Microbial Formation of Comenic acid from Kojic acid

Sir:

During the course of study on the utilization of kojic acid by microorganisms, it was found by the authors that comenic acid was directly formed from kojic acid by using a bacterium which was isolated from soil.

Since up to now comenic acid has not been known to be directly prepared from kojic acid neither by chemical nor by biochemical methods, consequently little knowledge has been obtained on the oxidation products of kojic acid at the early stage, the formation of comenic acid from kojic acid may be of particular interest on the metabolism of kojic acid.

The bacterium was grown on the bouillon agar medium containing 0.2% of kojic acid at 30°C for a few days. After being harvested and washed, the cells were suspended in distilled water. The cell suspension was added to a reaction mixture containing M/30 phosphate buffer (pH 7.0) and M/20 kojic acid. After the incubation on a shaker at 30°C for 70 hours, comenic acid was isolated from the reaction mixture by the following procedure: the reaction mixture was clarified by centrifuge and was neutralized. The neutralized mixture was

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**Fig. 1-a. Infrared Absorption Spectrum of (I.)**

**Fig. 1-b. Infrared Absorption Spectra of (III, IV.)**
concentrated in vacuo and remaining kojic acid was separated from the mixture by extracting with ether. The mixture was further acidified with sulfuric acid (pH 2) and was carried out the extraction with ether for 80 hours. The ether extract was concentrated in vacuo and the crude crystal obtained was recrystallized from hot water. The crystal (I) was faintly brownish coloured and small plates, which decomposed at 260–261°C. (I) gave a characteristic colour reaction with ferric chloride solution and showed the $R_F$ value of 0.48 on the paper chromatograph with a solvent system consisting of 1 volume of $n$-amylalcohol and 1 volume of 5$n$-formic acid.

Anal. Found: C, 45.75; H, 2.67. Calcd. for C$_6$H$_4$O$_5$: C, 46.15; H, 2.58.

The methyl ether (II) of (I) was prepared by Yabuta's procedure$^{1)},$ and it was converted to the corresponding ethyl ester (III). (III) melted at 155.5°C and no depression of the melting point was observed when mixed with the authentic ester (IV) which is the ethyl ester of comenic acid methyl ether, synthesized according to Yabuta$^{2)}$ from kojic acid.

Anal. Found: C, 54.22; H, 5.02. Calcd. for C$_9$H$_{10}$O$_5$: C, 54.54; H, 5.02.

The ultraviolet and infrared absorption spectra of the compounds (I, III, IV) are shown in Fig. 1 and 2.

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