Study in the Effect of Octacosanol on Athletic Ability

Yunpeng Tang

Guizhou University of Commerce, Guiyang 550014, China

Abstract

In this paper, four hypoxic animal models of atmospheric confined hypoxia (hypoxic hypoxia), sodium nitrite poisoning (blood hypoxia), cerebral ischemia hypoxia and myocardial hypoxia were established through experiments. The effects of octacosanol on the survival time of different types of acute hypoxia rats were observed. On the basis of this its effective dose was analyzed and its possible mechanism was discussed.

Keywords: octacosanol, athletic ability, hypoxia rats.

1. INTRODUCTION

After plains human enter in the plateau hypoxia environment, the ability to work and work efficiency decrease, some people may get altitude sickness due to poor acclimatization, serious and even life-threatening. Therefore, to find effective hypoxia acclimatization measures to improve the quality of life and labor capacity in the environment of plateau hypoxia as well as to promote its hypoxic acclimatization process, reduce the incidence of high altitude disease and mortality, has become the focus and frontier of plateau medicine Research. The use of drugs to promote plateau acclimatization, prevention and treatment of high altitude disease is simple, high feasibility, the relevant personnel has been committed to finding of anti-hypoxia drugs. Acetaminophen dexamethasone, theophylline and other drugs have mitigation effect on the plateau hypoxia symptoms, but the bigger side effects restrict their actual use. Anti-hypoxia food additives main component of which are Wolfberry fruit, Ginkgo biloba, ginseng and other raw materials for food and medicine can significantly prolong the survival time of hypoxia animals, reduce the incidence of acute plateau disease into the plateau, improve the ability of high altitude labor, but for the reason of less and expensive resources, which is not conducive to its promotion and use (Gonzalez et al., 1994).

Octacosanol is a naturally existing higher fatty alcohol widely distributed in the epidermis and viscera of animals, waxes secreted by insects, and lipids of plant roots, stems, leaves, shells and seed kernels, with a wide range of sources and without side effects, as a natural health food additives and broad-spectrum natural medicine, has been used for sports drinks, natural health products, medicines and cosmetics, and other fields. Studies have shown that octacosanol has a variety of physiological functions, including anti-sports fatigue, increased endurance, energy and physical strength, reduce blood fat, prevent cardiovascular disease, anti-platelet aggregation (Feng et al., 2005), cell protection and heart and brain ischemia protection. The results suggested that octacosanol could be an effective hypoxia-tolerant food. However, there has been no report on its anti-hypoxia effect in China and abroad. Therefore, this paper studies anti-anoxia effect of octacosanol, and to explore its mechanism of action (Zheng et al., 2012).

2. EFFECT OF OCTACOSANOL ON THE SURVIVAL TIME OF ACUTE HYPOXIA RATS

2.1 Experimental animals

124 Kunming male rats of clean grade, 6-7 weeks old, weighing 20±2g, were provided by the Experimental Animal Center of the Third Military Medical University. The laboratory animal license number is SYXK 2002-029, fed with ordinary fodder.

2.2 The main reagents and preparation

(1) Edible Oil: Chongqing Jiangbei Oil Grease Company north-river oil refineries produced;
2. Octacosanol: provided by Jiangsu Provincial Key Laboratory of Medicinal Plant Biotechnology, purity> 98%, prepared with edible oil, obtained concentration of 0.1% (m / v), 0.5% (m / v), 1% (m / v) of the suspension, 4 °C refrigerator reserved for use, fully oscillation mixed before use (Che, 2014);

3. Sodium Lime: Batch number 20011001, Chongqing Pengcheng Industrial Company calcification plant produced;

4. Vaseline: Batch number 900512, produced by Wuhan Petrochemical Plant (Dong, 2013);

5. Sodium nitrite: Batch number 20060317, Chengdu Kelon Chemical Reagent Factory produced, with double distilled water prepared into a concentration of 2% (m/v) solution, stored at room temperature for use;


2.3 The main instruments

1. Electronic balance: instrument number 02502, Shanghai Precision Science Instrument Co., Ltd.

2. Stopwatch: domestic.

2.4 The main indicators and experimental methods

1. Confined hypoxia experiment

52 male Kunming rats were randomly divided into experimental group A, experimental group B, experimental group C and control group, 13 rats in each group. In group A, 0.1% (m/v) octacosanol suspension was administered by 10mg·kg⁻¹, while in group B, 0.5% (m/v) octacosanol suspension was administered by 50mg·kg⁻¹. The rats in experimental group C were administered with 1% (m/v) octacosanol suspension at 100mg·kg⁻¹, and the control group was given 10mg·kg⁻¹ edible oil by intragastric administration. Continuous feeding 7d, free diet during the period, the 6th day of administration began to fast, free water. Body weight was measured before intragastric administration daily, and the feeding and drinking of rats were observed. After 30 min of administration on the 7th day, mice in each group were placed in a 150ml jar containing 10 g of sodium lime (all revised by capacity), each rat with one bottle, sealed with vaseline and immediately started timing. Using the last breath as the index, the rats were observed from the beginning of the sealed bottle to the apnea at when the breathing stopped (Li and Zhou, 2014).

2. Sodium nitrite poisoning experiment

24 male Kunming rat were randomly divided into control group and drug group, 12 rats in each group. The rats in the control group were fed with edible Oil at 50mg·kg⁻¹ and the drug group were administered with 0.5% (m/v) octacosanol suspension at dose of 10mg·kg⁻¹. Continuous feeding 7d, free diet during the period, the 6th day of administration began to fast, free water (Khan et al., 2010). 2h after the last administration conducted sodium nitrite poisoning experiments, the survival time of rats were observed after intraperitoneal injection of 2% (m/v) sodium nitrite solution at 20 mg·kg⁻¹.

3. Cerebral ischemia hypoxia experiment

Selected 24 healthy Kunming male rats, animal grouping, intragastric administration time and dose are all in line with sodium nitrite poisoning experiment. 2 hours after the last administration, the rats were killed with a pair of scissors by cutting head at the connection of binaural, and the duration of the mouth opening was observed.

4. Myocardial hypoxia experiment

Selected 24 healthy Kunming male rats, animal grouping, intragastric administration time and dose are all in line with sodium nitrite poisoning experiment. After 2 hours of the last administration, isoproterenol hydrochloride was injected intraperitoneally at 15mg·kg⁻¹. After 15min, the rats were placed in 5g sodium lime jars. The confined hypoxia experiment was conducted, the time of first respiration and the time of death were observed.
2.5 Results analysis

(1) Effects of octacosanol on the survival time of confined hypoxia of rats

The standard tolerance time of confined hypoxia in experimental group A and experimental group B was significantly higher than that in the control group (P < 0.01). There was no significant difference in the standard tolerance time between the experimental group A and the experimental group B (P > 0.05). (As shown in Figure 1, Table 1)

Table 1 Effects of octacosanol on the survival time of confined hypoxia of rats

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Standard tolerance time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>13</td>
<td>8.2±1.32</td>
</tr>
<tr>
<td>Experimental group A</td>
<td>13</td>
<td>10.22±1.52</td>
</tr>
<tr>
<td>Experimental group B</td>
<td>13</td>
<td>10.62±2.01</td>
</tr>
<tr>
<td>Experimental group C</td>
<td>13</td>
<td>8.15±1.52</td>
</tr>
</tbody>
</table>

![Figure 1. Effects of octacosanol on the survival time of confined hypoxia of rats]

(2) Effects of octacosanol on the sodium nitrite poisoning of rats

There was no significant difference in the survival time between the drug group and the control group (p> 0.05) after injection of sodium nitrite. (As shown in Table 2).

Table 2 Effects of octacosanol on the sodium nitrite poisoning of rats

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Survival time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>12</td>
<td>11.25±2.01</td>
</tr>
<tr>
<td>Drug group</td>
<td>12</td>
<td>10.36±1.14</td>
</tr>
</tbody>
</table>

(3) Effects of octacosanol on the survival time of cerebral ischemia hypoxia of rats

The duration of mouth opening in the rats given decapitation was significantly higher than that in the control group (P < 0.05). (As shown in Table 3).

Table 3 Effects of octacosanol on the survival time of cerebral ischemia hypoxia of rats

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Survival time of open mouth</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>12</td>
<td>18.25±2.01</td>
</tr>
<tr>
<td>Drug group</td>
<td>12</td>
<td>21.45±3.25</td>
</tr>
</tbody>
</table>

(4) Effects of octacosanol on the survival time of myocardial hypoxia of rats
The standard tolerance time of rats in the drug group after injection of isoproterenol was significantly higher than that in the control group (p < 0.05). (As shown in Figure 2, Table 4).

### Table 4 Effects of octacosanol on the survival time of myocardial hypoxia of rats

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Standard tolerance time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>12</td>
<td>7.25±0.25</td>
</tr>
<tr>
<td>Drug group</td>
<td>12</td>
<td>8.25±2.36</td>
</tr>
</tbody>
</table>

![Figure 2. Effects of octacosanol on the survival time of myocardial hypoxia of rats](image)

From the above experimental analysis can be obtained:

1. Octacosanol can significantly enhance the ability of hypoxia tolerance in rats, significantly prolonged the survival time of rats in atmospheric confined hypoxia environment;

2. The anti-hypoxic effect of octacosanol is not related to improving the oxygen transport efficiency of the organism, and its mechanism may be related to the regulation of metabolic and cytoprotective effects;

3. Octacosanol can reduce brain energy consumption, improve myocardial ischemia.

### 3. EFFECTS OF OCTACOSANOL ON CARDIAC FUNCTION IN CHRONIC HYPOXIA RATS

#### 3.1 The main indicators and experimental methods

1. **Experimental grouping**

   78 adult male SD rats of clean grade were randomly divided into control group (n=38) and drug group (n=40). The drug group were given intragastric administration of 0.5% (m/v) octacosanol suspension at a dose of 5 mg·kg\(^{-1}\) for 30 days, once a day, the control group were fed with 10 mL·kg\(^{-1}\) edible oil. During the period of intragastric administration, the rats in both groups were placed in simulated decompression chamber of 5000m altitude and depressurized for 23h/d. Every day put into the plain 1h for cleaning, adding water, adding food, measuring body weight and intragastric administration. 0.5h after the last administration, simulating 5000m highland environment, to conducted experiments.

2. **Animal anesthesia and surgery**

   Animals were anesthetized by intraperitoneal injection of 10% (m/v) urethane at 1.5mg·kg·1. After anesthesia was stabilized, the animals were supine fixed on the small animal operating table. Took the neck median incision, the left common carotid artery and the right external jugular vein were separated. The intubation was used to detect the left and right cardiac function.

3. **Arterial blood gas and lactic acid content detection**
0.1 ml arterial blood samples were catheter collected from the left carotid artery with 1000IU/ml heparin wetted injector, the air bubbles were removed, the nozzle was closed with a rubber stopper, the blood oxygen partial pressure (P02), oxygen saturation S02 and blood lactic acid (Lac) were measured by i-STAT blood gas analyzer.

(4) Blood routine determination

20 μl blood was took, 1 ml blood dilution was added, and blood routine indicators were measured on KX-21 three-class blood analyzer.

(5) Hemodynamic indicators determination

A cardiac catheter was inserted into aorta via the left common carotid artery, with an 8-channel physiological recorder to monitoring and recording heart rate (HR), aortic systolic pressure (ASP), aortic diastolic blood pressure (DAP) and mean pressure (MBP). The intubation was then continued, inserted into left ventricle. Left ventricle pressure (LVSP), left ventricular end diastolic pressure (LVEDP), maximum left ventricular pressure rise rate (+dp/dtmax) and maximal left ventricular pressure drop rate (-dp / dtmax) were measured.

3.2 Results analysis

(1) Effects of octacosanol on blood gas of chronic hypoxic rats

There were no significant differences in PaO\textsubscript{2}, SaO\textsubscript{2} and Lac between the drug group and the control group (P > 0.05). (As shown in Table 5).

Table 5 Effects of octacosanol on blood gas of chronic hypoxic rats

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>PaO\textsubscript{2}</th>
<th>SaO\textsubscript{2}</th>
<th>Lac</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>12</td>
<td>40.25±4.12</td>
<td>74.25±2.65</td>
<td>2.25±0.36</td>
</tr>
<tr>
<td>Drug</td>
<td>12</td>
<td>42.25±2.36</td>
<td>74.98±2.14</td>
<td>2.14±0.47</td>
</tr>
</tbody>
</table>

(2) Effects of octacosanol on blood routine of chronic hypoxia rats

The mean hemoglobin concentration (MCHC) of drug group was significantly higher than in the control group in the chronic hypoxic rats (P < 0.01), the hematocrit (HCT) was significantly lower than the control group (P < 0.05), the mean volume of red blood cells, mean corpuscular hemoglobin (MCH), red cell distribution width standard deviation (RDW-SD) and red blood cell distribution width coefficient variation (RDW-CV) were significantly lower than those of the control group (p < 0.01). (As shown in Table 6).

Table 6 Effects of octacosanol on blood routine of chronic hypoxia rats

|          | RBC  | HGB  |  |  |  |  |  |  |  |
|----------|------|------|  |  |  |  |  |  |  |
| Control  | 21.2±1.23 | 305.2±6.58 | 0.62±0.33 | 62.3±2.35 | 21.5±1.25 | 302.1±5.36 | 56.2±4.25 | 0.25±0.03 |
| Drug     | 16.2±0.25 | 346.2±6.51 | 0.60±0.25 | 62.3±2.12 | 18.5±0.48 | 316.5±2.01 | 45.2±3.68 | 0.24±0.09 |

(3) Effects of octacosanol on cardiac morphology of chronic hypoxia rats

HE staining showed the right ventricular wall in the control group was thicker, showing focal myocardial dissolution and myocardial interstitial edema (Figure 3). The volume of the right ventricular cardiomyocytes increased, the diameter of the ventricular cardiomyocytes increased, the eosinophilic granules in the cytoplasm increased; Van Gieson staining showed more collagen fibers around the blood vessels, and radial extension to the surrounding myocardial interstitium, but also the presence of focal collagen hyperplasia in myocardial tissue. While the changes of right ventricle wall thickness of the rats in the drug group were less than those of the control group, and there was no obvious change of myocardial dissolution and myocardial interstitial edema. The VG staining showed that the collagen fibers around the blood vessels were few (As shown in figure 4).
4. CONCLUSION

(1) Octacosanol can significantly enhance the ability of hypoxia tolerance in rats, reduce brain energy consumption, improve myocardial anoxia, at the same time significantly prolong the survival time of rats in atmospheric confined hypoxia environment;

(2) Octacosanol can’t improve the oxygen transport efficiency of the body, but can increase the concentration of hemoglobin in red blood cells, reduce the volume of red blood cells, help to reduce blood viscosity, improve microcirculation;

(3) Octacosanol can effectively prevent pulmonary hypertension induced by chronic hypoxia, reduce pulmonary vascular remodeling in chronic hypoxic rats, inhibit right ventricular hypertrophy in chronic hypoxic rats, and reduce the degree of myocardial fibrosis.

(4) Octacosanol can significantly increase the maximal (aerobic) athletic ability of chronic hypoxic rats. The mechanism may be related to the increase of myocardial mitochondrial oxidative respiration efficiency and ATP synthesis rate of chronic hypoxic rats induced by octacosanol, Improve myocardial function of chronic hypoxic rats.

REFERENCES


