with these agents results in a decreased production of acid. It is noteworthy that infection of HeLa cells by the latter viruses listed culminates in death and lysis of the host cells. In contrast, adenoviruses do not appear to lyse infected HeLa cells(2) and it has been suggested that these respiratory tract viruses do not actually kill the cells in which they multiply(2). It can be deduced, therefore, that the increased glycolysis and accumulation of carboxylic acids require a particular type of viral infection or cell injury. Whatever may be the mechanism of the increased accumulation of organic acids and utilization of glucose, the evidence is clear that the initiation of this phenomenon is an intrinsic characteristic of the adenoviruses and a constant effect upon HeLa cells which they infect.

Summary. Fluids from cultures infected with type 4 adenovirus were markedly more acid than fluids from non-infected control cultures. The lowered pH of infected cultures resulted from an accumulation of lactic, pyruvic, acetic and alpha ketoglutaric acids. The acids were separated by chromatography on a celite column, and measured by colorimetric and acid-base titration technics. Confirmation of the identity of the organic acids was obtained by paper chromatography. Concomitant with the increased accumulation of acid in fluids of infected cultures, an increased utilization of glucose by cells of the virus-infected cultures occurred.

1. Huebner, R. J., Rowe, W. P., Ward, T. G., Parrott, R. H., and Bell, J. A., *New Eng. J. Med.*, 1954, v251, 1077.

2. Ginsberg, H. S., *Ann. N. Y. Acad. Sci.*, in press.

3. Ginsberg, H. S., Badger, G. F., Dingle, J. H., Jordan, W. S., Jr., and Katz, S., *J. Clin. Invest.*, 1955, v34, 820.

4. Ginsberg, H. S., Gold, E., and Jordan, W. S., Jr., PROC. SOC. EXP. BIOL. AND MED., 1955, v89, 66.

5. Scherer, W. F., Syverton, J. T., and Gey, G. O., *J. Exp. Med.*, 1953, v97, 695.

6. Hullin, R. P., and Noble, R. L., *Biochem. J.*, 1953, v55, 280.

7. Swim, H. E., and Krampitz, L. O., *J. Bact.*, 1954, v67, 419.

8. Friedemann, T. E., and Haugen, G. E., *J. Biol. Chem.*, 1943, v147, 415.

9. Park, J. T., and Johnson, M. Y., *ibid.*, 1949, v181, 149.

10. Barban, S., and Schulze, H. O., *J. Biol. Chem.*, 1956, v222, 665.

11. Huang, C. H., PROC. SOC. EXP. BIOL. AND MED., 1943, v54, 160.

12. Robbins, F. C., Enders, J. F., and Weller, T. H., *ibid.*, 1950, v75, 370.

13. Robertson, H. E., Brunner, K. T., and Syverton, J. T., *ibid.*, 1955, v88, 119.

14. Lipton, M. M., and Steigman, A. J., *ibid.*, 1955, v88, 114.

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Guanine and Guanosine Deaminase Activity of Rat Mammary Gland Homogenates Through Pregnancy and Lactation.* (23114)

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Kirkham and Turner(1,2) reported changing levels of rat mammary gland DNA and PNA during pregnancy and lactation and in the mammary gland stimulated to growth with estrogen and progesterone. In rabbit mammary gland Yamamoto and Turner(3) showed increases in total DNA as glandular growth occurred. McShan *et al.*(4) have shown very striking increases in PNA content of pigeon crop gland following treatment with lactogenic hormone. Williams and Turner (5) reported apparent involvement of PNA in the action of lactogenic hormone on the mammary gland. From these studies PNA

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is implicated in the process of milk synthesis and increases in mammary gland PNA which have been noted probably correspond roughly to the amount of synthesis occurring in the mammary gland. There have been relatively few studies concerning the deaminases of guanine and guanosine. Schmidt(6) and Wakabayasi(7) reported separation of the two enzymes. Plentl and Schoenheimer(8) demonstrated guanine deaminase activity in various rat tissues. Block and Johnson(9) found guanine deaminase in rat skin. Schmidt (6) isolated guanine deaminase from rabbit liver. Even less work has been reported concerning guanosine deaminase; in fact, Kalckar doubts the existence of such an enzyme, believing the deamination of guanosine to be carried out by guanine deaminase and nucleoside phosphorylase. Guanosine deamination has been demonstrated in rabbit liver preparation by Schmidt(6) and in various tissues of the rabbit, cat and pig by Wakabayasi(7).

Methods and materials. Female albino rats, weighing approximately 150 g, were used. The mammary glands were studied in 4 physiological states; virgin non-growing, growing during pregnancy, lactating and involuting following lactation. The experimental animal was sacrificed, the mammary tissue rapidly dissected free and chilled on ice. The mammary tissue was then homogenized for 3 minutes in a Waring blendor in the cold with 9 volumes of isotonic NaCl, strained through 4 layers of cheese cloth and assayed immediately for guanine and guanosine deaminase activity. The assay was carried out using the manometric technic described by Zittle(10) for determination of liberated ammonia. The reaction flask contents for all determinations were as follows: 1.4 ml of 1.15 M $NaHCO_3$ gassed with CO_2, 1 ml of mammary gland of homogenate, and 5 mg of substrate. The gas phase was 100% CO_2 and a bath temperature of 38°C. The final flask volume was 3.4 ml and the flask pH 7. The flask contents were assayed for DNA content by the method of Webb and Levy(11). A DNA standard was used which assayed 7.8% phosphorus.

Results. Rat mammary gland homogenates were found to deaminate both guanine and guanosine in all physiological states studied. The rate of guanine deamination was consistently some 3 times that for guanosine.

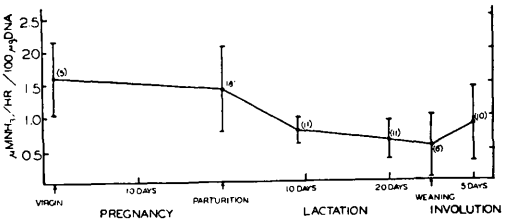

FIG. 1. Guanine deaminase activity during pregnancy, lactation and involution in rat mammary gland.

A change in rate of deamination was noted when mammary tissue from pregnant animals was compared to that from animals in lactation (Fig. 1). Guanine deamination declined from a mean value of 1.42 μM/hr/100 μg DNA during pregnancy to 0.72 μM/hr/100 μg DNA during lactation. This change was highly significant ($P < .0025$).

This same change in rate of deamination was noted for guanosine deamination when pregnancy and lactation were compared. Guanosine deamination declined from a mean of 0.44 μM/hr/100 μg DNA during pregnancy to a value of 0.24 μM/hr/100 μg DNA during lactation. This change was also highly significant ($P < .0025$).

Summary. Guanine deaminase activity and guanosine deaminase activity of rat mammary homogenates were studied through pregnancy, lactation and involution. Guanine and guanosine deamination occurred in mammary gland homogenates during all physiological states studied. The rate of deamination of guanine and guanosine exhibited a marked drop from the level of pregnancy to a lower level throughout lactation.

1. Kirkham, W. R., and Turner, C. W., PROC. SOC. EXP. BIOL. AND MED., 1953, v83, 123.

2. ———, *ibid.*, 1954, v87, 139.

3. Yamamoto, H., and Turner, C. W., *ibid.*, 1956, v92, 130.

4. McShan, W. H., Davis, J. S., Soukup, S. W., and Meyer, R. K., *Endocrinology,* 1950, v47, 274.

5. Williams, W. F., and Turner, C. W., PROC. SOC. EXP. BIOL. MED., 1955, v90, 531.

6. Schmidt, G., *Z. physiol. Chem.*, 1932, v208, 185.
7. Wakabayasi, Y., *J. Biochem. Japan*, 1938, v28, 185.
8. Plentl, A. A., and Schoenheimer, R., *J. Biol. Chem.*, 1944, v153, 203.
9. Block, W. D., and Johnson, D. V., *ibid.*, 1955, v217, 43.
10. Zittle, C. A., *ibid.*, 1946, v166, 499.
11. Webb, J. M., and Levy, H. B., *ibid.*, 1955, v213, 107.

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Pipetting Machine with Rapid Preset. (23115)

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Several immunochemical procedures require the addition of less than cc volumes of reagents to large series of tubes. In studies on production of hemolysin by mice or other laboratory animals following the method described by Taliaferro(1) large series of serum dilutions must be made. Pipetting machines are available* for repetitive delivery of fluid volumes but resetting for some new volume requires careful recalibration and adjustment. Ordinarily, there is not enough time for these manipulations during the course of a given test. The machine described here was constructed to reduce the time required for adding quantities such as 0.02, 0.04, 0.06 cc, etc. to sets of 20 to 30 tubes in the serial dilutions of serums for estimation of their hemolysin content.

Methods. Fig. 1 is a general view of the assembly and shows presetting button (white), toggle switch (black) for operating the machine and other features described in detail in Fig. 2. Details of construction of the plunger and associated parts of the machine are illustrated in Fig. 2, I. The stainless steel plunger, A, Fig. 2, I, was made 0.874 cm in diameter and displaces a 3 cc volume when inserted a distance of 5 cm in the fluid reservoir, B. Fluid volume displaced is directly proportional to movement of the plunger. Control of the forward movement and hence of the fluid displaced is provided in the preset counter by means of the micro-switch, C. Counting rate is determined by the 4-sided cam, D, and with the gear ratio and plunger shown here the preset counter records one count for each 0.01 cc delivered. Tapped holes in various positions around the cam allow one to obtain a larger or smaller number of counts per unit quantity of fluid delivered. Worm gears, E, with a 60 to 1 ratio and the pinion gear, F, advances the rack at the rate of 4.0 cm per revolution. By revolving the slotted ram, G, in a counterclockwise direction the rack and pinion become disengaged allowing the plunger assembly to be withdrawn for reloading.

An exploded view of the preset counter indicating the relationships between its 4 principal parts is shown from left to right in Fig. 2, II. Number of counts and thus volume to be delivered is determined by the setting of the dial, 1, which positions the stop block, 2. Immediately to the right of the stop in Fig. 2, II, is shown a single unit combining the following 4 structures: the cam, 3, that actuates the microswitch controlling the motor-drive; the positioning stop, 3S; the indexing tongue, 3L; and the axle of the instrument. The controlling element, shown at far right in Fig. 2, II, during normal operation lies in the position indicated by x - - - x. This structure is shown in place in Fig. 2, III. The ratchet wheel 4R and the toothed wheel 4S are machined as a single unit and are actuated by the relay armature, 5. The wheel, 4S, has 70-1/32 inch slots spaced about its periphery and the ratchet, 4R, has 70 teeth which correspond with the divisions

* Brewer automatic pipetting machine, Baltimore Biol. Lab., Baltimore, Md., and Sterling Automatic Pipette, Ivan Sorvall, N. Y. are 2 of the available mechanical pipettors in use in our laboratory.