

A large percentage of the guinea pigs which received the oestrin injections exhibited uteri in which the cellular and glandular picture was identical with that found in human hyperplasia. The glands, many of which were cystic, were very prominent throughout the greatly thickened mucosa. Figure 1 shows that these specimens were quite similar to the typical Swiss-cheese pattern found in human cases. As a rule, this picture was found in the uteri of guinea pigs which had received from 10 to 50 units of oestrin daily for 8 to 15 days.

In guinea pigs it was possible to induce both the glandular and cellular change of hyperplasia of the endometrium in all of the animals injected. In rats, the cellular changes were easily induced, but we were able to produce cystic dilatation of the glands in only 40% of the animals studied.

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Bactericidal Property of an Ultra-Violet Irradiated Petrolatum-Lanolin Mixture.

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Remarkable results on purulent wounds following the use of ultra-violet irradiated petrolatum, reported by Eising,¹ prompted us to determine whether such petrolatum had any bactericidal effect *in vitro*. We used a mixture of 2 parts petrolatum molle and one part lanolin, the latter being added to increase tissue absorption of the irradiated petrolatum.

Technic. A 2:1 petrolatum-lanolin mixture was melted and rendered sterile by infra-red heat. Twenty cc. of the mixture were placed in sterile petri dishes and irradiated directly for varying periods of time by a Burdick ultra-violet lamp at a distance of 12 inches. At the end of the time allotted for exposure 1 cc. of a normal saline suspension of organisms was thoroughly mixed with the petrolatum-lanolin. At varying intervals thereafter one loopful of the inoculated mixture was seeded on agar plates and the growth noted. For control we used: (1) Non-irradiated petrolatum-lanolin to which organisms had been added; (2) Petrolatum-lanolin to which organisms had been added before irradiation; (3) Petrola-

¹ Eising; E. H., *Annals Surg.*, 1931, **43**, 123.

tum-lanolin which was irradiated for the same varying periods of time as the directly irradiated samples but with the petri dish covers left on to filter out the shorter ultra-violet rays as much as possible. The organisms used were *Staphylococcus aureus* and *Bacillus pyocyaneus*.

Staph. aureus was practically always killed within 24 hours by a 2-hour directly irradiated mixture. *B. pyocyaneus* gave less consistent results, but was usually killed within 24 hours by the 4-hour directly irradiated mixture. The plates irradiated with their covers on showed no bactericidal property except those exposed for 4 hours, which consistently killed *Staph. aureus* within 24 hours, thus suggesting that over the prolonged exposure time sufficient rays passed through the glass to irradiate the mixture. The non-irradiated controls gave abundant growth, while those to which organisms had been added before irradiation gave no growth.

Time and facilities allowed only one test to determine how long the irradiated mixture could be effective, and this test showed that a 4-hour directly irradiated mixture one week old killed *Staph. aureus* and *B. pyocyaneus* within 24 hours.

That the effect of irradiation is bactericidal rather than bacteriostatic was shown by making broth cultures from the irradiated mixtures which had given no growth on agar and also from the non-irradiated controls. Warm broth was used on the theory that the grease would be melted, freeing the bacteria. Here again the non-irradiated controls gave growth while the irradiated mixtures did not.

We can only surmise the nature of the bactericidal agent in the irradiated petrolatum-lanolin. Montgomery² has shown that irradiated ergosterol is not bactericidal; hence the ergosterol in the lanolin of our mixture can be excluded as an active agent. Our non-irradiated controls show that the infra-red used in sterilization of the mixture plays no rôle. The fact that irradiation turns colorless petrolatum yellow suggests a change in molecular arrangement of that substance, but it is doubtful if such a chemical change renders the petrolatum capable of killing bacteria. A more appealing theory is that secondary ultra-violet emanations are held by the petrolatum-lanolin. The ultra-violet spectrum is between 1850 and 4000 Angstrom units. It is plausible to suppose that the secondary rays retained by the petrolatum-lanolin mixture are similar to the longer rays of the biologically active or so-called "vital" portion of the ultra-violet spectrum. This view is supported by the results of

² Montgomery, B. E., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 481.

Eising, who produced stencil designs on hermetically sealed X-ray films placed over irradiated petrolatum in a light-tight box, and by French.³

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Further Purification of the Adrenal Cortical Hormone.*

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In studying the problem of further purification of the adrenal cortical hormone we have used the following 3 types of fractionation procedure on the active material obtained from whole beef adrenal glands by the usual permutit fractionation: (1) distribution between an immiscible solvent and aqueous alkali, (2) distribution between an immiscible solvent and aqueous acid, and (3) fractionation with organic solvent mixtures. The starting material and the various fractions obtained were assayed on adrenalectomized dogs by the technique previously described.¹

Four hundred and seventy mg. of alcohol-soluble fraction obtained from 4000 gm. of beef adrenal glands and containing 8000 D. U. (dog units) were dissolved in ether and washed with 0.05 N NaOH. The aqueous alkaline solution was washed with fresh ether and the ether solutions combined. The ether-soluble fraction (200 mg.) was transferred to water (70 mg. water-soluble) for assay and was found to contain less than 500 D. U. The alkaline washings were adjusted to pH 5.6 and on assay contained between 500 and 1000 D. U. In this manipulation, therefore, about 6500 D. U. were apparently destroyed.

The above procedure was repeated on another aliquot of 470 mg. containing 8000 D. U. except that 0.05 N HCl was used instead of alkali. The ether soluble fraction (350 mg.) contained between 500 and 1000 D. U., while the aqueous acid washings after adjustment to pH 5.2 assayed between 6000 and 7000 D. U. The

³ French, F. S., *Clifton Med. Bul.*, 1932, **18**, 21.

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¹ Harrop, G. A., Pffner, J. J., Weinstein, A., and Swingle, W. W., *Proc. Soc. Exp. Biol. and Med.*, 1932, **29**, 449.