9508

Comparative Lipid Analyses of Cortex and Medulla of Beef Adrenals.

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The lipids of the adrenal gland have been assigned to a position in the cortical region of the gland. This conclusion has been based upon histological analytical techniques, employing various lipid differentiating stains, such as osmic acid, Sudan III, Nile pink or Nile blue sulfate. The effect of polarized light on the histological sections has been used as an additional criterion. In our work, described below, attempts to correlate histological lipid analyses with purely chemical analyses have resulted in rather paradoxical findings, namely that the quantitative histochemical analysis for lipids does not agree with a chemical assay for lipids in a similar tissue. Histologically lipids are demonstrable in the cortex of the gland but not in the medulla. Yet our chemical methods have shown that both the cortex and the medulla of beef adrenals contain lipids and that they are similar in composition.

I. Comparative Analysis of Cortical and Medullary Extracts for Total Lipid Content. Fourteen beef adrenals obtained fresh from a local packing house were cleaned of all extraneous body fat and then dissected into their separate cortical (85g) and medullary (24g) portions. A sample of each tissue was extracted with a 3:1 alcohol-ether mixture as described by Bloor[1] and made up to 500 cc. volume. The extracts of cortex and medulla lipids were separately analyzed for total lipid content according to Bloor's method[1] for the microdetermination of blood lipids. In addition the lipid content in each case was determined directly by evaporating the solvent from aliquots of the extract. The data are shown in Table I.

The data show the presence of nearly equal amounts of lipid in the cortex and in the medulla, calculated as percentage of the wet gland, in contrast to the analyses by histological staining methods which indicate that most of the lipids are in the cortex. The Bloor oxidative procedure gave results much lower than those obtained by direct weight. This lack of agreement is to be expected because the Bloor method estimates only total fatty acids and cholesterol and is

[1] Bloor, W. R., *J. Biol. Chem.*, 1928, **77**, 53.

TABLE I.
Comparative Analysis of Total Lipids of Cortex and Medulla.

	Total Lipids %
(1) Direct Method, wet basis	
Cortex	6.3
Medulla	5.6
(2) Direct Method, dry basis (calculated)	
Cortex	19.0
Medulla	28.2
(3) Bloor Oxidative Method, dry basis	
Cortex	12.3
Medulla	13.1

based on oxidation factors of characteristic blood lipids. We believe that the figures by the 2 procedures are of importance in that both show the presence of comparable amounts of lipids in the medulla and cortex.

II. Comparative Analysis of Cortical and Medullary Extracts for Differential Lipid Components. Aliquot portions of similarly prepared extracts of beef adrenal cortex and medulla were analyzed for the following lipid fractions: phospholipids according to Bloor,[2] the free and total cholesterol fractions by Okey,[3] and the total fatty acid portion as described by Man and Gildea.[4]

TABLE II.
Lipid Analysis of Cortex and Medulla.

Component	Cortex*	Medulla*
(1) Phospholipid %	3.09	2.66
(2) Total Fatty Acids %	1.76	1.46
(3) Free Cholesterol %	0.230	0.300
(4) Total Cholesterol %	0.255	0.354
(5) Combined Cholesterol (calculated) %	0.025	0.054

*All analyses on wet basis.

The results of these assays show a slight increase in phospholipids and total fatty acids in the cortex. The medulla, however, contains more cholesterol. Our results for free and combined cholesterol are lower than those usually reported; however, we have checked our methods against pure cholesterol with satisfactory agreement.

Summary. While the data above are not extensive, we believe them to be of importance in demonstrating lack of agreement between chemical findings and accepted histological technique. (1) Comparative analysis of extracts of both cortex and medulla indi-

2 Bloor, W. R., *J. Biol. Chem.*, 1929, **82**, 273.

3 Okey, R., *J. Biol. Chem.*, 1930, **88**, 367.

4 Man, E. B., and Gildea, E. F., *J. Biol. Chem.*, 1932-3, **99**, 43.

cates that the total lipid content of the medulla and cortex of beef adrenals is essentially the same. (2) Study of the individual lipids shows more phosphatide and fatty acids in the cortex and more total, free and combined cholesterol in the medulla.

9509 P

Use of Sulfanilamide in the Treatment of Type XIV Pneumococcus Infections in Mice.

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Rosenthal[1] showed that sulfanilamide (*p*-aminobenzenesulfonamide), used therapeutically, prolonged the lives of mice infected with 10 to 100 M.L.D. of types I, II, and III pneumococci. Cooper,[2] independently, made a similar finding in type III pneumococcus infections. Similar experiments of our own, which had been in progress prior to these publications, confirmed Cooper's results. This observation suggested the use of such therapy in infections with other types of pneumococci—IV to XXXII—and particularly in those types for which potent antisera are not readily available. The present report concerns experiments with type XIV pneumococci; this organism causes about 16% of all pneumococcus pneumonias in children less than 12 years of age and 2.5% of such pneumonias in persons older than 12 years.[3]

A type XIV pneumococcus, freshly isolated from the bloodstream of a patient suffering from bronchopneumonia, was passed through mice until maximal virulence was obtained. This organism was only moderately virulent for mice; 0.5 cc. of a 1-10,000 dilution of an 18-hour broth culture, injected intraäbdominally, invariably killed mice within 48 hours. Forty-eight mice were injected intraäbdominally with 0.5 cc. of a 1-1000 dilution of an 18-hour broth culture of this organism; 8 of these mice were untreated controls. The remaining mice were divided into 5 groups; all received subcutaneous injections of 5 mg. of sulfanilamide* (0.5 cc. of a 1% aqueous

1 Rosenthal, S. M., *Public Health Reports*, 1937, **52**, 48.

2 Cooper, F. B., Gross, P., and Mellon, R. R., PROC. SOC. EXP. BIOL. AND MED., 1937, **36**, 148.

3 Bullowa, J. G. M., *J. Clin. Invest.*, 1935, **14**, 373.

* Sulphonamid P, Burroughs, Wellcome and Co.