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Vascular Endothelial Growth Factor Concentration in Brain of Rat Treated with Anaerobic Exercises

Rostika Flora, Muhammad Zulkarnain, Yuliana
Faculty of Medicine, Universitas Sriwijaya, Indonesia

ABSTRACT
Anaerobic exercise is a high-intensity exercise that needs quick energy supplies obtained in a very short time. However, this exercise may result in tissue hypoxia which is characterized by the increase of HIF-1α concentration. The presence of HIF-1α will induce the secretion of VEGF and, eventually, trigger angiogenesis. Nevertheless, it is still unclear whether anaerobic exercise will also cause hypoxia in which this condition will increase the concentration of VEGF in brain tissues. The aim of this study was to find out the effect of anaerobic exercise frequency towards VEGF concentration of Wistar rat brain tissues. Brain tissues were taken from rats treated with anaerobic exercise using treadmill. This exercise was given in different frequencies; one time, three times, and seven times a week. The data collected were analyzed using independent t-test. The results of this study showed that anaerobic exercise done once a week could significantly increase VEGF concentration (p < 0.05) if compared with the one in control group (95.21 ± 31.99 vs. 63.36 ± 11.01 pg/mL). Meanwhile, VEGF concentration of treatment groups given exercise three times a week (47.97 ± 10.68 pg/mL) and seven times a week (40.56 ± 13.98 pg/mL) showed a significant decrease if compared with that of control group (63.36 ± 11.01 pg/mL). Anaerobic exercise affected VEGF concentration as an indicator of angiogenesis in brain tissue of wistar rats.

1. INTRODUCTION
Anaerobic physical exercise is a high-intensity activity acquiring immediate energy in a very short time, but it cannot be done continuously [1]. Anaerobic environment will trigger the accumulation of HIF-1α in tissues. When hypoxia occurs, HIF-1α will translocate to cell nuclei to make dimerisation with HIF-1β to form HIF-1 [2]. Through activation of HIF-1, the expression a number of genes, including Vascular Endothelial Growth Factor (VEGF), will be increased in order to lessen cell dependence to oxygen and to elevate oxygen supply to tissues as well.

VEGF is an essential regulator in developing new blood capillaries (angiogenesis) [3]. The development of these new capillaries appears as a response to meet oxygen demands in hypoxic muscle tissues. Other studies conducted both in vivo and in vitro had proved that VEGF was involved in the regulation of angiogenesis process [4]. A study of Flora et al indicated that there was an increase in VEGF concentration of cardiac muscle tissues of rats treated with anaerobic physical exercises. The increase of VEGF concentration in anaerobic groups was higher than that of aerobic ones [5].

Besides cardiac muscles, brain is one of the vital organs which also works hard to maintain body
balance when physical exercises occur. Other previous related study stated that angiogenesis occurred in brain motor cortex tissue of rats that were given aerobic physical exercise [6]. As previously mentioned, it is said that anaerobic physical exercise can cause angiogenesis in brain tissues, but this statement is still debatable [7]. In addition, the number of studies regarding the effect of anaerobic physical exercises towards angiogenesis is still small. In normal brain tissue, it is still unexplained whether or not anaerobic physical activities could also increase the secretion of VEGF considering the fact that brain is a vital organ that relies on oxygen supply in order to maintain its function. Therefore, it is needed to do a further study to find out the effect of anaerobic physical exercise towards VEGF concentration as an indicator of angiogenesis in Wistar rat brain tissue.

2. RESEARCH METHOD

This was an experimental study using post test only control group design. This study was conducted in animal house of Faculty of Medicine, Universitas Padjadjaran in February 2015. The sample of this study was twenty eight Wistar rats aged from 6-8 weeks and taken using random sampling. The sample was divided into four groups namely P1, P2, P3, and P4. P1 was control group and P2, P3, and P4 were treatment groups. The treatment groups were then given anaerobic physical activities on a treadmill which was set for twenty minutes in speed of 25 m/minute. The exercise was given in different frequencies; 1 time treatment (P2), 3 times a week (P3), and 7 times a week (P4). The research procedure for anaerobic physical exercises was based on a method developed by Fahrem and Flora et al [8],[5]. Before the treatments were implemented, all experimental animals were acclimatized for one week by putting them on a low-speed treadmill.

3. RESULTS AND ANALYSIS

There was a significant difference in VEGF concentration of brain tissues among groups of one-time treatment (p = 0.044), three-times a week (p = 0.034), and seven-times a week (p = 0.011) if compared with that of control group (Table 1). These findings indicated that anaerobic physical exercise with different frequencies had a significant effect to VEGF concentration of Wistar rat brain tissues.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Mean ± SD (pg/ml)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>63.30 ± 11.01</td>
<td></td>
</tr>
<tr>
<td>One-time treatment</td>
<td>95.21 ± 31.99</td>
<td>0.044</td>
</tr>
<tr>
<td>Three times a week</td>
<td>47.97 ± 10.68</td>
<td>0.034</td>
</tr>
<tr>
<td>Seven days a week</td>
<td>40.56 ± 13.98</td>
<td>0.011</td>
</tr>
</tbody>
</table>

Table 1 shows that there is an increased VEGF concentration of Wistar rat brain tissues in one-time treatment group if compared with that of control group. On the other hand, the concentration of VEGF was decreased in treatment groups of three times and seven times a week. The lowest concentration of VEGF was found in a group given seven-time treatment a week.

3.1. Discussion

The results of this study showed that there was an increased VEGF concentration of Wistar rat brain tissues in one-time treatment group if compared with that of control group. However, the concentration of VEGF was decreased in treatment groups of three times and seven times a week. This could possibly be explained that in one-time anaerobic exercise there was no body adaptation response to physical exercise given. The body responds this physical exercise as a stressor. The increase of oxygen demands during physical exercise will lead to tissue hypoxia. Tissue hypoxia will inhibit oxygen transfer to brain.

Brain is very vulnerable to insufficient supply of oxygen and glucose. In relaxation, our brain needs 20% oxygen from all oxygen demands and 70% glucose. The increased demand of oxygen is accompanied by continuous brain metabolic activities. It needs constant blood flow to brain in order to supply adequate and regular nutrients. In one minute, brain needs 800 cc of oxygen and 100 mg of glucose as energy sources [9].

The presence of hypoxia during physical exercises can increase the expression of VEGF through HIF-1α regulation. The elevation of mRNA VEGF has something to do with the increase of mRNA HIF-1α.

JPJHS Vol. 4, No. 3, September 2015 : 201 – 204
indicating that HIF-1α affects the expression of VEGF genes [10]. Similar studies on experimental animals showed that mRNA VEGF of rat muscles was increased four times after single bout treadmill run. The concentration was increased when physical activities were conducted in hypoxic environment [2].

The results of this study were in line with those of Flora et al in which there was an increased VEGF concentration of cardiac muscle tissues in one-time anaerobic physical exercises [5]. In addition, a study of Margariteescu on patients experiencing acute ischemic cerebral infarct also appealed an increased concentration of VEGF (8.2 x 10^6 to 7.5 x 10^7) as a protective mechanism towards stroke hypoxia [11]. The concentration of VEGF was also elevated on motor cortex and striatum of rats which were treated with aerobic physical activities [12].

VEGF is considered as an important angiogenic factor in that the regulation is increased after physical activities and electrical stimuli are given. Therefore, the increase of VEGF provides important stimuli for angiogenesis, especially in initial phase of training program. The VEGF concentration of brain tissue was increased in a treatment group of one-time anaerobic physical exercise [12][13].

VEGF serves as an important regulator in angiogenesis [14]. Angiogenesis itself refers to the development of new blood capillaries in responding muscle adaptation to lower level of oxygen. Angiogenesis occurs when there is an imbalance between tissue metabolic demands and the ability of blood vessel perfusion. The regulation of VEGF genes seems to have something to do with exercise intensity, and its concentration is increased significantly in hypoxic condition [2].

In treatment groups of three times and seven times a week the concentration of VEGF was decreased significantly (p < 0.05) if compared with control group. The result of this study was in line with Salceda et al that in acute hypoxia the expression of mRNA HIF-1α was increased, but in repeated hypoxia the expression of which was decreased again [15]. This result was also supported by a study of Harik et al. In their study, it was stated that vascularisation occurred 55% in rat brain tissues that experienced hypoxia for one week. However, the vascularisation was not occurred in repeated hypoxia [16].

The results of independent t-test showed that there was a significant difference (p < 0.05) between VEGF concentration of brain tissues in anaerobic treatment groups if compared with that of control one. Anaerobic treatment with different frequencies affected the VEGF concentration of Wistar rat brain tissues.

4. CONCLUSION

The results showed that anaerobic physical exercises performed in different frequencies could influence VEGF concentration of Wistar rat brain tissue.

ACKNOWLEDGEMENTS

This study was funded by a postgraduate grant from Ministry of Higher Education & Research and Technology in fiscal year 2015. Therefore, the researcher would be very grateful for the grant so that this study could be finished in time.

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