and they developed so severe a weakness that they were unable to walk. They became stuporous and remained quiet for some hours.

Di-n-butyl Phthalate. The L.D. 50 was found to be 5.5 ml per kg body weight (Table 1D).

Dioctanol 2 Phthalate. It was found by statistical analysis that the lethal dose for the average mouse is 46 ml of dioctanol 2 phthalate per kg body weight (Table 1E). The mice lived for several hours before any died.

Summary. The L.D. 50 doses for the mice used are as follows: Phthalic acid, 0.011 g; sodium phthalate, 0.042 g; dimethyl phthalate, 0.05 ml; dibutyl phthalate, 0.11 ml; and dioctanol 2 phthalate, 0.92 ml. Compared to phthalic acid, the L.D. 50s of the compounds are as follows: Phthalic acid, 1, sodium phthalate, 4; dimethyl phthalate, 5; dibutyl phthalate, 10; dioctanol 2 phthalate, 84.

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Bomskov Reports on Thymus Mediation of Pituitary Function.

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A series of papers by Bomskov and coworkers[1-5] in Freiburg reports the preparation and extensive investigation of an ether-soluble extract of the thymus. This extract was said to be "diabetogenic" (in rats, guinea pigs, pigeons) in that it showed marked glycogenolytic and hyperglycemic actions. These investigators claimed that a diabetogenic fraction of the anterior pituitary gland failed to lower the liver glycogen in rats whose thymus tissue had been destroyed by Roentgen irradiation.[1] They, therefore, postulated, and later claim to have shown, that the diabetogenic pituitary fraction is thymotrophic and exerts its recognized physiologic actions through stimulating the production of a hormone in the thymus. As the result of other work Bomskov and coworkers[2, 3] indicate that their diabetogenic-thymotrophic fraction from the pituitary gland is identical with growth hormone, and that physiologic effects of the latter

1 Bomskov, C., and Sladovic, L., *Deutsch. Med. Wochschr.*, 1940, **66**, 589.

2 Bomskov, C., and Hölscher, B., *Z. Klin. Med.*, 1940, **137**, 745.

3 Bomskov, C., and Sladovic, L., *Pfluger's Arch.*, 1940, **243**, 611.

4 Bomskov, C., and Brachat, F., *Endokrinologie*, 1940, **23**, 145.

5 Bomskov, C., and Karl-Heinz, K., *Pfluger's Arch.*, 1940, **243**, 623.

pituitary substance(s) are thus also mediated by the thymus hormone. In still other studies[4, 5] it is reported that a lymphocytosis and leucocytosis regularly follow the administration of thymus hormone to rats. On the basis of this finding, and of certain histological observations on thymus tissue, they develop the theory that thymus hormone is transferred to leucocytes during their passage through the thymus; thereafter it is transported exclusively within the leucocytes to points of utilization in the body. A number of relations of the thymus hormone to other endocrine secretions, notably thyroxin and sex hormones, have been stated in some detail but we have made no attempt to repeat those particular studies.

The international situation has made it impossible to secure all of the publications of the Freiburg investigators, but their strong claims to a resolution of problems in which this laboratory has long been interested have led us to this unsuccessful effort to confirm their results.

Preparation of Thymus Extract. Thymus glands of young calves were obtained from a slaughter house where they were frozen immediately after their removal from the animal and were delivered to us within 24 hours. The glands were finely ground while still partially frozen, and the tissue was then extracted in the cold with 5 times its volume of acetone for three 8-hour periods. The acetone was removed under reduced pressure and the residue taken up in ethyl ether. When the ether was distilled off *in vacuo* there remained a relatively large quantity of viscid yellow oil. This product, here called "thymus oil," was subsequently kept at low temperatures; for injection it was either undiluted or diluted with an equal quantity of propylene glycol. Three different extracts were prepared and tested. None of the publications available to us contained a detailed description of the method by which the thymus hormone was extracted, but the apparent simplicity of its preparation and available descriptions of its properties make it wholly probable that our "thymus oil" is identical with or quite similar to the product most commonly used by the Freiburg group.

Results and Discussion. Our first effort to discover physiologic activity in the thymus preparation was directed to total white cell counts in rats following subcutaneous administration of 0.2-0.5 cc of the thymus oil. We here used 2 groups of 6 adult male treated rats together with an equal number of control rats injected with sesame oil. As is to be expected in rats there was considerable variation in the white cell counts from time to time; but there was no indication of a leucocytosis, even within the physiologic range, following ad-

ministration of the thymus oil. The variations in leucocyte count observed by Bomskov[1] following injection of his thymus hormone are well within the normal range for rats. Significant differences within this range are exceedingly difficult to establish, and this would require a much larger number of observations than appears in the published data. It is also notable that the local reaction to the oil, often marked in our animals, is a factor which requires careful control.

In an attempt to observe a "diabetogenic" action of our thymus

TABLE I.
Blood Sugar and Liver Glycogen in Rats and Pigeons Injected Once with Thymus Oil. Time After Injection Usually Is Same as the Period of Fasting.

Test or Control	Dosage, cc	No. of animals	Blood sugar, mg% At hrs			Liver glycogen, mg% At hrs				
			12	17	24	5	12	17	24	36
Male rats aged 3-5 months.										
Thymus oil	.4*	13							.56	
Sesame "	"	9							.59	
Thymus "	.2	16				4.9	0.9		.93	.50
Sesame "	"	16				4.0	2.1		.90	.42
Thymus "	"	4					1.3			
Sesame "	"	4					1.1			
Thymus "	"	4					0.8			
Sesame "	"	4					1.6			
Thymus "	"	5					2.2			
Sesame "	"	5					2.3			
Thymus "	"	4					1.7			
Sesame "	"	4					2.7			
Thymus "	"	3					2.4			
Sesame "	"	4					2.9			
Prop. glycol	"	3					2.6			
Immature Carneau pigeons.										
Thymus oil	.2	9			203				.75	
Thymus "	"†	3			189				.85	
Sesame "	"	10			200				.71	
Thymus "	.4	3	210				4.4			
Sesame "	"	3	208				4.2			
Thymus "	"	3		205				2.8		
Sesame "	"	3		196				2.5		
Thymus "	"	3		194				1.0		
Prop. glycol	"	3		209				0.8		

*Most rats injected twice—at 17 and 10 hours before test; fasted 24 hours.

†Daily injections of 0.2 cc for 4 days preceding test; 3 of control here also injected 4 days.

preparations we administered varying doses at various stages of fasting. From 108 rats and 40 pigeons, treated and control, we obtained the liver glycogen and blood sugar values shown in Table I. In small groups of rats the averages for liver glycogen obtained after 12-hour fasts are more often below than above their control, but the difference seems hardly significant and the values obtained at other stages of fasting are mostly in the opposite direction, though likewise without significance. In pigeons measurable effects of thymus oil were obtained neither on liver glycogen nor on blood sugar.

In additional tests thymus oil did not produce ketonemia (24-hr fast, 10 hr after injection) in either rats or pigeons. From these and other results noted above it is clear that we have been unable to obtain a "diabetogenic" action of lipoid extracts of calf thymus.

With a view to a partial test of possible growth effects, or of influence on various organ weights, we administered moderate or large doses of the thymus oil to 21-day rats, 2-day chicks and to immature pigeons during 3-10 days. These tests were essentially negative; in Moon tests on rats of 21 days the suprarenal weights were unchanged and thymus weights (9 tests) were decreased by nearly 50%. Many years ago Romeis[6] made an extensive study of an acetone extract of the thymus and reported that it has growth-inhibiting properties.

Conclusions. With 3 different preparations of an ether-soluble extract of calf thymus unsuccessful attempts were made to confirm certain results reported by Bomskov and coworkers. No significant leucocytosis was obtained in rats. Short-term tests showed no appreciable effects on bodily growth in young rats, chicks and pigeons. Neither glycogenolytic, glycemic nor ketogenic activity was observed in rather extensive tests on rats and pigeons.

[6] Romeis, B., *Z. Exp. Med.*, 1918, **6**, 101.