

Will Invigorate Chinese Medicine Influence Amount of Superoxide Dismutase in the Hearts of the Mice?

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Abstract

The purpose of this study is to test whether invigorate Chinese medicine is able to reduce the superoxide dismutase inside the organisms' bodies. In the study, the mice are divided into 4 groups, and the variations of the experiment are taking in the invigorate medicine or not and experiencing strenuous motions or staying quiescent. The mice that take in the invigorate medicine have more, less or similar amount of superoxide dismutase inside their hearts compared to mice that do not take in the invigorate medicine. In addition, they are both under strenuous motions. The study is able to determine whether invigorate Chinese medicine is able to reduce the superoxide dismutase inside organisms that experience strenuous motions.

Keywords

Invigorate Chinese Medicine, Superoxide Dismutase, Strenuous Motion, Quiescent Condition.

1. INTRODUCTION

As the pace of life getting faster, men, especially young men, are consuming invigorate medicine to stimulate their sex behavior as pressure has reduced the sex drive. In addition, people have assumed that invigorate medicine can not only improve sex conditions but also improve the biological endurances. In that case, the invigorate medicine can be used to relieve the fatigue of athletes.

People who have a yang deficiency take invigorate medicine, and the external factors that cause yang deficiency are deficiency of sunlight, low temperatures and exposure to air-conditioners [1]. Under those circumstances, the small arteries and veins of the whole body skin contract and cramp, which causes the diameter of the blood vessels to narrow. When the diameter of the blood vessels narrows to a certain degree, the blood viscosity starts to increase sharply, and the blood flow rate slows down, resulting in microcirculation obstacles. [1] Since Chinese medicine serves to reduce the microcirculation obstacles, the essence behind it is that some components in the medicines are able to reduce the free radicals inside human's bodies; at the same time, components that contain element magnesium and zinc can prevent oxidation in the cells.

Among those invigorate medicine are pilose antler and cinnamon. [2] Pilose antler is a deer sika or red deer stag's dense velvet hairy horn without ossification [3], and cinnamon is the dried bark of Cinnamon cassia Presl [4]. In cinnamon, a matter called cinnamaldehyde is responsible for the invigorate effect.[5] In pilose antler, a matter called pilose antler blood is responsible for the invigorate effect[4], and the dominant chemical composition of pilose antler is essential amino acid and phosphatide[3]. In Chinese medicine, pharmacists frequently use compounds—the mixture of two or more kinds of Chinese medicine—to cure diseases, so a typical kind of invigorate medicine contains pilose antler and cinnamon[2]. With the material, researchers are able to develop a compound invigorate medicine to use in studies.

In the experiment, to justify whether the compound medicine that contains pilose antler and cinnamon is able to increase the biological endurance, researchers select 96 male mice in healthy and similar conditions and divide the mice into two groups. In one group, the researchers let the 48 mice take in a controlled amount of compound; in the other group, the researchers let the other 48 mice take in the same amount of water as the first group. Then, the mice are trained in the same condition for one week. After one week, the researchers divides the mice again into 4 groups—the first group is that mice that took in the compound under a quiescent condition; the second group is that mice that took in the compound under a strenuous exercise condition; the third group is that mice that took in water under a quiescent condition and the fourth group is that mice that took in water under a strenuous exercise condition. When the quiescent or strenuous exercise conditions are over, the mice are all killed immediately. As superoxide dismutase is a kind of anti-oxidize enzyme, if the amount of superoxide dismutase increases, the amount of free radicals will reduce, so the researchers extract the hearts of mice to examine the amount of superoxide dismutase in the hearts of mice. Eventually, the researchers find that the second group's mice's hearts have a higher amount of superoxide dismutase than the fourth group, but the first group's mice's amount of superoxide dismutase does not have much difference with that of the third group.[2]

Under the circumstances of the research, the conclusion is that when a living organism is under exercise condition, taking in a controlled amount of compound invigorate Chinese medicine is able to increase the amount of superoxide dismutase and decrease the amount of free radicals to improve the organism's endurance. But if the organism is under a quiescent condition, taking in the compound does not make a difference in the amount of superoxide dismutase. In that case, I assume that taking in the compound when the organism is under quiescent conditions will not improve the organism's endurance, but has a negative effect on the organism instead. Then, it can serve as a warning to those people who take in the compound that the compound will have a negative effect on their body when they take the drug but do not exercise.

2. CHEMICALS, MEDICAL MATERIALS AND ANIMALS

2.1. Chemicals

Superoxide dismutase is one kind of enzyme that resists the oxidization of metal ions inside the organisms' bodies, and there are two most common types of superoxide dismutase.

Cu/Zn-SOD	The hub of the molecule is a Cu ion and a Zn ion. The Cu ion bonds with four histidines' nitrogen atom and forms a quadrilateral, and the Zn ion bonds with three histidines' nitrogen atom and shares a bond with the Cu ion.
Mn-SOD	The hub of the molecule is a Mn ion, and the molecule consists of 203 amino acid residues.

2.2. Medical Materials

The materials to make compound invigorate Chinese medicine is antler pilose and cinnamon. The antler pilose as well as cinnamon are all shattered and dried, and after the previous process, the powder of antler pilose and cinnamon is blended with water and boiled for 4 hours. After the boiling process, the compound is cooled to room temperature. [2]

2.3. Animals

96 one-year-old male mice that are in similar health conditions are chosen as the experimental animals. They are fed under national-standard fodder, and the mice are beheaded

immediately after they finish the process in the experiment. After being killed, the mice's hearts are extracted. [2]

3. PROCESS OF THE STUDY

3.1. Dealing of the Mice Throughout the Whole Experiment

(1) The mice are divided into 4 groups. The first group and the second group are taking the invigorate compounds, and the third as well as fourth group are taking the same amount of clear water. The compounds are taken in by the first and second group at 9:00 a.m. and 6:00 p.m. every day for a whole week.[2] In addition, all the mice take in the same type and amount of national-standard forage every day for a whole week.

(2) Despite of taking in matters, the mice are trained during the same week. The mice are each put on an electronic runway and they will run for 15 minutes once and run 6 times throughout the day. The rest interval is controlled and it's 45 minutes. [2]The same training is happening every day during the experimental week.

(3) After a whole week, the mice are asked to stay under different conditions——the first group and the third group are put on the electronic runways and the speed of runways is raised 2 times than that of the previous week, while the second group and the fourth group are put under quiescent condition——they are confined in the cages, which doesn't allow them to move fast.

(4) When the first and third group of mice are stressed out, they are stopped and beheaded. The standard to judge whether the mice are stressed out is that when they respire much more frequently than they usually do, or they are blunt to stimuli.[2] The second and fourth group are beheaded at the time the as the first group and the third group.

3.2. Methods to examine the effect of invigorate Chinese medicine

(1) When the mice are beheaded, their hearts are extracted immediately and washed with physiological saline solution. After the dealing, they are frozen. In addition, the hearts are frozen by groups.

(2) The hearts are then extracted and the amount of superoxide dismutase is measured.

The basis of superoxide dismutase measurement is as following:[6]

Step 1: the hearts of mice are shattered in a machine.

Step 2: the shattered hearts are blended with phosphate buffer with a pH of 7.8, a density of 0.05 mol/L, and the buffer's volume is 3 times larger than that of shattered hearts.

Step 3: the mixture of hearts and buffer are put inside a centrifugal machine and they experience centrifugal movement for 15 minutes under room temperature.

Step 4: the clear solution after the centrifugal movement is extracted.

Step 5: the solution is blended with trichloromethane and ethanol solution with a volume quarter to the clear solution get in step 4, and then the new mixture is blended and it experiences a centrifugal movement for 15 minutes again. After the movement, the solution is put under a western blotting process to separate the SOD with other irrelevant proteins. Specifically speaking, the process is called polyacrylamide gel electrophoresis, which is able to select the molecule like SOD in this experiment. First of all, after protein electrophoresis separation, the protein on the gel is transferred to the solid support under the action of an electric field. Then, it is time to place the solid support in the blocking solution to block it and cover its non-specific sites. Last but not least, the in-situ enzyme color reaction is taken out to measure the amount of SOD in the solutions.[7]

Step 6: the four groups of solution's color reactions are measured to indicate the amount of superoxide dismutase, and if the amount of superoxide dismutase is higher, the color that represents superoxide dismutase is denser.

3.3. Possible Results and Analysis

Mice in group 1 take in the invigorate Chinese medicine compound and experience strenuous motions.

Mice in group 2 take in the invigorate Chinese medicine compound and stay quiescent.

Mice in group 3 take in the same amount of water and experience strenuous motions.

Mice in group 4 take in the same amount of water and stay quiescent.

3.3.1 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent. Mice that take in the invigorate Chinese medicine compound and stay quiescent have a larger amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions.

Analysis: Strenuous motions cause the rise of SOD in hearts of mice and the invigorate Chinese medicine is not able to reduce the amount of SOD in hearts of mice. If the mice take in the medicine when quiescent, it will not reduce the amount of SOD but increase the amount of SOD.

3.3.2 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have a larger amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent.

Analysis: Strenuous motions cause the rise of SOD in hearts of mice and the invigorate Chinese medicine is not able to reduce the amount of SOD in hearts of mice. If the mice is quiescent, the lower amount of SOD can be interpreted as the mice are not moving or the invigorate medicine reduces the amount of SOD only when the mice are quiescent.

3.3.3 Mice that take in the invigorate Chinese medicine compound and stay quiescent have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions. Mice that take in the invigorate Chinese medicine compound and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the same amount of water and experience strenuous motions.

Analysis: The amount of SOD will be higher when the mice do not move strenuously after they take the invigorate medicine, and the medicine will not reduce the amount of SOD in the moving mice's hearts, it will increase the amount instead.

3.3.4 Mice that take in the invigorate Chinese medicine compound and stay quiescent have a larger amount of SOD in the solution than mice that take in the same amount of water and experience strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions.

Analysis: The amount of SOD will be higher when the mice do not move strenuously after they take the invigorate medicine, and the medicine will effectively reduce the amount of SOD in the moving

3.3.5 Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions. Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent.

Analysis: The invigorate medicine can effectively reduce the amount of SOD in moving mice's hearts, and no matter whether the mice take in the invigorate Chinese medicine or not, the mice under quiescent conditions produce lower amount of SOD.

3.3.6 Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent. Mice that take in the invigorate Chinese medicine compound and stay quiescent have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions.

Analysis: The invigorate medicine can effectively reduce the amount of SOD in moving mice's hearts, and the effect is so strong that it can even reduce the SOD amount of mice to a lower level than quiescent ones.

3.3.7 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have roughly the same amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent. Mice that take in the invigorate Chinese medicine compound and stay quiescent have a larger amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions.

Analysis: The invigorate medicine has no apparent effect on the reduction of SOD when the mice are under strenuous conditions. In addition, if the mice stay quiescent, they have more SOD in the hearts than moving ones.

3.3.8 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have roughly the same amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent. Mice that take in the invigorate Chinese medicine compound and stay quiescent have a smaller amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions.

Analysis: The invigorate medicine has no apparent effect on the reduction of SOD when the mice are under strenuous conditions and if the mice stay quiescent, they have less SOD in the hearts than moving ones.

3.3.9 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have roughly the same amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent. Mice that take in the invigorate Chinese medicine compound and stay quiescent have roughly the same amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions.

Analysis: The amount of SOD in the mice's hearts is either influenced by the invigorate medicine or the movement conditions of the mice.

3.3.10 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have roughly the same amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent.

Analysis: The invigorate medicine has no apparent effect on the reduction of SOD when the mice are under strenuous conditions. When the mice are quiescent, the taking in of invigorate medicine is able to reduce the amount of SOD in the hearts.

3.3.11 Mice that take in the invigorate Chinese medicine compound and experiences strenuous motions have roughly the same amount of SOD in the solution than mice that take in the same amount of water and experiences strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a smaller amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and stay quiescent.

Analysis: The invigorate medicine has no apparent effect on the reduction of SOD when the mice are under strenuous conditions. When the mice are quiescent, the taking in of invigorate medicine increases the amount of SOD in the hearts instead of reducing it.

3.3.12 Mice that take in the invigorate Chinese medicine compound and stay quiescent have roughly the same amount of SOD in the solution than mice that take in the same amount of water and experience strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a larger amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions.

Analysis: Both moving and taking in the invigorate medicine have an overall effect on the mice that they will increase the amount of SOD in the hearts.

3.3.13 Mice that take in the invigorate Chinese medicine compound and stay quiescent have roughly the same amount of SOD in the solution than mice that take in the same amount of water and experience strenuous motions. Mice that take in the same amount of water and experience strenuous motions have a smaller amount of SOD in the solution than mice that take in the invigorate Chinese medicine compound and experience strenuous motions.

Analysis: Both moving and taking in the invigorate medicine have an overall effect on the mice that they will reduce the amount of SOD in the hearts.

3.4. Most Possible Results

Mice in group 1 take in the invigorate Chinese medicine compound and experience strenuous motions.

Mice in group 2 take in the invigorate Chinese medicine compound and stay quiescent.

Mice in group 3 take in the same amount of water and experience strenuous motions.

Mice in group 4 take in the same amount of water and stay quiescent.

3.4.1 Comparison between group 3 and 4

(1) Group3's amount of superoxide dismutase is lower than that of group 4.

(2) Group3's amount of superoxide dismutase is higher than that of group 4.

(3) Group3's amount of superoxide dismutase roughly equals to that of group 4.

The aim of this comparison is to prove that under strenuous motions, the mice's amount of superoxide dismutase is apparently larger than that of quiescent mice, so the predicted result should be (2).

3.4.2 Comparison between group 1 and 2

(1) Group1's amount of superoxide dismutase is lower than that of group 2.

(2) Group1's amount of superoxide dismutase is higher than that of group 2.

(3) Group1's amount of superoxide dismutase roughly equals to that of group 2.

The aim of this comparison is to prove that even if the mice groups both take in invigorate Chinese medicine, the group under strenuous motions has a higher amount of superoxide dismutase than that of the quiescent group. In that case, the result should form (2).

Otherwise, if the result form (1), the assumption that taking in invigorate medicine when staying still will increase the amount of superoxide dismutase is made.

3.4.3 Comparison between group 1 and 3

(1) Group1's amount of superoxide dismutase is lower than that of group 3.

Analysis: The result indicates the invigorate Chinese medicine can reduce the superoxide dismutase inside the mice's bodies when the mice are in strenuous motions.

(2) Group1's amount of superoxide dismutase is higher than that of group 3.

Analysis: The result indicates the invigorate Chinese medicine cannot reduce the superoxide dismutase inside the mice's bodies when the mice experience strenuous motions, but has a opposite effect by increasing the superoxide dismutase inside the mice's bodies instead.

(3) Group1's amount of superoxide dismutase roughly equals to that of group 3.

Analysis: The result indicates the invigorate Chinese medicine have no apparent effect on the reduction of superoxide dismutase inside the mice's bodies when they are under strenuous motions.

4. DISCUSSION AND CONCLUSION

The expected results do lead to the conclusion that invigorate Chinese medicine is able to reduce the amount of superoxide dismutase, but it is not enough to just focus on the positive effect of invigorate medicine within this experiment.

First of all, the amount of invigorate medicine is controlled, so there aren't any effects caused by overuse of medicine in the experiment. However, in daily lives people sometimes overuse medicine, and that might cause some side effects, so the damage caused by overuse of this medicine is expected to be studied and relevant experiments will be taken out.

Secondly, when some people are quiescent, maybe there will be side effects as well. On one hand, as I mentioned in the results of comparison 2, the medicine might not decrease the amount of superoxide dismutase inside the bodies but increase it instead. Then, the opposite effect may make people stressed out even if they do not move frequently. On the other hand, negative effects other than increasing superoxide dismutase might occur when people take in the invigorate Chinese medicine.

Last but not least, in the experiment, the chosen types of invigorate Chinese medicine are cinnamons and pilose antler, and they are representative invigorate medicine. However, there are other invigorate Chinese medicine categories, so experiments that contain other types of medicine should also be taken out in the future.

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