was filtered off and suspended in 7.7 cc. of boiling water for 20 mins. Water was added to the boiling suspension until all of the solid had dissolved. On cooling, (IV) crystallized out. Yield, 0.374 g.(62.0%).

Orotic〔2-^{14}C〕 Acid (V)—0.374 g. of (IV) was dissolved in 12.5 cc. of 1*M* KOH and allowed to stand in a constant temperature bath at 64° for 2 hrs. On acidifying the solution with conc. HCl, orotic-〔2-^{14}C〕 acid·H_2O precipitated out and was recrystallized from water. Yield, 0.323 g.(77.4%) of m.p. 320°(decomp.). Specific activity of the product was 72,400 counts/min./micromole by Q-gas flow counter.

Non-labeled orotic acid, synthesized by the same foregoing method, gave the following analytical values: *Anal.* Calcd. for $C_5H_4O_4N_2 \cdot H_2O$: C, 34.46; H, 3.47; N, 16.15. Found: C, 34.70; H, 3.56; N, 16.03.

Summary

Ureido〔^{14}C〕succinic acid and orotic〔2-^{14}C〕 acid, which are metabolic intermediates for nucleic acid pyrimidines, were synthesized by an improved method of Nyc and Mitchell starting with aspartic acid and urea〔^{14}C〕, which is more easily available than potassium cyanate〔^{14}C〕. The yield of orotic〔2-^{14}C〕 acid was 15.7% based on urea〔^{14}C〕.

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Tatsuo Kariyone and Keiichiro Hayashi: Studies on the Microanalysis of Essential Oil Components. I. The Paper Chromatography of Terpene Alcohol by using Anthranilate.

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All but a few of the reports in the past few years on the application of paper chromatography of alcohols were of compounds with larger carbon number such as terpene alcohol, obtaining well-separated spots on paper, while all these methods resulted in fine chromatograms with lower alcohols.[1,2]

In the present experiment, paper chromatography of terpene alcohols was carried out, using anthranilates which reveals dark violet spots under ultraviolet light or yellow coloration by spraying Ehrlich's reagent and a few micrograms of sample can be readily detected. Since esters of anthranilic acid are distributed widely among plants, e.g. in grape juice or as a component of miscellaneous essential oils such as neroli-bigarade, jasmin, ylang-ylang, and sweet orange oils, and since the synthetic anthranilates are also used widely in perfumes, their detection is becoming more important in recent years.

The method of Verley[3] or Roviralta[4] was applied after minor improvement by the authors, in which the esters of corresponding alcohols were obtained in higher yield by ester-exchange reaction between methyl anthranilate and alcohols using metallic sodium or sodium hydroxide. While the reaction was complete in three hours in the cases of borneol, citronellol, menthol, benzyl, and decyl alcohol, the others, among them geraniol, had to be heated at least five or six hours. The sample mix-

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tures obtained in this way were spotted onto the starting line of a filter paper strip and then developed with butanol saturated with 1*N* acetic acid. Calculated Rf values were as follows :

Rf Value of Alcohol Anthranilate

Toyo Roshi No. 50, 2×40 cm. Developed with BuOH saturated with 1*N* AcOH.

Borneol	0.22	Linalool	0.08
Benzyl alcohol	0.11	Menthol	0.20
Citronellol	0.13	Methanol	0.36
Decanol	0.20	Nerol	0.42
Ethanol	0.42	Octanol	0.23
Geraniol	0.18		

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Experimental

A mixture of 2.7~3.0 g. of sample alcohol, 0.1 g. of metallic Na, and 2.5 g. of methyl anthranilate was boiled in an oil bath at 140~150°. The amount of sample should be adjusted in the case of essential oils which contain some amount of terpene alcohol, so as to correspond to the amount of alcohol contained in the reaction mixture. The reaction was continued for 3~5 hrs. when formation of methanol ceased. The residual methyl ester was neutralized with 10% H_2SO_4 and then upper layer in which sulfate of methyl anthranilate was dissolved was separated and removed. The lower layer was neutralized with 50% H_2SO_4 and the solution was diluted with 0.1*N* H_2SO_4, adjusting the concentration of anthranilate to approximately 0.1%. The yield of anthranilate amounted to 60~70% of its theoretical value.[5]

The sample provided for the purpose of paper chromatography was prepared through a simplified way in which the mixture in above-mentioned equivalents was heated in a small test tube with an air reflux condenser provided with a $CaCl_2$-tube. MeOH formed was removed by gentle suction through a water jet pump which was connected to a $CaCl_2$ tube. After the reaction was complete the whole reactant was dissolved in H_2SO_4 solution and used as a sample of paper chromatography.

Paper chromatography—Several kinds of developing solvent were examined and it was found that BuOH saturated with 1*N* AcOH showed excellent separation. The sample solution which contained several to 10 γ of anthranilate was spotted on the starting line which was drawn at 5 cm. distance from the end of a paper strip of 2×40 cm. Toyo Roshi No. 50. After developing it for 15 hrs. or more, the paper strip was removed and dried. The locations of spots of anthranilate were examined by the irradiation of ultraviolet light of 3,650 Å or by spraying Ehrlich's reagent, prepared with 3 vols. of 5% EtOH solution of *p*-dimethylaminobenzaldehyde and 1 vol. of conc. HCl.

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