The results indicate, therefore, that an appreciable decrease in the adrenin content of the adrenals occurs in cats treated for a sufficiently long time at a very low oxygen tension. An additional cat subjected to an hour exposure at the same oxygen tension showed a value of 0.27 mg of adrenin per kg of body weight, which suggests that shorter exposure may not be effective; but since this value is within twice the standard deviation of the 3-hour series, the result cannot be considered significant. The possible effects from lighter degrees of anoxemia, for shorter periods, such as occur clinically during nitrous oxide anesthesia, need further investigation. Similarly, determinations of threshold degrees of anoxia and of time of exposure are beyond the scope of the present study.

Summary. Cats treated for 3 hours at an oxygen tension of 53 mm Hg show a 40% decrease in the adrenin content of their adrenals, as determined by an adaptation of the Moodey colorimetric technic.

9912 P

A Method for the Isolation of Pregnandiol from the Urine of Pregnant Mares.

PAUL G. WEIL. (Introduced by Carl G. Hartman.)

From the Surgical Pathological Laboratory, Department of Surgery, Johns Hopkins Hospital and University, Baltimore, Maryland.

The isolation of pregnandiol from the urine of pregnant mares has been reported by Marker, *et al.*[1] A method for the isolation of pregnandiol from the urine of pregnant mares based on a method for the quantitative determination of pregnandiol in human pregnancy urine[2] is herewith described because of its relative simplicity and possible application in the study of the function of the corpus luteum of other animals.

The fresh urine* without the addition of preservative is incubated at 37°C for 96 hours. It is filtered with suction and the precipitate is collected and heated with 100 cc of 95% ethyl alcohol. The alcoholic mixture is filtered with suction. This filtrate is evaporated to

1 Marker, R. E., Kamm, O., Crooks, H. M., Jr., Oakwood, T. S., Lawson, E. J., and Wittle, E. L., *J. Am. Chem. Soc.*, 1937, **59**, 2297.

2 Weil, P. G., to be published.

* In this determination a liter of urine of the sixth month of pregnancy was used.

dryness and the residue taken up in 10 cc of acetone and 10 cc of a 0.1 N solution of NaOH. When complete solution is effected with moderate heating, the volume is brought to 50 cc by the addition of 30 cc of 0.1 N NaOH. The solution is placed in the cold and pregnandiol completely precipitates when the solution is thoroughly chilled. The pregnandiol is collected by filtration with suction and the precipitate is washed with warm water. The precipitate is dissolved in a minimum amount of hot acetone and 2 volumes of 0.1 N NaOH added. This is put in the cold to effect precipitation. A third precipitation using a minimum amount of ethyl alcohol and two volumes of water usually give a pure product.

The essential points in the procedure are: (1) The liberation of the free form of pregnandiol from its conjugation form by enzyme action during incubation; (2) the insolubility of pregnandiol in aqueous solutions such as urine; (3) the purification of pregnandiol by precipitation from alkaline acetone.

9913

Effect of Nicotinic Acid, Its Isomers and Related Compounds upon Nutrition of *Staphylococcus aureus.*

MAURICE LANDY. (With the coöperation of Lester Szabo.)
(Introduced by N. Paul Hudson.)

From the Research Division, S.M.A. Corporation, Cleveland, and the Department of Bacteriology, Ohio State University, Columbus, Ohio.

Nicotinic acid has aroused widespread interest with the discovery of its rôle as a co-enzyme in tissue metabolism,[1, 2] later by the finding of its activity as a growth factor for *Staphylococcus aureus*,[3, 4] and most recently by its dramatic effect in the cure of canine black-tongue[5] and human pellagra.[6, 7, 8] In a series of unpublished ex-

1 Warburg, O., and Christian, W., *Biochem. Z.*, 1936, **287**, 291.

2 Schlenk, F., and Euler, H. v., *Arkiv. Kemi, Mineral Geol.*, 1936, 12B, No. 20.

3 Knight, B. C. J. G., *Biochem. J.*, 1937, **31**, 731.

4 Knight, B. C. J. G., *Biochem. J.*, 1937, **31**, 966.

5 Elvehjem, C. A., Madden, R. J., Strong, F. M., and Wooley, D. W., *J. Am. Chem. Soc.*, 1937, **59**, 1767.

6 Fouts, P. J., Helmer, O. M., Lepkovsky, S., and Jukes, J. H., PROC. SOC. EXP. BIOL. AND MED., 1937, **37**, 405.

7 Smith, D. T., Ruffin, J. M., and Smith, S. G., *J. A. M. A.*, 1937, **109**, 2054.

8 Spies, T. D., Cooper, C., and Blankenhorn, M. A., *J. A. M. A.*, 1938, **109**, 622.