

TABLE I.

Subject	Hippuric acid (expressed as benzoic acid) excreted in hour	Remarks
1	.71	Test done a.m.
	.73	Test done p.m.
	1.01	15 gm. gelatin ingested 1 hr. before test
2	.95	
	.91	
3	.76	
	.88	
4	.84	
	.92	
5	.92	
6	.72	
7	.99	
8	.82	
9	.81	
10	.79	
11	.74	
12	.82	
13	.91	
14	.74	

The intravenous injection can therefore serve as an alternative method to the oral administration of sodium benzoate in the determination of liver function by means of the hippuric acid synthesis.

9746 P

Acetylation of Sulfanilamide by Liver Tissue *in vitro*.

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Marshall, Emerson and Cutting¹ and Fuller² have demonstrated that sulfanilamide is partially converted by man and the rabbit into a conjugated form. The latter has been isolated from the urine of these species after oral ingestion and has been shown to be acetyl sulfanilamide. Ellinger and Hensel³ have demonstrated the acetylation of para-amino-benzoic acid by the rabbit, and Muenzen, Cerecedo, and Sherwin⁴ have concluded that acetylations are very likely confined to the liver.

¹ Marshall, E. K., Jr., Emerson, K., Jr., and Cutting, W. C., *J. Am. Med. Assn.*, 1937, **108**, 953.

² Fuller, A. T., *Lancet*, 1937, **1**, 194.

³ Ellinger, A., and Hensel, M., *Z. physiol. Chem.*, 1914, **91**, 21.

⁴ Muenzen, J. B., Cerecedo, L. R., and Sherwin, C. P., *J. Biol. Chem.*, 1924, **60**, XVI.

In an attempt to elucidate the course and site of the acetylation of sulfanilamide, tissue slices of rabbits and rats were suspended in Ringer bicarbonate solution (pH 7.4) containing 0.2% glucose and concentrations of sulfanilamide varying from 10 to 200 mg. %. The mixture was then shaken in a water bath at 37°C. for 4 to 5 hours in an atmosphere of 5% CO₂ and 95% O₂. Samples of the fluid were withdrawn and analyzed for free and conjugated sulfanilamide by the method of Marshall.⁵ Of the tissues used (liver, muscle, spleen, kidney, and blood) only the liver could convert sulfanilamide into the conjugated form, and of the preparations used (slices and broken cell suspensions) only the slices were effective. The conjugated form was isolated from the mixture in a crystalline state and was shown to be acetyl sulfanilamide by mixed melting point determinations and by the recovery of the theoretical amounts of acetic acid and sulfanilamide after acid hydrolysis. Conjugation was observed with each of 14 rabbit livers, but with only 2 of 4 rat livers.

The addition of sodium lactate, pyruvate, acetate, and acetoacetate increased the aerobic formation of the conjugated compound. Under anaerobic conditions, little or no acetylation took place except in the presence of added acetate or pyruvate.

Although the acetylation of sulfanilamide is thus dependent upon aerobic respiration, it was found, by the Warburg technique⁶ that the presence of sulfanilamide in concentrations up to 200 mg. % had no effect upon either aerobic or anaerobic tissue respiration.

The extent of the conversion under varying conditions is indicated in Table I.

TABLE I.
Mg. Sulfanilamide Converted into Conjugated Form per Mg. of Dry Tissue

Original conc. of sulfanilamide in mg. %	Control	Acetate	Pyruvate	Lactate	Acetoacetate
		Aerobically.			
20	.0059	.0068	.0081	.0082	.0068
50	.0063	.0148	.0113	.0133	.0112
200	.0060	.0097	.0193	.0082	.0133
50	.0075	.0146	.0139		
		Anaerobically.			
20	.0022	.0040	.0084		
50	None	.0048	.0043		
20	"				
20	"				

⁵ Marshall, E. K., Jr., *Proc. Soc. Exp. Biol. and Med.*, 1937, **36**, 422.

⁶ Warburg, O., *The Metabolism of Tumors*, Chapter 5, page 75, Constable and Co., Ltd., London, 1930.

The final concentration of added substances ranged from 0.02-0.04 M. 50 to 150 mg. wet rabbit liver slices were used per 3 cc. medium.

9747 P

Observations on Histologic Structure of Anterior Pituitaries of Old Female Rats.*

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Considerable information is now available concerning the histologic structure of the anterior pituitary of the female rat during the normal estrous cycle,¹ and after various experimental procedures.^{2, 3} However, as far as the writers are aware, there are available in the literature no reports of definite histologic studies on the anterior pituitaries of old female rats. Below are presented certain observations on a histologic condition found in the anterior pituitaries of old female rats which we have not observed in the anterior lobes of younger animals.

We have examined the pituitaries of 26 old breeding female rats of the experimental colony strain of the Wistar Institute. A majority had given birth to from 4 to 6 litters; all had delivered at least one litter. At autopsy the animals ranged in age from 543 to 848 days, the average age being 632 days. Three animals were found to have small subcutaneous tumors in the mammary region; subsequent histologic examination revealed that these were fibroadenomata of the mammary gland. The pituitaries were variable in weight, ranging from 10 to 45 mg., the majority weighing less than 17 mg. In 2 animals the right lateral halves of the anterior lobes were markedly and irregularly enlarged, causing a definite anterior and dorsal bulging of the anterior lobe tissue. The enlarged areas were extremely congested and very soft. In both, the posterior lobe appeared to be compressed and reduced in size. These enlarged

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¹ Wolfe, J. M., and Cleveland, R., *Anat. Rec.*, 1933, **55**, 233.

² Severinghaus, A. E., *Physiol. Rev.*, 1937, **17**, 556.

³ Wolfe, J. M., *Albany Medical Annals*, 1937, **56**, 102.