Serum Lipoprotein Patterns in Long-Term Anabolic Steroid Users

Thomas R. Sachtleben, Kris E. Berg, John P. Cheatham, Gary L. Felix, and Philip J. Hofschire

Key words: weight training, cholesterol, steroids

Anabolic steroid use has been recognized to cause adverse changes in blood lipid levels, particularly the high density lipoprotein fraction of cholesterol (HDL-C) (Baldzinski et al., 1990). A recent report suggested the importance of high HDL-C and a satisfactory total cholesterol to HDL-C ratio in preventing future cardiovascular disease (Stampfer, Sacks, Salvini, Willett, & Hennekens, 1991). Apolipoproteins are used also as measures of coronary heart disease risk and provide additional information to serum lipid assessment (Thompson, 1984). Therefore, reports of widespread anabolic steroid use, especially in the younger population (Terney & McLain, 1990; Windsor & Dumitru, 1989), may have important cardiovascular implications.

Few studies have reported long-term effects on lipoprotein levels with anabolic steroid use, and little is known about whether serum lipoproteins return to normal after individuals stop taking anabolic steroids. Peterson and Fahey (1984) demonstrated wide HDL-C variations with cyclical use of steroids in a small group of individuals who used steroids for at least 3 years. A group of 42 anabolic steroid users studied by Lenders, Demacker, and Vois (1988) exhibited a persistent elevation in the ratio of low density lipoprotein of cholesterol (LDL-C) to HDL-C even after discontinuing steroid use for an average of 5 months.

Evaluation of HDL-C and apolipoprotein A1/B ratios in this study remained nonsignificantly different from controls during the same period. In addition, Kleiner, Calabrese, Fielder, Naito, and Skibinski (1989) showed significant differences in lipoprotein and apolipoprotein values when comparing nonusers to users who had abstained from steroid use for at least 6 months. The average duration of prior steroid use in this group was 3 years.

In contrast, however, Webb, Laskarzewski, and Ghebeck (1984) studied 14 anabolic steroid users after an average of 7.3 months of abstinence and found not only that HDL-C values returned to normal levels also but that a majority of the participants had greater than 90th percentile age-race-and-sex HDL-C values. Costill, Pearson, and Fink (1984) also showed a return of HDL-C values among a small sample of steroid users after approximately 4 weeks of abstinence. Earlier work (Haffner, Kushwaha, Foster, Applebaum-Bowden, & Hazzard, 1983) also demonstrated significant reductions in HDL-C (51%) while participants administered anabolic steroids, but no evaluations were made after a period of abstinence.

Few studies have investigated the chronic effects of anabolic use on lipoprotein levels. Consequently, it is unknown if lipoprotein values return to their initial values after cessation of steroid use. Because of the inconsistent findings and limited number of studies on the topic, further study is needed to confirm previous findings. We measured serum lipoprotein and apolipoprotein levels in a group of weight trainers who used anabolic steroids and compared the levels with those in a group of weight trainers who did not use anabolic steroids. Lipoprotein and apolipoprotein levels of steroid users after more than 8 weeks of abstinence were compared with levels at the peak of their steroid cycle. The following research hypothesis was examined: serum lipoprotein and apolipoprotein

Submitted: September 19, 1995
Accepted: May 5, 1996

Thomas R. Sachtleben, John P. Cheatham, Gary L. Felix, and Philip J. Hofschire are with the Department of Pediatric Cardiology at the University of Nebraska Medical Center. Kris E. Berg is with the School of Health, Physical Education, and Recreation at the University of Nebraska–Omaha.
levels would be unfavorably different in anabolic steroid users while taking steroids and after 8 weeks of abstinence, as compared to nonusers.

Method

Participants

Twenty-four age-matched men with a 5-year history of weight training were recruited for evaluation of their lipid profiles. Eleven participants were anabolic steroid users, while 13 were not, thereby forming the two groups in the study (users and nonusers). All participants gave informed consent prior to participating in this study. The risks of steroid use were explained, and participants were advised to discontinue steroid use.

Design

A participant was included in the user group only after a positive history of current anabolic steroid use of at least 3 consecutive months or current use of at least 2 months with at least 2 additional months within the past year. A participant was included in the study as a nonuser if he had no previous history of anabolic steroid use and a negative urinalysis screen for anabolic steroids. Weight training eligibility for both user and nonuser groups included a 5-year lifting history of at least three times weekly. Inclusion criteria required participants to bench press 125%, squat 150%, and leg press 225% of their body mass. All users had a history of at least 12 months of anabolic steroid administration ($M = 59.1$ weeks, $SD = 18.8$) during cycles ranging in duration between 6–18 weeks.

Measures

Verification was done by gas chromatography-mass spectrometry on urine samples (Paul Ziffern Olympic Analytical Laboratory at the University of California–Los Angeles, Los Angeles, CA). Drugs taken either orally or by injection are listed in Table 1. A drug usage questionnaire was required, with specific drug history obtained by personal interview. Comparisons between the drug usage questionnaires and laboratory results revealed a single discrepancy, in which neither anabolic steroids or steroid metabolites were found in a user during the cycle peak.

After they had fasted for 12 hours, participants’ serum levels were evaluated using standardized methods of venipuncture in a Centers for Disease Control and Prevention-certified laboratory at the University of Nebraska Medical Center. Blood samples for lipoprotein values, liver function tests, and chemistry profiles were all drawn into a 5-cc lithium-heparin tube. A blood chemistry profile of all participants assessed the hepatic and renal systems. The nonuser group was evaluated during a one-time visit to the laboratory. The user group was tested both during peak cycle steroid use and after being off of drugs for at least 8 weeks. Apolipoproteins A1, B, and A1/B values were obtained also from both groups of participants. Blood for apolipoprotein profiles was drawn into 7-cc clot tubes and evaluated at Nichols Institute (San Juan Capistrano, CA). Calculated blood analysis (CALC) methodology was implemented in compliance with the international standardization of apolipoproteins A1 and B. All blood was drawn using 21-gauge needles and the vacutainer™ collection system. Internal quality control checks were performed daily for all serum and blood chemistry values. External quality control testing was performed on a regular basis in both laboratories by an external reviewer using duplicate equipment for comparative results documentation.

The user group participated both on and off-cycle in scheduled aerobic exercise at a level believed to attain cardiovascular fitness by American College of Sports Medicine (ACSM, 1991) criteria (aerobic exercise at least three times weekly, each of 15 min duration). The nonuser group completed 30-min aerobic sessions only 1.4 times per week. No participant had a history of symptoms or a systemic disease.

History regarding prior use of protein and amino acid supplements, illicit drugs, and other ergogenic

<table>
<thead>
<tr>
<th>Table 1. Anabolic steroids used</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral</td>
</tr>
<tr>
<td>Dianabol (methandrostenolone)</td>
</tr>
<tr>
<td>Anavar (oxandrolone)</td>
</tr>
<tr>
<td>Winstrol (stanozol)</td>
</tr>
<tr>
<td>Anadrol-50 (oxymetholone)</td>
</tr>
<tr>
<td>Metandren (methyltestosterone)</td>
</tr>
<tr>
<td>Halotest (fluoxymesterone)</td>
</tr>
<tr>
<td>Primobolin (methenolone acetate)</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>
Data Analysis

Statistical analysis was performed using independent t tests for comparing nonusers and users off-cycle. Dependent t tests were used for data comparing users off-cycle and users on-cycle. Due to the many comparisons made, the Bonferroni procedure was used to adjust the alpha level to .007 (.05/7 = .007), because seven dependent variables were compared in two sets of groups. Effect size was determined using the technique of Glass (1976) which expresses the difference in experimental and control groups in units of standard deviation.

Results

Patient demographics were not statistically different between the groups. Each group had the same mean age (nonusers, M age = 26.5 years, SD = ±5.8; users, M age = 26.5 years, SD = ±3.6). Body surface area was 2.08 ± 0.2 m² in the off-cycle nonuser and user groups and increased nonsignificantly to 2.12 ± 0.2 m² (p > .007) in the on-cycle user group. Nonuser mean body weight was 89.0 ± 12.2 kg, while the users averaged 92.7 ± 9 kg off-cycle and 95.5 ± 9 kg on-cycle, t(23) = 1.35 and t(25) = 1.78, respectively, p > .007 for both. The users employed a variety of anabolic steroids (see Table 1), self-administered orally and intramuscularly.

Total serum cholesterol and HDL-C values in the nonuser group were within normal reference limits (see Table 2). Only 2 of the 13 nonusers had a total cholesterol level greater than 200 mg/dl, and none of the values were greater than 220 mg/dl. No significant differences were observed in total cholesterol and LDL-C values. However, HDL-C and cholesterol-HDL values showed dramatic changes in the users-off and users-on conditions. The fasting HDL-C level in the nonuser group was significantly higher than the off-cycle users group, t(22) = 3.35, p < .007, effect size = 1.2. HDL-C levels were further depressed in the on-cycle users group, t(21) = 4.66, p < .007, effect size = 2.2. Nonuser HDL-C values were also within normal ranges, with no individual value less than 36 mg/dl. Although not significant, there was a trend for the cholesterol-HDL-C ratio to be significantly higher in the off-cycle user group compared to the nonuser group, t(22) = 2.52, p = .01. A similar trend, although not significant, t(21) = 1.47, p = .057, was found in the cholesterol-HDL-C for the on-cycle user group compared to the off-cycle group.

Apolipoprotein levels also were altered by anabolic steroid use (see Table 3). Apolipoprotein AII in the non-user group was significantly higher than the off-cycle user group, t(22) = 2.71, p < .007, effect size = 1.2. The on-cycle group had a lower apolipoprotein AII level than the off-cycle group, t(21) = 5.01, p < .007, effect size = 1.3. Comparisons of apolipoprotein B values were not significantly different. Apolipoprotein AII/B for the off-cycle user group was significantly lower than that for the nonuser group, t(22) = 2.72, p < .007, effect size = 1.2. The user group, while on steroids, had a significantly lower apolipoprotein AII/B than the off-cycle user group, t(21) = 3.89, p < .007, effect size = .8.

Nine participants in the steroid-user group had abnormal liver function studies, and six of the eleven had abnormal serum creatinine values. A summary of the blood chemistry values appears in Table 4. None of the non-user group had abnormal renal function serum tests, but two had evidence of mild liver dysfunction consistent with heavy protein supplement intake.
Discussion

Lipoproteins are conjugated protein carriers of cholesterol, phospholipids, and triglycerides. They are often used as markers for identifying cardiovascular risk, as a known relationship between dyslipoproteinemia and coronary heart disease has been established. A well-documented relationship has been reported between the combination of a low HDL-C value with a high cholesterol/HDL-C ratio and an increased incidence of coronary artery disease (Stampfer et al., 1991). In addition, some suggest that apolipoprotein levels may provide additional information in the evaluation of cardiovascular risk (Thompson, 1984). In a study involving over 200 men and women who underwent diagnostic coronary angiography, Kwiterovich et al. (1992) reported that apo-A1 and apo-B as well as apo-B/apo-A1 were all significant risk factors for premature coronary artery disease (CAD). It was also suggested that apolipoprotein levels are even better predictors of CAD than lipoprotein measurements.

Lipoprotein data from the present study show reduced levels of HDL-C and cholesterol/HDL-C ratio that substantiate previous reports (Baldo-Enzi & Stolley, 1979; Frolich, Kullmer, Urhausen, Bergmann, & Kindermann, 1989; Kleiner et al., 1989; Lenders et al., 1988; Webb et al., 1984). These significant alterations in the lipid profile were maintained even after at least 8 weeks of abstinence. Findings by Webb et al. (1984) have suggested that lipoprotein values return to antiatherogenic levels after a steroid cessation of at least 7.3 months occurs. However, steroid users in the present study typically spent only 8–12 weeks between consecutive steroid cycles. This pattern is similar to those exhibited by current and prior steroid users (O’Connor & Baldini, 1990). In the study by O’Connor & Baldini (1990) participants administered steroids during cycles lasting 9–12 weeks and repeated similar cycles 3–4 times per year. In addition, Frohlich et al. (1989) suggested that long-term use of anabolic steroids may alter lipoproteins and apolipoproteins permanently, if the periods of abstinence are relatively brief.

Previous investigations have determined that HDL-C values lower than 43.6 and cholesterol/HDL-C ratios greater than 5.3 are associated with an increased risk for myocardial infarction (Stampfer et al., 1991). The extremely low HDL-C values shown in several participants from the present study may forecast increased risk of coronary disease in these individuals. LDL-C values increased 14.7% during steroid cycles, although this increase was not statistically significant. The role of LDL-C in promoting atherosclerosis is well known. In addition, although the cholesterol/HDL-C ratio increase from 4.8–6.8 during steroid use was not significant, the effect size was large and, thus, demonstrates extreme alteration of cardiovascular risk by anabolic steroids.

When LDL-C data from the current study were compared to data from the Lipid Research Clinic (LaRosa et al., 1986) several striking trends were noted. All HDL-C values from the non-user group fall within 2 standard deviations of the mean. LDL-C values from the user groups all fall below the 50th percentile, with severe depression of HDL-C values during steroid use.

Although differences in apolipoprotein B levels did not vary significantly in the present study, an increasing trend from nonusers to off-cycle users to on-cycle users was demonstrated. The depressed apolipoprotein A1 and A1/B values confirm data reported in previous studies (Baldo-Enzi et al., 1990; Cohen, Faber, Benefe, & Noakes, 1986; Lenders et al., 1988). Baldo-Enzi & Stolley (1977) reported depressed A1/B levels in a group of weight trainers who had taken anabolic steroids for at least 2 months as compared to a similar group of non-users. Baldo-Enzi et al. (1990) also showed a decrease in A1/B during a steroid cycle when A1/B ratios were compared to the same group 3 months after the cessation of use. Cohen et al. (1986) also demonstrated lower apolipoprotein A1 levels in a group of steroid users who were compared to a nonuser reference group. Lenders et al. (1988) observed decreased A1/B values in a group of anabolic steroid users who were compared to a non-user control group. A separate group of users who had not taken steroids for at least 2 months showed dramatic falls in A1/B values following a 9-week cycle of steroid use.

It has been demonstrated that one of the most significant cardiovascular benefits from vigorous physical

| Table 4. Summary of participants with abnormal blood chemistry values |
|-------------------|----------------|----------------|
| Variable          | Nonusers (n = 13) | Users-off (n = 11) | Users-on (n = 11) |
| LDH               | 2               | 6               | 9               |
| AST               | 4               | 10              | 9               |
| ALT               | 1               | 0               | 0               |
| GGTP              | 1               | 1               | 0               |
| Bilirubin-direct  | 1               | 0               | 0               |
| Bilirubin-total   | 3               | 0               | 0               |
| BUN               | 0               | 4               | 2               |
| Creatinine        | 5               | 8               | 6               |

Note: LDH = lactate dehydrogenase; AST = aspartate aminotransferase; ALT = alanine aminotransferase; GGTP = gamma-glutamyl transpeptidase; BUN = blood urea nitrogen.
activity is an increase in levels of HDL-C (Haskell, 1984). In a longitudinal study involving only moderate physical activity, Peltonen, Marnniemi, Hietanen, Vuori, and Ehnholm (1981) found a significant increase in HDL-C after just 15 weeks. Although the value of aerobic exercise to serum lipids has been well documented, our user group showed no such effect on HDL-C levels perhaps because of the severe depression of the values. This lack of response to exercise may have been precluded by an effect of anabolic steroids or other individual factors, such as diet or baseline lipid concentrations. It is also well known that metabolic responses to physical training are subject to genetic variation. Previous research has demonstrated higher HDL-C levels in body builders but not power lifters (Hurley, Seals, and Hagberg, 1984).

Anabolic steroid use, when associated with weight training, is thought to result in greater gains in muscular strength and body weight as opposed to training alone (ACSM, 1984). Results from the current study support these contentions. Participants in the users-on group reported a 7% increase in their maximum bench press lift and a 12% gain in their maximum squat lift after 5–8 weeks of steroid use. This information was obtained through pre-investigation training questionnaires but was not directly assessed. However, possible strength benefits from use of anabolic steroids do not occur without side effects. It should be emphasized, particularly in the user group, that there seems to be evidence not only for adverse lipid effects but also widespread liver enzyme and renal abnormalities. These hepatic and renal effects have been observed in several other reports (Lenders et al., 1988; Strauss, Wright, Finerman, & Catlin, 1988).

The discrepancy between the drug questionnaire and urine analysis in one user probably is explained by the impurity of many anabolic preparations obtained on the black market. Six anabolic steroid preparations were sent to the Paul Ziffren Olympic Analytical Laboratory for identification and purity analysis. No anabolic steroids were detected in two of these samples, suggesting inaccurate labeling on many preparations of this type. Also revealed is the superiority of urinalysis to participative questionnaires in evaluating anabolic steroid use.

The effects of anabolic steroids are difficult to study, as ethics preclude the use of true experimental designs (i.e., randomization). This study was also limited by the wide variety and amounts of drugs used by participants and their failure to control diet and alcohol consumption. Consequently, the results should be interpreted with caution.

The use of anabolic steroids has become widespread, even among young adults (Terney & McLain, 1990; Windsor & Dumitru, 1989). Continued use of such drugs may predispose young people to early onset of cardiovascular disease. Our results demonstrated the significant deviation in serum lipid values in weight trainers using anabolic steroids as compared to a normal weight-lifting control group. Effect sizes were generally large, ranging from 8–2.2, where significant differences occurred. It was also demonstrated that alterations in lipoprotein and apolipoprotein values remain in a depressed state after a prolonged period of abstinence from steroid use. Our results suggest that long-term use of anabolic steroids, especially in athletes exposed to their atherogenic effects over long periods by frequent cycling protocols, have potentially deleterious effects on serum lipids. It follows that these athletes appear to be at risk for the development of early atherosclerosis.

References


**Authors’ Note**

Please address all correspondence regarding this article to Kris Berg, School of Health, Physical Education, and Recreation, University of Nebraska at Omaha, Omaha, NE 68182.