no statistically significant difference.

The content of urine potassium had no statistically significant difference before the supplement of drinks. The change of urine potassium is shown in Figure 9.

Figure 9 demonstrates that, the content of urine potassium significantly increased after exercise ($p < 0.05$); the content of urine potassium in the four groups had no statistically significant difference. It indicated that, different fluid supplement schemes had no obvious influence on the concentration of serum and urine potassium.

### *3.2.3. Change of chlorine ion in body fluid*

The content of serum chlorine in the four groups had no statistically significant difference before the supplement of drinks. The change of serum chlorine is shown in Figure 10. Figure 10 demonstrated that, the serum chlorine had no significant change after exercise compared to before exercise; serum chlorine of the four groups had no remarkable difference. The content of urine chlorine had no statistically significant difference before the supplement of drinks. The change of urine chlorine is shown in Figure 11. Figure 11 demonstrated that, the blood chlorine had no obvious change after exercise, and the difference of blood chlorine between four groups had no significant difference; the urine chlorine of the H-CHO group was much lower than that of the placebo group ($p < 0.05$). It indicated that, the addition of a certain concentration of chlorine ion was beneficial to the stability of chlorine ion in body fluid.

## 3.3 Change of acid-base balance

The content of lactic acid in venous blood had no statistically significant difference before the supplement of sports drinks. The change of blood lactic acid is shown in Figure 12. Figure 12 demonstrates that, the content of blood lactic acid had no obvious change after exercise compared to that before exercise; in pairwise comparison, the content after exercise was much higher than that before exercise ($p < 0.05$). It indicated that, lactic acid produced during exercise and the production of lactic acid had little influence on endurance performance.

The pH value had no statistically significant difference before the supplement of sports drinks. The change of pH value is shown in Figure 13. Figure 13 suggests that, pH value had no obvious change after exercise compared to before exercise; the effects on pH value in four groups had no obvious difference, suggesting the effects of different sports drinks on pH value were basically consistent.

## 3.4. Change of exercise capacity

Exhaustive time of exercise capacity of sportsmen taking sports drinks containing different components had no statistically significant difference ($p > 0.05$), as shown in Table 3.

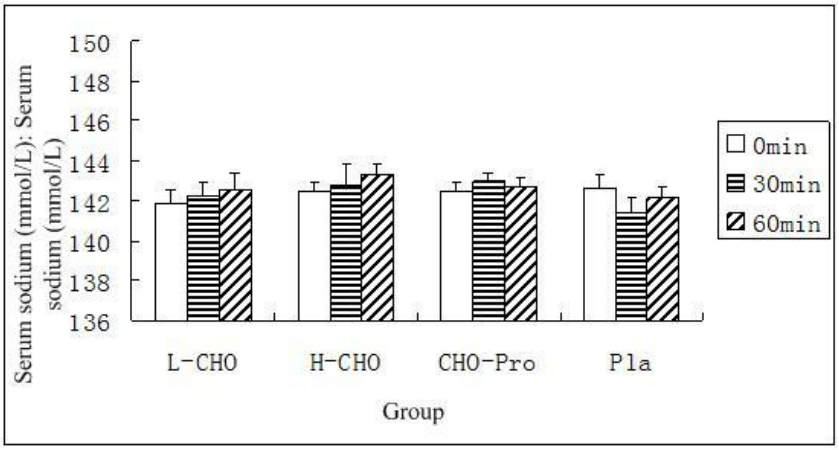


**Figure 6.** Effects of sports drinks containing different components on serum sodium

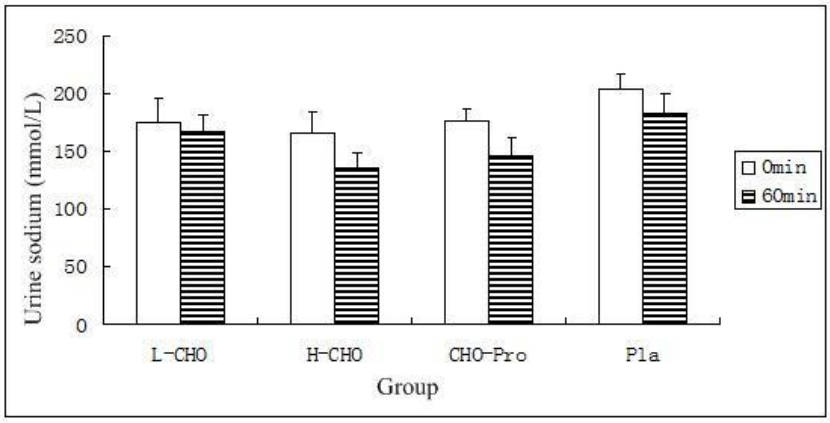


**Figure 7.** Effects of sports drinks containing different components on urine sodium

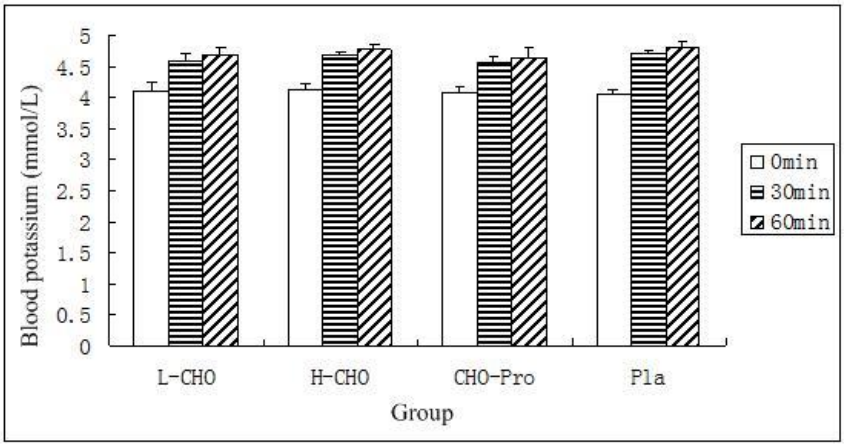


**Figure 8.** Effects of sports drinks containing different components on blood potassium

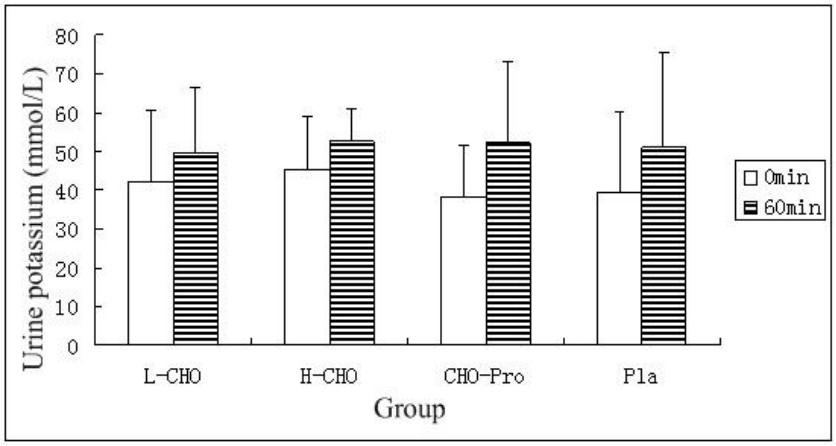


**Figure 9.** Effects of sports drinks containing different components on urine potassium

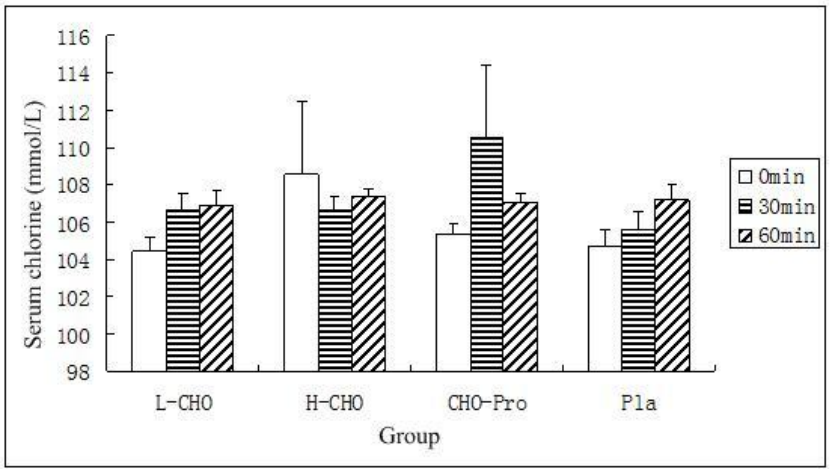


**Figure 10.** Effects of sports drinks containing different components on serum chlorine

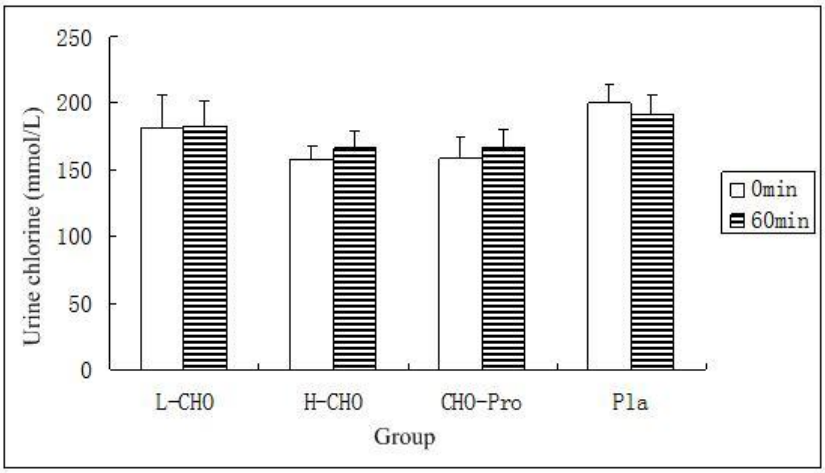


**Figure 11.** Effects of sports drinks containing different components on urine chlorine

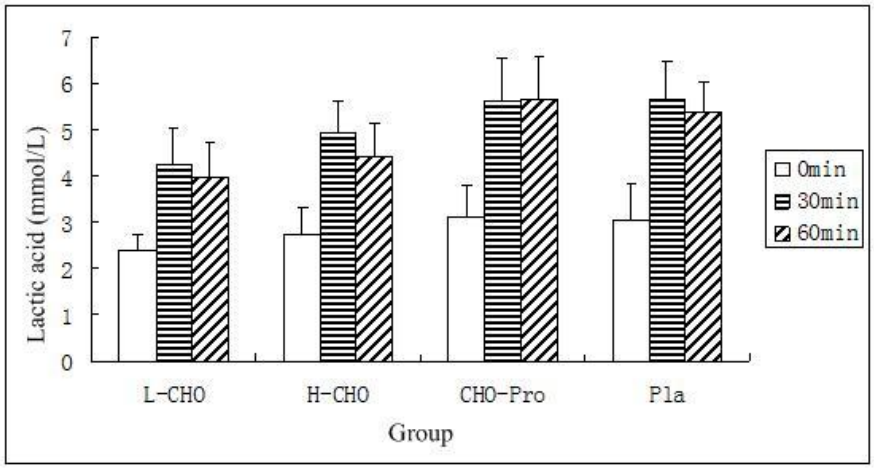


**Figure 12.** Effects of sports drinks containing different components on blood lactic acid

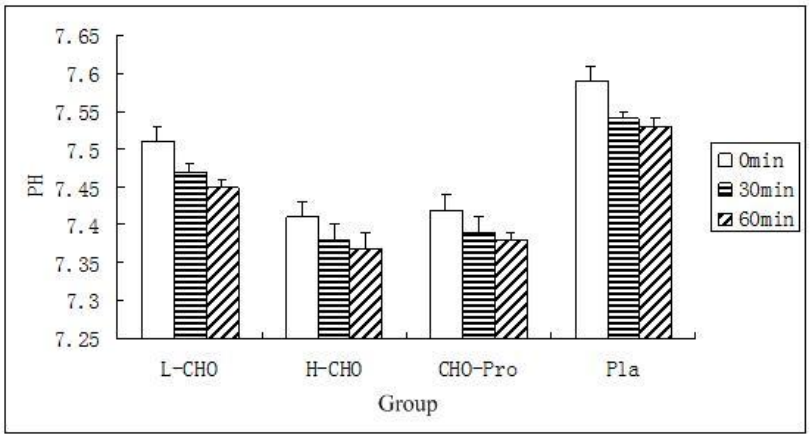


**Figure 13.** Effects of sports drinks containing different components on pH value

**Table 3.** Comparison of exhaustive time of sportsmen taking sports drinks containing different components

| Group | L-CHO | H-CHO | CHO-Pro | Placebo |
|---|---|---|---|---|
| Exhaustive time (s) | 3505.38±210.55 | 3317.68±273.21 | 3164.35±233.02 | 3237.34±179.78 |

It could be known from Table 3 that, the exhaustive time of sportsmen taking sports drinks containing different components had no statistically significant difference, suggesting the effects of different sports drinks on exercise capacity nearly had no difference.

## 4. Conclusions

To explore the effects of sports drinks containing different components on the body fluid balance of sportsmen engaging in endurance events, 20 sportsmen engaging in endurance events were given solid sports drinks containing different sugar components in this study. Research results suggested that, the supplement of L-CHO was more beneficial to the balance of water and electrolyte compared to other components; sports drinks containing three different components performed the same in regulating acid-base balance; sports drinks containing three different components had the same promotion effect on exercise capacity; the

three kinds of sports drinks has no effects on evacuation and absorption speed in gastrointestinal tract and subjective feeling during exercise.

The sports drinks analyzed in this study are of positive significance to the maintenance of acid-base balance and the prevention of hyperglycemia induced by long-term extensive exercise.

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# EFFECT OF DIETARY NUTRITION INTERVENTION ON THE PHYSICAL STAMINA OF FEMALE WRESTLING ATHLETES

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**ABSTRACT**

As the material basis for athletes to satisfy their nutritional needs and gain exercise capacity, diet nutrition can improve the metabolism process of the body and regulate organ functions. In addition, dietary nutrition is contributive to the recovery of physical stamina after exercise and the prevention of certain injuries. Therefore, only when dietary nutrition is taken seriously can the athletes bring better performance. In recent years, more and more scholars have paid close attention to the effects of dietary nutrition on physical stamina of athletes. In this study, we performed an eight-day investigation on the dietary nutrition status of 30 female wrestling athletes from a physical education school, which contributed to the monitoring on the daily dietary nutrition of the female wrestling athletes who took training every day. Thus we put forward targeted guidelines and effective suggestions on the unscientific diet habits of the athletes. Through the investigation, this study provided some basic data and suggestions on the problems regarding dietary nutrition and the influence of dietary nutrition on the physical stamina of athletes of different regions and different competition events.

## 1. Introduction

Through ingestion, digestion and absorption of nutrients from food, the body can function normally in vital movements such as growing development and metabolism (Jensen et al., 2013; Lima et al., 2012). The requisite nutrients for wrestling athletes can be supplemented by means of dietary nutrition intervention so that the wrestling athletes can achieve better performance and their somatic functions can recover more quickly after exercise. As the main source of athletes' nutrition supply, diet has been one of the main research issues in recent years.

For instance, Ho et al. (2013) suggested that wrestling athletes needed to consider the influence of their weight-loss modes on their physical fitness; during the weight control phase, their dietary arrangement and nutritional supplements should accord with scientificity and pertinence; for the tournaments, nutritional supplement of the wrestling athletes should be controlled according to their actual body weights; special nourishments were supposed to be supplemented according to the athletes' physical conditions and the specific problems emerging in the weight control phase. Based on the investigation on the diet structure, energy balance, body weights and body composition of teenager wrestling athletes and judokas as well as their ideal body weights and body fat percentages, Sled et al. (Sled et al. formulated an intervention recipe to improve the athletes' body composition. Eckner et al. (2011) stated that there was a gap between the physical function state of Chinese female wrestling athletes in the training period and that in pre-training period, which indicated that they needed to adjust their nutrition intake and resting schedules; they also found that

intervention with tea polysaccharide could promote the synthesis of red blood cells and hemoglobin during pre-match training period, protect skeletal muscle, cardiac muscle and liver from damage, improve the body's metabolic state and the rheological property of blood, and accelerate the recovery of body function. In this paper, we analyzed the influence of intervention of some dietary nutrients on the physical stamina of female wrestling athletes.

## 2. Subjects and methods

### 2.1. Subjects

In the study, we selected 30 female wrestling athletes (aged between 18 and 24 years old) from a school of competitive physical education. They were healthy without any medical history of acute or chronic diseases, including 15 top athletes, 8 first grade athletes and 7 second grade athletes; their average age was 21.24±2.4 years old; their average training duration was 5.12±1.29 years; their average height and weight were respectively (163.29±5.03) cm and (58.23±7.16) kg. They participated in the survey between March 21 and 28 in 2016.

### 2.2. Experimental instruments

The experimental instruments include an electronic balance, a weight scale, an energy monitor and a body composition analyzer.

#### *2.2.1. Inquiry method*

In a one-to-one manner, the athletes were inquired about their dietary structure and habits of the latest year. In addition, they needed to tell whether they took dietary supplements; if any, they would be inquired about the types and the dosage (Kitano *et al*., 2010; Pereira *et al*., 2014). By issuing dietary questionnaires, health conditions of the athletes were learned, including their dietary habits, food preference, whether they and their families had chronic diseases such as hypertension and hyperlipidemia.

#### *2.2.2. Weighing and recording method*

The investigators weighed and recorded the raw weight (before cooking) and cooked weight (before eating) of each meal of the wrestling athletes every day; moreover, they weighed the leftovers respectively. The common recording method of the weights is to record the dietary weights for three to four days, which is the most accurate and reliable method. However, it costs a lot of manpower and time, so it is not suitable for large-scale surveys.

#### *2.2.3. Chemical analysis method*

Chemical analysis method is applied by analyzing the subjects' daily food intake in a laboratory. It takes multiple procedures and the accuracy of measurement has to be guaranteed. In general, chemical analysis method is user-friendly without costing much manpower. It is suitable for long-term researches on diets; however, it is not as accurate as the weighing and recording method, which makes it unsuitable for in-depth studies on nutritional problems (Sun et al., 2011; Lee et al., 2015).

In summary, inquiry method is the most convenient and adaptable method for individuals; it is featured by high accuracy, while there might be great errors. Weighing method is accurate and suitable for the collective units, individuals and families with special requirements or for demanding researches. However, as it consumes a lot of time and efforts, it is unsuitable in the cases with large investigation scope and scale. Chemical analysis is a good choice when it is necessary to perform accurate measurement; however, it is disadvantageous in complex procedures and high cost.

#### *2.2.4 Statistical analysis*

Statistical software SPSS 17.0 was used for calculational analysis of the data. All the data were expressed as mean ± standard deviation.

**Table 1.** Basic information of the female wrestling athletes (N = 20)

| Age (years old) | Training duration (years) | Weight (kg) | Height (cm) |
|---|---|---|---|
| 21.24±2.4 | 5.12±1.29 | 58.23±7.16 | 163.29±5.03 |

**Table 2.** Body composition of the athletes in intervention group and control group before and after the experiment

| | Before experiment (intervention group) | After experiment (intervention group) | Before experiment (control group) | After experiment (control group) |
|---|---|---|---|---|
| Body weight (kg) | 63.75±6.27 | 58.93±8.31 | 61.94±7.26 | 57.81 ± 4.16 |
| Water content in the body (kg) | 36.61±8.38 | 37.59±4.04 | 38.12±4.19 | 38.55 ± 3.46 |
| Percentage of body fat (%) | 15.82±4.29 | 12.03±3.12 | 15.02±4.19 | 13.16 ± 3.19 |
| Weight of skeletal muscle (kg) | 29.01±3.77 | 26.11±4.16 | 27.88±2.91 | 23.02 ± 3.54 |
| Weight of body fat (kg) | 10.11±3.26 | 7.29 ± 2.44 | 10.02 ± 2.16 | 7.24±2.15 |

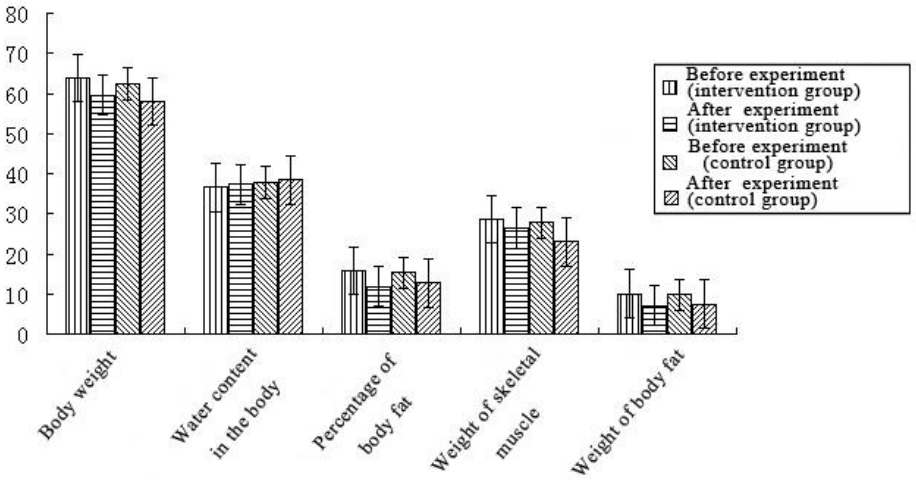


**Figure 1.** Body composition of the athletes the intervention group and control group before and after the experiment

Paired sample t test was used for the comparison between groups of the same type (including the intervention group and control group), while independent sample t test was used for the comparison between different types of groups (namely, the intervention group and control group); $p \leq 0.05$ means there is significant difference; $p \leq 0.01$ means there is insignificant difference.

## 3. Results and discussions

### 3.1. Body composition test

Table 2 shows the body composition of intervention group and control group before and after the experiment. Figure 1 shows the differences of data more distinctly.

**Table 3.** Recommended intakes of calories for the daily meals of the athletes and their actual intake

| Daily meals | Recommended intake (%) | Actual intake (%) |
|---|---|---|
| Breakfast | 25~30 | 27.1 |
| Lunch | 40 | 35.9 |
| Dinner | 30~35 | 36.3 |

### 3.2. Recommended intake of calories for the daily meals of the athletes and their actual intake

According to the recommended caloric intake proportions for daily meals shown in Table 3, it can be seen that the wrestling athletes' caloric intake for breakfast basically accorded with the recommended values; however, their caloric intake for lunch was insufficient compared with the recommended values, while their caloric intake for dinner was slightly beyond the recommended values. From the perspective of training, insufficient caloric intake of lunch could affect the wrestling athletes' performance for the whole afternoon, thus affecting the overall training effect.

### 3.3. Dietary nutrition intake of the athletes

Based on the data in Table 4, we found that an athlete's actual intake of calories was about 1361 Kcal higher than the recommended value; as for the protein intake, an athlete's intake was about 250 g more than the recommended value; moreover, fat intake of an athlete was nearly seven times the recommended value.

**Table 4.** Dietary nutrition intake of the athletes

| Category | Calories (kcal) | Protein (g) | Fat (g) | Sugar (g) |
|---|---|---|---|---|
| Actual intake (A) | 3950.23±578.92 | 331.24±100.69 | 574.29±234.39 | 602.29±133.78 |
| Recommended value (B) | 2589.12±148.29 | 80 | 80 | 550 |
| Difference value (A-B) | 1361.11±430.63 | 251.24.2±100.69 | 494.29±234.39 | 52.29±133.78 |
| T value | 11.791 | 12.823 | 10.927 | 2.103 |
| P value | 0.000 | 0.000 | 0.000 | 0.049 |

### 3.4. Changes of energy balance before and after intervention

Observing the data in Table 5, we found that after dietary nutrition intervention on the intervention group, energy intake and energy expenditure were basically in a balanced state, which showed remarkable effect in comparison with the unbalanced state in control group before dietary nutrition intervention ($p < 0.05$).

**Table 5:** Energy balance conditions of the wrestling athletes before and after dietary nutrition intervention

| | Sample size (people) | Energy balance before intervention (Energy intake- energy consumption) | Energy balance after intervention (Energy intake- energy consumption) |
|---|---|---|---|
| Intervention group | 15 | -165.8±153.15 | -19.1±91.25 |
| Control group | 15 | -161.2±133.96 | -165.9±122.91 |

## 4. Conclusions

Comparing the body fat percentages, skeletal muscle weights and body fat weights before and after the experiment, we found that there were decreases in the body weight, average body fat percentage and body fat weight in intervention group after the experiment and the differences were significant; there was no significant difference in water content of the body and skeletal muscle weight. The results suggested that through dietary intervention, the lean body mass of athletes was maintained well, while their body fat percentages were reduced significantly. In the low-speed weight control phase, there was no dehydration (Peterson et al., 2011; Mettler et al., 2010; Leblanc et al., 2011; Bann et al., 2014). As for the athletes in control group, their body weights, body fat percentages and body fat weights decreased obviously after the experiment, and the differences were significant. The athletes without systematic dietary intervention could reduce their body weight and body fat content; however, their skeletal muscle weights decreased greatly after the experiment, which caused the loss of lean body mass (Liu et al., 2014; Yoo et al., 2015; Saleh et al., 2014). In addition, some adverse conditions should also be taken seriously, such as dehydration and inorganic salt loss which might bring serious damage to the athletes' physical stamina.

In the process of investigation, we found that some wrestling athletes had unhealthy eating habits— they chose their favorite food over the food they disliked, which could result in nutrition imbalance. For a lot of athletes, meat consumption accounted for the majority of their diet structure while their intake of vegetables and fruit fell far behind. The reason why meat was their favorite was probably that they were affected by the custom of their families or hometowns or they misunderstood nutritional intake theory and considered that meat and other fatty food were more nutritional (Manabe et al., 2015; Lees et al., 2013). It indirectly indicated that the wrestling athletes, coaches and restaurant staff were lack of scientific knowledge of dietary nutrition, or perhaps they were unaware of the importance of dietary nutrition to athletes' physical stamina (Kim et al., 2012; Ganio et al., 2010) . After the investigation, it was suggested that the staff should lay emphasis on dietary nutrition intake, thus to avoid extremely unhealthy diet structure which would affect the training effect in future.

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# ANALYSIS OF RELIEF EFFECT OF CERVUS ELAPHUS BLOOD WINE ON PHYSICAL ACTIVITY INDUCED FATIGUE

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**ABSTRACT**
This study detected quality indexes of Cervus elaphus blood wine and analyzed its effect on physical activity induced fatigue of mice, providing a theoretical basis and data support for the further development and value improvement of Cervus elaphus blood wine. Several major quality indexes of Cervus elaphus blood wines from three batches were detected and analyzed. Besides, the effect of Cervus elaphus blood wine in relieving physical activity induced fatigue was analyzed by comparing the effects of Cervus elaphus blood wines in three different doses and wine base on the resistance of physical activity induced fatigue of mice with a wine base control group. It was found that, effects of Cervus elaphus blood wines in different doses on the weight of mice had no significant difference, but Cervus elaphus blood wine could effectively prolong the loaded swimming time of mice, significantly increase the level of hepatic glycogen of mice, reduce the content of serum urea and blood lactic acid of mice, and relieve physical activity induced fatigue of mice. Cervus elaphus blood wine containing many nutritional components such as amino acid, vitamin and lipid can enhance immunity and produce fatigue resistance effect. Cervus elaphus blood wines in different doses can relieve fatigue of mice to various degrees; hence Cervus elaphus blood wine has good edible value and broad market.

## 1. Introduction

Cervus elaphus blood refers to the blood of Cervus elaphus deer or red deer. A large number of animal experiments and clinical researches in recent years have suggested that, Cervus elaphus blood, in indeed, has many healthcare functions including maintaining beauty, keeping young, treating anemia, regulating immunity, delaying aging, relieving fatigue and improving memory, which provide a new development space and opportunity for Cervus elaphus industry and relevant medical and health care product industries (Gombotz, 2012; Liu et al., 2013; Cook et al., 2010). Wine made from Cervus elaphus blood, commonly known as healthcare wine, is a kind of valuable medicinal wine which integrates the edible value and medical value of Cervus elaphus blood; it has significant healthcare and medicinal efficacy and functions of enhancing immune functions, improving working ability, relieving fatigue, and improving sleep (Wiebrecht et al., 2014; Hou et al., 2011).

Physical activity induced fatigue refers to temporary decline of the maximum contraction or the maximum output power of muscle induced by physical activities. The occurrence of physical activity induced fatigue and the degree of fatigue are determined by considering the functional level and motor activity of

tissues and organs in human body. Causes for physical activity induced fatigue mainly include energy exhaust, metabolite accumulation and disordered endocrine regulation (Oliveira et al., 2013; Ishii et al., 2013). As the physical structure of mice is similar to that of human body, this study selected mice as experimental subjects for the analysis of anti-fatigue function of Cervus elaphus blood wine.

In recent years, many experts and scholars in China and abroad have explored the functions of Cervus elaphus blood. In 2015, Esbaugh A J et al. (Esbaugh et al., 2015) pointed out that, Cervus elaphus blood could promote the formation and maturity of T4 cells, which suggested it might have the function of enhancing the immune function of human body. Chang CW et al. (Chang et al., 2016] found that, oral administration of Cervus elaphus blood could accelerate the healing of ulcer and wound, strengthen the regeneration process of skin, further promote the healing of bone fracture as well as the metabolism of carbohydrate and nitrogen matters, improve memory, gastrointestinal function, sleep and vision, regulate blood pressure, resist radiation, reduce toxic and side effects of chemotherapy, and clear and nourish throat. It is of great significance to carry out studies on healthcare foods which are mainly made of Cervus elaphus blood and discuss over its processing techniques and specific healthcare functions, because those actions have important economic significance and scientific values to the improvement of deep processing level of Cervus elaphus blood and the expansion of its application approaches (Kanter et al., 2014). This work provides theoretical and data support for the further development and utilization of Cervus elaphus blood wine through analyzing the quality of Cervus elaphus blood wine and studying the effect of Cervus elaphus blood wine on the resistance of physical activity induced fatigue of mice.

## 2. Materials and methods

### 2.1. Experimental subjects and materials

(1) Cervus elaphus Linnaeus, Chinese date, arillus longan, Chinese wolfberry fruit and ginseng (Beijing Chinese traditional medicine wholesale market, China).

(2) Cervus elaphus blood wine (Yanghe Wine Co., Ltd., China).

(3) One hundred and sixty male Kunming mice, weighed 20 g, were randomly divided into four groups, i.e., wine base control group (wine base 15% v/v), Cervus elaphus blood gavage groups (gavage using Cervus elaphus blood wine in doses of 4.6 mL/kg.Bw/d, 9.2 mL/kg.Bw/d and 18.4 mL/kg.BW/d), 40 in each group. All mice could take food and water freely. All animal experiment operations followed the requirements of experimental animal management committee, reviewed and approved by the animal ethics committee and verified by pathologists.

### 2.2. Experimental instruments and reagents

#### *2.2.1 Experimental instruments*

Instruments included microbiological incubator (Shanghai Heheng Instrument and Equipment Co., Ltd., China), spectrophotometer (Hangzhou Qianjiang Instrument and Equipment Co., Ltd., China), alcoholmeter (Shanghai Shangbi Experimental Instrument Co., Ltd, China), atomic absorption spectrophotometer (AAS) (Beijing Dafengrui Instrument Co., Ltd., China), swimming box (50 cm × 50 cm × 40 cm), thermostat water bath (Beijing Xinbiao Tengda Instrument and Equipment Co., Ltd., China), second chronograph, electronic scale, etc.

#### *2.2.2. Experimental reagents*

Reagents included serum urea detection kit, hepatic glycogen detection kit, whole blood lactate detection kit, sodium sulfite, sodium hydroxide, absolute ether, sulfuric acid, acetonitrile, methyl alcohol, etc.

## 2.3. Preparation technique of Cervus elaphus blood wine

Firstly, serum was separated from Cervus elaphus blood and then the PH value of Cervus elaphus blood was decreased to 7. After one-day enzymolysis using protease, the Cervus elaphus blood was mixed with white wine (65% v/v) and immersed in a closed container. Fifteen days later, it was filtered after the removal of supernate.

Secondly, Chinese date and arillus longan selected were denucleated and beaten to pieces; ginseng, Chinese wolfberry fruit and cervus elaphus linnaeus selected were dried and grinded. All the processed raw materials were mixed up, mixed with white wine (65% v/v), and immersed in a closed container for 15 days. The mixture was stirred for 15 min every day. Prefiltration was performed fifteen days later.

The Cervus elaphus blood and raw materials which were processed by prefiltration were mixed up and blended with honey for the second filtration.

The liquid obtained after the second filtration was tested and encapsulated. Finally, Cervus elaphus blood wine was obtained.

## 2.4. Properties of Cervus elaphus blood wine finished products

Cervus elaphus blood wine obtained was red brown, clear, mellow, warm and tasty. Besides, it had functions of nourishing blood, boosting essence, promoting circulation of blood, removing stasis, eliminating swelling, resisting aging, maintaining beauty and extending life. The Cervus elaphus blood wine contained 80 % of water and 16% ~ 17% of organic matters; protein was the major component. The alcohol degree of the Cervus elaphus blood wine was 35% (v/v) and 1 ml of Cervus elaphus blood wine contained 0.97 g of herbal component. The wine had a high medical value and edible value.

## 2.5. Experimental method

### *2.5.1. Mouse loaded swimming experiment*

Each mouse was gavaged with 0.4 ml of Cervus elaphus blood wine, once each day, totally for one month. Half an hour after the last time of gavage, lead sheath whose weight was 5% that of a mouse was bounded on the tail of mouse. Then the mice were put into the swimming box. The swimming time of each mouse from the beginning of swimming to death was recorded.

### *2.5.2. Detection of serum urea of mice*

Half an hour after the last time of gavage, mice were put into the swimming box for 90 min-swimming.

At the end of swimming, the mice were taken out of the swimming box. Blood was collected from eyeballs of the mice after one hour of rest. After the blood froze, it was centrifuged at 14000 rpm. Ten minutes later, the upper-layer serum was taken for detection. Serum urea was detected using urea detection reagents.

### *2.5.3. Detection of hepatic glycogen of mice*

Half an hour after the last time of gavage, the liver was removed and put into a liquid nitrogen container for detection.

Then liver specimens were rinsed with normal saline. After the removal of water using filter paper, the weight of tissue was weighed precisely.

The specimen and alkaline liquor (weight of specimen: volume of alkaline liquor: 1:3) were added into a tube and cooked with boiling water. Twenty five minutes later, it was cooled using flowing water.

Finally, hepatic glycogen of mice was detected using glycogen detection liquid made of glycogen hydrolysate.

### *2.5.4. Detection of blood lactic acid of mice*

Half an hour after the last time of gavage, blood was collected from eyeballs of mice through angular vein using capillary glass tubes.

The mice were put into the swimming box for unloaded swimming after blood collection. Ten minutes later, blood was collected from eyeballs of the mice for the detection of the content of blood lactic acid.

After half an hour of rest, 20 μl of blood was collected from eyeballs of each mouse for the detection of blood lactic acid.

### 2.6. Data statistics

SPSS ver. 19.0 was used to statistically analyze experimental data. The difference between the experimental groups and the control group was analyzed; the relevant data were processed by one-way analysis of variance using SPSS ver. 19.0. The measured data were expressed as mean ± standard deviation (SD). The comparison between groups was statistically processed by analysis of variance. $p < 0.05$ indicated the difference had statistical significance.

## 3. Results and discussions

### 3.1. Detection of physicochemical indexes of Cervus elaphus blood wine (Table 1)

**Table 1.** Statistics of the measured indexes of Cervus elaphus blood wine

| Test item | Unit | Batch No. 20080310 | Batch No. 20080517 | Batch No. 20080819 |
|---|---|---|---|---|
| Cervus elaphus blood wine identification test | - | Positive | Positive | Positive |
| Panaxoside Re | Mg/L | 110 | 109 | 105 |
| The degree of alcohol (v/v) | % | 35.76 | 35.84 | 35.97 |
| Acetic acid | g/L | 1.35 | 1.43 | 1.39 |
| Methyl alcohol | g/mL | 0.6 | 0.6 | 0.5 |
| Lead | Mg/L | <0.1 | <0.1 | <0.1 |
| Manganese | Mg/L | <1 | <1 | <1 |
| Arsenic | Mg/L | <0.1 | <0.1 | <0.1 |
| Total number of bacterial colony | cfu/mL | <1 | <1 | <1 |
| Yeast | cfu/mL | <1 | <1 | <1 |

The above statistical data suggested that, all values of the indexes were up to specifications.

### 3.2. Detection results of microbacterial indexes (Table 2)

**Table 2.** Statistical table of detects results of microbiological indexes of Cervus elaphus blood wine

| Test item | Unit | Batch No. 20080310 | Batch No. 20080517 | Batch No. 20080819 |
|---|---|---|---|---|
| Total number of bacterial colony | cfu/mL | <1 | <1 | <1 |
| Coli group | MPN/100mL | <3 | <3 | <3 |
| Mycete | cfu/mL | <1 | <1 | <1 |
| Saccharomycetes | cfu/mL | <1 | <1 | <1 |

It could be known from Table 2 that, the total bacterial count was smaller than 1 cfu/ml, the total count of coli group was smaller than 3 MPN/100 ml, and the total count of mould and saccharomycetes was smaller than 1 cfu/ml, conforming to food microbiological detection standard.

### 3.3. Effects of Cervus elaphus blood wine on swimming time of mice

Mice were put into swimming box for loaded swimming.

Compared to the control group, mice in the experimental groups swam for a longer time, suggesting Cervus elaphus blood wine could slow down the appearance time of fatigue and strengthen the exercise tolerance. The time of loaded swimming was a key index for evaluating physical activity induced fatigue. The results of effects of Cervus elaphus blood wine on the swimming time of mice are shown in Table 3.

**Table 3.** Effects of Cervus elaphus blood wine on the swimming time of mice

| Group | Wine base control group | Low dose group | Medium dose group | High dose group |
|---|---|---|---|---|
| The number of animals | 10 | | | |
| Time of loaded swimming | 4.3±1.4 | 5.0±1.2 | 5.7±1.4 | 6.4±1.3 |

Through the results of the one-way analysis of variance, as shown in Table 3, we found that, the loaded swimming time of mice in the experimental groups had remarkable difference ($p < 0.05$) and the loaded swimming time of three experimental groups were longer than that of the wine base control group, suggesting the intake of Cervus elaphus blood wine could effectively improve the exercise tolerance of mice, prolong loaded swimming time and relieve physical activity induced fatigue. Besides, 18.4 mL/kg.BW/d was considered as the best intake amount.

### 3.4. Effects of Cervus elaphus blood wine on the weight of mice

Table 4 demonstrates the weight change of mice in the initial stage and later stage of test.

**Table 4.** Statistical table of weight of mice in groups in the initial stage and later stage of test

| Group | Swimming group | | | Serum urea group | | | Hepatic glycogen group | | | Blood lactic acid group | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | The number of mice (n) | Weight (g) | | The number of mice (n) | Weight (g) | | The number of mice (n) | Weight (g) | | The number of mice (n) | Weight (g) | |
| | | Initial stage | Later stage | | Initial stage | Later stage | | Initial stage | Later stage | | Initial stage | Later stage |
| Wine base control group | 10 | 21.1 | 35.0 | 10 | 21.1 | 35.5 | 10 | 21.0 | 35.6 | 10 | 21.5 | 35.5 |
| Low dose group | 10 | 21.3 | 35.0 | 10 | 20.9 | 35.4 | 10 | 21.1 | 35.4 | 10 | 21.6 | 35.4 |
| Medium dose group | 10 | 20.8 | 35.6 | 10 | 21.5 | 35.3 | 10 | 21.3 | 35.2 | 10 | 21.3 | 35.2 |
| High dose group | 10 | 21.0 | 34.8 | 10 | 21.3 | 35.0 | 10 | 21.1 | 35.2 | 10 | 21.3 | 35.7 |

Table 4 demonstrates that, one-way analysis of variance found that, the weight of the swimming group, serum urea group, hepatic glycogen group and blood lactic acid group was not significantly different with three dose groups, so did the three dose groups and the wine base control group ($p > 0.05$). These data indicated that, Cervus elaphus blood wine had no remarkable effect on the weight of mice.

### 3.5. Effects of Cervus elaphus blood wine on serum urea and hepatic glycogen of mice (Table 5)

One-way factor analysis of variance found that, the content of serum urea and hepatic

glycogen of mice between groups had significant difference; the content of serum urea in the high dose group was much lower than that of the medium dose group and the medium dose group. the content of serum urea and hepatic glycogen of the medium dose group was much lower compared to that of the low dose group after physical activity; the content of serum urea of the three dose groups was much lower than that of the wine base control group after physical activity; the level of hepatic glycogen of the three dose group was much higher than that of the wine base control group after physical activity; results indicated that, Cervus elaphus blood wine could effectively improve the adaptability of mice to exercise load, increase the level of hepatic glycogen, accelerate the removal of serum urea, and relieve physical activity induced fatigue.

### 3.6. Effects of Cervus elaphus blood wine on blood lactic acid of mice

One-way analysis of variance found that, the average value of area under the curve of blood lactic acid of mice in different experimental groups had remarkable difference; the area under the curve of blood lactic acid of the low dose group was much higher than that of the medium group before swimming, at the end of swimming and after rest and the area under the curve of blood lactic acid of the medium dose group was much higher than that of the high dose group in the three stages; the content of blood lactic acid of the three dose groups was significantly lower than that of the wine base control group. The findings suggested that, Cervus elaphus blood wine could effectively reduce the content of blood lactic acid after physical activity and relieve fatigue.

**Table 5.** Effects of Cervus elaphus blood wine on serum urea and hepatic glycogen of mice

| Group | Wine base control group | Low dose group | Medium dose group | High dose group |
|---|---|---|---|---|
| The number of mice (n) | 10 | | | |
| Hepatic glycogen (mg/g liver tissue) | 18.5±1.4 | 20.0±1.6 | 21.9±1.8 | 23.0±1.2 |
| Serum urea (mmol/L) | 9.0±1.1 | 8.1±0.9 | 7.2±0.3 | 6.8±0.8 |

Cervus elaphus blood wine made by novel preparation technique looks red-brown and clear, smells mellow and tastes soft, sweet and clear (Cui et al., 2013; Vilgis, 2013; Xiong et al., 2012). This study explored the effect of Cervus elaphus blood wine in relieving exercise induced fatigue by detecting and analyzing several major quality indexes of Cervus elaphus blood wine from three different batches and comparing the effects of Cervus elaphus blood wine in different doses and wine base on exercise induced fatigue (Peng et al., 2012). Each milliliter of the prepared Cervus elaphus blood wine contained 0.97 g of herbal components; there was 55 ml in each bottle and the degree of alcohol was 35% (v/v). Loaded swimming experiment was performed on mice and the content of serum urea, hepatic glycogen and blood lactic acid of mice gavaged with Cervus elaphus blood wine was detected. Results demonstrated that, the intake of Cervus elaphus blood wine could effectively strengthen the exercise tolerance of mice, prolong the loaded swimming time of mice, relieve exercise induced fatigue, improve the adaptability of mice to exercise load, accelerate the elimination of urea, reduce serum urea of mice, increase the level of hepatic glycogen, and effectively reduce the generation of blood lactic acid of mice. These results all suggest that, Cervus elaphus blood wine can relieve exercise induced fatigue, strengthen resistance, reduce the occurrence of fatigue phenomenon and improve athletic ability.

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# EXPERIMENTAL RESEARCH ON THE ANTI-FATIGUE EFFECT OF TRIBULUS TERRESTRIS IN SPORTS FOOD

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**ABSTRACT**

Objective: This paper aims to study the anti-fatigue effect of *Tribulus terrestris* in sports food, which provides theoretical and practical basis for the prevention of exercise fatigue. Methods: 24 healthy SD rats were divided into control group, exercise fatigue group and *Tribulus terrestris* + exercise fatigue group. After seven-day adaptive feeding, the two experimental groups took five-week increasing load treadmill training. The rats in *Tribulus terrestris* + exercise fatigue group took intragastric administration of *Tribulus terrestris* extract once a day before exercise. By the fourth week, 24 hours after the training, the blood was obtained from the orbital venous plexus of each narcotized rat; the liver and spleen of each rat were frozen for later use; their abdominal venous blood was collected; the numbers of natural killer cells and natural killer T cells of the rats were detected by a flow cytometer; the content of hemoglobin and muscle glycogen in the rats was detected by Sysmex automatic blood cell analyzer.
Results: In comparison with control group, there was an evident decrease in the weights and NK cell numbers of the rats in exercise fatigue group, indicating that in the exercise fatigue group, the exercise-induced immune of the rats with exercise fatigue was suppressed. In comparison with exercise fatigue group, numbers of NK and NKT cells of the rats in *Tribulus terrestris* + exercise fatigue group increased significantly (even greater than those of the control group), which showed that *Tribulus terrestris* extract could enhance the immunity of the rats which took excessive exercise and improve their fatigue resistance. In comparison with the control group, hemoglobin and muscle glycogen content of the rats in exercise fatigue group decreased more obviously, while a significant increase of hemoglobin and muscle glycogen level was found in *Tribulus terrestris* + exercise fatigue group (with no significant difference from the control group). Conclusion: *Tribulus terrestris* extract can improve the exercise-induced immune-suppression of the rats with exercise fatigue and enhance their anti-fatigue ability.

## 1.Introduction

Due to its distinct clinical effect and extensive pharmacological effects, *Tribulus terrestris* has been applied as a kind of medicine for a long time in China (Talasaz et al., 2010). Since the 1960 s, a lot of foreign scholars have studied the chemical composition of *Tribulus terrestris*; in China, the relevant studies were not carried out until the 1980 s (Hu et al., 2009; Berkman et al., 2009). Gradually, the researches on the composition of *Tribulus terrestris* have become more refined and penetrated. According to a large number of studies, it is generally recognized that *Tribulus terrestris* mainly contains compounds (saponin, flavonoid, biology class and polysaccharide) as

well as amino acids, terpenes, fatty acid, inorganic salt (Hamed et al., 2009; Ranjithkumar et al., 2013). Saponin compounds, whose content is the highest in *Tribulus terrestris*, are endowed with the advantages of depressurization, antitumor, strengthening sexual function, anti-aging and protecting myocardium (Chan et al., 2014).

*Tribulus terrestris* is widely applied in sports owing to the fact that it contains no prohibited ingredients while it can improve the blood testosterone level of the body and promote the synthesis of protein, thereby increasing the athletes' muscle strength and improving their athletic ability (Koncic and Tomczyk, 2013). A lot of studies have revealed that the saponin in *Tribulus terrestris* could increase the content of sex hormones in human body (Bourke, 2012); especially, it could increase the content of male hormones which contributed to enhancing anabolism, promoting the recovery of athletes and preventing exercise-induced fatigue. Wang et al. (Wang *et al.*, 2013) found that the nourishment containing *Tribulus terrestris* could improve the athletic ability of long-distance runners by improving synthesis and oxygen carrying capacity of their bodies. Esfandiari et al. (Esfandiari et al., 2011) learned that Tri*bulus terrestris* extract could inhibit the decomposition of protein and amino acid, promote protein synthesis and increase hemoglobin content. Borrione (Borrione et al., 2012) et al. revealed that *Tribulus terrestris* extract was able to increase hemoglobin content and glycogen reserve; in addition, it could accelerate protein synthesis as well as inhibit the decomposition of protein and amino acids, which further demonstrated the anti-fatigue effect of *Tribulus terrestris.* In view of the research situation, we selected 24 Sprague-Dawley (SD) rats as the subjects, aiming to study the anti-fatigue effect of *Tribulus terrestris* extract as a kind of sports food.

## 2. Materials and methods

### 2.1. Subjects

We selected 30 healthy male SD rats (180~220 g) from the Experimental Animal Center of Liaoning Province and purchased standard rodent feed. The rats were kept in a clean animal room (relative humidity: 45-65%; room temperature: 22-27 °C). After being fed adaptively for 7 days, all the rats were screened through exercise (15 m/min, 10 min/day), and 24 of them were selected for the experiment on account of their good adaptability. They were randomly divided into control group, exercise fatigue group and *Tribulus terrestris* + exercise fatigue group, each group including eight rats. No exercise load was exerted on the control group; 30 min before each training, each rat in *Tribulus terrestris* + exercise fatigue group was given intragastric administration of the solution (25 mg/ml) of *Tribulus terrestris* extract (total saponins > 60%), while the control group and the exercise fatigue group took the same volume of normal saline by gavage.

### 2.2. Experiment method

The rats in the control group were given conventional feeding without exercise; the rats in the other two groups took treadmill training bearing increasing load from Monday to Saturday every week and took rest every Sunday. The specific training plan is shown in Table 1.

**Table 1.** The training plan for the exercise fatigue group and *Tribulus terrestris* + exercise fatigue group

| Weeks | Intensity (m/min) | Gradient (°) | Exercise duration (min) |
|---|---|---|---|
| The first week | 15 | 5 | 25 |
| The second week | 20 | 5 | 45 |
| The third week | 25 | 10 | 65 |
| The fourth week | 30 | 10 | 85 |
| The fifth week | 30 | 15 | 85 |

Every Sunday, the rats in each group were weighed. By the fifth week, 24 hours after the final training, all the rats were treated with moderate anesthesia, and their orbital venous plexus blood was collected and preserved in a cryogenic refrigerator at -50 °C for later detection. Then, all the rats were put to death; their abdominal cavity vein blood, livers and spleens were all collected, labeled and preserved in the refrigerator at -50 °C for later use. The numbers of natural killer (NK) cells and natural killer T (NKT) cells of the rats in the three groups were detected: 100 μl of rat venous blood was added to a flow tube; with antibody labeling added, the flow tube was kept away from light for 30 min; 500 μl of Optilyze C hemolysis reagent was added; after shaking, the tube was kept away from light for 30 min; 500 μl of ISOTON III was added; then, the sample was detected by a flow cytometry.

In this study, Sysmex automatic blood cell analyzer was used to detect the hemoglobin (Hb) and muscle glycogen in rats. For the statistical analysis on the experimental data, SPSS 17.0 was used; the inter-group differences in mean values were compared using the method of single factor analysis of variance; the results were presented as mean ± standard deviation ($x \pm SD$); $p < 0.05$ means the differences were significant.

## 3. Results and discussions

### 3.1. Changes of apparent conditions and weights of the rats in the three groups

During the five-week exercise with increasing load, as the exercise time and intensity were under control, there was no death in the three groups. In the fourth and fifth weeks, all the rats in exercise fatigue group showed depression, slow reaction and hair loss; in *Tribulus terrestris* + exercise fatigue group, such phenomena were rare. Before the experiment, there was no difference in the weights of the rats in the three groups; since the third week of the experiment, weights of the rats in exercise fatigue group were obviously lower than those in the control group, the difference was significant ($p < 0.05$); the difference was the maximum at the fifth week. By supplementing *Tribulus terrestris* extract, weights of the rats in *Tribulus terrestris* + exercise fatigue group were markedly lower than those of control group ($p < 0.05$). In the fourth and fifth weeks, weights of the rats in *Tribulus terrestris* + exercise fatigue group were higher than those of exercise fatigue group; the difference was significant ($p < 0.05$). The results are shown in Table 2.

**Table 2** Weights of the rats in three groups with increasing load training and supplement of *Tribulus terrestris* extract (g)

| Week | Control group (n=8) | Exercise fatigue group (n=8) | *Tribulus terrestris* + exercise fatigue group (n=8) |
|---|---|---|---|
| One week before exercise | 197.82±7.15 | 199.21±9.36 | 200.84±4.58 |
| The first week | 262.26±19.78 | 257.18±12.21 | 260.27±11.42 |
| The second week | 305.26±10.77 | 289.54±17.25 | 291.04±10.48 |
| The third week | 368.12±8.95 | 312.83±15.62 | 325.91±15.20 |
| The fourth week | 379.47±21.54 | 315.95±16.51 | 336.49±20.54 |
| The fifth week | 398.43±23.72 | 324.59±21.82 | 355.19±24.51 |

### 3.2. Changes in the numbers of NK and NKT cells of the rats in three groups

After the increasing load exercise, numbers of NKT and NK cells of the rats in three groups are shown in Figure 1. It can be seen that the numbers of NKT and NK cells of the rats in *Tribulus terrestris* + exercise fatigue group were still lower than those in control group ($p < 0.05$); however, they were in the tendency to increase in comparison with the exercise fatigue group. In comparison with exercise fatigue group, numbers of NKT ($p < 0.05$) and NK ($p < 0.05$) cells, which were higher than those of the control group, increased significantly in *Tribulus terrestris* + exercise fatigue group.

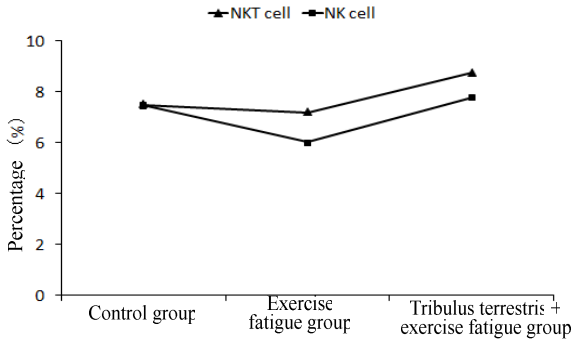


**Figure 1.** Numbers of NKT and NK cells of rats in three groups after increasing load training and supplement of *Tribulus terrestris* extract

### 3.3. Changes in hemoglobin and muscle glycogen of the rats in three groups

Table 3 shows the changes in hemoglobin and muscle glycogen of the rats in three groups. In comparison with control group, hemoglobin content of the rats in exercise fatigue group declined significantly ($p < 0.05$). Apparently, the tendency could be inhibited by taking in *Tribulus terrestris* extract, which was reflected in the fact that the hemoglobin content of the rats in *Tribulus terrestris* + exercise fatigue group increased ($p < 0.05$) in comparison with exercise fatigue group. In comparison with the control group, the muscle glycogen content of the rats in *Tribulus terrestris* + exercise fatigue group increased significantly ($p < 0.05$); due to exercise fatigue, muscle glycogen content of the rats decreased obviously ($p < 0.05$); nevertheless, *Tribulus terrestris* extract could obviously inhibit the decrease of muscle glycogen caused by exercise fatigue.

Health conditions can be reflected in weight changes. In motion experiments, body weights can be used to analyze the influence of training on the body and the adaptability of the body (Buijsse *et al.*, 2009). If there is a progressive decrease in body weight, it is assumed that the training might be arranged improperly or there are diseases in the body. Under normal conditions, body weights might fluctuate by 10%. If the fluctuating value is greater than 10%, the weight is abnormal (Schafer *et al.*, 2011). In this study, in the later period of increasing load training, the weights of the rats were in a declining trend in exercise fatigue group and *Tribulus terrestris* + exercise fatigue group. By the end of the training, in contrast with the control group, the weights of the rats in the two experiment groups decreased by 18.5% and 10.9% (both beyond the normal range of weight fluctuation). In this study, the weights of the rats in *Tribulus terrestris* + exercise fatigue group increased significantly compared with those in exercise fatigue group, which indicated that *Tribulus terrestris* extract could restrain the decreasing trend of weights. The data regarding the change of weight indirectly indicated that *Tribulus terrestris* extract might be able to benefit muscle growth and a well-built body.

As the lymphocytes with natural kill ability, NK cells are important for the immune defense responses of the body. NKT cells are T cell subgroup of special type; on the surface of NKT cells, there are both T cell receptors and NK cell receptors. Movement can lead to the change of the number and function of immune cells among which NK cells are the most sensitive to such changes; accordingly, NK cells can be used as a monitoring index for the early diagnosis of exercise fatigue (Paust et al., 2010; Vonarbourg et al., 2010).

**Table 3. Content of hemoglobin and muscle glycogen of the rats in three groups**

| Week | Control group (n=8) | | Exercise fatigue group (n=8) | | *Tribulus terrestris* + exercise fatigue group (n=8) | |
|---|---|---|---|---|---|---|
| | Hemoglobin (g/dl) | Muscle glycogen (mg/g tissue) | Hemoglobin (g/dl) | Muscle glycogen (mg/g tissue) | Hemoglobin (g/dl) | Muscle glycogen (mg/g tissue) |
| One week before exercise | 14.56±0.89 | 1.32±0.52 | 14.51±0.98 | 1.35±0.21 | 14.71±1.20 | 1.36±0.19 |
| 1st week | 14.62±1.23 | 1.35±0.65 | 14.22±1.21 | 1.30±0.09 | 14.65±1.65 | 1.30±0.29 |
| 2nd week | 13.92±2.56 | 1.34±0.54 | 13.85±2.55 | 1.21±0.15 | 14.03±2.06 | 1.26±0.13 |
| 3rd week | 14.78±1.75 | 1.36±0.71 | 13.24±3.01 | 1.11±0.29 | 13.86±1.87 | 1.21±0.22 |
| 4th week | 14.59±2.02 | 1.33±0.46 | 12.63±2.98 | 1.08±0.32 | 14.12±1.34 | 1.17±0.34 |
| 5th week | 13.66±2.71 | 1.35±0.39 | 12.51±3.54 | 1.05±0.34 | 14.02±1.46 | 1.14±0.51 |

NKT cell is a sensitive index that reflects the change of immune function after exercise, under the influence of intensity and amount of exercise. In the study, we found that with the supplement of *Tribulus terrestris* extract, there was a noticeable increase in the amounts of NK and NKT cells which were even greater than those of the control group, indicating that *Tribulus terrestris* extract could improve the immune function of over-exercise rats by increasing the numbers of NK and NKT cells. The increasing NKT cells not only benefits the differentiation of $CD_8^+$ killer T cells, but also greatly enhances the killing activity of NK cells (Witte et al., 2010) and improves the immune function of the body.

Hemoglobin, a kind of oligomeric protein (containing iron) in red blood cells, mainly plays its physiological function by transporting oxygen and carbon dioxide; it has a buffer effect on acidoid and can regulate the intracorporal potential of hydrogen (Parshina et al., 2013). Some researches (Garvican et al., 2010; Holden, 2013) found that hemoglobin content was influenced by the nutrition intake, exercise load and rest time of athletes during training and competition. Therefore, it is necessary to determine the hemoglobin content in order to grasp the nutritional status and physical function of athletes (Joseph et al., 2013). The data in Table 2 showed that after intense exercise, intracorporal hemoglobin content of the rats in exercise fatigue group decreased more obviously than that of *Tribulus terrestris* + exercise fatigue group, which indicated that *Tribulus terrestris* extract was beneficial to the synthesis of hemoglobin or was able to inhibit the injury of hemoglobin caused by free radicals, thus hemoglobin content was relatively high. Muscle glycogen reserves can directly influence the athletic ability of the body. Previous studies (Maga *et al.*, 2013) considered that the increase of muscle glycogen content might be one of the reasons why *Tribulus terrestris* extract prolonged the exercise duration of rats and improved their exercise capacity. In this study, muscle glycogen content was higher in *Tribulus terrestris* + exercise fatigue group than in exercise fatigue group ($p < 0.05$), which shares some similarity with the opinion of the previous studies.

## 4. Conclusions

Excessive exercise can result in exercise fatigue which reduces immunity of the body; consequently, there is exercise-induced immune-suppression which leads to the

decrease of athletes' resistance as well as infectious diseases, which is harmful to their training or competition. Therefore, for the sake of athletes' exercise capability, it is significant to find a solution to avoid the negative effect of exercise fatigue. Considering that *Tribulus terrestris* extract is a kind of sports food, we studied its effect of resisting exercise fatigue based on increasing load training on 24 SD rats. The results showed that *Tribulus terrestris* extract could enhance the inhibited immune function of the rats with exercise-induced fatigue by increasing the numbers of NK and NKT cells; it could effectively restrain the decrease of hemoglobin content caused by excessive exercise; in addition, it could increase muscle glycogen content, thus to improve energy reserves and aerobic exercise ability of the body, and eventually to improve the fatigue resistance of the body.

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# THE EFFECT AND PROTECTION MECHANISM OF LYCIUM BARBARUM POLYSACCHARIDE SUPPLEMENT ON IMMUNITY OF ATHLETES AFTER HIGHLY INTENSE TRAINING

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**ABSTRACT**

This study aims to discuss the effect and the protection mechanism of lycium barbarum polysaccharide (LBP) supplement after highly intense training on immunity of athletes. We selected 18 experiment athletes from colleges and divided them into the control group (A), pure training group (B) and LBP supplement group (C). Athletes in group B and C went through highly intense training everyday. The athletes in group C took LBP capsules (5 g each day) morning and night everyday, while the athletes in group B took placebos (5 g) morning and night everyday. Athletes in group A did not need any training but took placebos (5 g) morning and night everyday. T-lymphocyte subsets, immune globulin and interleukin-2 (IL-2) activities of athletes were detected before and after the experiment for further analysis.

In the meantime, in order to detect the effect of LBP on immune organs damage caused by highly intense training, we took mice as the research objects and divided them into three groups (group D, E and F) in the same way, thus to detect the spleen and thymus indexes of mice for results analysis. Results showed that, after highly intense training, compared with group A, the specific values of CD4+%, CD4+ and CD8+ of athletes in group B declined remarkably ($p < 0.01$); the specific value of CD8+% also decreased but only slightly ($p > 0.05$). The specific values of CD4+%, CD8+%, CD4+ and CD8+ of athletes in group C increased significantly compared with group A; moreover, compared with group A and B, detected data of immunoglobulin A (IgA), immunoglobulin M (IgM) and immunoglobulin G (IgG) of group C increased remarkably ($p < 0.05$). Compared with group A and B, activities of IL-2 of athletes in group C enhanced substantially. The thymus indexes of mice in group E were lower than that of the other two groups ($p < 0.05$); no significant difference was discovered in spleen and thymus indexes between group F and group D ($p > 0.05$). In conclusion, supplement of LBP can effectively prevent the decrease of immunity of athletes after highly intense training, as well as has significant inhibiting effect on the immune organs damage caused by highly intense training.

## 1. Introduction

Lycium barbarum is one of the traditional Chinese medicinal materials, and according to the Compendium of Materia Medica, it has various kinds of effects. Moreover, based on researches on lycium barbarum in recent years, we found that the lycium barbarum polysaccharide (LBP) extracted from the dried lycium barbarum after a series of process like

degreasing, etc., had significant effect on human immunity (Castiel and Gueniche, 2010). Some researchers in China, such as Yang Xinbo, et al. (Yang and Huang, 1998) believed that LBP had the hypoglycemic effect. Bian Lun, et al. (Sun et al., 2013) carried out relevant experiments and the results indicated that, the LBP had the recovery effect on acute liver injury induced by carbon tetrachloride as well as an inhibiting effect on growth and proliferation of human cervical cancer and human gastric adenocarcinoma cells; in addition, it could decrease the Methane Dicarboxylic Aldehyde (MDA) and lipofuscin content in the mice body, thus it also had anti-aging effect. Besides, the LBP also has immunomodulatory, anti-hyperlipidemia, hypotensive, anti-fatigue, anti-radiation and hemopoiesis accelerating effects (Edge and Xin, 1996), etc. Therefore, the LBP has become an important kind of additive in health food.

In order to solve the immune suppression and other problems which occur after highly intense training, physical culture workers and researches in China have carried out lots of studies. Some researches indicated that the best method to avoid low immune functions was to supplement nutritional agents after intensive training to avoid exercise-induced immune suppression (Gonçalves et al., 2012). This study was carried out to discuss the effect of LBP supplement on immunity of athletes as well as its protective effect on organ damage of athletes.

## 2. Experimental objects and methods

### 2.1. Research objects

We selected 18 athletes who majored in physical education and were (20±2) years old as the research objects. All research objects took part in the experiment voluntarily. After examination, no object had the disease history of metabolism and immune system. Besides, 24 8-week old healthy male mice which weighted (20±2) g were also selected.

### 2.2. Research methods

Eighteen athletes were divided into the control group (A), pure training group (B) and LBP supplement group (C) randomly, 6 athletes for each group. During training, athletes in group C took the LBP capsules (5 g each day) according to the specification morning and night. Athletes in group A and B took placebos (empty capsules which had the same shape and color as LBP capsules) morning and night. Except the normal diet, all athletes in three groups did not take any other drugs. This experiment lasted one month. After the experiment, 5 ml of venous blood samples were extracted from each athlete and added with heparin anticoagulant and sent to a hospital for detection of lymphocyte subsets, immune globulin concentrations and IL-2 activities. The 24 mice were also divided into three groups (group D, E and F). Mice in group D did not have any exercise and were fed with normal saline (0.9%). Mice in group E were fed with normal saline (0.9%) after exercise. Mice in group F were fed with LBP after exercise, thus to study the effect of LBP on spleen and thymus damage caused by highly intense exercise.

#### *2.2.1. Detection of lymphocyte subsets*

According to tagged molecules on the cell surface, a flow Cytometer (produced by American Beckman Coulter, Inc.) as well as the monoclonal antibody (McAb) indirect immunofluorescence were adopted for the detection of specific values of CD4+%/CD8+% and CD4+/CD8+ (Benetatos *et al.*, 2009).

#### *2.2.2. Detection of serum immune globulin*

The concentrations of immunoglobulin A (IgA), immunoglobulin M (IgM) and immunoglobulin G (IgG) in serum protein were detected by immunoturbidimetry. A semiautomatic biochemistry analyzer produced by American Beckman Coulter, Inc. was used.

#### *2.2.3. Detection of IL-2 activities*

The heparin anticoagulant blood was mixed with an equal amount of phosphate buffer

saline (PBS) and then slowly added with an equal amount of lymphocyte separation medium. After that, the mixed solution was centrifuged in 2000 rpm for 20 min. Then cells in the lymphocyte layer were drawn and washed by PBS twice and added with a moderate amount of Roswell Park Memorial Institute-1640 culture medium (RPMI-1640) (10% fetal calf serum). The number of lymphocytes was counted by microscopic examination and lymphocytes were diluted to $2\times10^6/mL^{-1}$, i.e., lymphocytes suspension (Suo *et al.,* 2015). Then the lymphocytes suspension was added to a 96-well plate, 100 μL for each well. Phytohaemagglutinin (PHA) was then added and the final concentration reached $100\mu g/mL^{-1}$; each group had duplicate wells (Walsh *et al.,* 2003). After three days of culture, the cultural supernatant was collected for detection of IL-2 level using the enzyme-linked immunosorbent assay. The optical density at 570 nm wavelength was measured by the microplate reader and the number of cytokines in the cultural supernatant was calculated.

#### *2.2.4. Detection of spleen and thymus indexes*

After the training, the spleen and thymus were separated from the mice and the indexes of spleen and thymus were calculated. Thymus index=thymus weight/mice weight; spleen index=spleen weight/mice weight (unit was mg/g) (Jia *et al.,* 2014).

### 2.3 Statistical analysis

Software SPSS 15.0 was used to statistically analyze the obtained experimental data. The significant difference level p was 0.05. $p < 0.05$ indicated that the difference had statistical significance.

## 3. Results and discussions

### 3.1. The effect of LBP on T- lymphocyte subsets

After the highly intense training, the effect of LBP supplement on T-lymphocyte subsets was shown in Table 1.

**Table 1.** Effect of LBP supplement on T-lymphocyte subsets

| Group | CD4+% | CD8+% | CD4+/CD8+ |
|---|---|---|---|
| Control group (A) | 50.68±2.25 | 28.08±2.65 | 1.80±0.85 |
| Pure training group (B) | 46.09±2.04 | 27.42±2.28 | 1.68±0.89 |
| LBP supplement group (C) | 56.60±3.14 | 28.54±1.84 | 1.98±1.71 |

Note: the CD4+%, CD4+/CD8+ of group C were compared with that of group B, $p < 0.05$

As shown in Table 1, compared with group A, the specific values of CD4+% and CD4+/CD8+ of group B decreased remarkably ($p < 0.01$), indicating that highly intense training could lead to lower immune functions. However, compared with group A and B, specific values of CD4+%, CD8+% and CD4+/CD8+ of group C increased, which increased significantly compared with group B; the difference had no statistical significance ($p > 0.05$). Therefore, LBP had inhibiting effect on the decrease of T-lymphocyte immune functions caused by highly intense training (Dingjuan et al., 2016).

**Table 2** Effect of LBP supplement on immune globulin

| Group | $IgA/g\cdot L^{-1}$ | $IgM/g\cdot L^{-1}$ | $IgG/g\cdot L^{-1}$ |
|---|---|---|---|
| Control group (A) | 1.61±0.43 | 1.20±0.13 | 10.93±2.58 |
| Pure training group (B) | 1.65±0.34 | 1.16±0.15 | 10.72±2.04 |
| LBP supplement group (C) | 1.89±0.64 | 1.24±0.13 | 11.21±2.42 |

Note: the $IgA/g\cdot L^{-1}$ and $IgG/g\cdot L^{-1}$ of group C were compared with that of group B, $p < 0.05$.

### 3.2. The effect of LBP on immune globulin

In Table 2, compared with group A, the IgA concentration of group B increased while the concentrations of IgM and IgG decreased; however, no significant differences were found ($p > 0.05$). Therefore, highly intense training could result in the increase of IgA concentration and the decrease of IgM and IgG concentrations. The IgA concentration of group C was significantly higher than that of group A and B; there was a significant difference ($p < 0.05$). The concentration of IgG of group C was higher than that of the other two groups ($p < 0.05$) and the difference had statistical significance. There was no significant difference between the concentration of IgM of group C and group A and B ($p > 0.05$). Therefore, the results indicated that the LBP could accelerate the synthesis of immune globulin (Luo et al., 2014).

**Table 3.** Effect of LBP on IL-2 activities

| Group | Right after training | 2 hours after training | 4 hours after training |
|---|---|---|---|
| Pure training group (B) | 4.25±0.18 | 4.19±0.83 | 3.85±0.14 |
| LBP supplement group (C) | 9.10±0.34 | 14.74±0.67 | 18.12±0.43 |

Note: IL-2 activities of the control group A at the same time points were 4.73±0.85, 4.70±0.86 and 4.71±0.83.

### 3.3 The effect of LBP on IL-2 activities

As shown in Table 3, right after training, 2 hours after training and 4 hours after training, the IL-2 activities of group C increased significantly compared with that of group A, which had statistical significance ($p < 0.01$). The IL-2 activities of group B after training showed a decreasing tendency, but the difference had no statistical significance ($p > 0.05$).

**Table 4.** Effect of LBP on spleen and thymus of mice

| Group | Number | Weight of thymus | Weight of spleen |
|---|---|---|---|
| Mice control group (D) | 6 | 5.122 | 12.973 |
| Mice pure training group (E) | 6 | 3.846 | 12.314 |
| Mice LBP supplement group (F) | 6 | 4.756 | 12.814 |

**Table 5.** Effect of LBP on spleen and thymus of mice after highly intense training

| Group | Thymus index (average value of thymus/weight ) | Spleen index (average value of spleen/weight) |
|---|---|---|
| Mice control group (D) | 0.259 | 0.655 |
| Mice pure training group (E) | 0.197 | 0.631 |
| Mice LBP supplement group (F) | 0.237 | 0.638 |

Note: the weight of mice in each group was the average value: D group was 19.8 g, E group was 19.5 g and F group was 20.1 g.

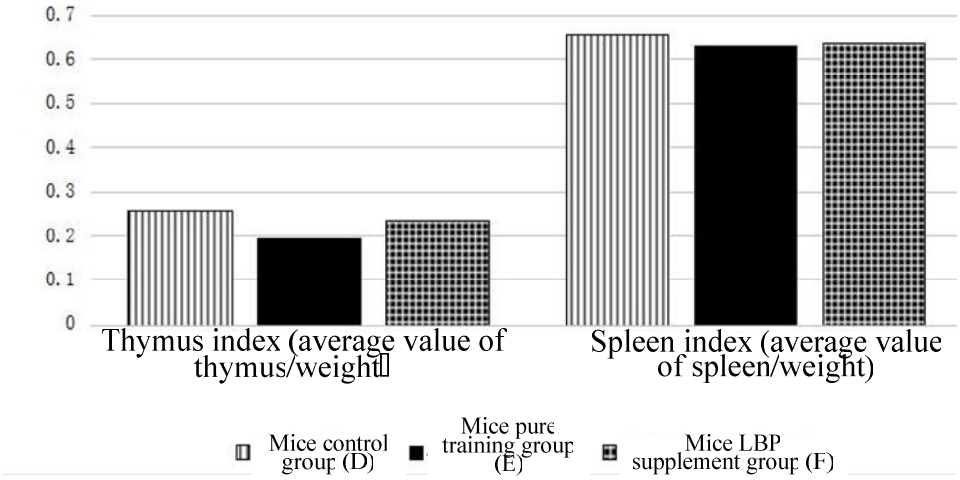


**Figure 1**. Spleen and thymus indexes of mice after the supplement of LBP

### 3.4. The effect of LBP on spleen and thymus of mice

The thymus indexes of group E decreased significantly compared with group D ($p < 0.05$), indicating that highly intense training could result in the decrease of thymus weight and thus affect the immune functions. No significant differences were found in spleen indexes and there was no statistical significance. Compared with group D, the indexes of thymus and spleen of group F had no statistical significance ($p > 0.05$). Therefore, the supplement of LBP could recover the immune organ damage caused by highly intense training (So et al., 2011).

As people are paying more and more attentions to the relationship between exercise and immunity, how to enhance the immunity has become an important subject. Highly intense training or competition can result in inhibition of cellular immunity tissues and humoral immunity functions, thus the immunity to pathogenic microorganism can be weakened (Peters, 2004). As an important component of lycium barbarum, LBP contains various kinds of microelements and amino acid (Li et al., 2007), which has significant effect on human immunity.

Results in this study show that, the functions of T-lymphocyte and its subsets are depressed with the increase of training intensity and time. In the experiment, after the supplement of LBP, specific values of CD4+%, CD8+% and CD4+/CD8+ of athletes show increasing tendency, indicating that LBP can improve the proliferation ability of T-lymphocytes. In the experiment of detecting the effect of LBP on immune globulin, the immune globulin is taken as the glycoprotein molecules secreted by activated and proliferated plasmocyte (Jackson and Elsawa, 2015), and its indexes (IgA, IgM, IgG) are important evidences of the effect of LBP on tissues and organs and humoral immunity (Wallukat et al., 2012). Detection of IgA, IgM and IgG in serum shows that, after the supplement of LBP, the values of IgA and IgG increase significantly, indicating that the supplement of LBP can accelerate the synthesis of IgA and IgG and thus improve immunity. Human peripheral blood mononuclear cell IL-2 is a kind of glycoprotein released by helper lymphocytes (Anta et al., 2012), which can accelerate the proliferation of T and B lymphocytes as well as the generation of antibodies. Thus it is an important kind of lymphokine that can improve immunity (Bonini and Bondanza, 2011). LBP can accelerate the synthesis of antibodies through enhancing activities of IL-2 (Corrigall et al., 2004). Moreover, LBP also has recovery and protective effect on damaged thymus caused by intensive training. In experiment of mice, although the

thymus indexes of mice that receive LBP supplement are lower than mice that do not have training, the difference is not significant. Therefore, the supplement of LBP can reduce the damage of thymus caused by highly intense training (Yan et al., 2002).

In conclusion, the supplement of LBP can inhibit the decrease of immune functions caused by highly intense training, as well as effectively relieve and control the body infection caused by massive exercise. In addition, it has a powerful accelerating effect on immunity of athletes and an inhibiting and protective effect on damaged organs caused by highly intense training.

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# ANTI-EXERCISE FATIGUE EFFECT OF BAZHEN DECOCTION ADDED WITH CINNAMON AND FRUCTUS PSORALEAE

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**ABSTRACT**

The relief effect of Bazhen decoction added with cinnamon and fructus psoraleae on exercise induced fatigue was analyzed by establishing rat exercise-induced fatigue models. Ninety Sprague dawley (SD) male rats were selected and randomly divided into three groups, i.e., control group, model group and test group, according to experimental requirements. Rats in the control group were not trained, but chronic exercise induced fatigue models were set up in the other two groups. Rats in the control group were fed normally, under the condition of no exercise training; rats in the model group were gavaged with normal saline as placebo once within half an hour after daily training; rats in the test group were gavaged with a compound traditional Chinese medicine, i.e., Bazhen decoction added with cinnamon and fructus psoraleae once within half an hour after daily training. Results suggested that, Bazhen decoction added with cinnamon and fructus psoraleae could increase the content of hemoglobin in blood, inhibit the catabolism of heme, keep the metabolism of hemoglobin at a relatively low level, increase the synthesis of hemoglobin, enhance the immune function of red blood cells, improve hematopoietic function, and effectively relieve exercise-induced anemia. Besides, Bazhen decoction added with cinnamon and fructus psoraleae could relieve or eliminate exercise-induced fatigue by promoting the reduction of creatine kinase (CK) content, lowering the permeability of cytomembrane, and inhibiting the release of CK in cells.

## 1. Introduction

With the constant improvement of modern competitive level, competition has become increasingly intense and the intensity of training of sportsmen is increasing; under such an environment, such kind of overload training and competition will result in higher risks of exercise-induced fatigue (Sargent et al., 2014; Halson, 2014; Zhai, 2013). Fatigue will accumulate if body cannot be effectively relieved. If exercise-induced fatigue cannot recover timely and the functions and immunity of body cannot be improved timely, the risks of diseases will be higher, which can directly affect the fulfillment of skilled movement and competition performance of sportsmen and even increase the risks of exercise injuries (Joshi et al., 2013). Thus searching for effective methods that can delay or eliminate exercise induced central fatigue is the key point that is urgent for study. Bazhen decoction added with cinnamon and fructus psoraleae is an improvement on Bazhen decoction, which is a convenient and simple way to resist exercise-induced fatigue.

The mechanism for the generation of exercise-induced fatigue is complex. When exercise induced fatigue generates, the

functional levels of tissues and organs in human body will decline; therefore, fatigue can be evaluated based on the changes of functional level of organs and tissues in human body (Radiloff et al., 2014; Benson and Mushtaq, 2015; Tomlinson et al., 2014). In the field of competitive sports, how to strengthen and enhance the immunity of sportsmen so that they can keep a healthy constitution and a good competitive state is a key problem that needs to be solved (Goodall et al., 2014; Meneses-Echávez et al., 2014; Durcan et al., 2014). Traditional Chinese medicine as the traditional medicine of Chinese nation has its special advantages in promoting the recovery of exercise-induced fatigue and improving exercise capacity for its high safety without any side effects (Wang et al., 2014). Erqun Song et al. (Erqun et al., 2014) found that, Bazhen decoction had functions of notifying qi, nourishing blood, and supplementing kidney; cinnamon and fructus psoraleae were highly effective in resisting oxidation and regulating immune; the addition of cinnamon and fructus psoraleae in Bazhen decoction could improve the efficacy of Bazhen decoction.

Bazhen decoction added with cinnamon and fructus psoraleae is made from ginseng, white paeony root, Angelica sinensis, Rehmannia Glutinosa, Ligusticum wallichii, white atractylodes rhizome, Poria cocos, liquorice, Eucommia ulmoides, cinnamon, radix achyranthis bidentatae, Radix Dipsaci Asperoidis, Chinese yam and fructus psoraleae, which has functions of nourishing kidney, enriching qi, strengthening muscles and bones, tonifying liver, replenishing yin and tonifying yang. Through an experiment on rats with exercise-induced fatigue, this study discussed the relief effect of Bazhen decoction added with cinnamon and fructus psoraleae on exercise-induced fatigue.

## 2. Experimental subjects and methods

### 2.1. Experimental subjects

Ninety healthy male Sprague-Dawley (SD) rats, weighed 180 ~ 220 g, were selected. They were fed by national standard dried feed for rodent and trained indoor. They were raised in different cages, in a ventilated room and under natural lighting. The test lasted for 6 weeks. During experiment, the exercise of the rats was observed timely; besides, the rats were motivated to run continuously by stimulation using electricity or instruments. The temperature and humidity of the laboratory, ingestion, water drinking, defecation, fur and exercise capacity were checked every day. Besides, weights of the rats were weighed and recorded before exercise at the first day of every week.

### 2.2. Experimental method

#### *2.2.1. The preparation of Bazhen decoction added with cinnamon and fructus psoraleae*

Ginseng, white paeony root, Angelica sinensis, Rehmannia Glutinosa, Ligusticum wallichii, white atractylodes rhizome, Poria cocos, liquorice, Eucommia ulmoides (9 g each), cinnamon (3 g), radix achyranthis bidentatae, Radix Dipsaci Asperoidis, Chinese yam (6 g each) and fructus psoraleae (9 g) were taken and prepared by means of immersion and decoction to control and reduce the toxic and side effect of drugs. Finally it was condensed into a solution with a concentration of 100%. Then the solution was stored at 4 °C.

#### *2.2.2 Experimental grouping*

The room temperature was controlled at around 23 °C and the indoor humidity was kept at about 55%. The lighting was free. The laboratory was cleaned and disinfected every two days. The rats were weighed once every week. After one week of adaptive raising, rats in the control group were fed in natural way every day, rats in the model group were gavaged with 0.1 ml/10 g of normal saline every day, and rats in the test group were gavaged with 800 mg/kg Bazhen decoction added with cinnamon and fructus psoraleae two hours before exercise every day.

Except the control group, rats in the other two groups were required for swimming training once each day. The swimming box

used in the experiment had a size of 80 cm × 46 cm × 40 cm. The water depth was kept equal to or higher than 30 cm, and the water temperature was kept at 28 °C. Rats did exercise from am 7:30 to am 11:30 at the first six days of every week and rested on Sunday. The physical condition before and after exercise was recorded. The rats swam for 30 min at the first week, 10 min more afterwards till the fourth week. At the fourth week, the rats swam for 60 min every day; after the fourth week, the duration of swimming was kept at 60 min, for one week.

At the last day of experiment, one hour after the administration of Bazhen decoction added with cinnamon and fructus psoraleae of the test group, all rats did exhaustive swimming group by group. Rats were determined as exhaustive if they were immersed into water for more than 8 s and could not do righting reflex on plane. The duration of exhaustive swimming was recorded.

### 2.3. Material analysis

#### *2.3.1 Index testing method*

Chloral hydrate solution with a concentration of 10% was used to narcotize rats in group through abdominal cavity, in a dose of 0.3 ml/10 g. Then the abdominal skin and muscles were cut apart to expose abdominal aorta; 5 ml of artery blood was collected through abdominal aorta puncture. After one hour of standing at room temperature, it was centrifuged at 3000 r/min for 10 min. The supernate was isolated, transferred to a 2 ml freezing tube, and stored at - 80 °C. After the collection of blood, liver and muscle tissues were resected immediately, washed by normal saline, dried using filter paper, and stored at - 80 °C. Hemoglobin (Hb) and the content of creatine kinase (CK) in plasma and relevant liquid samples were detected using enzyme-linked immunosorbent assay (ELISA) kit and biotin double antibody sandwich technology.

### 2.4. Statistical processing

Data were expressed as mean ± standard error (SE). SPSS for Windows 16.0 was used for statistical analysis.

## 3. Results and discussions

### 3.1. The observation of general condition and weight change of rats in groups

Rats in the control group demonstrated normal food taking and water drinking states, weight increase and activity as well as smooth fur; rats in the model group and the test group were observed with good mental state and lively and active action, and symptoms such as decreased amount of food taking or water drinking, weight loss, squintm, fatigue and withered and dried fur were not observed. Compared to the control group, the model group and the test group had no abnormal situation. It could be seen from Table 1 that, all groups showed an increasing tendency of weight, suggesting progressive increasing load training did not induce excessive fatigue and rats showed no adverse reactions to the medicine.

**Table 1.** The weight change of rats in groups

| | 1st week | 2nd week | 3rd week | 4th week | 5th week |
|---|---|---|---|---|---|
| Control group | 216.33±11.32 | 299.47±21.98 | 321.87±36.11 | 345.14±26.32 | 369.21±18.49 |
| Model group | 214.14±16.11 | 271.56±28.54 | 301.45±30.74 | 314.95±33.32 | 331.78±30.32 |
| Test group | 219.45±14.75 | 267.14±13.63 | 294.87±18.36 | 313.54±20.96 | 338.23±15.23 |

### 3.2. Effects of Bazhen decoction added with cinnamon and fructus psoraleae on exhaustive swimming time of rats

Table 2 demonstrates that, the duration of exhaustive swimming of rats in the test group was longer than that of the model group, but there was no statistically significant difference ($p > 0.05$), suggesting Bazhen decoction added with cinnamon and fructus psoraleae could extend exercise time and improve endurance.

**Table 2.** The duration of exhaustive swimming of rats in groups

| | Duration of exhaustive swimming (min) |
|---|---|
| Model group | 168.23±28.65 |
| Test group | 211.45±30.21 |

### 3.3. Test results of Hb

Hb can reflect the oxygen carrying capacity of blood during exercise. Excessive training can damage red blood cells, reduce the content of Hb and even induce exercise-induced anemia in severe cases. For middle and long-distance runners, the content of Hb is required to be at a normal level or higher than middle level. The enhancement of oxygen transport and carrying capacity of blood is helpful to the improvement of sports performance and excise capacity of sportsmen engaging in middle and long-distance running events (Ronghui, 2015; Matos et al., 2014). Table 3 shows that, Hb of the model group at the 5th and 6th week was lower than that at the 1st week and 3rd week, and the difference was highly statistically significant ($p < 0.01$); at the 5th week, Hb of the test group showed a decreasing tendency, and the difference with the control group was highly statistically significant ($p < 0.01$); at the 6th week, Hb of the test group was lower than that of the control group ($p < 0.05$), but higher than that of the model group, and the difference was highly statistically significant ($p < 0.01$). It suggested that, Bazhen decoction added with cinnamon and fructus psoraleae promoted the content of Hb in blood of rats, inhibited the catabolism of heme, enhanced the immune function of red blood cells, improved hematopoietic function, and effectively relieved exercise-induced anemia.

**Table 3.** The change of Hb of rats in groups (g/L)

| | 1st week | 3rd week | 5th week | 6th week |
|---|---|---|---|---|
| Control group | 145.45±6.39 | 146.14±10.36 | 147.25±6.47 | 140.21±9.14 |
| Model group | 148.14±6.21 | 142.58±14.14 | 115.98±14.74 | 118.21±3.56 |
| Test group | 150.21±4.47 | 142.45±16.39 | 131.47±12.41 | 133.87±10.96 |

### 3.4. Test results of CK

CK involving in the control of glycolysis as well as the contraction and energy supply of muscle is one of the key enzymes in ATP-CP energy supply system metabolism. Distributed in skeletal muscle, brain tissue and heart of animals, CK can satisfy the physiological need of organs and tissues. Stimulation, no matter high strength or low strength, can result in the increase of serum CK; hence serum CK is considered as one of the most sensitive indexes for training load (Hody et al., 2013; Leite et al., 2013). Table 4 demonstrates that, the value of CK of the model group at the 5th week was higher than that at the 3rd week ($p < 0.05$) and extremely higher than the control group ($p < 0.01$), suggesting body had been fatigued; at the 5th week, the value of CK in the test group was no remarkably different with that in the control group, but much lower than that in the model group ($p < 0.01$); the CK values of the control group, model group and test group at the 6th week were much lower than those at the 1st, 3rd and 5th week, and the differences were statistically significant ($p < 0.05$); at the 6th week, the CK value of the test group was extremely lower than that of the control group ($p < 0.01$) and much lower than that of the model group ($p < 0.05$). It indicated that, Bazhen decoction added with cinnamon and fructus psoraleae could reduce the content of CK and the permeability of cytomembrane, and restrain the release of CK in cells, thereby relieving exercise-induced fatigue.

**Table 4.** The change of CK in groups (μ/l)

| | 1st week | 3rd week | 5th week | 6th week |
|---|---|---|---|---|
| Control group | 557.23±125.14 | 506.32±115.47 | 462.35±50.68 | 256.47±23.84 |
| Model group | 658.89±167.46 | 584.22±122.63 | 699.98±59.96 | 260.74±9.65 |
| Test group | 705.32±142.51 | 631.89±169.58 | 480.63±66.41 | 233.49±18.63 |

## 4. Conclusions

Research results demonstrated that, Bazhen decoction added with cinnamon and fructus psoraleae could increase the content of Hb in blood, restrain the catabolism of heme, enhance immune functions of red blood cells, improve hematopoietic function, and effectively relieve exercise-induced anemia. Besides, the decoction could reduce the content of CK of rats with exercise-induced fatigue, reduce the permeability of cytomembrane, and inhibit the release of CK in cells. To sum up, the supplement of Bazhen decoction added with cinnamon and fructus psoraleae can improve the fatigue resistance of the body.

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# EFFECT OF TRADITIONAL CHINESE MEDICINE ON THE ENERGY METABOLISM OF ATHLETES AFTER RUNNING

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**ABSTRACT**

Over a long history, some traditional Chinese medicine (TCM) has evolved into health food for humans. Dating back to the Tang Dynasty, in his medical book Dietetic Materia Medica, Meng Xian introduced a lot of TCM diet therapies which were effective in tonifying qi and yin, nourishing spleen and stomach, reinforcing the kidney and lung. Moreover, TCM diet therapies can enhance the capability of athletes. In this study, we observed the effects of TCM intervention on the body function level and recovery ability of exercise rats. We prepared two kinds of TCM decoction (A and B). The experimental rats were divided into quiet group A (QA), movement group A (MA), quiet group B (QB), movement group B (MB), quiet control group (QC) and movement control group (MC). The rats in groups MA and QA took TCM decoction A; the rats in groups MB and QB took TCM decoction B; the rats in groups MC and QC took the same amount of clean water. By observing the influence of TCM intervention on the energy metabolism of the exercise rats, we deduced the action mechanism of TCM intervention in improving the exercise capacity of rats.

## 1. Introduction

While competitive sports has developed constantly, scientific researchers have attempted to explore the mechanism of over-training from different aspects and the approaches to enhance athletes' endurance to high-intensity training (Scharhag et al., 2013). The ability to carry and use oxygen of an athlete's body is an important factor influencing the athletic ability of the body (Wang, 2012). Therefore, the mechanism of over-training can be explored by studying the changes in the form and functions of red blood cells (Dass et al., 2012; Lombardi et al., 2012). In addition, skeletal muscle is the main tissue that completes competitive activities in sports training; consequently, skeletal muscle is the victim of over-training (Loucks et al., 2011). When an athlete is under the influence of over-training, his/her body might suffer physiological or pathological changes due to a series of overloading training (Artioli et al., 2011). Accordingly, the relevant studies can be performed from the aspect of energy metabolism (Sabatini et al., 2011).

In recent years, more and more researchers have devoted themselves to the studies on the effect of traditional Chinese medicine in enhancing athletic ability. The concept of traditional Chinese medicine is to regulate the balance of Yin and Yang, qi and blood on the whole (Carlsohn et al., 2010), which differs from the concept of modern medicine. In competitive sports, the body function of athletes may be in a declining trend with the increase of exercise load (Zadik et al., 2009). How to improve the recovery ability of body function under overload training is one of the

major concerns in the field of competitive sports. Therefore, in this study we confected a TCM decoction of medicinal herbs according to the principle of traditional Chinese medicine (TCM). Through intragastric administration of the TCM decoction, we fed the rats which took long-term exercise training. By observing their red blood cell metabolism, gene expression of metabolic enzyme in skeletal muscle and the change of issue form, we aimed to study the effect of Chinese medicine intervention on the body function level and recovery ability of rats.

## 2. Materials and methods

### 2.1. General materials

This study included 66 male rats which were purchased from Nanjing Better Biotechnology Co., Ltd. The rats were divided into two groups (movement group and quiet group), including six subgroups: quiet group A (QA), movement group A (MA), quiet group B (QB), movement group B (MB), quiet control group (QC) and movement control group (MC). Two kinds of TCM decoction (A and B) were prepared according to a certain proportion of ingredients. TCM decoction A consisted of American ginseng, acanthopanax, dodder, fructus schisandrae and etc. TCM decoction B consisted of American ginseng, wolfberry fruit, fructus schisandrae, epimedium, Radix Ophiopogonis, Polygonum multiflorum and etc.

### 2.2. Experiment methods

#### *2.2.1. Movement mode*

The rats took load exercise on a treadmill for seven weeks; the running speed was 28 m/min. In the first week, the initial exercise time was 20 minutes; subsequently, it increased by 5 minutes every day. In the second week, the running speed increased to 32 m/min (increased by 50%) and it increased at this rate until the fifth week when the speed remained unchanged (till the end of the training).

During the first two weeks, the rats took training once a day; in the other five weeks, twice a day (respectively in the morning and evening). If a rat showed any symptoms of severe exhaustion, it should be given mechanical stimulation; if it still could not continue to run or there was an obvious mark in the shape of a soft-shelled turtle in its abdomen after it touched the ground, it was allowed to take a rest for 5 minutes before it continued training.

#### *2.2.2. Blood sampling and test method*

Every time after training, blood was collected from each rat by cutting off its tail. A hematology analyzer was used to determine the blood indexes, including hemoglobin (Hb), red blood cell (RBC) count, hematocrit (Hct) and etc. Testosterone was determined using enzyme-linked immunosorbent assay.

#### *2.2.3. Statistical methods*

Software SPSS19.0 was used for the statistical analysis on the experimental data which were all expressed in the form of mean±standard deviation. For the T test on the data, $p < 0.05$ means significant difference, while $p < 0.01$ means highly significant difference.

## 3. Results and discussions

### 3.1. Effect of TCM intervention on the basic indexes of the exercise rats

As can be seen from Figure 1, after seven-week overload training, in the movement groups, the increase in the weights of the rats was not obvious; however, in the quiet groups, there was a much more evident increase in the weights of the rats. Moreover, the rats in the quiet groups had glossy fur while the rats in the movement groups had matted hair and hair slip. It was observed that the rats in movement groups showed mild fatigue after three-week exercise; in the sixth and seventh weeks, they even showed symptoms of severe fatigue, accompanied by shortness of breath and decrease of movement coordination.

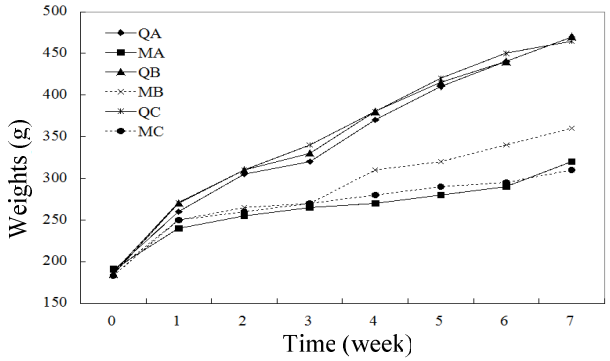


**Figure 1** Influence of overload training on the weights of rats

### 3.2. Effect of TCM intervention on the red blood cell parameters of exercise rats

As shown in Table 1, there was a decrease in Hb concentration, RBC count and Hct of the rats in movement groups, and the level of these indexes was significantly lower than that of quiet groups ($p < 0.05$). Accordingly, it was considered that the overload training method used in this study had an evident effect on the body functions of exercise rats. Hb, RBC and Hct of the exercise rats in MA and MB groups were remarkably higher than those of MC group, suggesting that Chinese medicine intervention could significantly improve the body function of rats. As can be seen in table 1, there was a difference in the serum testosterone of the rats in movement and quiet groups: it was significantly higher in groups A and B than in group C. Based on the comparison between group A and group B, it was found that TCM decoction B was more effective in increasing serum testosterone than TCM decoction A. Both of them could improve the serum testosterone level of exercise rats and give rise to adverse effect on the secretion of testosterone in the rats.

**Table 1.** Blood cell parameters of different groups

| Groups | Concentration of serum testosterone (ng/mL) | Hb(g/L) | RBC($\times 10^{12}$/L) | Hct($\times$10L/L) |
|---|---|---|---|---|
| QA | 1.86±1.07* | 143.84±7.06 | 7.69±0.50 | 0.45±0.03 |
| MA | 3.23±1.86*# | 145.79±14.74* | 7.68±0.66* | 0.44±0.04* |
| QB | 1.74±1.42* | 150.01±7.59 | 7.96±0.63 | 0.45±0.05 |
| MB | 3.78±2.93*# | 148.13±7.34* | 7.38±1.08* | 0.44±0.04* |
| QC | 1.49±0.48 | 145.62±6.68 | 7.76±0.62 | 0.46±0.05 |
| MC | 1.01±0.73 | 125.15±20.95# | 6.38±1.13# | 0.40±0.06# |

Note: For the comparison between exercise groups and quiet groups, # means $p < 0.05$, ## means $p < 0.01$; for the comparison of groups A, B and C, * means $p < 0.05$, ** means $p < 0.01$.

### 3.3. Effect of load exercise on the metabolism of free radicals in red blood cells of rats

According to Table 2, after taking load exercise, the malondialdehyde (MDA) level in the red blood cells of the rats in MA and MB groups was significantly higher than that of MC group; MDA level was evidently higher in the movement groups than in the quiet groups; there was no significant difference in either of the comparisons ($p > 0.05$). The activity of catalase (CAT) in movements was significantly lower than that of quiet groups, which suggested that large load exercise led to the decrease of CAT activity in mice. The fact that the CAT activity of MA and MB groups was higher than that of MC group suggested that Chinese medicine could enhance the antioxidant capacity of the body; as for the activity of superoxide dismutase (SOD), there was no significant difference between groups.

**Table 2.** CAT, MDA and SOD level in the RBC of exercise rats in different groups

| Group | CAT(U/mgHb) | MDA(mmol/mL) | SOD(U/mgHb) |
|---|---|---|---|
| QA | 3.90±1.92* | 1.14±0.46 | 8891.09±2481.51 |
| MA | 3.91±2.31* | 2.10±1.32 | 10556.84±3965.65 |
| QB | 2.45±1.37** | 2.12±0.83 | 8293.77±4400.05 |
| MB | 4.12±0.42* | 2.24±0.87 | 11084.58±2973.64 |
| QC | 5.61±1.95 | 0.98±0.63 | 7945.21±2341.24 |
| MC | 2.38±0.98# | 2.87±0.51## | 9820.34±4623.64 |

Note: For the comparison between exercise groups and quiet groups, # means $p < 0.05$, ## means $p < 0.01$; for the comparison of groups A, B and C, * means $p < 0.05$, ** means $p < 0.01$.

### 3.4. Effect of Chinese medicine intervention on the gene expression of energy metabolism enzyme in skeletal muscle of exercise rats

Figure 2 shows that the messenger RNA (mRNA) expression quantity of adenosine triphosphate (ATP) inhibitor peptide (IF1) was higher in movement groups than in quiet groups, indicating that exercise training could increase the expression of IF1. In addition, with the application of TCM decoction A and TCM decoction B, there was no such phenomenon as the increase of IF1 gene expression caused by the decrease of load exercise. Chinese medicine intervention resulted in an increasing trend of F0 protein gene expression in the rats of MA group; accordingly, it was speculated that Chinese medicine intervention might promote the increase of F0 protein. The increase of GLUT-4 gene expression caused by long-term load exercise was an adaptative change of exercise, which could improve the energy metabolism ability of the body.

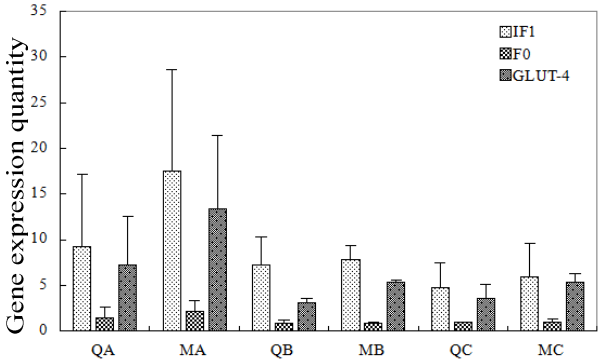


**Figure 2** .Effect of Chinese medicine intervention on gene expression of metabolic enzymes in skeletal muscle of exercise rats

### 3.5. Effect of Chinese medicine intervention on LD content, LDH activity and MDH activity in serum of exercise rats

According to the data in Table 3, there was no significant difference in serum lactic dehydrogenase (LD) content, serum lactate dehydrogenase (LDH) activity and serum malate dehydrogenase (MDH) activity between quiet and movement groups. Under the same condition, serum LD content in group A and group B was lower than in group C ($p < 0.05$), while serum LDH and MDH activity in group A and group B increased evidently in comparison with group C ($p < 0.05$).

### 3.6. Effect of TCM intervention on the phosphorylation of RBC membrane band 3 protein in exercise rats

Figure 3 shows that due to the long-term load exercise, the phosphorylation of erythrocyte membrane band 3 protein of the rats in MC group was significantly higher than that of QC group. It was pointed out that long-term load exercise led to evident changes in the structure of erythrocyte membrane and seriously affected the normal frame structure of RBC membrane band 3 protein. Furthermore, it had an impact on the structure, function and metabolism of RBC and RBC membrane. Based on the experiment, it proved that TCM intervention was beneficial to improving the structure and function of RBC membrane of rats.

**Table 3** Effect of TCM intervention on LD content, LDH activity and MDH activity in the serum of rats

| Group | LD(mmoL/L) | LDH(U/mL) | MDH activity (U/mL) |
|---|---|---|---|
| QA | 3.01±0.43 | 2321.05±462.25 | 0.14±0.03 |
| MA | 2.48±0.54* | 3434.64±373.56** | 0.22±0.04* |
| QB | 3.14±0.51 | 2325.64±458.31 | 0.15±0.04 |
| MB | 2.48±0.54** | 3452.34±375.32* | 0.21±0.04* |
| QC | 3.12±1.16 | 2353.68±275.64 | 0.14±0.03 |
| MC | 3.15±0.61 | 2551.54±317.84 | 0.18±0.04 |

Note: For the comparison of groups A, B and C, * means $p < 0.05$, ** means $p < 0.01$.

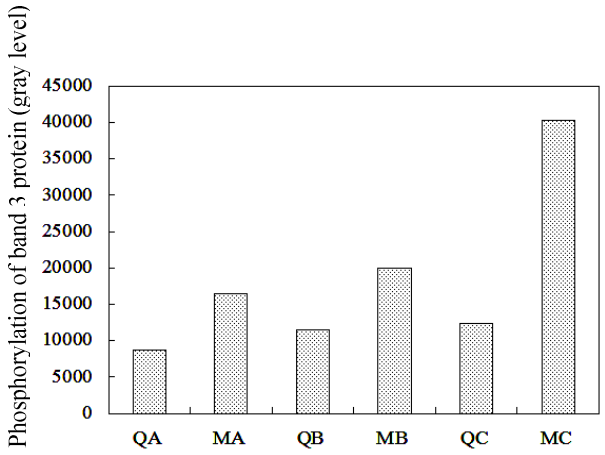


**Figure 3** Effect of TCM intervention on the phosphorylation of RBC membrane band 3 protein in exercise rats

As an important approach to examine the effect of training on body functions, treadmill exercise is easy to control, has a similar mode as competitive sports and the mortality of experiment animals is low. A lot of studies (Ke-Tien, 2012; Maughan, 2010; Miccheli et al., 2009) have shown that treadmill exercise could result in a significant decrease in hemoglobin concentration. However, there was a difference in the exercise intensity, exercise frequency and duration of those studies. As the treadmill exercise load was increasing, it was easier for the experimental rats to have exercise-induced anemia or low hemachrome, which was because the rats were highly adaptive (Branth et l., 2008; Corbett, 2008). In this study, due to the long-term load exercise, Hb concentration, RBC count and Hct of the rats in movement groups were lower than those of quiet groups, indicating that long-term load exercise could result in the low hemachrome state of rats, further leading to the decrease in their body function and recovery ability, thus the effect of exercise training was seriously affected.

We found that long-term load exercise was also the reason why MDA in the RBC of the rats in MC group was evidently higher than that of QC group, while CAT activity of MC group was significantly lower than that of QC group, indicating load exercise resulted in the metabolic disorder of free radical in the rats of MC group as well as the increase of generated free radicals which was a major cause of the structure change of RBC membrane. In this case, the supplement of antioxidants can enhance the antioxidant capacity of the body. Quite a few studies have revealed that vitamin E, C and glutathione have a desirable antioxidant effect (Zouhal et al., 2010; Laabes et al., 2008). After seven-week TCM intervention, the MDA level of the exercise rats in groups MA, OA, MB and QB was lower than that of groups MC and QC, indicating that TCM could adjust the metabolic disorder of free radicals caused by long-term load exercise. Although the effect of gastroenteric administration on SOD activity in RBC was insignificant, its effect was reflected in that the CAT activity of the rats in groups MA, OA, MB and QB was significantly higher than that of groups MC and QC.

The cross-linking effect of IF1 with endogenous IF1 is the main factor influencing ATP synthetase. IF1 inhibitor might interfere the rotation of ATP synthase center stem (Perseghin et al., 2009; Kelly et al., 2011). So far, some studies have stated that IF1 could affect the activity of ATP synthetase as IF1 had an inhibitory effect on the influence of F1F0 complex of ATP synthase (Garthe et al., 2011; Erdman et al., 2012; Wong et al., 2012). However, there is no study reporting the influence of exercise training on IF1 yet. In this study, it was observed that the expression quantity of IF1 in exercise groups was higher than that of quiet groups, from which we speculated that exercise training could increase the expression quantity of IF1.

Skeletal muscle is the main movement organ of the body as well as a major part where lactic acid is generated in the movement state (Nosaka et al., 2009). In this study, TCM intervention could reduce the serum lactic acid level in the rats, which indicated that TCM decoction could reduce the generation of lactic acid during exercise and improve the body's ability to remove lactic acid. TCM intervention could improve the activity of serum LDH either in a quiet state or under long-term exercise load. Serum LDH is a marker enzyme of anaerobic oxidation enzymes. Its activity is usually used to evaluate the anaerobic metabolism ability of skeletal muscle, kidney and myocardium. In the study, we also found that TCM intervention could increase the serum MDH activity of exercise rats. We deduced that the TCM decoction promoted the rise of certain hormones which might have important effects on the change of MDH activity. Another speculation was that the TCM ingredients in this study contained some active substances which had inducing or stimulating effects on the activity of aerobic oxidase. In addition, this study also showed that TCM intervention led to a significant decrease in the phosphorylation level of RBC membrane band 3 protein in exercise rats. In comparison with QC group, the phosphorylation level in groups QA and QB was in a decreasing trend, indicating that TCM could improve the structure and function of RBC membrane of rats.

To sum up, with TCM intervention, we studied the energy metabolism of exercise rats after treadmill exercise, which provides theoretical support for mastering the energy metabolism of athletes in running competition.

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# RESEARCH ON INFLUENCE OF PHYSICAL EXERCISE AND NUTRITION INTERVENTION ON PHYSICAL QUALITY OF COLLEGE STUDENTS

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**ABSTRACT**
The study, setting the freshmen of a college as the objects for body-building and exercise combined with nutrition intervention and observing the difference before and after the intervention, mainly focuses on the difference of influence of body-building and sport combined with nutrition intervention on the body shape, physical function, physical quality and physical prowess of teenagers, so as to provide helpful theoretical and practical basis for nutrition intervention scheme that is used to strengthen the physical quality of teenagers.

## 1. Introduction

In recent years, according to the tracking investigation of physical exercise conditions for many years made by governments, the investment in public exercise and promotion of national body-building program are good for obtaining the economic and social benefits. Many countries has formulated body-building plan organized by the government in accordance with the national condition, and our country also has carried out the physique test on the whole people separately in 1995 and 2000. All of those activities are aimed to improve the physique of people. Many scholars think that people's physical are mainly affected by nurture, acquired disposition, environmental & social conditions and sports, but the largest factor that affects the physical is the rationality of nutrition ingestion. Reasonable nutrition can help to keep health and prolong lifespan, but the malnutrition will bring bad effect on the body or even lead to some diseases. Yang Zeyi, an expert in Sport & Exercise Nutrition, said that life lies in movement and nutrition, which made people gradually understand that the healthy living method with scientific exercise and adequate nutrition is essential and helpful for perfecting and improving people's physique. To improve people's physique scientifically and effectively, many experts, scholars and researchers have conduct a large amount of relevant study (Niu, 2013).

According to some existing literatures, college students in different places are obviously different in physical function, which is not only affected by hereditary factor, but also closely related to the local environment and economic condition (Donaldson and Hill, 2003). As is known to all, students from the south have are obviously distinguished from students from the north due to their height, weight and nutrition condition. Students from the south are poorer than students from the north in terms of height and weight, and the students from economically developed area are better than students from developing area (Kathy, 2007). Height, weight, vital capacity and other relevant indexes are the typical reference indexes to judge the physical condition of student and also the important factors for the student physique test. The south of

Yangtze River belongs to subtropical moist climate, it is warm throughout the year and rich in rains, but how are the body shape, physical quality and physical function of teenagers in this region?

In order to promote the physical health level of teenagers in Anhui Province, it is important to get aware of the physical quality, body shape and physical function, exercise features as well as the influencing factors on the physical quality of teenagers in this region and the weak point, and make them have targeted physical exercise and conduct nutrition intervention based on the existing level (Susan, 2007).

Therefore, based on the existing literatures, this paper presents the test and analysis on physical quality, physique state, physical function and other indexes of teenagers in Anhui Province after body-building exercise and nutrition intervention, and further gives the existing problems and the influencing factors. Moreover, the specific influence of intervention on physical quality of teenagers are determined through exercise and nutrition interventions in order to give suggestions to correct lifestyle and exercise way for teenagers as well as excellent exercise and diet habits for students (Wengstrom and Wahren, 2009). The study of influence of physical exercise and nutrition intervention on physical quality of teenagers in the paper can be also used for providing some practical suggestions for improving the physical quality of teenagers in our country and further perfect the health for millions of teenagers in China.

## 2. Materials and methods

For the whole society, there are lots of complex factors that affect the health of teenagers such as inheritance, material condition, physical exercise, environment, lifestyle, habit and so on. Teenagers' physical quality has become poorer and poorer in the past continuous twenty years because of not only the comprehensive factor, but also the multi-factor dynamic influence. In this paper, the influencing factors on physical quality of teenagers are divided into macro factor and micro factor according to the different levels of the teaching staff. The micro factor generally refers to what leads to the decrease of teenager's physical quality thanks to the mistakes of education decision makers, including the education PE policy and development strategy in the country or in a region (Kathy, 2007). Fox example, the relationship between PE and the politics & economy are not well done, the public body-building activity and promotion of public physical status are not set as the basis for administration of the country at the right moment, or the safety education and psychological health education are not developed as early as possible. The macro factor may have an influencing scope of the whole country, causing bad effect to many generations and seriously hindering the healthy development of the society (Wengstrom and Wahren, 2009). Micro factor refers to the mistakes of school educators in teaching process which results in the physical quality decrease of teenagers, for example, the purpose of PE course cannot meet the demand of social development, school leaders think little of PE, the PE teaching contents are out of date, the PE course is not launched on schedule but subject to other courses, PE teachers fails to run new teaching method, and PE evaluation is unreasonable and not objective(Li et al. 2010). The micro factor will bring direct impact on the well-developed in mind and body of the educatees, affecting the benefits of millions of parents or even the stable development of the whole society. Macro factor is an important aspect that affects the health of teenagers, but the micro factor shall not be neglected, as is shown in Figure 1.

### 2.1. Objects

According to the demand of the research, 120 freshmen at age of 17-20 years old of a college were selected as the objectives (Montanari, 2008). All objects were informed the research purpose and experimental method in advance and they were ensured voluntarily to take part in the experiment. The experimental members had

physical exercise and nutrition intervention for 12 weeks.

## 2.2. Methods

Though the combination of experimental and theoretical study, the data in the paper are true and reliable though the experiments and the experimental results were analyzed according to the relevant theoretical knowledge to discuss the influence of physical exercise and nutrition intervention on the physical quality of teenager.

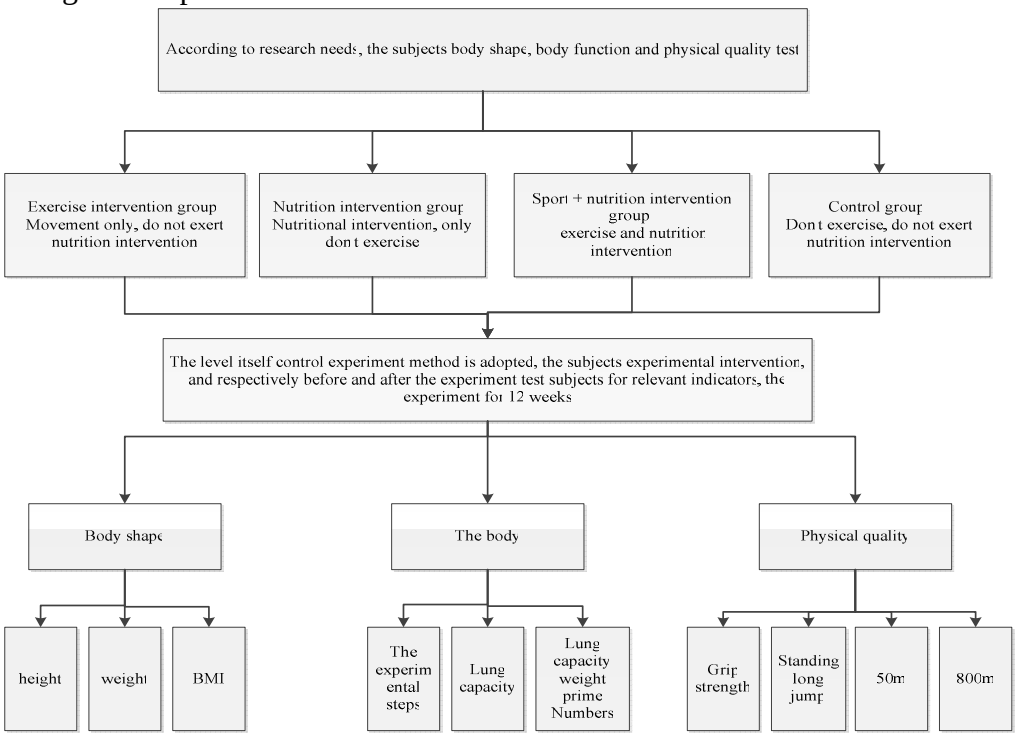


**Figure 1:** Study design road map

Specific research methods include literature study, experimentation and mathematical statistics (Palacio and Nuin, 2009).

## 2.3. Contents

According to the *Student Physical Health Standard*, the test items were: height, weight, BMI, vital capacity, step test, vital capacity/weight ratio, grip, standing long jump, 800m, and 50m.

## 2.4 Specific sport training schedule

Each time of training was launched as planed and the condition was recorded by the tester day to day. After training, the tester communicated with subjects timely to observe the body reaction and ask the feelings, and adjustment was made when necessary to ensure the normal training schedule. The specific training plan is shown in Table 1.

**Table 1.** Specific training schedule

| Weeks | Exercise | Exercise time | Exercise intensity | Exercise place |
|---|---|---|---|---|
| 1 | Slow rope skipping, transform the terrain to run, game | 60min | 120-140 time/min | playground |
| 2 | Slow rope skipping, 3000 jogging ,game | 60min | 120-140 time/min | playground |
| 3 | Slow rope skipping, 15 min jogging, game | 60min | 120-140 time/min | playground |
| 4 | Basketball dribbling running, The stairs, Slow running, game | 60min | 120-140 time/min | playground |

**Table 1.** - continue

| Weeks | Exercise | Exercise time | Exercise intensity | Exercise place |
|---|---|---|---|---|
| 5 | Aerobics, Strength training, game | 60min | 120-140 time/min | playground |
| 6 | Basketball dribbling running, Strength training, game | 60min | 120-140 time/min | playground/outdoor |
| 7 | aerobics, Strength training, game | 60min | 120-140 time/min | playground/outdoor |
| 8 | aerobics, swiming, game | 60min | 120-140 time/min | playground/outdoor |
| 9 | Basketball dribbling running, game | 60min | 120-140 time/min | playground/outdoor |
| 10 | Slow running, game | 60min | 120-140 time/min | playground/outdoor |
| 11 | aerobics, game | 60min | 120-140 time/min | playground |
| 12 | Rope skipping, leapfrog, game | 60min | 120-140 time/min | playground |

The training plan is divided into three stages according to the exercise strength: the first stage (1~3 weeks) is the adaption stage, emphasizing on low intensity of training; the second stage (4~7 weeks) is the improvement stage mainly on medium intensity as well as small amount of high intensity exercise; and the third stage (8~12 weeks) is the consolidation stage mainly for medium and high intensity training (Montanari, 2008). The training time is 5:00-6:00 pm of each day. The subjects wear heart rate monitor during the training process to maintain their heart rates within the applicable scope as far as possible. The researcher recorded the content, attendance, reaction of trainees each time and shall also duty the supervision at the same time. Considering that the trainees are teenager students and in order not to impact their physical and psychological heath, attentions shall be paid to the following matters:

(1) Warm up before exercise and do some relaxed movements after exercise to avoid exercise injury;

(2) Communicate with trainees timely and observe the changes during training; conduct proper treatment for the trainees with special condition, and adjust the training plan as per the specified condition (Palacio and Nuin, 2009);

(3) Check the dressing of the trainees to prevent their body injury because of clothing.

### 2.5. Formulation and implementation of nutrition intervention plan

The nutrition intervention scheme is formulated after surveying the dietary condition and knowing the nutrition level and eating habit of the subjects.

According to Table 2, we can see that the carbohydrate, protein and total energy intake of the subjects everyday are lower than the recommended nutrient intake, with the differences of 3.02g/kg, 0.58g/kg and 0.65g/kg respectively, indicating that the daily diet of the subjects is not very appropriate. The scope of ratio of energy supply of carbohydrate to the total energy supply is within the recommended values while that of fat has exceeded the recommended maximum, indicating that fat intake should be reduced in the diet. The energy supply ratio of protein is higher than 25%, close to the recommended top limit, and there is an unbalanced phenomenon where the actual protein intake is little while the energy supply ratio is high, which will cause reduction of the protein level of an organism for a long time (Chen, 2013).

It can be seen from Table 3 that only the vitamin E intake of the subjects is consistent with the recommended value while other vitamin intake is less than that. These elements are mainly from fruits and vegetables. According to the dietary statistics of the subjects, we can see that most of them eat less coarse food grain, fruits or other foods, which is one of the main reason why they lack of vitamins. Moreover, the mineral

intake is also relatively small, only with the selenium intake close to the recommended amount, indicating that the mineral intake of the subjects is insufficient. There are two subjective reasons causing these phenomena, one being no knowledge of foods containing these elements and the other no scientific diet consciousness, so theoretical guidance and learning about a rational diet should be strengthened.

**Table 2.** Subjects were total calorie intake and the three major nutrients TAB

| | Carbohydrates | Fat | Protein | The total energy |
|---|---|---|---|---|
| Intake | 3.95 | 1.59 | 1.36 | 29.36 |
| The recommended intake | 6.25 | 1.56 | 1.25 | 56.12 |
| The proportion of power | 52.13 | 11.32 | 26.35 | |
| Recommended power ratio | 50-60 | 13-16 | 25-32 | |

**Table 3.** Before the trial subjects body form basic statistics

| Indicators | The control group | Sports group | Nutrition group | Sport + nutrition group | T | P |
|---|---|---|---|---|---|---|
| Height | 160 | 160.7 | 160.2 | 160.3 | 0.000 | 0.999 |
| Weight | 52.21 | 56.32 | 55.23 | 55.20 | -0.163 | 0.939 |
| BMI | 23.12 | 23.21 | 23.21 | 23.65 | 0.788 | 0.413 |

## 3. Results and discussions

### 3.1 Pre-experiment homogeneity analysis of physical function

Before the experiment, physical function indexes of the control group, motion group, nutrition group and motion & nutrition group had been tested, and the test results indicate that there is no significant difference in physical function indexes of the four groups of students, i.e. students' physical function indexes are almost consistent with each other.

### 3.2 Results of post-experiment body shape change of the subjects and analysis

It can be seen from Table 4 that there is no significant difference in the height of the control group, nutrition group, motion group and motion & nutrition group before and after the experiment. In weight, there is no significant difference for the control group before and after the experiment while a significant difference ($P<0.01$) for the motion group, group & nutrition group before and after the experiment; a significant difference also exists for the nutrition group before and after the experiment. In the aspect of BMI, there is no significant difference for the control group before and after the experiment while a significant difference for the motion group ($P<0.01$) as well as the nutrition group and group & nutrition group ($P<0.05$) before and after the experiment.

It can be seen from Table 5 that there is no significant difference in the height of the control group, nutrition group, motion group and motion & nutrition group before and after the experiment. It shows that the intervention plan of 12 weeks of aerobic exercise with the heart rate of 120-170 times/min companied with the recipe and nutrition education oriented nutrition intervention plan has little impact on college students' height (Zhang et al. 2012). It is mainly because that the height of 70% of teenagers is influenced by congenital factors while less influenced by acquired factors such as environments, nutrition, and exercise.

It can be seen from Table 5 that the weight of the motion group and the motion & nutrition group has decreased obviously with an significant difference (PCO.OI) compared with the control group and the weight of the nutrition group has increased obviously with an significant difference ($P<0.05$) after the experiment compared with the motion group.

The experiment shows that scientific diets and physical exercise are good for weight loss while only diet control or physical exercise does not have a good effect. By analyzing the reason, I think this is mainly because that the subjects have maintained a certain amount of exercise and a rational diet during the experiment. Relevant data show that the weight of a human body decreases obviously in the initial stage of physical activities resulting from that the moisture and fat of the body have been consumed during exercise and then decreases slowly. From an energy supply standpoint, according to the intervention plan in the study, the subjects' heart rates are kept at 120-150 times/min and even an amount of exercise with the heart rate of 160 times/min is reached which is equal to a long run at the speed 4km/h. This intense exercise is within the scope of aerobic energy supply, so the energy consumed is almost supplied by steatolysis. Therefore, the subjects' weight has decreased at different levels. This point can also provide a scientific theory basis for obesity people to lose weight. In addition, it was also discovered during the experiment that some students in the motion group had a bigger demand for foods after having consumed a lot of energy in exercise due to no nutrition intervention plan and their weight fails to reduce obviously and even increases due to no control of their diet and excessive intake of calories higher than the consumption of calories. Therefore, teenagers should also pay attention to diet control during physical exercise to avoid weight increase due to overeating.

It can be seen from Table 5 that, compared with the control group, the BMI of the motion group and the motion & nutrition group ($P<0.01$) as well as the nutrition group ($P<0.05$) increases significantly after the experiment. BMI is closely related to the weight and height, so is change in BMI. The three indexes almost increase.

**Table 4.** After the experiment subjects body form basic statistics

| Indicators | The control group | Sports group | Nutrition group | Sport + nutrition group | T | P |
|---|---|---|---|---|---|---|
| Lung capacity | 2958.32 | 2921.65 | 2914.25 | 2945.45 | 0.004 | 0.998 |
| Vital capacity index weight | 33.23 | 33.54 | 32.12 | 32.65 | 0.030 | 0.888 |
| Bench test | 45.12 | 44.12 | 44.56 | 44.75 | 0.625 | 0.524 |

**Table 5.** Before the trial subjects body form basic statistics

| Indicators | The control group | Sports group | Nutrition group | Sport + nutrition group | T | P |
|---|---|---|---|---|---|---|
| Height | 160 | 160.7 | 160.2 | 160.3 | 0.000 | 0.999 |
| Weight | 56.23 | 48.12 | 52.68 | 50.20 | 5.03 | 0.011 |
| BMI | 23.56 | 26.36 | 26.48 | 26.65 | 5.50 | 0.013 |

### 3.3. Results of post-experiment physical function change of the subjects and analysis

The vital capacity/weight ratio is the ratio of vital capacity of a human body to the weight, i.e. a relative value of vital capacity per 1kg reflecting the degree of correlation between the vital capacity and weight, which is used to carry out objective quantitative comparative analysis of individuals and groups at different ages and of different genders (Qi and Tian, 2011). The ratio can also provide a reference for material selection for oxygen metabolism sport athletes and students' physical general evaluation. The calculation formula: vital capacity/weight, unit: milliliter (ml) for vital capacity and kilogram (kg.) for weight. The step test index is an important index reflecting individual cardiovascular status. The greater the ratio is, the higher the skill level is, on the contrary then is lower (Zhang et al. 2013).

It can be seen from Table 6 that there is no significant difference for the control group and nutrition group in the aspect of vital capacity index before and after the experiment while a significant difference for the motion group and the motion & nutrition group ($P<0.01$). There is no significant difference for the control group and nutrition group in the aspect of vital capacity/weight ratio before and after the experiment while a significant difference for the motion group and the motion & nutrition group ($P<0.01$). There is no significant difference for the control group and nutrition group in the step test index before and after the experiment while a significant difference for the motion group and the motion & nutrition group ($P<0.05$).

It can be seen from Table 7 that compared with the control group, the vital capacity of the motion group has increased obviously with a significant difference ($P<0.05$) and that of the motion & nutrition group after the experiment also has increased obviously with a significant difference ($P<0.01$); compared with the motion group, the vital capacity of the motion & nutrition group has increased obviously with a significant difference ($P<0.05$), and compared with the nutrition group, the vital capacity of the motion & nutrition group has increased obviously ($P<0.01$). The vital capacity of the subjects after nutrition intervention has slightly increased and the motion group and motion & nutrition group have a greater increase range, indicating that physical exercise can increase teenagers' vital capacity. The vital capacity is closely related to exercise intensity. With the increase of exercise intensity, the vital capacity will increase. Under the exercise intervention plan, the subjects have been in exercise with moderate intensity for a long time and the muscle body can generate a lot of $CO_2$ while generating energy. To remove $CO_2$ out of the body, the respiratory system in muscle body is required to increase the workload. With the increase of depth of respiration, the respiratory muscle is exercised and its strength is increased. With the increase of exercise time, the respiratory rate will decrease and slow and deep respiratory status will appear where the respiratory muscle can get a full rest when lung is assured to have oxygen, which effectively increases the respiratory capacity and further increases the vital capacity and also promotes the subjects free of asthma during exercise.

**Table 6.** Each body function index change before and after the experiment table

| | | The control group | T | P | Sports group | T | P | Nutrition group | T | P | Sport + nutrition group | T | P |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Lung capacity | Before | 2947.21 | 0.029 | 0.989 | 2865.32 | 2.855 | 0.007 | 2915.65 | 0.588 | 0.625 | 2945.32 | 5.231 | 0.000 |
| | after | 2950.23 | | | 3121.23 | | | 2994.23 | | | 3430.21 | | |
| Vital capacity index weight | Before | 33.21 | 0.237 | 0.852 | 33.52 | 2.798 | 0.008 | 32.15 | 0.914 | 0.362 | 32.14 | 3.126 | 0.003 |
| | after | 33.12 | | | 40.26 | | | 35.21 | | | 41.25 | | |
| Bench test | Before | 45.21 | 0.234 | 0.845 | 44.56 | 2.212 | 0.038 | 44.15 | 0.727 | 0.419 | 45.36 | 2.512 | 0.018 |
| | after | 46.36 | | | 49.12 | | | 46.32 | | | 50.02 | | |

**Table 7.** Before the trial subjects body form basic statistics

| Indicators | The control group | Sports group | Nutrition group | Sport + nutrition group | T | P |
|---|---|---|---|---|---|---|
| Lung capacity | 2945.23 | 3125.36 | 2991.23 | 3436.21 | 6.123 | 0.007 |
| Vital capacity index weight | 33.25 | 42.12 | 35.42 | 41.36 | 5.321 | 0.009 |
| Bench test | 46.32 | 49.23 | 46.81 | 50.12 | 4.412 | 0.032 |

## 4. Conclusions

In the paper, through exercise intervention and nutrition intervention test on 120 freshmen and post-experiment data analysis, it is obtained that different plans have influences with different degrees on college students' body shapes, physical functions and physical quality which is detailed as follows:

(1) In the aspect of physical functions, the subjects' vital capacities increase slightly.

Physical exercise increases the capacity of respiratory system, besides, some foods in the nutrition intervention plan such as apples and tomatoes also have obvious promotion functions on vital capacity. However, only nutrition intervention has no effect. Step tests indicate that the cardiovascular function has improved after the subjects experience intervention.

(2) In the aspect of physical quality, what is different from other literatures is that the subjects' upper body strength has improved after exercise intervention, which mainly results from that the items promoting the upper body strength in the exercise intervention plan for the subjects with weak upper body strength. The lower body strength of the subjects in different groups has increased, indicating that intervention plans can promote the lower body strength of teenagers. Standing long jump results of the subjects in different groups before and after the experiment are different from each other significantly, indicating that exercise intervention and nutrition intervention can obviously promote the speed quality of the subjects.

## 5. Acknowledgements

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# ESTABLISHMENT AND GUARANTEE OF FOOD LOGISTICS SYSTEM FOR BIG SPORTS EVENT

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**ABSTRACT**
This paper analyzed the detailed chains and agents involved in the food logistics framework at big sports event and pointed out the main problems which should be noticed in the logistics food framework. Finally, detailed measures of establishing food logistics security system and framework at big sports event were proposed.

## 1. Introduction

The holding of big sports event has great significance upon the development of sports, a nation's economic and cultural advancement and the promotion of people's sportsmanship. Especially, the internationally big sports event can expand these influences into the whole world, and are important to improve China's international status. To achieve the above goals, ensuring the safety of food logistics at big sports events must be paid high attention by the organizers (Vlachos, 2014).

The security of food logistics at big sports event is not only directly related with the dietary health of all personnel, but also directly influences the performance of contestants. If any accidents caused by food logistics safety happened at any international big sports event held by China, any opportunity of China continuing to obtain the right to hold a similar event as well as the international influence shall be significantly negatively affected.

This paper analyzed the detailed chains and agents involved in the food logistics framework at big sports event and pointed out the main problems which should be noticed in the logistics food framework. Finally, detailed measures of establishing food logistics security system and framework at big sports event were proposed.

## 2. Materials and methods

The food logistics at big sports event usually comprises two aspects, namely logistics of purchasing ingredients demanded by food production and logistics of distributing finished food products.

### 2.1. Security for the logistics of purchasing

Purchasing logistics is comprised of ingredients choosing, transportation and storage. Since it directly affects the quality and quantity of ingredients, it's the foundation for food logistics security at big sports event.

**(1)Safety Guarantee for the Quantity of Ingredients**

Big sports event usually involves people from different races, religions and regions, which makes greatly different demands for food, thus more types of ingredients are required. The organizers of big sports events must consider the

great differences in food requirements, and notice what ingredients can't be locally purchased or need special treatment. The quantity of ingredients purchased at other places or specially treated should be paid more attention so as to insure the corresponding requirements can be met.

**(2) Safety Guarantee for the Quality of Ingredients**

It mainly includes the quality of ingredients as well as quality of ingredients at the transportation and storage. After the purchasing contracts being established, the ingredients must be chosen according to the quality standards specified at the agreed purchasing contract. Then the ingredients must be transported to the warehouses of organizer as soon as possible, and distributed to the food manufacturer rapidly (Amani et al. 2013). The most frequently occurring problems at transportation and storage period are decomposition and secondary pollution of ingredients. On one hand, on the basis of shortening time required for transportation and storage, cold-chain transpiration & cold storage should be employed to prevent ingredients from being decayed. On the other hand, on the basis of guaranteeing basic food hygiene, different kinds of ingredients should be transported by dedicated vehicles to prevent them from affecting each other, resulting in secondary pollution (Durak and Ünverdi, 2014).

## 2.2. Security of the Logistics for Distributing Finished Food Products

It means the process of finished food products being eventually distributed to contestants and other related personnel at big sports event, during which the quality and quantity of food should also be guaranteed. It's the final and the most important stage for guaranteeing the food logistics safety (Soysal et al. 2012).

**(1) Safety Guarantee for the Quantity of Distributed Finished Food Products**

If the ingredients have been purchased and manufactured according to the dietary habits of personnel attending the sports event, the total quantity safety of logistics for food distribution can be guaranteed. One of the major quantity safety issues is the quantity guarantee issue of food distribution both at different times and spaces, which involves learning in detail and scientifically arranging the different dietary habits, especially where and when these people with special dietary habit shall have their food. Different people should be guaranteed to have their desired and enough food at normal times and places.

**(2) Safety Guarantee for Distribution Process after Food Being Manufactured**

It mainly includes two aspects, namely transportation and storage. The most basic measure is to shorten the time required by these two processes as much as possible. For hot food, on the basis of shortening time required by storage and transportation, heat-preserving issue should also be pay much attention. However, for food of other sorts, to prevent them from being decayed and on the basis of shortening required time, the best way is to employ cold-chain transportation and cold storage(seen in Figure 1).

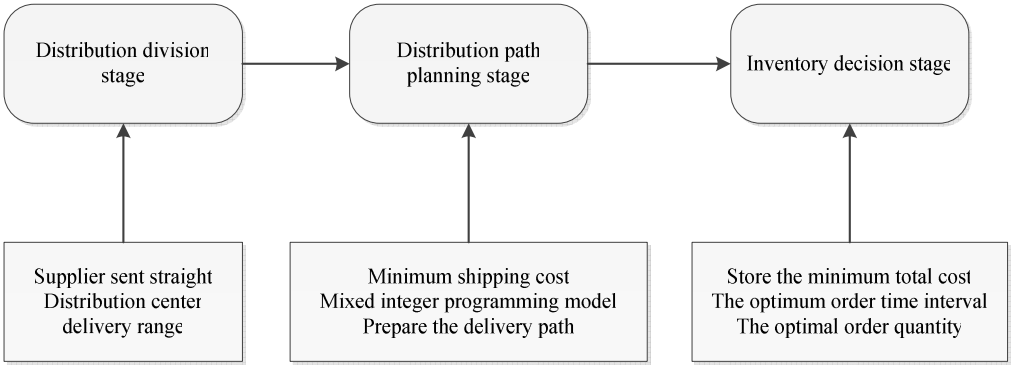


**Figure 1:** Big events logistics operation mode process

## 3. Results and discussions

Assumption: the sequence of the number of participants over the years in similar large-scale sports events

$$a^{(0)} = \left(a^{(0)}(1), a^{(0)}(2), \cdots, a^{(0)}(n)\right) \quad (1)$$

To use accumulated generating transform data:

$$a^{(1)}(k) = \sum_{m=1}^{k} a^{(0)}(m) \qquad k = 1, 2, \cdots, n \quad (2)$$

To calculate the stage ratio variance of data, and determine whether it is suitable for the grey forecasting model, the relevant formula is as follows:

$$\delta(k) = \left|1 - \frac{a(k-1)}{a(k)}\right| \quad (3)$$

If the stage ratio variance is in an admissible scale (0.1353, 7.389), the GM (1, 1) modeling can be done. After data processing and inspection, the number of future participants will be predicted, as shown in formula (4), (5).

$$a^{(1)}(k+1) = \left(a^{(0)}(1) - \frac{\beta}{\alpha}\right)e^{-\alpha k} + \frac{\beta}{\alpha} \quad (4)$$

$$a^{(0)}(k+1) = a^{(1)}(k+1) - a^{(1)}(k) \quad (5)$$

$$z^{(1)}(k) = 0.5\left(a^{(1)}(k) + a^{(1)}(k-1)\right) \quad (6)$$

$$C = \sum_{k=2}^{n} z^{(1)}(k) \quad (7)$$

$$D = \sum_{k=2}^{n} a^{(0)}(k) \quad (8)$$

$$E = \sum_{k=2}^{n} z^{(1)}(k) a^{(0)}(k) \quad (9)$$

$$F = \sum_{k=2}^{n} z^{(1)}(k)^2 \quad (10)$$

$$\alpha = \frac{CD - (n-1)E}{(n-1)F - C^2} \quad (11)$$

$$\beta = \frac{DF - CE}{(n-1)F - C^2} \quad (12)$$

After getting the number of future participants and the amount of arrived luggage from all participating delegations, with the grey forecast method, the average amount of carried luggage per person is necessarily required, whose influencing factors are divided into two kinds: the inner factors of sports events, depending on demands and projects of sports events, for example, more equipment is needed in the Olympic Games than in an individual world championship or similar World Cup projects. More equipment is needed in winter games than in summer games; the external factors of sports events, mainly referring to the difference among participating countries, that is, the difference among sports equipment and luggage comes from the difference in the national economic level and emphasis degree (Morganti, 2011). For example, the average amount of carried luggage per person is 2.783 [1.333, 4.923] in the 21st university sports meeting of Beijing, and its deviation is 0.9727, which shows the difference is more obvious (Zunder et al. 2013).

According to the principle of homomorphism, because the large-scale sports events are mostly mature and relatively fixed, the amount of luggage is relatively fixed for similar sports events, and it fluctuates up and down with the national economic cycle and event influence (Ellinger, 2006). So, a simple average method is adopted to predict the average amount of luggage per person, as follows:

$$X_t = \left(\frac{1}{t-1}\right)\sum_{i=1}^{t-1} D_i \quad (13)$$

In order to get the average amount of luggage per person in current sports event, we only need the actual average amount of luggage per person for previous sessions. Therefore, the amount of arrived luggage from all participating delegations is as follows:

$$Y_t = aX_t = \left(\frac{a}{t-1}\right)\sum_{i=1}^{t-1} D_i \quad (14)$$

### 3.1. Quality safety problems of food materials from individual suppliers

In general, the suppliers of food material must be some reputable supply companies of food material through strict certification in order to guarantee the quality safety of food logistics. Due to a variety of unplanned situations, some certificated companies still cannot completely meet the demands of food material supply. Under special circumstances, therefore, the

supplement of individual suppliers is required. With the lack of technical facilities and guarantee abilities of quality safety, there inevitably will be some problems in food material quality, especially the health conditions do not conform to the standard.

### 3.2. Storage temperature control

Both the food material and the processed food should be stored for some time, if they can't be immediately eaten. In order to retain freshness, the food material generally requires hot storage for hot food or cold storage for cold food, for which the temperature should be controlled in a certain range (Pasternak and Pellissier, 2014). The equipment can automatically control the temperature in hot storage room or cold storage room, without absolute accuracy occasionally, which cannot meet the standard and lead to deterioration of food or food material if the disposal is not in time (Vander et al.2005).

### 3.3. Shortage of key equipment

If the cold storage facilities and thermal sterilization equipment are input largely in one go, the processing will become difficult after events. In view of cost, the organizers reduce the purchase of these devices as far as possible. As the key equipment assuring the food logistics safety, the lack of cold storage facilities and thermal sterilization equipment will influence the efficiency of food logistics safety management. And for the efficiency, some necessary steps of food logistics safety are omitted, causing the potential security problems.

### 3.4. The insufficient staff of logistics safety management

For the control power of the safety of food logistics, the large-scale sports events are far more than general group activities, which need a lot of staff to meet the needs of strict safety management(Jack and Vander, 2009). Generally speaking, the organizers usually have a science arrangement for human, rarely suffering from understaffed problems. However, there exist some abnormal situations in the large-scale sports events (Vorst et al. 2005). For example, when the organizer’s inspection agency of food material finds that a group of food material cannot meet the health indicator, in order to eliminate the potential safety problems it will deal with the unqualified food material, disinfection of the corresponding transport vehicles and personnel, etc, which largely increases the organizer's work. The peak of these abnormal situations will cause the insufficient staff of logistics safety management.

### 3.5 The health management of relevant volunteers and temporary employers

The volunteers and temporary employers only attend the knowledge training about the food logistics safety, temporarily organized by the organizer. The training is very important for the food logistics safety ensured by them, but it is not enough to help them form a habit fully conforming to the safety standards of food logistics. In many large-scale sports events of China, some volunteers enter into work places without disinfection in accordance with the health regulations.. In order to keep the job, some temporary employers don’t report to the higher level in accordance with the relevant provisions when suffer from the influenza, dysentery and other infectious diseases. The accidents of food logistics safety haven’t appeared so far, though, the potential safety problems should be paid attention to by organizers, as is shown in Figure 2.

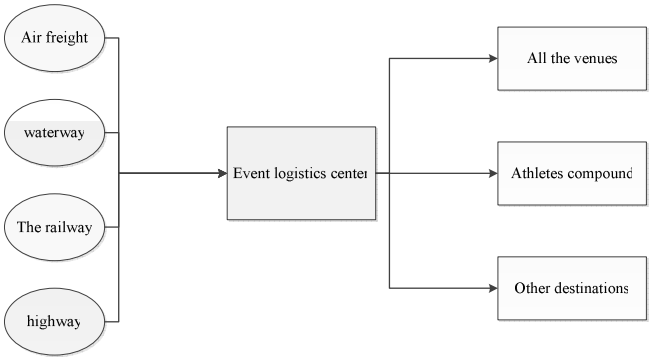


**Figure 2. E**vent logistics center system traffic connection diagram

### 3.6. Unexpected problems caused by water, power and gas supply

The normal supply of water, power and gas is the most basic condition for the normal operation of food logistics in large-scale sports events, for which the construction should have been very mature in those cities holding large-scale sports events (An and Ruck, 1999). However, there are still some abnormal situations, such as the water-break, power-break, gas-break, water pollution and so on, which cannot meet the corresponding standard of food logistics safety. For the food logistics safety, if the abnormal situations of water, power and gas cannot be solved in time, it will be likely to cause the collapse of the food logistics safety system, resulting in a serious consequence. At present, the organizers have more rich experience in safety management of food logistics in large-scale sports events of China, which is conducted in strict accordance with the requirements of the HACCP management system, without big safety problems generally. But once some problems appear, some potential safety problems will accordingly appear, and even resulting in some bad safety accidents about food logistics, so the corresponding measures are expected for its prevention.

## 4. Conclusions

In large-scale sports events, we must place a strict safety control on links, subjects and potential problems of food logistics safety in accordance with the requirements of HACCP management strictly, in order to build a scientific and effective system framework of food logistics safety. And we have to do the pre-arranged planning for all sorts of unexpected accidents to ensure the smooth progress of events.

**(1) To set up the leading agency with a unified management and coordination of food logistics safety.**

The leading agency has a supreme power to manage and coordinate the safety of food logistics in large-scale sports events, and takes charge of building a safety framework of food logistics system and ensuring its normal operation, including the safety planning and managing supervision of all links and subjects involved by food logistics safety, formulation and audit of all management provisions, coordination with other relevant departments, disposal of abnormal situations, etc. The establishment of the agency can ensure a smooth operation of food logistics safety under the condition of unified management and coordination.

**(2) To set up an independent agency of health detection and disinfection for food logistics safety.**

In addition to agencies of procurement logistics and food distribution, we required an independent agency of health detection and disinfection for food logistics safety. The agencies of procurement logistics and food distribution possibly don't disinfect the personnel and facilities in strict accordance with the provisions for their own interests or human saving, and for some existing problems or potential safety problems of food logistics, they

don't report to the superior and dispose them timely according to the regulations.

**(3) To establish a perfect training system of food logistics safety**

The food logistics safety training should not be limited to the temporary induction training of the staff in safety positions. In order to guarantee the employers and university volunteers to carry out food logistics safety ideologically, and form the correct behavior patterns, more training and education are required after induction, which can combine with the daily work meeting system, and can aim at specific problems to organize the small specialized training for the relevant staff. The system of continuous training combines with specific work closely, which is conductive to make the related personnel keep a profound impression and form a correct behavior of food logistics safety.

**(4) To improve the propaganda and education of food logistics safety to contestants**

As the direct consumers of food, the contestants are most concerned about the food logistics safety. We should improve the propaganda and education of food logistics safety to them, which can both make them take correct measures timely in food logistics safety accidents, avoid damage increase, and make themselves an important force in safety monitoring of food logistics. A method is provided for the curriculum education reform of food chemistry, based on the basic knowledge of food logistics safety for contestants, evaluation methods of the students' comprehensive quality and so on. So, a teaching effect is achieved, in which we can cultivate students' autonomous learning ability of food chemistry, personal ability, interpersonal communication ability as well as the whole CDIO ability.

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# ANALYSIS OF THE METABOLIC REGULARITY OF BODY IMPORTANT INDEX OF OBESE ADOLESCENTS FROM THE ANGLES OF SPORTS AND NUTRITIONAL INTERVENTION

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**ABSTRACT**
This paper has compared the results of various indicators before and after experiments of implementing three different intervention programs of simple exercise, simple nutrition and combination of both for ten weeks among overweight and obese adolescents. It has been found out that different programs have different influence on the body composition, circumference, physiological function, physical quality and partial metabolic indicators of overweight/adolescents by observing the differences in physique and metabolism between overweight/obese adolescents and non-overweight/obese adolescents aging from 12 to 16.

## 1. Introduction

In recent years, with the changing of people's way of life, the detection rate of overweight and obese children is increasingly rapidly year by year, coming with the decreasing of physiological function and physical quality of children as well as the increasing of metabolic diseases such as hypertension, diabetes, dyslipidemia and metabolism syndrome etc. Children obesity not only has influence on their social adaption and psychology, for the obesity during childhood, the relative risk of obesity after grown up is obviously higher than those who have normal weight during childhood, which has increased the morbidity rate and mortality rate of adult chronic disease and cardiovascular diseases. Therefore, it is very important for the current and future health of obese adolescents to seek for effective and safe intervention means to improve physical quality and decrease the happening of metabolic related diseases.

Numerous researches have shown that suitable decrease of weight can improve body composition, strengthen physical quality and change metabolic characteristics etc. Till today, a lot of scholars both at home and abroad have adopted sports, nutrition, psychology, medicine and operations etc. (Mallinson et al. 2013). To decrease the weight of obese children, while sports and nutritional intervention are means adopted most at present. There is still controversy in the selection of suitable sports intensity, design for sports program as well as strategy and means of nutritional intervention etc. for reducing the weight of children (Kleiner, 2003). Study and compare the effects of simple sports, simple nutrition as well as the combination of two for reducing the weight of obese children and for reducing the fat content. In the process of reducing weight, the research on physical quality and metabolic characteristics is not adequate (Naidoo et al. 2009); especially the research results of effects of sports and nutritional intervention on related indicators of

fat and glucose metabolism are not exactly the same. Considering about the individual differences existed in physical quality and metabolic disorder of obese children, further research needs to be made on how to adopt suitable intervention means and design suitable individual program based on different physical and metabolic disorders.

This research has taken middle school students in urban area aged between 12 and 16 who are in critical period of weight growth as research objects, made intervention of simple nutrition, simple sports as well as combination of both for ten weeks through making aerobic exercise program and nutritional intervention program and observed the changes of body composition, physiological function, physical quality, blood lipid and blood glucose and other metabolic indicators before and after intervention and tried to discuss the following problems:

(1) Influence of sports and nutritional intervention on physique of overweight and obese adolescents;

(2) Influence of sports and nutritional intervention on partial indicators of fat and glucose metabolism of overweight and obese adolescents. Decrease the degree of obesity of overweight and obese adolescents through research on above two aspects; discuss the differences in the influences of sports, nutrition and combination of sports and nutrition on their body composition, physiological function, physical quality and metabolic influence, and then provide theory and practical material for the design of individualized intervention program to enhance physical fitness and improve metabolism of overweight and obese adolescents.

## 2. Materials and methods

Physique refers to the quality of human body, which is the human form structure expressed on the basis of hereditary and acquisition. The comprehensive characteristics of physiological function and physical quality are the material basis of human production and life. The physique mainly includes the following five aspects: body shape development, physiological function level, physical quality level, psychological balance state and adaptability. The physique of students is always the focus of education and sports department. It has been pointed out in the national physical fitness monitoring report that compared with 1995, at present, the physical fitness of our students is decreasing, which is mainly presented in speed, endurance, flexibility, explosive force and strength etc, while the obese students are increasing obviously, which has great influence on the decreasing of the overall quality of adolescents. At present, improving the physique especially the physique of obese students has drawn the attention of all social circles.

The body composition of obese children has the characteristics of large weight and high percentage of body fat etc. Feng Ning and others determined the body composition for 356 children of 7 years old with normal weight and overweight with adoption of double energy X-ray absorption method; results have shown that compared with the children with normal weight with the same age the gender, the obese children have the characteristics of high fat-free mass index (FFMD), high fat mass index (FMI), high BMI and high body fat percentage). The body fat percentage of obese children is between 30% and 45%, while the percentage of children with normal weight is between 10% and 30%. The FMI and BMI of obese children present high degree of positive correlation and the correlation coefficient is bigger than that of children with normal weight; FFMI and BMI of obese children present moderate positive correlation. Compared with children with normal weight, the obese children present different degrees of decrease in body shape, physiological quality and physical quality. In the aspect of cardio-pulmonary function, the obese children have the characteristics of low $VO_2$max, big quiet pulse and high systolic pressure etc. Nianhong Yang and others have made investigation on obesity as well as related influencing factors among 56, 150

students from middle and primary schools in 11 big and medium-sized cities; investigation results have shown that there is no significant difference in the weight of the two groups of children, but the weight, bust, pulse, blood pressure and vital capacity of obese group are obviously higher than that of the group with normal weight; vital capacity index is obviously lower than non-overweight obesity group and the differences have significant meaning. McGavock and others have studied the obese children aging from 5 to 19 and the research results have shown that the systolic pressure (SBP) of obese children is obviously higher than children with normal weight (Łagowska, 2014). The tracking survey from 2004 to 2006 has shown that in these two years, the increased SBP value of people with the most BMI increase is 4.5 times of that with the minimum BMI increase; SBP will increase by 0.77mmHg with the increasing of weight by 1kg. Research made by Zhengzhen Wang and others has shown that the cardiac and pulmonary function of overweight and obese adolescents aging between 12 and 14 is obvious lower than children of normal weight at the same age. People with hypertension account for 32.92%, however, the correlation between increased blood pressure and change of body composition is not obvious;

The vital capacity index of obese adolescents has decreased obviously, the vital capacity index of obese male is in moderate negative correlation with body fat percentage (Chen, 2015); the relative maximum oxygen intake of obese adolescents decreases obviously. Research made by Rizzo and others has shown that the fat weight of children body is in negative correlation with cardiac and pulmonary function; the physical activity level of girls is in negative correlation with metabolic risk factors (HDL, TG, FBG and others are higher than the mean plus standard deviation of people with normal weight). The author suggests that the correlation of CPF and metabolic risk factors is bigger than the correlation degree of CPF and physical activity level. In addition, in the correlation of CPF and metabolic risk factors, the body fat weight plays a negative role.

Children obesity can be divided into simple obesity and secondary obesity. Secondary obesity refers to obesity with clear cause. However, the simple obesity is closely related to way of life with characteristics of excessive eating, less physical activity and behavioral biases and it's a chronic disease of hyperplasia of body adipose tissue. At present, two main methods of defining children obesity at home and abroad are weight for height and body mass index. In 1997, International Obesity Task Force (IOTF) suggests to adopt body mass index (BMI) to reflect the body fat index of school age children and adolescents as well as the parameter of estimated incidence of obesity. At present, two BMI classification standards made by National Center for Health Statistics (NCHS) and IOTF recommended by World Health Organization (WHO) are widely applied international. In Nov. 2003, working group on obesity in China (WGOC) of International Society for Life Science has made defined standards for overweight and obese children aging between seven to eighteen suitable for our children; the definition for obese children in domestic generally adopts this standard.

## 3. Results and discussions

This research takes the students from the first and second grade of one middle school in Xicheng district of Beijing city as research objects. Inclusion standards and exclusion standards of research objects are as Table 1.

**Table 1.** Basic information of each group subjects

| | The overweight /obese group | Sports group | Fewer ten patients experienced septic | Sport + and fewer ten patients experienced septic complications | Obesity in the control group |
|---|---|---|---|---|---|

| | | | complications | | |
|---|---|---|---|---|---|
| age | 13.26 | 12.95 | 13.26 | 13.69 | 13.48 |
| height | 161.23 | 166.23 | 165.21 | 163.54 | 163.47 |
| weight | 52.12 | 71.58 | 78.02 | 74.25 | 75.21 |
| BMI | 19.25 | 25.12 | 27.25 | 26.32 | 27.95 |

### 3.1. Exclusion standards

Patients of secondary obesity, patients diagnosed with diabetes, patients with severe organic diseases of heart, brain, lung, kidney and motion system etc, patients with hepatic dysfunction, progressive fatal disease, patients with a history of alcoholism or drug abuse, patients with chronic disease and taking drugs, patients unwilling to sign informed consent (Malayil, 2014).

Based on entrance examination data and basic testing results of students, finally 124 students have been selected for this research, in which there are 94 overweight/obese people and 30 non-overweight/obese people. On the voluntary basis of participants, divide the overweight and obese students into five groups. There is no statistical difference in BMI of overweight/obesity groups, details as shown in Table 2.

**Table 2.** The stages of exercise group and exercise + and fewer ten patients experienced septic complications exercise intensity and its corresponding heart rate

| | Sports group | Sports+nutrient group |
|---|---|---|
| 40%VO2max(ml/kg/min) | 14.25 | 15.65 |
| Corresponding to the heart rate | 128.25 | 128.54 |
| 50% VO2max(ml/kg/min) | 18.54 | 18.84 |
| Corresponding to the heart rate | 140.36 | 141.25 |
| 60% VO2max(ml/kg/min) | 22.52 | 22.84 |
| Corresponding to the heart rate | 154.23 | 156.32 |
| 70% VO2max(ml/kg/min) | 25.85 | 26.51 |
| Corresponding to the heart rate | 167.25 | 167.45 |

### 3.2. Research Methods

Research procedures and technical routes are as shown in Figure 1 and the research methods adopted in this research mainly include literature review, questionnaire and experiment method.

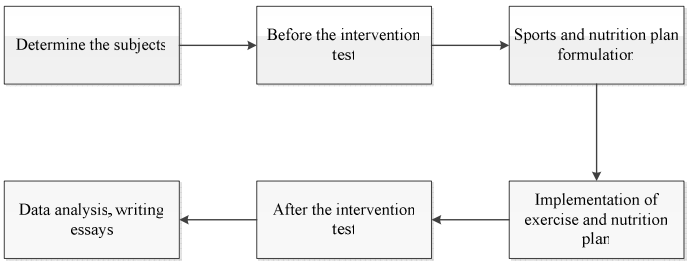


**Figure 1:** The research steps and technical route

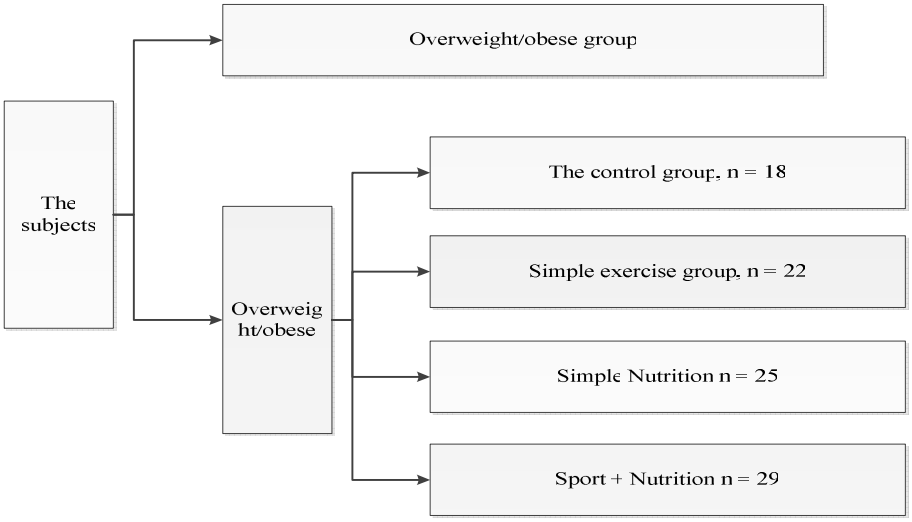


**Figure 2.** The experimental group

### 3.3. Experimental grouping

There are totally five groups in this research, which include non-overweight/obese control group, overweight/obese control group, sports group, nutrition group and sports + nutrition group, see details in Figure 2.

**(1) Non-overweight/obese control group**: only participate in the test and health education seminars before research; educate them the means and methods of preventing obesity and improving physical quality (Deibert et al. 2007).

**(2) Overweight/obese control group**: the subjects participate in all tests before and after research voluntarily, but are reluctant to participate in any interventions. But before the research, they also participate in health education seminars, which focus on educating them the harms of obesity, common methods for weight loss and fat loss and suggestions for weight loss and fat loss.

**(3) Sports group**: subjects participate in sports intervention program focusing on aerobic exercise within ten weeks; they only participate in health education seminars before intervention and are not offered with nutritional guidance and recipes.

**(4) Nutrition group**: implement nutritional intervention of ten weeks based on nutrition intervention program, participate in health education seminars before intervention and do not participate in the exercise of the research organization.

**(5) Sports + nutrition group**: offer nutrition intervention program, at the same time, participate in ten weeks' exercise class organized by this research, make health education before intervention.

### 3.4. The preparation and implementation of sports program

The selection of modes of exercise focuses on aerobic exercise, including brisk walking, jogging, walking up and down the stairs, meanwhile, combining strength, flexibility and agility exercise and inserting with sports games, in which strength exercise adopts the method of overcoming its own weight (such as push-up and sit-up etc.) and trying to avoid static exercise. In addition, adding one aerobic exercise per week from the sixth week to increase the difficulty and interest of aerobic exercise (Duncan et al. 2012).

The setting of exercise intensity takes the maximum oxygen uptake attained from exercise load test as the basis, takes decreasing the fat weight and improving cardiac and pulmonary function and physical quality of subjects as purpose and selects 50%-60%V02max as the main intensity range of ten weeks' aerobic

exercise intervention (Andrade et al. 2014). Moreover, divide the implementation of this exercise program into three stages based on the characteristics of physical quality of obese children compared with that of normal children:

First stage: the exercise intensity before two weeks of implementation of exercise program is 40%-50%VO2max;

Second stage: increase to 50%-60%VO2max after two weeks;

Third stage: insert exercise with great intensity in short time (60%-70%VO2max) in the exercise intensity of 50%-60%VO2max after six weeks.

Use the heart rate corresponding to the above intensity to monitor the intensity during exercise; the exercise intensity and heart rate corresponding to this intensity in three stages of sports group and sports + nutrition group are as shown in Table 3; there is no obvious difference in the item values of these two groups.

**Table 3.** Ten patients experienced septic complications and sport + energy and fewer ten patients experienced septic complications recipes key nutrients

| | Sports group | Sports+nutrient group |
|---|---|---|
| The total energy | 1982.36 | 2015.23 |
| protein | 82.36 | 85.42 |
| fat | 60.23 | 61.45 |
| carbohydrates | 274.56 | 281.69 |
| The total energy | 254.26 | 16.32 |
| protein | 17.26 | 27.23 |
| fat | 27.85 | 55.26 |
| carbohydrates | 55.62 | 55.21 |

### 3.5. Content of nutrition program

Nutrition intervention program mainly includes nutrition recipe and nutrition guidance. Calculate the daily energy requirement of normal children based on the recommended value (moderate physical activity) of energy of Chinese children and adolescents RNI (recommended intake standard). Based on the differences between overweight and obesity, offer 80% and 70% of the daily total energy of children with ideal weight respectively and for ideal weight, please refer to "standard height and weight for normal male and female"(Hansen et al. 2014). Make daily diet recipes for simple nutrition group and exercise + nutrition group in this research. In the total daily energy, the protein accounts for 10%-20%, fat accounts for 25%-30% and sugar accounts for 50%-60%. The details of daily total energy and main nutrient conditions of nutrition group and exercise + nutrition group are as shown in Table 4. There is no obvious difference in each indicator between these two groups.

Make nutrition education to subjects through "nutrition guidance". Eat based on the principles of "having more meals a day but less food at each", "fixed time and fixed amount" and "adding meals without adding amount" etc. In addition, teach the subjects and parents to exchange the food and adjusting the diet flexibly based on keeping the same total energy intake.

The above nutritional intervention program is prepared and accomplished by department of nutrition in Beijing Hospital leaded by Ministry of Health.

**Table 4.** The overweight/obese and overweight/obese compared the basic situation and body composition

| | Sports group | Sports+nutrient group |
|---|---|---|
| age | 13.62 | 13.36 |
| height | 161.52 | 164.52 |

| | | |
|---|---|---|
| weight | 50.23 | 75.45 |
| BMI | 19.54 | 26.35 |
| Fat mass | 10.23 | 44.52 |
| Lean body mass | 40.26 | 46.32 |
| Body fat percentage | 19.23 | 37.25 |

### 3.6. Research results

Before the experiment, there is no obvious difference in the age and height between non-overweight/obese group and overweight/obese group. However, the weight, BMI, fat weight, lean body weight and body fat percentage of overweight/obese group are higher than that of non-overweight/obese group and with obvious difference (P<0.01), see details in table 5.

The SBp and DBp of overweight/obese girl group is obviously higher than that of non-overweight/obese group (p<0.01, P<0.05). The vital capacity of overweight/obese girl group is obviously higher than that of non-overweight/obese group (P<0.01). The vital capacity index of both overweight/obese girl group and overweight/obese boy group is obviously lower than that of non-overweight/obese group with the same gender (P<0.01, P<0.01). The maximum oxygen uptake of overweight/obese boy group is obviously lower than that of non-overweight/obese boy group (P<0.05).

**Table 5:** Not overweight/obese and overweight/obese male and female physiology

| | The overweight/obese group Male | Overweight/obese Male | The overweight/obese group Female | Overweight/obese Female |
|---|---|---|---|---|
| Quiet pulse | 77.22 | 82.62 | 84.52 | 85.41 |
| Systolic blood pressure | 110.23 | 113.25 | 99.52 | 110.23 |
| Diastolic blood pressure | 68.25 | 69.25 | 65.48 | 75.23 |
| Lung capacity | 3215.23 | 3125.23 | 2635.23 | 1985.23 |
| Vital capacity index | 62.35 | 44.85 | 52.12 | 41.23 |
| Vo2 Max | 48.21 | 40.26 | 36.52 | 23.36 |
| Reaction time | 0.40 | 0.42 | 0.45 | 0.45 |

## 4. Conclusions

This paper has compared the results of various indicators before and after experiments of implementing three different intervention programs of simple exercise, simple nutrition and combination of both for ten weeks among overweight and obese adolescents. It has been found out that different programs have different influence on the body composition, circumference, physiological function, physical quality and partial metabolic indicators of overweight/adolescents by observing the differences in physique and metabolism between overweight/obese adolescents and non-overweight/obese adolescents aging from 12 to 16. Conclusions are as following:

(1) The cardiac and pulmonary function as well as physical quality of overweight/obese adolescents in this research is weaker than that of normal adolescents. Ten weeks of interventions of exercise, nutrition as well as combination of both can reduce the obesity degree of overweight/obese adolescents effectively. Exercise and combination of exercise and nutrition intervention can improve the cardiac

and pulmonary function of overweight/obese adolescents. Exercise combined with nutrition intervention is with the best effect in decreasing visceral fat and improving physical quality.

(2) The blood lipid, glucose, insulin and other metabolic indicators of overweight/obese adolescents in this research are within the normal range, but its regulation role for fat and glucose metabolism is obviously lower than non-overweight/obese.

(3) Ten weeks of exercise combined with nutrition has the best effect on improving blood lipid metabolism. Nutrition intervention has good effect on improving the regulation of blood glucose. Exercise and exercise combined with nutritional intervention can improve the secretion function of insulin cell of overweight/obese adolescents effectively. However, these three intervention programs do not have obvious effect on regulating the role of insulin resistance.

## 5. Acknowledgment

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# ANALYSIS OF ERYTHROCYTE FUNCTION CHANGE DURING MOTION FROM NUTRITIONAL INTERVENTION PERSPECTIVE

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**ABSTRACT**
In this paper, first set up sports anemia model through animal experiment; make research on the oxidative stress state and energy metabolism function of erythrocyte based on sports anemia model and anti-sports anemia agent; make qualitative and quantitative research on the aging erythrocyte with adoption of advanced flow cytometry and laser co focal technique; at the same time, observe the change of protein of erythrocyte membrane with membrane one-dimensional and two-dimensional electrophoresis technology; adopts advanced image analysis system to make quantitative analysis of protein of erythrocyte membrane. Make a series of erythrocyte index test for 12 sports anemia athletes and 12 normal athletes through human experiment and make anti-sports anemia agent treatment for them by one month to explore how exercise causes erythrocyte damage, the mechanism of causing sports anemia and also how to present.

## 1. Introduction

Higher incidence of anemia exists among athletes. Anemia has serious influence on exercise ability, training effect, recovery and immunity after exercise and other functional status; it becomes the cause of excessive training sometimes. The relationship between anemia and physical load as well as nutritional status has aroused extensive attention of the medical community (Lukaski, 2005). The purpose of this research is to establish animal model of sports anemia and it studies the erythrocyte membrane changes of rats which make long-term training during different periods to understand the influence of exercise training on erythrocyte, especially the changing regularity of erythrocyte membrane when sports anemia or potential sports anemia happens to the rats, which can provide sensitive monitoring index for reflecting potential anemia accurately and preventing the happening and development of sports anemia(Pan, 2007). At the same time, combine hemoglobin, parameters of iron metabolism and other indexes to evaluate sports anemia and increase the diagnosis accuracy for sports anemia and provide basis for preventing the happening and development of sports anemia. Make research on the erythrocyte membrane change of rats which has made eight weeks of training and further discuss the mechanism of sports anemia.

## 2. Materials and methods

The simplest index of anemia evaluation is hemoglobin. Usually, the hemoglobin of male is higher than that of female. The standards of diagnosing sports anemia at home and abroad are different (Neuberger et al., 2007). The hemoglobin standard for diagnosing anemia in European countries and America is female (120g/l,) male<140g. The domestic adult standard is female<105g, male<105g, for children below 14 years old, both are (120g/l. Hematocrit and blood viscosity are in close relationship with hemoglobin. Physiology

believes that the best hematocrit is at the high value part within the normal range, which is about 45%. When the hematocrit is at 45%, the value of hemoglobin equals to about 16g%. Normally, hematocrit and blood viscosity present a rising curve, while the situation changes at high hematocrit (such as globalism). So it can't be simply believed that the higher hemoglobin, the better (Lewis et al., 1988).

In 1959, the Japanese scholar Yoshimura firstly put forward "sports anemia", which did not draw the attention of sports medicine. In recent twenty years, the development of research on people with sports blood deepens people's understanding. For most of athletes, sports anemia is a kind of relative anemia. The premise of proposing sports anemia is considered from the ideal value of hemoglobin when the athletes engaged in endurance make aerobic exercise (Mao et al. 2011). Because the function of hemoglobin is to transfer oxygen, which is the main factor deciding the max oxygen uptake of athletes. Therefore, the quantity of hemoglobin affects the motion ability obviously and hemoglobin is also used to judge the functional status of athletes (Konstam et al., 1982).

**(1) Relative anemia caused by plasma dilution**

There is evidence proves that the endurance athletes have bigger plasma volume, while the endurance training 11 increases with the plasma volume. There are also researches indicate that the athletes are with bigger total hemoglobin, while the training increases with total hemoglobin. The decrease of hemoglobin concentration is caused by the disproportionate increase between plasma volume and erythrocyte or total hemoglobin. Brotherhood has compared the material of endurance runners and the control group and observed the similar results. The total hemoglobin of these athletes is higher than control group by 20%, but the hemoglobin concentration is relatively low. It can be seen that for some athletes, the increase of plasma volume caused by training is higher than the increase of total hemoglobin. The increase of plasma volume is an adaptive response of body and its result is to increase the stroke volume and max output of heart, which is helpful to transfer the oxygen to the surrounding tissues during strenuous exercise, where a situation of one reaction compensating the other reaction appears. Because the strenuous exercise can increase the maximum oxygen uptake, the positive adaptive response of cardiovascular function compensates the negative adaptive response causing relative anemia to a great extent. At the moment, the problem is that the training effect will be improved greatly if the dilution of hemoglobin can be avoided.

**(2) Exercise worsens the destruction of erythrocyte in the blood**. Exercise training of high intensity causes permeable and oxidative damage to erythrocyte membrane, decreases the deformation of erythrocyte and causes hemolytic; during anaerobic fermentation training of high intensity, the blood PH decreases due to lactic acid accumulation, which has influence on erythrocyte membrane with -3 protein; exercise affects the activity of $Na^{+}K$ eleven ATP enzyme on erythrocyte, and then changes the osmotic pressure of erythrocyte and decreases the deformability. The adverse effect of exercise training on the structure of erythrocyte membrane and deformability worsens the erythrocyte damage (Shin et al., 2015).

**(3) Sports iron deficiency and iron-deficiency anemia,** the iron deficiency of athletes may be possibly caused by three reasons: 1. Demand of iron increases during exercise, for example, the demand of iron of female increases due to the menstruation; athletes who need to reduce the fat usually eat low calorie diet, causing the inadequate intake of iron. 2. The decrease of iron absorption, under normal situation, the body will increase the iron absorption when the storage of iron is not enough, but the athletes with iron deficiency are different, who are usually with absorption barrier (Mooren et al. 2003). Ehn and others have observed that the iron absorption rate of long distance runners with iron deficiency is only 16.4%, while the control group with inadequate iron storage is 30%. The observation result of

Clement and others for female long distance runners is similar and the reason needs to be elaborated. 3. The increase of iron loss is caused by the great sweating of athletes (iron lass increases by 100%), stool excretion, appearance of hemoglobinuria, myohemoglobinuria, hematuria as well as the menstruation of female athletes etc. (Weder and Egan, 1988).

**(4) Exercise causes the change of hormone**. Exercise stress causes the increase of adrenaline, adrenaline causes spleen contraction and releases hemolytic factor, which increases the damages to erythrocyte.

## 3. Results and discussions

### 3.1. Experimental methods

This animal experiment is made based on sports anemia model of incremental load treadmill exercise which has been established successfully but not published yet. There are 65 male Westar rats, weighted 200g and provided by Institute of Medical Laboratory Animals of Chinese Academy of Medical Sciences, animal license number SCXKn-00-0006 and with animal grade of grade II. Select and divide into three groups randomly, which include control group (21 rats) and incremental load treadmill exercise group (short as sports group, 44 rats). The animal feed is nutrient pellet feed at full price, which is provided by Beijing Keao Xieli Feed Co., Ltd; environmental temperature of animal feeding is 23 ℃ with humidity 40-60%; raised in separate case and each with five rats, free diet, natural lighting. The nutrient components include ginseng, herb distances, yinyanglei, radix astragal, meddler, home iron, lycoypene and compound vitamin etc; the supplement time and method is gavages after training until the accomplishment of the experiment. The grouping of experimental animals is as shown in Table 1.

The rats of training group make incremental treadmill training (BCPT-96 model) by five weeks with treadmill degree at zero degree, speed at 30m/m, training for 6 days per week and for the first two weeks, training one time per day and in the following weeks, training one time in the morning and in the evening respectively and rest on Sunday (Sharif et al., 2010); The training arrangement is as following: the training time for the first time is one minute, followed by increase of 2 minutes/time and the last training time is 97 minutes (Hassel et al., 2013). If serious exhaustion symptom happens to rats during training, such as continuous mechanical stimulation make the rats can't run any more or their abdomens touch the ground seriously after get off the treadmill presenting "turtle type", then they are allowed to rest for 2 to 5 minutes. Select 22 rats randomly after five weeks to make nutritional intervention with anti-sports anemia complex and kill them in the 8$^{th}$ week, seen in Figure 1.

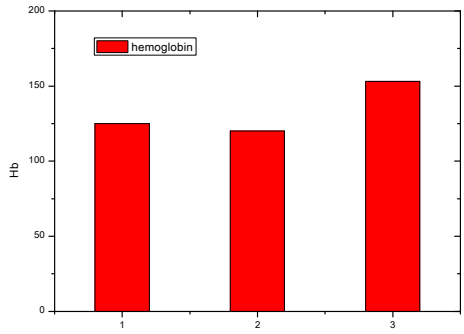


**Figure 1.** Running, swimming movement of hemoglobin

Select four rats randomly from control group, sports group and sports + nutrition group to make scanning electron microscope observation (Da et al., 2014). Erythrocyte classification refers to RBC form classification method reported by Keji Lias well as classification method introduced in clinical hematology written by Jading Deng; observe abnormal RBC form and each observed sample should not be less than 1000 RBC and calculate abnormal rate of erythrocyte (Kanda et al., 2015). The specific methods for treatment of erythrocyte by scanning electron microscope are in Table 2.

(1) Put 10Oul (about one drop) whole blood into pre-mixed SOOul normal saline at room temperature and EP tube of SOul biotin,

supplement to 1.5ml with normal saline at 37℃ complex; the time is 20 minutes(Mcmorrow et al. 2012);

(2) Low speed centrifugation (less than 150OrPm), add 0.25% glutaraldehyde solution to fix after removing supernatant, room temperature, coincidence minutes, as is shown in Table 3;

(3) Make centrifugal separation for erythrocyte and use 30k glutaraldehyde for the 2nd fixation, room temperature, and coincidence minutes;

(4) Centrifugation, water bath;

(5) Overlay;

(6) 50%, 70%, 90%, 100% gradient ethanol dehydration;

(7) Critical point drying;

(8) Spraying;

(9) Observe erythrocyte form under JEOLJSM-5600Lv scanning electron microscope.

**Table 1.** Experimental animal group

| group | N | weight |
|---|---|---|
| The control group | 20 | 204.3 |
| Animal groups | 15 | 205.9 |

**Table 2.** Treadmill and swimming exercise influence on rat blood index and weight

| group | N | hemoglobin | Red blood cell count | Red blood cells deposited | weight |
|---|---|---|---|---|---|
| The control group | 7 | 126.3 | 9.01 | 50.23 | 362.59 |
| The treadmill group | 8 | 112.5 | 6.32 | 44.26 | 345.25 |
| The swimming team | 9 | 153.6 | 9.25 | 44.85 | 353.68 |

**Table 3.** Exercise and nutrition intervention effects on erythrocyte antioxidant enzyme activity

| group | SOD | GSH-PX | CAT | Ery-SOD | Ery-GSH-PX | Ery-CAT |
|---|---|---|---|---|---|---|
| 1 | 985.32 | 123.65 | 32.65 | 906.23 | 7.06 | 0.52 |
| 2 | 858.23 | 69.85 | 21.58 | 785.21 | 4.58 | 0.26 |
| 3 | 904.68 | 85.45 | 33.69 | 779.36 | 5.62 | 0.48 |

### 3.2. Data statistical method

Make data analysis with SPSS10.0 statistical software. Each index is expressed with mean and standard deviation. Make variance analysis and correlation analysis of each index. P<0.05 with significance.

It can be seen from Figure 2 that the sports change the antioxidant enzyme activity of plasma and erythrocyte. (1) Compared with control group, the plasma SOD and erythrocyte SOD decrease obviously, decreasing from 951.48±20.33 and 903.97±234.42 to 898.45 ± 80.98 and 728.54 ± 129.95, decreased by 5.62% and 19.44% respectively, with obvious significance (P<0.05); the plasma SOD and erythrocyte SOD of sports + nutritional intervention group increase, increased by 0.83% and 6.96% respectively with obvious trend, but there is no significant difference; however, the plasma SOD and erythrocyte SOD of sports + nutritional intervention group decrease when compared with that of control group, but there is no significant difference (P(0.05). (as shown in Figure 2 and 3)

(2) The changing trend of GSH-PX is in line with SOD. Compared with control group, the

plasma GSH-PX and erythrocyte GSH-PX of sports group decrease obviously, decreasing from 123.04±71.80 and 7.09±3.51 to 63.87±56.87 and 4.82±2.03 respectively, decreased by 48.9% and 31.98% respectively, the difference is with obvious significance (P<0.05); the plasma GSH-PX and erythrocyte GSH-PX of sports + nutritional intervention group increase, increased by 33.28% and 16.23%, the trend is obvious but without obvious significance. However, the plasma GSH-PX and erythrocyte GSH-PX of sports + nutritional intervention group present decreasing trend compared with control group (as shown in Figure 4 and 5).

(3) Compared with control group, the plasma CAT and erythrocyte CAT of sports group decrease obviously, decreasing from 36.26±13.13 and 0.52±0.14 to 21.52±10.98 and 0.29±0.045, decreased by 40.73% and 44.16% respectively, the difference is with high degree of significance (P<0.01); the plasma CAT and erythrocyte CAT of sports + nutritional intervention group increase obviously then sports group, increasing from 21.52±10.98 and 0.29±0.045 to 33.13±13.24 and 0.41±0.17, increased by 34.97% and 41.44% respectively, the difference is with significance ((P<0.05)). However, the plasma CAT and erythrocyte CAT of sports + nutritional intervention group decrease compared with control group, but with no obvious significance(as shown in Figures 6 and 7).

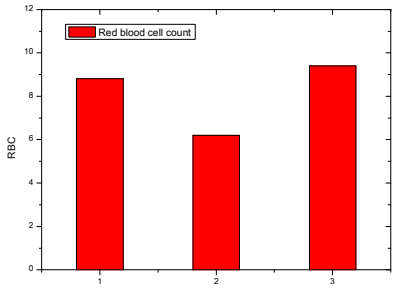


**Figure 2.** Running, swimming exercise effect on the number of red blood cells

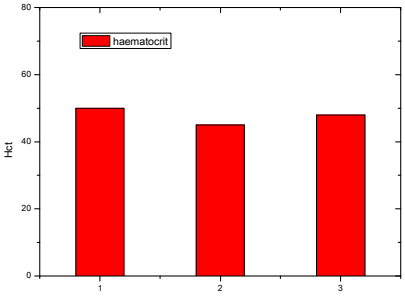


**Figure 3.** Run, the influence of the backlog of swimming exercise on blood cells

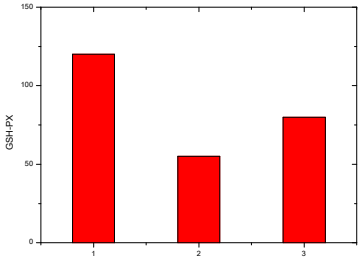


**Figure 4** Sports and anemia agent affect plasma GSH – PX

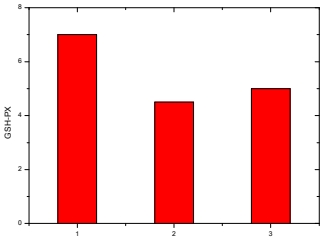


**Figure 5.** Sports and anemia agent effects on erythrocyte GSH – PX

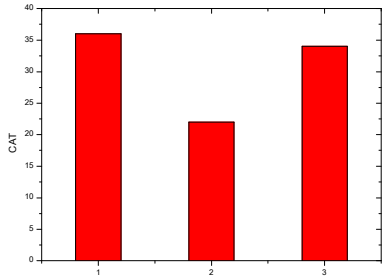


**Figure 6.** Sports and anemia agent affect plasma CAT

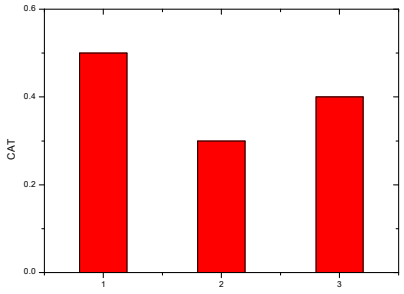


**Figure 7.** Sports and anemia agent effect on red blood cells to the CAT

## 4. Conclusions

In this experimental research, first set up sports anemia model through animal experiment; make research on the oxidative stress state and energy metabolism function of erythrocyte based on sports anemia model and anti-sports anemia agent; make qualitative and quantitative research on the aging erythrocyte with adoption of advanced flow cytometry and laser co focal technique; at the same time, observe the change of protein of erythrocyte membrane with membrane one-dimensional and two-dimensional electrophoresis technology; adopts advanced image analysis system to make quantitative analysis of protein of erythrocyte membrane. Make a series of erythrocyte index test for 12 sports anemia athletes and 12 normal athletes through human experiment and make anti-sports anemia agent treatment for them by one month to explore how exercise causes erythrocyte damage, the mechanism of causing sports anemia and also how to present. The following conclusions have been reached:

(1) This research results show that for the three standard indexes Hb for evaluating anemia in the 10 weeks' exhaustion of heavy load, treadmill sports group and control group presents very obvious statistical significance ($P<0.01$), for RBC and/or Hct in the 10 weeks' exhaustion of heavy load, treadmill sports group and control group do not have obvious statistical significance. Therefore, this sports anemia model is good. In addition, the author finds out that this model consumes too much time during experiment and the utilization of treadmill is no economical, therefore, it is suggested to adopt sports anemia model of incremental load treadmill.

(2) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that the erythrocyte abnormal rate of rats in sports group is higher than that of control group through observations with different times of electron microscope; moreover, each abnormality of sports group is higher than that of control group.

(3) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that erythrocyte free radical and lipid peroxide products increase, oxidation resistance ability decreases, presented as the decreased ability of antioxidant enzyme system and non-enzyme system, which explains the serious imbalance between erythrocyte oxidation and anti-oxidation.

(4) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that both erythrocyte $Na^{+}$-K-ATP enzyme activity and $Ca^{2+}Mg^{2+}$-ATP enzyme activity decrease, and ion imbalance appeared in erythrocyte, which has affect the permeability of erythrocyte membrane.

(5) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that both the erythrocyte glycolytic potential and phosphoric acid bypass metabolism of sports anemia athletes decrease, ATP and NADPH generations decrease, affecting body energy metabolism and GSH-PX activity; two energy metabolisms of erythrocyte glycolysis and pentose phosphate shunt decrease, causing the damage to erythrocyte.

(6) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that exercise has speeded up the aging process of erythrocyte; the SA of sports anemia group decreases obviously than that of control group; PS extrusion improves obviously than that of control group. The increase of erythrocyte aging after exercise is related to the increase of oxidative stress level of erythrocyte as well as the accumulation of calcium ion in the cell.

(7) Based on sports anemia model caused by long time incremental exercise, the sports anemia athletes experiment proves that when free radical generation increases and superoxide dismutase SOD, catalane CAT, glutathione peroxides GSH-Px etc. decrease, oxygen free radical damages many biomacromolecules, such as nucleon and protein membrane polyunsaturated acid, causes super oxidation reaction, decreases the content of actins and protein with -6 and brings about damages to skeleton structure and function of erythrocyte membrane. The observation result of erythrocyte membrane protein change with two-dimensional electrophoresis techniques reveals the change of erythrocyte membrane protein with -3. Statistics can't be made for lot of samples in this experiment, but the advantages can't be neglected.

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# RESEARCH AND PROSPECT ON THE DEVELOPMENT OF SPORTS NUTRITION FOOD INDUSTRY IN CHINA

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**ABSTRACT**
This paper has made deep analysis of various factors affecting the development of sports nutrition food industry after summarizing the development history of our sports nutrition food industry systematically (Rostami et al., 2000), proposed strategic thinking which promotes the development of our sports nutrition food industry on this basis and aimed to provide valuable suggestions for the development of related departments and enterprises.

## 1. Introduction

Our sports nutrition food industry originated from the demand of serving for the improvement of competitive sports performance and the consumer group at that time was mainly professional athletes. Therefore in a period of time, sports nutrition food market was regarded as “niche market” however, with the change of situation, our sports nutrition food industry needs to change from “niche market” to “mass market” gradually, which requires systematic research on the market demand and supply, market positioning, market structure and enterprise behavior etc. of the whole industry to adapt to the change of market demand, this is also the realistic significance of this research (Senchina et al., 2011).

The writer has found out when checking the literature both at home and abroad that in the foreign research on sports nutrition food industry, most of literatures study the microscopic behavior from the perspective of management and market(Goodman et al. 2011). The development of sports nutrition food industry abroad is relatively mature, which enables the researchers to analyze the consumer composition, technology development process, marketing program, government supervision, industry association effect and quality system etc. of sports nutrition food industry with comprehensive adoption of investigation, comparison, cases and other research methods(Yin, 2015). The research has adopted various theoretical tools, such as industrial spatial agglomeration theory and value chain analysis etc, which is of great reference significance and value. Most of domestic researches on sports nutrition food industry are focusing on the definition and classification of sports nutrition food, the development history of our sports nutrition industry and the size and development trend of foreign sports nutrition food market etc, while from an angle combining macro analysis and micro analysis together, systematic analysis of the market demand and supply, market structure, enterprise behavior and industry competitiveness of our sports nutrition food industry has not been found and the comparative analysis with foreign sports nutrition food industry is almost blank (Yang,

2011). In this sense, the research in this paper has more important theoretical significance.

This paper has made deep analysis of various factors affecting the development of sports nutrition food industry after summarizing the development history of our sports nutrition food industry systematically, proposed strategic thinking which promotes the development of our sports nutrition food industry on this basis and aimed to provide valuable suggestions for the development of related departments and enterprises.

## 2. Materials and methods

### 2.1. Market demand of our sports nutrition food industry

Market demand is the basis of enterprise survival and development. The development of sports nutrition food market can't be separated from market demand either, should be based on market demand and decided by the consumption structure formed by the demand market as well as the consumption amount brought about by this in the end (Maughan et al., 2004). The demand of sports nutrition food refers to the demand amount of the public for sports nutrition food. Sports nutrition food demand takes the purchase desire as premise and is restricted by the payment ability (Antonio et al., 2001). The demand of sports nutrition food can be divided into potential demand and effective demand. Potential demand is people's demand for sports nutrition food objectively, which includes the demand with purchase desire but restricted by payment ability and other factors temporarily as well as the demand without purchase desire temporarily (Devlin and Belski, 2014); the effective demand of sports nutrition food refers to the demand with purchase desire, payment ability and can be transferred to practical purchase behavior at current stage. The potential demand of sports nutrition food is the premise of effective demand, but the potential demand needs to be transferred into effective demand and finally cause sports nutrition consumption, which is the basic demand for the development of sports nutrition food industry (Bingham et al., 2015).

### 2.2. Economic factors affecting the market demand of our sports nutrition food industry

**(1) Economic development and income level**

Based on principles of economics, the increase of consumption demand and income increase present a positive correlation. Generally speaking, the lower income level of residents, the higher marginal propensity to consume (Liang, 2015); the higher income level of residents, the lower marginal propensity to consumer. In the aspect of social-economic structure, people with high income and low income should be few and the middle class accounts for the most, occupying the mainstream status. The distribution of overall population presents "olive" structure, which is good for the increase of consumption demand. Our current population distribution presents bell structure, where middle class is few (Liang, 2015); people with high income and low income are more. However, with the increasing income gap, most of the wealth concentrates on few people of high income, but the people with high income have low average propensity for sports nutrition food and the purchasing power is relatively surplus (Du, 2015); a lot of middle and low income people with potential consumption demand have high average propensity for sports nutrition food, but the purchasing power is seriously inadequate, the non-coordination between income level and average propensity to consume makes it difficult to transfer the potential demand for sports nutrition food to effective demand with payment ability, which has caused the decrease of average propensity to consume for sports nutrition food and then caused the limited demand amount and inadequate effective demand.

**(2) Consumer preference**

The preference of consumers for sports nutrition food is also an important factor affecting the market demand for sports nutrition food and this preference is related to the cognition of physical exercise group for the function of sports nutrition food (Chen, 2015). Among sports group in China, most of people believe that they can exercise well without special sports nutrition food, while it is not the

same in developed countries; taking America as example, the sports group with adoption of sports nutrition food can be divided into four categories, which include professional athletes, amateur sports enthusiasts, bodybuilder enthusiasts and bodybuilders; the consumption proportions of sports nutrition food for these four categories are 23%, 19%, 35% and 23% respectively(Campbell et al. 2013). The sum of proportions of amateur sports enthusiasts and bodybuilder enthusiasts is 54%, exceeding half of the total consumption, which proves that in America, both professional athletes and amateur enthusiasts realize the functions and advantages of sports nutrition food and then consume sports nutrition food in a long run.

**(3) Prices of sports nutrition products and prices of related products** (Potgieter, 2013).

In economics, related products mainly refer to complementary and substitutes. Complementary refers to that the price increase (or decrease) of one product will cause the demand decrease (or increase) of the other product; substitute refers to that the price increase (or decrease) will cause the demand increase (or decrease) of the other product. In view of sports nutrition products, related products mainly refer to substitutes, including general health food and traditional food etc.

## 3. Results and discussions

### 3.1. The market supply of sports nutrition food industry in China

In 2010, the sales amount of sport nutrition food in China has reached 222 million Yuan (as shown in Table 1).

From 2005 to 2010, the sales of sports nutrition food in China has increased by 98% in total nearly doubled. In 2010, it has increased by 17.6% (as shown in table 2). In view of the sales channels of health products from 2005 to 2010, the sales based on stores account for the most, but its proportion presents a downtrend(as shown in table 3); in 2010, the proportion is 68.4%; the sales without stores present an increasing trend, in 2010, the proportion is 31.6%, in which the direct sales is 30.6%, accounting for 96.8% of non-store sales channels. Other non-food retailers (gyms and slimming club etc.) are also main sales channels for sports nutrition products, which account for 90% of the sales amount. The market share of e-commerce is increasing due to its convenience as well as the promotion support of leading companies.

**Table 1.** Sports nutrition food sales of 2005-2005

| Time | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 |
|---|---|---|---|---|---|---|
| sales | 112.3 | 117.2 | 154.3 | 162.3 | 198.5 | 212.6 |

**Table 2.** Sports nutrition food sales growth rate of 2005-2010

| Time | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 |
|---|---|---|---|---|---|---|
| sales | 112.3 | 117.2 | 154.3 | 162.3 | 198.5 | 212.6 |

**Table 3.** 2005-2010 analysis of consumer health products sales channels

| Time | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 |
|---|---|---|---|---|---|---|
| Based on the store's sales | 67.2 | 74.2 | 70.2 | 70.6 | 68.2 | 68.5 |
| Discount grocery retailers | 7.8 | 8.5 | 8.6 | 9.5 | 7.5 | 7.1 |
| Health food store | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| A large supermarket | 0.5 | 0.5 | 0.4 | 0.6 | 0.5 | 0.4 |
| Small grocery store retailers | 4.5 | 5.2 | 5.6 | 4.8 | 4.6 | 4.4 |
| The super market | 0.5 | 0.4 | 0.6 | 0.5 | 0.5 | 0.4 |
| Other grocery retailers | 2.4 | 2.3 | 2.5 | 2.6 | 3.2 | 3.0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| Non-store sales | 56.3 | 52.1 | 50.9 | 58.4 | 49.6 | 54.2 |
| Beauty of health | 58.9 | 59.4 | 55.1 | 54.9 | 59.6 | 60.2 |
| A pharmacy/drugstore | 31.3 | 34.9 | 35.3 | 34.7 | 31.2 | 34.9 |
| The grocery store | 9.5 | 8.6 | 8.9 | 7.6 | 8.0 | 8.2 |
| Other medical | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| Specialist retailers | 0.1 | 0.2 | 0.2 | 0.1 | 0.3 | 0.2 |
| Star market | 32.5 | 28.6 | 27.1 | 26.9 | 29.5 | 25.6 |
| No other grocery store sales | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| Vending machines | 1.4 | 1.9 | 1.8 | 1.6 | 1.5 | 1.4 |
| The network shopping | 0.2 | 0.2 | 0.4 | 0.1 | 0.3 | 0.6 |
| Direct selling | 31.6 | 25.7 | 27.9 | 28.6 | 24.3 | 30.0 |
| Total | 100 | 100 | 100 | 100 | 100 | 100 |

### 3.2. Research on sports nutrition food in Europe and America

First, analyzing from the supply side of sports nutrition food, the increasing point of the whole sports nutrition food market focuses on supplementing moisture quickly, including instant drink type sports drink, water drink with high concentration nutrient supplement, sports drink with fast supplementary energy, past diet drink and rod products which can supplement energy quickly after sports.

All sports nutrition foods can be divided into two categories based on the directions of its influence on health, the first category is to promote health with increase, such as improving or increasing products with body materials, vitamins, dietary fiber, antioxidants, amino acids, creatine and protein etc or increase body function, such as improving the products of energy level, body water level and promoting recovery etc; the other category is to promote healthy products by decreasing some substances of the body, such as products decreasing body sodium, fat, sugar, trans fats and synthetic pigment etc.

Based on the research report of BCC Investigation Company, in global sports nutrition food market, the America accounts for 49% and the Europe (Russia is excluded) accounts for 26%, while in European market (Russia is excluded), Germany, Britain and Italy account for 60%. The growth situation of sports nutrition food is as shown in Figure 1.

In the field of sports and health nutrition, sports drinks market accounts for the most and develops rapidly.

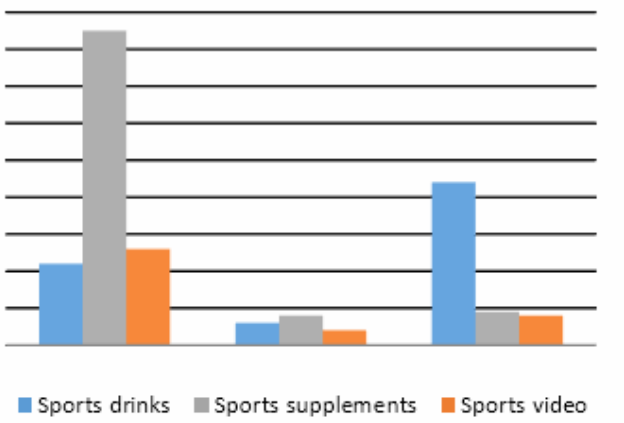


**Figure 1.** Nutrition proportion

In 2007, the worldwide income is 24.9 billion US dollars, which is expected to reach 87 billion US dollars in 2013 with annual growth rate of 25.6%. In 2007, the global value of sports food is 1.2 billion US dollars, which achieved 1.5 billion US dollars in 2013 and will achieve 2.5 billion US dollars in 2013, increasing at a speed of 10% every year.

In view of the demand terminal of sports nutrition food, consumers are paying more and more attention to healthy and convenient way of life, while sports nutrition food offers perfect selection to meet the demands of this aspect. Consumers of sports nutrition food, sports drinks

and sports supplements can be divided into following four different groups: bodybuilders, professional or amateur athletes, leisure athletes and consumers pursuing for way of life. Bodybuilders pursue body-building exercise, which achieves the purpose of muscle growth through combining the weight training and increasing the caloric intake together. Athletes include all professional and amateur athletes (bodybuilders are excluded). Leisure athletes represent non-professional athletes, who can't reach the same level as athletes and bodybuilders in physical exertion. This group of people generally takes exercise as recreation of weekend or focuses on maintaining the body. Consumers pursuing way of life use sports nutrition food but not take exercise as the purpose. The consumed sports nutrition food by this group is mainly used as refreshing drink and replaces daily meal or consumes a healthy snack occasionally. This group of consumers can improve the energy level in short time with adoption of sports nutrition food. This group of consumers seeking for way of life is consumers for sports nutrition food with the fastest growth speed and has become an important component of the market. These consumers wish to experience the enjoyment of products brought about to their health.

### 3.3. Strategic thinking of the development of sports nutrition food industry in China

There are various ways of satisfying people's demand for health, which include public medical products and services provided by the government, public sports facilities and services provided by the government as well as the health care products and sports products and services provided by market mechanism. In view of the voice for changing the governmental functions at the moment, government departments in the future make great efforts in providing public sports products and service to ensure the basic physical training needs of the public and then make more and more people participate in the exercise. Besides the basic needs of the public, there are also a lot of diversified and personalized sports demands caused by different income levels. The sports nutrition food industry of China originated from competitive sports and has formed subdivided and specialized market positioning mode gradually, which is to say at the moment, the overall positioning of the market still focuses on niche market. With the development of economy, it is possible for the industry positioning changing from niche market to mass market. Especially for middle aged and old people who do exercise frequently, they have a very strong awareness of health and often participate in the exercise. If complemented with sports nutrition food of protein type, it will increase their muscle strength and will avoid many chronic diseases such as multiple diabetes mellitus in middle and old age. For adolescents, they are facing the pressure of growth and learning and it will be very helpful for their growth and leaning effect if they can participate in the exercise actively and use sports nutrition foods at the same time. However, the sports nutrition food industry can make great achievements only by making strategic adjustment and transferring the target market positioning from professional athletes to mass consumers. In other words, the whole industry can only grow up by changing the sports nutrition food to mass consumer goods.

## 4. Conclusions

(1) In view of the market demand and supply of our sports nutrition food industry, first, the market demand of sports nutrition food industry can be divided into potential demand and effective demand. Changing from potential demand to effective demand is affected by economic factors and non-economic factors. The economic factors mainly include economic development and income level of residents, change of residents consumption structure, consumption concept and awareness, consumer preference, prices of related products, industrial structure and sports industry structure etc. Non-economic factors mainly include different consumer groups, number of sports population, knowledge, standards and safety of sports nutrition food, government policy and public

opinion etc. In general, the effective demand of our sports nutrition food is inadequate at the moment. The main reasons are that the consumption concept and awareness of residents does not change, the number of people participated in exercise does not increase obviously; the functional understanding of sports nutrition food is limited; the food safety supervision is not standardized. Second, main factors affecting the market supply of our sports nutrition food industry include cost and price, marketing channel and marketing method and investment of research and development. The subjects of market supply of our sports nutrition food industry are less and the size is relatively small; compared with developed countries, the category, quantity and quality of sports nutrition food need to be improved.

(2) In view of the market structure of our sports nutrition food industry, first, the market concentration degree of our sports nutrition food industry is high. In the sports nutrition food market of professional athletes, CPT, Vita, MET-Rx, EAS and other brands account for over 80% of market share. Second, certain entry barrier exists in our sports nutrition food industry. As technology-intensive industry, capital barrier, technology barrier and policies and regulations barrier exist. Third, product homogeneity is serious. There is small overall differentiation in our sports nutrition food and the homogeneity is very serious, which is mainly presented as functional repetition and prescription repetition. Forth, the above analysis has shown that the market structure of our sports nutrition food industry belongs to market structure of oligopoly. The future development tends to be a market structure of monopolistic competition, which should increase competition subjects, expand industry size and strengthen technological innovation.

(3) The related experience of Europe and America has important reference significance for the development of our sports nutrition food industry. First, adjust product structure and marketing method continuously based on the consumer demand and market change. Second, it needs to put emphasis on the improvement of the core competitiveness of research and development ability. Third, the government should improve the laws and regulation system continuously for the supervision of sports nutrition food, establish comprehensive and strict industry standards and national standards.

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# THE ANALYSIS OF HIGH-LEVEL ATHLETES' NUTRITION AND DIET

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*Food logistics.*

**ABSTRACT**
This paper compared research works of sports nutrition to other countries, Comparing the intakes of vitamins by female wrestlers and boxers in a particular province and the recommended amounts of vitamins for Chinese athletes, the results show that the average intakes of Vitamin A, PP and E by athletes are obviously higher than the correspondingly recommended amounts for Chinese athletes. The intake of Vitamin B2 happens to fall into the recommended range. Although the intake of Vitamin B1 hasn't reached the recommended standard, the gap is small. The intake of Vitamin C is simply less than the recommended standard for Chinese athletes.

## 1. Introduction

Nutrition refers to the whole process in which human organism ingests, digests, absorbs and utilizes the nutrients in the food so as to maintain the normal physiological, biochemical and immunologic function and life activities including the growth development and metabolism. Nutrients refer to beneficial substances the diet contains (Maughan, 2002). These substances can maintain life and promote human organism's growth development and health. For example, seven nutrients required in the human body are protein, fat, carbohydrates, vitamin, mineral element, dietary fiber and water. Neurology is the science of researching human organism's nutrition law and improvement measures, which is to study food components which are beneficial to the human body and the law and mechanism of the human body's ingesting and utilizing these components to maintain and improve health. On this basis, people can take various measures to enhance the human health and improve life quality (Jamaluddin et al. 2014). Nutrition and diet are both closely related to people's daily life. People must ingest a certain amount of food to meet their body's nutritional requirements and maintain their own activities and health.

Sports nutrition refers to an academic discipline which studies how athletes enhance their body functions' athletic ability, eliminate fatigue and regain their strength in a short time through ingesting, absorbing and utilizing various nutrients. At the same time, it also helps athletes prevent several diseases to improve sport achievements (Panza et al., 2007). This discipline mainly conducts reasonable collocation based on athletes' training conditions at different periods and dietary nutrition during the competition and provides sports nutritional supplements as the auxiliary supply. Its main purpose is to provide athletes with required energy and various nutrients for adapting to the intensive training and reasonable diet collocation to improve training effects (Grahampaulson et al., 2015).

Our country has started the research of sports nutrition since the late 1950s. The Sports Nutrition Research Center was established in National Research Institute of Sports Medicine which was founded in 1987. This research center is used for studying athletes' nutritional problems.

Compared to research works of sports nutrition of other countries, those of our country are started relatively late. However, we have also made impressive achievements in this field. For example, the distinguished sports nutritionist Professor Jidi Chen is the father of China's sports nutrition and is the first person to establish the sports nutrition biochemical laboratory in China. She has dedicated his life to researching in the field of sports nutrition. Besides, her research achievements have won the First Award of State Physical Culture and Sports Commission's Scientific and Technological Progress for several times. It is observed that the development prospect of sports nutrition is quite bright (Lloyd et al., 1987).

In recent years, as the key component of athletes' nutrition, the diet has been an emphasis of sports nutrition researches. Among them, reasonable dietary and nutritional requirements of athletes in various sports have become a research focus in recent years (Potgieter, 2013). For example, The Survey and Countermeasures of High-level Throwing Athletes' Meal and Nutrition Supplementation, Analysis of Dietary Nutrition, Body Composition and Blood Biochemical Indexes of Female Wrestling Athletes during Weight Control Period and The Comprehensive Analysis of Dietary Status of Male Athletes in Heavy Athletics Team of Hubei Province (Mountjoy et al., 2014).

Nowadays, the level of sports competition in China has been rapidly improved and Chinese athletes have repeatedly accomplished splendid works in various competitions around the world. China is now marching from a sports power into a sports giant. This can not be separated with the scientific guidance of sports nutritional knowledge and the proper use of methods and means. Along with the continuous social improvement and the booming development of sports undertakings, sports nutrition will surely accomplish more stupendous achievements in the future.

## 2. Materials and methods

### (1) Research object

Taking 13 female wrestlers and 14 women boxing athletes of one province as examples. Among 13 female wrestlers, there are 1 National Master Athlete, 1 National First-Level Athlete and 11 National Second-Level Athlete. Among 14 women boxing athletes, there is 1 International Master Athlete, 5 National Master Athletes, 4 National First-Level Athletes and 4 newly entered athletes whose levels have not been recognized yet. The basic information of research objects is as follows.

### (2) Investigation methods

A survey is conducted by issuing questionnaire of nutritious diet to investigate athletes' daily diets so that their dietary habits and nutritional knowledge can be comprehensively understood and researched. This research uses 13 female wrestlers and 14 women boxing athletes of Nei Monggol Autonomous Region as the research object and issues 81 return visit questionnaires of 24-hour diet. These questionnaires are issued at three different times in 3 days and 27 questionnaires are issued each time in one day. Among them, there are 81 valid questionnaires and the recovery rate of those valid questionnaires is 100 % (Table 1).

### (3) Weighed dietary record method

Researchers use libra to weigh the raw weight and cooked weight of the food that respondents eat every meal each day respectively. At the same time, they should also pay attention to weighing the remaining food to obtain more accurate intake results of the food that respondents eat every meal each day(Dueck et al. 1996). The weighed dietary record method can be conducted in 3, 4, 5, 7 or 20 days continuously. The weighed dietary record method of 3-4 days is the most widely and frequently used and takes the top spot. This method is accurate and reliable, yet its human cost and time cost are relatively high and is not suitable for the large-scale survey (Howe et al., 2014). However, it is still fit for the small-scale population that possesses special nutritional

requirements, for example, the aged, children and athletes.

Varieties and quantities of the food supplied by the canteen every meal per day and the food ingested by athletes should both be precisely weighed and recorded in great details (Beck et al., 2014). Then the software of Athletes and Public Diet Application Analysis and Management System should be applied to conduct the data analysis of various nutrients on acquired data.

**(4) Data processing method**

Athletes' intake of various food recorded in the related questionnaire survey should be entered into the software of Athletes and Public Diet Application Analysis and Management System which is researched and developed by General Administration of Sport of China to calculate the data of various nutrients. SPSS 17.0 software should be used to conduct the data processing and analysis and all data should be expressed as average ± standard deviation. In the end, a pared-samples T test should be conducted among statistical results, Chinese Resident Dietary Nutrition Recommended Standard and Chinese Athletes Dietary Nutrition Recommended Standard. If $P \leq 0.05$, then the significant difference exists; if $P \leq 0.01$, then non-significant difference exists (Gao and Zhang, 2015).

**Table 1.** Athletes the material average energy intake

| Player number | Energy | Protein | Fat | Sugar |
|---|---|---|---|---|
| 1 | 3620 | 120 | 163 | 357 |
| 2 | 2625 | 106 | 235 | 623 |
| 3 | 4144 | 389 | 223 | 365 |
| 4 | 3452 | 236 | 2361 | 605 |
| 5 | 3625 | 402 | 524 | 702 |
| 6 | 4421 | 409 | 412 | 554 |
| 7 | 3620 | 410 | 925 | 754 |
| 8 | 4521 | 312 | 625 | 565 |
| 9 | 4120 | 265 | 666 | 236 |
| 10 | 4265 | 366 | 485 | 648 |
| 11 | 4258 | 331 | 652 | 569 |
| 12 | 4520 | 420 | 645 | 668 |
| 13 | 2323 | 452 | 611 | 669 |
| 14 | 3525 | 306 | 459 | 545 |
| 15 | 3958 | 311 | 605 | 658 |
| 16 | 4712 | 308 | 695 | 458 |
| 17 | 4256 | 369 | 459 | 625 |
| 18 | 2565 | 296 | 552 | 474 |
| 19 | 3362 | 452 | 454 | 625 |
| 20 | 4420 | 268 | 628 | 565 |
| 21 | 3362 | 369 | 564 | 352 |
| 22 | 3636 | 452 | 268 | 645 |
| 23 | 4850 | 245 | 525 | 525 |
| 24 | 4421 | 333 | 703 | 664 |
| 25 | 4456 | 358 | 625 | 756 |
| 26 | 2658 | 542 | 459 | 541 |
| 27 | 4012 | 333 | 563 | 762 |
| Average | 3621.32 | 320.32 | 575.25 | 602.42 |
| The standard | 574.26 | 101.25 | 254.23 | 135.21 |

| | | | | |
|---|---|---|---|---|
| deviation | | | | |
| Recommended value | 4472 | 160 | 115 | 655 |

**Table 2.** Athletes average vitamin intake

| Player number | Vitamin A | Vitamin B1 | Vitamin B2 | Vitamin PP | Vitamin C | Vitamin E |
|---|---|---|---|---|---|---|
| 1 | 751.23 | 1.6 | 1.9 | 37.2 | 86.3 | 51.3 |
| 2 | 251.32 | 1.7 | 1.9 | 32.6 | 71.3 | 35.2 |
| 3 | 752.36 | 1.5 | 2.1 | 46.9 | 95.3 | 119.6 |
| 4 | 4521.3 | 2.5 | 1.7 | 60.2 | 114.3 | 356.2 |
| 5 | 4162.3 | 2.3 | 2.5 | 64.3 | 147.5 | 312.6 |
| 6 | 2754.1 | 2.5 | 7.8 | 50.3 | 104.3 | 247.5 |
| 7 | 4210.6 | 2.3 | 3.5 | 45.3 | 88.5 | 225.6 |
| 8 | 2730.6 | 2.1 | 1.6 | 103.6 | 160.3 | 278.3 |
| 9 | 3345.3 | 3.6 | 2.0 | 78.6 | 88.2 | 256.3 |
| 10 | 4512.3 | 2.2 | 2.1 | 28.6 | 160.3 | 225.3 |
| 11 | 5212.2 | 1.8 | 1.8 | 45.6 | 129.5 | 541.6 |
| 12 | 3212.2 | 1.4 | 2.6 | 25.3 | 97.5 | 352.6 |
| 13 | 1232.3 | 2.5 | 3.0 | 78.3 | 158.2 | 298.3 |
| 14 | 4253.3 | 2.3 | 2.9 | 103.6 | 92.3 | 295.3 |
| 15 | 2563.3 | 2.6 | 3.2 | 45.6 | 152.3 | 365.3 |
| 16 | 5223.6 | 2.5 | 3.5 | 75.0 | 114.3 | 525.3 |
| 17 | 3633.2 | 2.8 | 3.3 | 28.5 | 88.5 | 264.3 |
| 18 | 3625.3 | 2.4 | 2.1 | 49.3 | 93.2 | 232.6 |
| 19 | 3021.3 | 2.2 | 2.2 | 28.6 | 175.6 | 336.8 |
| 20 | 3632.2 | 2.7 | 1.5 | 45.3 | 93.6 | 284.5 |
| 21 | 5232.6 | 3.2 | 2.2 | 55.3 | 154.9 | 452.3 |
| 22 | 4253.3 | 1.9 | 2.6 | 56.3 | 142.7 | 254.6 |
| 23 | 2315.2 | 2.0 | 3.5 | 58.6 | 185.3 | 295.6 |
| 24 | 4251.3 | 1.2 | 2.2 | 45.3 | 113.6 | 269.5 |
| 25 | 3205.2 | 2.8 | 1.5 | 47.3 | 100.8 | 365.3 |
| 26 | 2536.4 | 2.6 | 1.9 | 65.3 | 147.6 | 266.3 |
| 27 | 3625.3 | 2.6 | 2.0 | 66.8 | 165.3 | 298.5 |
| Average | 3521.4 | 2.4 | 2.3 | 57.5 | 115.3 | 83.6 |
| The standard deviation | 1136.3 | 2.0 | 1.3 | 26.3 | 30.2 | 85.6 |
| Recommended value | 1500 | 3-5 | 2-2.5 | 20 | 140 | 30 |

## 3. Results and discussions

**(1) Athletes' ingestion conditions of three major energy substances** and **energy intake**

As seen in Table 2, athletes' daily ADI of protein and fat is higher than the recommended intake standard for the most part. 3 athletes' ADI of protein is lower than the standard, which accounts for 11% of the overall number of athletes. Only 1 athlete's ADI of fat is lower than the standard and other athletes' ADIs are all around 1-9 times higher than the standard value. The intake of 13 athletes' ADI of carbohydrate reaches the recommended standard and accounts for 48% of the overall number of athletes. As for

the energy intake, only 5 athletes' ADIs are higher than the recommended standard, which accounts for 18.5% of the overall number of athletes. We can learn from Table 3 that: athletes' intake conditions of various energy substances exist different levels of inadequate intake phenomenon. Especially, the intake of energy and that of carbohydrate both commonly exists the inadequate intake phenomenon. Carbohydrate is the most easily digested and assimilated substance and plays the vital role in the elimination of athletic fatigue. Research results show that the competitive sports training of large load intensity consumes sugar and uses it as the main energy source. Besides, the inadequate carbohydrate reserve before the training is the most important factor that makes sports fatigue happen in advance and affects training effects. In consideration of the insufficient carbohydrate intake, from now on, athletes should pay attention to ingesting more food which contains high carbohydrate content in the daily diet.

**Table 3.** Athletes' ADIs of vitamin PP and vitamin E

| Player number | Potassium | Calcium | Iron | Zinc | Selenium |
|---|---|---|---|---|---|
| 1 | 2540 | 535 | 32 | 34 | 170 |
| 2 | 1720 | 452 | 25 | 20 | 100 |
| 3 | 2560 | 563 | 128 | 24 | 75 |
| 4 | 9256 | 2112 | 112 | 64 | 256 |
| 5 | 7512 | 1525 | 78 | 64 | 236 |
| 6 | 8805 | 1356 | 58 | 70 | 185 |
| 7 | 7152 | 1852 | 86 | 52 | 156 |
| 8 | 7562 | 1116 | 84 | 56 | 178 |
| 9 | 9125 | 1168 | 82 | 84 | 156 |
| 10 | 6235 | 3625 | 54 | 42 | 156 |
| 11 | 7012 | 1425 | 96 | 26 | 123 |
| 12 | 9912 | 1652 | 54 | 53 | 256 |
| 13 | 7025 | 1563 | 116 | 33 | 207 |
| 14 | 8856 | 2546 | 69 | 69 | 236 |
| 15 | 8812 | 2654 | 83 | 95 | 253 |
| 16 | 7025 | 2512 | 125 | 66 | 117 |
| 17 | 6636 | 2654 | 129 | 68 | 156 |
| 18 | 7025 | 1265 | 136 | 56 | 185 |
| 19 | 5623 | 1525 | 90 | 84 | 235 |
| 20 | 8566 | 1865 | 85 | 53 | 227 |
| 21 | 8562 | 1763 | 75 | 66 | 256 |
| 22 | 8152 | 1140 | 74 | 85 | 196 |
| 23 | 8412 | 1360 | 110 | 65 | 227 |
| 24 | 6596 | 1425 | 96 | 56 | 186 |
| 25 | 6032 | 1565 | 56 | 44 | 200 |
| 26 | 6060 | 1253 | 69 | 36 | 227 |
| 27 | 7025 | 1752 | 69 | 58 | 193.2 |
| Average | 6563 | 1632.2 | 86 | 56 | 47.5 |
| The standard deviation | 3263 | 625.3 | 28.3 | 13.6 | 48.2 |
| Recommended value | 3000-4000 | 1000-1500 | 20-25 | 20-25 | 50-150 |

**(2) Athletes' ingestion conditions of vitamin**

Table 3 shows that athletes' ADIs of vitamin PP and vitamin E are all 1-10 times higher than the recommended value. 3 athletes' ADIs of vitamin A are insufficient, which accounts for 11% of the overall number of athletes. Only 3 athletes' ADIs of vitamin B1 reach the standard, among them, 1 athlete's ADI of vitamin B1 is more than 2 times higher than the standard. 14 athletes' ADIs of vitamin B2 are lower than the recommended standard, which accounts for 52% of the overall number of athletes. Only 7 athletes' ADIs of vitamin C reach the recommended standard, which accounts for 26% of the overall number of athletes. We can learn from this table that athletes' intake conditions of vitamin A, vitamin B1, vitamin B2 and vitamin C all exist different levels of inadequate intake phenomenon, which indicates that athletes pay no attention to the sufficient vegetables and fruits' intake in their daily dietary nutrition.

**(3)Athletes' ingestion conditions of mineral substances**

Table 4 shows that athletes' ADIs of mineral nutrientsall reach or exceed the recommended value. Only 3 athletes' ADIs of K and Ca are lower than the recommended standard, which accounts for accounts for 11% and 11% respectively. We can learn from this table that there are great variations between different athletes' intake conditions of mineral elements.

**Table 4.** Athletes energy substance actual intake with all the Chinese people's energy intake recommendations

| Category | Energy | Protein | Fat | Sugar |
|---|---|---|---|---|
| The actual intake | 3625.2 | 332.3 | 525.3 | 602.3 |
| Recommended value | 2563.3 | 80 | 80 | 550 |
| difference | 1362.1 | 253.2 | 456.3 | 52.3 |
| T value | 11.255 | 12.522 | 10.233 | 2.013 |
| P values | 0.000 | 0.000 | 0.000 | 0.045 |

**Table 5.** Athletes vitamin actual consumption compared with Chinese residents vitamin intake

| Category | Vitamin A | Vitamin B | Vitamin C | Vitamin D | Vitamin E |
|---|---|---|---|---|---|
| Athletes intake | 3252.3 | 2.7 | 2.2 | 57.2 | 266.3 |
| Residents are recommended | 700 | 1.3 | 1.2 | 13 | 14 |
| difference | 2362.3 | 1.4 | 1.0 | 13.2 | 253.2 |
| T value | 11.520 | 3.363 | 4.233 | 2.200 | 15.261 |
| P values | 0.000 | 0.001 | 0.000 | 0.038 | 0.000 |

**(4) Comparison between Results of Dietary Nutrition Survey and Chinese DRIs**

Table 5 shows that the athletes' intake of energy, protein and fat is much higher than the recommended amount for Chinese citizens. Athletes' intake of energy is 1-2 times higher than the recommended amount for Chinese citizens while the protein intake is 1-7 times higher. Besides, athletes' intake of fat is 1-10 times higher than the recommended amount for Chinese citizens. Only one athlete intakes fat over 10 times more than average Chinese citizens, making 3.7% of the total population. The intake of carbohydrate by 10 athletes is less

than the recommended amount for Chinese citizens, covering 37% of the total population. From these data, it can be seen that the intakes of energy, protein and fat by athletes are more than the recommended amounts for Chinese citizens, meaning that athletes' demands for these substances are much larger than that of average Chinese citizens, and the gaps of demands for energy substances between athletes and average Chinese citizens are great. From the data on carbohydrate intake, the percentage of number of athletes whose intakes of carbohydrate have approached or surpassed the amount recommended for average Chinese citizens is only 67%, which means that athletes don't intake enough carbohydrate to different degrees.

It can be seen from Table 5 that the actual energy intake of athletes is higher than the recommended amount for Chinese resident by approximately 1,400 Kcal, and the intake of protein is 4 times higher while the fat intake is about 7 times higher. The intake of carbohydrate is higher than the recommended amount for Chinese citizens by 50g. By applying the comparative T-test to the actual intake of energy substances by athletes and to the Chinese DRIs, it can be found that the difference in carbohydrate intake between by athletes and by Chinese citizens isn't significant ($t=2.015$, $p\geq0.05$). The results of T-test applied to other energy substances (excluding carbohydrate) as well as their corresponding recommended amount for Chinese citizens show that: for energy, $t = 11.781$ and $p < 0.05$; for protein, $t = 12.811$ and $p < 0.05$; for fat, $t = 10.944$ and $p < 0.05$, which means differences between athletes' intake of other two energy substances (excluding carbohydrate) and recommended amount for Chinese citizens are significantly obvious.

As Table 5 shows, the actual intakes of Vitamin A, B1, B2, PP, C and E are much higher than their correspondingly recommended amount for Chinese citizens. By applying T-Test analysis into athletes' actual intake of vitamin and the recommended amounts in Chinese DRIs, for Vitamin A, B1, B2, PP, C and E, t is respectively 11.503, 3.551, 4.356, 15.192, 2.200 and 15.616, and p of all these vitamins is $< 0.05$, which means the differences between athletes' actual intake of each sort of vitamin and the correspondingly recommended amount for average Chinese citizens are significantly obvious.

## 4. Conclusions

(1) From the distribution proportions of calories for three meals (i.e. breakfast, lunch and dinner) each day from the survey results, compared with the recommended proportions, the intake of calories at breakfast is less than the recommended proportion, so does the intake of calories at lunch. Although the actual intakes at breakfast and lunch aren't enough, they're basically qualified. Meanwhile, the calories intake at dinner surpasses the recommended proportion by 0.3%, which is acceptable.

(2) According to actual intakes of nutrients by athletes, the fat intake has met the recommended standards. However, the intakes of energy, protein and carbohydrate by some athletes aren't enough to different degrees, especially the intake of carbohydrate. Athletes' intakes of vitamin PP and E are above the recommended standards while the intakes of Vitamin A, B1, B2 and C are below the standards to different degrees, especially the intakes of B1, B2 and C. Athletes' intakes of Fe, Zn and Se have already reached the recommended standards while only 11% of athletes don't have enough intakes, which means that a few athletes don't take enough mineral elements.

(3) Comparing the actual intakes of nutrients by athletes and Chinese DRIs, results show that athletes' intakes of energy substances, vitamins and mineral elements are above the recommended intakes for average Chinese citizens, and the in-between gaps are significantly obvious.

(4). Comparing the actual intakes of three energy substances and the ADIs for Chinese athletes, results have made it clear that the average intakes of protein and fat by athletes have approached or even exceeded the recommended amount while the intakes of

energy and carbohydrate are less than the recommended amount.

(5) Comparing the intakes of vitamins by female wrestlers and boxers in a particular province and the recommended amounts of vitamins for Chinese athletes, the results show that the average intakes of Vitamin A, PP and E by athletes are obviously higher than the correspondingly recommended amounts for Chinese athletes. The intake of Vitamin B2 happens to fall into the recommended range. Although the intake of Vitamin B1 hasn't reached the recommended standard, the gap is small. The intake of Vitamin C is simply less than the recommended standard for Chinese athletes.

(6) Comparing the intakes of mineral elements by female wrestlers and boxers in a particular province and the recommended amounts of vitamins for Chinese athletes, results show that the average intakes of all mineral elements by athletes have met and exceeded the recommended standards for Chinese athletes. Only a few athletes may lack some kinds of mineral elements to different degrees.

However, some problems in the survey may affect the results. (1). Due to the limits of manpower and material resources, only 27 athletes from two teams were chosen as subjects which was a relatively small sample. (2). In the surveying process, it has found out many athletes wrongly wrote the words, thus the names of food they ate couldn't be accurately understood. (3). Female athletes may be shy and weren't capable of honestly telling the actual amount of food they consumed. (4). Since a few athletes didn't cooperate well with researchers, some errors may occur to affect the accuracy of the results. (5). It was found out that the restaurant didn't supply fruit. For the above problems, we discussed and coordinated with coaches so that they could persuade the athletes to take this survey seriously and honestly tell the food they consumed each day. Meanwhile, we made every effort in ensuring the accuracy of this survey. During the research, we also found that excellent athletes usually possessed good qualities and had food according to regulated quota, which means they paid much attention to this dietary nutrition survey. On the contrast, athletes with bad performance couldn't have the healthy and reasonable diet, and they may be picky eater, reflecting their lack of understanding the significance of dietary nutrition.

By referring to related literature, we found that many athletes at the province-level or municipal athletic teams had different degrees of misunderstanding of dietary nutrition. Besides, their neglect of dietary nutrition was alarming. Athletes had a bad knowledge of nutrition, especially those about everyday dietary nutrition, for which some measures were proposed:

(1). The managers should find some time to educate the athletes about the knowledge about nutrition.

(2). Posters about dietary nutrition should be placed inside the restaurant, dinner table, training facility and dormitory so that they can learn the related knowledge whenever and wherever they can.

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# INVESTIGATION AND ANALYSIS ON OUTDOOR SPORTS AND DIETARY NUTRITION OF COLLEGE STUDENTS

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**ABSTRACT**
This paper shows that the balanced diet and adequate nutrition of sophomores and juniors have great significance for college students in daily work and study as well as the future work, this paper presents the investigation of 500 sophomores and juniors in five universities in a city to know the dietary characteristics, structures as well as the impact on physical quality by means of 24-hour recall and KAP questionnaire, and also gives some comments and suggestions through analyzing the existing problems of the investigation results. Through relevant researches, the subject is purposed to attract attention in Changsha about balanced diet of college students to improve the health for college students.

## 1. Introduction

Human beings cannot exist without food and nutrition, and the food and nutrition status is closely related with population quality, having great impact on prosperity of a nation. Study of epidemiology and nutrition proves that the growth & development, physique, working efficiency of human body as well as many chronic diseases like gastric cancer, diabetes, esophageal cancer and renal failure are not separated from the daily dietary structure.

Since reform and opening-up, our country has developed the economy very rapidly while the living standard of people has been improved greatly (Burford, 2004). College students can enjoy abundant food in canteens as well as many other snacks from some restaurants in or near campus (Parsons et al., 2005). However, college students are so particular that most of them like to choose food relying on their favorites, and they tend to eat meats, convenient food and quick meal instead of milk, beans and seafood. College students are mainly at the age of 19~23, the critical period for growth and development, longing for all kinds of nutrition elements. Unreasonable dietary structure may lead to obesity and other problems because of excessive energy, or will cause malnutrition due to lack of some necessary microelements such as calcium, iron, vitamins, high-quality protein, or further result in hypoglycemia because of insufficient energy and protein, therefore, students may not concentrate their attention during study and will get failed in examination (Williams, 2005). Long-term lack of nutrition elements will lower the immunity of body, inducing many types of infectious diseases and weakening the metal work capability, so the normal college students will be affected on their physique, or even their normal study as well as their life will be negatively impacted (Burke et al., 2010).

On premise that the balanced diet and adequate nutrition of sophomores and juniors have great significance for college students in daily work and study as well as the future work, this paper presents the investigation of 500 sophomores and juniors in five universities in a city to know the dietary characteristics, structures as well as the impact on physical quality in Changsha by means of 24-hour recall and KAP questionnaire, and also gives some comments and suggestions through analyzing the

existing problems of the investigation results. Through relevant researches, the subject is purposed to attract attention in Changsha concerning on balanced diet of college students to improve the health for college students (Castell, 2010).

## 2. Materials and methods

The paper presents the questionnaire survey to 500 students in five universities in Changsha. The dishes in the university canteens are weighed before and after cooking to work out the included nutritional elements by comparing to the Food Composition Table 1 and 2, besides, the student received the dietary survey for continuous three days by means of "investigation on dietary frequency" and "24-hour recall", then the ingestion of nutritional elements and heat energy each day were recorded in detail as per the investigation results as the reference data for analysis of relationship between dietary structure and physical quality of college students. The students will be regarded as light manual workers for conducting nutritional evaluation as per Chinese DRIs issued by Chinese Nutrition Society. The appraisal index for evaluation of dietary nutrition status included heat energy and other nutritional elements as specified in Table 3.

**Table 1.** Dietary nutrition evaluation standard

| Index/percentage | Inadequate intake | Serious lack |
|---|---|---|
| Heat energy | 92 | 85 |
| Other nutrients | 84 | 65 |

**Table 2.** City University students' breakfast

| Breakfast | Every day or eat mostly | Occasionally or almost don't eat | P value |
|---|---|---|---|
| sophomore | 84 | 18 | 0.148 |
| Junior | 95 | 10 | |
| Total | 86.3 | 13.9 | |

**Table 3.** City University students' breakfast foods

| Types of food choice | Sophomore | Junior |
|---|---|---|
| eggs | 44.2 | 60.2 |
| milk | 26.3 | 26 |
| cereal | 78.4 | 80.5 |
| vegetables | 50.2 | 41.2 |
| fruit | 1.6 | 0 |
| fish | 0 | 0 |
| meat | 0.8 | 0.6 |

The 500 sophomores and juniors, or the objects of investigation take meals in the canteens and shops in campus every day, so the dining table 2 in canteens of Central South University, Hunan Normal University, Central South University of Forestry and Technology, Hunan University of Chinese Medicine and Changsha University are used as the samples (Stear et al., 2009). Every university has several canteens, but generally they provide meals as the mixture of rice, noodle, vegetable and fruit with different cooking methods. The meals can basically satisfy the tastes of students at low price although some students think that they cannot stand the excessive spices such as salt and oil.

### 2.1 Dietary structure status of college students

Breakfast is very important for person as it can supplement energy and all kinds of

nutritional elements after the whole-night of metabolism. The survey of breakfast is shown in Table 3, and there is no obvious different between the sophomores and juniors according to the Chi-square test (P >0.05).

Refer to Table 4 for the food categories of breakfast for college students in Changsha, which proves that the sophomores are obviously different from junior students in terms of food selection (Senchina et al., 2011).

According to the nutrition theory, the following selections of breakfast are regarded to be unreasonable during Chi-square test for relevance: only select cereal, only select milk, only select egg, only select fish added to meal, and only select vegetable added to fruit; and the selections of breakfast below will be regarded as reasonable: choose cereal added to milk, choose cereal added to egg, chose cereal, milk and egg, or choose milk added to egg (Senchina et al., 2011). The investigation results are shown in Table 5, proving that the sophomore are more rational than juniors in terms of breakfast category (P <0.01).

**Table 4.** The rationality of the college students' breakfast food choices

| Food choices | Reasonable | Unreasonable | P value |
|---|---|---|---|
| sophomore | 45.36 | 56.36 | 0.00036 |
| Junior | 21.25 | 78.45 | |
| Total | 32.69 | 65.23 | |

**Table 5.** City university students lunch and dinner meal

| Types of food | Twice a day | | Once a day | | Once every two days | |
|---|---|---|---|---|---|---|
| choose | sophomore | Junior | sophomore | Junior | sophomore | Junior |
| cereal | 24.3 | 30.5 | 44.8 | 60.3 | 14.3 | 10.2 |
| eggs | 45.2 | 34.5 | 56.3 | 52.4 | 21.5 | 12.3 |
| meat | 39.6 | 25.1 | 26.3 | 48.5 | 36.2 | 21.5 |
| vegetables | 17.5 | 40.6 | 29.5 | 52.1 | 19.6 | 45.6 |
| soy | 20.6 | 16.9 | 36.5 | 12.3 | 19.4 | 51.2 |
| dairy | 51.0 | 25.8 | 42.1 | 32.5 | 32.5 | 23.6 |
| fruit class | 0.5 | 1 | 21.3 | 21.4 | 70.1 | 25.2 |
| fish | 0.8 | 0.6 | 15.3 | 12.6 | 30.2 | 12.3 |

**Table 6.** College students' physical quality index comparison in 2012

| | Male | | | Female | | |
|---|---|---|---|---|---|---|
| | 50m | 100m | The experimental steps | 50m | 100m | The experimental steps |
| sophomore | 7.23 | 233.5 | 84.2 | 8.94 | 245.6 | 66.3 |
| Junior | 7.25 | 235.3 | 85.3 | 9.52 | 248.9 | 67.4 |

Refer to Table 6 for the dietary condition of launch and dinner. According to Chi-square test, sophomores intake more milk, meat, fish and fruit than juniors with obvious different (P< 0.05 or 0.01), but juniors intake more cereal and fruit than sophomores (P<0.01). Both the sophomores and juniors intake little fish, egg product, dairy product and bean product in daily diet.

### 2.2. Analysis on dietary structure college students in Changsha

The subject, in line with the study demand, designed the KAP questionnaire to survey the

current dietary structure of 500 sophomores and juniors in five universities in a Changsha, including the daily three meals (breakfast, lunch and dinner), snack, supper, as well as acquisition of meat, fish and daily nutritional elements. Based on the above condition, this section will give the analysis of dietary structure for sophomores and juniors. The investigation shows that people may become short of some nutritional elements or have excessive heat energy, protein and fat due to their unreasonable diet, while may lead to hazards of chronic diseases in a long term (Ranchordas et al., 2010).

**(1) Analysis on breakfast**

Breakfast can greatly perfect the ingestion of nutritional elements for students and help them to keep their mind active and easier to understand and master knowledge. However, the cognitive ability, physique and other aspects of college students will be badly influenced if they do not eat breakfast or have unscientific breakfast (Lowery, 2004). The investigation results in Table 6 shows that juniors tend to have scientific breakfast as there are only 10% students in the five universities who do not eat breakfast or seldom eat breakfast, and this data is the lower limit of the literature proportion at 10% and at the same time lower than the proportion reported by Hu Xiaoqi about the problem that primary and secondary students who do not eat breakfast (King et al., 2013). Among the 500 sophomores and juniors, the proportion of sophomores and juniors who do not eat breakfast are 17% and 10% separately, and there is no obvious different between the two groups as per the Chi-square test ($P > 0.05$).

As for the sophomores and juniors who tend to eat breakfast, the proportion of sophomores and juniors choose cereal are 79.8% and 80.5% separately, and there is no obvious different between the two groups as per the Chi-square test ($P > 0.05$)(Pilis et al., 2014). Nevertheless, the breakfast of juniors is unscientific, simple, lacking nutrition. According to researches, scientific breakfast should consist of cereal+ egg, cereal+ milk, cereal+ egg+ milk, or egg+ milk. The Chi-square test shows that the sophomores tend to be more reasonable than junior in terms of breakfast category ($P < 0.01$).

**(2) Analysis on launch and dinner**

Dietary nutrition status is closely related to the physical quality. According to the diet pagoda in China, an adult shall intake cereal of 300~500g, vegetable of 400~500g, fruit of 100~200g, fish of 50g, egg and poultry of 50~100g, egg of 25~50g, milk of 100g, bean & bean product of 50g, and oil & fat not exceeding 25g. According to the investigation results (as shown in Fig 5 & 6), the daily consumption of seafood, egg product, milk product and bean product is somewhat low, which is in compliance with the conditions in China.

The sophomores have meat and fish more than the juniors and the ingestions of meat and fish between the two groups are not different greatly based on the Chi-square test ($P> 0.05$). Fish contains high protein and low heat energy rich in unsaturated fatty acid, mineral substance and Vitamin D, greatly helpful to human body especially for students who are busy in examination and heavy schoolwork. Juniors intake more cereal than sophomores, and there is an obvious difference in terms of cereal ingestion based on the Chi-square test ($P< 0.01$).

Sophomores intake more milk than juniors, and there is an obvious difference in terms of cereal ingestion based on the Chi-square test ($P< 0.01$).

## 3. Results and discussions

### 3.1. Physical quality

The investigation tests physical fitness and endurance of female sophomores and juniors through women's 50m & 800m and also tests physical fitness and endurance of male sophomores and juniors through men's 50m & 1000m, moreover, the muscle tolerance of sophomores and juniors is reflected in step experiment. Refer to Table 7 for the testing results of physical fitness of sophomores and juniors.

### 3.2. Physical function

The vital capacity and body mass index are used in the investigation to show the vital function of sophomores and juniors. The overall vital function standard of sophomores and juniors in 2012 is shown in Table 8, indicating that the difference between sophomores and juniors in terms of vital capacity is great ($P < 0.01$), and another index of body function-vital capacity /body mass index (vital capacity index) is also lower than the overall average in our country, presenting obvious different ($P<0.01$).

The results obtained by this method is different to that of BM method and method of weight for height, in which the low body fat is 4.47%, and overweight or obesity weight is 21.23%, among which the obesity rate is relatively high while the low-weight rate is low.

**Table 7.** Sophomore, junior functional index contrast in 2012

| | Male | | Female | |
|---|---|---|---|---|
| | Lung capacity | Vital capacity index | Lung capacity | Vital capacity index |
| sophomore | 3362.52 | 54.21 | 2248.12 | 43.65 |
| Junior | 3265.54 | 51.69 | 2128.10 | 45.58 |

**Table 8.** Rise in 2012, weight table

| | Male | | Female | |
|---|---|---|---|---|
| | height | weight | height | weight |
| sophomore | 174.26 | 62.31 | 159.45 | 49.52 |
| Junior | 175.36 | 62.59 | 157.26 | 44.23 |

**Table 9.** Percentage of weight status of sophomore and junior

| | malnutrition | The low weight | The normal weight | overweight | obesity |
|---|---|---|---|---|---|
| sophomore | 10.25 | 22.59 | 54.28 | 10.26 | 2.45 |
| Junior | 13.23 | 18.45 | 52.69 | 11.59 | 3.69 |

### 3.3. Height & weight

**(1) Normal weight**

Refer to Table 8 for height and weight information of sophomores and juniors in five universities of a city in 2012.

**(2) Standard weight converted from height**

The standard height is converted by height based on the certain rate between height and weight. Standard weight can reflect the circumference and intensity of human body, and it is very important to evaluate the development standard, nutrition and symmetry of body shape. Refer to Table 9 for percentage statistics of standard weight of all grades for sophomores and juniors converted by height.

### 3.4. Analysis on physical health of college students in Changsha

The investigation tests speed quality of female sophomores and juniors through women's 50m & 800m and leg muscle tolerance through step experiment, and also tests physical fitness and endurance of male sophomores and juniors through men's 50m & 1000m. The physical fitness testing results of sophomores and juniors is shown in Table 9, showing that female sophomores get the women's 50m grade better than female juniors by 0.05s, resulting in no obvious difference, while male sophomores get the men's 50m grade better than male juniors by 0.35s, resulting in obvious difference. 50m is a simple exercise, but it can reflect the flexibility, body harmony, suppleness of joint & muscle and strength & tolerance of muscle of human body,

therefore, 50m can show comprehensive quality of human body to some extent. 50m examination is targeted to know running speed, sensitivity and flexibility level of nervous system of students.

Female sophomores get the women's 800m grade better than female juniors by 3s, resulting in obvious difference ($P< 0.01$), while male sophomores get the men's 1000m grade better than male juniors by 4s, also resulting in obvious difference ($P<0.01$). 800m and 1000m are mainly purposed to test endurance of students, and it can also reflect whether the function of cardiovascular and respiratory systems as well as the muscle tolerance of college students. Generally the step experiment is used to evaluate the muscle strength and tolerance as it can work out the muscle strength and tolerance in a safety way. Female sophomores get the step experiment grade better than female juniors by 2/min., resulting in obvious difference ($P< 0.05$), while male sophomores get the step experiment grade better than male juniors by 1.7/min., also resulting in obvious difference ($P<0.05$), proving that sophomores is superior than juniors regarding to their strength at waist and legs.

The vital capacities between sophomores and juniors in 2012 are largely different from each other ($P < 0.01$) as shown in Table 9. According to the theoretical analysis, the absolute value of vital capacity is related to the following three factors: anatomical vital capacity, lung ventilation ability and the self-contraction strength of respiratory muscle. Except being affected by growth and development condition, the body vital capacity is also connected with exercise condition and living habit (for example smoke or not), however, juniors are faced with employment difficulty and heavy schoolwork, and they cannot concentrate themselves into physical exercise, thus their vital capacity will be affected. The investigation indicates that 70.13% of juniors treat the exercise randomly, 20.7% have exercises once a week, 6.49% have exercises twice a week, only 2.59% have exercises three times or more every week, and none of them have exercises four times or more every week, showing that juniors have not get the good awareness for having exercise and insisting on exercise, as a result, their vital function is poor. Sophomores do better than juniors on having exercise as they have one PE class every week (Lan and Xue, 2013). To strengthen the exercise of heart-lung function for sophomores and juniors, enhance exercise and regulation beyond PE class and perfect the body function for sophomores and juniors through the way like morning exercise can not only perfect the heart-lung function but also let college students have the awareness for having exercise in all of their life (Guerra et al., 2001).

## 4. Conclusions

The investigation and analysis on dietary structure and physical quality of college students in Changsha proves that the sophomores and juniors have many problems in dietary structure and physical quality such as insufficient ingestion of Ca, vitamin C, vitamin B1 and vitamin B2, which causes nutritional deficiency because they pay too much attention to their stature and insist on weight loss on the one hand, and on the other hand, they do not have enough consumption ability. For example, they are still rely on their parents in terms of economy, they tend to not have breakfast or eat breakfast so late or even in class, besides, the light-out time in dormitories is so last as lasting to 1:00-2:00 in midnight that they will eat supper at excessive amount such as snacks, which will result in obesity or other diseases. College students choose food relying on their tastes and daily dietary habit instead of the contents of nutritional elements in food. The dietary management and intervention are necessarily strengthened to guide college students for reasonable diet, and the specific measures below can be taken: strictly manage the schedule in dormitories and specify the light-out time at 11 pm in night, avoid some bad habit such as having food before sleep; insist on drill in the morning while make them eat breakfast in correct time to help them get enough nutrition; limit or ban snake shops or retail departments in campus to make them do not has access to snacks, or limit the supply or adopt

fixed supply in snake shops or retail department in campus.

To sum up, sophomores and juniors do not have good living habits as they are faced with heavy schoolwork as well as pressure for postgraduate examination or employment, and they are mainly lack of milk, bean food, animal food, vegetable and fruit. Meanwhile, some students also have several food or nutrition in excessive high amount such as over ingestion of oily, salty and high-calorie food, which will lead to obesity, high blood pressure and other unfavorable physical conditions. Students do not have access to food of many categories, and they do not know enough knowledge about food nutrition and nutrition element, and also fail to gain high dietary quality for themselves. Therefore, the publicity on nutrition knowledge to sophomores and juniors or even to the freshmen is necessary in order to help them choose reasonable food categories, improve dietary structure as well as physical fitness.

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# A RESEARCH ON INFLUENCES OF NUTRITION INTERVENTION ON TAEKWONDO ATHLETES' PHYSICAL CAPACITY RECOVERY

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**ABSTRACT**
In this paper, main physical function indexes reflecting qualities including athletes' strength, speed, flexibility, endurance, coordination and sense of rhythm are monitored through sports nutrition intervention in order to study on its major influencing factors for especially aerobic metabolism capacity, find out nutrition intervention methods suitable for high-level college taekwondo athletes' physical capacity promotion, and explore more effective training methods so as to achieve the goal of enhancing college taekwondo athletes' physical capacity.

## 1. Introduction

With rapid development of modern society, sports have become an indispensable part of people's daily life, and competitive sports have received more and more attention. Taekwondo was firstly a sport promoted to the whole world by Koreans (Fleming and Costarelli, 2009). As a competitive sport with leg actions as its main offensive skills, a delicate body art and effective fitness means, taekwondo boasts extremely high martial arts practicability and ornamental traits. Currently, this new sport and workout has been favored by the majority of youths. Since it was officially named as taekwondo in 1955, this sport has developed at an astonishing speed and started a taekwondo craze all around the world. After several decades of development, it has formed completely independent international sports organizations and regular international games. In 1966, the International Taekwondo Federation was found. In 1973, the World Taekwondo Federation was found. In 2000, taekwondo was listed into an official event of the Olympic game (Pitriani, 2012). At present, the International Taekwondo Federation has 147 member states, which is among the best single sports in the world. The number of people practicing taekwondo around the world has reached 70 million (Chen et al., 2016). Along with prosperous development of taekwondo and its confirmation as a formal sport, our national sports field started to realize the importance and necessity of developing taekwondo. Presently, taekwondo has been listed into the Program of Striving for Olympic Glory (Moreira et al., 2012). Sports colleges from all parts of the country and professional sports associations have all created professional teams, among which participating teams and the number of athletes have hit the best of domestic individual championships for multiple times. Taking Chen Zhong's women's 67kg weightlifting champion at the 2000 Sydney Olympic Games as a symbol, our national taekwondo competitive level has been greatly improved (Chen et al., 2016). Up to date, China's athletes have won 5 Olympic gold medals for individual sport (2 gold medals from Taiwan included) and dozens of world medals (intercontinental) in the Olympic Games, world championships, the World Cup, the Asian Games, etc. With excellent performance of Chinese athletes in arenas, popular taekwondo

has entered a rapid developmental stage. Taekwondo associations all over the world have been established, and a great many of taekwondo clubs and venues have sprung up like mushrooms in various places. There are over 500 taekwondo venues in Beijing and Hangzhou, excluding taekwondo classes in amateur sports schools. Taekwondo has gradually become a fashionable fitness exercise and been favored by the wide public, especially those highly active youths and students. According to incomplete statistics, population practicing taekwondo in China (including Hong Kong, Macao and Taiwan) has surpassed 5 million (Kang, 2014). Continuous development and standardization of public taekwondo have provided solid mass foundation for high-level competitive taekwondo. It is predicted that taekwondo has stepped in to a fast track of healthy development. However, with meanwhile rapid growth of taekwondo, we still cannot ignore those problems and disadvantages existing during the development. However, China is a relative newcomer in taekwondo. Although our taekwondo athletes have taken a share of gold medals in the previous two Olympics, there is a certain difference between them and elite athletes from other countries. In order to adapt to development and enhance competitiveness of taekwondo (Su, 2015), we must find solutions and countermeasures. Nowadays, taekwondo has become a potential advantage program of our national "Program of Striving for Olympic Glory", and we are planning to win 2 gold medals in the 2008 Beijing Olympic Games. China's taekwondo is facing unprecedented opportunities and challenges. Thus, we should find problems and deficiencies existing in development of our taekwondo, seek solutions, prepare our national taekwondo team for competitions in the 2008 Beijing Olympic Games, and strive for greater results.

Rising level of competitive taekwondo in colleges has put forward higher requirements for physical functions of athletes. Elite taekwondo team in Southwest Jiaotong University (hereinafter referred to as the "Southwest Jiaoda") has won champions for several times in national, provincial, municipal taekwondo matches. Before participating in the First National Students' Health & Vigor Competition in 2004 and International Taekwondo Competition in China, Japan and Korea, in order to enhance athletes' potential athletic abilities and promote fatigue tolerance, we have carried out nutritional intervention for participating members(Qu, 2014), tested varied abilities including speed endurance and strength, so that they can quickly recover physical power and get into next phase of training after intensive training. Major physical function indexes inflecting taekwondo athletes' qualities including strength, speed, flexibility, endurance, coordination and sense of rhythm are monitored through sports nutrition intervention, and its major influencing factors, especially aerobic metabolism capacity, have been studied through principal component analysis, so as to find out nutrition intervention methods suitable for promoting high-level college taekwondo athletes' physical and provide foundation for high-level college taekwondo athletes' training.

## 2. Materials and methods

### 2.1. Research Objects

There are 12 athletes (female) in Southwest Jiaoda taekwondo team who have won taekwondo champion in "the First National Students' Health & Vigor Competition" and the second place of "International Invitational Tournament in China, Japan and Korea", and they are divided into the experimental group (8 cases) and the control group (4 cases).

### 2.2. Research Methods

**(1) Nutritional Supplements**

Investigate dietetic condition of athletes, determine hemoglobin, and regulate dietary structure and energy intake of students. From the beginning of intensive training until competition, provide on a daily basis 600mg chalybeate, 25g protein, 40f carbohydrates, electrolytes and 500ml microelement beverage for experimental athletes on the basis of regular diets. Provide no supplement for athletes in the control group.

**(2) Effectiveness Observation**

Observe physical capacity recovery of different training intensities in terms of physiological function indexes, it major index maximal exercise capacity would be tested through America margut te Cordioof electrocardiogram treadmill analysis system test of sports psychology. V.O2max would be calculated through EGH-□constant-motion-rate bicycle measuring and calculation; Maximal anaerobic power would be tested through GC811 bicycle with constant-resistance capacity; height, weight, lung capacity and hemoglobin would be tested through regular checkup equipment.

**(3) Training Intensity and Testing Time**

Carry out regular training for all members of the experimental group and control group: two months before the competition, three times a week, 2h each time (□regular warm-up, □interval training of set movements, □taekwondo set movements, □set movements of cheering squad, □specialized fitness exercise, □relaxing); Intensive training: one month before the competition, five times a week, 2.5h each time (□warm-up of taekwondo movement combination, □route of set movements,□4 sets of optional and defined competition movements for 2 ~ 3 times, □specialized fitness exercise, □relaxing, test for once one week before the competition during excessive recovery phase)

## 3. Results and discussions

### 3.1. Shapes and Physical Capacity of Elite College Taekwondo Athletes

Physique constitution of elite college taekwondo athletes requires their relatively high comprehensive qualities including speed, endurance, flexibility, coordination and rhythmic ability. From a standpoint of body anatomy, it requires aesthetic shape, line and muscle; in terms of aerobic metabolism ability, taekwondo belongs to aerobic exercises but also requires a fine speed explosive force; from the perspective of nutriology, the diet shall not only satisfy athletes' demands for intensive training and cultural course study but also meet the needs of shape. Shapes and physical capacity indexes can be seen in Table 1.

**(1) Composition of Physique Components**

With heights of around 165cm and weights of about 51kg, female college athletes have showed their fine body shapes through the Body Mass Index. Body fat has accounted for 21% of total body weight. With certain but not much fat, those athletes boast graceful body lines. With a large Lean Body Mass proportion, they are equipped with good muscle strength (Moreira et al. 2012).

**(2) Hemoglobin**

As an important index in nutrition evaluation, hemoglobin can not only know about nutritional status of athletes, but also assess amount of training from its variation. Due to hematoclasis caused by irrational dietary structure of athletes in the above-mentioned school and perennial heavy-load training, athletes have been in a state of anemia. And this is a common phenomenon among college students (Cho et al., 2013).

**(3) Anaerobic Metabolism Capacity**

Evaluate speed strength and explosive power of athletes in terms of anaerobic power. Anaerobic power can increase the load through Wing ate method; when athletes pedal at a full speed, their metabolism will focus on anaerobic type. Use V.O2max for evaluating aerobic metabolism capacity of athletes, and assess maximal exercise capacity and endurance through maximal exercise capacity; Lung capacity reflects ventilation amount of lung at a single breath which is irrelevant to anaerobic condition and reflects anaerobic metabolism capacity from another perspective. From Table 1, it is shown that lung capacity, anaerobic function, V.O2max and maximal exercise capacity of college taekwondo athletes are over one standard deviation higher than ordinary female students, while their body fat percentage is two standard deviations slightly lower than that of ordinary girls, and hemoglobin level is one standard deviation lower than that of ordinary girls.

**Table 1.** Outstanding college students taekwondo athletes form function index statistics

| | | Height | Weight | Hemoglobin | Lung capacity | VO2 | Anaerobic work | The biggest sports ability | Body fat |
|---|---|---|---|---|---|---|---|---|---|
| fFmale | The average | 165.32 | 50.21 | 10.25 | 3365.25 | 2.65 | 356.45 | 1457.21 | 21.14 |
| | The standard deviation | 1.25 | 3.25 | 0.46 | 115.24 | 0.36 | 45.36 | 130.69 | 2.36 |
| Ordinary female | The average | 153.26 | 51.24 | 11.25 | 2.36 | 2.36 | 318.25 | 1.325 | 26.36 |
| | The standard deviation | 1.25 | 6.36 | 0.75 | 213.58 | 0.33 | 51.25 | 147.58 | 4.12 |

### 3.2. Influences of Nutrition Intervention on Aerobic Metabolism Capacity at Different Training Intensities

Physical growth of college athletes aged between 20 and 23 has been stabilized, and variations in its morphological indexes are non-specific. This paper will focus on analyzing influences of nutrition intervention on aerobic metabolism capacity of athletes at different training intensities.

**(1) Influences of Training on Aerobic Metabolism Capacity of Athletes in the Control Group.**

Athletes in the control group have carried out simple sports exercises only according to training plan, without any additional conditions in training of any intensity. From Table 2, after intensive training, hemoglobin and maximal exercise capacity of athletes in the control group would markedly decline (Bürger-Mendonça et al., 2015). Decline degree of hemoglobin is directly proportional to training intensity; that is, the greater the intensity is, the quicker the hemoglobin will decline. After one-month intensive training, hemoglobin of athletes in this group decline by 30.5 ~ 1g than regular training, and oxygen transport and fatigue resistance of them would be then affected. During intensive exercises, oxygen consumption would multiplied to 10 ~ 15 times larger than that in regular training, which may lead to increase of free radicals by 2 ~ 3 times (Burton et al., 2006). Free radicals would cause lipid peroxidation of cell membrane (mainly myocytes and erythrocytes) and subsequent premature emergence of fatigue. Athletes in the control group themselves felt overwhelmed by fatigue and caught colds at varying degrees. Under an exhausted state, the decline of maximum exercise capacity turns to be self-evident. Under the same assessment, V. O2max of aerobic metabolism capacity didn't turn on an obvious downward trend, and anaerobic power and lung capacity haven't apparently enhanced. The results were due to that they all belong to anaerobic metabolism and their instantaneous powers have been tested. It is reasonable that all indexes in excessive recovery phase have restored to above conventional level.

**(2) Influences of Intensive Training on Athletes' Physical Capacity under Nutritional Intervention**

From the beginning of intensive training until competition, athletes from the experimental group have been provided with nutritional supplements (Hashemvarzi et al., 2014). A large amount of data can demonstrate that their improvement of their lung capacity is relevant to

genetic factor and varied training exercises. This paper focuses on discussing hemoglobin. From Table 3, hemoglobin of athletes in the experimental group hasn't evidently increased, which indicates that training intensity was just suitable. Iron demand of athletes is higher than that of ordinary people. In continuous intensive training, catabolism of protein was then strengthened, and sweat and iron lost. Sports anemia is due to intake deficiency from increasing protein demand and consequential erythrocyte hemolysis. In high-intensity training, athletes were on the one part expending protein and losing iron, and on the other part maintaining a balance between enough protein and iron supplements. Evident increase of hemoglobin indicates that training intensity can be enhanced. Higher hemoglobin can better exert to the maximal aerobic capacity of human body. V.O2max, maximal exercise capacity and anaerobic power have obtained obvious rise in the experimental group, and the results are also closely related to maintenance of nutritional supplement and hemoglobin besides training. During intensive training, athletes in the experimental group were all energetic without any fatigue, and no one has caught a cold.

**Table 2.** The control group in different training intensity test of significance

| Lung capacity | Regular training | Intensive training | t | Excessive adjustment before reply |
|---|---|---|---|---|
| Hemoglobin | 3365 | 3352 | —— | 3500 |
| VO2 | 12 | 10.54 | 3.25 | 10.47 |
| Anaerobic work | 2.58 | 2.58 | —— | 3.11 |
| The biggest sports ability | 365.69 | 356.47 | —— | 3.59 |
| Body fat | 1785.25 | 1520 | 3.25 | 1158 |

**Table 3.** The control group, experimental group intensive training fitness matching test

| | The control group | experimental group | t |
|---|---|---|---|
| Hemoglobin | 3350 | 3124 | ——— |
| VO2 | 10.25 | 11.54 | 2.36 |
| Anaerobic work | 2.545 | 3.254 | ——— |
| The biggest sports ability | 317.25 | 390.45 | 2.58 |
| Body fat | 1502 | 1752 | 4..12 |

**(3) Index Comparison between Experimental and Control Group after Training**

The two groups were trained under the same intensity. From Table 3, it can be seen that both two groups were without nutritional supplements during regular training and there was no obvious discrepancy between indexes of the two groups (Hashemvarzi et al., 2014). However, in intensive training, after the experimental group was provided with nutritional supplement, we can easily find that there was obvious discrepancy in its hemoglobin, anaerobic power, V.O2max and maximal exercise capacity. Taking hemoglobin as an example: in regular training, there was no obvious discrepancy in indexes of the two groups; in intensive training, hemoglobin of the experimental group declined while that of the control group increased, and no significant

difference existed among the two groups. The reason for between-group discrepancy in V.O2max, anaerobic power and maximal exercise capacity was also caused by the decline of one group and rise of another group, which has fully reflected the great significance of nutritional supplement on promoting students' aerobic metabolism capacity. If enough nutrition was supplemented, athletes would become energetic and be able to accomplish intensive training and cultural courses' study.

## 4. Conclusions

China is a relative newcomer in taekwondo. Although our taekwondo athletes have taken a share of gold medals in the previous two Olympics, there is a certain difference between them and elite athletes from other countries. In order to adapt to development and enhance competitiveness of taekwondo, we must find solutions and countermeasures. Nowadays, taekwondo has become a potential advantage program of our national "Program of Striving for Olympic Glory", and we are planning to win 2 gold medals in the 2008 Beijing Olympic Games. China's taekwondo is facing unprecedented opportunities and challenges. Thus, we should find problems and deficiencies existing in development of our taekwondo, seek solutions, prepare our national taekwondo team for competitions in the 2008 Beijing Olympic Games, and strive for greater results.

Rising level of competitive taekwondo in colleges has put forward higher requirements for physical functions of athletes. Elite taekwondo team in Southwest Jiaotong University (hereinafter referred to as the "Southwest Jiaoda") has won champions for several times in national, provincial, municipal taekwondo matches. Before participating in the First National Students' Health & Vigor Competition in 2004 and International Taekwondo Competition in China, Japan and Korea, in order to enhance athletes' potential athletic abilities and promote fatigue tolerance, we have carried out nutritional intervention for participating members, tested varied abilities including speed endurance and strength, so that they can quickly recover physical power and get into next phase of training after intensive training. Major physical function indexes inflecting taekwondo athletes' qualities including strength, speed, flexibility, endurance, coordination and sense of rhythm are monitored through sports nutrition intervention, and its major influencing factors, especially aerobic metabolism capacity, have been studied through principal component analysis, so as to find out nutrition intervention methods suitable for promoting high-level college taekwondo athletes' physical and provide foundation for high-level college taekwondo athletes' training.

(1) Physique constitution of college taekwondo athletes: the Body Mass Index shall be 165:51, and body fat percentage shall be around 20%; their anaerobic power, V.O2max and maximum exercise capacity shall be higher than those of ordinary students by 1 ~ 2 standard deviation.

(2) During intensive training, if without nutritional supplement, hemoglobin will decrease 0.5 ~ 1g each month, and maximal exercise capacity will significantly decline; Within nutritional supplement, hemoglobin will maintain its original level; V.O2max and maximal exercise capacity will increase visibly; Athletes will be energetic, with enhancing disease resistance.

(3) Among the experimental group and control group, hemoglobin, anaerobic power, V.O2max and maximal exercise capacity of the former group have taken on an increasing tendency while those of the latter group decreasing after intensive training. The significant group discrepancy has confirmed the importance of nutritional supplements on aerobic metabolism capacity of human body.

(4) Physique constitution of college taekwondo athletes can serve as the foundation of athlete selection.

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# DETECTION OF SYNTHESIZED COLORANTS IN SPORTS DRINKS

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**ABSTRACT**

This study aimed at detecting synthesized colorants in sports drink to ensure the health of consumers. Synthesized colorants which are unbeneficial to human body in commonly seen sports drinks were taken as research subjects; detection and analysis method was used. First, the study discussed over the current research status using literature data method, expert interview method, interdisciplinarity method and experimental method. Then the pre-processed sports drink was detected using high pressure liquid chromatography (HPLC) and ultra-high pressure liquid chromatography (UPLC). Finally, the results obtained by detection based on HPLC and UPLC were compared and analyzed. Research results demonstrated that, the value of the linear correlation coefficient R for the detection of the pre-processed sports drink samples using HPLC was between 0.9906 and 0.9995, and the average recycling rate was between 80.5% and 97.1%. The linear correlation coefficient R for the detection of the pre-processed sports drink samples using UPLC was between 0.9909 and 0.9999, and the average recycling rate was between 71.3% and 113.5%. Compared to HPLC, UPLC was more sensitive. UPLC makes up the blank of multi-group detection of colorants and provides a technical support for the detection of colorants in sports drink.

## 1. Introduction

Food additives is a kind of material that can improve the color, favor and taste of food. The synthesized colorant, one kind of food additives, is able to control the color change of food and drink. Revision on the National food safety standard-standards for uses of food additives (GB 2760—2011) (Wang et al., 2011) includes 11 water-soluble artificially synthesized colorants (Liao et al., 2012; Cong et al., 2015) including quinoline yellow, brilliant blue, lemon yellow, sunset yellow, acid red, amaranth, new red, carmine, allura red, indigo blue and erythrosine. There are only 5 international standards for water-soluble artificially synthesized colorants in the current stage in China; besides, the components which require detection account for a small proportion and only cover a small part of foods in China.

Globally, many countries pay much attention to the application of colorants in food and drink. In 1960, American parliament passed Federal Food, Drug, and Cosmetic (FD&C) Act (Casadevall et al., 2014), emphasizing the importance of isolating colorants for management. The regulation claims that, colorants used in food, drugs and cosmetics must be approved by the Food and Drug Administration of America before entering into the market and samples of every batch produced by domestic or foreign producers should be strictly inspected for purification to confirm whether the colorants satisfy relevant limit standard. The European Union released 89/107/EEC which was related to food colorants (Türküler Isiksel, 2010). 94/36/EC which was proposed to normalize the use of food colorants gave a definition of colorants, and moreover the appendix exhibited the lists of colorants which were licensed to be used, forbidden to be used, limited to be used in usage and can be used in a moderate amount.

China also proposed the Edible Synthesized Dye Management Method (Wedge, 2013) in 1960 to temporarily monitor the application of food colorants in China. GBN 50-1977 The Applied Sanitary Standard of Food Additives (Sato, 2010) was formulated and put into trial use from 1973 to 1977. In 1980, Revision on the National Food Safety Standard-Standards for Uses of Food additives (GB 2760—2011) was released; afterwards, it was supplemented and amended for several times. Currently, GB 2760-2014 The Applied Sanitary Standards of Food Additives is taken as the national standard, and moreover the synthesized colorants allowed for use, the ones allowed for use within a limited quantity and those allowed for use in food according to the production needs.

Artificially synthesized coloring matters refer to organic coloring matters prepared using artificial chemical synthesis method. Most of the synthesized coloring matters that are allowed to be used globally are water-soluble coloring matters (Dupuy and Stéphanie, 2014). But the recent studies suggested that, nearly all the synthesized coloring matters could not provide human body with nutritional substances and some of them might even threaten the health of human body for its carcinogenicity (Verhagen et al., 2011; Boobis et al., 2013). In this study, synthesized colorants in sports drink were detected using high pressure liquid chromatography (HPLC) and ultra-high pressure liquid chromatography (UPLC). This work provides relevant departments with a reference and some suggestions in the aspect of the detection of synthesized colorants.

## 2. Study of detection methods for artificially synthesized colorants in sports drink

The intake of artificially synthesized colorants can block the absorption of vitamins, which can thereby damage nervous system and induce a series of symptoms such as the absence of mind. Moreover, artificially synthesized colorants are not easy to be oxidized and eliminated when accumulating in human body. Some colorants can affect liver functions as well and they will combine with target cells in human body to form tumor cells after decomposition. Therefore, the detection of colorants in sports drink is especially important.

### 2.1. Referable instrument method

With the development of science and technology, more and more novel instruments and methods have been developed. The detection methods of artificially synthesized coloring in sports drink include HPLC (Svec and Frechet, 2012), high-performance liquid chromatography-mass spectrometry (HPLC/ES-MS) (Vicente et al., 2015), electrochemical analysis method (Mattsson, 2015), capillary electrophoresis, fluorescence analytical method (Gostishchev et al., 2012), thin-layer chromatography and polyamide adsorption method. In this study, HPLC, HPLC-EC-MS, electrochemical analysis method and fluorescence analytical method were used.

HPLC adopts a novel infusion pump including a high-sensitive detector and a high efficiency particle stationary phase. It operates to elute and isolate solutes with different absorption capacity and molecular size through their multiple exchanges between stationary phase and moving phase. Combining the advantages of both HPLC and MS, HPLC/ES-MS not only has the high efficient isolation ability of liquid chromatography, but also can provide structural information like MS. Electrochemical analysis method is applied for analyzing the electrochemical properties of substances in solution which is also usually used for isolating and identifying substances such as protein, nucleic acid and amino acid. Fluorescence analytical method is established based on different absorption wavelengths of substances under electromagnetic radiation. It is featured by high sensitivity, less sampling volume and convenient use, but there are many interference factors.

## 2.2. Referable pre-processing method

Through looking up relevant literature data and interviewing experts, we found that, the preprocessing of sports drink before experiment was in certain relationship with the detection of some physiological indexes. In this study, the adopted methods included polyamide absorption method, liquid-liquid extraction method, direct sample introduction method, microwave assisted extraction method and solid phase extraction.

Polyamide absorption method completes the extraction of coloring matters through optimizing the dose of polyamide adsorbent, apparatus and eluent. Liquid-liquid extraction method purifies the samples by making use of the characteristic that the detected components and impurities are incompatible. Though it is easy to be operated, poor repeatability, low recycling rate and emulsification can frequently occur. Direct sample introduction method simplifies the complex preprocessing process, which can significantly improve working efficiency (Tokita et al., 2014) and save cost, and moreover, more microelements which are difficult to be detected can be detected using the method (Gomes, et al., 2015). Microwave assisted extraction method is featured by short extraction time, high extraction precision, high extraction efficacy and small dose of extractants. When solid phase extraction is used, substances in detected samples which have influence on target chemical are isolated effectively using non-liquid adsorbing agents at first and then the absorption of target chemical is relieved using eluent. The method can isolate target chemicals effectively (Behbahani et al., 2013).

## 2.3. Sample processing and condition optimization

Before HPLC and UPLC, the samples were pre-processed according to the national standard (Chang et al., 2013). The samples were scanned when the detection wavelength of HPLC was set as 500 nm and 20 mmol of ammonium acetate solution (A), acetonitrile (B) and methyl alcohol (C) were taken as flow phase and when the detection wavelength of UPLC was set as 260, 312, 300, 400, 427, 500, 540 and 634 nm respectively and 20 mmol of ammonium acetate solution (A) and methyl alcohol (B) were taken as flow phase. The set of elution procedures are shown in Table 1.

**Table 1.** Gradient elution procedures

| | | HPLC | | | UPLC | |
|---|---|---|---|---|---|---|
| Time | Flow speed mL/min | A Ammonium acetate (%) | B Acetonitrile (%) | C Methyl alcohol (%) | A Ammonium acetate (%) | B Methyl alcohol (%) |
| 0 | 1 | 90 | 0 | 10 | 90 | 10 |
| 8 | 1 | 75 | 6 | 24 | 75 | 25 |
| 11 | 1 | 70 | 5 | 25 | 70 | 30 |
| 13 | 1 | 43 | 0 | 57 | 68 | 32 |
| 30 | 1 | 38 | 4 | 59 | 55 | 45 |
| 38 | 1 | 18 | 2 | 80 | 0 | 80 |
| 42 | 1 | 18 | 2 | 80 | 0 | 100 |
| 50 | 1 | 90 | 0 | 10 | 90 | 10 |

## 2.4. Two liquid chromatography methods

### *2.4.1 The detection of colorants using UPLC*

The working principle of HPLC (Nugroho, 2011) is basically the same with that of general traditional detection methods; its characteristic lies on the application of infusion pump with relatively large pressure. In this study, the commonly seen sports drinks were taken as research subjects; five kinds of colorants, i.e., acid orange 10, acid red 1, acid red 14, acid black 1 and acid yellow 2 were detected using HPLC.

### *2.4.2 The detection of colorants using HPLC*

Compared to HPLC, UPLC (Hung et al., 2011) can analyze compounds which are finite or exist in complex matrix, thus to achieve better isolation effect; the isolation of substances which is difficult to be realized using high performance liquid chromatograph in short time can be realized using ultra-high performance liquid chromatograph; besides, UPLC improves resolution and sensitivity, shortens time taken for analysis, reduces the dose of solvent and lowers analysis cost, which can effectively save cost and realize the optimization of time benefit.

## 3. Results and discussions

### 3.1. Chromatogram of HPLC and UPLC

The subjects of this study were all polar substances which are soluble in water or methyl alcohol. Therefore, reversed-phase chromatography was selected for separation and detection. A spectrogram was obtained based on the above chromatographic conditions, as shown in Figure 1.

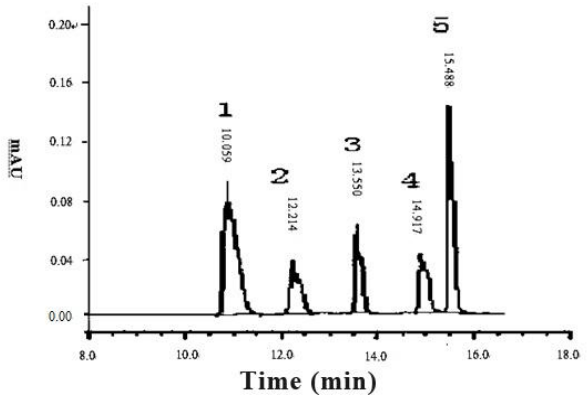


**Figure 1.** High performance liquid chromatograph of samples

As shown in Figure 1, 1 represents acid orange 10, 2 represents acid red 1, 3 represents acid red 14, 4 represents acid black 1, and 5 represents acid orange 2.

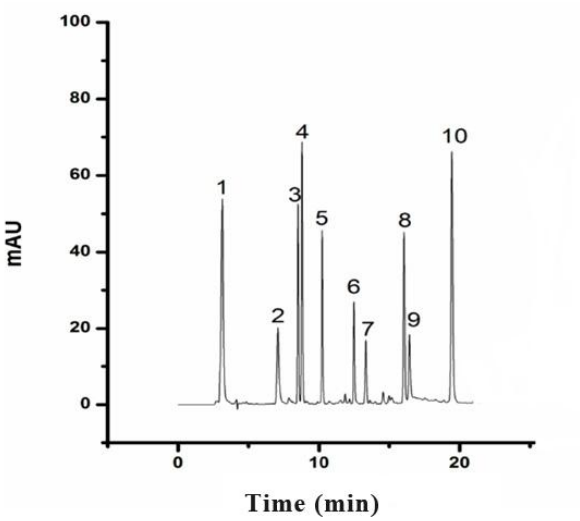


**Figure 2.** Ultra-high performance liquid chromatograph of samples

As shown in Figure 2, 1 represents acid yellow 23, 2 represents acid red 18, 3 represents acid purple 7, 4 represents acid orange 10, 5 represents acid red 35, 6 represents acid red 14, 7 represents acid green 5, 8 represents acid red 73, 9 represents acid black 1, and 10 represents acid orange 7. The experimental results demonstrated that, UPLC was applicable to the quantitative and qualitative detection and analysis of drink as it could detect multiple kinds of colorants.

### 3.2. Detection limit and linear range of colorants

HPLC: Five kinds of colorants were diluted into 5.0 mg/L, 10 mg/L and 100.0 mg/mL respectively using methanol solution. Then a coordinate was established, taking mass concentration (mg/ml) as the horizontal coordinate and peak area as the vertical coordinate. A standard curve was drawn after relevant data were substituted; then the linear regression equation was calculated. The correlation coefficients and detection items are shown in Table 2.

**Table 2.** Linear regression equations, correlation coefficients and detection limits of five kinds of colorants

| Colorants | Linear range (mg/L) | Linear equation | Correlation coefficient | Detection limit (mg/kg) |
|---|---|---|---|---|
| Acid orange 10 | 5~100 | y=19.7785x-3.6885 | 0.99995 | 0.016 |
| Acid red 1 | 5~100 | y=14.3568x-8.6271 | 0.99988 | 0.016 |
| Acid red 14 | 5~100 | y=18.8842x-12.1968 | 0.99995 | 0.061 |
| Acid black 1 | 5~100 | y=12.6795x-21.9655 | 0.99982 | 0.007 |
| Acid yellow 2 | 5~100 | y=41.5095x-9.8566 | 0.99996 | 0.092 |

UPLC: Ten kinds of colorants were diluted into 5.0 mg/L, 10 mg/L, 20 mg/L, 40 mg/L and 80 mg/L respectively using methanol solution. A coordinate was established, taking mass concentration (mg/mL) as the horizontal coordinate and peak area as the vertical coordinate. A standard curve was drawn after relevant data were substituted. The results are shown in Table 3.

**Table 3.** Linear regression equations, correlation coefficients and detection limits of 10 kinds of colorants

| Colorants | Linear range (mg/L) | Linear equation | Correlation coefficient R |
|---|---|---|---|
| Acid yellow 23 | 5~50 | y=25.0556x+33.2477 | 0.99096 |
| Acid red 18 | 5~50 | y=11.8875x-17.0491 | 0.99468 |
| Acid purple 7 | 5~50 | y=21.0102x-2.7975 | 0.99993 |
| Acid orange 10 | 5~50 | y=19.2741x-6.3333 | 0.99973 |
| Acid red 35 | 5~50 | y=5.51136x-2.0595 | 0.99952 |
| Acid red 14 | 5~50 | y=18.9478x-4.9321 | 0.99951 |
| Acid green 5 | 5~50 | y=68.1438x-2.4865 | 0.99968 |
| Acid red 73 | 5~50 | y=28.0635x+3.8665 | 0.99963 |
| Acid black 1 | 5~50 | y=20.3515x-32.1906 | 0.99536 |
| Acid orange 7 | 5~50 | y=39.2795x+2.4155 | 0.99983 |

### 3.3. Preciseness and recovery test

To test the preciseness of HPLC, mixed standard solutions made of five kinds of colorants (0.05, 0.1 and 0.2 mg/L) were added into blank drink samples respectively. The detection was performed according to the test method. After six times of parallel detection, the average value was calculated. The results are shown in Table 4. Results demonstrated that, the average recovery rates were larger than 83.5% and the relative standard deviations (RSD) were smaller than 1.55%, suggesting HPLC was accurate and reliable. Seven kinds of acid industrial dyes were extracted using alkalescence solvent. The experimental results demonstrated that, the recovery rate was high. Besides, a detection and analysis method which could be used for detecting seven kinds of acid industrial dyes in functional drink, i.e., HPLC, was set up. The method was simple, high-efficient and highly sensitive and moreover the RSD and recovery rate satisfied the requirements of relevant departments.

**Table 4:** The preciseness of adding standard recovery rate of HPLC

| Component | Adding standard matter amount (mg/L) | Measured value (mg/L) | Relative standard deviation (%) | Recovery rate (%) |
|---|---|---|---|---|
| Acid orange 10 | 0.05 | 0.0416 | 0.33 | 83.6 |
| | 0.1 | 0.0806 | 1.58 | 80.4 |
| | 0.2 | 0.1733 | 0.26 | 86.6 |
| Acid red 1 | 0.05 | 0.0405 | 0.45 | 81.3 |
| | 0.1 | 0.0829 | 0.27 | 82.6 |
| | 0.2 | 0.1765 | 0.99 | 88.1 |
| Acid red 14 | 0.05 | 0.0415 | 0.32 | 82.6 |
| | 0.1 | 0.0918 | 0.58 | 91.5 |
| | 0.2 | 0.1964 | 0.40 | 98.3 |
| Acid black 1 | 0.05 | 0.0407 | 0.52 | 81.1 |
| | 0.1 | 0.0901 | 1.25 | 89.8 |
| | 0.2 | 0.1914 | 0.33 | 95.7 |
| Acid orange 2 | 0.05 | 0.0416 | 1.03 | 83.3 |
| | 0.1 | 0.0903 | 0.41 | 90.3 |
| | 0.2 | 0.1925 | 0.30 | 96.1 |

In the process of new product development, effects of the concentration of organic solvent, the category of buffer solution, the concentration of buffer salt, the acid value of flow phase, controlled temperature and the PH value of flow phase on products should be paid attention to.

In UPLC test, mixed standard solutions made of five kinds of colorants (5.0, 10.0 and 20.0 mg/L) were added into blank drink samples respectively. Parallel detection was performed for 6 times. The results are shown in Table 5.

**Table 5.** The preciseness of adding standard recovery rate of UPLC

| Component | Adding standard matter amount (mg/L) | Measured value (mg/L) | Relative standard deviation (%) | Recovery rate (%) |
|---|---|---|---|---|
| Acid yellow 23 | 5 | 5.24 | 3.26 | 106.5 |
| | 10 | 10.19 | 3.42 | 101.9 |
| | 20 | 22.76 | 0.89 | 113.6 |
| Acid red 18 | 5 | 4.88 | 1.86 | 102.5 |
| | 10 | 10.87 | 1.92 | 108.6 |
| | 20 | 19.75 | 1.51 | 98.8 |
| Acid purple 7 | 5 | 5.026 | 1.72 | 100.5 |
| | 10 | 10.4 | 1.51 | 100.1 |
| | 20 | 19.72 | 1.45 | 98.6 |
| Acid orange 10 | 5 | 4.88 | 2.24 | 97.5 |
| | 10 | 9.88 | 2.36 | 98.6 |
| | 20 | 19.16 | 4.01 | 95.8 |
| Acid red 35 | 5 | 5.01 | 2.09 | 100.2 |
| | 10 | 10.68 | 2.55 | 106.8 |

| | | | | |
|---|---|---|---|---|
| | 20 | 19.58 | 3.77 | 98.0 |
| Acid red 14 | 5 | 4.73 | 3.38 | 94.3 |
| | 10 | 9.75 | 1.59 | 97.5 |
| | 20 | 17.51 | 5.18 | 87.6 |
| Acid green 5 | 5 | 4.41 | 1.99 | 87.9 |
| | 10 | 9.43 | 1.02 | 94.3 |
| | 20 | 17.42 | 4.42 | 87.2 |
| Acid red 73 | 5 | 4.88 | 1.71 | 97.5 |
| | 10 | 9.91 | 1.38 | 98.8 |
| | 20 | 19.56 | 3.62 | 97.8 |
| Acid black 1 | 5 | 5.06 | 2.06 | 100.9 |
| | 10 | 10.55 | 7.51 | 105.2 |
| | 20 | 20.81 | 1.05 | 103.8 |
| Acid orange 7 | 5 | 4.33 | 2.66 | 96.5 |
| | 10 | 9.39 | 0.82 | 93.9 |
| | 20 | 19.16 | 1.32 | 95.8 |

Results demonstrated that, the average recovery rates were larger than 89.8% and the RSD was between 1.50% and 3.58% (n = 4), suggesting UPLC was accurate and reliable. With the assistance of ultrasonic wave, 10 kinds of acid industrial colorants were extracted. Through optimization, the detection of sports drink became simpler. Compared to HPLC, UPLC is featured by simple operation, high efficacy, high sensitivity, high recovery rate and good repeatability, which provides relevant departments with a technical support in the aspect of the detection of sports drink.

## 4. Conclusions

In this study, HPLC and UPLC were applied to detect and analyze the artificially synthesized colorants in sports drink. The research results suggested that, the number of categories of synthesized colorants detected out by UPLC was more than that by HPLC; detecting colorants with UPLC was more sensitive than HPLC; moreover, the average recovery rate of samples detected by UPLC was larger. Thus, it can be concluded that, UPLC is more suitable for the detection and analysis of artificially synthesized colorants in sports drink. UPLC makes up the deficiency of colorant detection and provides relevant departments with a technical support in the aspect of colorants detection in sports drink, which can ensure the personal safety interest of consumers.

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