Beroza is much lower in this procedure than in the chromotropic acid method⁸. This is due largely to the fact that with chromotropic acid the wave-length of maximum absorption of the glucose colour is the same as for piperine (580 mµ), whereas with the Labat reagent the glucose colour has maximum absorption at 540 mµ, which is appreciably removed from the absorption maximum (660 mµ) for the piperine colour.

This method, in conjunction with the development of colorimetric methods based on colour complexes formed by piperine with alkaloid reagents, and coloured salts formed with some of the concentrated acids4,9, can provide useful and easily executed alternatives to the methods now being used for the determination of the piperine content of pepper. The reagent as described is more stable than the chromotropic acid reagent which is usually prepared daily.

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Use of Methylene Blue for Staining Sialomucoproteins on Paper

For some time we have used in our laboratory a simple and sensitive method for the location of sialomucoproteins separated by electrophoresis on paper.

Whatman No. 3 $\dot{M}M$ paper strip (10 cm wide \times 34 cm long) was moistened with a volatile buffer solution such as pyridine-acetic acid-water (10:0.4:90, by volume) buffer of pH 6.5 (ref. 1) and then pressed between two sheets of filter paper to remove the excess of the buffer. The strip was then placed in an electrophoretic apparatus. Sample solution containing sialomucoprotein was applied at the origin. Electrophoresis was carried out in the buffer by the horizontal open strip method at 200 V for 7 h and platinum electrodes were used. After electrophoresis the strip was allowed to dry overnight at room temperature. The dry strip was immersed in a solution of methylene blue (100 mg of Merck's methylene blue 'B' was dissolved in 200 ml. of 90 per cent methanol) for 20 min, and excess dye was removed by washing with 90 per cent methanol until the strip had only a light blue tint. The strip was then dried at room temperature. The sialomucoprotein gave a blue, while acid mucopolysaccharides such as chondroitin, chondroitin sulphate, hyaluronic acid and heparin gave each a blue violet, metachromatic spot under the same conditions.

Methylene blue gave better results than toluidine blue and alcian blue for the staining of sialomucoproteins under the same conditions.

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¹ Ryle, A. P., Sanger, F., Smith, L. F., and Kitai, R., Biochem. J., 60, 541 (1955).

Portal Venous Transport of Free Pelargonic Acid following Intestinal Instillation of Tripelargonin

SATURATED odd-chain fatty acids such as valerate (C5) and pelargonate (C9) are metabolized differently from even-numbered saturated fatty acids in the same range of carbon chain length, caproate (C6) and caprylate $(C\overline{8})^{1-4}$. The three terminal carbons of the odd-chain acids appear to be metabolized as a unit, which can serve as a carbohydrate precursor, in contrast to the behaviour of the terminal two-carbon units of even-chain acids. Digestion of glycerides of odd-chain fatty acids and absorption and transport of the digestion products appear to have been little studied.

This communication describes experiments with tripelargonin suggesting that, in the dog, this triglyceride is hydrolysed in the gut with the resulting C9 acids transported in the portal vein in the free fatty acid (FFA) form.

Dogs were lightly anaesthetized with sodium pentobarbital and a polyethylene catheter was implanted in the portal vein. Thirty-five g tripelargonin homogenized with 20 ml. of a 2.5 per cent aqueous solution of sodium taurocholate was injected directly into the duodenal lumen. The pylorus was occluded by a non-traumatic clamp during the 5-min instillation period. After the injection the portal vein catheter was exteriorized and the abdomen closed. The catheter was kept open by means of a slow infusion of 0.9 per cent saline solution. Samples of blood were obtained from the portal vein before, and at frequent intervals for 4 h after, intraduodenal instillation of the glyceride. The lipids were extracted from each plasma sample and the lipid residue separated by thinlayer silicic acid chromatography into phospholipid, free cholesterol, FFA, triglyceride and cholesterol ester. The lipid esters and FFA were eluted with chloroform-methanol and the fatty acid moieties were identified by gas-liquid chromatography (GLC) following methylation. To provide a sufficient quantity of lipid ester for fatty acid analysis, the post-prandial samples were pooled by lipid ester class. Samples were run at both high (175° C) and low (100° C) column temperatures in order to obtain a more complete spectrum of fatty acids.

Results in one experiment are shown in Figs. 1 and 2. In Fig. 1 are shown the fatty acid patterns by GLC of the FFA from portal venous plasma in the post-absorptive state and 2 h after intraduodenal instillation of tripelargonin. Under both high and low column temperatures, post-absorptive portal plasma FFA did not contain any fatty acid below C14. However, in the post-prandial sample, pelargonic acid was readily demonstrated in the FFA fraction. In contrast, the lipid ester fractions from the post-prandial plasma did not contain any detectable amount of pelargonic acid under either high or low temperature column conditions (Fig. 2).

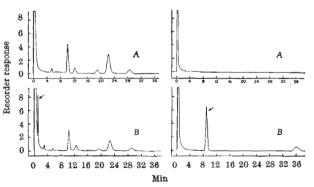


Fig. 1. Gas-liquid chromatograms of free fatty acids in canine portal venous plasma before (A) and 2 h after intraduodenal instillation of tripelargonin (B). Pelargonic acid is readily identified (arrow) in the post-prandial plasma at both high (left) and low (right) column tem-peratures