

A full account of this work will be published in due course.

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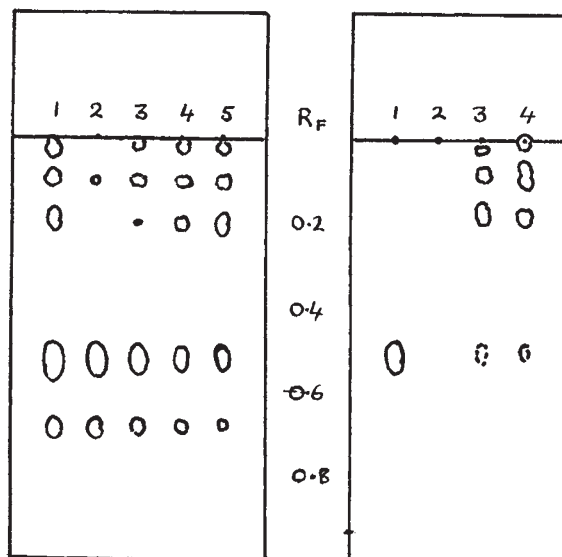
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Chlorogenic Acid and the Enzymic Browning of Apples and Pears

Joslyn and Ponting¹ have recently stressed the meagreness of the information regarding individual phenolic compounds involved in the enzymic browning of fruits, although they suggested that chlorogenic acid was a possible substrate (cf. Ingraham and Corse²). Since chlorogenic acid is known to be acted on by the polyphenoloxidase of sweet potatoes³, and to be widely distributed in many plant tissues including apples and pears⁴, we have investigated its role in the enzymic browning of these two fruits.

Paper chromatograms run in butanol-acetic acid-water (4:1:5) of crude concentrated acetone extracts of Bramley's seedling apples show the presence of five fluorescent components when viewed in ultra-violet light in the presence of ammonia, three of which (R_F 0.02, 0.08 and 0.20) show only faintly (Fig. 1). The major compound (R_F 0.54) was separated by large-scale paper chromatography and shown to be chlorogenic acid by identity of R_F values in several solvents, colour reactions⁵, and ultra-violet absorption spectra in ethanol and 0.002 *M* sodium ethoxide solution⁶.



(1) Crude acetate extract of apple tissue; (2)-(5) chromatograms of apple during course of browning

(1) Chlorogenic acid; (2) apple enzyme extract; (3) mixture of (1) and (2) after reaction; (4) mixture of (1) and pear enzyme extract after reaction

When samples of the tissue are minced in phosphate buffer pH 6.7 and air is bubbled through the suspension, a rapid browning takes place. Chromatograms of the extract show that as browning proceeds the intensity of the compounds of low R_F value increases and that of chlorogenic acid and the other component decreases (Fig. 1,a). Tissues minced in the presence of ascorbic acid or thiourea in which no browning takes place show the presence of chlorogenic acid and the faster running component only.

Tests were carried out in phosphate buffer with pure chlorogenic acid plus either a suspension of minced tissue which had been pre-washed with acetone at -20°C . until free of phenols, or the polyphenoloxidase extracted from it with 1 per cent sodium carbonate solution⁷. In both cases passage of air through the mixture gave rapid browning. Chromatograms of the mixture showed that the three compounds of low R_F values found above were produced during the course of the reaction (Fig. 1,b). When catechol was used as a substrate, no formation of the three slow-moving components was found, although strong browning was observed.

Similar results have been obtained with chlorogenic acid and an enzymic preparation from Conference pears. Thus it is apparent that chlorogenic acid is one of the substrates involved in the enzymic browning in both apple and pear, and that the three fluorescent compounds (R_F 0.02-0.20) are formed from it during the reaction.

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Glycogen in the Dividing Cells of the Liver of the Chicken Embryo

VON GROPP¹ has investigated the distribution of glycogen in hepatic cells of the rat after the administration of the carcinogen butter yellow (*p*-dimethyl-aminoazobenzol). Using Best's carmine technique and the periodic acid-Schiff sequence, he concluded that the amount of glycogen in dividing cells was considerably less than in non-dividing cells. It was further found that when individual dividing cells were examined glycogen was found in 84 of 200 cells (42 per cent), whereas it was present in nearly all the non-dividing cells. The same difference in the glycogen content of dividing and non-dividing cells has been recorded in a transplantable tumour of the rat liver by Strong and Smith². This note records the results of equivalent investigations on the liver of the chicken embryo.

Glycogen can be demonstrated histologically in the liver of the chicken embryo at the end of the seventh day of incubation³. In the present investigations the liver was removed from the embryo during the eighth and ninth days and immediately fixed in Carnoy's fluid. Sections were stained for glycogen by the periodic acid-Schiff sequence, as described