proximately 5 days after the first inflammation.

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Effect of Anti-Rheumatic Drugs on Synovial Membrane Permeability.* (19275)

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Alterations in synovial membrane permeability have been described following the administration of various drugs. Seifter and co-workers(1) found that in rabbits hyaluronidase and desoxycorticosterone (DOCA) produced an increase in the rate of absorption and excretion of phenolsulfonphthalein (PSP) injected into the joint, and that Cortisone, ACTH, and an adrenal cortical extract inhibited absorption and excretion of the dye. They concluded from these studies that one of the controlling factors in membrane permeability is an antagonism between DOCA and Cortisone-like substances. In a later study(2), these investigators reporting on the effects of many steroid substances on synovial membrane permeability suggested that compounds impairing this permeability should produce an anti-rheumatic effect in patients.

Our study was designed to further evaluate the relationship between synovial permeability and the anti-rheumatic effect of cortisone and ACTH in animals and man. We undertook to explore synovial permeability by studying the rate of absorption and excretion of PSP following its injection into a joint. Since Seifter's studies had described its effective use, this appeared to be a convenient and accurate index. However, our results lead us to question the validity of this procedure as an accurate measure of synovial permeability.

Methods and materials. Studies on rabbits were conducted in male animals weighing 3 to 5 kg. Each animal was anesthetized with sodium amytal 100 mg/kg intramuscularly, and hydration was attained by the administration of 25 ml per kg of water by stomach tube. An indwelling catheter was secured in the bladder for collection of urine. PSP was injected into the ankle joint using 0.25 mg per kg, and after the injection, collections of urine were made by irrigation and manual expression of the bladder every 15 minutes for 2 hours. Urine samples were adjusted to a pH of 10.5 and diluted to a standard volume, filtered, and the optical density measured on a Coleman Junior Spectrophotometer at a wave length of 500 millimicrons. Excretion of the dye was calculated for each fifteen minute period as a percentage of the total dye injected. Cortisone was given to 7 rabbits for 11 courses in the doses shown in Fig. 1.

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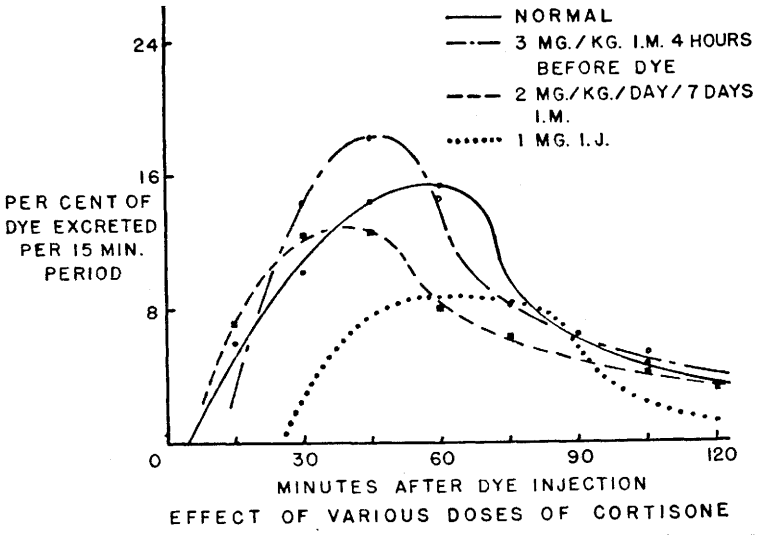

FIG. 1. Absorption and excretion of dye in rabbits. Parenteral cortisone failed to alter normal curve. Cortisone injected into the joint delayed absorption.

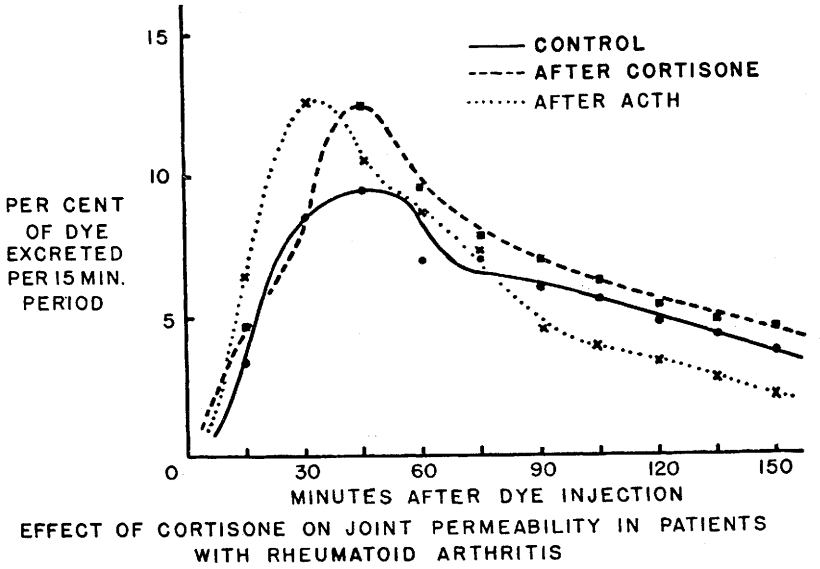

FIG. 2. Comparison of absorption and excretion of dye injected into knee joints of patients with rheumatoid arthritis before and after therapy with cortisone and ACTH.

Three of the animals received 1 mg of Cortisone intra-articularly, and the remainder received it intramuscularly. ACTH was given for 5 courses in 2 rabbits. Renal function was determined by intravenous administration of the dye before and after Cortisone and ACTH therapy. Studies were made of the effects of hyaluronidase administered into the joint along with the dye in 3 normal animals, after Cortisone in 2 animals, and after ACTH in one animal. The presence of dye in the joint was confirmed by careful dissection of the joints of several animals. The method used for investigating synovial membrane permeability in patients was similiar to that outlined above. All patients studied suffered from severe rheumatoid arthritis. In each case the knee joint was injected with 6 mg (1 ml) of PSP and urine collections were made at fifteen minute intervals with a retention

TABLE I. Joint PSP in Rabbits.

Periods		1	2	3	4	5	6	7	8	Total
Control	Avg	5.8	10	15	13.8	8.3	5.8	3.9	3.1	63.3
	SD*	4.6	4.8	4.8	5.6	.7	1	3.8	1	6.7
Cortisone										
3 mg/kg	Avg	0	14	17.5	14	7.5	8	4.5	—	65
3 hr	SD	0	3	2.5	3	2.5	2	.5	—	3
2 mg/kg	Avg	6.5	12	12	7.5	5.5	5.5	4	3	56
7 days	SD	.5	2	2	.5	.5	.5	1	1	6
1 mg IJ	Avg	0	3	5.8	15	7.5	7.5	2	.7	38
	SD	0	2	4.8	4	4	4.7	1	—	8.7
ACTH										
.3 mg/kg	Avg	3	21	13.5	9	4.5	4.5	2	2.5	60
8h-3 days	SD	3	6	4.5	0	2.5	1	2	2.5	7
Hyaluronidase										
150 TRU IJ	Avg	10.8	17.5	14.2	13	9	5	5	3	71.2
	SD	2.3	7	6.2	3	2.5	1	—	—	4
After corti-	Avg	7	13.5	13.5	8.5	4.5	3.5	2	2	54.5
sone	SD	1	.5	.5	1.5	.5	.5	0	0	4.5
After ACTH	Avg	0	13	14.2	9.1	6.4	4	4.1	1	51.8
	SD	—	—	—	—	—	—	—	—	—

* SD = Stand. dev.; TRU = Turbidity reducing units; IJ = Intra joint.

catheter in the bladder. Adequate oral fluid intake was insured throughout the two and one-half hour period of study. Urine samples were tested, as in the rabbit studies, to determine the percentage of dye in each 15 minute sample. Initial clinical observations included degree of joint deformity, blood counts, sedimentation rates, and synovial permeability to PSP. Cortisone was given to 9 patients for a total of 12 courses in a dose schedule of 300 mg intramuscularly the first day, then 100 mg daily for a total of 14 days at which time studies were repeated. Identical studies were performed on 3 patients before and after a course of ACTH consisting of 20 mg every 6 hours for 14 days. Renal function was determined before and after administration of ACTH and Cortisone in all patients, and no change was noted.

Results. Excretion of the dye following its intra-articular administration to normal untreated rabbits reached a maximum output between 45 and 60 minutes, and excretion was not altered by treatment of animals with Cortisone in doses of 2 mg/kg for 7 days or 3 mg/kg for 3 hours before the study. (Fig. 1) However, 1 mg of Cortisone injected into the joint with the dye resulted in a retardation of its excretion. ACTH in doses of 0.3 mg/kg every 8 hours for 3 days also failed to alter excretion of the dye. Renal function was not inhibited by Cortisone or ACTH.

The administration of 150 turbidity reducing units (TRU) of hyaluronidase into the joints of normal animals resulted in a probably insignificant increase in the rate of excretion of the dye. In animals treated with ACTH or Cortisone, studies of the effects of hyaluronidase showed little alteration in the rate of excretion. Total amounts of dye excreted over the 2-hour period bore no correlation to therapy.

In patients with rheumatoid arthritis, the clinical response to Cortisone and ACTH therapy was marked, but renal excretion of PSP was not altered by therapy. Excretion of PSP injected into the joint reached a maximum in 45 minutes, and was unaltered by Cortisone and ACTH therapy. White blood counts, sedimentation rates and eosinophil counts showed the expected response(3) to therapy with these two substances, but the magnitude of clinical response to anti-rheumatic therapy did not correlate with even minimal variations in the synovial permeability as measured by PSP.

Discussion. The report of Seifter described definite changes in the synovial membrane permeability of rabbits. Hyaluronidase accelerated and ACTH or Cortisone retarded dye

TABLE II. Joint PSP in Patients.

Periods		1	2	3	4	5	6	7	8	9	10	Total
Cortisone												
Pre-treatment	Avg	3	8.3	5.8	6.7	6.7	6	5.3	5	4.2	3.6	56.7
	SD*	2.4	5.5	5.6	3.2	3.6	2.8	3.3	2.6	1.5	2	26.2
Post	Avg	5	7.7	12.3	9.3	7	7	6	5.2	4.2	4	69.1
	SD	5.4	4.5	10	5.7	2.4	2.9	1.9	1.3	2.1	2.6	20.2
ACTH												
Pre-treatment	Avg	3	11.3	10	14	7	6.3	5.3	5.2	3.8	3.8	68.3
	SD	1.6	3.7	3.2	7.5	2.8	.9	.9	.9	.5	.9	9.4
Post	Avg	7	12	10	8.5	6.7	4.8	3.9	3.6	2.7	2.5	65
	SD	3.3	2.8	.9	.8	.9	1.2	.3	.4	.4	.6	8.3

* SD = Stand. dev.

absorption and excretion. From these results he inferred that permeability studies provided an accurate measure of the anti-rheumatic action of certain drugs. Considering the selective permeability of synovial membranes to naturally occurring joint substances (proteins, electrolytes, etc.)(4), it would seem that permeability to PSP might be an inappropriate measure of synovial membrane change. In view of the known histologic changes of rheumatoid arthritic joints following ACTH and Cortisone therapy(5) and the lack of changes in permeability to PSP, our results further suggest that this agent is an inadequate measure of synovial membrane permeability. For greater accuracy, we chose to collect urine at fifteen minute intervals and to calculate the percent of the administered dye excreted during each period rather than record only the times of appearance, peak excretion, and disappearance of the dye. The possibility of depression of renal excretion of PSP by ACTH or cortisone has been eliminated by kidney function studies. The membrane permeability to PSP was not altered to any significant degree in those animals treated with ACTH or Cortisone. The one exception to this statement occurred in 3 animals given Cortisone directly into the joint. We believe that the injection of particulate matter such as Cortisone could have mechanically altered permeability, and we are unwilling to draw conclusions from this limited study. Hyaluronidase injected intra-articularly in the rabbits resulted in a questionable increase in PSP dye excretion. Previous administration of ACTH or Cortisone did not alter this effect appreciably.

The human experiments closely paralleled those done in rabbits. The failure of ACTH and Cortisone to alter joint permeability in these patients was equally apparent regardless of marked clinical improvement.

Summary and conclusions. (1) Permeability studies were performed in rabbits and in patients with rheumatoid arthritis using the method of Seifter which employs injection of PSP into the joint, and recovery of the dye from the urine. (2) Cortisone and ACTH were given to rabbits and patients to determine any effect upon synovial membrane permeability and to detect any anti-hyaluronidase effect in rabbits. (3) Our results fail to demonstrate significant alterations of synovial permeability in rabbits or humans following parenteral therapy with ACTH or Cortisone. There was no evidence of hyaluronidase inhibition in rabbits. (4) Cortisone injected into the joints of rabbits impaired the absorption of PSP. A mechanical blocking effect was not excluded. (5) The injection of PSP into joints is not necessarily a valid measurement of permeability of synovial membranes to physiologic substances.

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