

## SHORT REPORTS

### SACCHAROPINE FROM TOBACCO LEAVES

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(Received 3 October 1977)

**Key Word Index**—*Nicotiana tabacum*; Solanaceae; tobacco leaf; saccharopine.

In the course of a study of oligopeptides in tobacco leaves, several constituents which gave more than two ninhydrin-positive compounds by  $\text{KMnO}_4$  oxidation were observed. One of these was isolated by sequential chromatography on Sephadex G-10, anion exchange resin and cellulose powder, and was identified as *N*-(2-glutaryl)-L-lysine (saccharopine). It has been reported that saccharopine is concerned with lysine metabolism in both yeast [1, 2] and animal tissue, but not in higher plants which are known to have the alternative diaminopimelate pathway. However, saccharopine was isolated from buckwheat seeds and its possible participation in lysine metabolism in higher plant was suggested [3]. The present finding suggests this compound may be more widely distributed in plants and may be of importance in relation to lysine metabolism.

#### EXPERIMENTAL

Ca 60 kg of fr. tobacco leaves (*N. tabacum*, BY-4) was extracted with 200 l. of 70% MeOH. After conc, metal ions and neutral substances were removed with Dowex 50,  $\text{H}^+$ . The crude amino acid fraction (400 ml) was applied on a Sephadex G-10 (10 × 150 cm) and eluted with  $\text{H}_2\text{O}$ . Saccharopine, as well as many peptides, was eluted just before glutamic acid. The whole peptide

fraction was applied to a column of Dowex 1 (AcOH form, 2.5 × 40 cm). After washing thoroughly with  $\text{H}_2\text{O}$ , the absorbed compounds were fractionated by stepwise elution with 0.1, 0.3, 1 and 2 M HOAc. The eluate with 0.3 M HOAc was collected, concd (4 ml) and applied to a cellulose column (Avicel, 2.5 × 90 cm) and eluted with  $\text{BuOH-HOAc-H}_2\text{O}$  (12:3:5) at room temp. The fraction containing saccharopine (1–1.1 l.) was evapd to dryness at 45° *in vacuo* and dissolved into a small amount of hot  $\text{H}_2\text{O}$ . After keeping overnight at room temp., colorless needles appeared which were recrystallized 3 × giving, 40 mg. Anal. (Found: C, 46.82, H, 7.67, N, 9.92. Calcd. for  $\text{C}_{11}\text{H}_{20}\text{N}_2\text{O}_6$ : C, 47.82, H, 7.24, N, 10.14 %); its parent peak in HDMS,  $m/e$  277 ( $\text{M}^+$ , +1). PMR  $\delta_{\text{HDO}}^{0.2\text{NDCl}}$  4.0 (2H, *tt*), 3.1 (2H, *t*), 2.5 (2H, *t*), 2.1 (2H, *m*), 1.5–1.8 (8H, *m*). CMR  $\delta_{\text{dioxane}}^{0.2\text{NDCl}}$  176.6 (s), 172.3 (s), 171.2 (s), 59.7 (d), 53.3 (d), 47.1 (t), 30.2 (t), 30.0 (t), 26.0 (t), 24.7 (t), 22.4 (t). The IR spectrum was identical with previous data [2–4]. Mp 257–259°, decomp.  $[\alpha]_{\text{D}}^{23} +31.4^\circ$  (c 1 in 0.5 N HCl).

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### METABOLISM OF 5-ACETOAMINO-2-HYDROXYVALERIC ACID IN TOBACCO LEAVES

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(Received 6 October 1977)

**Key Word Index**—*Nicotiana tabacum*; Solanaceae; tobacco leaf; 5-acetoamino-2-hydroxyvaleric acid;  $\beta$ -acetylornithine; metabolism.

**Abstract**—5-Acetoamino-2-hydroxyvaleric acid, (5-AHV) was metabolized to  $\delta$ -acetylornithine in tobacco leaves. On the other hand,  $\delta$ -acetylornithine fed to tobacco leaves was metabolized into at least 5 components, one major component being 5-AHV. These results show that tobacco plant has a reversible metabolic pathway between 5-AHV and  $\delta$ -acetylornithine.

#### INTRODUCTION

Recently we isolated a new compound, 5-acetoamino-2-hydroxyvaleric acid (5-AHV) from green tobacco leaves

[1]. Preliminary experiments showed that not only young green leaves but also aged yellow leaves contain appreciable amounts of this compound. Since  $\alpha$ -hydroxy