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Estrogenic Effects of 18-Norestradiol -17β and 18-Norestrone. (26085)

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Several workers have studied the effects of structural modification on biological actions and effectiveness of phenolic estrogens. Huggins and Jensen have discussed significance of carbon 17 hydroxyl groups of estradiol and of various modifications, related particularly to oxygenation at carbon 6 or carbon 16 (1,2); they showed that 17-desoxyestradiol was an active estrogen, albeit a material of low potency, and that 6 and 16 oxygenation resulted in estrogens that had shallow dose-response curves, *i.e.*, increment of uterine growth per unit increase in dose was less for these *impeded* estrogens than for estrone or estradiol. In mouse studies it was shown that there may be a whole series of possible dose-response curve slopes, extending from steep estrone slope, through intermediate slope of estradiol-17β, to shallow slope of estriol(3); similar results were obtained from spayed rats(4,5) and Drill, Cook and Edgren(6) have discussed several miscellaneous series of steroids; their finding most germane to present discussion was that 16-halogenation of 3-methoxyestrone does not produce an *impeded* or shallow slope. Most of these structural modifications have involved addition, removal or transposition of oxygenated functions. Recently synthesis of 18-norestrone and 18-norestradiol-17β has been completed(7). The uterine growth stimulating effects of these materials will be discussed here.

Materials and methods. Technics employed here have been described previously. Uterine growth was determined from tests employing immature female mice that received test compound daily for 3 days and were sacrificed on fourth. At autopsy uteri were removed, cleaned of adherent tissues and weighed wet on a torsion balance(3).

Results and discussion. As discussed previously(3,4), estrone had steepest slope of compounds treated; remaining materials had slopes that were essentially similar (Fig. 1). Slopes for estradiol-17β and estrone were appreciably more shallow than those cited earlier(3); this probably is result of change in mouse strain between the two studies (unknown strain from small breeder to Carworth mice from Small Animal Farms, Des Plaines, Ill.). The extremely high potency for estradiol-17β is the result of slope of dose-response curve and low response level at which 2 materials were compared; at 50 mg uterine weight level estrone and estradiol-17β would be equal in potency. Removal of angular methyl group at carbon 13 results in a sharp decrease in potency for each compound (Table I) with the estradiol derivative about

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TABLE I. Dose Response Curves and Potency for Estrone, Estradiol-17β, 18-Norestrone and 18-Norestradiol-17β.

Compound	Curve	Potency*
Estrone	Uterine wt = $-.2 + 30.5$ (log 100 dose)	100%
18-norestrone	" " = $5.9 + 11.6$ (log dose)	.028
Estradiol-17β	" " = $17.4 + 12.6$ (log 1000 dose)	2500
18-norestradiol-17β	" " = $8.3 + 13.0$ (log dose)	.058

* At response equivalent to about 100% increase in uterine wt over oil-treated controls.

twice as potent as the estrone. The more remarkable finding, however, is decrease in slope of dose-response curve for 18-norestrone, which was similar to that for estradiol-17β. Why such structural modifications as removal of angular methyl group at carbon 13 or incorporation of oxygenated functions at carbon 6 or carbon 16 should produce decreases in slope of dose-response curve is unclear at present time.

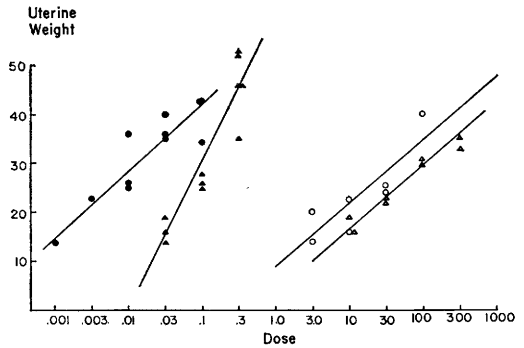

FIG. 1. Effects of estrone (▲), estradiol-17β (●), 18-norestrone (△) and 18-norestradiol-17β (○) on uterine growth in immature intact female mice. Each point represents one mean of 8-10 mice. Curves were fitted by the method of least squares.

Summary. Removal of the angular methyl group of estradiol-17β to form 18-norestradiol-17β results in a decrease in potency. A similar structural modification of the estrone molecule, forming 18-norestrone, produces both potency decreases and alterations in slope of the dose-response curve.

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Absence of Sialic Acids and Other Carbohydrates in Crystalline Papain.* (26086)

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Studies from this laboratory have been concerned for some years with the enzymic activity and structure of crystalline papain (1). Studies of the composition of this protein by Smith, Stockell and Kimmel(2) indicated that non-amino acid constituents were probably absent since the recovery of amino acids accounted satisfactorily for the weight and nitrogen content of the protein. Moreover, both Close, Moore and Bigwood(3) and Smith *et al.*(2) could detect no carbohydrate by the carbazole method. Recently, Loo, Lee and Flemming(4) reported that crystalline papain contains sialic acid by the direct Ehrlich method. It would be surprising indeed if sialic acid were present in a protein which

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