α-Tocopherol is secreted from rat liver in very low density lipoproteins

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Abstract Three separate studies were carried out to test the hypothesis that rat liver secretes vitamin E (α-tocopherol) within very low density lipoproteins (VLDL). *i*) When the clearance of plasma chylomicrons (CM) and VLDL was blocked by the administration of Triton WR-1339, α-tocopherol concentrations increased linearly with time in both classes of triacylglycerol-rich lipoproteins, although accumulation rates within VLDL exceeded those within CM. For fasted rats, appearance of α-tocopherol in VLDL persisted at slightly reduced rates. α-Tocopherol and triglycerides in the VLDL fraction responded to Triton WR-1339 administration by coordinate increases. In contrast to the situation in serum, α-tocopherol concentrations decreased in the liver following injection of Triton. *ii*) In order to inhibit the secretion of hepatic lipoproteins containing apolipoprotein B (apoB), rats were fed a diet containing orotic acid. This resulted in a reduction of apoB and α-tocopherol concentrations in serum and VLDL, whereas the vitamin E content of liver was increased. *iii*) In primary cultures of hepatocytes, α-tocopherol was secreted into the culture media predominantly within VLDL. We, therefore, conclude that the liver secretes α-tocopherol within VLDL and in this way contributes to the maintenance of serum vitamin E concentrations. —**Cohn, W., F. Loechleiter, and F. Weber.** α-Tocopherol is secreted from rat liver in very low density lipoproteins. *J. Lipid Res.* 1988. **29:** 1359–1366.

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α-Tocopherol (vitamin E) is transported in blood within plasma lipoproteins and there is no evidence for the existence of a specific carrier protein as has been found for vitamins A and D (1, 2). In healthy human subjects (3) and in rats (4) all the lipoprotein fractions contain vitamin E, and its distribution parallels the total lipid content in each lipoprotein class. In the rat, the vitamin was found to be most abundant in the HDL fraction followed by VLDL (4, 5). In hyperlipoproteinemic subjects, vitamin E is also predominantly incorporated into the lipoprotein fraction which contains the largest amount of total lipid (3, 6). As α-tocopherol distribution in lipoproteins does not parallel individual lipid classes such as triglycerides, cholesterol, or phospholipids, specific studies are necessary to understand the distribution of vitamin E within lipoproteins. At present it is not known how the distribution of α-tocopherol within lipoproteins is adjusted, but mechanisms such as spontaneous exchange and transfer of the vitamin between lipoproteins have been postulated (7). Alternatively, the presence of vitamin E in plasma lipoproteins might be related to metabolic events such as the appearance of dietary α-tocopherol in newly synthesized CM (2). Similarly, the portion of vitamin E in VLDL might reflect its secretion from the liver in that lipoprotein. An hepatic contribution to the serum transport of vitamin E has been previously considered by Davies et al. (8). These workers demonstrated that feeding orotic acid to rats caused a reduction in serum concentrations of α-tocopherol while liver levels of the vitamin were increased. Malloy et al. (9) also suggested a role for the liver in the maintenance of serum α-tocopherol concentrations when reporting the case of a patient with normotriglyceridemic abetalipoproteinemia. In this patient the serum levels of α-tocopherol were extremely low although CM formation was normal. The larger species of apoB, denoted apoB-100 (10), was absent and since apoB-100 is associated with hepatic VLDL and LDL (10) it was concluded that the low α-tocopherol concentrations reflected a requirement for VLDL and LDL synthesis.

In the present investigation the hypothesis was tested that rat liver secretes α-tocopherol within VLDL. The results of studies both with intact animals and with isolated hepatocytes are presented and are in agreement with our working hypothesis.

Abbreviations: CM, chylomicrons; VLDL, very low density lipoprotein(s); TRL, triglyceride-rich lipoprotein(s) (including CM and VLDL); LDL, low density lipoprotein(s); HDL, high density lipoprotein(s); apoB, apolipoprotein B.

METHODS

Animals and diets

Male SPF "Fü" albino rats from the Institut für Biologisch-Medizinische Forschung AG, CH-4414 Füllinsdorf, were used in all experiments. Animals were fed a commercial diet (NAFAG 850) containing 65 mg vitamin E (all-*rac*-α-tocopheryl acetate) per kg. In some experiments orotic acid at a level of 2% (w/w) was added to the diet (11). In these studies rats (initially weighing 180–220 g) were fed ad libitum for 26 days on diets with or without orotic acid before they were exsanguinated as described below.

When Triton WR-1339 was administered, the rats (250–300 g) were either fed ad libitum or fasted overnight before experiments were started between 7:30 and 8:30 AM. Triton WR-1339 (Serva) dissolved in 0.15 M NaCl, pH 7.4, was injected into the tail vein at a dose of 250 mg/kg body weight (12). At various times after injection, four animals were exsanguinated. Each experiment included a control group of four rats which did not receive an injection of Triton. Rats were bled from the abdominal aorta under Halothane® anesthesia and livers were excised and homogenized. Samples of sera and of homogenized livers were kept at −20°C for lipid analysis. To obtain sufficient starting material for lipoprotein analysis, serum samples from each group of animals were pooled and 11.5 ml was used for the separation procedure.

Isolation and incubation of hepatocytes

Male rats (230–250 g) fed ad libitum were used. Isolation of the hepatocytes of one liver was started at 8:00 AM after induction of Nembutal anesthesia. The procedure of tissue dissociation was as described by Walter et al. (13), but collagenase perfusion was performed without the addition of hyaluronidase. After the hepatocytes had been removed from the surrounding capsule, the cell suspension was filtered through a nylon screen (250 μm mesh size) into precooled centrifugation tubes and the filtrate was centrifuged at 31 *g* for 3 min at 4°C. The cells were washed twice at 4°C and resuspended as described (13). Yields varied between 5 and 8 × 10^8 cells per liver. Cell viability was approximately 90% as judged by Trypan Blue exclusion. For each experiment 50–100 × 10^6 cells were incubated in 25-ml Erlenmeyer flasks in a shaking water bath at 37°C; the cells were suspended in 3 ml of a modified Krebs-Henseleit bicarbonate buffer supplemented with 1.5% fat-free bovine albumin (Sigma) and equilibrated with O_2–CO_2 95:5. The final composition of this buffer was (mM): NaCl, 115.5; KCl, 5.4; $MgSO_4$, 0.8; $NaHPO_4$, 0.8; $NaHCO_3$, 25; $CaCl_2$, 1.3; glucose, 17.0. After incubation for various periods, cells were harvested by centrifugation at 31 *g* for 3 min at 4°C. The hepatocyte incubation medium was carefully removed. Cell pellets were washed with the modified Krebs Henseleit bicarbonate buffer and aliquots were kept at −20°C for protein and lipid analysis.

Isolation of lipoproteins

The pooled sera were subjected to preparative ultracentrifugation at 15°C and the lipoproteins were isolated by flotation (14) through buffer (pH 7.4) containing 10 mM Tris, 1 mM EDTA, 0.2 mM 1,4-dithioerythrite, 0.2 mM phenylmethanesulfonylfluoride, and 0.05% NaN_3 adjusted to d 1.006 g/ml with NaCl (buffer A). TRL from the pooled sera were prepared by ultracentrifugation in a Kontron TGA 65 using a Kontron TST 41.14 rotor at 2.19 × 10^8 g_{av}-min. TRL were recovered by tube slicing and after centrifugation in a Kontron TFT 65.13 rotor at 10^6 g_{av}-min, the floating CM were removed. The residual TRL fraction was concentrated by reverse dialysis at 4°C (15) before the lipoproteins were washed by recentrifugation in a Kontron TST 41.14 rotor at 2.19 × 10^8 g_{av}-min, yielding the VLDL fraction. Both CM and VLDL fractions were concentrated by reverse dialysis and further dialyzed against fresh buffer A. VLDL fractions from hepatocyte incubation medium were isolated by ultracentrifugation using a Kontron TST 54 rotor at 2.33 × 10^8 g_{av}-min and the d > 1.006 g/ml infranate was concentrated by reverse dialysis. CM and VLDL fractions were free of contaminating serum albumin as revealed by sodium dodecyl sulfate polyacrylamide gel electrophoresis. Aliquots of the lipoprotein fractions were collected for protein and lipid analysis.

Analytical methods

Protein in the hepatocytes and lipoprotein fractions was usually determined by the method of Markwell et al. (16) using bovine serum albumin as a reference standard. Interference of sulfhydryl compounds in the protein assay was circumvented by preincubation of the samples with iodoacetate (17). Hepatocyte protein was solubilized in 0.1 N NaOH containing 1% sodium dodecyl sulfate. VLDL isolated from the hepatocyte medium was quantitated by a fluorescent method using *o*-phthalaldehyde (18). VLDL recovered from the hepatocyte medium was precipitated in trichloroacetic acid (5.8%), sodium phosphotungstate (0.3%), and $MgCl_2$ (0.154 M). After centrifugation the sediment was washed twice with 1 N HCl, extracted with methanol–hexane 1:2 (v/v) and washed with methanol. The dried protein was then hydrolyzed in 6 N HCl at 115°C under reduced pressure for 18 hr. The protein hydrolyzates were dried over solid KOH and quantitated using the *o*-phthalaldehyde macroassay (19). ApoB concentrations were determined by the electroimmunoassay technique of Laurell (20) as modified by Bar-On, Roheim, and Eder (21). Anti-apoB antiserum was prepared as described by Ishikawa and Fidge (12).

Cholesterol and triglyceride concentrations were determined enzymatically (22, 23), using commercial kits (Roche Diagnostica, Switzerland). Phospholipids were assayed by a semi-automated procedure (24). α-Tocopherol concentrations were determined by high performance liquid chromatography (HPLC) (25). For the hepatic lipid analyses, homogenized tissues or hepatocytes (300 mg wet weight) were lyophilized and extracted twice with 2 ml of hexane–isopropanol 3:2 (v/v) containing butylated hydroxytoluene (250 μg/ml). Extraction was promoted by sonication (5 × 30 sec) at 4°C. Aliquots of the combined hexane–isopropanol extracts were dried under nitrogen and dispersed in 10 mM Tris buffer, pH 7.4, containing 150 mM NaCl and 1 mM EDTA prior to the enzymatic determination of cholesterol and triglyceride. α-Tocopherol dissolved in hexane was determined by HPLC analysis. Unless stated otherwise the results are expressed as means ± SD. The significance of differences between mean values was calculated by the Student's *t*-test.

RESULTS

Effect of Triton injection on serum and liver α-tocopherol

To study hepatic and intestinal secretion of α-tocopherol associated with TRL, the serum clearance of CM and VLDL was blocked with Triton WR-1339 (26, 27). Triacylglycerol concentrations were then monitored as a measure of TRL accumulation in the plasma. Serum triglycerides and α-tocopherol concentrations increased simultaneously after Triton injection and both lipids accumulated mainly in the TRL fraction (**Table 1**). Recoveries of these lipids from TRL plus infranate fractions varied between 57 and 77% for α-tocopherol and between 59 and 77% for triglycerides. The vitamin E concentration in the d > 1.006 g/ml infranate was increased 1 hr after Triton injection but declined to its initial level when the experiment was continued for 2 hr. In contrast to its accumulation in the serum, α-tocopherol concentrations were significantly reduced in liver 2 hr after Triton injection. After administration of Triton to fed rats, α-tocopherol in VLDL and CM increased linearly with time (**Fig. 1A**). In fasted rats, where VLDL is of predominantly hepatic origin, appearance of α-tocopherol in VLDL persisted at slightly reduced rates whereas it was absent in CM. The secretion of α-tocopherol and triglycerides within VLDL appears to be coupled since the correlation between these components resulted in linear plots passing through the origin (Fig. 1B). The mean ratios of α-tocopherol to triglyceride in VLDL at the different time points were similar in the fed and fasted rats, being 2.41 ± 0.07 and 2.15 ± 0.19 mg/g, respectively. Similar results were obtained when rats were fed an ICN diet (#903079) supplemented with 65 mg vitamin E per kg (data not presented).

Inhibition of hepatic VLDL secretion by orotic acid

Orotic acid was used to study whether a diminished secretion of hepatic VLDL would result in lowered serum α-tocopherol levels. Orotic acid inhibits the release of VLDL and LDL from the liver into blood whereas the capacity for synthesis of intestinal lipoproteins remains unchanged (11, 28). After feeding rats orotic acid, the serum α-tocopherol concentrations decreased continuously for 18 days after which levels remained constant, suggesting that a steady state had been reached (data not presented). Results are shown after 26 days of orotic acid feeding at which time the concentrations of α-tocopherol, triacylglycerol, cholesterol, and apoB in whole serum were reduced (**Table 2**). α-Tocopherol, triacylglycerol, and phospholipids associated with VLDL showed approximately a 50% decrease in concentration after orotic acid administration whereas the fall in apoB concentration was

TABLE 1. Changes in α-tocopherol and triacylglycerol concentrations in serum and liver following Triton administration

Time after Triton	Liver[a]		Serum		TRL[b] (d 1.006 g/ml)		Infranate[b] (d 1.006 g/ml)
	α-T	TG	α-T	TG	α-T	TG	α-T
hr	*μg/g*	*mg/g*	*mg/l*	*g/l*	*mg/l*	*g/l*	*mg/l*
0[c]	21.64 ± 2.4	7.48 ± 0.79	8.10 ± 0.75	0.58 ± 0.10	1.18	0.38	3.90
1	19.15 ± 3.15	6.79 ± 1.02	17.59 ± 0.65[d]	4.87 ± 0.73[d]	6.65	3.65	6.84
2	13.52 ± 0.99[d,e]	7.25 ± 1.02	22.28 ± 2.11[d,e]	11.15 ± 0.75[d,e]	10.94	6.59	3.78

Rats were fed ad libitum. Data for liver and whole serum are means ± SD of four rats; α-T, α-tocopherol; TG, triacylglycerol.
[a]Liver lipid concentrations are expressed per g wet weight.
[b]Concentrations in serum fractions are expressed per liter of the pooled serum; data are means of four determinations. CV was less than 5% of the mean in all cases.
[c]Control, no Triton injection.
[d]Significantly different from controls ($P < 0.01$).
[e]Significantly different from 1 hr treatment ($P < 0.05$).

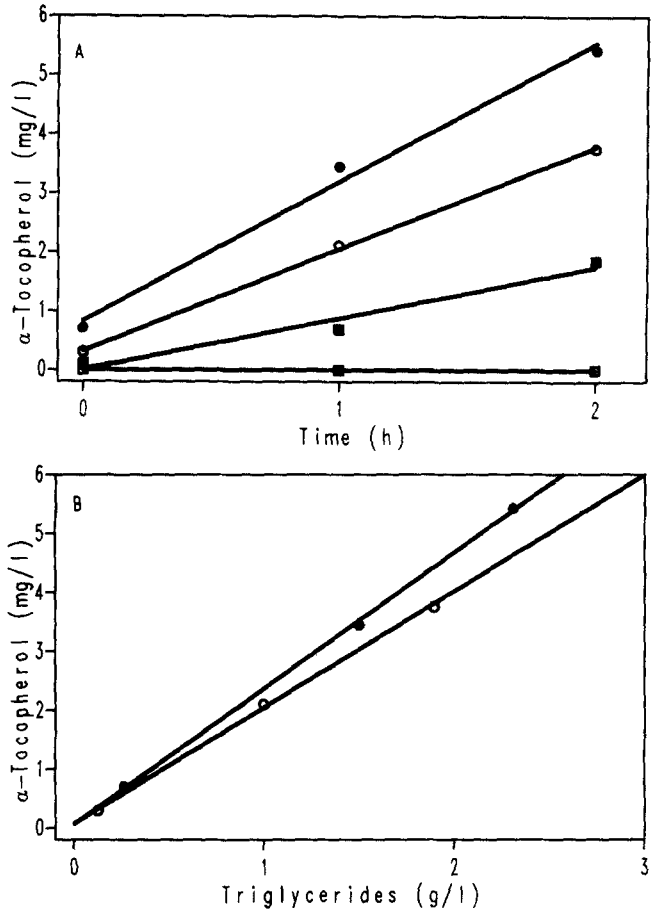

Fig. 1. α-Tocopherol accumulation within triglyceride-rich lipoprotein fractions following Triton administration. Concentrations are expressed in terms of 1 liter of pooled serum from which the lipoprotein fractions had been separated. Data are means of four lipid determinations in samples of VLDL of fed rats (●), VLDL of rats fasted for 16 hr (○), and CM of fed (■) and fasted (□) rats. Time course of α-tocopherol accumulation (A), and correlation of α-tocopherol and triglycerides associated with VLDL (B).

greater (72%). Concomitantly, orotic acid feeding caused an increase in liver concentrations of α-tocopherol and cholesterol, and also induced a slight gain in liver weight (**Table 3**). There was no significant change in triglyceride concentrations.

α-Tocopherol secretion from isolated rat hepatocytes

The secretion of α-tocopherol from isolated rat hepatocytes was predominantly associated with VLDL, as only a small amount of vitamin E was recovered in the d > 1.006 g/ml infranate fraction (**Fig. 2A**). When cells had been incubated for 150 min, 77.7% of the α-tocopherol (or 12.7 ng/mg cell protein) and 80.6% of the apoB (or 67.5 ng/mg cell protein) were incorporated in the VLDL fraction and the residue was isolated in the d > 1.006 g/ml infranate. The α-tocopherol content of the VLDL fraction increased linearly with time for at least 60 min (Fig. 2A), whereas hepatocyte α-tocopherol levels did not alter significantly with incubation time (Fig. 2B). Linear kinetics of appearance were also observed for VLDL triacylglycerol and apolipoproteins; the secretion rates were 6.93×10^{-3}, 0.27 and 2.44 $\mu g \times h^{-1} \times (\text{mg cell protein})^{-1}$ for α-tocopherol, apolipoproteins, and triacylglycerols, respectively. The parallel secretion of these components of VLDL resulted in linear correlation plots (**Fig. 3**) which gave incorporation ratios of 2.84 and 25 μg/mg apolipoprotein for α-tocopherol and triacylglycerol, respectively.

A decrease in α-tocopherol levels within the hepatocytes had been expected after secretion into the medium. However, after an incubation period of 60 min, only 8.8% of the cellular α-tocopherol was released into the medium and the resulting vitamin E reduction was well within the detection error for corresponding cellular levels. There were no significant changes in intracellular triglyceride content during the incubation period; concentrations were 38 ± 7 and 43 ± 8 μg/mg cell protein at 10 and 60 min, respectively (n = 3).

DISCUSSION

Three complementary approaches have been applied to elucidate the contribution of hepatic VLDL to α-tocopherol transport from the liver into the plasma. To study α-tocopherol influx within TRL in the intact animal, we measured the secretion of the vitamin into serum after injection of Triton WR-1339. The capacity of

TABLE 2. Lipid and apoB concentrations in serum and VLDL following orotic acid administration

Fraction	Diet	α-T	TG	PL	CH	ApoB
		mg/l	*g/l*	*g/l*	*g/l*	*mg/l*
Serum	BD	7.15 ± 0.80	0.54 ± 0.11	1.51 ± 0.09	0.53 ± 0.06	64.2
Serum	OA	4.00 ± 1.20[a]	0.31 ± 0.13[a]	1.44 ± 0.08	0.36 ± 0.09[a]	24.2
VLDL	BD	1.09	0.36	0.19	0.03	25.9
VLDL	OA	0.48	0.17	0.08	ND	7.3

Rats were fed Nafag 850 as a basal diet (BD) or Nafag 850 containing 2% orotic acid (OA) for 26 days. With the exception of apoB values, serum concentrations are means ± SD of 12 rats. VLDL fractions were separated from pooled serum and both VLDL and apoB concentrations are expressed as explained in Table 1 for lipoprotein fractions; α-T, α-tocopherol; TG, triacylglycerol; PL, phospholipids; CH, total cholesterol; ND, not determined.

[a]Significantly different from rats fed BD ($P < 0.01$).

TABLE 3. Lipid levels in liver of rats fed orotic acid

Diet	Liver weight	α-T	TG	CH
	g	*μg/g*	*mg/g*	*mg/g*
BD (n = 12)	11.9 ± 1.2	21.7 ± 2.4	7.48 ± 0.79	2.58 ± 0.18
OA (n = 12)	13.7 ± 1.4[a]	36.6 ± 5.5[b]	8.72 ± 1.41	4.55 ± 0.68[b]

Abbreviations as in Table 2; lipid concentrations are expressed per g wet weight. The values represent the means ± SD; n, number of animals.
[a]Significantly different from rats fed BD ($P < 0.05$).
[b]Significantly different from rats fed BD ($P < 0.01$).

this nonionic detergent to block the intravascular metabolism of TRL has previously been employed to measure both cholesterol and triglyceride secretion into plasma (27, 29, 30) and also production of intestinal lipoproteins (31, 32). Following the injection of Triton, α-tocopherol accumulated predominately within the TRL fraction (Table 1). The finding of a parallel increase of α-tocopherol and triacylglycerol in the TRL fraction is suggestive evidence for secretion of the vitamin together with nascent TRL. Since the concentrations of α-tocopherol associated with the d > 1.006 g/ml infranate did not vary consistently (Table 1) it appears unlikely that the elevation of the vitamin within the TRL fraction was related to exchange processes from HDL. Such processes have, however, been demonstrated in vitro (7) when HDL containing ^{3}H-α-tocopherol was incubated with LDL or VLDL.

α-Tocopherol is known to enter the circulation by means of CM and intestinal VLDL (2). After Triton administration α-tocopherol accumulated predominantly in VLDL and not in CM (Fig. 1A). Since VLDL represented the major lipid compartment in serum (data not presented), the high concentrations of α-tocopherol in this fraction might have been related to thermodynamic partitioning between CM and VLDL. However, in fasted rats, when no CM are formed and when VLDL is of predominantly hepatic origin, appearance of α-tocopherol in VLDL persisted (Fig. 1A). This indicates that hepatic VLDL are carriers of α-tocopherol. As Triton WR-1339 is known to prevent both TRL catabolism (27) and remnant binding to liver plasma membranes (33), the accumulation of α-tocopherol in the plasma should coincide with a reduced influx of the vitamin into the liver. The finding of reduced hepatic α-tocopherol levels (Table 1) is consistent with the secretion of the vitamin into the plasma or bile (34), but might also reflect its metabolism.

Although our results suggest a contribution of hepatic VLDL to α-tocopherol plasma concentrations, studies using Triton must also take into account the marked changes in lipoprotein structure and apolipoprotein content that result from its use (12). We confirmed that Triton injection into rats caused a dissociation of apoE and apoC from VLDL (data not presented). In order, therefore, to ascertain whether hepatic VLDL plays a major role in the transport of α-tocopherol in serum, additional evidence was required.

The use of orotic acid demonstrated that the inhibition of hepatic VLDL secretion resulted in reduced α-tocopherol levels in serum and confirmed the finding of Davies et al. (8). The addition of orotic acid to purified diets induces severe fatty liver in rats (35); therefore, in the present study animals were fed a diet based on natural ingredients. In consequence, liver triacylglycerols were only marginally raised (Table 3). This suggested that secretion of hepatic VLDL was only partially blocked.

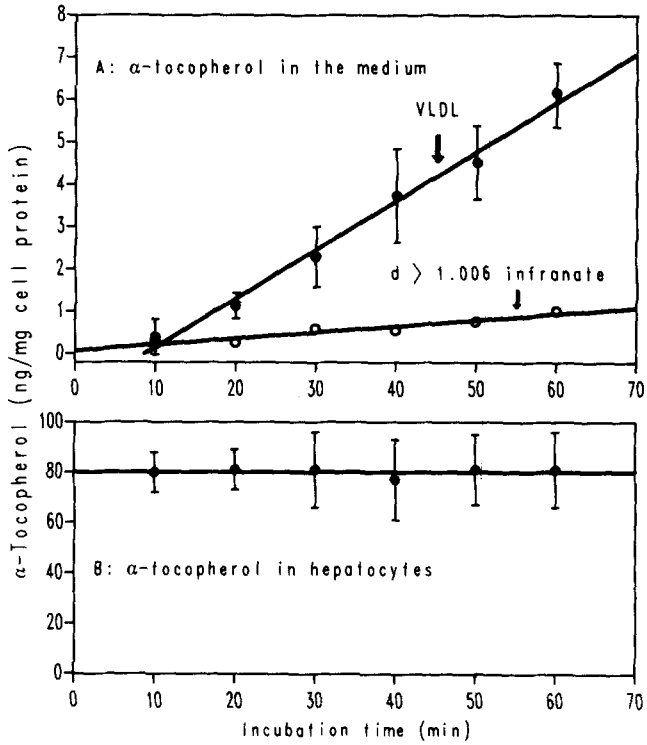

Fig. 2. Time course of α-tocopherol secretion by hepatocytes into the culture medium. α-Tocopherol associated with VLDL (●) or within the d > 1.006 infranate (○) (A) and α-tocopherol concentration in hepatocytes (B). For VLDL and hepatocytes, values are the mean ± SD of three independent hepatocyte preparations. For the d > 1.006 g/ml infranate, points represent the mean of two experiments. Cell viability after 10 and 60 min incubation were similar, 93 ± 2% and 90 ± 3%, respectively.

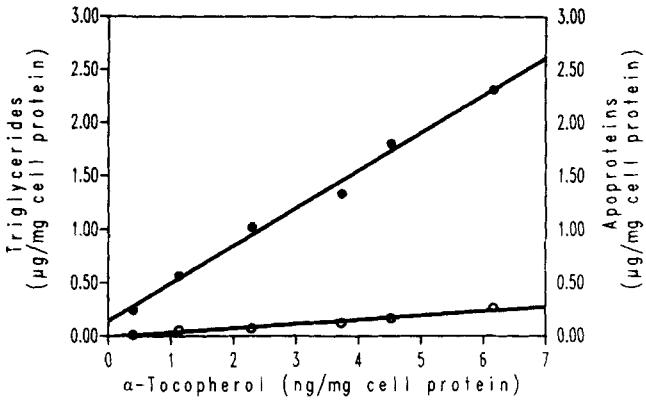

Fig. 3. Coordinate secretion of α-tocopherol and triglycerides (●) or apolipoprotein (○) as components of isolated VLDL released by hepatocytes. Values represent the means of three determinations.

Nevertheless, orotic acid caused a decrease of serum lipid and apoB levels and, in particular, VLDL concentrations were lowered (Table 2). Since tocopherol concentrations were reduced by a similar percentage in both serum and VLDL, we concluded that orotic acid feeding inhibited the secretion of the vitamin within hepatic VLDL, and as a consequence the liver α-tocopherol content was increased (Table 3). Orotic acid treatment appears to be analogous to the situation observed in a patient with normotriglyceridemic abetalipoproteinemia. In both situations the secretion of hepatic VLDL is inhibited, whereas the synthesis of intestinal lipoproteins remains unchanged (9, 11, 28, 36). Moreover, in both situations there is a reduction in serum α-tocopherol concentrations, emphasizing the role of α-tocopherol secretion within hepatic VLDL in the maintenance of serum vitamin E concentrations.

Studies with isolated rat hepatocytes avoid interference from nonhepatic plasma lipoproteins and provide a suitable system for confirming that α-tocopherol is secreted within VLDL. The data obtained do not allow us to distinguish whether VLDL are synthesized and secreted or just secreted during the incubation periods. We confirmed (37) that the release of VLDL could be inhibited by cycloheximide (data not presented). The release of VLDL is, therefore, subject to a regulatory mechanism and it was not just leaking from broken cells. VLDL are the major lipid-carrying particles released by hepatocytes (37) and these cells do not metabolize VLDL at an appreciable rate (38). Vitamin E secreted into the culture medium was almost exclusively recovered in the VLDL fraction (Fig. 2A), confirming that VLDL was the predominant vehicle for α-tocopherol transport from the liver. The composition of the secreted VLDL in terms of α-tocopherol, triacylglycerol, and apoB did not vary with incubation time (Fig. 3). This is consistent with the findings from the Triton experiments and confirms their parallel incorporation into VLDL. Our results supplement the findings of Kempen (37) who reported that primary hepatocytes release VLDL with a constant molar proportion of triacylglycerols, phospholipids, and cholesterol. At present we do not know how α-tocopherol is incorporated into VLDL during its assembly but the incorporation rate for α-tocopherol in VLDL is likely to depend on the vitamin's supply in liver. Under our experimental conditions, only small amounts of cellular α-tocopherol were released into the medium (Fig. 2). A relatively constant intracellular α-tocopherol concentration might, therefore, be a prerequisite for a uniform incorporation rate of the vitamin into VLDL.

The combined results from this study provide conclusive evidence that VLDL is the predominant vehicle for α-tocopherol transport from the liver and is, therefore, an important contributor to the circulating α-tocopherol level. It appears that the entry of α-tocopherol into serum depends on the secretion of both CM and VLDL. Since the delivery of dietary α-tocopherol by CM fluctuates with the load of absorbed vitamin E, hepatic VLDL are important for maintaining α-tocopherol serum concentrations. Hepatic VLDL are the precursors of LDL. We therefore assume that the α-tocopherol concentration within LDL is related to the vitamin secreted in hepatic VLDL. Recently, the high affinity receptor for LDL has been recognized to function as a mechanism for delivery of vitamin E to fibroblasts in culture (39, 40). Thus, lipoprotein species have a defined role in vitamin E secretion, transport, and distribution to tissues though the contribution of HDL to α-tocopherol transport is not yet known. Furthermore, during lipid transport in plasma and subsequent transfer to tissues, polyunsaturated fatty acyl chains are always protected from free radical attack by the presence of vitamin E.

ADDENDUM

After this manuscript was submitted Bjørneboe et al. (41) confirmed that α-tocopherol is secreted within VLDL from cultured rat hepatocytes.

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