The method thus provides a unique opportunity for the systematic study of physiological and psychological functions at different levels of neurological integration.

These moderately prolonged comas administered at intervals of several days have resulted in improvement in patients whose psychoses were resistant to the ordinary full course of insulin treatment. The therapeutic value of the procedure must, however, await more extended study. We are gradually increasing the duration of coma in successive trials. Further modifications in the technique may become necessary. Since numerous cases of accidentally induced protracted coma have amply demonstrated its therapeutic effectiveness,[1] we hope that a single prolonged coma of one or more days, of the controlled type described above, may prove to be the equivalent of long periods of ordinary treatment. At any rate, it can be reasonably anticipated that this modification of the so-called insulin shock treatment will shorten its duration, simplify its management and still further extend its range of usefulness.

13489

Presence of Estrogenic Hormone(s) in Testicular Material.

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Although a number of studies have been made on male urine to determine the presence or absence of estrogens, comparatively few have been made upon testes. This is rather surprising in the light of the work of Zondek[1] who reported enormous quantities of estrogenic substance (more recently identified as alpha-estradiol and estrone by Beall[2]) in the testes of the stallion.

Zondek,[1] while more often credited with the report on the stallion testes, examined also the bull testes. Perhaps because of the relatively minute amounts (less than 0.1%) found in the latter as compared to the former it was not thought to be worth recording in the later literature. Zondek assumed that "the mass production and excretion of the estrogenic hormones in the testes is a peculiarity of the equines."

1 Zondek, B., *Nature*, 1934, **133**, 209.

2 Beall, D., *Bioch. J.*, 1940, **34**, 1293.

There is considerable difference of opinion among those who have examined testes for estrogens. A number of workers have failed to secure any results from their extracts, but on the other hand sufficient data have been recorded to indicate that estrogens occur in the testes of several species. Fellner,[3] Brouha and Simonnet,[4] Dorfman, *et al.*,[5] all reported the presence of estrogens in bull testes, while Laqueur and deJongh[6] report them in human testes and Dodds, *et al.*,[7] in "mammalian" testes. With the exception of Zondek all of the workers who have found estrogens in the testes have reported only qualitative results.

The desirability of further quantitative study is evident, but any quantitative study based upon extraction and bio-assay is bound to be more or less inaccurate. About the most one can say is that there is at least as much hormone as the assay indicates.

During the course of this study we were fortunate enough to secure two lots of fresh deer testes. These were prepared and assayed in the same manner as the bull testes. We thus had 4 preparations as follows:

Extract	I:	2103 g of frozen bull testes.
"	II:	2119 " " " " "
"	III:	671 " " fresh deer "
"	IV:	531 " " " " "

The fresh or frozen material was extracted according to the Gallagher and Koch[8] method. Both androgens and estrogens were present in the crude extract and they were separated by alkali fractionation according to the Gallagher, Koch, and Dorfman[9] procedure. The alkali-estrogen portion was neutralized with HCl and again fractioned with ether. The ether was evaporated and the residue taken up in olive oil.

Ovariectomized rats were used for assay with the following results:

EXTRACT I.

1 rat	given	equiv.	of	400	g	bull	testes,	positive
1 "	"	"	"	300	"	"	"	"
4 "	"	"	"	200	"	"	"	all positive
2 "	"	"	"	100	"	"	"	both negative

[3] Fellner, O. O. *Pflugers Arch.*, 1921, **189**, 199.

[4] Brouha, L., and Simonnet, H., *C. R. Soc. Biol.*, 1928, **99**, 41.

[5] Dorfman, R. I., Gallagher, T. F., and Koch, F. C., *Endocr.*, 1935, **19**, 33.

[6] Laquer, E., and de Jongh, S. E., *J. A. Med. Assn.*, 1928, **91**, 1169.

[7] Dodds, E. C., Greenwood, A. S., and Gallimore, E. J., *Lancet*, 1930, **228**, 683.

[8] Gallagher, T. F., and Koch, F. C., *J. Biol. Chem.*, 1929, **84**, 495.

[9] Gallagher, T. F., Koch, F. C., and Dorfman, R. I., PROC. SOC. EXP. BIOL. AND MED., 1935-36, **33**, 440.

EXTRACT II.

1 " " " " 636 g bull testes, positive
3 " " " " 424 " " " all positive
1 " " " " 212 " " " negative

EXTRACT III.

2 " " " " 122 g deer testes, both positive
3 " " " " 61 " " " 1 neg., 2 positive
3 " " " " 30 " " " all negative

EXTRACT IV.

4 " " " " 106 g deer testes, 1 neg., 3 positive

The shortage of material prevented a closer determination of the minimal effective dosage, but any correction would indicate a greater concentration of estrogens in the testes than shown herein.

It is to be noted that the second extracts of both bull and deer testes are far less potent than the first extracts. This may be due in part to the fact that in the second extracts a much larger lipoidal content was encountered. Efforts to free the hormones from the lipoid by the Thayer, Jordan and Doisy[10] procedure may not have been fully effective in our hands. We are at a loss to explain the presence of this large lipoid content since all the coverings with attached fat were removed from the testes prior to extraction.

The data confirm the observations of Fellner, Brouha and Simonnet, Zondek, and Dorfman, *et al.*, that estrogens occur in the bull testes, and the quantitative results on Extract I agree remarkably well with those of Zondek.

We have demonstrated for the first time, we believe, that estrogens occur in fresh deer testes and that they have a potency about 3 times as great per gram of tissue as those that occur in frozen bull testes.

13490

Action of Urea and Ether on Agent and Complement-fixing Antigen of *Lymphogranuloma venereum.*

CLARA NIGG. (Introduced by Geoffrey Rake.)

From the Biological Laboratories of E. R. Squibb & Sons, New Brunswick, N.J.

That the complement fixation reaction is a valuable aid in the diagnosis of *Lymphogranuloma venereum* has been firmly established.[1-3] Two antigens were found to be satisfactory, viz., (a) the

[10] Thayer, S. A., Jordan, C. N., and Doisy, E. A., *J. Biol. Chem.*, 1928, **79**, 53.