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Separation of 3-Chloro-4,6-dimethoxysalicylic Acid from the Urine of Rabbit administered Griseofulvin

Griseofulvin, the antifungal antibiotic agent isolated from *Penicillium griseofulvum*,[1] *P. janczewskii* (=*P. nigricans*),[2] and *P. patulum*,[3] has proved to be effective by oral administration in the treatment of certain superficial mycoses. However, metabolic fate of griseofulvin in the animal body has not yet been determined.

In the present series of work, 3-chloro-4,6-dimethoxysalicylic acid was isolated from the urine of rabbit, infected with *Tricophyton asteroides* on the back, and administered orally with griseofulvin. To separate the metabolite from the urine, the method of Kamil, Smith, and Williams[4] was used.

The collected urine was filtered through cotton, the filtrate was brought to about pH 4 with glacial acetic acid, and then treated with saturated aqueous lead acetate solution until precipitation was complete. The precipitate was discarded by filtration. The filtrate was brought to about pH 8 with ammonia and saturated aqueous basic lead acetate solution was added in excess. The basic lead that precipitate was collected by filtration, washed with water, made into a fine suspension in 50% ethanol, and lead removed by saturation with hydrogen sulfide. Lead sulfide was filtered off and the filtrate was evaporated to dryness *in vacuo* at 45°. The residue was dissolved in a small volume of water and extracted with ether. The ether solution was shaken repeatedly with 5% sodium hydrogencarbonate solution. The hydrogencarbonate solution was acidified, reëxtracted with ether, the ether residue was purified by sublimation under a reduced pressure, and the sublimate was crystallized from ethyl acetate to colorless needles, m.p. 215~220° (*Anal.* Calcd. for $C_9H_9O_5Cl$ (3-Chloro-4,6-dimethoxysalicylic acid) : C, 46.45; H, 3.91. Found : C, 46.58; H, 4.02. IR ν_{max}^{KBr} cm^{-1} : 3225, 1692, 1220, 806).

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