Microbial Oxidation of Ecdysones. A Convenient Preparation of Rubrosterone

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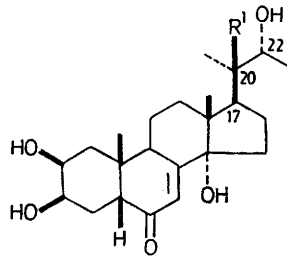
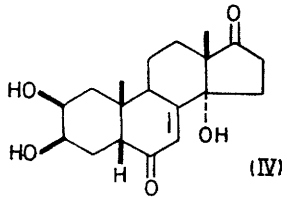
Summary Ponasterone A (I) is converted by the micro-organism *Fusarium lini* ATCC 9593 into rubrosterone (IV) in 15% yield.

ALTHOUGH microbial oxidations of numerous steroid side-chains are well documented, there have been very few such investigations reported on ecdysones, insect moulting hormones. The recent communication by Canonica and his co-workers[1] prompts us to report our observations on microbial oxidations of ecdysones by another micro-organism. We report that, among *ca.* 30 micro-organisms so far screened, *Fusarium lini* ATCC 9593 oxidatively catabolizes the 20*R*,22*R*-dihydroxy-side-chain in ponasterone A (I) to yield rubrosterone (IV) efficiently (*ca.* 15%). Although rubrosterone has been isolated from the plants,[2] its predicted occurrence as a metabolite of ecdysones in *Arthropoda* or *Crustacea* has never been demonstrated, and so its biological role remains to be appraised.

F. lini ATCC 9593 was cultured[3] in a medium containing 2% corn steep liquor and 1% glucose. Ponasterone A was incubated by shaking at room temperature in the 2-day-old culture for 48 h. Extraction of the culture medium with n-butanol revealed that ponasterone A was completely degraded. Purification of the metabolite by silica gel



(I) R^1 = OH, R^2 = H
(II) R^1 = H, R^2 = OH
(III) R^1 = OH, R^2 = OH



(IV)

column chromatography, preparative t.l.c. (HF_{254}, Merck), and recrystallization (MeOH–EtOAc) gave rubrosterone,[2]† m.p. 235—237°, M^+, m/e 334. Similar incubation of β-ecdysone (III) for 96 h also gave rubrosterone, but conversion was much less efficient (<5%); α-ecdysone (II) produced no rubrosterone.

Since ponasterone A is available in large quantities from certain *Podocarpaceae* and *Taxaseae* species (*e.g.*, 0·2% and 0·04% from *Podocarpus nakaii* Hay[4] and *P. macrophyllus* D. Don[5] leaves, respectively) the present microbiological conversion provides an alternative and convenient means for the preparation of rubrosterone.[6]

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† The identity was validated by comparison with the authentic compound. We thank Professor H. Hikino for a gift of rubrosterone.

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