Impact of Exercise on Some Haematological and Cellular Immune Markers in Male Athletes

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Authors’ contributions

This work was carried out in collaboration among all authors. Author KUN designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author ANC managed the analyses of the study and author FSAT managed the literature searches. All authors read and approved the final manuscript.

ABSTRACT

Due to conflicting reports on the impact of exercise on haematological and immunological indices, this study investigated the effects of exercise on some haematological and cellular immune markers in male athletes. Blood samples were collected from 86 apparently healthy male athletes before and after exercise training using standardized methods. Similarly, blood samples were obtained from 100 male non-athletes and served as control. Blood samples collected from athletes and non-athletes were subjected to experimental evaluation of some haematological and cellular immune biomarkers using standard techniques. Results showed that, with the exception of neutrophils that significantly increased after exercise in athletes, lymphocytes, monocytes, eosinophils and clusters of differentiation counts significantly (p<0.05) decreased in athletes after exercise. Also, with the exception of platelets, other haematological parameters assayed in this study significantly (p<0.05) decreased in athletes after exercise. However, there was no significant change in these parameters between athletes at rest and non-athletes. This study concludes that heavy training could lead to an open window of immunodepression leading to susceptibility to infection in athletes and non-athletes alike.
1. INTRODUCTION

It is widely believed that physical activity enhances the cardiovascular system. However, new studies have pointed to what seemed like adverse effects of prolonged heavy exercise upon both resistance to and the course of various viral and bacterial diseases [1]. Upper respiratory tract infection (URTI) is a major ailment that has been reported in athletes following participation in marathon or ultra-marathon events [2,3]. Examinations show that the pathogens normally responsible for URTI could not be identified in these athletes, suggesting that changes in the immune system and not pathogens could be responsible for these post exercise traumas [4]. To further explain this, some studies targeted at the specific immune response to exercise, have reported that light regular physical exercises have favourable effects on the physiological, psychological and immunological functions [5] and therefore could increase resistance of the body against infections [6]. Conversely, vigorous exercise produces negative effects on the aforementioned functions. Moderate exercise has been reported to boosts immune functions via chemotaxis and phagocytosis whereas extreme exercise on the other hand reduces these functions, with the exception of chemotaxis and degranulation [7]. Increased lymphocyte concentration has been reported to be likely due to the recruitment of all lymphocyte subpopulations (CD4 T cells, CD81 T cells, CD191 B cells, CD161 natural killer (NK) cells, and CD561 NK cells) to the vascular compartment [8,9]. Earlier studies have shown that NK cells with a high IL-2 response capacity were recruited to the blood during bicycle exercise [10].

On immunoglobulin, lower concentrations of the salivary IgA have been reported in cross-country skiers after a race [11]. This finding was confirmed by a 70% decrease in salivary IgA that persisted for several hours after completion of intense, long-duration ergometer cycling [12]. Decreased salivary IgA was also found after intense swimming [13] and after incremental treadmill running to exhaustion [14]. Submaximal exercise had no effect on salivary IgA [14]. The percentage of B cells among blood mononuclear cells (BMNC) does not change in relation to exercise; suggesting that the suppression of immunoglobulin-secreting cells is not due to changes in numbers of B cells. Many of these reported studies were on Caucasian non-athletes in the temperate environment. Research needs to be extended to performing athletes especially in the tropical environment. This is therefore partly covered in this study.

2. MATERIALS AND METHODS

2.1 Study Population and Sample Size

With ethical approval from the University of Port Harcourt Research ethics committee, the targeted population for this study was the 112 registered Nigerian athletes that partook in the 14th West African University Games (WAUG) held at the University of Port Harcourt between October and November, 2018. The sampling adopted was total population purposive-sampling technique, in which case, there is the possibility of selection all the elements in the study population. The basis for purposive sampling is to concentrate on subjects with particular desired characteristics that would be able to assist with the generation of relevant information to achieve the research objectives. Therefore, the justification for the adoption of total population purposive sampling technique is because it is a non-probability sampling technique that could include all members, within the population of interest. Hence, all the elements in the population were given equal opportunity to participate in the selection. Based on the sampling technique, 86 apparently healthy subjects between the ages of 22 and 30 years volunteered and were recruited for the study. The subjects were drawn from the following sporting events: fitness exercises, soccer, long-distance running, short-distance running and swimming. The athletes have been in the profession for not less than two years; and were at the peak of their training, in preparation for the West African University games (WAUG) held at the University of Port Harcourt between October and November, 2018. Training sessions were not less than 3 days per week for a minimum of 60 minutes per session as recommended by the [15]. In addition, the subjects were encouraged to avoid smoking, use of tobacco products or anti-inflammatory drugs. Subjects also completed a health history, drug usage, and physical activity questionnaires to determine eligibility. Also, 100 volunteer non-athletes were recruited and served as the control. Prior to participation in this study, each subject was informed of all procedures, potential risks, and benefits associated with the study and an informed consent form was signed.
2.2 Anthropometrics

For the participants that met the selection criteria, weight was measured using an electronic scale (Hanover, MD) while the subjects were wearing shorts with bare feet. Height was measured using a Seca stadiometer with 1 cm spaced leaning to the wall. Body mass index (BMI) for each participant was calculated using the formula: weight/height². All measurements were taken in the morning from 7 a.m. to 8 a.m.

2.3 Collection of Blood Sample and Analysis of Biomarkers

Using standard venipuncture procedures [16], about 5 ml of blood was collected from the antecubital veins of subjects using sterile syringes before exercise; between the hours of 7 am-8 am and immediately after exercise between 10-11 am. The blood was dispensed in Ethylenediaminetetraacetic Acid (EDTA) and stored at -4°C pending analysis.

The cells that make up the cellular immune system; namely: lymphocytes, neutrophils, monocytes and eosinophils were assayed. Also, total white blood cell (WBC) count, red blood cell (RBC) count, hemoglobin (Hb), packed cell volume (PCV), platelets (PLT) were also assayed. These parameters were assayed using the hematology autoanalyzer (CellDyn 3700 (Abbott, Chicago, IL, USA), which makes use of the Coulter principle [17]. The CD4 count was determined manually by microbead separation of CD4 T lymphocytes from other blood cells, followed by standard manual cell counting techniques using a light microscope.

2.4 Statistical Analysis

Data collated on selected variables from the study were statistically analyzed using statistical package and service solution (SPSS, version 20.0). The mean of descriptive statistics was recorded as mean± standard deviation. Comparison of parameters between athletes and non-athletes were done using paired t-test with the level of statistical significance accepted at p<0.05. Multiple comparison between non-athletes, athletes before exercise and athletes after was done using post-hoc multiple comparison test (LSD).

3. RESULTS

The result of the study is presented in tables and the figure below. Table 1 showed the anthropometric measurement of the athletes and non-athletes. Table 2 showed the effect of exercise on haematological parameters. Table 3 showed the effect of exercise on cellular immunophysiological marker while Fig. 1 showed percentage changes in both haematological and some immunophysiological parameters of male athletes after exercise.

4. DISCUSSION

Body Mass Index (BMI): The BMI is a calculation that compares weight relative to height. The index however, is not perfect because a body builder’s BMI may indicate overweight, but the extra weight is muscle rather than fat [18]. For everyone else, however, BMI is a reasonable measure of fitness level, demonstrating whether one is overweight, underweight or just right. The result of this study showed that there was no statistically significant p<0.05 difference between the BMI of male athletes compared with male non-athletes (Table 1); suggesting that BMI alone cannot be used to determine fitness levels in athletes because hypertrophied muscle may give perception of overweight. This agrees with a study of which concluded that the intensity of physical activity does not matter in lowering or maintaining BMI [19]; therefore, one can appear to be fat and yet be fit.

Haematological parameters: Investigators have suggested that the value of haematological indices in athletes varies as a result of different training regimes [20,21]; usually higher at the beginning of the competition, then declined in well-trained athletes [22]. Therefore, haematological indices is key determinants of optimal exercise performance in athletes. Result of this study showed that with the exception of platelets, haemoglobin, PCV, RBC and WBC diminished significantly in athletes after exercise; as shown in Table 2. This result is consistent with a study which reported low values of haematological variables in athletes during intensive training periods compared with clinical norms [23]. However, the study disagrees with other investigations that reported normal concentrations of hematological indices throughout training programmes [20,24].

The significant decrease in Hb, PCV and RBC immediately after heavy exercise might be due to training induced haemodilution as a result of plasma volume expansion associated with aerobic sports, endurance and ultra-endurance sports.
events. Plasma volume expansion is a compensatory mechanism to increase cardiac output and reduces blood viscosity, thereby optimizing microcirculation and improving oxygen delivery to the working muscles.

**Innate immunological markers:** Figure 1 showed that lymphocyte, monocyte, eosinophil and CD4 counts significantly p<0.05 decreased after exercise. However, there was significant p<0.05 increase in neutrophils count. The significant decrease in lymphocyte and Cd4 (cluster of differentiation) cells seen in this study is consistent with the studies which reported a rapid exercise-induced decrease in lymphocyte number within 30 minutes following a prolonged or high-intensity exercise [25,26]. However, there are contrary reports of lymphocytosis occurring during or immediately after exercise; depending on the intensity and the duration of the exercise [25,27]. The significant decrease in lymphocyte count, rather than being a real reduction, may be secondary to lymphocyte redeployment to peripheral tissues as a result of stress induced cortisol release.

Monocytes are mobilized during exercise; they are rapidly redeployed to skeletal muscle and differentiate into tissue-resident macrophages that facilitate repair and regeneration after an intensive bouts of exercise that cause significant skeletal muscle damage [28]. This redeployment of monocytes to muscles may account for the significant decrease in monocyte count immediately after exercise.

Eosinophils are a type of white blood cells that defend against parasites and infectious agents. In this study, we observed a significant decrease in eosinophil count which is consistent with a similar study in the elderly. However, it differs with the result of a similar study in asthmatic subjects [29].

The significant increase in neutrophil count seen in this study agrees with a report which showed that running at 75% of maximum heart rate (HRmax.) for 15 min increased neutrophil number [30]. Neutrophils recruited into circulation during exercise may be responsible for controlling the elevated levels of oxidative stress in plasma after exercise [31]. This impact of exercise on neutrophil count may be mediated by the activation of catecholamine and cortisol which are known to have adverse effect on the immune system [32,33].

<table>
<thead>
<tr>
<th>Variables</th>
<th>Male athletes (n=86)</th>
<th>Male NON-athletes (n=100)</th>
<th>Percentage change (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Weight (kg)</td>
<td>72.43±5.76</td>
<td>75.20±6.47</td>
<td>3.68</td>
</tr>
<tr>
<td>Height (m)</td>
<td>1.69±0.05</td>
<td>1.7101±0.06</td>
<td>1.18</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>25.41±1.52</td>
<td>25.69±1.56</td>
<td>1.09</td>
</tr>
</tbody>
</table>

Values are expressed as Mean ± S.D. *=statistically significant at p < 0.05, n= sample size

Fig. 1. Changes in haematological and some immuno-physiological parameters of male athletes after exercise

* shows that there was a statistically significant change at p≤0.05
Table 2. Effect of exercise on haematological parameters

<table>
<thead>
<tr>
<th>Variables</th>
<th>Non-athletes (n=100)</th>
<th>Athletes before exercise (n=86)</th>
<th>Athletes after exercise (n=86)</th>
<th>Change in athletes after exercise (%)</th>
<th>Change between athletes after exercise and non-athletes(%)</th>
<th>Change between athletes before exercise and non-athletes(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBC (x10^12/L)</td>
<td>6.22±0.29</td>
<td>6.19±0.27</td>
<td>5.08±0.40*</td>
<td>-21.85</td>
<td>-22.44</td>
<td>-0.48</td>
</tr>
<tr>
<td>PCV (%)</td>
<td>40.59±1.71</td>
<td>41.76±1.68</td>
<td>39.45±1.16</td>
<td>-5.89</td>
<td>-2.89</td>
<td>2.80</td>
</tr>
<tr>
<td>HB (g/dl)</td>
<td>13.91±0.41</td>
<td>13.98±0.46</td>
<td>12.37±0.55*</td>
<td>-13.02</td>
<td>-12.45</td>
<td>0.50</td>
</tr>
<tr>
<td>Platelets (x10^7/L)</td>
<td>210.52±11.71</td>
<td>210.06±12.24</td>
<td>253.20±6.20*</td>
<td>17.04</td>
<td>16.85</td>
<td>-0.22</td>
</tr>
<tr>
<td>WBC (x10^9/L)</td>
<td>6.40±0.21</td>
<td>6.39±0.19</td>
<td>4.07±0.38*</td>
<td>-57.00</td>
<td>-57.25</td>
<td>-0.16</td>
</tr>
</tbody>
</table>

All values are expressed as Mean ± S.D. *=statistically significant at p < 0.05, n= sample size

Table 3. Effect of exercise on cellular immuno-physiological markers

<table>
<thead>
<tr>
<th>Variables</th>
<th>Non-athletes (n=100)</th>
<th>Athletes before exercise (n=86)</th>
<th>Athletes after exercise (n=86)</th>
<th>Change in athletes before and after exercise (%)</th>
<th>Change between athletes after exercise and non-athletes(%)</th>
<th>Change between athletes before exercise and non-athletes(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NEUT (%)</td>
<td>46.15±4.97</td>
<td>46.20±4.77</td>
<td>60.17±3.45*</td>
<td>23.23</td>
<td>23.30</td>
<td>0.11</td>
</tr>
<tr>
<td>LYMPH (%)</td>
<td>32.69±1.95</td>
<td>32.95±2.14</td>
<td>22.98±1.62*</td>
<td>-43.39</td>
<td>-42.25</td>
<td>0.79</td>
</tr>
<tr>
<td>MONO (%)</td>
<td>6.10±1.11</td>
<td>6.08±1.23</td>
<td>4.07±0.27*</td>
<td>-49.39</td>
<td>-49.88</td>
<td>-0.33</td>
</tr>
<tr>
<td>EOSINO (%)</td>
<td>2.09±0.85</td>
<td>2.09±0.85</td>
<td>2.07±0.82</td>
<td>-0.97</td>
<td>-0.97</td>
<td>0</td>
</tr>
<tr>
<td>CD4(cells/μL)</td>
<td>704.21±31.96</td>
<td>688.05±54.14*</td>
<td>508.98±38.85*</td>
<td>-35.18</td>
<td>-38.36</td>
<td>-2.35</td>
</tr>
</tbody>
</table>

Results are presented as mean±standard deviation; n= sample size; * shows that there was a statistically significant change at p<0.05 when compared with the non-athletes
5. CONCLUSION

The available evidence from this study showed that exercise decreases hematological indices in the vascular compartment and also has modulatory effects on immunocyte dynamics. These effects may cause a temporary decline in the cellular immune system and create an open window, during which the exerciser may be susceptible to infections.

CONSENT

Prior to participation in this study, each subject was informed of all procedures, potential risks, and benefits associated with the study through both verbal and written form and signed an informed consent form.

ETHICAL APPROVAL

With ethical approval from the University of Port Harcourt Research ethics committee.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

18. Kesavachandran C, Bihari V, Mathur N. Can physical activity maintain normal


