groups receiving x-ray treatment an initial increase is tumor size, attributed to tissue edema, was noted. This was followed by regression and ultimate tumor regrowth. At time of death all animals in each group exhibited huge tumors. These tumors contributed to approximately one-third of animals' total body mass (Fig. 2).

Control groups gained weight more rapidly than the group receiving x-ray treatment. In groups receiving irradiation, initial slight weight gain might be attributed to tissue edema. This was followed by weight loss which paralleled tumor regression. Weight gain accompanied tumor growth in all groups but several days prior to death weight loss was observed. The group receiving T3 plus x-ray averaged 5 g less in weight than the other groups (Fig. 3).

Two weeks following irradiation maximum tumor regression and skin reaction were noted. In the group receiving T3 plus x-ray, moist epidermitis and superficial ulceration were observed, while in the group which received x-ray alone epilation and a brisk erythema were most commonly seen. In several animals receiving both T3 and x-ray limb amputation occurred subsequent to irradiation.

Conclusions. 1. Growth rate of transplanted mammary tumor in ZxC57 black F_1 hybrid mice treated with L-triiodothyronine was increased. 2. Average life span of tumor-bearing mice receiving L-triiodothyronine was significantly shortened. 3. In mice receiving L-triiodothyronine and x-ray, increased skin reaction was noted. 4. There was no significant difference between average life span of tumor-bearing groups receiving combined irradiation plus triiodothyronine and the group which received local irradiation only.

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Effect of Single Dose of DL-Serine on Lipide Phosphorylation in Kidney of Rats.* (25811)

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Interference of plasma lipide metabolism in degenerative kidney lesions has been reported (1). The present study is, to our knowledge, the first to investigate metabolism and distribution by column chromatography of phospholipides in experimental renal necrosis produced by administration of a single dose of DL-Serine.

Methods. Experimental necrosis was produced by administration of a single dose of DL-Serine by the method of Fishman and Artom(2). Male albino rats of Sprague-Dawley strain weighing 80-100 g were fed 10% casein diet(3) for 10 days supplemented daily with a B vitamin solution(4) not containing pyridoxine. Control animals received a similar diet daily supplemented with Vit. B solution containing pyridoxine. After being on the diet for 10 days, the rats were stomach-tubed with single dose of 100 mg of DL-Serine; control animals received water. Forty-

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TABLE I. Effect of Single Dose of DL-Serine on Lipide Phosphorylation in Kidney of Rats.

		Phospholipide phosphorus	
	No. of rats	mg/g wet wt	Relative specific activity
Control	20	.74 ± .15	.094 ± .009
Necrosis	27	.59 ± .12*	.309 ± .079*

Test of significance was applied to difference between mean of control and experimental values. Probability for chance occurrence of this difference was * <0.01.

eight hours later the rats were killed by decapitation. All animals were injected intraperitoneally with 1 ml of physiological saline containing 4 μc of P^{32} as NaH_2PO_4 6 hours before sacrifice. Kidneys were removed, rapidly weighed, a small sample taken for histological examination, and the remainder divided into 2 fractions. From one fraction, acid soluble phosphorus was removed by extracting with 10% TCA containing 0.4 M $MgCl_2$(5), and radioactivity and total P(6) were determined. The other fraction was covered with alcohol, and lipides extracted with alcohol-ether and purified with chloroform(7). Radioactivity(7) and phosphorus(6) were determined on aliquots of the chloroform solution. As a measure of phospholipide turnover, relative specific activities(8) were calculated. Additional groups of rats (240-400 g) were fed similar diet for 18 days, then they received, by stomach-tube, either a single dose of 100 mg of DL-Serine (experimental) or water (controls). Thirty hours later animals were killed by decapitation. Six hours before sacrificing 20 μc of P^{32} was administered intraperitoneally. Kidneys were removed, rapidly weighed and a small section taken for histological examination. Kidneys were pooled from 4 rats for each experiment except for one control group which contained 2 rats. The kidneys were homogenized in the cold for 2 minutes in 95% ethanol in Potter-Elvehjem homogenizer then allowed to stand in the dark at room temperature 4 hours. Lipides were extracted, chromatographed on silicic acid columns and eluted with chloroform-methanol by the method of Hanahan(9) using a Gilson Medical Electronics fraction collector. Radioactivity(7) and phosphorus(6) were determined on aliquots of the chloroform-methanol fractions. Data are reported as means ± standard dev.

Results. Histological examination of all kidneys showed marked degenerative lesions as reported by Morehead *et al.*(10). The effect of a single dose of DL-Serine on lipide phosphorylation in kidney of rats is shown in Tables I and II. To evaluate the significance of results, the *t* test of significance(11) was applied to the difference between mean of controls receiving saline and experimental values. The data in Table I show that DL-Serine necrosis produces a statistically significant decrease in phospholipide content. There is a statistically significant increase in lipide phosphorylation in the necrotic kidneys. Phospholipides of the kidney were separated by column chromatography to elucidate which class of phospholipides is involved in production of the necrosis. Hanahan's(9) method of column chromatographic separation of the classes of phospholipides fractionates them into: lecithins, phosphoinositides and sphingomyelins. However, by this method phospha-

TABLE II. Kidney Phospholipide Composition and Specific Activities Following Administration of Single Dose of DL-Serine.

			% total lipide phosphorus			
	No. of exp.*	mg/g wet wt	Phosphatidyl-ethanolamine-serine	Phospha-tidyl-inositol	Lecithin	Sphingomyelin
Control	4 (14)	.68 ± .05	34.9 ± 3.0	12.9 ± 2.7	46.8 ± 4.4	3.1 ± 1.9
Necrosis	3 (12)	.33 ± .20‡	57.6 ± 4.5†	22.2 ± 5.0†	18.4 ± 6.1†	2.2 ± .2
			Specific activities of lipide phosphorus			
Control	4 (14)	18.9 ± 2.0	4.0 ± 1.5	7.3 ± 1.6	8.7 ± 2.1	5.1 ± 2.3
Necrosis	3 (12)	28.5 ± 4.9 †	10.0 ± 4.8‡	12.9 ± 6.4	11.0 ± 1.1	11.6 ± 3.0†

* Values in parentheses are total No. of rats/series.

Test of significance was applied to difference between mean of control and experimental values. Probability for chance occurrence of this difference was † <0.01; ‡ <0.05.

tidyl serine is mixed with phosphatidyl-ethanolamine. There is a significant increase in % of total lipide phosphorus in the necrotic kidney in phosphatidylethanolamine-serine and phosphatidylinositol fractions (Table II). However, a decrease occurs in the lecithin fraction. The increase in lipide phosphorylation in the necrotic kidney apparently involves only phosphatidylethanolamine-serine and sphingomyelin fractions. These results demonstrate that phospholipides of the kidney are involved in production of kidney necrosis from administration of a single dose of DL-Serine. These phospholipide changes may be a reflection of the degenerative process since these lipides make up 16-32% dry weight of the cellular mitochondria, microsomes and nuclei (12).

Summary. 1) The effects of a single dose of DL-Serine on lipide phosphorylation in kidney were studied in the rat. 2) A statistically significant decrease occurred in phospholipide phosphorus of the necrotic kidney as compared to controls. However, there was a 3-fold increase in lipide phosphorylation as shown in the relative specific activities values. 3) A statistically significant increase occurred in % of total lipide phosphorus in phosphatidylethanolamine-serine and phosphatidylinositol fraction, in the necrotic kidney as compared to controls. However, a decrease occurred in the lecithin fraction. This increase in lipide phosphorylation in the necrotic kidney apparently involved only the phosphatidylethanolamine-serine and sphingomyelin fractions.

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Isolation of α- and β-Melanocyte Stimulating Hormones (MSH) on a Preparative Scale.* (25812)

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The need for large amounts of α- and β-MSH for investigation of their physiological significance in mammals and particularly for the study of effects of β-MSH on the central nervous system(1) and for further studies on biosynthesis of ACTH from α-MSH(2), prompted us to develop still other methods for isolation‡ of these 2 substances on a preparative scale.

Methods and materials. Starting materials for purification were an MSH concentrate

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‡ For review on isolation and structure of α- and β-MSH, see Harris(3).