intraperitoneal administration and disappears completely within 24 hours. Acetylation becomes apparent at about 3 hours after the injection. The deposition of relatively increasing amounts of the extremely insoluble acetylated derivative may be responsible for the renal irritation.

The data presented demonstrate the extreme rapidity of absorption and excretion of sulfathiazole and lend further support to the view which has been expressed elsewhere: that the acute precipitation in the urinary tract is due mainly to a high rate of elimination of sulfathiazole from the body. Thus after concentration of the glomerular filtrate by reabsorption of water, precipitation occurs in the collecting tubules.

A similar picture could not be produced with sulfanilamide, sulfapyridine, sulfamethylthiazole or their sodium salts.

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# SCIENTIFIC APPARATUS AND LABORATORY METHODS

## A MODIFICATION OF RIDDLE'S METHOD OF PROLACTIN ASSAY

WHILE we were comparing the alkaline alcoholic extraction method of Bates and Riddle<sup>1</sup> with the acid acetone extraction method of Lyons<sup>2</sup> for preparing lactogenic hormone of the anterior pituitary, it became necessary to use a large number of pigeons for accurate assaying of various fractions used in the investigations.<sup>3</sup> The purchasing and keeping of the pigeons, however, became so expensive that a way had to be found to reduce the cost as much as possible. The method described below is the result of such an attempt and enables one to assay the hormone without sacrificing the birds. Thus the cost of investigation was greatly reduced.

As in the original method of Riddle, Bates and Dykeshorn,<sup>4</sup> we used both male and female pigeons. On the previous day, before the injection of the sample, the feathers on the pectoral regions of the birds as well as on the area around the crop sac on the neck are carefully plucked. Before the injection of the sample, the skin layer over the crop sac on either right or left side of the median line is incised to the length of about two centimeters, so that the crop sac can be clearly seen. The crop sac is usually colorless and transparent and the presence of food material such as wheat, oats and barley within the sac is clearly discernible. If the crop sac is opaque and thick as in the nesting and nursing period, the pigeon is not suitable for the assay of the hormone and should be exchanged for another bird. Only the pigeons with clear transparent sac are used for the assay. After examination of the sac, the opening is closed by sewing with surgical suture and an appropriate antiseptic,

such as tincture of iodine or merthiolate solution, is applied. As in the original method, a definite quantity of the sample solution is injected once daily for four days. On the fifth day (ninety-six hours after the first injection) the crop sac located on the other side of the one previously examined is exposed and examined. According to the condition of the thickening of the sac membrane, the potency of the hormone solution is evaluated.

The method is easy to execute with a little practice. It should be borne in mind, however, that the examination of the sac should be made as soon as it is exposed. The irritating operations such as rubbing of the exposed area with alcohol-soaked cotton swabs or cheesecloth must be avoided, since a slight irritation of the crop sac membrane will result in the thickening of the membrane and may cause an erroneous evaluation of the potency of the sample. It is important to note that during the incising of the skin layer over the sac, great care should be exercised to avoid tearing the crop sac membrane. If, however, the crop sac membrane is cut, the area should be immediately closed by holding together the surrounding membrane and tying with sterile silk thread.

After the examination of the sac, the incised skin layer is closed by sewing with surgical suture and an antiseptic is applied. The pigeon is then kept in a large cage for three to four weeks or until the incised area is perfectly healed. They are then ready to be used again for prolactin assay.

The authors are aware of the fact that an accurate assaying of the hormone by this method or any other biological method is a difficult task. This is due to the variation in the sensitivity of the individual birds, even among the same strain.<sup>1,2</sup> The difficulty can be overcome if a large number of birds are used for the assay of a sample and also by using a standard preparation for comparison as recommended by Bates and Riddle or by the Commission on Biological Standardization of the League of Nations. The authors believe that by the use of the method described here and with

<sup>&</sup>lt;sup>1</sup> R. Bates and O. Riddle, Jour. Pharm. Exp. Therap., 55: 365, 1935.

<sup>&</sup>lt;sup>2</sup> W. R. Lyons, Proc. Soc. Exp. Biol. and Med., 35: 645, 1937.

<sup>&</sup>lt;sup>3</sup> The results of the study will be published in the *Journal of Biochemistry* (Japan).

<sup>4</sup> O. Riddle, R. Bates and S. W. Dykeshorn, Amer. Jour. Physiol., 105: 191, 1933.

the use of a standard preparation, a great deal of saving will be accomplished without sacrificing the accuracy of the assay.

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### CELLOPHANE COVER SLIPS AND A METHOD FOR MOUNTING

BECAUSE the price and paucity of cover glasses used in the preparation of microscope slides have become almost prohibitive of their use, an efficient, economical substitute has been sought. O'Brien and Hance<sup>1</sup> have stated that isobutyl methacrylate may be used to replace the cover. This has not been found practical when slides are produced in large numbers. The solution seems also to detract from the brilliancy of the stain. Suntzeff and Smith<sup>2</sup> recommend a plastic, cellulose acetate, obtained in sheets the approximate thickness of No. 1 cover glasses. They experience some difficulty in its use because the plastic tends to curl unless the slides are carefully dried at room temperature for five to six days.

Plain transparent Cellophane<sup>3</sup> has been used in our laboratory during the past seven months. Cellophane covers have a refractive index of 1.53, dry rapidly and without curling, are very thin and inexpensive. Cellophane can be obtained in rolls 15/16ths inch wide, slightly less than the width of a slide. Strips of the desired length are cut as needed. No. 300 plain Cellophane seems to be most satisfactory. It is .00088 inch (0.022 mm) thick. (No. 1 cover slips average 0.13 mm in thickness.) It is possible to take photomicrographs from slides having Cellophane covers. Oil immersion does not affect Cellophane but for high magnifications replacements with glass covers may prove desirable.

The only difficulty so far encountered in the use of Cellophane as a substitute for glass cover slips has been the storage of the roll. During the winter months it can be kept at ordinary room temperatures, but under more humid conditions the Cellophane absorbs moisture, the edges of the roll wrinkle and will not lie smoothly. This wrinkling can be avoided by storing the roll of Cellophane in a desiccator, but continued storage over a long period causes warping and eracking.

Moistureproof Cellophane is less susceptible to atmospheric changes, but it too will wrinkle, and does not seem to be quite as clear as the plain. Cellulose acetate<sup>4</sup> No. 88 was tried, but was found not to adhere to the slide when dry. This is probably caused by the xylol used to mount the covers.

A method long used for mounting glass covers is successful with Cellophane as well. Instead of leaving the slides to dry in trays they are turned face down on clean paper towels and xylol is pipetted between them. An extra towel on top readily absorbs any excess on the back of the slide. In 15-30 minutes the slides are ready for use. This method has several advantages: (1) it insures a thin layer of mounting medium under the cover, (2) the slides are ready for immediate use. Further drying in an oven is entirely unnecessary, (3) it removes any excess mounting medium, leaving the edges and ends of the slide clean and dry. Canada balsam was first used in this manner but since the appearance of an article by Groat<sup>5</sup> we have used Clarite exclusively. It dries more rapidly than balsam, is clear and adheres well to Cellophane.

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- <sup>4</sup> Courtesy of du Pont Company.
- <sup>5</sup> R. A. Groat, Anat. Record, Vol. 74, No. 1, 1939.

### **BOOKS RECEIVED**

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<sup>&</sup>lt;sup>1</sup> O'Brien and Hance, SCIENCE, 91: 412, 1940.

<sup>&</sup>lt;sup>2</sup> Suntzeff and Smith, SCIENCE, 92: 17-18, 1940.

<sup>&</sup>lt;sup>3</sup> A du Pont product obtained through H. D. Catty Distributing Company, 3311 Carroll Avenue, Chicago, Illinois.