a small part of it remains with the *s* antigen.

Henle and other investigators reported that toxic activity of the total virus particle is neutralized by the specific antiserum. In our experiments, however, only the decrease in total phosphorus was neutralized by the specific antiserum.

A similar variation in phosphocreatine level was found by Pinchot and Bloom(7) after inoculating diphtheria toxin into rats subcutaneously, but this action was neutralized by the antitoxin.

Perhaps the toxic activity observed in our work can be attributed to one of the virus components against which antiserum has no action. On the other hand, the possibility of an interferon action must be considered. It is already known that normal cells inoculated with interferon increase their oxygen uptake and their lactic acid production, but glycolysis is related to ATP production. It seems fair to suggest that this is the way interferon acts against the virus, lowering the ATP level, so that virus synthesis cannot be accomplished(8). In any event interferon produced by the inoculated animals could be the agent responsible for the ATP and phosphocreatine decrease.

Conclusions. 1. Analysis of inorganic phosphorus, phosphocreatine, ATP and total acid soluble phosphorus was carried out in white mice previously inoculated with influenza virus and also in control animals. 2. A significant decrease was observed in muscle phosphocreatine, ATP and total phosphorus in the infected animals.

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Effect of Adrenalectomy and Corticoid Replacement on Lactational Performance in Rats.*† (27514)

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Importance of adrenocortical hormones in maintenance of lactation has been(1-4) unieequivocally substantiated in adrenalectomized rats. Levels of corticoids required for complete maintenance of lactation have been found to be 0.85 mg cortisone acetate and 0.36 mg desoxycorticosterone acetate (DOCA) per day in rats adrenalectomized on day 4 of lactation as judged by absorption of pellet implants(5). On daily injection basis, 1.1 mg cortisone and 0.43 mg DOCA per day maintained normal lactation(6).

Maintenance of normal lactation in adrenalectomized rats was accomplished with 3 mg DOCA per day while 0.1 or 1 mg were not sufficient(3). Maintenance of lactation in rats at 80% of normal was obtained with 0.46 mg cortisone per day(2). However, other investigations found that 0.5 mg 11-dehydrocorticosterone or 0.47 mg cortisone did not maintain lactation(4).

9α-Chlorocortisol at 100 μg per day maintained almost complete lactation; aldosterone at 50 μg per day maintained lactation at a lower level(7). 9α-fluorocortisol acetate at 200 μg per day maintained normal lactation

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TABLE I. Effect of Adrenalectomy (4th Day of Lactation) and Corticoid Replacement on Lactational Performance in Rats.

Group	No. of dams (day 4)	Litter wt, g (day 4)	No. of dams (day 14)	Litter wt, g (day 14)	No. of dams (day 20)	Litter wt, g (day 20)	No. of dams (day 14)	Milk production, g (day 14)	Pup mortality, % (day 14)	Pup mortality, % (day 20)
1. Normal	16	51.9 ± 1.5*	16	149.3 ± 5.6	15	203.7 ± 15.9	16	8.9 ± .7	7	18
ADRX 4th day lactation										
2. No treatment	15	53.4 ± 2.0	13	87.0 ± 6.7‡	0		13	2.0 ± .6‡	16	100
3. 1% NaCl water	9	52.3 ± 1.7	6	106.5 ± 7.2‡	2	112 ± 8.1	6	3.8 ± .8‡	24	78
4. 1-250 μg HCA inj.; 1% NaCl water	8	48.2 ± 2.8	6	105.8 ± 9.7‡	1	100			35	92
5. 250 μg pred./day; 1% NaCl water	13	50.2 ± 2.1	13	134.4 ± 7.4	13	199.8 ± 19.2			2	12
6. 250 μg HCA/day; 1% NaCl water	19	49.5 ± 2.0	19	157.3 ± 8.5	17	219.1 ± 18.3			5	16
7. 1 mg HCA/day; 1% NaCl water	16	49.1 ± 1.4	16	148.8 ± 4.5	16	195.9 ± 7.8	16	13.2 ± .8‡	1	5
8. 250 μg DOCA/day	14	50.6 ± 1.4	14	114.7 ± 7.4‡	2	104 ± 78.5	14	4.9 ± .5‡	8	92
9. 100 μg HCA/day; 50 μg DOCA/day	16	50.6 ± 1.7	16	118.1 ± 9.0‡	13	138.5 ± 13.2‡	16	6.1 ± .7†	8	26
10. 250 μg HCA/day; 100 μg DOCA/day	17	45.9 ± 1.4	17	122.6 ± 6.4‡	16	147.3 ± 12.7‡	17	7.9 ± .5	5	17
11. 500 μg HCA/day; 250 μg DOCA/day	15	50.7 ± 1.4	15	161.5 ± 6.8	15	208.2 ± 8.8	15	9.2 ± .6	1	1
12. 500 μg HCA/day	17	52.4 ± 1.4	17	125.1 ± 5.4‡	16	159.0 ± 9.9†	17	6.2 ± .5‡	0	10

* Mean ± stand. error. † Significantly different from normal ($P < .05$). ‡ Significantly different from normal ($P < .01$).
HCA = Hydrocortisone acetate. Pred. = Prednisone. DOCA = Desoxycorticosterone acetate.

in adrenalectomized rats from day 4 to day 12(8).

Present study was undertaken to test combinations of hydrocortisone acetate (HCA) and desoxycorticosterone acetate (DOCA) in regimen of daily injection to determine lowest levels of these 2 steroids which in combination maintained normal lactation from day 4 to day 20 in adrenalectomized rats.

Materials and methods. Albino rats of Sprague - Dawley - Rolfsmeyer strain were maintained on Purina lab pellets in animal room at constant temperature of 78 ± 1°F. On day 4 of lactation dams were adrenalectomized and litters reduced to 6 pups. Twelve groups of 8 to 19 litters per group were subjected to treatments as outlined in Table I. Group 4 differed from Group 3 in that the dams received one injection of 250 μg HCA immediately after adrenalectomy on day 4.

Corticosteroids were suspended in physiological saline with aid of gum arabic as described by Nandi(9) and injected subcutaneously in 0.1 ml volume making total of 0.2 ml injected in animals receiving both HCA and DOCA.

Criteria of lactational performance included litter weights on days 4, 14 and 20 of lactation and mortality of pups on days 14 and 20. Pups had access to mothers' feed at all times. Milk production of dams was estimated on day 14. This was done by isolating dam from pups for 10 hr, weighing dam and pups at end of period, reuniting and injecting dam with 2 × 1 I.U. oxytocin 10 minutes apart by intraperitoneal route, and allowing a suckling period of 30 minutes. Pups and dam were weighed again. Differences in body weights of dam and pups before and after nursing were averaged to obtain estimate of milk production.

Pups were weaned on day 20 of lactation. Dams were sacrificed on day 20 or on day when all pups were dead and autopsied for completeness of adrenalectomy.

Results. Non-treatment of dams after adrenalectomy on day 4 of lactation resulted in pup mortality of only 16% on day 14 but 100% by day 20 (Table I). Maintenance of dams on 1% NaCl drinking water resulted in significant decrease in litter weight and milk production on day 14 and mortality of 78% of pups by day 20. Single injection of 250 μg HCA on day of operation plus 1% NaCl drinking water was also ineffective in maintaining lactation. Injection of 250 μg prednisone per day plus saline drinking water or 250 μg HCA per day plus saline maintained lactation at normal level as judged by weights of litters and mortality rates of pups.

One mg HCA per day plus saline maintained normal lactation as shown by litter weights and pup mortality. This treatment resulted in a significant increase above control in milk production on day 14 of lactation.

Injection of 250 μg DOCA per day did not aid much in supporting lactation. Pup mortality was 92% on day 20. Treatment of 500 μg HCA per day enabled dams to maintain a fairly high level of lactation as judged by mortality rate of only 10% by weaning time at day 20.

Combination of 100 μg HCA and 50 μg DOCA per day supported low level of lactation; 250 μg HCA and 100 μg DOCA was somewhat better but significantly less than litter weights of control group. When 500 μg HCA and 250 μg DOCA were injected per day, adrenalectomized dams maintained lactation at level similar to normal.

Two treatments produced marked body weight changes of dams. Those receiving 250 μg DOCA per day gained an average of 29 g from day 4 to 14, while those receiving 500 μg HCA lost 38 g from day 4 to 14 and 10 g from day 14 to 20. Body weight changes in other groups did not vary over 15 g between days 4 and 20.

Discussion. Maintenance of lactation in adrenalectomized rats from day 4 to day 20 with 500 μg HCA and 250 μg DOCA per day was accomplished. These levels of adrenocorticoids are approximately 70% of the levels of cortisone and DOCA found by Cowie (5) to be required for lactation in rats. Glucocorticoid (HCA in this case) is more effective than mineralocorticoid (DOCA in this case) in supporting lactation. These findings support work of Gaunt *et al.*(2). In addition to action on carbohydrate metabolism, glucocorticoid may have ability to control transfer of electrolytes and water between extra- and

intracellular compartments of body(10).

Initiation of lactation in hypophysectomized rats was accomplished by Bintarningsih *et al.*(11) using combinations of lactogenic hormone and HCA or prednisolone acetate and growth hormone. Five mg lactogenic hormone plus 1 mg HCA or 0.4 mg prednisolone acetate maintained lactation at approximately 50% of normal. On basis of present work, levels of glucocorticoids used in above study were adequate for maintenance of normal lactation.

Significantly higher than normal milk production was found on day 14 of lactation in dams maintained on 1 mg HCA per day and 1% saline. Since the litters of these dams did not weigh any more than normal litters, question arises as to the explanation for this. Perhaps the composition of milk was changed; pups did not consume all milk that was available; or pups did not begin to eat solid food on day 16 of lactation because milk was more readily available.

Summary. Sprague-Dawley-Rolfsmeyer rats were adrenalectomized on day 4 of lactation and lactation was maintained by daily subcutaneous injections of corticosteroids from day 4 to 19. Data suggest that requirements for normal lactation in rats are 250 to 500 μg hydrocortisone acetate or equivalent in glucocorticoid activity and 250 μg desoxycorticosterone acetate or equivalent in mineralocorticoid activity. Either steroid alone at these levels was unable to support lactation but combination was effective.

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Treponemal Antigens as Related to Identification and Syphilis Serology. (27515)

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Since the introduction of the Treponema Pallidum Immobilization (TPI) test by Nelson and Mayer(1), a renewed interest in treponemal antibodies has been evidenced by the development of new test procedures. The objective has been to develop methods resulting in greater sensitivity and specificity of antibody detection in terms of syphilis. The Treponema Pallidum (T.p.) Agglutination (TPA) test(2), the T.p. Complement Fixation (TPCF) test, the Reiter Protein Complement Fixation (RPCF or KRP) test and the Fluorescent Treponemal Antibody (FTA) test are presently being used or evaluated(3). In spite of new developments and apparent improvements in the field of syphilis serology the need for more basic information in this area led to the present inquiry into the antigenic relationship of treponemes and their operation in current treponemal tests for syphilis. The following treponemes were selected for investigation: The "Reiter treponeme" (R.t.), a representative of several named spirochetes originally isolated from syphilis lesions and purported to be a cultivatable, non-pathogenic