Effects of Different Intensities of Circuit Resistance Training on Plasma level of High-Density Lipoprotein Subfractions and Apolipoprotein M in Untrained Young Men

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ABSTRACT

Background and Objectives: Coronary artery disease (CAD) is the leading cause of death worldwide. It is well established that low level of high-density lipoprotein-cholesterol (HDL-C) is a strong and independent risk factor for CAD. Apolipoprotein M (apoM) is a component of HDL, which is involved in pre-β-HDL formation and cholesterol efflux to HDL. It is believed that resistance and aerobic exercise can significantly reduce risk of cardiovascular disease, especially by increasing serum levels of HDL-C. However, little is known about effects of these activities on HDL-apoM levels. The aim of this study was to investigate effects of circuit resistance training at different intensities on HDL-associated apoM levels in young untrained men.

Methods: Forty-five age- and weight-matched healthy untrained men were randomly assigned to a control group (n=10) and four training groups: 20% 1 repetition maximum (1RM) (n=9), 40% 1RM (n=8), 60% 1RM (n=7) and 80% 1RM (n=8). The subjects performed circuit resistance training consisting of barbell bench press, underarm flab, seated barbell curl, triceps exercise with chains, lying leg curl, squats, hyperextension, abs workout, sit-ups and quadriceps workouts (30 seconds each) in three bouts without rest between stations and with active rest (3 minutes) between sets or bouts. The training protocol was carried out for 45 minutes per session, three sessions a week, for five weeks.

Venous blood samples were taken 48 hours before the first exercise session and 48 hours after the last training session. After separating plasma, HDL-associated apoM was measured using commercial ELISA kits. SPSS 16 was used for analysis of data using two-way ANOVA and Tukey’s post hoc test at significant level of 0.05.

Results: After the training intervention, the exercise groups had higher apoM levels in total HDL and HDL-2 compared to the control group (P<0.05). However, no significant difference in HDL-associated apoM level was observed between the study groups.

Conclusion: The results of this study indicate that various intensities of circuit resistance training can alter HDL-associated apoM levels. The decreased HDL-3-associated apoM level could indicate increased rate of apoM transfer to HDL-2, which could potentially prevent development of atherosclerosis and CAD by enhancing the antioxidant effects of HDL.

Keywords: Circuit Resistance Training, Total HDL-M, HDL3-M, HDL2-M.
INTRODUCTION
Homeostasis imbalance is one of the main causes of heart attack (1). Coronary artery disease is the leading cause of mortality in the world (2). Coronary artery disease and metabolic syndrome are common among Iranians (3). Studies have indicated a reverse and significant relationship between high-density lipoprotein-cholesterol (HDLC) levels and risk of atherosclerosis (4). Atherosclerosis is a chronic inflammatory condition that occurs when cholesterol ester accumulates in macrophage foam cells due to the inability of the cells to remove excess cholesterol (5).

In ultracentrifugation, human HDL can be separated into HDL2 and HDL3 based on their density. HDL2 and HDL3 can then be separated into HDL2b and HDL2a, HDL3a, HDL3b and HDL3c (6). HDL has antiatherogenic properties and plays a critical role in reverse cholesterol transport. It can induce endothelial synthesis of nitric oxide and prevent low-density lipoprotein (LDL) oxidation and inflammatory responses. Proteolytic analyses have revealed presence of 56 HDL-associated proteins, including apolipoproteins and lipid transfer proteins, but the exact function of these proteins is still not fully understood (7). Apolipoprotein M (apoM) is a novel HDL-associated human apolipoprotein, which is also found in triglyceride-rich lipoproteins and LDL (8). Although apoM is present in only 5% of HDL particles, recent studies have demonstrated a positive correlation between plasma apoM levels and HDL-C concentrations. This indicates that apoM might be involved in HDL metabolism. ApoM plasma is mainly associated with α-HDL and affects its interconversion to pre-β-HDL particles. It has been demonstrated that a 2-fold increase in apoM concentration can decreases atherosclerosis progression (9).

The positive effects of exercise on factors associated with cardiovascular disease are well established. However, most of these studies have been focused on the effects of endurance training, and little attention has been given to the effect of resistance training (10). Currently, it is not clear how HDL and its components will respond to different intensities of resistance training. Therefore, the purpose of this study was to determine effects of different intensities of circuit resistance training on plasma level of HDL, HDL subfractions and apoM in untrained young men.

MATERIALS AND METHODS
We carried out this semi-experimental study on 45 non-active male students who were studying at the Golestan University of Medical Sciences in Gorgan (Iran). Mean age and weight of subjects was 19.52 ± 0.96 years and 16.40 ± 78.42 Kg, respectively. Study procedures were explained in detail, and then written informed consent was taken from all participants. Exclusion criteria included having drug/alcohol addiction, regular exercise activity in the past six months, a history of diabetes, kidney, liver and cardiovascular diseases, and any type of physical disability. The subjects were randomly divided into five groups: control (n=10), 20% one-repetition maximum (1RM) (n=9), 40% 1RM (n=8), 60% 1RM (n=7) and 80% 1RM (n=8) (Table 1).

The training protocol was designed based on a previous study (11). The subjects first became familiar with the training environment and equipment. Value of 1RM of the intended movements (bench press, seated cable row, arm cable curl, triceps cable curl, lying leg curl, barbell squat, lumbar extension, abdominal, decline sit-up and quadriceps) was calculated using the following formula and through trial and error:

\[
1\text{RM} = \frac{\text{displaced weight}}{n-(\text{repit}d) \times 25}\%
\]

After warm up, the subjects performed the movements in 10 stations at different intensities (20, 40, 60 and 80% of 1RM) for 30 seconds without rest between the stations. The exercises were performed in three sets with three active rest periods between each set. Blood samples (10 ml) were taken from subjects’ forearms in a comfortable sitting position after at least eight hours of overnight fasting. Sampling was done 48 hours before the first training session and 48 hours after the last training session. The samples were collected in EDTA-coated tubes. Plasma was separated by centrifugation at 1500g for 15 minutes and then stored at -70 °C. were measured using the Prestige 24i automated analyzer (Japan). The mg/ml, respectively. The rest of the parameters normality assumption. Data were analyzed the measurement method
ApoM was measured using commercial ELISA kits. HDL-C was measured by photometric method (ParsAzmun Co., Iran). The coefficient of variation and sensitivity of were 0.81% and 1.

Kolmogorov–Smirnov test was used to test the with SPSS (version 16) using one-way ANOVA, two-way repeated measures ANOVA and Tukey’s test at a significant level of 0.05.

Table 1- Characteristics of subjects in each group

<table>
<thead>
<tr>
<th>Group</th>
<th>Time</th>
<th>Control</th>
<th>20% one-repetition maximum (1RM)</th>
<th>40% 1RM</th>
<th>60% 1RM</th>
<th>80% 1RM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Variable</td>
<td>Mean ± standard error</td>
<td>Mean ± standard error</td>
<td>Mean ± standard error</td>
<td>Mean ± standard error</td>
<td>Mean ± standard error</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>-</td>
<td>20 ±1</td>
<td>19 ±1</td>
<td>19 ±1</td>
<td>18 ±1</td>
<td></td>
</tr>
<tr>
<td>Height (cm)</td>
<td>-</td>
<td>177 ±8</td>
<td>178 ±3</td>
<td>176 ±6</td>
<td>183 ±7</td>
<td>181 ±4</td>
</tr>
<tr>
<td>Weight (Kg)</td>
<td>Pretest</td>
<td>76 ±17</td>
<td>77 ±20</td>
<td>76 ±16</td>
<td>81 ±13</td>
<td>81 ±15</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>77 ±16</td>
<td>75 ±19</td>
<td>76 ±16</td>
<td>80 ±12</td>
<td>81 ±15</td>
</tr>
<tr>
<td>Body mass index (Kg/m²)</td>
<td>Pretest</td>
<td>24 ±4</td>
<td>24 ±6</td>
<td>24 ±3</td>
<td>24 ±2</td>
<td>24 ±5</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>25 ±4</td>
<td>23 ±5</td>
<td>24 ±3</td>
<td>23 ±2</td>
<td>24 ±4</td>
</tr>
<tr>
<td>Back Extensions (Kg)</td>
<td>Pretest</td>
<td>24 ±5</td>
<td>21 ±5</td>
<td>18 ±6</td>
<td>18 ±10</td>
<td>18 ±8</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>41 ±7</td>
<td>49 ±11</td>
<td>50 ±11</td>
<td>41 ±15</td>
<td>49 ±8</td>
</tr>
<tr>
<td>Abdomen (Kg)</td>
<td>Pretest</td>
<td>28 ±6</td>
<td>22 ±8</td>
<td>25 ±6</td>
<td>24 ±7</td>
<td>21 ±10</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>41 ±9</td>
<td>50 ±23</td>
<td>56 ±17</td>
<td>62 ±13</td>
<td>49 ±15</td>
</tr>
<tr>
<td>Back arm (Kg)</td>
<td>Pretest</td>
<td>42 ±7</td>
<td>31 ±5</td>
<td>37 ±7</td>
<td>44 ±15</td>
<td>35 ±6</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>47 ±8</td>
<td>47 ±9</td>
<td>50 ±11</td>
<td>53 ±7</td>
<td>48 ±4</td>
</tr>
<tr>
<td>Barbell Bench Press (Kg)</td>
<td>Pretest</td>
<td>30 ±6</td>
<td>26 ±11</td>
<td>28 ±10</td>
<td>33 ±11</td>
<td>24 ±10</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>33 ±9</td>
<td>36 ±9</td>
<td>42 ±6</td>
<td>40 ±10</td>
<td>37 ±8</td>
</tr>
<tr>
<td>Leg squats (Kg)</td>
<td>Pretest</td>
<td>57 ±11</td>
<td>54 ±15</td>
<td>53 ±12</td>
<td>64 ±14</td>
<td>54 ±11</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>69 ±22</td>
<td>75 ±20</td>
<td>90 ±15</td>
<td>83 ±21</td>
<td>91 ±16</td>
</tr>
<tr>
<td>Leg (Kg)</td>
<td>Pretest</td>
<td>95 ±35</td>
<td>145±209</td>
<td>61±14</td>
<td>63±12</td>
<td>61±11</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>121±59</td>
<td>177±59</td>
<td>163±36</td>
<td>148±27</td>
<td>181±39</td>
</tr>
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<td>Front arm (Kg)</td>
<td>Pretest</td>
<td>16 ±3</td>
<td>13 ±5</td>
<td>11 ±5</td>
<td>14 ±6</td>
<td>11 ±6</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>21 ±6</td>
<td>24 ±3</td>
<td>27 ±4</td>
<td>30 ±7</td>
<td>28 ±4</td>
</tr>
<tr>
<td>Back foot (Kg)</td>
<td>Pretest</td>
<td>21 ±2</td>
<td>23 ±9</td>
<td>16 ±5</td>
<td>19 ±2</td>
<td>18 ±5</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>21 ±5</td>
<td>23 ±5</td>
<td>24 ±3</td>
<td>28 ±4</td>
<td>24 ±3</td>
</tr>
<tr>
<td>Front leg (Kg)</td>
<td>Pretest</td>
<td>34 ±7</td>
<td>36 ±12</td>
<td>31 ±12</td>
<td>38 ±4</td>
<td>35 ±10</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>39 ±8</td>
<td>48 ±13</td>
<td>48 ±6</td>
<td>53 ±7</td>
<td>52 ±7</td>
</tr>
<tr>
<td>Armpit (Kg)</td>
<td>Pretest</td>
<td>49 ±6</td>
<td>47 ±8</td>
<td>44 ±5</td>
<td>48 ±6</td>
<td>44 ±6</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>46 ±7</td>
<td>46 ±7</td>
<td>51 ±6</td>
<td>52 ±7</td>
<td>50 ±4</td>
</tr>
<tr>
<td>Total power (Kg)</td>
<td>Pretest</td>
<td>324±59</td>
<td>349±80</td>
<td>328±57</td>
<td>364±73</td>
<td>325±66</td>
</tr>
<tr>
<td></td>
<td>Posttest</td>
<td>483±81</td>
<td>578±124</td>
<td>582±98</td>
<td>596±77</td>
<td>613±90</td>
</tr>
</tbody>
</table>

RESULTS

ApoM level in HDL2 and total plasma HDL increased significantly in all exercise groups and decreased in the control group. Table 3 shows alteration of apoM, HDL2 and HDL3 in the study groups. Results of repeated measures ANOVA showed that time, group and their combination had no significant impact on the variables except for HDL3(Table 3) . Regarding the results of the ANOVA with repeated measures, the effect of time was significant in comparison with the control group before and after (P=0.021).
According to human tissue expression array studies, apoM is predominantly present in adult liver and kidney. Liver-derived apoM is mainly secreted into the plasma and accumulates in lipoproteins, while kidney-derived apoM is connected to a multiligand, endocytic receptors megalin in the renal proximal tubule (16). Nevertheless, mechanisms that regulate transcription of the human apoM gene are not well-understood (17). Recently, it has been shown that plasma level of apoM has a positive correlation with plasma leptin levels and a negative correlation with cholesterol levels in obese individuals. In fact, it has been suggested that leptin can stimulate hepatic apoM expression (18). According to Lappalainen et al., intensive isokinetic exercise can significantly reduce leptin levels (19). Hence, it can be assumed that a possible decrease in leptin levels through resistance training could contribute to the apoM reduction.

ApoM can delay LDL oxidation, alter HDL metabolism by increasing the production of pre-β-HDL and exert anti-atherosclerotic effects (9, 21). There is a strong correlation between plasma apoM and cholesterol concentrations, but the mechanisms of this association remain unknown (14). ApoM may facilitate the reverse cholesterol transfer process by converting small pre-β-HDL into larger pre-β-HDL (22). A study reported overexpression of the apoM gene in LDL.
receptor-knockout rats, which prevented progression of early atherosclerotic lesions (23). Furthermore, the protective activity of HDL against atherosclerosis could be related to its antioxidant and anti-inflammatory properties (21).

CONCLUSION
The 5-week circuit resistance training intervention at different intensities not only improved body mass index, muscle strength and weight loss, but also altered plasma

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