Lipoprotein metabolism in the ovariectomized rat

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Abstract The hyperlipoproteinemia observed after ovariectomy in rats was previously shown to be associated with increased concentrations of cholesterol, triglycerides, and apolipoproteins B, E, and C. In the present study, it was shown that increases in low density lipoproteins and high density lipoproteins were almost entirely responsible for the changes in plasma lipids and apolipoproteins after ovariectomy. The size of the low density lipoproteins and high density lipoproteins isolated from the plasma of ovariectomized rats as determined by agarose chromatography appeared to be somewhat different from that of control rats. Specifically, the apolipoprotein B appeared to be associated with somewhat smaller particles, whereas the apolipoprotein E from those rats appeared to be associated with larger particles than that of control rats. To determine the mechanism for the increased plasma low density lipoproteins, apolipoprotein B pool sizes and turnover rates were calculated and compared. In addition to an increased mass of low density lipoproteins in ovariectomized rats, the turnover rate of low density lipoproteins was increased almost twofold, indicating an increased low density lipoprotein synthesis and catabolism in those animals. We postulate that the increased low density lipoprotein levels of ovariectomized rats are due to an initial increased production of low density lipoproteins, followed by an enhanced catabolism of low density lipoproteins to establish a steady state at higher plasma low density lipoprotein concentrations.—**Van Lenten, B. J., G. W. Melchior, and P. S. Roheim.** Lipoprotein metabolism in the ovariectomized rat. *J. Lipid Res.* 1983. **24:** 1475–1484.

Supplementary key words hyperlipoproteinemia • low density lipoprotein • ovariectomy • lipoprotein synthesis

Although there is a great deal of literature on the possible role of sex hormones in the development of cardiovascular disease, a cause and effect relationship remains to be established. Both the natural (1) and surgical (2) menopause have been associated with an increased risk of CHD, but the mechanisms by which estrogen deficiency could cause or accelerate the progression of CHD are not clear.

We previously showed in rats (3) that adult females have lower plasma concentrations of apoB, cholesterol, and triglyceride than males, which parallels the human sex differences (4–6). Ovariectomy in the rat resulted in elevations of these same plasma constituents (7), supporting gonadal influences on plasma lipoprotein metabolism. In the present study, we have characterized the hyperlipoproteinemia of OVX rats and compared the turnover rates of ^{125}I-labeled LDL in OVX and in sham-operated control rats. The results show that the hyperlipoproteinemia of OVX rats is due, in part, to an increased synthesis of LDL.

MATERIALS AND METHODS

Animals

In experiments involving the chemical characterization of the plasma lipoproteins, 48 rats were used. To study the effects of ovariectomy on lipoprotein metabolism in rats, two identical studies were performed. Thirty rats were used for Study I, and 36 rats for Study II. In all studies, 12-week-old female Wistar rats weighing 230–240 g were divided into two equal groups. A bilateral ovariectomy was performed on one group whereas the other group received a sham operation. The animals were allowed to recover for 30 days after the surgery during which time they were fed Purina rat chow (Ralston Purina Company, St. Louis) ad libitum. The rats were weighed weekly and vaginal smears were performed to confirm that the estrus cycle had been interrupted in OVX rats. At the time of the turnover studies, the body weights had increased to 255 ± 4 g in controls and to 292 ± 4 g in OVX rats. The animals were fasted for 14 hr and exsanguinated from the abdominal aorta under light ether anesthesia. Atrophic uteri documented the estrogen deficiency in the OVX rats.

Lipoprotein separation

Blood collected from the rats was allowed to clot and the serum was separated from the cells by centrifugation.

Abbreviations: CHD, coronary heart disease; VLDL, very low density lipoproteins; IDL, intermediate density lipoproteins; LDL, low density lipoproteins; HDL, high density lipoproteins; OVX, ovariectomized rats; SDS-PAGE, sodium dodecyl sulfate polyacrylamide gel electrophoresis; QEIA, quantitative electroimmunoassay; A.U., arbitrary units; $apoB_l$, low molecular weight apoB; $apoB_h$, high molecular weight apoB; EDTA, ethylene diamine tetraacetic acid; TMU, tetramethylurea.

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EDTA (1 mg/ml serum) and sodium azide (1 mg/ml serum) were routinely added as preservatives. The lipoproteins were separated by ultracentrifugation at 15°C in a 40.3 rotor at 39,000 rpm as described by Havel, Eder, and Bragdon (8). The rotor was fitted with special adaptors (Beckman Instrument Co., Palo Alto, CA) so that lipoproteins from 1 ml of serum could be isolated. The lipoproteins were isolated by centrifuging separate serum aliquots as follows: VLDL, 18 hr at a solvent density of 1.006 g/ml; VLDL + IDL, 18 hr at a solvent density of 1.030 g/ml; VLDL + IDL + LDL, 20 hr at solvent density 1.063 g/ml; and total lipoproteins, 44 hr at solvent density 1.21 g/ml. The apolipoprotein concentrations were measured in the infranatants of the lipoprotein fractions separated by ultracentrifugation and thus the apoprotein content of a given fraction was determined by subtracting the apoprotein content of its infranatant from that of the preceding infranatant; e.g., the apoprotein content of the IDL was obtained by subtracting the d 1.030 g/ml infranatant from the d 1.006 g/ml infranatant apoprotein concentration. The use of the infranatant has the additional advantage that the apoproteins are measured in their native milieu (9), rather than in an artificial medium as would be the case were the supernatant fractions used. Although this procedure required four separate serum aliquots for a complete lipoprotein profile, it substantially reduced losses since each fraction was centrifuged and sliced only once.

Rat serum was also fractionated using agarose gel chromatography on a 0.9 × 90 cm column of 6% agarose (Bio-Gel A-5M, 200–400 mesh, Bio-Rad Laboratories, Richmond, CA). Chromatography was carried out on 2-ml samples of freshly separated serum using 0.15 M NaCl, 0.01% EDTA, 0.01% sodium azide, pH 7.4, as equilibrating and running buffer. The column was operated at 23°C and calibrated using blue dextran and the d < 1.210 g/ml fraction of human serum. The void volume was calculated to be 25.3 ml and a flow rate of 5.3 ml/hr was maintained for all runs with 1.3 ml/hr collection. Human ^{125}I-labeled LDL and ^{125}I-labeled albumin were added to each serum sample as internal standards. ^{125}I-labeled LDL consistently peaked at fraction #34, and the peak for the radioactive albumin was at tube #47.

Analytical methods

Cholesterol and triglyceride concentrations were determined enzymatically by the methods of Allain, et al. (10) and Bucolo and David (11), respectively. Protein was determined using the technique of Lowry et al. (12), using bovine serum albumin as the standard.

Apolipoproteins were measured in ultracentrifuge and column fractions by QEIA according to the technique of Laurell (13) as modified by Bar-On, Roheim, and Eder (14). A non-ionic detergent, Nonidet 40 (NP 40), was added to a final concentration of 1% in the samples and standards, and 0.05% in the agarose for apoE and apoA-I measurements (9).

The antisera used in this study were prepared as previously described (14). Apolipoprotein values from the determinations, expressed as A.U., were calculated as percentages of a rat plasma standard pool run simultaneously on the plates with the samples and a secondary standard. As used here, 100 A.U. indicates that the concentration of the apoprotein in the sample being assayed was the same as in the standard pool.

In those instances where the apoprotein content of isolated fractions was measured (the apoB content of LDL, for example), the fraction was mixed with fresh rabbit serum prior to the assay.

The apolipoprotein concentrations were also estimated using SDS-PAGE (15) of the d < 1.210 g/ml fraction. The SDS-PAGE was carried out using a Hoefer tube gel apparatus (Hoefer Scientific Instruments, San Francisco, CA). The apoproteins were stained with Coomassie brilliant blue for 2 hr at room temperature and destained in 9% acetic acid overnight (18 hr). The gels were scanned with an E-C densitometer (E-C Apparatus Corp., Indianapolis, IN). Ten-percent gels were used to separate the major apoproteins (apoB, apoA-IV, apoE, apoA-I, and the C apoproteins); 3.5% gels were used to subfractionate the apoB into the high molecular weight apoB (B_h) and the low molecular weight apoB (B_l) (16–18). Total apoB mass was determined by TMU precipitation (19).

LDL turnover

Rat LDL (d 1.030–1.050 g/ml) was isolated by preparative ultracentrifugation using an SW41 rotor. The lipoproteins were washed once by reflotation and dialyzed against 0.15 M NaCl, pH 7.4. The purity of the fraction was checked by agarose electrophoresis (20) and a single β-migrating band was present.

For the iodination procedure, the LDL solutions contained 2–3 mg of protein in 2–3 ml of saline and were diluted with an equal volume of 1 M glycine buffer (pH 10.0) just prior to iodination. The lipoproteins were radiolabeled with ^{125}I as described by McFarlane (21) and modified by Bilheimer, Eisenberg, and Levy (22). Iodine monochloride (3.3 mM, 3 μl/mg protein) was added to 5 mCi carrier-free Na ^{125}I (New England Nuclear, Boston, MA) dissolved in 0.5 ml 1 M glycine buffer (pH 10.0). The ^{125}ICl solution was aspirated into a Pasteur pipette and forcibly ejected into the protein solution while the latter was being mixed. Free iodine was then removed by dialysis in saline containing EDTA (pH 7.4). The radioactivity was measured using a Beckman

Gamma 4000 well counter (Beckman Instruments). Ninety-two to 96% of LDL radioactivity was precipitated by trichloroacetic acid, and 8 to 16% was recovered in the chloroform phase after extraction (23).

SDS-PAGE of the ^{125}I-labeled LDL preparation showed that 80% of the protein radioactivity was in apoB, of which 93% was in apoB$_h$. The majority of the remaining radioactivity was associated with the C apoproteins. Flotation of the ^{125}I-labeled LDL at d 1.063 g/ml resulted in 85% of the radioactivity being recovered in the supernatant.

The radiolabeled LDL (300,000 cpm/100 g body weight) was injected into the tail veins of the rats. The animals were killed by exsanguination at 1.5, 3, 6, 12, and 24 hr (Study I); or 5 min, 30 min, 1, 4, 10, and 18 hr (Study II) after isotope administration. Three rats were killed at each time point. Four ml of serum from each rat was used for LDL isolation.

The d 1.030–1.063 g/ml fractions (LDL) were dialyzed against 0.15 M NaCl, 0.01% sodium azide, pH 7.4, and brought up to equal volumes (2 ml). Equal aliquots (200 μl) of the LDL from each rat were applied to 3.5% SDS-PAGE gels to separate the major apoB bands from other apolipoproteins. Gels were stained in 0.1% Coomassie blue and the bands were cut out and counted. In this way, the counts in apoB$_h$, apoB$_l$, and in other apolipoproteins could be determined and directly compared from animal to animal.

The radioactivity in the lipids was determined as described by Mans and Novelli (24). The apoB content of each LDL fraction was determined by electroimmunoassay as described above. Thus, although radioactivity was found in other apoproteins (most of which apparently exchanged freely among the various lipoprotein fractions), our results were not affected since we followed changes in the LDL apoB specific activity only. The apoB specific activity averaged 2.5×10^8 cpm/mg of apoB.

Calculations

The Log$_e$ of the normalized apoB specific activity (cpm/apoB A.U. [as determined by QEIA] ÷ dose) was plotted against time (t) and the resulting curves were described by the equation:

$$Y = \sum_{i=1}^{N=2} A_i e^{-\alpha_i t} \qquad \text{Eq. 1}$$

where Y is the normalized apoB specific activity, A_i is the intercept and α_i the slope of each exponential term. The turnover of LDL apoB (R_t) was determined by Steward-Hamilton analysis (25) where:

$$R_t = \frac{1}{\int_0^\infty Y\,dt} \qquad \text{Eq. 2}$$

The mean transit time ($\bar{t}$) was determined from the equation:

$$\bar{t} = \frac{\int_0^\infty t \cdot R_t dt}{\int_0^\infty R_t dt} \qquad \text{Eq. 3}$$

The total traced mass (M_T) was determined from the equation:

$$M_T = R_t \cdot \bar{t} \qquad \text{Eq. 4}$$

The mass of apoB in the rapidly miscible compartment (M_a; the compartment which includes the plasma) was determined from the equation:

$$M_a = \frac{1}{A_1 + A_2} \qquad \text{Eq. 5}$$

The mass determined in this way is in A.U. and the turnover is in A.U./hr. Since A.U. represents a direct measure of apoB mass, this method provides a valid comparison of the apoB mass and turnover rate in control and OVX rats.

Comparisons between control and OVX groups were tested for significance using the independent sample *t*-test. The null hypothesis was rejected if $P < 0.05$.

RESULTS

Lipoprotein and apoprotein concentrations

Table 1 shows the effects of ovariectomy on the total lipoprotein protein concentrations, the TMU-precipitable protein concentrations, and the concentrations of the major apoproteins as estimated by densitometry. In general, the results are in good agreement with the changes in apoprotein levels measured by QEIA reported previously (7). After ovariectomy, the mean increase in those apoproteins measured was 55%. This was due to an increase in apoB, apoE, apoA-I, and apoC levels. The change in apoA-IV levels was not significant.

Apolipoprotein and lipid distribution

Fig. 1 depicts the distribution of plasma cholesterol and triglyceride in sham-operated and OVX rats. OVX rats exhibited a slight but significant increase in VLDL triglyceride (23%) (Fig. 1B); however, LDL cholesterol was doubled (Fig. 1A).

Fig. 2 shows the distribution of apoB, apoE, apoA-I, and apoA-IV among four lipoprotein fractions separated by ultracentrifugation. The majority of the increase in apoB (Fig. 2A) was in the LDL. A 120% increase in IDL apoB (Fig. 2A) further contributed to the increase in total apoB concentrations after ovariectomy.

TABLE 1. Concentrations of the major apolipoproteins in the d < 1.210 g/ml fractions of sham-operated and ovariectomized rats[a]

	Sham-Operated	Ovariectomized
	mg/dl	
ApoB[b]	6.8 ± 0.8[c]	9.7 ± 0.3*
ApoA-IV[d]	4.4 ± 0.1	6.4 ± 1.5
ApoE[d]	9.2 ± 0.3	14.1 ± 1.2*
ApoA-I[d]	21.7 ± 1.8	31.7 ± 5.0*
ApoC[d]	8.9 ± 0.2	17.4 ± 3.2*

[a] N = three pools of eight rats per pool.
[b] TMU-precipitable protein in d < 1.210 g/ml fraction.
[c] Values are mean ± SEM.
[d] Proportion of TMU-soluble protein determined by densitometry.
* $P < 0.05$.

The increases in the apoE levels (Fig. 2B) appeared to be distributed among all of the lipoprotein density fractions with the major increases in the HDL and VLDL. A significant quantity, however, of the apoE (18–26%) was recovered in the d > 1.21 g/ml fraction, most likely as a result of ultracentrifugation (26), since it was not present in the lipoprotein-free fraction from the column shown below. It is not known from which lipoprotein fractions this apoE was lost.

To employ a method of lipoprotein separation that would avoid ultracentrifugal artifacts (26), 5M agarose gel chromatography was used. **Fig. 3A** shows increases of apoB-containing particles of smaller average diameter in the plasma of OVX rats than found in sham-operated control rats. In Fig. 3B, the elution profile for apoE reveals that the increase is predominantly associated with the larger apoE-containing particles. No significant changes were observed in the elution profiles for either apoA-I (Fig. 3C) or apoA-IV (Fig. 3D).

LDL turnover

To determine the mechanism by which ovariectomy increases the LDL concentration, LDL from control rats was radiolabeled and injected into both control and ovariectomized recipients. Semilog plots of the normalized LDL-apoB specific activity, followed for 24 hr in both control and OVX rats, are shown in **Fig. 4.** Both curves were biphasic indicating that the administered LDL equilibrated with two metabolic compartments. Visual comparison of the two curves showed that the apoB specific activity in OVX rats was significantly lower than that of control rats at almost every time point, clearly an effect of the larger quantities of LDL apoB present in those animals (Fig. 2A). Furthermore, the time required for the apoB specific activity to reach its final exponential rate of decay was delayed in ovariectomized rats. Nonetheless, the rate constant for the second component of the equation (α_2) was not significantly different in the two groups (0.135 in controls vs. 0.147 in OVX rats; Fig. 4).

Mathematical analysis of the two curves by standard techniques (25) showed that the total mass of LDL apoB (M_T) and the mass of the LDL apoB in the rapidly miscible compartment (M_a) were increased in the OVX rats (**Table 2**). However, the mean transit time ($\bar{t}$) was more

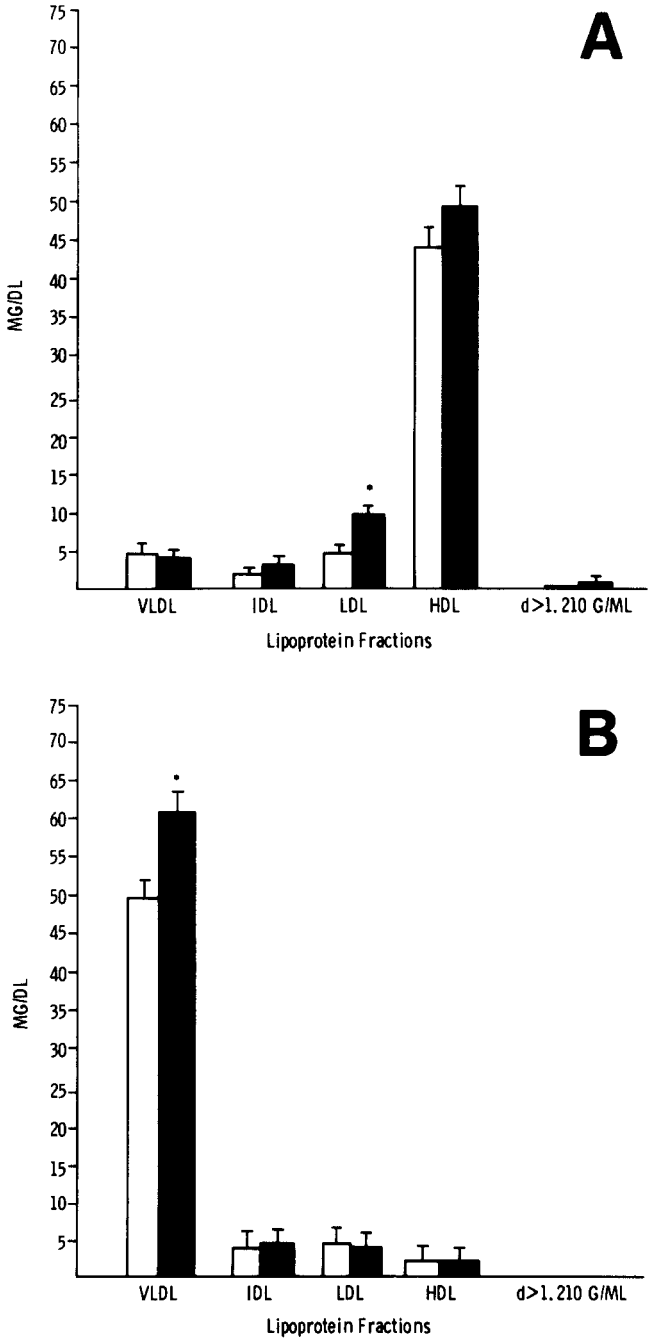

Fig. 1. Distribution of plasma cholesterol (A) and triglyceride (B) among the lipoprotein fractions of sham-operated [□] and OVX [■] rats. Values based on six pools of four rats per pool. *, $P < 0.05$.

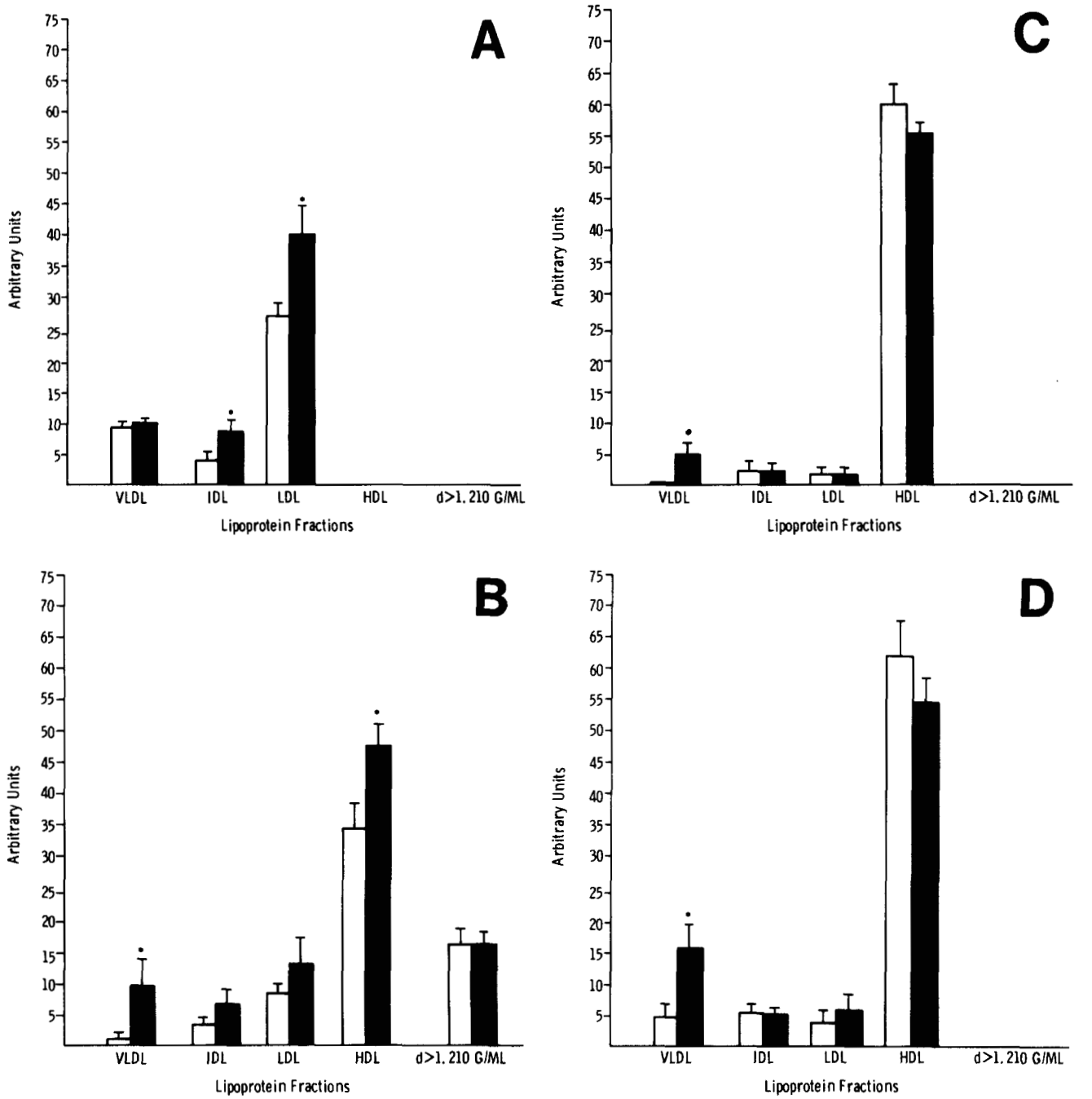

Fig. 2. Distribution of plasma apoB (A), apoE (B), apoA-I (C), and apoA-IV (D) among the lipoprotein fractions of sham-operated [□] and OVX [■] rats. Values based on six pools of four rats per pool. *, $P < 0.05$.

than an hour less in OVX animals than in controls (Table 2), indicating that the fraction of the LDL pool turning over per unit time was greater in OVX animals. Thus, the absolute turnover of LDL apoB (R_T) was 1.7 times greater in OVX rats than in controls.

When the radiolabeled LDL proteins were separated by SDS-PAGE on 3.5% gels, 7% of the radioactivity in apoB was found to be associated with $apoB_l$, and this radioactivity distribution between $apoB_h$ and $apoB_l$ did not change significantly in either group during the ensuing 24 hr (**Fig. 5**). Thus, the intravascular–extravascular distribution, and the turnover of the B_l isolated from the 1.030–1.063 g/ml fraction was the same as that of the B_h in both normal and OVX rats, and the curves in Fig. 4 can be used to describe the turnover of either form of apoB.

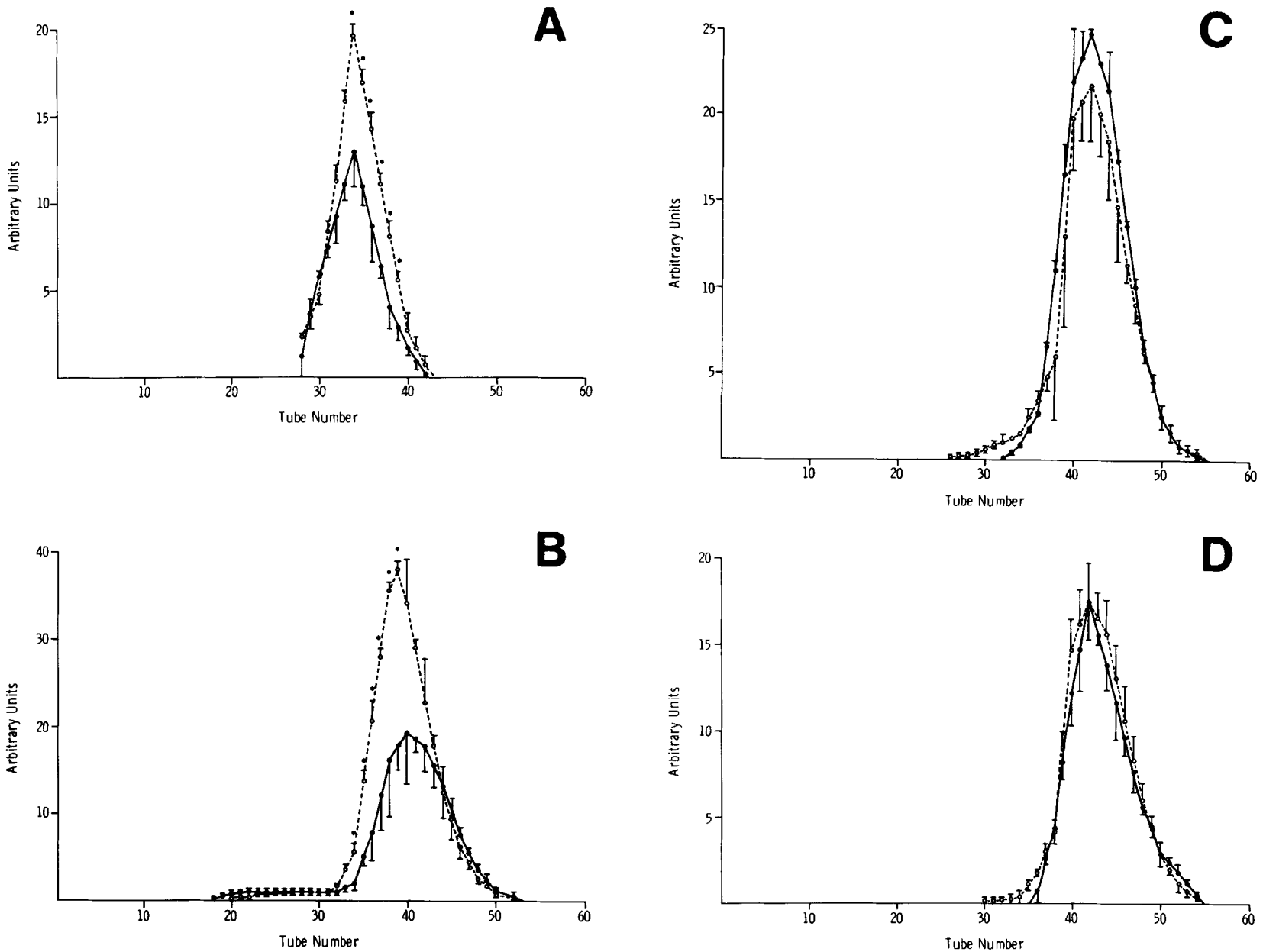

Fig. 3. 5M agarose gel chromatography elution profiles for apoB (A), apoE (B), apoA-I (C), and apoA-IV (D) of sham-operated [●] and OVX [○] rats. Values based on three rats per group. Human ^{125}I-labeled LDL and ^{125}I-labeled albumin were added to each serum sample as internal standards. ^{125}I-labeled LDL consistently peaked at tube #34, and the peak for the radioactive albumin was at tube #47.*, $P < 0.05$.

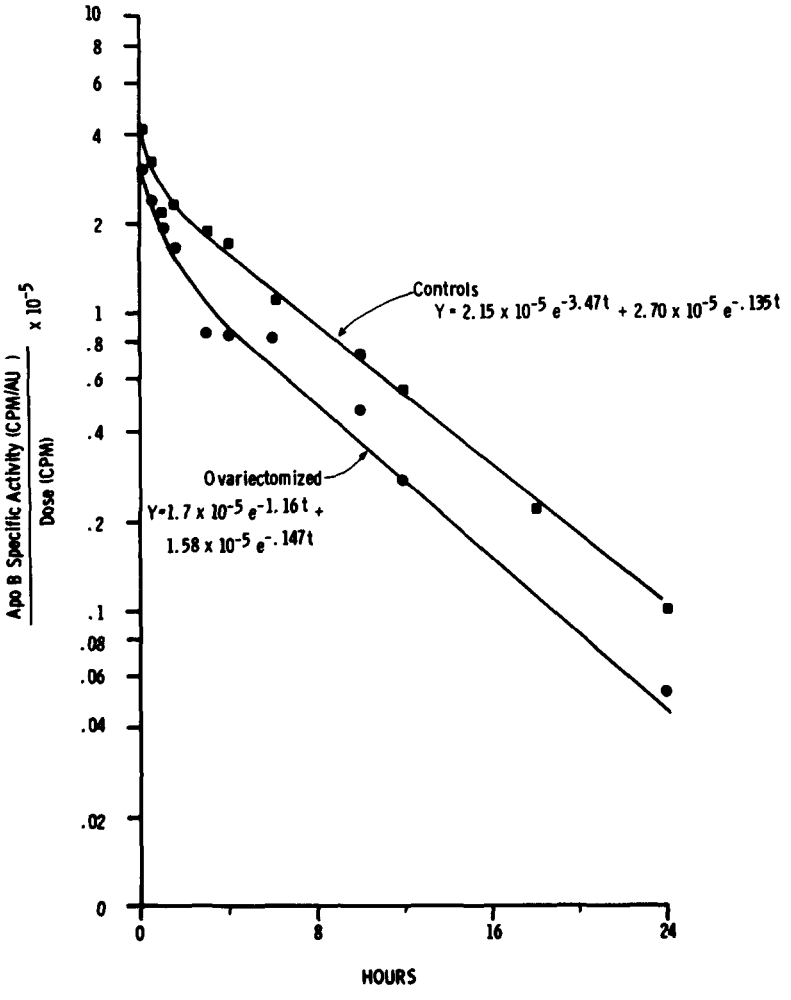

Fig. 4. The specific activity decay of ^{125}I-labeled LDL from the plasma of control [■] and OVX [●] rats. Each point represents the mean value from a subgroup of three rats killed at that time. In each case the points were adequately fit by the sum of two exponential terms and the equation describing each curve is shown.

DISCUSSION

Ovariectomy of the adult rat results in hyperlipoproteinemia. We previously showed (7) that, in addition to increased cholesterol and triglyceride concentrations, apolipoproteins B, E, and C were increased in the plasma, whereas apoA-I and apoA-IV changed little or not at all. In the present study, these results were confirmed and extended to show that increases in LDL and HDL contributed to the changes observed in plasma lipids and apolipoproteins.

When ultracentrifugal isolation and agarose column chromatography were compared, some variations of apoprotein distribution were observed that were clearly due to differences in the methods of separation. The chromatographic elution patterns for apoB and apoE showed increases in the smaller apoB-containing particles and the larger apoE-containing particles, suggesting that the LDL and HDL may overlap to a greater extent in OVX rats. That is to say, ovariectomy may result in the formation in the plasma of a spectrum of apoB- and apoE-containing particles not normally present in female rats. Davis et al. (27) have shown in rats that removing the influence of

TABLE 2. The total traced mass, mass of the rapidly-miscible pool, mean transit time, and turnover rate of LDL apoB in ovariectomized and control rats

	M_T[a]	M_a[a]	$\bar{t}$[b]	R_T[c]
Ovariectomized	5.0	3.0	6.0	8.2
Control	3.5	2.1	7.2	4.9
Ovariectomized/Control	1.4	1.5	0.8	1.7

[a] The total traced mass (M_T) and mass of the rapidly-miscible pool (M_a) in arbitrary units × 10^{-4}.
[b] The mean transit time in hours.
[c] The turnover rate in arbitrary units/hr ($\times 10^{-3}$).

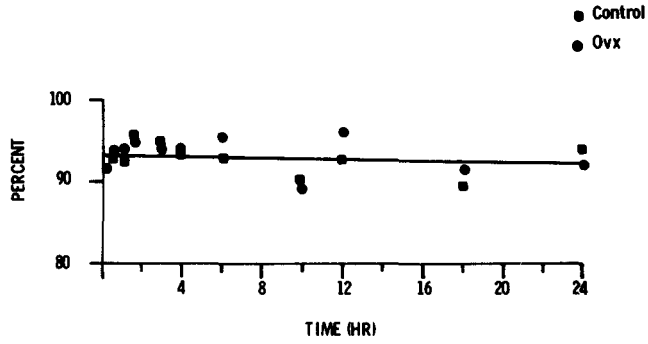

Fig. 5. The percent of the total LDL-apoB radioactivity migrating with $apoB_l$ during SDS-PAGE in 3.5% gels.

the liver by hepatectomy results in the accumulation of apoE-containing particles larger than apoA-I-rich HDL, but smaller than LDL. Patsch et al. (28) have also presented data indicating that a shift in the apoE distribution occurs in OVX female rats. The majority of the increase in plasma apoE levels was in the apoE-containing HDL (Fig. 2B), and the larger of these particles showed the greater increase (Fig. 3B), suggesting that HDL_1 might be the fraction of the apoE-containing HDL most susceptible to decreased estrogen levels.

To determine the mechanism responsible for the increased apoB concentrations in OVX rats, we compared the LDL-apoB pool sizes and turnover rates in control and OVX rats. Analysis of the apoB specific activity time-course curves confirmed that the total mass of LDL was increased in OVX rats. Thus, the increased plasma LDL levels in those animals were not due simply to a redistribution of LDL between the intra- and extravascular compartments. Furthermore, the turnover rate of LDL was increased 1.7-fold in OVX rats, which indicates that both the LDL synthetic and catabolic rates were increased in those animals, assuming the presence of a steady state.

Based on this observation, we postulate that the initial event leading to the increased LDL levels in OVX rats was an enhanced synthesis, since an increased catabolic rate would be expected to decrease, not increase, the LDL levels. The LDL levels presumably continued to increase until the catabolic rate was eventually able to compensate. At that point a new steady state was established at the higher LDL levels. These observations were based on the assumption that the control LDL was catabolized in ovariectomized rats by the same process as their native LDL; i.e., that the tracer was in fact tracing the turnover of LDL in ovariectomized rats. Fig. 3 shows that the distribution of LDL from ovariectomized rats was slightly different from that of control rats; however, we have no reason to assume that this affected their ability to compete with the tracer for the catabolic sites. Nonethelss, it is possible that the ovariectomized LDL were in fact heterogeneous, and that we were over- or underestimating LDL turnover to some extent in those animals.

Because the specific activity of LDL apoB was expressed as cpm/A.U. of apoB (Fig. 3), the data in Table 2 give only a comparison of the pool sizes and turnover rates between OVX and control rats. However, a reasonable estimate of these values in absolute units (mg and mg/hr, respectively) can be made using the data in Table 1 and Fig. 4 as follows: Table 1 shows that the plasma apoB concentrations were 6.8 and 9.7 mg/dl in control and OVX rats, respectively, 68% of which was in LDL apoB (Fig. 2A). Thus, the LDL-apoB concentrations averaged 4.6 mg/dl in controls and 6.7 mg/dl in OVX rats. the apoB mass, as estimated by QEIA, averaged 7.37×10^4 A.U./dl in controls, and 10.7×10^4 A.U./dl in OVX rats. Therefore, a conversion factor can be obtained by dividing the A.U./dl by apoB mass/dl which, for these studies, is approximately 1.6×10^4 A.U./mg of apoB. Applying this conversion factor to Table 2, one obtains (OVX vs. controls): M_T, 3.1 vs. 2.2 mg; M_a, 1.9 vs. 1.3 mg; R_t, 0.5 vs. 0.3 mg/hr.

The increased LDL synthesis may indicate that VLDL synthesis and secretion in general are increased, or that just the synthesis of those VLDL destined to become LDL is increased. Rats differ from humans in that a comparatively small fraction of the total VLDL apoB secreted by the liver is retained in the plasma as LDL apoB (29). The metabolic basis for this difference between rats and humans is not known, although it indicates that rat VLDL are more heterogenous; i.e., a mixture of VLDL, some of which is rapidly cleared from the plasma after triglyceride removal, and some of which is converted to circulating LDL. Ovariectomy may, therefore, stimulate the production of those VLDL destined to become "long-lived" LDL. Alternatively, there may be a direct effect of decreased estrogen levels on the interconversion of VLDL to LDL or direct secretion of LDL by the liver. Hamosh and Hamosh (30), for example, observed in rats that the adipose tissue lipoprotein lipase activity was significantly increased after ovariectomy.

It should be noted at this point that although the classic interpretation of the biphasic specific activity time-course curves has been that the two phases of the curves represent equilibration of the tracer with the intravascular and extravascular pools of tracee, a nonhomogeneous tracer (in this case LDL apoB) could produce similar results; i.e., the two phases of the curve could be due to subspecies of the tracer turning over at different rates. Two subspecies of apoB that are metabolically different have been demonstrated to exist in the rat (17, 18). $ApoB_l$ turns over more rapidly than does $apoB_h$. About 7% of the radioactivity in the plasma at any time was associated with

apoB_l in the present study, and the ratio of radioactivity in the two subspecies of apoB did not change significantly with time (Fig. 5). Thus, it does not appear likely that the presence of B_l in our samples had a significant effect on the shape of the specific activity decay curves. Our data, showing that the turnover rates of LDL apoB_l and LDL apoB_h were identical, did not contradict those previous reports, but simply suggested that during conversion of VLDL to LDL in the rat, some apoB_l is retained and, thereafter, its turnover is identical to that of the LDL particle.

Women experience an elevated plasma cholesterol after the menopause due to an increase in LDL cholesterol (1, 2). Rats in our study also showed an increased LDL cholesterol after ovariectomy. Thus, changes in ovarian function may produce an altered lipoprotein metabolism resulting in higher plasma concentrations of LDL. However, the question remains as to whether the influence of the ovarian hormones is exerted directly or indirectly. Of the ovarian hormones, the administration of estrogen has major effects on lipoprotein metabolism, whereas, progesterone may act either synergistically or antagonistically to estrogen (31–33). The OVX rat is a good model for studying the effects of ovarian hormones on lipoprotein metabolism without the complications of existing endogenous hormones.

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