

cies and of *Carica papaya* are to be attributed to the proteinases of these plants. However, there is some evidence on hand that certain other plant proteinases which are as proteolytically active as papain do not possess this peculiar ability to digest live worms.

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### CONVERSION OF ESTRADIOL TO ESTRONE IN VIVO<sup>1,2</sup>

WESTERFELD and Doisy measured the estrogenic activity of the phenolic fraction of the urine of monkeys injected with estradiol.<sup>3</sup> They found that 30 to 45 per cent. of the activity of the excreted estrogens was present in the ketonic fraction. After the administration of estradiol to estrous, hysterectomized estrous, or pregnant rabbits, Pincus showed the possible presence of estrone in the urine.<sup>4</sup> He was unable, however, to find any evidence for estrone in the urine of ovariectomized animals injected with estradiol. Neither the work of Westerfeld and Doisy nor that of Pincus was supported by chemical isolation and identification of metabolic products. This communication deals with the isolation and identification of estrone from the urine of long-time ovariectomized guinea pigs to which estradiol had been administered.

Ten mg of estradiol dipropionate in oil solution were administered by subcutaneous injection daily for 5 days to each of 5 adult guinea pigs which had been ovariectomized for at least one year. The urine was quantitatively collected during the injection period and for the following 5 days. After acidification with 10 per cent. of concentrated hydrochloric acid, the mixture was refluxed for 15 minutes and thoroughly extracted with benzene. That portion of the extract representing the phenolic compounds was separated by the Girard-Sandulesco reagent into ketonic and non-ketonic fractions.

The ketonic fraction contained more than 50,000 I. U. of estrogenic material when assayed by the vaginal smear test on the spayed adult mouse. The ketonic substances were subjected to high vacuum sublimation and the fraction subliming at 150° C. and 3 micra of mercury was collected. This semicrystalline

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<sup>1</sup> This work was supported by grants from the Committee for Research in Problems of Sex, National Research Council: Grant administered by Dr. William C. Young; by the Rockefeller Foundation, and by the Fluid Research Fund of Yale University School of Medicine.

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<sup>3</sup> W. W. Westerfeld and E. A. Doisy, *Ann. Int. Med.*, 11: 267, 1937.

<sup>4</sup> G. Pincus, *Cold Spring Harbor Symposia on Quant. Biol.*, 5: 44, 1937.

material was crystallized from methanol and a crop of approximately 5 mg of crystals, m. p. 242–245° C., was recovered. After recrystallization from methanol, the melting-point was raised to 245–246° C. The melting-point of a mixture of this compound with an authentic sample of estrone (m. p. 255–258° C.) was 247–249° C. The benzoate melted at 211–214° C. When mixed with a sample of estrone benzoate (m. p. 215–217° C.), the melting-point was 211–214° C. All melting-points are uncorrected.

In a second experiment 50 mg of estradiol dipropionate were administered orally for 2 days to each of 5 adult guinea pigs spayed for at least one year. A procedure similar to that outlined above was used with the following modification: The phenolic compounds were fractionated between 0.1 N sodium hydroxide and 10 per cent. sodium hydroxide and the material soluble in the latter solvent separated into ketonic and non-ketonic portions by the Girard-Sandulesco reagent. About 12 mg of crystalline estrone, m. p. 256–257° C., were obtained from the ketonic fraction without resorting to high vacuum sublimation. It did not depress the melting-point of an authentic sample of estrone.

Thus it appears from this work that, at least in the guinea pig, estradiol may be converted to estrone even in the absence of the ovary. This and further work on the metabolism of the estrogenic hormones will be reported in detail.

We are indebted to Ciba Pharmaceutical Products, Inc., for the supply of estradiol dipropionate and estrone benzoate, and to the Schering Corporation for estrone.

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### TOMATO POMACE IN THE DIET

TOMATO pomace is the term applied to the dried residues that remain after the preparation of tomato juice. These residues contain the seeds, skin and some of the original pulp. The composition of the material<sup>1</sup> used in our studies was the following: Protein, 24 per cent.; ether soluble, 14 per cent.; fiber, 33 per cent.; ash, 4 per cent., and moisture, 7 per cent. An analysis for pectin by Z. I. Kertesz, of Geneva, N. Y., showed 3.8 per cent. of this substance.

Three properties of this tomato pomace attracted our attention while improved feeding mixtures were being developed for dogs, foxes and minks. A sample of tomato pomace was ground for a rat assay and for a carotene determination. After these were made the material was left in a mason jar without a rubber for a year in a warm laboratory. At the end of this time

<sup>1</sup> C. M. McCay, "The Nutritional Requirements of Dogs," p. 27. Ithaca, 1939.