

Table 1. SUBSTRATE ACTIVITIES AND R_F VALUES IN AQUEOUS SOLVENTS OF FLAVONOIDS

Substrate	Structure	R_F value	Oxidized by enzyme
Quercetin	Planar	0.00	Yes*
Quercetin-3-rhamnoside	Non-planar	0.19	No
Quercetin-3-rhamno-glucoside	Non-planar	0.29	No
Myricetin	Planar	0.00	Yes
Myricetin-3-rhamnoside	Non-planar	0.23	No
Luteolin	Planar	0.00	Yes
Luteolin-7-diglucoside	Planar	0.00	Yes

* Roberts and Wood (ref. 5) stated that quercetin was not oxidized by the tea oxidase. This was an inaccurate observation due to the very low solubility in water of quercetin.

in plant tissues, and it is suggested that glycosylation in the 3-position is necessary for translocation to take place. A paradoxical situation may now arise, for the planarity of the molecule, which is necessary for any physiological activity dependent upon enzymic oxidation, will hinder translocation. On the other hand, the more mobile glycoside may be physiologically inactive unless there is a glycosidase present to regenerate the active aglycone. Considerations such as these may also be relevant to the activity of certain flavonoids on vascular resistance or capillary permeability in animals^{6,7}.

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E. A. H. ROBERTS

Indian Tea Association (London),
Butler's Wharf,
Lafone Street, London, S.E.1.

¹ Roberts, E. A. H., Symp. Soc. Leather Trades' Chemists, 87 (Cambridge, 1956).

² Roberts, E. A. H., Cartwright, R. A., and Wood, D. J., *J. Sci. Food Agric.*, 7, 637 (1956).

³ Geissman, T. A., in "Modern Methods of Plant Analysis", by Paech, K., and Tracey, M. V., 3 (Berlin, Göttingen, Heidelberg: Springer-Verlag, 1955).

⁴ Baruah, P., and Swain, T., *J. Sci. Food Agric.*, 10, 125 (1959).

⁵ Roberts, E. A. H., and Wood, D. J., *Nature*, 167, 608 (1951).

⁶ Lockett, M. F., "The Pharmacology of Plant Phenolics", 81 (Academic Press, London, 1959).

⁷ Lavollay, J., and Neumann, J., "The Pharmacology of Plant Phenolics", 103 (Academic Press, London, 1959).

Need for Inorganic Phosphate in Oxidations stimulated by Dinitrophenol

Recently we have reported, in disagreement with the widely accepted view, that the maximal rate of oxygen uptake by respiring rat-liver or rat-heart mitochondria with glutamate as substrate is not obtained with 2,4-dinitrophenol unless phosphate is also present¹. With succinate as substrate, on the other hand, the same initial rate of oxygen uptake was found with dinitrophenol as with inorganic phosphate. We concluded that a link in the respiratory chain between glutamate and cytochrome *b* required phosphate even in the presence of dinitrophenol, a conclusion which seemed to have important implications for theories of the mechanism of the uncoupling of oxidative phosphorylation by dinitrophenol.

We now wish to report that further studies have provided evidence that dinitrophenol can replace inorganic phosphate in the respiratory chain. There is, therefore, no need to modify our earlier view² that dinitrophenol reacts with an energy-rich intermediate

in oxidative phosphorylation formed prior to the intervention of inorganic phosphate.

The new evidence we have obtained is:

(1) Substrates, such as malate and β -hydroxybutyrate, which operate through nicotinamide-adenine dinucleotides, are oxidized at the same rate in the presence of 2,4-dinitrophenol alone as with dinitrophenol and inorganic phosphate together.

(2) The results with glutamate are explained by the finding that a large part of the measured oxygen uptake with this substrate is not concerned with the oxidation of glutamate to α -ketoglutarate, but with the further oxidation of the α -ketoglutarate. This is shown by the fact that the α -ketoglutarate (μ moles) found in the presence of phosphate was less than 1 per cent of the oxygen uptake (μ atoms). Malonate (0.02 M) inhibited by 70 per cent the rate of oxygen uptake with liver mitochondria. The requirement for inorganic phosphate for maximal rates of oxygen uptake is explained by the fact that dinitrophenol cannot replace inorganic phosphate in the 'substrate-linked' phosphorylation step associated with α -ketoglutarate oxidation³.

The rapidity with which the α -ketoglutarate formed from glutamate is oxidized by mitochondrial preparations was unexpected. It is clear that in the absence of an inhibitor of α -ketoglutarate oxidation, glutamate is not a suitable substrate for studying oxidative phosphorylation and respiratory control associated with the oxidation of reduced nicotinamide-adenine dinucleotides.

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P. BORST

E. C. SLATER

Laboratory of Physiological Chemistry,
University of Amsterdam.

¹ Borst, P., and Slater, E. C., *Nature*, 184, 1396 (1959).

² Slater, E. C., *Nature*, 172, 975 (1953).

³ Judah, J. D., *Biochem. J.*, 49, 271 (1951). Hunter, F. E., in "Phosphorus Metabolism", 1, 297 (The Johns Hopkins Press, Baltimore, 1959).

PHARMACOLOGY

Histamine Sensitivity in Children after Pertussis Infection

It has been shown in recent years that mice and rats are rendered hypersensitive to the action of histamine, 5-hydroxytryptamine and anaphylactic sensitization by pre-treatment with *Haemophilus pertussis* vaccine¹⁻⁴. These properties are present only in organisms in the smooth phase and are not shared by other members of the *Haemophilus* family^{5,6}. The sensitizing action is not seen in the guinea pig and the rabbit; on the other hand, there is some evidence that these two species are rendered slightly more resistant to histamine after such treatment⁷. Rats and mice are relatively resistant to histamine and anaphylactic shock, whereas guinea pigs and rabbits are susceptible. It was therefore of interest to study whether the reaction in man follows a similar pattern to the former two species or the latter. Recently there was an epidemic of whooping cough in Darbhanga and advantage was taken of this for such a study.