

Reduction of Plasma Cholesterol Levels in Male Retired Breeder Rats by an Anabolic Steroid, Oxandrolone (37698)

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Oxandrolone (17 β -Hydroxy-17 α -methyl-2-oxa-5 α -androstan-3-one), a synthetic steroid-like lactone, has six times the anabolic and one-third the androgenic activity of methyl testosterone (1). Like certain other anabolic steroids, oxandrolone exerts a hypolipidemic effect. In several clinical trials oxandrolone effectively lowered plasma triglycerides of most patients with Types III, IV, and V hyperlipoproteinemia, but produced little or highly variable effects on their plasma cholesterol levels (2, 3). For this and other reasons, oxandrolone currently is regarded primarily as a triglyceride-lowering agent, and much work is being devoted to its effect on triglyceride removal mechanisms (4-6). As a result, very little attention has been given to oxandrolone's hypocholesterolemic properties.

The purpose of the present paper is to report the effect of oxandrolone on the plasma cholesterol levels of male retired breeder rats as well as to draw attention to the advantages of using retired breeder rats for studies of hyperlipidemia.

Materials and Methods. Since oxandrolone is administered orally to human patients, we used this route of administration. Preliminary experiments revealed that rats will readily eat the desired dosages of oxandrolone, an exceedingly insoluble compound, when the steroid is mixed with 250 mg corn oil margarine (Fleischmann brand). The ingestion of this amount of margarine vehicle per day for 21 days produced no significant effect on the plasma lipid levels of the rats and circumvented the hazards associated with oral intubation (unpublished data).

Thirty-two 500-g male albino, recently re-

tired breeder rats (Holtzman Co., Madison, WI) were divided into two groups and housed in individual cages. They were allowed to consume Purina Laboratory Chow (Purina Ralston Co.) and tap water *ad libitum*. Each member of the control group received 250 mg corn oil margarine per day, while each member of the experimental group was fed 250 mg corn oil margarine mixed with 100 mg oxandrolone/kg body weight per day.

Six 200-g male albino rats (Holtzman Co., Madison, WI) housed in individual cages also were allowed to consume Purina Laboratory Chow and tap water *ad libitum*. Each rat in this group received 250 mg corn oil margarine per day.

The rats were weighed daily. On the morning of Day 21, food was removed from all animals. Tap water was allowed *ad libitum*. The last dose of margarine or margarine plus oxandrolone was given 12 hr later. The animals were anesthetized with ether and killed by decapitation on the morning of Day 22, *i.e.*, 12 hr after the last dose and 24 hr after the initiation of the fast. Plasma samples were collected, extracted immediately in chloroform-methanol (2:1), and analyzed for cholesterol according to the method of Rudel and Morris (7).

Results. The results summarized in Table I show that the old male recently retired breeder rats had higher plasma cholesterol levels (91.3 mg/100 ml) than the young adult males from the same strain (65.2 mg/100 ml). Oxandrolone exerted a profound hypocholesterolemic effect on the plasma of the retired breeder rats. The 41% reduction in plasma cholesterol produced during the 21

TABLE I. Effect of Oral Administration of Oxandrolone on Plasma Cholesterol Levels of Male Retired Breeder Rats.

Group	Oxandrolone (100 mg/kg/ day)	No. of rats	Mean plasma cholesterol (mg/100 ml)	Standard error of the mean	% Effect of oxandrolone
Retired breeder, 500 g	0	16	91.3	6.547	—
Retired breeder, 500 g	+	16	53.9 ^a	2.595	41
Young adult, 200 g	0	6	65.2 ^b	2.141	—

^a $p < 0.001$ vs retired breeder control and < 0.02 vs young adult control.

^b $p < 0.05$ vs retired breeder control.

days of oxandrolone treatment resulted in more than a statistically significant drug effect: it returned the plasma cholesterol concentration of the old retired breeder rats to a level significantly below that of the young adults (*i.e.*, 53.9 mg/100 ml). No differences in body weight were found between the oxandrolone-treated and control retired breeder rats during the course of this experiment.

Discussion. The observations that plasma lipids increase with age in the rat and are elevated in repeatedly bred rats are not new (8, 9). Boberg *et al.* (10) have presented data indicating that in male rats serum phospholipids and cholesterol increase steadily from birth to 20 months of age, while triglycerides increase from birth to 10 months and then plateau. Unfortunately, the latter workers did not mention the breed or strain of rats used in their investigation, and the lipid profiles on retired breeder rats are incomplete; hence, we are now attempting to document these changes in our animals.

In our study the young 200-g male rats were an average of 50 days old at the beginning of the experiment, whereas the 500-g retired male breeder rats were at least 6 months of age. According to our data, then, the mean plasma cholesterol level of male Sprague-Dawley Holtzman strain rats increases 1.4-fold from the second to the sixth month of age. The extent to which repeated breeding *per se* elevates plasma lipids in male rats above that normally accrued by aging is under study in our laboratory. In any event, an increase in plasma lipid concentration with age, similar to that occurring in the male rats, also has been observed in many human populations (11). These findings suggest that the male rat with its limited 2-year life span

may be an appropriate model for investigating the interrelationships between aging and blood lipids.

In this study, oxandrolone treatment for 21 days reduced the plasma cholesterol levels of the male retired breeder rats to a level below that of the young adult males. The lowering is statistically significant and, to our knowledge, represents an unprecedented reduction of plasma cholesterol levels by oxandrolone in man or experimental animals in the absence of dietary manipulation or overt disease. Others have reported that oxandrolone lowers the plasma cholesterol levels of young (170–180 g) female rats fed high cholesterol diets, but does not lower blood cholesterol in rats of the same age-weight group when they are fed low cholesterol diets or are rendered hypercholesterolemic via Tween 80 or alloxan diabetes (12).

Recently, Lavietes *et al.* (13) suggested that a rise in serum lipids of men is not obligatorily coupled to aging, but is directly related to weight gain during adult life. They observed that men who stabilized their weight over a period of 30 years did not have significant elevations in their plasma cholesterol. The results of our study are of interest in this regard because there was no difference in the rate or amount of weight gained by the oxandrolone-treated retired breeder rats and their corresponding controls during the 21-day period. Additional experiments will be required to elucidate the mechanism of action of oxandrolone and the extent to which the oxandrolone-induced lowering of cholesterol is maintained with increasing age and weight. In one long-term study, the serum cholesterol levels of weight-stable geriatric patients eventually escaped the hypocholesterole-

lemic action of oxandrolone (14). Nevertheless, even a temporary decrease of serum cholesterol levels may have therapeutic value in certain critically ill individuals. Hence, the ultimate aim of our future investigations will be to answer: what is the *biological* significance of the oxandrolone-induced lowering of plasma cholesterol in retired male breeder rats and by what mechanism(s) is it accomplished.

Summary. The plasma cholesterol levels of 6-month-old 500-g male retired breeder rats were significantly higher than the plasma cholesterol levels of 71-day-old 200-g adult male rats of the same strain. Oxandrolone administration daily for 21 days reduced the plasma cholesterol of the retired breeder rats 41% to a level below that of the young adult males. There was no difference in weight between the oxandrolone-treated retired breeder rats and their controls. The hypocholesterolemic properties of oxandrolone and the virtues of retired breeder rats as an experimental model are discussed.

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