TABLE I.
Summary of Injection Experiments

	Material inj.	Amt	Vehicle	No. of rats	Tumors	%
I.	Sulfanilamide	30 mg	Lard	10	2	20
II.	Sulfapyridine	30 "	"	40 (18)*	3	7.5 (16⅔)*
III.	Dibenzanthracene	30 "	"	10	6	60
IV.	Lard	0.5 cc		30	0	0
V.	No injection			30	0	0

* Only 18 rats survived the entire experiment (see text).

fonamides carcinogenic for man? This question which occurred to the early workers remains as yet unanswered. Some analogy may perhaps be drawn from the experience with aniline dyes and carcinoma of the bladder in man. Hueper[4] has summarized the studies and experience in this field and it would seem that aniline dye workers develop bladder tumors in a statistically significant higher incidence than other people their age who are not exposed to these dyes. The average time between exposure and appearance of the tumor was 18 years. So many concomitant factors operate in such a period that conclusive evidence can be obtained only by statistical methods. However, if sulfonamides exert a similar influence and have the same latent period, it may be that an increase in the incidence of bladder tumors may be seen in or about 1955. Although highly speculative, this possibility deserves consideration because of obviously grave implications.

Conclusions. 1. Under the conditions listed it was observed that albino rats injected with sulfanilamide or sulfapyridine in lard developed sarcomata at the injection site. This incidence was less than the number of tumors occurring following the injection of dibenzanthracene under identical conditions. 2. The occurrence of tumors in Swiss albino mice following the injection of sulfanilamide in lard is reported in confirmation of previous work.

[4] Hueper, W. C., *J. Indust. Hyg.*, 1934, **16**, 255.

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Lipids of the Fasting Mouse. V. Liver Phospholipid, Lecithin and Cephalin.

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The young, adult mouse has been shown to mobilize available carcass fat in the first 2 days of a fasting period.[1] The liver total lipid increases during this period perhaps due to the failure of a phosphorylating mechanism to meet the emergency by converting the neutral fat to phospholipid prior to burning or transporting it. The incorporation of radio phosphorus (P32) into liver phospholipid metabolism is increased during fasting: this increase has been found in a series of experiments in which the total phospholipid, lecithin and cephalin fractions of the livers of mice fasted 5 days were studied.

Experimental. Three months old, male, albino mice were fasted as described previously.[1] Twenty-four hours before sacrifice, a small volume of a solution containing P32 as Na_2HPO_4 was injected intraperitoneally;

[1] Hodge, H. C., MacLachlan, P. L., Bloor, W. R., Stoneburg, C. S., Oleson, M. C., and Whitehead, R., *J. Biol. Chem.*, 1941, **139**, 897.

radioactivity determinations were made with a Bale dipping type counting tube.[2] At the time of sacrifice, the livers were removed quickly and analyzed by standard procedures.[3]

Liver Phospholipid. The amount of phospholipid in the liver decreased approximately 43% during fasting; the normal liver contained about 36 mg; the liver after 5 days fasting contained about 20 mg of phospholipid. This loss of phospholipid was almost exactly parallel to the loss of liver weight, so that the concentration of phospholipid in the liver remained approximately constant; this was in complete agreement with previously-reported data.[1,4] The normal percentage of phospholipid was about 3.24 (moist weight basis); with fasting, the values for succeeding days ran 2.97, 3.35, 3.20, 3.35 and 3.46. The tendency of the phospholipid percentage to go through a minimum on the first fasting day has been observed before; however, no reason for such a decrease has been surmised.

It is noteworthy that the % P32 in the liver phospholipid increased as the amount of phospholipid decreased during fasting. The magnitude of the increase in % P32 dose was unexpectedly large; a 2-fold increase was observed on the second day and a 4-fold increase on the fifth day (Table I).

Lecithin and Cephalin. Determined separately, both the lecithin and the cephalin fractions of the liver phospholipid showed a significant decrease on the first day of fasting and again on the fourth and fifth days. This conformed with the observed decrease in the total liver phospholipid; in fact, the combined values of lecithin and cephalin equalled over 90% of the total phospholipid values taken day by day.

Lecithin normally achieves higher values of % P32 dose per mg than does cephalin; this ratio is of the order of 2. Riley reported a similar finding in normal young rats; lecithin phosphorus had a specific activity of 825-966,

TABLE I.
Data on Liver Lipids and % P_{32} Dose in the Liver Phospholipid of Normal and Fasting Mice.*

	Total lipid		Phospholipid				Lecithin				Cephalin			
			Total		% P_{32} Dose per mg		Total		% P_{32} Dose per mg		Total		% P_{32} Dose per mg	
	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
Normal	77.0	14.2	35.6	5.7	.056	.012	19.8	4.2	.064	.014	15.0	1.5	.033	.010
Fasted 1 day	108.5	32.0	27.3	5.8	.088	.020	14.8	3.0	.108	.022	11.1	2.7	.059	.020
" 2 days	139.4	18.9	28.0	2.4	.113	.025	15.1	2.1	.156	.028	12.2	1.4	.066	.020
" 3 "	63.4	22.5	27.7	4.6	.136	.085	14.5	2.4	.097	.029	12.5	3.0	.050	.017
" 4 "	43.7	12.4	23.3	5.1	.086	.036	11.9	3.8	.114	.052	9.1	2.3	.053	.039
" 5 "	38.0	4.9	20.0	1.7	.188	.070	10.6	1.0	.215	.121	8.3	1.1	.138	.055

* The numbers of mice in each group are given in Table II of "Lipids of the Fasting Mouse. IV. Liver Total Lipid Content." PROC. SOC. EXP. BIOL. AND MED., 1948, **67**, 137-139.

[2] Bale, W. F., Haven, F. L., and LeFevre, M., *Rev. Scient. Instruments*, 1939, **10**, 193.

[3] Welch, E. A., *J. Biol. Chem.*, 1945, **161**, 65.

[4] MacLachlan, P. L., Hodge, H. C., Bloor, W. R., Welch, E. A., Truax, F. L., and Taylor, J. B., *J. Biol. Chem.*, 1942, **143**, 473.

whereas cephalin phosphorus from the same livers averaged 363-735.[5]

The changes in the % P32 dose per mg of liver lecithin and cephalin paralleled each other. The values increased during the first 2 days of fasting, fell on the third day, and increased again markedly on the fifth day. With the exception of the value obtained on the third day of fasting, the % P32 dose per mg total liver phospholipid represented approximately the average of the values for the % P32 dose per mg lecithin and cephalin.

During the first 2 days of fasting when depot fat was being rapidly mobilized, the increased phospholipid metabolism in the liver was evidenced by an increase in the % P32 dose per mg liver phospholipid. As previously observed[6] the liver total lipid reached a maximum on the second day of fasting. In some respects a similar condition obtains on a low choline diet, *viz.*, the liver neutral fat is increased and the lecithin level falls below normal; the administration of choline reverses this relationship.[7]

The accumulation of fat on the second day of fasting might account for the decrease in the % P32 dose in the liver lecithin and cephalin on the third day. Later, as the fat moved out of the liver (third to fifth day of fasting) the phospholipid metabolism presumably increased and, by the fifth day, the value for the % P32 dose in the liver phospholipids attained very high values (4 times normal). In the light of recent transmethylation studies it seems reasonable to suppose that more of the choline precursors were made available when protein metabolism was accelerated during the last days of fasting.

Summary. Young, adult, male albino mice lost almost half the liver total phospholipid during a 5-day fast. The liver weight also decreased in such a fashion that the phospholipid concentration remained approximately constant. The amounts of lecithin and cephalin decreased in proportion. The % P32 dose per mg of phospholipid increased markedly during fasting and, although the % P32 dose per mg lecithin is normally higher than that of cephalin, during fasting the values increased markedly for both fractions.

5 Riley, R. F., *J. Biol. Chem.*, 1944, **153**, 535.

6 Hodge, H. C., MacLachlan, P. L., Bloor, W. R., Welch, E., Kornberg, S. L., and Falkenheim, M., PROC. SOC. EXP. BIOL. AND MED., 1948, **67**, 137.

7 Fishman, W. H., and Artom, C., *Federation Proc.*, 1945, **4**, 90.

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Effect of Asphyxia on Blood Coagulation in Dogs.

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Several manuals on postmortem examinations contain statements to the effect that a fluid state of the blood in a body is suggestive of oxygen deficiency at the time of death. For example, in describing the conditions found after death from asphyxia, Ross[1] stated "The blood is fluid and very dark in color owing to its being chemically almost completely reduced." Under the caption "Drowning," the same author said "The blood on the right side is dark in color and fluid (asphyxia)." In the section on asphyxia, Warthin's "Practical Pathology"[2] contains the following: "The lips, skin of the face and neck, and the finger nails may be deeply cyanotic; the blood is dark and fluid."

This experimental study was undertaken to determine the effect of severe asphyxia on the clotting mechanism as judged by *in vitro* tests on blood withdrawn from living animals

1 Ross, J. M., *Post-mortem Appearances*, New York and London, Oxford Univ. Press, 1939.

2 Warthin, A. S., *Practical Pathology*, Ann Arbor, George Wehr, 1928.