plain this result would assume that there appears in the plasma of the malarious chickens an increased concentration of some nonspecific reaction product which has growth-inhibiting properties. An increase in lipoproteins has in fact been demonstrated in birds undergoing malarial infections(6). This may provide a clue as to the type of substance to be looked for.

It is of interest to note the recent observation(7) that the life of leukemic mice was prolonged when they were infected with *Plasmodium berghei,* a malarial parasite of certain rodents.

Summary. Two-week-old chickens were inoculated in the breast muscle with a suspension of chicken tumor I tissue. They were subsequently left untreated or given a series of intravenous injections of either normal chicken plasma or plasma from corresponding chickens which were near the peak of an acute infection with *Plasmodium lophurae.* On the day after the last treatment the tumors which had developed were weighed. In a series of 5 experiments, the mean tumor weight of the chickens left untreated or treated with normal plasma was 1.5 to 1.8 times greater than that of the chickens treated with malarious plasma.

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Serum Complement Levels in Dogs Undergoing Kidney Homotransplantation.* (20357)

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Success with corneal(1), bone(2), and aortic(3) homotransplantation has further stimulated interest in the possibility of skin and renal transplantation in human medicine. Advances in surgical technics which have made possible autotransplantation of kidneys in experimental animals(4) as well as temporary homotransplantation in man(5) have focused new attention on the phenomenon of individual tissue specificity(6).

The mysterious failure of skin and kidney homotransplants days to weeks following an initial period of functional survival is suggestive of an immune response to an antigenic stimulus. Coincident with the development of experimental renal lesions induced by large doses of serum globulin a temporary depression of serum complement levels has been observed(7). Similar depressions of complement have also been noted in nephrotoxic nephritis(8) and human glomerulonephritis (9,10).† In the present study serial serum complement levels were determined in a group of dogs undergoing kidney transplantation. A comparison of these findings with patterns

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† Suppression of circulating complement is not unique for renal disease. Studies of C. A. Ianewav and co-workers (12) have shown this phenomenon to be a feature of "serum sickness" reactions produced by relatively pure antigens (albumin) which involve the arterioles. The most marked and sustained serum complement suppression observed in humans occurs in disseminated *lupus erythematosis* (11) with or without obvious renal disease.

seen in spontaneous and experimental renal disease is of interest for the understanding of the biology of organ homotransplantation.

Materials and methods. Homotransplantation. Healthy mongrel dogs weighing 10 to 14 kg were used as donors and recipients for kidney transplantation. Under nembutal anesthesia one kidney was removed from the prospective donor with preservation of adequate length of renal artery and of renal vein for anastomosis. In some earlier experiments, arterial perfusion of the donor kidney *in vivo* was done, using physiologic saline, with or without penicillin (5 cc with 5000 U/cc) and heparin (5 cc with 10 mg/cc). Since this procedure had no noticeable beneficial effect on the results, it was discontinued in later experiments. End-to-end suture anastomoses of the renal vessels to the iliac vessels were carried out in all cases reported (except Dog 47-52 where the neck site was used), and the ureter brought out to the skin. This location in the iliac fossa made possible gravity drainage of urine, a well anchored and protected graft, and helped prevent ureteral angulation. At the conclusion of the operation, the tip of the ureter bled freely, and clear urine appeared in a steady drip. *Complement determination.* Freshly drawn venous blood was allowed to clot at room temperature and the serum decanted within 90 minutes for storage at 8°C. The majority of determinations were carried out on the day blood was collected. Specimens examined at a later date were stored frozen at –20°C. No appreciable change in complement level was apparent under these conditions. Groups of sera containing specimens from normal controls were titered concurrently. A preliminary group of the complement titrations (Table I) was done by the 50% end-point method of Kabat and Mayer(13) modified by using a 5% instead of a 10% sheep cell suspension in the initial part of the procedure and by reducing the final volume of the test to 4 ml (2 ml sensitized cell suspension, 2 ml serum dilution) to facilitate hemoglobin determinations in the Klett-Summerson Colorimeter (5500 Å filter). Analysis of data on these 7 animals indicated that relatively high and variable readings resulted from the estimations in the upper range

TABLE I. Series I. Complement Levels in Dogs Undergoing Renal Homotransplantation.

Days	1	1		5			10				15			20			25			30
Dog 11	36		51	60		58		27	47	47			34	80*		51			76			74
14	41			41		38	94	64	68	60			84			*		D				
35	51		51		60	65	57	61	55	60	57	74	73	68	105	105	92	104	90		60		
47	59		67	80	94	90	82	100	88	120	140	140	110	110		120		96		80			
75	61			60				110	115	48		53		90						130	106		*	
92	66		48	29		86			130		160				145					50		105	115	118
98	55			70			90									62							
Avg	53	↑	55	58	64	88	66	90	81	67	93	83	74	78	101	113	84	80					
		Operation																					

* Graft removed. D = Animal died.

TABLE II. Series II. Complement Levels in Dogs Undergoing Renal Transplantation.

	Days	3	2	1	1		5			10			15				20				25			Transplant	Avg
Dog 130		19	18	18	24	20	22	24		30	22	22	21	21	18	16		17	16	15	16	17	20	20	17	14	25	Auto-	
141				10	11	20	21	36	31	25	30	26	26	14	23	27	27	20	23	28	28	23	23	21				"	
131	AM		25	18		22	22	15	23	23	23	24	22	23	22		24	24	25	26	19		28	28	19*	22	36	30	20	27	22	Homo-	24
	PM		20		12	14	16	28	23	23	31	26	26	27	26	26	29	22	30	22		22									"	23
137	AM	12	14	14	14	20	22	26	31	24	19	19	23		28	31	25	26	26	22	18	19	22	12*23						"	21
	PM			17	12	20	24	36		24	23	23	24		31	23	25													"	24
	Avg		14	19	15	14	19	21	26	27	25	25	23	24	21	25	25	24	23	24	23	20	20	23	20	21					
	Avg auto-				14																										
	Avg homo-		22	18																											

* Graft removed.

of optical density. Taken as a group, however, readings occur in a normal distribution curve, following roughly the pattern obtained by the unmodified 50% end-point complement method used thereafter (Table II). The main experiment of this report was carried out on 4 dogs in which 2 homotransplants were compared with 2 autotransplants. The homotransplanted animals received a third kidney in the groin, and the autotransplanted animals had one of their own kidneys transplanted to the same location. Complement titers were determined in these 4 animals by the method of Kabat and Mayer without modification.

Results. All homotransplants stopped producing urine within 2 to 6 days. As urine flow ceased, the ureters retracted, blackened and finally disappeared into the wound. Coincidentally the transplant enlarged, forming a firm, palpable abdominal mass. Less commonly transplants disintegrated, leaving only remnants of necrotic tissue. The prompt establishment of urine flow and evident effective functional survival of the two autotransplants was in marked contrast to the course of events in the homotransplanted kidneys. Ten values on 8 normal control dogs,‡ and single preoperative values on the 7 animals operated upon, were employed to find the complement level of normal dogs (Table I). By the modified method the mean complement level was 53 units with a σ of 18 units. When the daily or the grouped postoperative values for the first 16 days are compared with the 17 normal values, no statistically significant change is noted.

In the second series of animals studied by the unmodified complement titration procedure, uniformly lower and more closely grouped values were obtained (Table II). The course of events for a typical experiment is given in Fig. 1. Normal dog complement for this method was found from 14 preoperative levels on 4 animals and single values on 2 other normal dogs.§ The mean for these 16 determinations was 23 with a σ of 4.5 units of complement. There is no significant differ-

‡ Values for normal dogs not operated upon are omitted in the tables.

§ Values for normal dogs not operated upon are omitted in the tables.

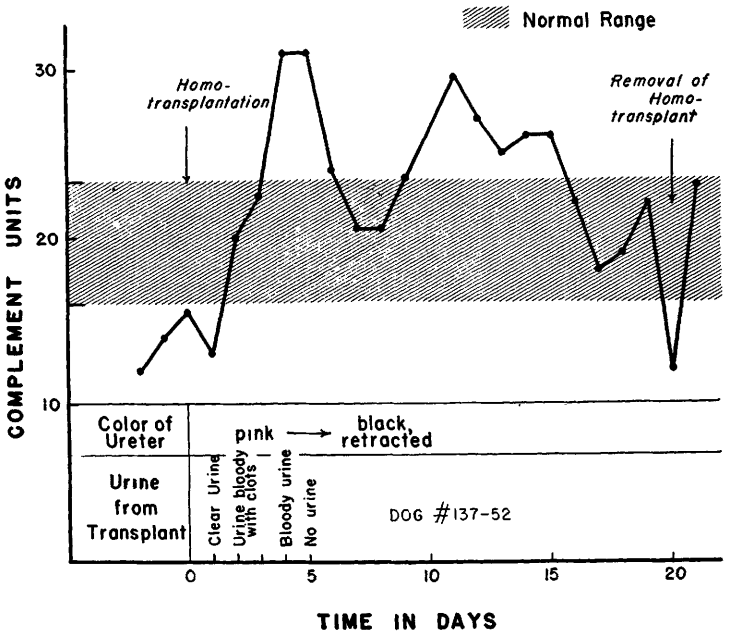

FIG. 1. Complement levels in a dog undergoing homotransplantation of a kidney into the iliac fossa.

ence between these 16 normal values and 30 postoperative values, days 1 through 16, for autotransplants ($t = 0.06$). On the other hand, a significant rise in complement followed homotransplantation (16 normal values compared to 56 values from days 1 through 16 give a t of 12.8 and P of >0.01). There is a significant difference between 16 normal values and values for many of the days after homotransplantation. The first day after operation there was a significant fall in complement ($t = 9.2$; $P = >0.01$). For the next 3 postoperative days complement lay within the normal range ($t = 1.8$; P 0.1. $t = 1.0$. $t = 0.8$). For days 5 through 16 complement was elevated significantly ($t < 4.0$; $P > 0.01$).

Discussion. In 9 dogs which were subjected to a kidney homotransplantation a uniform failure of the transplant was seen. In contrast to the 2 successful autotransplantations which functioned continuously following surgery, the urine flow of all homologous transplants ceased within 5 days. Various evidences of necrosis and infection promptly followed. Although there was a transient fall of complement on the first postoperative day in 2 of the 9 animals undergoing homotransplantation, the pattern of complement depression seen in human and experimental glomerulonephritis was not observed. This initial change could be due to many nonspecific causes. In these same animals a significant rise in complement in the late postoperative period was noted. During this later period the graft was undergoing dissolution. Similar complement rises associated with inflammatory processes have been seen by other observers(9). There was no change in complement postoperatively in 2 control dogs subject to autotransplantation. Neither was there a significant postoperative change in complement in 7 dogs undergoing renal homotransplantation who were studied with somewhat different method of complement titration. These patterns of complement levels after renal homotransplantation are quite unlike the "serum sickness" type of response in which a fall in complement ordinarily occurs a week or two after exposure to an antigen.

Summary. Homotransplantation of kidneys in normal dogs was followed by prompt renal shutdown and progressive failure of the graft. Frequent complement determinations were performed during the 3-week postoperative period. Unlike the situation in human glome-

rulonephritis and experimental nephritis a fall in serum complement 1 to 2 weeks later was not observed. Instead there was sometimes seen a significant rise in serum complement following homotransplantation.

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Addendum

After this work was submitted for publication we learned that a similar study of serum complement levels in dogs undergoing kidney homotransplantation had been presented by M. Simonsen, Biological Incompatibility in Kidney Transplantation in Dogs, (E. Munksgaard. Copenhagen 1953).

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Phase Contrast Microscope Study of Oral Epithelium of Normal and Vitamin A-Deficient Rats.* (20358)

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A recent investigation into the structure of the epithelial attachment to the tooth revealed that in fresh, undenatured oral epithelium prominent tonofibrils secure cohesion of the epithelial cells among themselves and with neighboring tissues such as the lamina propria and the surface of the tooth(1). These observations, made in man and monkey with the phase contrast microscope, suggested a corollary study of the behavior of the tonofibrils in oral epithelium of normal and vit. A deficient rats.

Experimental. Twenty male rats of the Long-Evans strain were fed a vit. A-deficient diet† beginning the week before weaning at 14 days of age. Their weight curves showed a plateau between the ages of 50 to 60 days. A maintenance dose of 3 μg of vit. A alcohol in oil was administered *per os* when they showed signs of extreme exhaustion. Four rats which survived the deficient diet for 86 days (100

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† The vit. A deficient diet had the following percentage composition: vit.-free casein 24, sucrose 62, salts 4, cottonseed oil, vit. D, mixed tocopherol 10. The following vit. were added per 10 g diet: vit. D 10 USP units, tocopherol (alpha and delta) 3 mg, menadione 50 μg, thiamine 20 μg, pyridoxine 20 μg, riboflavin 50 μg, p-aminobenzoic acid 50 μg, niacin amide 100 μg, inositol 2 mg, choline citrate 10 mg, biotin 6 μg, pteroylglutamic acid 20 μg, pantothenic acid 350 μg, B_{12} 0.2 μg.