Metabolism of the Serum Amyloid A Proteins (SAA) in High-Density Lipoproteins and Chylomicrons of Nonhuman Primates (Vervet Monkey)

JOHN S. PARKS, PhD, and
LAWRENCE L. RUDEL, PhD

Arteriosclerosis Research Center, Department of Comparative Medicine, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, North Carolina

Serum amyloid protein (SAA) has been reported to be an apoprotein of high-density lipoprotein (HDL), but little is known concerning its metabolism. In this study, apoSAA was induced in nonhuman primate plasma HDL and thoracic duct lymph chylomicra by overnight chair restraint of the animals. There was a 3–6-fold increase in plasma HDL apoSAA in chair-restrained animals when compared with caged (control) animals. Lymph chylomicrons of chaired animals also contained significant amounts (~20% of total protein) of apoSAA. For study of the metabolism of HDL apoSAA, animals were given injections of ^{131}I-labeled lymph chylomicrons and autologous ^{125}I-HDL. HDL were isolated from the plasma of recipient animals between 1 minute and 5 days after injection, and the specific activity of the apoSAA was determined. The turnover of apoSAA from plasma was biphasic, the initial phase having a t½ (0.39–0.48 days) similar regardless of the source (chylomicrons versus HDL) of the injected dose. The second phase of the turnover was significantly faster (t½ = 2.5 days) for apoSAA of ^{125}I-HDL origin than that of ^{131}I-chylomicron origin (t½ = 4.3 days). This difference also was suggested by the fractional catabolic rates (FCR) of ^{125}I-HDL and ^{131}I-chylomicron apoSAA (1.02 versus 0.74 d^{-1}, respectively). From these studies it was concluded that 1) apoSAA can be rapidly induced in plasma HDL and lymph chylomicrons of nonhuman primates by chair restraint; 2) HDL apoSAA is catabolized more rapidly than HDL apoA-I and apoA-II; 3) and the catabolic rate of HDL apoSAA may be determined, in part, by its lipoprotein origin (chylomicrons versus HDL). (Am J Pathol 1983, 112:243–249)

HIGH-DENSITY LIPOPROTEINS (HDL) are the smallest of the plasma lipoproteins and are approximately 50% protein by mass.[1] The protein component of HDL is heterogeneous with two major apoproteins, apoA-I and apoA-II, and a number of minor apoproteins.[2] Several years ago two new apoproteins, called the threonine-poor apoproteins because of their low content of the amino acid threonine, were described in human and nonhuman primate HDL.[3,4] The threonine-poor apoproteins have a molecular weight similar to the C apoproteins (~14,000) but a pI that is more basic than other HDL apoproteins.[3,4] These proteins contain no detectable sialic acid and have no known cofactor function.[4]

Recently, Eriksen and Benditt[5] isolated and characterized serum amyloid proteins (SAA) from human HDL and found them to correspond to the previously reported threonine-poor apoproteins.[3] This assignment was based on amino acid analysis and partial sequence, isoelectric point, and molecular weight of the SAA apoproteins. The SAA proteins have been reported to behave as plasma acute-phase reactants and can be induced by various stimuli.[6–9]

Little is known about the relative contribution of different tissues to the whole-body synthesis of apoSAA or to its catabolic fate. SAA is the presumed precursor of amyloid protein, which can deposit in tissues in response to acute and chronic inflammatory

Supported by the National Heart, Lung and Blood Institute (Grants HL-14164 and HL-24736).

This work was carried out, in part, during completion of the requirement for a PhD degree (Dr. Parks) in Comparative and Experimental Pathology from the Bowman Gray School of Medicine of Wake Forest University. Dr. Parks was an R. J. Reynolds Industries Special Research Fellow during completion of this work.

Accepted for publication March 31, 1983.

Address reprint requests to Dr. John S. Parks, Arteriosclerosis Research Center, Department of Comparative Medicine, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem, NC 27103.

0002-9440/83/0907-0243$01.15

events.[6,10] The relationship between apoSAA catabolism and the deposition of amyloid is unknown. One study has shown that *in vitro* proteolytic digestion of apoSAA by monocytes can lead to amyloidlike intermediates.[11] Several potential sites of synthesis of apoSAA have been identified,[8,9,12] and hepatic apoSAA synthesis has been shown to be stimulated by a factor released from macrophages that had been previously exposed to an inflammatory agent.[13,14] One study has shown a rapid disappearance of apoSAA-rich HDL from mouse plasma.[15]

Because HDL is purported to be a protective factor for coronary heart disease,[16] interest in the metabolism of HDL has increased recently. A better understanding of the pathogenesis of atherosclerosis and secondary amyloidosis may be obtained with a more detailed knowledge of the metabolic fate of HDL apoSAA. This study was undertaken to characterize the *in vivo* metabolism of HDL apoSAA in the nonhuman primate. This animal model was chosen because of its close phylogenetic relationship to man and its key role as a model for atherosclerosis research. In this animal model apoSAA has been found to associate with thoracic lymph duct chylomicrons as well as HDL. The results suggest that HDL apoSAA is metabolized much more rapidly than HDL apoA-I and apoA-II and that apoSAA from two different sources, chylomicrons and HDL, may have different catabolic rates in the plasma.

Methods

Induction of apoSAA

The experimental animals used for this study were adult male African green monkeys of the vervet subspecies (*Cercopithecus aethiops*). The animals' diets were described previously.[17] Plasma HDL SAA concentrations were induced by placing the animals in restraining chairs (Plas Laboratories, Lansing, Mich) for a 16-hour or longer period. All the animals were previously chair-trained for a total of at least 15 nonconsecutive days. It was found that after this amount of chair training the urinary 17-hydroxy steroids returned to normal levels, suggesting that restraint-induced stress had decreased.[17a] Caged animals had low amounts of HDL apoSAA (see below) and served as control animals.

Dose Preparation

For metabolic studies, autologous HDL was isolated from the plasma of 4 chair-restrained animals.[18] Chylomicrons (Sf 400–2000) were isolated from the lymph of thoracic-duct-cannulated animals who were chair-restrained during the collection of lymph.[18] Chylomicrons and HDL were iodinated with ^{131}I and ^{125}I, respectively.[18]

Metabolic Studies

Doses of ^{131}I-chylomicrons and autologous ^{125}I-HDL were mixed at 4° C and immediately injected into the saphenous vein of chair-restrained recipient animals. Blood samples were withdrawn at various times (1, 5, 10, 20, and 40 minutes; 1, 3, 5, 8, and 12 hours; and 1–6 days) via a femoral artery cannula.[18] HDL were isolated from plasma by heparin-manganese precipitation and ultracentrifugation.[18] HDL were delipidated and analyzed by isoelectric focusing (IEF) for apoSAA as previously described.[18] Stained gel slices corresponding to these apoproteins were counted for ^{131}I and ^{125}I. The relative amount of SAA was found by scanning Coomassie-blue-stained IEF gels at 550 nm. A linear response of area units to the amount of purified $apoSAA_1$, or $apoSAA_2$[4] applied to the IEF gels was found by this method. Specific activity was calculated as cpm/area units for $apoSAA_1$, and $apoSAA_2$.

Kinetic Analyses and Statistics

The specific activity of individual apoproteins of interest was normalized to the 1-minute specific activity, since 97% ± 9% (mean ± SEM) of the injected HDL count was in the plasma 1–5 minutes after injection. Plots of the percentage of 1-minute specific activity versus time were analyzed by "curve peeling" (see Figure 2 inset). In this method the log-linear part of the terminal turnover curve was subjected to log-linear regression analysis, and the best fit line was extrapolated back to time zero; the slope of this line is commonly referred to as β, and the y-intercept is called B. Ln $2/\beta$ is equal to the half-life ($t_{½}\beta$) of the slowly decaying label. Data points at early times were then subtracted from the extrapolated line and plotted as a function of time. The slope (α) and y-intercept (A) of the newly created line was determined by log-linear regression analysis. Ln $2/\alpha$ is equal to the half-life ($t_{½}\alpha$) of the rapidly decaying label. Fractional catabolic rates (FCRs) were calculated as $100(A/\alpha + B/\beta)^{-1}$.[19]

Tests for statistical significance were made between different sources of apoproteins (^{131}I versus ^{125}I) or between different apoproteins ($apoSAA_1$ versus $apoSAA_2$) with the use of a paired t test.[20]

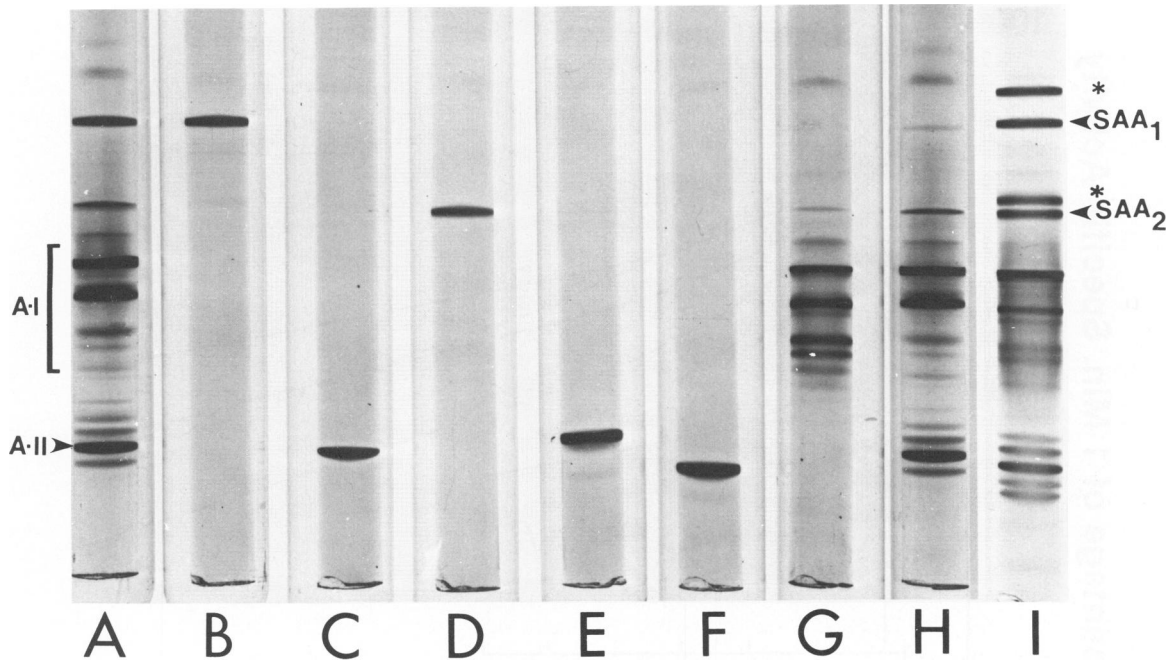

Figure 1—Isoelectric focusing gels of apoHDL and purified HDL apoproteins (pH range, 4-7). **A**—Induced HDL (from a chair-restrained animal). **B**—ApoSAA$_1$. **C**—ApoA-II. **D**—ApoSAA$_2$. **E**—ApoCII. **F**—ApoCIII. **G**—ApoA-I. **H**—Control HDL (from caged animal). **I**—Induced HDL (from a chair-restrained animal). Some animals exhibited additional bands (marked by the asterisk in gel **I**) in the basic region of the IEF gels with chair restraint. These bands were presumed to be isoforms of apoSAA$_1$ and SAA$_2$.

Results

The isolation and characterization of HDL apoSAA apoproteins from African green monkeys were reported previously.[4] In that report the apoSAA proteins were identified as two threonine-poor proteins, DI-1, and DII-1. This nomenclature identified the proteins as eluting from a DEAE-cellulose column in Peaks I and II, respectively. The arabic number indicated the respective proteins as being the slowest migrating protein on urea polyacrylamide gel electrophoresis. To simplify existing nomenclature, we will use the names originally proposed by Erikson and Benditt.[5] Thus, DI-1, and DII-1 will be referred to as apoSAA$_1$, and apoSAA$_2$, respectively. The two apoproteins will be collectively referred to as apoSAA.

Table 1—Percentage Apoprotein Composition of HDL and Chylomicrons*

	AI	AII	SAA$_1$	SAA$_2$
HDL (n = 6)	74 ± 1†	12 ± 1	4 ± 1	4 ± 1
"Induced" HDL‡ (n = 5)	56 ± 1	6 ± 0.4	25 ± 3	12 ± 2
Chylomicrons (n = 2) (S$_f$ 400-2000)	20-23§	11-14	9-10	9-12

* Percentage composition was determined by densitometric scanning of isoelectric focusing gels as described in Materials and Methods. Other peptides (apoCs) are not included.

† Mean ± SEM. ApoA-I and A-II percentages were significantly reduced, and SAA percentages were significantly increased for HDL versus "induced" HDL.

‡ HDL samples in which SAA has been "induced" by chair restraint of the monkeys (see Materials and Methods).

§ Values for chylomicrons are the range for 2 animals.

In our previous study[4] it was found that the amount of apoSAA proteins in the HDL fraction was highly variable (the apoSAA$_1$/apoA-I ratio ranged from 0.01-0.81). The variability in the amount of the apoSAA proteins has subsequently been found to correlate with the chair-restraining of the animals. Figure 1 shows the IEF gels of apoHDL and purified HDL apoprotein samples. The amount of individual proteins in apoHDL was estimated by scanning IEF gels (Table 1). This method is only semiquantitative, because a given amount of the various individual apoproteins may bind different amounts of Coomassie blue. However, a comparison of staining intensity between caged and chaired animals can be made.

Caged animals contained 4% of their total HDL apoprotein as apoSAA$_1$ or apoSAA$_2$ (Figure 1H and Table 1). Three of the six caged animals had no detectable HDL apoSAA. However, "induced" HDL isolated from chair-restrained animals had an average of 25% of the total HDL protein as apoSAA$_1$ and 12% as apoSAA$_2$ (Figure 1A and Table 1). This represented a 3-6-fold increase in the percentage of HDL apoSAA. In a series of control experiments it

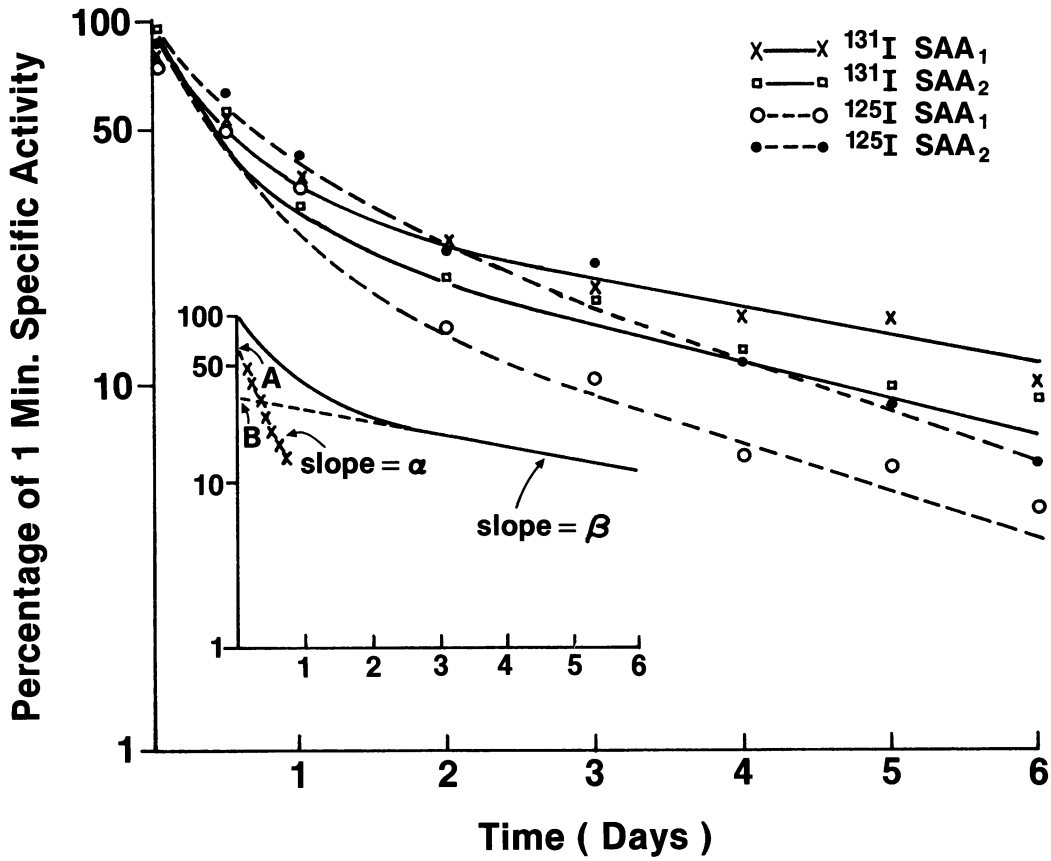

Figure 2 — Decay of HDL apoSAA after the simultaneous injection of ^{131}I-chylomicrons and autologous ^{125}I-HDL into recipient animals. Details are given in the Materials and Methods section. The time points represent the mean experimental data from 4 animals. The curves represent the predicted turnover based on the slopes and intercepts derived from the experimental data via curve peeling. The equation for a biexponential turnover ($y = Ae^{-\alpha x} + Be^{-\beta x}$) was used to predict the curves. The insert shows an example of the curve peeling process for the predicted turnover of HDL ^{131}I-SAA.

was found that the dramatic HDL apoSAA induction was associated with chair restraint of the animals and not other environmental factors such as ketamine immobilization, type of diet (fat saturation or cholesterol concentrations), or time after a high-fat meal. The induction of apoSAA was rapid; the change from 4% to 25% of the total HDL protein occurred in less than 16 hours. The amount of apoSAA in HDL remained elevated for the duration of chair restraint (up to 6 days).

Polymorphism of apoSAA amino acid sequence has been reported in the mouse[21] and in man.[22] Certain individual animals in this study exhibited apparent microheterogeneity of the apoSAA proteins as additional bands appeared on IEF gels with chair restraint. Figure 1 (Gel I) shows a representative gel of apoHDL from one such animal. Instead of the usual two bands representing apoSAA$_1$ and apoSAA$_2$ there were four bands; two of the bands corresponded to the usual pI of the apoSAA proteins; however, the two additional bands had a slightly more basic pI. The doublet banding pattern was reproducible over time for those animals that showed it.

Concomitant with the induction of apoSAA by chair restraint of the animals was an apparent decrease in the percentage composition of the major apoproteins of HDL, apoA-I and apoA-II (Table 1). In a larger subset of animals (6 control and 8 chaired) a similar decrease in the percentage of HDL protein as apoA-I (43%) and apoA-II (35%) was found by immunoassay. Per-particle compositions would be necessary for determination of whether apoA-I and apoA-II are being displaced from the HDL particle by apoSAA or are becoming a proportionally smaller amount of the particle protein mass as a result of the apoSAA addition.

Thoracic lymph duct chylomicrons were investigated by IEF also. During the collection of thoracic duct lymph, animals were chair-restrained. Percentage composition values for apoA-I, A-II, SAA$_1$ and SAA$_2$ are shown in Table 1. Because of the necessity to chair-restrain animals during collection of lymph,

Table 2—Half-Life and Fractional Catabolic Rate of HDL apoSAA$_1$ and apoSAA$_2$ Die-Away*

Origin of label	HDL apoSAA$_1$ $t_{1/2}\alpha$ (days)	HDL apoSAA$_1$ $t_{1/2}\beta$ (days)	HDL apoSAA$_1$ FCR (days^{-1})	HDL apoSAA$_2$ $t_{1/2}\alpha$ (days)	HDL apoSAA$_2$ $t_{1/2}\beta$ (days)	HDL apoSAA$_2$ FCR (days^{-1})
Chylomicrons (^{131}I)	0.39 ±0.06	4.3† ±0.3	0.74 ±0.23	0.31 ±0.06	3.5 ±0.9	0.72 ±0.18
HDL (^{125}I)	0.39 ±0.08	2.5† ±0.3	1.02 ±0.36	0.48 ±0.07	2.5 ±0.5	0.68 ±0.15

* ^{131}I-chylomicrons and ^{125}I-HDL were injected simultaneously into recipient animals, and HDL were isolated from plasma at various times. The specific activity was determined (see Materials and Methods) for each protein and plotted versus time on semilog paper. The half-lives of the first ($t_{1/2}\alpha$) and second ($t_{1/2}\beta$) exponentials were determined by "curve peeling" (see Materials and Methods). Values are the mean ± SE of 4 animals.

† ^{125}I-SAA$_1$ $t_{1/2}\beta$ is significantly different ($P < 0.01$) from ^{131}I-SAA$_1$ $t_{1/2}\beta$ as determined by a paired t test. No other significant differences in half-lives of the tracers or FCR were found with SAA$_1$ versus SAA$_2$ or ^{131}I versus ^{125}I comparisons.

comparable samples from caged animals were not collected. These data illustrate that the apoSAA proteins were present on lymph chylomicrons. In fact, lymph chylomicrons contained nearly equal molar amounts of apoSAA$_1$, SAA$_2$, A-I, and A-II if dye uptake was approximately equal for all proteins.

To study the metabolism of the SAA apoproteins, doses of ^{131}I-chylomicrons and ^{125}I-HDL were injected, and the specific activity of HDL apoSAA$_1$ and apoSAA$_2$ was followed with time. The resulting semilog plots, normalized to the percentage of 1 minute of specific activity, are shown in Figure 2. The ^{131}I-specific activity of apoSAA$_1$ and apoSAA$_2$ in the HDL fraction peaked within 20 minutes after injection (time points not shown) and decayed rapidly thereafter. Thus, the apoSAA label that was injected on ^{131}I-lymph chylomicrons was rapidly exchanged and/or transferred to plasma HDL. The HDL SAA proteins, regardless of the source (chylomicrons or HDL), had essentially identical kinetic behavior over the first 24 hours after injection. The initial turnover of the HDL SAA proteins was rapid, with a half-life ranging from 0.31 to 0.48 days ($t_{1/2}\alpha$; Table 2). There was no significant difference in the half-life of the rapid phase of the turnover ($t_{1/2}\alpha$) between apoSAA$_1$ and apoSAA$_2$ or between the HDL SAA apoprotein from chylomicrons and that of HDL origin (^{131}I versus ^{125}I). The half-life of the slow decay or second exponential is also given in Table 2. The decay rate of apoSAA$_1$ of HDL origin (2.5 days) was significantly shorter ($P<0.01$) than apoSAA$_1$ of chylomicron orgin (4.3 days). A similar trend was found for apoSAA$_2$ of chylomicron versus HDL origin in 3 of 4 animals. If a metabolic steady state is assumed, FCR can be calculated. There was no statistically significant difference between the FCR for apoSAA proteins, although the FCR for the ^{125}I-HDL apoSAA$_1$ was greater than that of ^{131}I-HDL apoSAA$_1$ (Table 2). This was consistent with the shorter $t_{1/2}$ values for the ^{125}I-HDL apoSAA. The FCR calculated by measuring the area under the turnover curves[23] was within 10% of the values calculated by curve peeling.

Discussion

Induction of HDL apoSAA concentrations has been produced in the African green monkey by chair restraint. The induction of apoSAA was rapid (<16 hours) and was sustained for the duration of chair restraint. In addition, chair-restrained animals had a significant amount of apoSAA in thoracic duct lymph chylomicrons. Presumably the increased levels of apoSAA were the result of compression trauma at the point of contact between the skin and chair. After chair restraint the animals were found to have bruising at the points of contact between the skin and chair. Past studies on SAA have relied on invasive means of induction such as injection of casein,[8,24,25] lipopolysaccharide,[21,24] or dextrose.[7] The nonhuman primate promises to be a good animal model for further studies on the metabolism of apoSAA because of lipoprotein distribution, composition,[26,27] and metabolism[18] are more similar to those of man than are those of lower animal forms.

The disappearance of HDL apoSAA from plasma was biphasic; the rapid initial phase (Figure 2 and Table 2) was essentially the same for both apoSAA$_1$ and apoSAA$_2$ regardless of lipoprotein origin (HDL versus chylomicrons). The decay of the second exponential ($t_{1/2}\beta$) was much slower than the first exponential ($t_{1/2}\alpha$). This presumably was due to slow interchange of apoSAA between the plasma compartment and other compartments of the body in equilibrium with the plasma.[23] The metabolism of apoSAA also has been studied in the mouse, and a rapid monophasic disappearance ($t_{1/2}$ = 38 minutes) of apoSAA has been found.[15] However, the $t_{1/2}$ in

the mouse and that in the monkey are not directly comparable because of the difference in the basal metabolic rate. The FCR of vervet HDL apoSAA (Table 2) was more similar to that of low-density lipoprotein (LDL) apoB (1.3 d^{-1}; see Melchoir and Rudel[28]) than that of HDL apoA-I (0.32–0.38 d^{-1}; see Parks and Rudel[18]) and apoA-II (0.42–0.51 d^{-1}; see Parks and Rudel[18]). Thus, the existing data support the conclusion that monkey HDL apoSAA is metabolized more rapidly than the other major apoproteins of HDL. In addition, the data suggest that the metabolism of HDL, as an intact particle, does not occur for all HDL subfractions. Rather, the metabolism of individual protein constituents occurs on different time scales.

Indigenous HDL apoSAA (^{125}I) was found to decay more rapidly than HDL apoSAA of chylomicron origin (Table 2). These data suggest that there may be an intrinsic difference between apoSAA of HDL and chylomicron origin. This hypothetical difference could occur by at least two mechanisms. First, apoSAA may be modified while circulating on plasma HDL; this modification may lead to a decreased half-life (increased FCR) in plasma. The HDL dose was isolated from plasma, and so the apoSAA may have a shorter $t_{½}$, compared with the apoSAA of the lymph chylomicron dose, which was not previously exposed to plasma, if this hypothesis is correct. The second possibility is that the intestine synthesized chylomicron apoSAA, which is slightly different from that which exists in plasma HDL. However, since HDL apoSAA can transfer to chylomicrons[28a] and presumably can transfer from plasma HDL to lymph chylomicrons, the presence of lymph chylomicron apoSAA does not necessarily imply intestinal synthesis of apoSAA. Additional experiments are necessary for one to determine whether differences between plasma and lymph apoSAA exist and whether the intestine is a contributor to the apoSAA pool.

When HDL apoSAA was induced by chair restraint, there was a significant reduction in the percentage of HDL apoA-I and apoA-II. ApoSAA may displace HDL apoA-I and apoA-II and result in a lower percentage composition of these two proteins. Another possibility is that apoA-I and apoA-II become a proportionally smaller amount of the total HDL protein with the addition of large amounts of apoSAA. If apoSAA were merely added to the HDL particle with no displacement of apoA-I and apoA-II or lipid, an increase in the density of the particle would have resulted. No such shift toward denser HDL subfractions in the presence of elevated SAA levels has been found in preliminary studies.[28a] However, Hoffman and Benditt[29] have recently shown that apoSAA tends to be distributed in the denser HDL subfractions of the mouse.

HDL has been reported to bind bacterial lipopolysaccharides[30,31] and to contain elastase type activity[32] in addition to binding apoSAA. These data suggest that HDL may play an important role in the inflammatory response, perhaps in the transportation of factors to the site of injury. It also has been suggested that HDL may function to remove nonpolar toxins or cellular constituents from the site of injury and transport them to the liver for detoxification.[15] ApoSAA appears to associate preferentially with HDL_3 regardless of the relative amount of HDL_3 subfractions.[28a] This preferential association of apoSAA may relate to the ability of HDL_3 to be transformed to HDL_2 like particles by the acquisition of nonpolar constituents.[33] The transformation of HDL_3 to HDL_2 could function to increase the capacity of the HDL particle for transporting nonpolar constituents back to the liver.

References

1. Skipski VP: Lipid composition of lipoproteins in normal and diseased states. Blood, Lipids and Lipoproteins: Quantitation, Composition and Metabolism. Edited by GJ Nelson. New York, Wiley-Interscience, 1972, pp 471–583
2. Osborne JC Jr, Brewer HB Jr: The plasma lipoproteins. Adv Protein Chem 1977, 31:253–337
3. Shore VG, Shore B, Lewis SB: Isolation and characterization of two threonine-poor apolipoproteins of human plasma high density lipoproteins. Biochemistry 1978, 17:2174–2179
4. Parks JS, Rudel LL: Isolation and characterization of high density lipoprotein apoproteins in the non-human primate (vervet). J Biol Chem 1979, 254:6716–6723
5. Eriksen N, Benditt EP: Isolation and characterization of the amyloid-related apoprotein (SAA) from human high density lipoprotein. Proc Natl Acad Sci USA 1980, 77:6860–6864
6. McAdam KPW, Elin RJ, Sipe JD, Wolff SM: Changes in human serum amyloid A and C-reactive protein after etiocholanolone-induced inflammation. J Clin Invest 1978, 61:390–394
7. Malmendier CL, Ameryckx JP: S-peptides-new apoproteins of human high density lipoproteins induced by glucose infusions. Clin Chim Acta 1981, 111:267–270
8. Benson MD, Kleiner E: Synthesis and secretion of serum amyloid protein A(SAA) by hepatocytes in mice treated with casein. J Immunol 1980, 124:495–499
9. Rosenthal CJ, Sullivan L: Serum amyloid A: Evidence for its origin in polymorphonuclear leukocytes. J Clin Invest 1978, 62:1181–1186
10. Rosenthal CJ, Franklin EC, Frangione B, Greenspan J: Isolation and partial characterization of SAA: An amyloid-related protein from human serum. J Immunol 1976, 116:1415–1418
11. Lavie G, Zucker-Franklin D, Franklin EC: Degradation of serum amyloid A protein by surface-associated enzymes of human blood monocytes. J Exp Med 1978, 148:1020–1031

12. Morrow JF, Stearman RS, Peltzman CG, Potter DA: Induction of hepatic synthesis of serum amyloid A protein and actin. Proc Natl Acad Sci USA 1981, 78:4718–4722
13. Sipe JD, Vogel SN, Ryan JL, McAdam KPWJ, Rosenstreich DL: Detection of a mediator derived from endotoxin-stimulated macrophages that induces the acute phase serum amyloid A response in mice. J Exp Med 1979, 150:597–606
14. Selinger MJ, McAdam KPWJ, Kaplan MM, Sipe JD, Vogel SN, Rosenstreich DL: Monokine-induced synthesis of serum amyloid A protein by hepatocytes. Nature 1980, 285:498–500
15. Benditt EP, Eriksen N, Hoffman JS: Origin of protein AA, Amyloid and Amyloidosis, Proceedings 3rd International Symposium on Amyloid. Edited by GG Glenner et al. Amsterdam, Excerpta Medica, 1980, pp 397–405
16. Miller GJ, Miller NE: Plasma-high-density lipoprotein concentration and development of ischaemic heart disease. Lancet 1975, 1:16–19
17. Rudel LL, Reynolds JA, Bullock BC: Nutritional effects on blood lipid and HDL cholesterol concentrations in two subspecies of African green monkeys (*Cercopithecus aethiops*). J Lipid Res 1981, 22:278–286
17a. Klein RL, Rudel LL: Unpublished observations
18. Parks JS, Rudel LL: Different kinetic fates of apolipoproteins A-I and A-II from lymph chylomicra of nonhuman primates: Effect of saturated vs. polyunsaturated dietary fat. J Lipid Res 1982, 23:410–421
19. Matthews CME: The theory of tracer experiments with ^{131}I-labeled plasma proteins. Phys Med Biol 1957, 2:36–53
20. Snedecor GW, Cochran WG: Statistical Methods. Ames, Iowa, Iowa State University Press, 1976.
21. Gorevic PC, Levo Y, Frangione B, Franklin EC: Polymorphism of tissue and serum amyloid A (AA and SAA) proteins in the mouse. J Immunol 1978, 121: 138–140
22. Bausserman LL, Herbert PN, McAdam KPWJ: Heterogeneity of human serum amyloid A proteins. J Exp Med 1980, 152:641–656
23. Shipley RA, Clark RE: Tracer methods for in vivo kinetics: Theory and applications. New York, Academic Press, 1972, pp 77–92
24. McAdam KPWJ, Sipe JD: Murine model for human secondary amyloidosis: Genetic viability of the acute-phase serum protein SAA response to endotoxins and casein. J Exp Med 1976, 144:1121–1127
25. Baumal R, Sklar S, Wilson B, Laskov R: Casein-induced murine amyloidosis: Amyloidogenesis *in vitro* by monolayer spleen explants of casein-injected mice. Lab Invest 1978, 39:632–639
26. Rudel LL: Plasma lipoproteins in atherogenesis in nonhuman primates, Proceedings of the First Annual Symposium on the Use of Nonhuman Primates in Cardiovascular Research. Edited by SS Kalter. Austin, Texas, University of Texas Press, 1980, pp 37–57
27. Rudel LL, Parks JS, Carroll RM: Effects of polyunsaturated vs. saturated dietary fat on nonhuman primate HDL. Am Oil Chem Soc, 1983 (in press)
28. Melchior GW, Rudel LL: Heterogeneity in the low density lipoproteins of cholesterol fed African green monkeys (*Cercopithecus aethiops*). Biochim Biophys Acta 1978, 531:331–343
28a. Parks JS, Rudel LL: Unpublished observations
29. Hoffman JS, Benditt EP: Changes in high density lipoprotein content following endotoxin administration in the mouse: Formation of serum amyloid protein-rich subfractions. J Biol Chem 1982, 257:10510–10517
30. Munford RS, Hall CL, Dietschy JM: Binding of salmonella typhimurium lipopolysaccharides to rat high density lipoproteins. Infect Immun 1981, 34:835–843
31. Ulevitch RJ, Johnston AR, Weinstein DB: New function of high density lipoproteins: Isolation and characterization of a bacterial lipopolysaccharide-high density lipoprotein complex formed in rabbit plasma. J Clin Invest 1981, 67:827–837
32. Jacob MP, Bellon G, Robert L, Hornbeck W, Ayrault-Jarrier M, Burdin J, Polonovski J: Elastase-type activity associated with high density lipoproteins in human serum. Bioch Biophys Res Commun 1981, 103: 311–318
33. Patsch JR, Gotto AM Jr, Olivecrona T, Eisenberg S: Formation of high density lipoproteinα-like particles during lipolysis of very low density lipoproteins *in vitro*. Proc Natl Acad Sci USA 1978, 75:4519–4523

Acknowledgments

The authors wish to thank Mrs. Linda Odham for her assistance in manuscript preparation.