

Effect of Kidney Beans, Weight Gains and Unsaturated Fat on Incidence of Liver Necrosis in Rats (34597)

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Liver necrosis of rats fed a torula yeast diet can be prevented by the addition of vitamin E or selenium. Kidney beans have been reported to influence the vitamin E nutrition of sheep and chickens (1), but the effect of beans on liver necrosis in rats has not been reported. However, raw kidney beans reduce feed intake of rats (2), and Naftalin (3) reported that food restriction decreased the incidence of liver necrosis in rats fed a vitamin E deficient casein diet. The following trials were conducted to determine the effect of kidney beans or food restriction on the incidence of liver necrosis in rats fed torula yeast diets.

Materials and Methods. Four trials were conducted with Sprague-Dawley weanling rats individually housed in wire cages with water available *ad libitum*. The basal diet contained 30% torula yeast, 50.5% glucose monohydrate, 1% CaCO₃, 15% cod liver oil (CLO), 3% cellulose,¹ and 0.5% NaCl. The various treatments are shown in Table I. Beans and cod liver oil were added to the diets at the expense of glucose. The incidence of liver necrosis, survival time, weight gains, and feed intake were recorded.

Results. Trial 1. When intake of the basal diet was limited to produce weight gains similar to those of rats fed the raw bean diet, raw beans significantly influenced the incidence of liver necrosis (Table I). After 59 days only 1 of 16 rats fed a limited intake of the basal diet developed liver necrosis, compared to 11 of 16 rats fed the bean diet. However, 16 out of 16 rats fed the basal diet *ad libitum* developed liver necrosis.

Trial 2. Trial 2 was designed to study the effect of long term feeding of diets at restrict-

ed levels to maintain constant body weight. The results were consistent with those of Trial 1, in that animals fed diets containing raw beans had a higher incidence of liver necrosis and decreased survival time when compared to those fed diets with similar levels of vitamin E when equal gains were produced. The effect of rate of body weight gain is quite striking. In the previous trial rats fed the basal diet *ad libitum* lived an average of 27.6 days, whereas in Trial 2 when intake was limited the rats lived an average of 146.8 days. However, the diets containing raw beans were not efficiently utilized. Consequently, more raw bean diet was required to produce equal gains and this provided a slightly greater unsaturated fat intake. The beans contained 2% fat, which was 57% unsaturated C₁₈ fatty acid and 15% unsaturated C₁₆ fatty acids. Therefore, the majority of the fat intake was from the CLO. It was assumed the CLO contained 29% unsaturated C₁₈, 26% unsaturated C₂₀, 20% unsaturated C₁₆ and 10% unsaturated C₂₂ fatty acids (4). The level of unsaturated fat in the diet has been demonstrated to influence the status of vitamin E nutrition (5). Therefore, Trial 3 was conducted to determine if the decreased survival time was produced by the increased fat intake rather than by a specific factor(s) in the beans.

Trial 3. Eight rats were assigned to each of six treatments: (a) basal (10% CLO)—8 g/rat/day, (b) basal—4 g/rat/day, (c) 30% cod liver oil—4 g/rat/day, (d) basal + 30% raw beans—4 g/rat/day, (e) basal + 30% beans autoclaved at 1 kg/cm² for 30 min—4 g/rat/day, (f) basal + 30% raw beans + 0.3% sodium taurocholate—4 g/rat/day. Sodium taurocholate was added because in trials with chicks the addition of sodium tau-

¹ Solka flocc, Brown Paper Co., Durham, New Hampshire.

TABLE I. Effect of Raw Kidney Beans, Food Restriction and Cod Liver Oil (CLO) on Incidence of Liver Necrosis in Young Rats.

Treatment	Liver necrosis ^a	Survival time (days) ^b	Avg daily gain (g)	Avg daily feed intake (g)	Avg daily fat intake (g)
Trial 1 (59 days)					
Basal	16/16	27.6	2.30 (a) ^c	8.1 (a)	1.22 (c)
Basal (intake limited)	1/16	43.1	0.67 (b)	3.9 (c)	0.59 (a)
Basal + raw beans	11/16	33.1	0.52 (b)	5.0 (b)	0.77 (b)
Trial 2—Limited intake (215 days)					
Basal	10/10	146.8 (c)	-0.01	4.3 (a)	0.65 (a)
+ raw beans	10/10	64.4 (a)	-0.07	5.3 (b)	0.81 (b)
+ vit. E ^e (10 IU/kg)	5/10	140.6 (c)	0.02	4.4 (a)	0.72 (a,b)
+ raw beans + vit. E	10/10	85.6 (b)	-0.06	5.2 (b)	0.80 (b)
Trial 3 (97 days)					
Basal + 15% CLO—8 g/day	8/8	23.2 (a)	0.69 (b)	8.0 (b)	1.20 (b)
+ 15% CLO—4 g/day	8/8	70.8 (c)	-0.16 (a,b)	4.0 (a)	0.60 (a)
+ 30% CLO	8/8	42.0 (b)	-0.08 (b)	4.0 (a)	1.20 (b)
Basal + raw beans (15% CLO)	8/8	36.1 (b)	-0.05 (b)	4.0 (a)	0.62 (a)
+ autoclaved beans (15% CLO)	3/8	46.3 (b)	+0.23 (c)	4.0 (a)	0.62 (a)
+ raw beans + 0.3 taurocholate (15% CLO)	8/8	41.9 (b)	-0.30 (a)	4.0 (a)	0.62 (a)
Trial 4 (159 days)					
CLO (%)					
3 (<i>ad lib.</i>)	5/8	91.5 (d)	0.98 (b)	10.3 (c)	0.31 (a)
3 (limited)	4/8	125.0 (c)	0.40 (a)	6.5 (b)	0.20 (a)
12 (<i>ad lib.</i>)	6/9	52.5 (b)	1.00 (b)	9.9 (c)	1.19 (c)
12 (limited)	4/8	87.5 (d)	0.25 (a)	5.2 (a)	0.62 (b)
24 (<i>ad lib.</i>)	8/8	37.5 (a)	0.98 (b)	7.4 (b)	1.78 (e)
24 (limited)	8/8	72.6 (c)	0.41 (a)	4.4 (a)	1.06 (c)
3 + vit. E (100 IU/kg)	0/4	—	0.80 (b)	12.2 (d)	0.40 (a,b)
12 + vit. E (100 IU/kg)	0/4	—	0.81 (b)	11.4 (d)	1.37 (d)
24 + vit. E (100 IU/kg)	0/4	—	0.82 (b)	9.6 (c)	2.30 (f)

^a No. of animals with liver necrosis/no. of animals per treatment.

^b Survival time of rats diagnosed as having liver necrosis.

^c The fat was calculated to be composed of 85% unsaturated fatty acids.

^d Means with different letters in parentheses are significantly different ($p < .05$).

rocholate decreased the effect of raw beans on vitamin E requirement (6). Raw beans demonstrated an effect on liver necrosis even when unsaturated fat levels were equalized. Survival time was decreased from 70.8 to 36.1 days by the addition of raw beans, but autoclaving the beans eliminated the effect. No effect of sodium taurocholate was observed in this trial. The effect of the raw beans was similar to that of adding an additional 15% CLO to the basal diet.

It is well established that dietary linoleic acid increases the vitamin E requirement of

rats (5). However, Green *et al.* (7) reported that although nutritional liver necrosis apparently does not occur when the diet is completely free of fat, it is rapidly precipitated by amounts of dietary linoleic acid hardly greater than the rat's minimal requirement and the velocity of onset is unaffected by large increases in dietary unsaturated fat beyond this minimum. The results of Trial 3 did not appear to be consistent with this idea. The rats fed 4 g of diet containing 30% CLO had a mean survival time of 42.0 days compared to 70.8 days for rats fed 4 g of the

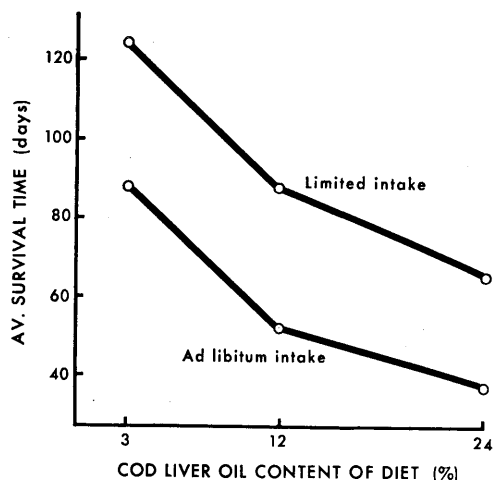


FIG. 1. Effect of cod liver oil and *ad libitum* feeding on average survival time of rats fed a vitamin E deficient diet.

diet containing 15% CLO. Therefore, Trial 4 was conducted to further study the effect of the level of cod liver oil on incidence of liver necrosis and survival time.

Trial 4. Increasing the level of CLO from 3 to 12 to 24% significantly decreased survival time (Fig. 1). Rats gaining less than 0.5 g per day fed 3, 12, or 24% CLO, survived 125.0, 87.5 and 72.6 days, respectively. Rats fed the diets *ad libitum* containing 3, 12, or 24% CLO survived 105.0, 52.5, and 37.5 days, respectively. Thus, the level of CLO influenced survival time under both *ad libitum* and limited feeding conditions. Limited feeding again resulted in an increased survival time (Fig. 1).

Discussion. The addition of raw beans to diets containing zero or marginal levels of vitamin E increased the incidence of liver necrosis or decreased survival time only when the rate of gain of the control rats was limited to that of rats fed raw beans. In contrast the addition of raw beans to chick diets induced nutritional muscular dystrophy in spite of reduced weight gains (1). Thus, the effect of beans on vitamin E requirements did not appear to be as great with rats as that previously reported with chicks.

Some of the effect of beans on vitamin E requirement in chicks has been attributed to decreased absorption of vitamin E (6, 8)

which could be alleviated by the addition of sodium taurocholate. However, decreased absorption of vitamin E does not appear to be the major factor with rats. Raw legume seeds exert a much greater influence on fat absorption in chicks than in rats (9, 10), and the addition of sodium taurocholate, which improves fat absorption in young chicks but not in rats fed raw legumes, did not alleviate the effect of rawbeans on the incidence of liver necrosis in rats.

The results demonstrate that when the incidence of liver necrosis or survival time is used as a criterion the rate of body weight gain must be considered. Increasing the level of cod liver oil, even when supplying more than the rat's minimum requirement for linoleic acid, hastened the onset of liver necrosis and decreased survival time.

Summary. Rats fed torula yeast diets containing 30% raw kidney beans had shorter survival times and a higher incidence of liver necrosis than rats fed comparable diets without beans when body weight gains were similar. Food restriction increased survival time and decreased the incidence of liver necrosis, whereas increasing the level of cod liver oil had the opposite effect. It was concluded that when incidence of liver necrosis or survival time is used as a criterion of vitamin E nutrition the rate of gain and unsaturated fat content of diet must be considered.

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