Turnover of Lipoprotein (a) in Man

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ABSTRACT An elevated concentration of lipoprotein (a) [Lp(a)] in the serum has been considered a risk factor for coronary heart disease by various investigators. In the present study, the turnover of Lp(a) was investigated in nine individuals with serum Lp(a) levels ranging from 1 to 68 mg/100 ml. After intravenous injection of radioiodinated Lp(a), the radioactivity time-curve of the serum and the specific activitity time-curves of the isolated Lp(a) and Lp(a) apolipoproteins were measured for 14 d. More than 97% of the label was found in the protein moiety of Lp(a). During the entire study period, the serum radioactivity remained with Lp(a), only insignificant amounts of radioactivity were detectable in other lipoprotein fractions. The serum radioactivity time-curves and the specific activity time-curves of the isolated Lp(a) and Lp(a) apolipoproteins were identical.

The kinetic parameters of Lp(a) turnover were calculated in terms of a two-compartment model. 76.5±5.1% (mean±1 SD) of total Lp(a) was contained in the intravascular space. The biological half-life of Lp(a) was 3.32±0.52 d, the fractional catabolic rate (FCR) was 0.306±0.054/d, and the rate of synthesis was 5.00±3.37 mg/kg/d. A positive correlation was found between serum concentration and synthetic rate of Lp(a) apoprotein. No relationship could be demonstrated between serum level and FCR of Lp(a).

The results of this study indicate that Lp(a) is not converted to other serum lipoproteins. From the correlations between serum concentration and kinetic parameters of Lp(a) it is concluded that an elevated Lp(a) level is the consequence of an increased Lp(a) apoprotein synthesis.

INTRODUCTION

When the lipoprotein (a) [Lp(a)][1] was first described (1), it was considered a genetic variant of low density lipoprotein (LDL). Other studies, however, have demonstrated that Lp(a) is a separate lipoprotein, although it exhibits some physicochemical and immunological properties that are similar to those of LDL. The lipid composition of these two lipoproteins, as determined by several investigators (2–4), is nearly identical. On the other hand, Lp(a) shows a several-fold higher content of hexose, hexosamine, and sialic acid than LDL (5). This may explain why Lp(a) migrates faster than LDL on agarose gel or cellulose acetate and, therefore, forms a separate band between β- and pre-β-lipoproteins, which was named "pre-β_1" band (6–8). The main apolipoprotein of Lp(a) has been found to be identical with apolipoprotein B of LDL (5, 9). Lp(a), however, contains an additional, specific Lp(a) antigen, which is dissociable from apolipoprotein B by several agents (5, 10). In contrast to apolipoprotein B, the specific Lp(a) antigen is soluble in 6 M urea (11). By ultracentrifugation Lp(a) is found in the fraction of density 1.055–1.110 g/ml (11), which is somewhat higher than the density of LDL and overlaps that of LDL and high density lipoproteins (HDL).

It is well documented that Lp(a) is present in the serum of all individuals with apolipoprotein B, although in different individuals its serum level may vary to a considerable degree (12, 13). Lp(a) has gained extensive clinical interest since a relationship between the concentration of Lp(a) in the serum and the occurrence of coronary heart disease has been postulated by several investigators (6, 7, 13, 14). Lp(a) has also been demonstrated by immunohistochemical methods in the intima of human arteries (15).

Although Lp(a) might play an important role in the development of atherosclerotic vascular lesions, only scarce information is available about the metabolism of this lipoprotein. In a previous study the half-life of radiomethylated Lp(a) has been determined (16). It has been shown recently that Lp(a) is not a metabolic product of other apolipoprotein B-containing lipoproteins (17). Therefore it has been suggested that Lp(a) is synthesized as a separate lipoprotein. No information, however, exists about the regulation of

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[1] *Abbreviations used in this paper*: FCR, fractional catabolic rate; HDL, high density lipoprotein; LDL, low density lipoprotein; Lp(a), lipoprotein (a); VLDL, very low density lipoprotein.

the serum concentration and the catabolic pathway of Lp(a).

METHODS

Subjects. The clinical data of the subjects are given in Table I. None of them revealed thyroid, renal, or hepatic dysfunction or hyperglycemia. All individuals were checked for Lp(a) by immunodiffusion with specific antisera to Lp(a) (see below). All subjects were "Lp(a)-positive" except subject No. 1. In this individual no Lp(a) could be detected by immunodiffusion and no measurable Lp(a) concentration was found using Laurell's one-dimensional immunoelectrophoresis (see below). All individuals were hospitalized during the investigation. They were kept on a "normal" hospital diet containing 45–50% of total calories as carbohydrates, 30–35% fat, and 15–20% protein. None of them received any treatment (except thiazide diuretics) known to influence lipid metabolism. Body weight and the serum concentration of Lp(a), total cholesterol, and triglyceride were constant throughout the study (Table I). Potassium iodide (3 × 60 mg daily) was administered to each individual beginning 3 d before the study and continuing throughout the entire experiment. Informed consent to the study was obtained from each subject.

Separation and iodination of Lp(a). Lp(a) was isolated according to the method of Ehnholm et al. (18). After an overnight fast, plasma was obtained by plasmapheresis using citrate-dextrose as anticoagulant and dialysed against 0.15 M NaCl containing Na_2EDTA (1 mg/ml) and NaN_3 (1 mg/ml). The plasma was then brought to a solution density of 1.055 g/ml by addition of solid NaCl and centrifuged at 42,000 rpm for 22 h. The densities of the sera were measured at 15°C with a calculating digital density meter (Anton Paar K.G., Graz, Austria). All ultracentrifugal procedures were performed in a Beckman L5-50 centrifuge with a Ti 50.2 rotor at 15°C (Beckman Instruments, Inc., Fullerton, Calif.). The top fraction was removed by tube slicing, the lower fraction was brought to a density of 1.110 g/ml by addition of solid NaCl and then centrifuged at 44.000 rpm for 22 h. The top fraction, which contained the lipoproteins with densities of 1.055–1.110 g/ml, was concentrated to a volume of about 4 ml by dialysis against a 30% solution of polyethyleneglycol (mol wt, 20.000) containing Na_2EDTA (1 mg/ml) and NaN_3 (1 mg/ml). Lp(a) was then separated from other lipoproteins by filtration on an agarose column (0.9 × 100 cm) using Bio Gel A-5m (Bio-Rad Laboratories, Richmond, Calif.). Elution of the lipoproteins was carried out at room temperature with 0.15 M NaCl containing Na_2EDTA (1 mg/ml) and NaN_3 (1 mg/ml), pH adjusted to 8.5 by addition of NH_4OH. The elution profile is shown in Fig. 1. The peaks were identified by immunodiffusion using monospecific antisera (16, 17, 19). The Lp(a) peak was further characterized by its electrophoretic mobility on agarose (single band between β- and pre-β-lipoproteins of normal serum), immunoelectrophoresis [precipitation line at identical position with anti-Lp(a) and anti-Lp B], and chemical composition as described recently (16, 17). To avoid contamination of Lp(a) with LDL, only the front portion of the Lp(a) peak was used for iodination and analytical procedures. The Lp(a) eluted from the column was concentrated to ~1 mg protein/ml by dialysis against polyethyleneglycol and then dialysed against glycine buffer, pH 10. The isolated and concentrated Lp(a) consisted of a clear solution. By electrophoresis in agarose gel, 95% of the material migrated as a narrow band in the position between β- and pre-β-lipoproteins of normal serum. Iodination with ^{125}I (The Radiochemical Centre, Amersham, England) was performed according to McFarlane (20) as modified by Bilheimer et al. (21). Free iodine was removed by filtration on Sephadex G-25 and subsequent dialysis against 0.15 M NaCl with several bath changes. Isolation and iodination of Lp(a) took 4 d. Labeled Lp(a) was injected on the morning of the 5th d after plasmapheresis.

TABLE I
Clinical Data of the Subjects

Subject	Sex	Age	Weight*	Height	Serum Lp(a)*	Serum cholesterol*	Serum triglycerides*	Diagnosis	Medication
		yr	*kg*	*cm*	*mg/100 ml*	*mg/100 ml*	*mg/100 ml*		
1	Male	21	63.2±0.1	174	<1	227±18	176±16	Bronchitis‡	—
2	Male	43	67.0±0.1	167	3±0.9	192±10	154±12	Normal	—
3	Male	57	77.7±0.1	179	44±4.9	225±13	130±15	Emphysema	—
4	Male	70	75.2±0.0	172	41±5.1	196±15	142±17	Normal	—
5	Male	66	62.2±0.1	166	71±6.2	212±14	112±10	Hypertension	Amiloride, hydrochlorothiazide
6	Male	52	74.5±0.2	178	35±2.6	231±21	123±19	Angina pectoris	Nitrate
7	Male	57	70.0±0.1	172	28±1.9	243±19	98±9	Hypertension	Amiloride, hydrochlorothiazide
8	Male	21	65.0±0.0	163	68±4.3	195±11	158±14	Normal	—
9	Male	59	75.5±0.1	176	56±3.8	205±16	102±16	Angina pectoris	Nitrate

* Mean±1 SD.
‡ Patient was recovered at the time of the study.

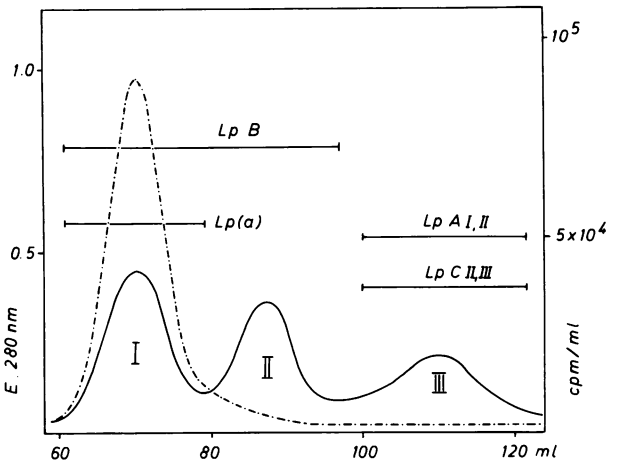

FIGURE 1 Elution profile of the lipoproteins of density 1.055–1.110 g/ml on Bio-Gel A-5m. The sample was taken 24 h after the intravenous injection of labeled Lp(a). ——, extinction at 280 nm; ·–·–·–·, radioactivity (cpm/ml). The immunological reactivities of the eluted fractions against antibodies to Lp(a), apolipoprotein A I and A II, B, C II, and C III are indicated. Peak I represents Lp(a), peak II lipoprotein B (part of the serum LDL), and peak III part of the HDL. Void volume 60 ml.

Characterization of the labeled Lp(a). Labeled Lp(a) showed the same electrophoretic mobility on agarose gel and identical immunoreactive behavior as unlabeled Lp(a). When labeled and unlabeled Lp(a) were mixed and applied to an agarose column (Bio-Gel A-5m), the labeled Lp(a) eluted in the same fraction as the unlabeled Lp(a). 0.5–3% of the radioactivity in Lp(a) was found in the lipid moiety after extraction with chloroform:methanol (2:1) (22). 14–17% of the radioactivity in Lp(a) was found in the "soluble" and 80–85% in the "insoluble" apolipoproteins (see below).

Study protocol. Labeled Lp(a) was sterilized by passage through a Millipore filter (0.45 nm; Millipore Corp., Bedford, Mass.) before the injection. After an overnight fast, 50–70 μCi labeled Lp(a) was injected intravenously. Autologous Lp(a) was injected in subjects 3–6 and 9. Subjects 1 and 2 received labeled Lp(a), which was obtained from subject 5, and subjects 7 and 8 received labeled Lp(a) from subject 9. Venous blood was drawn 10 min after the injection of the labeled Lp(a) and at various intervals during the 1st d and then every morning for up to 14 d. Two subjects were studied for 21 d. In these samples the radioactivity of the serum was measured. In subjects 3–6, 50 ml of blood was obtained 10 min, 6 h, and 24 h after the injection of labeled Lp(a), and then at different intervals throughout the study, as shown in Fig. 3. In these samples, Lp(a), very low density lipoproteins (VLDL) and LDL were isolated and the radioactivity in these lipoproteins and in the fraction with a density > 1.110 g/ml was estimated as follows: Serum was brought to a density of 1.055 g/ml by addition of solid NaCl and centrifuged for 22 h at 42,000 rpm. The top fraction, which contained VLDL and the bulk of LDL, was collected by tube slicing, dialyzed against saline with a density of 1.006 g/ml, and then centrifuged for 20 h at 42,000 rpm. The top fraction, was removed by slicing the tube. From the bottom fraction LDL was purified from contaminating Lp(a) by gel filtration on Bio-Gel A-5m as described above. The fractions containing VLDL, the purified LDL, the purified HDL, which eluted from the agarose column (Fig. 1), and the density > 1.110 g/ml fraction were checked for radioactivity. Lp(a) was isolated and purified by ultracentrifugation and gel filtration as described above. In the purified Lp(a), protein content and radioactivity were measured. In addition, part of the purified Lp(a) was dialyzed against distilled water, lyophilized, and the lipids removed with diethylether:ethanol (1:3, vol/vol). From the resultant apolipoproteins, the soluble apolipoproteins were obtained by extraction with 6 M urea. The insoluble apolipoproteins were washed several times with 6 M urea. Removal of soluble apolipoproteins was considered complete when no radioactivity could be detected in the urea solution. The remaining insoluble apolipoproteins were solubilized by addition of sodium dodecyl sulfate. The protein content and the radioactivity in both the soluble and insoluble apolipoproteins were measured to obtain the specific activity. The mean ratio of the specific activities in soluble and insoluble apolipoproteins was 2:3 and remained constant throughout the study.

In three subjects (Nos. 7–9) 24-h urine specimens were collected for calculation of the fractional catabolic rate of Lp(a) from the urinary radioactivity excretion. KI, $NaHSO_3$, and NaOH was added to the urine samples as a preservative (23).

Chemical and immunological analysis. Protein concentrations were measured according to Lowry et al. (24), total cholesterol by the Lieberman Burchard kit from Boehringer Mannheim GmbH, Mannheim, West Germany, and triglycerides according to Eggstein and Kreutz (25). Agarose electrophoresis was carried out in 1% agarose gel using 0.05 M barbital buffer, pH 8.2. Immunodiffusion was performed in 1% agarose using monospecific antisera (19). Quantitation of serum Lp(a) was carried out by Laurell's electroimmunoassay (rocket electrophoresis) (26) on glass plates (7 × 7 cm) in 1% agarose gels, 0.05 M barbital buffer, pH 8.2. The antibody was monospecific for the Lp(a) antigen and identical with the specimen described earlier (16, 17, 19); it gave no cross-reactivity with apo B or any other apolipoprotein of the A–F class. 0.05 or 0.1 ml of antiserum was used per 6 ml of agarose and per plate. Each plate was used for 14 analyses and contained three highly purified Lp(a) standards of different concentrations as well as 1–3 reference Lp(a)-positive sera, which had been standardized and kept at 4°C for <1 mo. The purified Lp(a) standard was prepared as described above and was homogenous according to the following criteria: single band in agarose gel electrophoresis, no reaction with antibodies other than anti-Lp(a) and anti-Lp B, chemical composition characteristic for Lp(a) (11, 16, 17). The concentration of the standard was measured gravimetrically by determination of the residual weight after equilibration dialysis (coefficient of variation was <1%). Electrophoresis was carried out at a field strength of 5 V/cm for 3 h at 15°C in 0.05 M barbital buffer, pH 8.2. The plates were rinsed after electrophoresis in 0.15 M saline for 2–4 h, dryed, and stained with Coomassie Blue R 250. The evaluation of the Lp(a) concentrations was performed by comparing the rocket area (height times width at half height) of the samples with that of highly purified standards. The relationship between the rocket area (mm^2) and protein concentration was linear from 5–25 mg Lp(a)/100 ml in the low antibody containing gels and from 20–60 mg Lp(a)/100 ml in the high antibody containing gels. Samples of higher concentrations were diluted accordingly. The electroimmunoassay worked with a day-to-day coefficient of variation of <5%.

Calculations. The fraction of the injected dose of radioactivity remaining in the plasma was plotted semilogarithmically against time (Fig. 2). The fractional catabolic rate (FCR = fraction of intravascular Lp(a) catabolized per day) was calculated according to the method described by Matthews (27): $FCR = 1/(C_1/b_1 + C_2/b_2)$, where C_1, C_2 = intercepts of

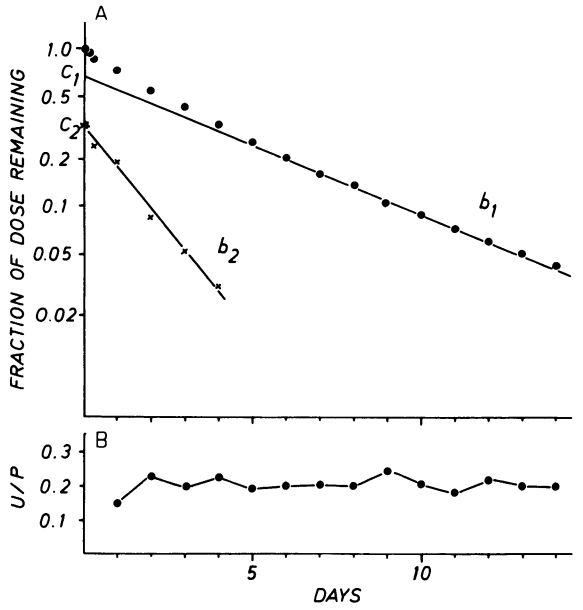

FIGURE 2 Serum die-away curve and urine/plasma (U/P) radioactivity ratios of subject 9 after intravenous injection of ^{125}I-Lp(a). (A) The fraction of injected radioactivity remaining in the serum is plotted semilogarithmically against time. The first exponential (b_1) was calculated from the terminal log-linear portion of the decay curve. The second exponential (b_2) was obtained by subtraction of the first exponential from the serum die-away curve. C_1 and C_2 are the intercepts of the first and second exponential with the vertical axis. (B) Daily U/P ratios are shown.

the first and second exponential with the vertical axis and b_1, b_2 = slopes of the first and second exponential (Fig. 2). The proportion of the total pool of Lp(a) that was in the intravascular space was calculated using the formula (27):

$$\text{percent intravascular} = \frac{\left(\frac{C_1}{b_1} + \frac{C_2}{b_2}\right)^2}{\frac{C_1}{(b_1)^2} + \frac{C_2}{(b_2)^2}}.$$

Under steady-state conditions, the rate of synthesis of Lp(a) is equal to its absolute rate of catabolism. The absolute rate of synthesis or catabolism (milligram per day) of Lp(a) was calculated as the product of FCR times plasma volume times Lp(a) concentration. The plasma volume was estimated using Edelman's formula (28). FCR was also calculated as the ratio of the daily urinary excretion of radioactivity to the total amount of radioactivity in the plasma (29). Statistical calculations were performed using standard methods (30).

RESULTS

Throughout the study, >96% of the total radioactivity in the serum remained with Lp(a). Less than 0.05% of the serum radioactivity was found in VLDL. In LDL, HDL, and in the salt volume eluting from the column, only traces of radioactivity were detectable after purification on Bio-Gel A-5m. In the unpurified density fraction > 1.110 g/ml, a total amount of <3% of the serum radioactivity was found. Presumably, this was due to contamination with labeled Lp(a), in that the radioactivity in purified HDL was negligible. Under the assumption that the labeled apolipoprotein has not been altered by iodination, these findings seem to indicate that no conversion of serum Lp(a) to other serum lipoproteins or exchange of apolipoproteins between Lp(a) and other lipoproteins occurred.

In four subjects the specific activities of the isolated intact Lp(a) molecule and of the soluble and the insoluble apolipoproteins were determined at different times of the study as shown in Fig. 3. These values are expressed as fraction of the specific activities at 10 min after the injection of labeled Lp(a). The values obtained for the whole Lp(a) molecule, the soluble and the insoluble apolipoproteins were compared with the corresponding values of the serum radioactivity decay curve. The mean difference between the values of the intact Lp(a) and the corresponding values of the serum radioactivity decay curve was + 0.3±4.8% (mean±1 SD), between the soluble apolipoproteins and the serum radioactivity was +0.5±7.5%, and between the insoluble apolipoproteins and the serum radioactivity was −3.0±6.5%. This shows that the decay of the specific activity of the whole Lp(a) molecule and of the individual apolipoproteins was practically identical with the decay of the serum radioactivity. The percentage of radioactivity in the lipid moiety of the Lp(a) molecule showed some decline during the first days of the experiment, but was too low to influence the calculation of the kinetic parameters at any time of the study.

Therefore, it was permissible to calculate the kinetic parameters of the Lp(a) turnover from the serum radioactivity-time curves. The concentration of Lp(a) in the serum was determined by immunoelectrophoresis repeatedly from the beginning until the end of the study and showed little variation as indicated in Table I. In subjects 3–6, the serum level of Lp(a) apolipoprotein could also be calculated from the radioactivity of the serum and the specific activity of the isolated Lp(a), since >96% of the radioactivity in the serum was associated with the protein moiety of Lp(a). Also from this calculation, a constant Lp(a) concentration was found in each case. The constancy of the Lp(a) concentration during the study period was considered to reflect a steady state in Lp(a) turnover.

Fig. 2 shows a representative serum radioactivity time-curve. In all cases, the curves showed an initial rapid decline merging with a log-linear portion about 4 d after the injection of the labeled Lp(a). Since the radioactivity decay curves could always be resolved into two exponentials by curve peeling (Fig. 2), the kinetic parameters were calculated in terms of a two-

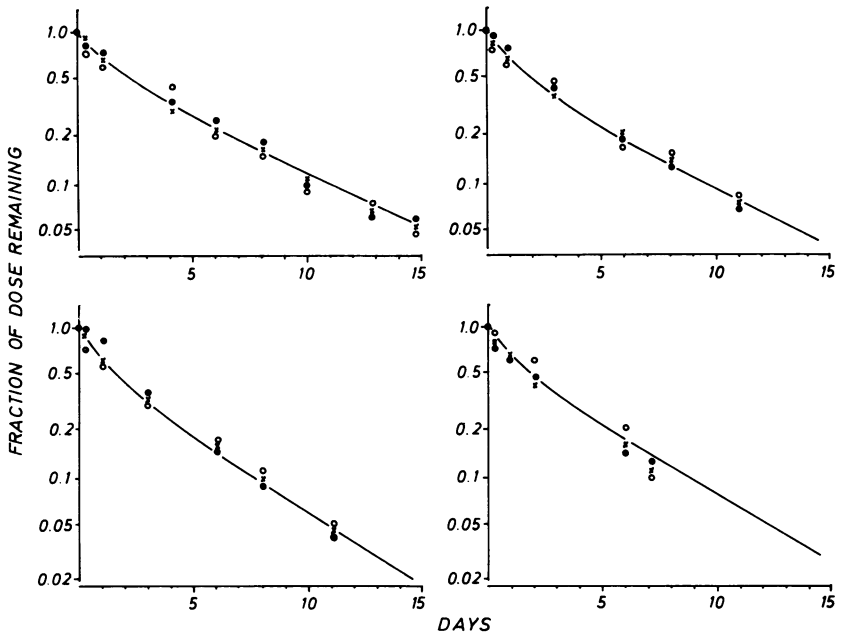

FIGURE 3 Die-away curve of the serum radioactivity (——) and decay of the specific activities of the isolated intact Lp(a) (×), the soluble (○) and insoluble (●) apolipoproteins of Lp(a) after intravenous injection of ^{125}I-Lp(a) in subjects 5, 4, 6, and 3. The decay of the whole Lp(a) molecule and of its individual apolipoproteins closely follows the die-away curve of the serum radioactivity.

compartment model (27). The regression line of the first exponential was calculated from the values obtained from the 5th to last day of the study. In two cases the radioactivity time-curve was followed for 21 d and the decay curves could also be resolved into two exponentials. The kinetic parameters were identical when calculated from the values of the first 14 or 21 d.

The kinetic parameters of Lp(a) are presented in Table II. In subjects 7–9, the FCR was also calculated from the urine/plasma radioactivity ratio. This method can be applied for the estimation of

TABLE II
Kinetic Parameters of Lp(a) Turnover

Subject No.	Serum Lp(a)	Half-life of* second exponential	Half-life of* first exponential	FCR	Distribution of Lp(a)	Rate of synthesis§
	mg/100 ml	*d*	*d*	*Fraction of i.v. pool/d*	*% intravascular*	*mg/kg/d*
1	<1	0.72	3.05	0.343	70.7	0.16
2	3	0.63	2.64	0.392	73.8	0.51
3	44	0.57	3.04	0.308	77.9	5.89
4	41	1.11	3.71	0.258	82.6	5.03
5	71	0.44	3.91	0.236	78.8	7.97
6	35	0.71	2.90	0.336	76.9	5.11
7	28	0.96	4.20	0.272 (0.248±0.017‡)	67.9	3.31
8	68	1.10	3.02	0.350 (0.335±0.020‡)	73.8	10.75
9	56	1.13	3.45	0.257 (0.228±0.043‡)	86.0	6.26
Mean		0.82	3.32	0.306	76.5	5.00
SD		0.26	0.52	0.054	5.1	3.37

* Half-life of the first (b_1) and second (b_2) exponential of the serum radioactivity time-curve.
‡ Values calculated from urine/plasma radioactivity ratio, mean±1 SD of the values from day 2 to 14.
§ Units for Lp(a) synthesis refer to the entire Lp(a) molecule.

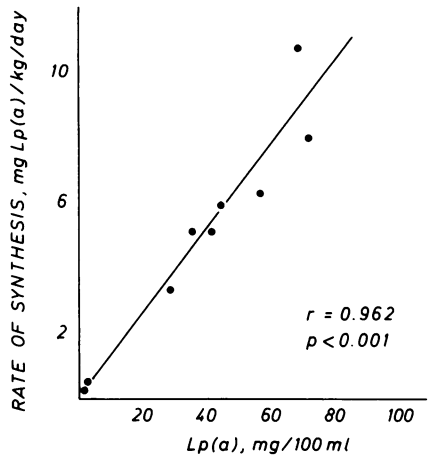

FIGURE 4 Relationship between Lp(a) concentration in the serum and rate of synthesis of Lp(a).

Lp(a) turnover, because practically all the radioactivity of the serum was contained in Lp(a) over the entire study period. The values obtained by this method were in close agreement with the values for FCR obtained from the die-away curve of the serum radioactivity (Table II).

A positive correlation was found between the concentration of Lp(a) in the serum and the absolute rate of synthesis of serum Lp(a) apoproteins (Fig. 4). There appeared to be a tendency toward an inverse relationship between the serum level of Lp(a) and FCR, but this correlation could not be proven statistically (Fig. 5). No relationship existed between the serum concentration and half-lives of Lp(a) in the serum.

The mean FCR of Lp(a) was somewhat lower in the five subjects who received autologous Lp(a) as compared with the four subjects who received nonautologous Lp(a). This difference, however, was not statistically significant.

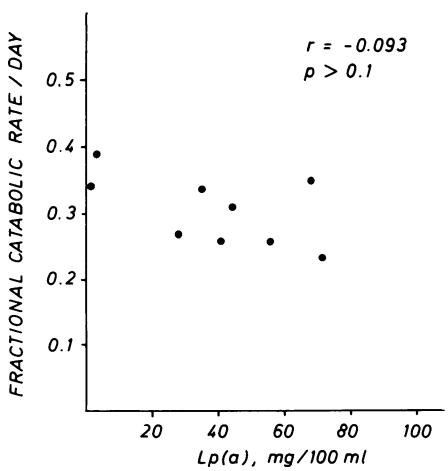

FIGURE 5 Relationship between Lp(a) concentration in the serum and FCR of Lp(a).

DISCUSSION

After the intravenous injection of labeled Lp(a) only insignificant amounts of radioactivity were detectable in lipoproteins other than Lp(a) during the study period. The specific activity time-curves for the isolated whole Lp(a) molecule, the soluble and the insoluble Lp(a) apolipoproteins, and the radioactivity time-curve of the serum were identical. Apparently, Lp(a) is not converted to another lipoprotein and no exchange of apolipoproteins between Lp(a) and other lipoproteins takes place. Lp(a) appears to leave the plasma pool as an intact particle as suggested in a recent study (16). Other experiments, in which labeled VLDL has been injected to Lp(a)-positive individuals, have shown that Lp(a) is not derived from the catabolism of other apolipoprotein B-containing lipoproteins (17). Therefore, it is suggested that Lp(a) is synthesized and metabolized independently of other plasma lipoproteins. This conclusion could explain the findings of others that the serum level of Lp(a) is not influenced by various dietary manipulations that are known to cause considerable changes in the plasma concentrations of other lipoproteins (13).

The half-life of Lp(a) has already been estimated using reductive alkylation as method of labeling (16). Due to the low radioactivity of Lp(a) in this former study, the radioactivity time-curve of Lp(a) could be followed for only 4 d. Therefore, only the initial rapid decline of the whole die-away curve could be observed and hence a monoexponential plasma decay of Lp(a) was assumed (16). This explains that the half-lives obtained in this earlier study are shorter than those calculated in terms of a two-pool model in this study. During the first 4 d after the injection of labeled Lp(a), the decay of radiomethylated and radioiodinated Lp(a) is similar.

The kinetic parameters of Lp(a) obtained in this study are very similar to those of LDL in normal subjects as found by others. The mean FCR of Lp(a) is ~20–30% lower and the average percentage of the total Lp(a) pool that was in the circulation is 15–20% higher than the corresponding values found for LDL by Langer et al. (31), Simons et al. (32), and Sigurdsson et al. (33). The mean values for FCR and for the distribution of Lp(a) between intravascular and extravascular space were practically identical with those found for LDL by Packard et al. (34). The biological half-life of Lp(a) was slightly longer than that of LDL as reported by Langer et al. (31) and Simons et al. (32), and slightly shorter than that of LDL found by Sigurdsson et al. (33) and Packard et al. (34). Apparently, there

are not only immunological and chemical similarities between LDL and Lp(a). From the comparison of the kinetic parameters of these two lipoproteins one may suggest that these two lipoproteins also appear to be acted upon by similar catabolic mechanisms. To prove this assumption kinetic studies of LDL and Lp(a) in patients with familiar hypercholesterolemia would be of interest.

It has been reported by others that the concentrations of Lp(a) vary in a wide range among the individuals of a population (13, 18, 35). The Lp(a) level in a single individual, however, remains remarkably constant (13, 18). Principally, the concentration of Lp(a) in the serum is dependent on the rate of synthesis and the FCR of the lipoprotein. The individuals in this study also showed very different serum Lp(a) levels. Their FCR, however, varied to a much lesser degree and there was no relationship between the concentration and FCR of Lp(a). On the other hand, a highly significant correlation was found between the serum concentration and the absolute synthetic rate of Lp(a). Therefore, it is concluded that the mechanism that is responsible for an elevated concentration of Lp(a) in the serum of these subjects is not a defective catabolism but an increased synthesis of Lp(a). It should be stated that the studies of this paper were performed in males. Until now no information is available on the Lp(a) turnover in females.

Studies with ^{125}I-labeled LDL in patients with an elevated LDL concentration indicated a defect of LDL catabolism in these subjects. In homozygote and heterozygote patients with familial hyperbetalipoproteinemia, a reduced FCR of LDL was found (31–34). A marked increase in the synthesis of LDL in the homozygotes has also been reported (32). In this study, the Lp(a) concentration was not dependent on the FCR of Lp(a). However, the possibility that a defective catabolism, as found in familial hyperbetalipoproteinemia, is responsible for an increased Lp(a) level cannot be excluded. Until now, no information is available about the site and regulation of Lp(a) synthesis or catabolism.

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REFERENCES

1. Berg, K. 1963. A new serum type system in man—the Lp system. *Acta Pathol. Microbiol. Scand.* **59:** 369–382.
2. Simons, K., C. Ehnholm, O. Renkonen, and B. Bloth. 1970. Characterization of the Lp(a) lipoprotein in human plasma. *Acta Pathol. Microbiol. Scand. Sect. B. Microbiol. Immunol.* **78:** 459–466.
3. Vogelberg, K. H., G. Uterman, and F. A. Gries. 1973. Zur Differenzierung des Lp(a) Lipoproteins mit Hilfe der Agargel-Elektrophorese. *Z. Klin. Chem. Klin. Biochem.* **11:** 291–296.
4. Jürgens, G., and G. M. Kostner. 1975. Studies on the structure of the Lp(a) lipoprotein. Isolation and partial characterization of the Lp(a) specific antigen. *Immunogenetics.* **1:** 560–574.
5. Ehnholm, C., H. Garoff, O. Renkonen, and K. Simons. 1972. Protein and carbohydrate composition of Lp(a) lipoproteins from human plasma. *Biochemistry.* **11:** 3229–3232.
6. Dahlen, G., K. Berg, U. Ramberg, and A. Tamm. 1974. Lp(a) lipoprotein and pre-β_1-lipoprotein in young adults. *Acta Med. Scand.* **196:** 327–331.
7. Berg, K., G. Dahlen, and M. H. Frick. 1974. Lp(a) lipoprotein and pre-β_1-lipoproteins in patients with coronary heart disease. *Clin. Genet.* **6:** 230–235.
8. Albers, J. J., V. G. Cabana, G. R. Warnik, and W. R. Hazzard. 1975. Lp(a) lipoprotein: Relationship to sinking pre-β-lipoprotein, hyperlipoproteinemia and apolipoprotein B. *Metab. Clin. Exp.* **24:** 1047–1054.
9. Albers, J. J., C. H. Chen, and F. Aladjem. 1972. Human serum lipoproteins. Evidence for three classes of lipoproteins in S_f 0-2. *Biochemistry.* **11:** 57–63.
10. Uterman, G., K. Lipp, and H. Wiegandt. 1972. Studies on the Lp(a) lipoprotein of human serum. IV. The disaggregation of the Lp(a) lipoprotein. *Humangenetik.* **14:** 142–150.
11. Kostner, G. M. 1976. Lp(a) lipoproteins and the genetic polymorphisms of lipoprotein B. *In* Low Density Lipoproteins. C. E. Day and R. S. Levy, editors. Plenum Publishing Corp., New York. 229–269.
12. Albers, J. J., and W. R. Hazzard. 1974. Immunochemical quantification of human plasma Lp(a) lipoprotein. *Lipids.* **9:** 15–26.
13. Albers, J. J., J. L. Adolphson, and W. R. Hazzard. 1977. Radioimmunoassay of human plasma Lp(a) lipoprotein. *J. Lipid Res.* **18:** 331–338.
14. Dahlen, G., C. Ericson, C. Furberg, L. Lundkvist, and K. Svärdsudd. 1972. Studies on extra pre-β lipoprotein fraction. *Acta Med. Scand. Suppl.* **531:** 1–29.
15. Walton, K. W. 1974. Identification of lipoproteins involved in human atherosclerosis. *In* Atherosclerosis III. G. Schettler and A. Weizel, editors. Springer-Verlag, Berlin. 93–95.
16. Krempler, F., G. Kostner, K. Bolzano, and F. Sandhofer. 1978. Studies on the metabolism of the lipoprotein Lp(a) in man. *Atherosclerosis.* **30:** 57–65.
17. Krempler, F., G. Kostner, K. Bolzano, and F. Sandhofer. 1979. Lipoprotein (a) is not a metabolic product of other lipoproteins containing apolipoprotein B. *Biochim. Biophys. Acta.* **575:** 63–70.
18. Ehnholm, C., H. Garoff, K. Simons, and H. Aro. 1971. Purification and quantitation of the human plasma lipoprotein carrying the Lp(a) antigen. *Biochim. Biophys. Acta.* **236:** 431–439.
19. Kostner, G. M., J. R. Patsch, S. Sailer, H. Braunsteiner, and A. Holasek. 1974. Polypeptide distribution of the main lipoprotein density classes separated from human plasma by rate zonal ultracentrifugation. *Eur. J. Biochem.* **45:** 611–621.
20. McFarlane, A. S. 1958. Efficient trace-labelling of proteins with iodine. *Nature (Lond.).* **182:** 53.
21. Bilheimer, D. W., S. Eisenberg, and R. I. Levy. 1972. The metabolism of very low density lipoprotein proteins. I. Preliminary in vitro and in vivo observations. *Biochim. Biophys. Acta.* **260:** 212–221.

22. Folch, J. M., M. Lees, and G. H. Sloane-Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.* **226:** 497–509.
23. Steinfeld, J. L., R. R. Paton, A. L. Flick, R. A. Milch, and F. E. Beach. 1957. Distribution and degradation of human serum albumin labeled with I^{131} by different techniques. *Ann. N. Y. Acad. Sci.* **70:** 109–121.
24. Lowry, O. H., N. J. Rosebrough, A. L. Farr, and R. J. Randall. 1951. Protein measurement with the Folin phenol reagent. *J. Biol. Chem.* **193:** 265–275.
25. Eggstein, M., and F. H. Kreutz. 1966. Eine neue Bestimmung der Neutralfette im Blutserum und Gewebe. I. Mitteilung. Prinzip, Durchführung und Besprechung der Methode. *Klin. Wochenschr.* **44:** 262–265.
26. Laurell, C. B. 1972. Electroimmunoassay. *Scand. J. Clin. Lab. Invest. Suppl.* **124:** 21–37.
27. Matthews, C. M. E. 1957. The theory of tracer experiments with ^{131}I-labelled plasma proteins. *Phys. Med. Biol.* **2:** 36–53.
28. Edelman, I. S., and J. Leibman. 1959. Anatomy of body water and electrolytes. *Am. J. Med.* **27:** 256–277.
29. Berson, S. A., and R. S. Yalow. 1957. Distribution and metabolism of I^{131} labeled proteins in man. *Fed. Proc.* **16:** 13S–18S.
30. Snedecor, G. W., and W. C. Cochran. 1967. Statistical Methods. The Iowa State University Press, Ames, Iowa. Sixth edition.
31. Langer, T., W. Strober, and R. I. Levy. 1972. The metabolism of low density lipoproteins in familial type II hyperlipoproteinemia. *J. Clin. Invest.* **51:** 1528–1536.
32. Simons, L. A., D. Reichl, N. B. Myant, and M. Mancini. 1975. The metabolism of the apoprotein of plasma low density lipoprotein in familial hyperbetalipoproteinemia in the homozygous form. *Atherosclerosis.* **21:** 283–298.
33. Sigurdsson, G., A. Nicoll, and B. Lewis. 1976. The metabolism of low density lipoprotein in endogenous hypertriglyceridemia. *Eur. J. Clin. Invest.* **6:** 151–158.
34. Packard, C. J., J. L. H. C. Third, J. Shepherd, A. R. Lorimer, H. G. Morgan, and T. D. V. Lawrie. 1976. Low density lipoprotein metabolism in a family of familial hypercholesterolemic patients. *Metab. Clin. Exp.* **25:** 995–1006.
35. Schultz, J. S., D. C. Schreffler, C. F. Singer, and N. R. Harvie. 1974. The genetics of the Lp antigen. Its quantitation and distribution in a sample population. *Ann. Hum. Genet.* **38:** 39–46.