

Biochemical and ultrastructural alterations in platelets, reticulocytes, and lymphocytes from rats fed vitamin E-deficient diets

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Abstract Effects of vitamin E-deficiency, dietary fat (corn oil versus lard), and incubation on ultrastructure of platelets, red and white cells, and on selected biochemical parameters of platelets and plasma were studied. Platelets from vitamin E-deficient rats had no obvious morphological defects, but reticulocytes and lymphocytes had swollen and deformed mitochondria. Fatty acid and glycogen levels of platelets were not affected by the deficiency but total lipid levels in plasma were decreased or increased depending upon the type and level of dietary fat. In comparison with supplemented controls, the proportion of stearate increased in the phospholipid fraction of plasma from vitamin E-deficient, lard-fed but not corn oil-fed rats. In platelets, total fatty acids per mg protein were 8–12% lower with lard than with corn oil as fat source. Oleate and linoleate were higher and lower, respectively, although not to the same degree, and arachidonate was not affected. With incubation of platelet-rich plasma at 37°C for 6 hr, there were no obvious morphological changes in platelets from control or from deficient, lard-fed rats, but platelets from deficient, corn oil-fed rats contained mitochondria that were swollen and deformed. Incubation did not affect fatty acid, glycogen, or tocopherol levels of platelets or tocopherol levels of plasma, regardless of type or amount of dietary fat.—**Lehmann, J. and M. McGill.** Biochemical and ultrastructural alterations in platelets, reticulocytes, and lymphocytes from rats fed vitamin E-deficient diets. *J. Lipid Res.* 1982. 23: 299–306.

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Published reports have consistently described ultrastructural changes in mitochondria of several tissues as one effect of vitamin E deficiency (1–3). In our studies of platelets, however, we could not detect similar changes in mitochondria of fresh platelets from vitamin E-deficient rats. Subsequently, we induced ultrastructural abnormalities in mitochondria of vitamin E-deficient platelets by incubating platelet-rich plasma (PRP) at 37°C. The rationale for this approach was that if platelet mitochondria had qualitative abnormalities, stimulation of mitochondrial activity by incubation at 37°C (4) might initiate morphological changes representative of the defects. In this report, morphological alterations in platelets

from rats fed vitamin E-deficient diets containing two levels of either lard (low linoleic acid) or corn oil (high linoleic acid) are described and possible relationships between these observations and measurements of changes in fatty acid, tocopherol, and glycogen content in platelets and plasma are discussed. Both lard and corn oil were used as fat sources in order to vary the stress on vitamin E-deficiency that is associated with dietary and tissue levels of polyunsaturated fatty acids (PUFA). In the course of these studies we observed abnormalities of mitochondria in reticulocytes and lymphocytes from vitamin E-deficient rats, which are also reported.

MATERIALS AND METHODS

Diet and treatment of animals

Male weanling (50–55 g) Wistar rats (Hilltop Lab Animals, Inc., Scottsdale, PA.), eight per group were placed two to a cage for 6 weeks and then separated to individual cages and fed, ad libitum, a basal vitamin E-deficient diet (5) supplemented with either 0 or 50 ppm D- α -tocopheryl acetate (ICN Pharmaceuticals, Inc., Life Science Group, Cleveland, OH). Tocopherol-stripped corn oil or lard (5% or 15%) was added at the expense of sucrose. Fatty acid composition of the diets is shown in **Table I**. After 15 weeks on the diets, the rats were fasted overnight and anesthetized with sodium pentobarbital.

Sample preparation

Blood was drawn from the hearts of the anesthetized animals into 20-ml plastic syringes containing 1 ml of ACD anticoagulant (0.8 g citric acid, 2.2 g sodium citrate, and 2.45 g glucose in 100 ml of distilled water), divided equally between two 12-ml plastic centrifuge

Abbreviations: ACD, acid citrate dextrose; PRP, platelet-rich plasma; PPP, platelet-poor plasma; GLC, gas-liquid chromatography; PUFA, polyunsaturated fatty acids.

TABLE 1. Dietary fatty acid composition

Fatty Acid	Diet			
	5% CO ^a	15% CO	5% L	15% L
	<i>g/100 g diet</i>			
16:0	0.5	1.5	1.1	3.2
16:1			0.2	0.5
18:0	0.2	0.6	0.6	1.7
18:1	1.2	3.6	2.1	6.2
18:2	3.0	8.9	0.6	1.7

^a CO, corn oil; L, lard.

tubes and diluted 1:9 with additional ACD. Whole blood was centrifuged at 1200 rpm for 12 min at 20°C. The buffy coat was resuspended by swirling gently, and the tubes were recentrifuged for 5 min at 700 rpm. Platelet-rich plasmas (PRP) from all animals in the same group were combined in a 50-ml plastic centrifuge tube and diluted with platelet-poor plasma (PPP) to a platelet count of $6.0\text{--}6.5 \times 10^7$ cells per ml. PPP was obtained by centrifuging the remaining plasma and red blood cell concentrate at 3000 rpm for 10 min at 20°C. The PRP (36 ml) was then divided equally among four 12-ml plastic tubes, capped tightly and incubated for 0, 2, 4, or 6 hr at 37°C in a water bath.

Electron microscopy

Platelet-rich plasma or buffy coats with red cells were fixed with equal volumes of 0.2% glutaraldehyde (Tousimis Research Corp., Bethesda, MD) in Millonig's buffer for 1 hr at room temperature and centrifuged at 1500 rpm. The resulting pellet was resuspended in 5 ml of 3% glutaraldehyde in Millonig's buffer for a minimum of 1 hr. Samples were post-fixed with 1% osmium tetroxide, pre-stained with uranyl acetate, dehydrated in ethanol, and embedded in Epon by standard techniques.

Biochemical analyses

Platelets were isolated from the incubation mixture and plasma and platelets were both analyzed immediately for α -tocopherol by a gas-liquid chromatographic (GLC) method (6). Lipids were extracted from plasma and platelets with chloroform-methanol 2:1. Internal standards for fatty acid analyses (trimyristin, cholesteryl arachidate, and diheptadecoyl lecithin for plasma, and methyl tricosenoate for platelets) were added with the extraction solvents. Individual lipid classes in plasma were separated on silica gel HR plates developed in hexane-acetone-acetic acid 79:11:3. Methyl esters were prepared by treatment with boron trifluoride-methanol (7) and analyzed by GLC (8).

Glycogen content of platelets was analyzed by the micromethod of Van Handel (9) with anthrone reagent.

Susceptibility of red blood cells to hemolysis was determined by the method of Draper and Csallany (10).

Statistics

A representative sampling of platelets for electron microscopy was obtained by pooling platelet-rich plasma from eight rats fed the same diet. Where appropriate groupings of analytical values for these pooled samples were possible, statistical significance was evaluated by the Student's *t*-test. Even though the variability within these pooled samples cannot be evaluated statistically, the analyzed values represent the mean values for eight rats.

Sizes and shapes of platelet mitochondria were estimated by measuring, on each mitochondrial profile, the longest line which could be drawn between two points on the profile surface (maximum diameter), and the length of a second line perpendicular to and passing through the midpoint of the first line, connecting two other points on the profile circumference (secondary diameter). Comparisons of mean (\pm SD) diameter values were performed by the Student's *t*-test.

RESULTS

Electron microscopy

Platelets from control rats possessed ultrastructural features described for other species. They were discoid in shape and contained numerous surface-connecting system channels (invaginations of the plasma membrane), secretory granules, and mitochondria (Fig. 1). When platelets from rats fed adequate vitamin E were suspended in plasma and incubated at 37° for 6 hr, they retained their discoid configurations and ultrastructural features of organelles were unchanged (Fig. 2). Maintenance of rats on vitamin E-deficient diets with either corn oil or lard also did not induce obvious morphological changes in platelets (Fig. 3). However, when platelets from deficient rats fed corn oil were incubated at 37°C for up to 6 hr, abnormalities in mitochondria were visualized (Figs. 4–6). Mitochondria in these vitamin E-deficient platelets were swollen, contained fewer cristae, and did not display the usual electron density. Examination of many thin sections indicated that the extent and frequency of mitochondrial abnormalities increased with time at 37°C. A quantitative morphological analysis for 2 and 4 hr of incubation was not performed. At 6 hr, however, all mitochondrial profiles in platelets from these vitamin E-deficient, corn oil-fed rats showed morphological abnormalities such as those illustrated in Figs. 4–6 and the maximum and secondary diameters (μ m) were increased from 367 ± 66 (SD) to 483 ± 67 (*P*

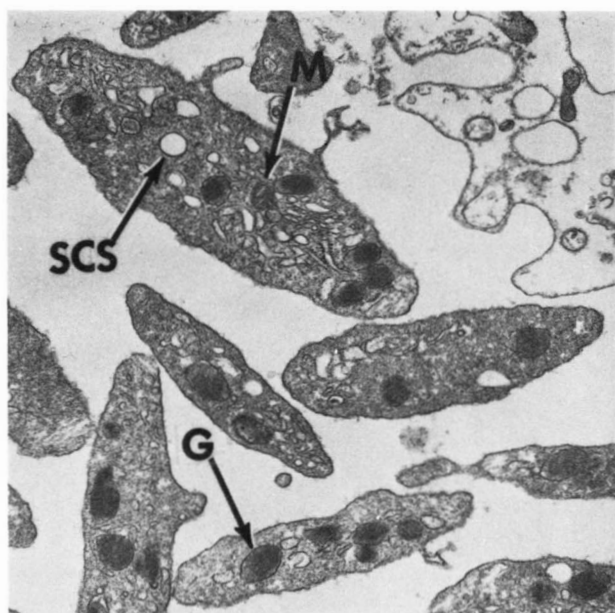


Fig. 1. Transmission electron micrograph of pooled platelets from rats maintained on vitamin E-adequate diets (controls). Morphologically, rat platelets are similar to those of other mammalian species. At this magnification, surface-connecting system membrane (SCS), secretory granules (G), and mitochondria (M) can be identified. $\times 14,600$.

< 0.001) and from 264 ± 49 to 338 ± 72 ($P < 0.02$), respectively. No morphological changes induced by incubation were observed in the pooled samples from rats receiving vitamin E-deficient diets containing lard (**Fig.**

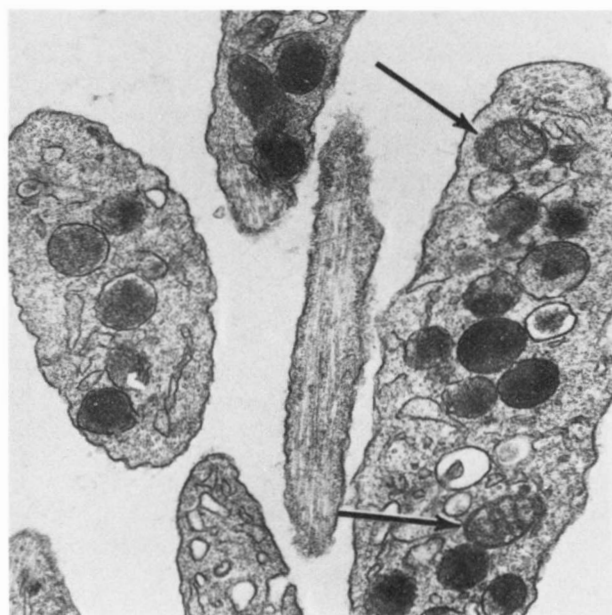


Fig. 2. Platelets from control rats after incubation in platelet-rich plasma for 6 hr at 37°C . Incubation did not induce any obvious changes in ultrastructure. Mitochondria (arrows) were not increased in size and displayed the usual electron density peculiar to platelet mitochondria. $\times 23,300$.

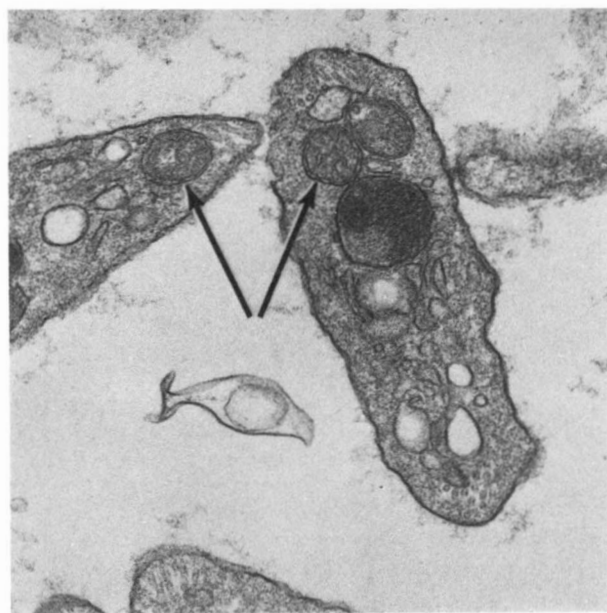


Fig. 3. Platelets from rats fed vitamin E-deficient diets were identical, morphologically, to control platelets. Mitochondria are at arrows. $\times 34,500$.

7). Erythrocytes and lymphocytes from buffy coat preparations from rats fed both levels of corn oil were also observed in the electron microscope. Samples from vitamin E-deficient rats contained lymphocytes in which all mitochondria were abnormal (**Fig. 8**), and numerous reticulocytes containing large numbers of mitochondria exhibiting swelling and loss of cristae (**Fig. 9**).

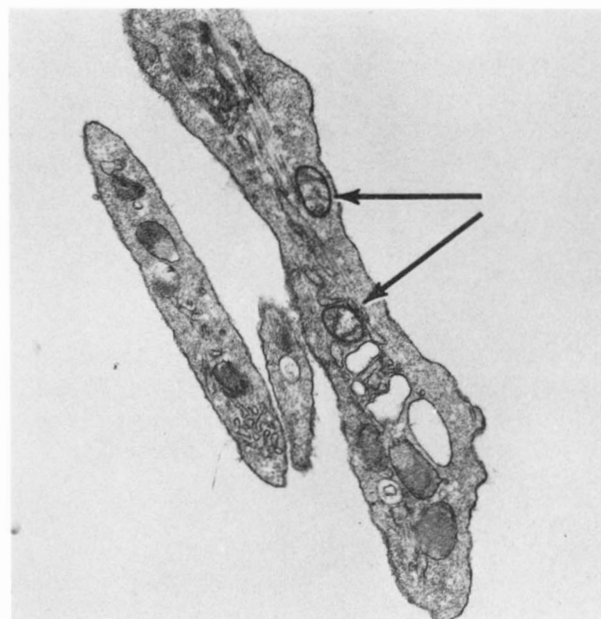


Fig. 4. Platelets from animals fed vitamin E-deficient diets (corn oil) after incubation at 37°C for 2 hr. Mitochondria (arrows) exhibited obvious ultrastructural abnormalities. $\times 23,000$.

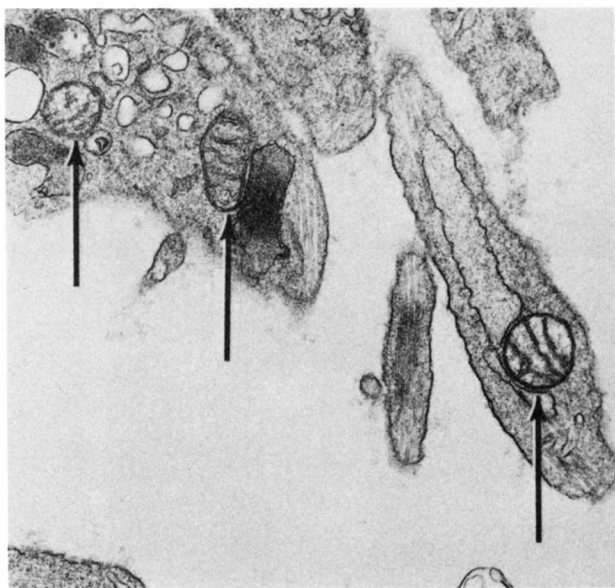


Fig. 5. Platelets from animals fed vitamin E-deficient diets (corn oil) after incubation at 37°C for 4 hr. Mitochondria (arrows) were swollen and exhibited loss of cristae. $\times 23,000$.

Biochemical

Effects of vitamin E deficiency. Vitamin E deficiency was confirmed by greater than 90% hemolysis of red blood cells subjected to the in vitro spontaneous hemolysis test. Appetite, body weight, and testes weight were not affected by the deficiency.

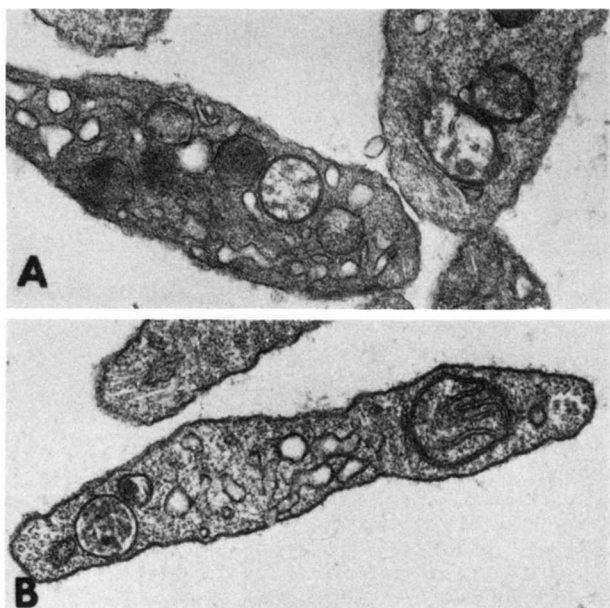


Fig. 6. Platelets from animals fed vitamin E-deficient diets (corn oil) after incubation at 37°C for 6 hr. All mitochondria exhibited morphological abnormalities including swelling, loss of cristae, and decreases in electron density. A, $\times 26,600$; B, $\times 35,600$.



Fig. 7. Platelets from rats fed vitamin E-deficient diets (lard) did not show changes in mitochondrial morphology after incubation at 37°C. Mitochondria are at arrows. $\times 21,000$.

Compared to their respective controls, total lipids in pooled plasmas were lower for vitamin E-deficient rats fed either level of corn oil or the higher level of lard, and were higher for vitamin E-deficient rats fed the lower level of lard (**Table 2**). With the deficiency, total 20:4 on an absolute basis (mg/100 ml plasma) was lower in

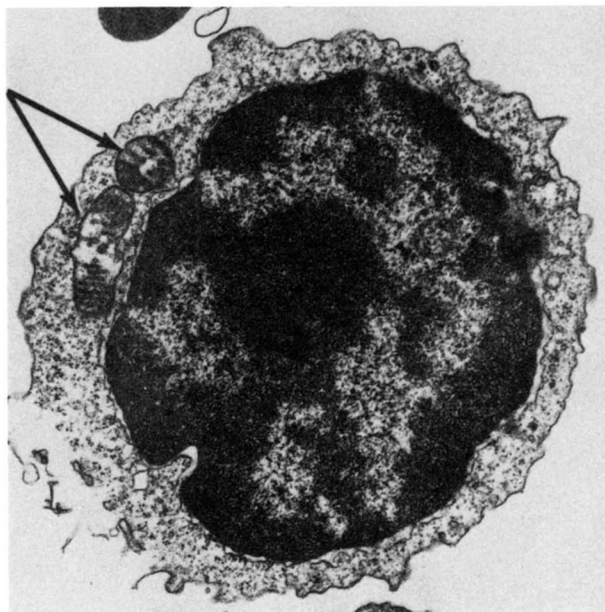


Fig. 8. Blood cells from buffy coat layer of whole blood from rats fed vitamin E-deficient diets (corn oil) fixed without incubation at 37°C. Mitochondria (arrows) in all lymphocytes exhibited swelling and loss of cristae. $\times 13,800$.

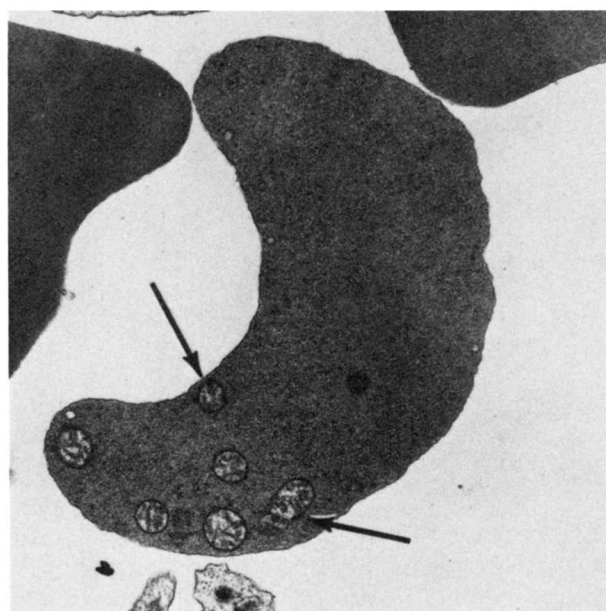


Fig. 9. Blood cells from buffy coat layer of whole blood from rats fed vitamin E-deficient diets (corn oil) fixed without incubation at 37°C. Thin sections of reticulocytes contained numerous mitochondrial profiles (arrows) displaying the same ultrastructural abnormalities observed in lymphocytes. $\times 12,700$.

plasma from animals fed corn oil ($P < 0.05$), but not from lard-fed rats. However, absolute changes with the deficiency of most fatty acids of the major lipid fractions generally reflected the differences in total lipids, and, as a percentage of total fatty acids, 20:4 was not affected by the deficiency with either fat. The only fatty acid in plasma notably affected by vitamin E deficiency was 18:0 which, in comparison to supplemented controls, was increased ($P < 0.05$) proportionally in the phospholipid

fraction of deficient, lard-fed animals. Vitamin E deficiency did not affect the levels of fatty acids (Table 3) or glycogen in pooled platelets.

Tocopherol content of pooled platelets and plasma are shown for the vitamin E-supplemented rats in Table 4. Only trace amounts of tocopherol were detected in vitamin E-deficient platelets ($<0.05 \mu\text{g}/\text{mg}$ protein and plasma ($<0.2 \mu\text{g}/\text{ml}$).

Effects of type and level of fat. Platelet total fatty acid content per mg protein was 8–12% lower ($P < 0.001$) with lard than with corn oil as the dietary fat (Table 3). Most of the difference was accounted for by decreases in 16:0 ($P < 0.01$), 18:0 ($P < 0.05$), and 18:2 ($P < 0.001$), while 20:4 was not affected. Oleate, 18:1, increased ($P < 0.1$) slightly as would be expected to reflect the higher level of 18:1 in lard (Table 1), but the increase did not compensate for decreases of the other fatty acids. Increasing corn oil from 5 to 15% of the diet resulted in an increase in 18:2 that was compensated for by a decrease in 18:1. Glycogen levels in platelets were not affected by changes in either the type or amount of fat.

As shown in Table 2, with lard compared to corn oil as the dietary fat, 18:2 in plasma was lower in both the triglyceride ($P < 0.01$) and phospholipid ($P < 0.001$) fractions. However, there was no significant effect of type or amount of fat on levels of 20:4.

Effects of incubation. Tocopherol levels of either plasma or platelets were not affected by 6 hr of incubation at 37°C. Incubation also did not affect either fatty acid or glycogen levels in platelets from any of the groups including those deficient in vitamin E. Since incubation had no effect on tocopherol or fatty acid levels of platelets, the means presented are pooled for each diet group (Tables 3 and 4). Also, since none of the experimental

TABLE 2. Plasma^a levels of total lipid and the principal fatty acids of the major plasma lipid fractions from rats fed 5 and 15% corn oil or lard diets with and without vitamin E

		Total Lipid	Fatty Acids									
			Triglyceride				Phospholipid					Cholesteryl ester
			16:0	18:1	18:2	20:4	16:0	18:0	18:1	18:2	20:4	20:4
mg/100 ml												
5% CO	+E	377	27	34	29	9	25	29	8	11	36	30
	−E	316	25	30	23	7	19	22	9	10	28	23
15% CO	+E	311	15	15	25	7	21	28	5	11	33	25
	−E	272	16	14	20	4	17	23	4	9	27	24
5% L	+E	339	33	61	8	5	20	19	14	6	27	21
	−E	372	37	70	11	5	22	32	14	5	26	25
15% L	+E	376	41	65	17	8	21	26	9	6	34	21
	−E	330	31	50	9	5	17	30	9	6	30	24

^a Each sample is composed of plasma pooled from eight rats.

TABLE 3. Fatty acid content of platelets from rats^a fed diets containing either corn oil or lard with or without vitamin E

Fatty Acids	+Vitamin E				-Vitamin E			
	Corn Oil		Lard		Corn Oil		Lard	
	5%	15%	5%	15%	5%	15%	5%	15%
	$\mu\text{g}/\text{mg protein}$							
16:0	29.9 \pm 2.7 ^b	29.6 \pm 1.8	28.0 \pm 1.4	26.5 \pm 2.4	30.5 \pm 2.2	30.2 \pm 0.8	26.8 \pm 0.4	23.9 \pm 0.5
18:0	16.9 \pm 1.1	17.8 \pm 0.6	15.5 \pm 1.1	16.9 \pm 2.0	17.6 \pm 0.9	19.9 \pm 0.6	16.4 \pm 0.1	16.3 \pm 1.9
18:1	9.0 \pm 0.5	7.1 \pm 1.3	9.9 \pm 0.7	9.1 \pm 0.8	9.6 \pm 0.4	6.6 \pm 0.5	10.8 \pm 0.4	8.7 \pm 1.0
18:2	5.8 \pm 0.3	7.6 \pm 0.8	2.1 \pm 0.2	3.3 \pm 0.4	6.0 \pm 0.3	7.5 \pm 0.2	2.8 \pm 0.5	3.3 \pm 0.3
20:4	26.7 \pm 0.9	26.3 \pm 1.0	25.0 \pm 1.9	27.1 \pm 2.6	26.8 \pm 1.3	27.3 \pm 0.9	25.3 \pm 0.5	26.3 \pm 1.6
Total	88.3	88.4	80.5	82.9	90.5	91.5	82.1	78.5
P/S ^c	0.69	0.71	0.65	0.70	0.68	0.69	0.65	0.74

^a Each sample is composed of platelets pooled from eight rats.^b Mean \pm SD, N = 4. Because incubation had no effect, fatty acid values for 0, 2, 4, and 6 hr were averaged.^c P/S = ratio of polyunsaturated to saturated fatty acids.

variables affected glycogen levels in platelets, the data for all four diets with and without vitamin E were pooled with a mean of 21.7 μg glycogen/mg protein \pm 1.2 (SD). There was a decrease in pH in all plasmas from 7.4 to not less than 6.9 during the 6-hr incubations. (Platelet ultrastructure is unchanged by decreases in plasma pH to below 6.5 (11))

There were no changes during incubation attributable to type or level of fat or to vitamin E deficiency. Changes in fatty acid compositions of the major plasma lipid fractions from all diet treatments were typical of plasma that stands at room temperature before analysis (12). There was a slight but not significant tendency toward loss of triglyceride with no one fatty acid affected more than another. As shown in Table 5, 20:4 decreased \sim 25% in phospholipids ($P < 0.01$) and increased \sim 20% in cholesteryl esters ($P < 0.001$), a transfer that would be expected because of LCAT (lecithin:cholesterol acyltransferase) enzyme activity. However, there was no significant change in total 20:4.

TABLE 4. Alpha tocopherol content of pooled platelets and plasma from vitamin E-supplemented rats fed 5 and 15% corn oil or lard diets

	Alpha Tocopherol	
	Platelets	Plasma
	$\mu\text{g}/\text{mg protein}$	$\mu\text{g}/\text{ml}$
5% Corn Oil	1.6 \pm 0.13 ^a	10.6 \pm 0.8
15% Corn Oil	1.2 \pm 0.09	6.4 \pm 0.7
5% Lard	1.6 \pm 0.09	9.7 \pm 0.2
15% Lard	1.5 \pm 0.06	8.3 \pm 0.2

^a Each sample is a pooled composite from eight rats. Since incubation had no effect on tocopherol levels, values for the four incubation periods were combined. Mean \pm SD.

DISCUSSION

Plasma

Published reports on the effects of vitamin E deficiency on lipid levels of plasma are conflicting. Depending upon the age, sex, species, and diet of the experimental animals, plasma lipid levels that were either higher (13), lower (14, 15), or unchanged (13) by the deficiency have all been reported. The evidence presented here suggests that changes in lipid levels of plasma that are associated with vitamin E deficiency may depend not only on the absence of the vitamin but also on the type and amount of dietary fat, and might explain some of these inconsistencies. These data, combined with data from a previous study (5) in which plasma lipids increased as vitamin E was varied in increments from 5 to 500 ppm of the diets, indicate that vitamin E probably does not affect lipid metabolism solely as an antioxidant. This rationale is further supported by the increase with vitamin E deficiency in 18:0 in the plasma phospholipid fraction of lard-fed, but not corn oil-fed, animals.

Platelets

Under the conditions of this study, platelets from vitamin E-deficient rats did not have ultrastructural ab-

TABLE 5. Effects of incubation on 20:4 levels in cholesteryl esters and phospholipids in plasma from rats (n = 8)

	Time, hr		Δ
	0	6	
	<i>mg 20:4/100 ml plasma</i>		
Cholesteryl ester	26 \pm 1.4 (SE)	31 \pm 1.5	+5 ^a
Phospholipid	30 \pm 1.4	22 \pm 1.3	-5 ^b

^a $P < 0.01$, paired *t*-test.^b $P < 0.001$, paired *t*-test.

normalities of mitochondria that have been described for other tissues (1, 2, 3). Only after incubation at 37°C did similar abnormalities appear and only then in platelets from rats fed corn oil. Incubation did not affect the ultrastructure of platelets from rats fed vitamin E-supplemented diets with either corn oil or lard or vitamin E-deficient diets with lard. These findings disagree with those of Arimori and Sumitomo (16) who described structural destruction of mitochondria and other morphological abnormalities of fresh platelets from vitamin E-deficient rats. Possibly the disagreement could be due to differences in methods for harvesting and fixing platelets. That possibility is suggested by differences in morphology of control platelets from the two studies. Platelets shown by Arimori and Sumitomo (16) were rounded, rather than discoid, which suggests that they had aggregated during processing.

Even though mitochondrial membranes of many tissues are labilized by vitamin E deficiency, our data do not explain why platelets required incubation to demonstrate this instability. The need for a stress such as incubation might be explained, however, by the much lower respiration rate of mitochondria in platelets than of mitochondria in more active tissues such as liver. Mitochondria from vitamin E-deficient rat livers are unable to maintain normal respiratory activity in vitro (17). Possibly the relatively less active platelet mitochondria require stimulation before changes in mitochondrial ultrastructure become obvious.

The biochemical data suggest one possible explanation for the failure of platelets from vitamin E-deficient, lard-fed rats to exhibit ultrastructural defects even after incubation: the 18:2 level was less than half that of platelets from corn oil-fed rats. In platelets from lard-fed rats, a concomitant decrease in saturated fatty acids resulted in a relatively constant ratio of polyunsaturated to saturated fatty acids (P/S) but decreased the ratio of total fatty acid to protein by about 12%. The levels of unsaturated fatty acids in membranes affect both fluidity of the membrane and the activities of several membrane-bound enzymes. Possibly the higher level of 18:2 and of lipid-to-protein ratio in platelets from corn oil-fed rats resulted in an increased membrane fluidity and permeability that was modulated by vitamin E. That possibility is strengthened by the report that vitamin E may also affect membrane properties such as permeability and transport (18). Differences in 20:4 levels in both vitamin E-deficient platelets and plasma were negligible with respect to fat type. Therefore, the observed disparity in ability to maintain mitochondrial membrane integrity depending upon type of fat cannot be attributed to variations in amounts of this fatty acid. Differences in 18:2 levels in both platelets ($P < 0.001$) and plasma ($P < 0.01$) were significant

with respect to fat type. However, the lower level of 18:2 compared to 20:4 in both plasma and platelets and the apparent stability of the PUFA in both vitamin E-deficient plasma and platelets even after 6 hr of incubation would seem to rule out oxidative stress related to PUFA as an explanation. PUFA in vitamin E-deficient platelets may have been more stable to incubation than expected because of continued peroxidase and catalase enzyme activity in plasma. Small but biologically important oxidative losses of PUFA in platelets at the mitochondrial level were possible but would have been masked by larger levels of PUFA in the more abundant cellular membranes. Attempts to isolate mitochondria from these platelets in adequate yields for fatty acid analysis were unsuccessful.

The most striking observation, after incubation of PRP from control rats, was the stability of tocopherol in platelets regardless of type or level of dietary fat, especially since our experience has been that, when isolated platelets are stored in buffer overnight at 4°C, the tocopherol is unstable. Even after 6 hr of incubation of PRP no losses of α -tocopherol were detected in either plasma or platelets from control rats. This stability of a compound considered by some to function solely as an antioxidant also might be due to continued catalase and peroxidase activity, as well as to other endogenous antioxidant activity in plasma (19).

Red and white cells

Many pathological effects of vitamin E-deficiency on erythrocytes and erythropoiesis of several species have been reported (20–23). These effects differ among species but collectively include: 1) interference with red cell maturation as evidenced by the presence in bone marrow of multinucleated precursors (21, 24) and elevated RNA and DNA (25); 2) increased susceptibility of erythrocytes to lysis under in vitro oxidant stress (20, 26, 27); 3) either reticulocytosis, as in rats and premature human infants (27), or an acute anemia, as in monkeys (21, 26) and swine (22); and 4) a decreased red cell half-life that is attributed to an intrinsic defect in the erythrocytes (27–29). We found no reports, however, that described the large numbers of reticulocytes with identifiable but deformed mitochondria such as those shown in Fig. 9. The presence in reticulocytes of mitochondria with structural abnormalities is further evidence that in vitamin E deficiency red cell maturation is interfered with and that there is an intrinsic defect in the red cells beyond a susceptibility to hemolysis with oxidant stress. Possibly because of changes in membrane properties (18), reticulocytes are unable to extrude these organelles.

Reports of effects of vitamin E deficiency on the morphology of white blood cells are limited (21, 22, 24). In

both vitamin E-deficient monkeys and swine, neutrophil counts were elevated and in monkeys they were not hypersegmented. The electron micrograph in Fig. 8 shows swollen and deformed lymphocyte mitochondria. That was typical of all the mitochondria observed in vitamin E-deficient lymphocytes.

In our comparison of the various blood elements from vitamin E-deficient rats, fresh platelets differed from both white cells and reticulocytes by maintaining the structural integrity of their mitochondrial morphology. As suggested above, this difference may reflect the lower level of mitochondrial activity in platelets than in more active tissues such as reticulocytes and lymphocytes.

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