

cal factors control the oxygen combining power of hemoglobin. The K of the Hill-Barcroft equation is known to vary, however, with different bloods at identical temperatures and hydrogen ion concentrations.<sup>7</sup> These fluctuations may be wide and of importance in pathological states.

*Conclusions.* 1. A modified aerotonometer method for determining the partial pressure of oxygen can be applied to blood drawn from a human artery and is reasonably accurate. 2. In patients thought to be normal a certain variation obtains. 3. Partial pressure results which are high in relation to the oxygen saturation are commonly seen in the presence of anoxemia.

## 8461 C

### Intravaginal Assay of Urinary Estrin.

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Since estrin is not commonly standardized on the basis of its ability to produce behavioristic estrus, but rather by virtue of its growth-promoting influence on the vaginal epithelium, there seems but little need to saturate the entire animal to gain this end. The vaginal epithelium reacts to extremely minute quantities of estrin applied locally,\* although Pratt and Smeltzer<sup>1</sup> and Powers, *et al.*,<sup>2</sup> found their methods of vaginal administration only one-third to one-half as efficacious as the subcutaneous.

We have used the intravaginal method of assaying urinary estrin not only because minute amounts may be detected (even in male urine), but also to avoid the toxic effects produced by parenteral injection of crude extracts. Twenty-four-hour urines were obtained from 4 women† on days 7, 14, 21, and 28 of the menstrual

<sup>7</sup> Judd, E. S., Snell, A. M., and Hoerner, M. T., *J. A. M. A.*, 1935, **105**, 1653.

\*Cornification of the vaginal epithelium (without uterine growth) may be produced by vaginal administration of 1/200th of a rat unit of "Progynon" (kindly supplied by the Schering Corporation, New York).

<sup>1</sup> Pratt, J. P., and Smeltzer, M., *Endocrinol.*, 1929, **13**, 320.

<sup>2</sup> Powers, H. H., Varley, J. R., and Morrell, J. A., *Endocrinol.*, 1929, **13**, 395.

† We are indebted to the Nursing Department of the Providence Hospital, Oakland, California, for obtaining urine samples from 4 nulliparous nurses from 19 to 21 years of age. Their physical and dietary habits were as equal as hospital routine would allow. They had normal menstrual cycles varying from 25 to 29 days with the period of flow varying from 4-6 days. Cases 3 and 4 had acne vulgaris, otherwise no gross deviations from the normal were observed.

cycle, extracted immediately, or stored at 0°C. Half of the 24-hour sample was evaporated *in vacuo* over a steam bath to a salty sludge, (a few drops of capryl alcohol being added to lessen foaming). The sludge was extracted with two 100 cc. portions of hot absolute C<sub>2</sub>H<sub>5</sub>OH and the insoluble material discarded. The alcohol was distilled off, and the residue neutralized with N/1 or stronger NaOH. A graded series of dilutions of this extract was prepared with distilled water so that the 0.02 cc. used in testing represented original urinary volumes of from 0.05 to 1.0 cc. 0.01 cc. of each dilution was introduced twice (24-hour interval) into the vaginae of 3 full-grown rats ovariectomized at least 10 days previously.‡ The minimal effective urinary equivalent was considered the smallest amount causing cornification in the vaginae of at least 2 of the 3 rats, 24 hours after the second dose. After an interval of 5 days or more, the rats were used again.

TABLE I.  
Vaginal Units of Estrin Excreted Daily by Women.

| Case | Day of Cycle | 24 hr.<br>Urine<br>cc. | Minimal Effective<br>Urine Equivalent<br>cc. | "Vaginal" Units<br>per 24 hour | Units<br>per litre |
|------|--------------|------------------------|----------------------------------------------|--------------------------------|--------------------|
| 1    | 7            | 1300                   | 0.8                                          | 1620                           | 1250               |
| "    | 14           | 600                    | 0.4                                          | 1500                           | 2500               |
| "    | 21           | 1680                   | 0.4                                          | 4200                           | 2500               |
| "    | 28           | 1700                   | 0.4                                          | 4250                           | 2500               |
| 2    | 7            | 1200                   | 0.4                                          | 3000                           | 2500               |
| "    | 14           | 1000                   | 0.1                                          | 10000                          | 10000              |
| "    | 21           | 960                    | 0.2                                          | 4800                           | 5000               |
| "    | 28           | 930                    | 0.6                                          | 1550                           | 1666               |
| 3    | 7            | 950                    | 0.4                                          | 2375                           | 2500               |
| "    | 14           | 1300                   | 0.4                                          | 3250                           | 2500               |
| "    | 21           | 1160                   | 0.4                                          | 2900                           | 2500               |
| "    | 28           | 1140                   | 0.6                                          | 1900                           | 1666               |
| 4    | 7            | 1920                   | 0.4                                          | 4800                           | 2500               |
| "    | 14           | 1700                   | 0.1                                          | 17000                          | 10000              |
| "    | 21           | 1930                   | 0.2                                          | 9650                           | 5000               |
| "    | 28           | 2000                   | 0.4                                          | 5000                           | 5000               |

Table I contains the figures for the number of minimal effective doses of estrin per litre, as well as the daily estimates. They show that one may detect a rat vaginal-cornification unit in from 0.1 to

‡ A Breed and Brew milk pipette (U. S. Public Health Association Standard) calibrated to deliver 0.01 cc. milk was used. A preliminary swabbing of the vaginal mucosa with dry cotton allows for better retention of the extract. If the tip of the pipette is at the cervix, and if the pipette is rotated in the vagina during and after the administration, a more even distribution of the hormone is ensured, and the likelihood of reflux minimized.

0.8 cc. of a normal woman's urine. The 2 samples showing the highest unitage happened to have been taken in the mid-interval and therefore when a ripened or recently ruptured Graafian follicle may have been influential.

### 8462 C

#### Reactions of Mammary Glands of Normal and Hypophysectomized Male Guinea Pigs to Female Sex Hormone.

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It has become increasingly important during these last few years of intensive experimentation on the hormonal control of the mammary apparatus to discriminate between the respective rôles played by the primary female sex hormone (estrin) and the anterior pituitary mammary-stimulating hormone (mammatropin), in inducing growth and secretory activity of the breast.

Several years ago in the course of experiments on the effects of purified gonadotropic hormone in hypophysectomized rats<sup>1</sup> it was noticed that although the female genital tract responded to the hormones of the stimulated ovaries, the mammary apparatus showed only a poorly-developed duct system. These findings pointed to a greater importance of the anterior pituitary as a source of a mammary growth-promoting factor than is admitted by some investigators. To further determine the degree of mammary stimulation possible through estrin, it was decided to use hypophysectomized male guinea pigs, since Laqueur and others<sup>2</sup> have shown that the intact male guinea pig responds to estrin treatment by mammary growth and lactation. Asdell and Seidenstein<sup>3</sup> found the development of the mammary glands of ovariectomized, hypophysectomized rabbits injected daily for 15 days with 25 rat units of estrin plus 4 rabbit units of progestin almost the same as ovariectomized, non-hypophysectomized rabbits similarly treated. Apparently they did not study the effect of estrin alone.

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<sup>1</sup> Evans, H. M., Pencharz, R. I., and Simpson, M. E. Unpublished observation.

<sup>2</sup> Laqueur, E., Borchardt, E., Dingemanse, E., and De Jongh, S. E., *Deut. med. Wchnschr.*, 1928, **54**, 465.

<sup>3</sup> Asdell, S. A., and Seidenstein, H. R., *PROC. SOC. EXP. BIOL. AND MED.*, 1935, **32**, 931.