

r_f VALUES OF CARBOXYLATE IONS IN BUTANOL-1.5 N AMMONIUM HYDROXIDE AT 15° C. WHATMAN NO. 1 PAPERS. 1 PER CENT SOLUTIONS

Anion	r_f	Anion	r_f
Formate	0.09	Salicylate	0.50
Acetate	0.10	β -Phenylpropionate	0.52
Propionate	0.19	Lactate	0.07
<i>n</i> -Butyrate	0.33	Oxalate	0
<i>n</i> -Valerate	0.45	Malate	0
<i>n</i> -Caproate	0.61	Tartrate	0
<i>n</i> -Caprylate	0.74	Adipate	0
Benzoate	0.39	Citrate	0

The r_f values of the anions of the lower fatty acids are shown in the accompanying table. Formate and acetate have similar r_f values; but confusion between the two can be avoided by making use of the reaction between the former and ammoniacal silver nitrate. The other anions of the series give well-separated spots.

The r_f values of several other carboxylate ions are also included in the table. It is interesting to note that dicarboxylate ions do not move from the starting line in the solvent used. The method appears to be applicable to the sodium salts of both aromatic and aliphatic monocarboxylic acids, although these could probably be more conveniently separated as the free acids.

The cations (in this case the sodium ion) moved only short distances from the starting line. An interesting feature was, however, the increase in excursion of the cation as the r_f value of the anion increased. Full experimental details will be published elsewhere.

F. BROWN
L. P. HALL

Bristol University Fruit and Vegetable
Preservation Research Station,
Campden, Glos.
March 10.

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Hydrolysis of Laminarin by Wheat β -Amylase

THE only enzymes so far found to hydrolyse laminarin are those of the snail¹ and the limpet². In 1915 Kylin³ stated that he had hydrolysed it with malt diastase; but he has recently withdrawn this claim⁴, and in this Laboratory also it has been found that malt diastase is without action on laminarin.

We have now found that the β -amylase of wheat hydrolyses laminarin, yielding glucose (70 per cent) and a disaccharide. The rate of hydrolysis is slow. An extract of the enzyme which hydrolysed starch in a few hours took several days to hydrolyse laminarin. Since malt diastase is inactive, the action must be due to some factor other than the small amount of α -amylase in the wheat enzyme.

This ability of wheat β -amylase to hydrolyse 1:3 linkages is of interest in connexion with recent work on the action of amylases on starch. Thus, Barry, Halsall, Hirst and Jones⁵ found that crystalline β -amylase had little effect on floridean starch, whereas

we have found that wheat β -amylase hydrolyses it to maltose to the extent of 50 per cent. Again, Halsall, Hirst, Hough and Jones⁶ found that the limit dextrans produced from amylopectins by crystalline β -amylase have a much higher molecular weight than those from the crude wheat enzyme. These facts suggest that the amylopectins contain 1:3 linkages which are hydrolysed by the factor in wheat β -amylase that hydrolyses laminarin. The occurrence of such linkages in glycogen has already been suggested by Bell⁷.

THOMAS DILLON
PROINNSIAS O'COLLA

Chemical Laboratory,
University College,
Galway.
March 4.

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Separation of NorAdrenalin and Adrenalin

A METHOD of separating these two substances on paper chromatograms, using phenol as solvent, has been described earlier¹. No satisfactory second solvent, enabling a two-way chromatogram to be performed, was found at the time, and it was necessary to use the substances as hydrochlorides, the free bases being insufficiently stable. Movement in *n*-butanol was very slight. More recently, von Euler and Hamberg² have reported separation of adrenalin and *l*-nor-adrenalin, using *n*-butanol saturated with *N* hydrochloric acid. Their results are recorded in a photograph of a small part of the chromatogram and no data are given for the r_f values or the time necessary to obtain separation. On repeating their experiment, we found that movement was very little greater than in our original *n*-butanol-water system, and that, to obtain separation, the chromatogram had to be run for forty-eight hours or longer, allowing the solvent to drip from the end of the paper. It was, therefore, not possible to determine r_f values.

It appeared probable that greater travel, allowing a faster separation, might be obtained by using an organic acid instead of hydrochloric acid. Acetic and trichloroacetic acids were chosen for trial. For experiments with the first, the bases were applied as solutions in 5 per cent acetic acid, and the solvent was an *n*-butanol-acetic acid mixture made by shaking four parts *n*-butanol with one part glacial acetic acid and five parts water, and rejecting the lower layer on separation. For trichloroacetic acid experiments, a 5 per cent solution was used to dissolve the bases, and the solvent was made up with 5 gm. trichloroacetic acid dissolved in 100 ml. water and added to 80 ml. *n*-butanol. After shaking, the lower layer was discarded. 0.015 ml. solution containing about 0.03 mgm. adrenalin or noradrenalin was applied with an 'Agla' pipette and the run made in a saturated atmosphere over the appropriate mixture.

Drying the papers in gentle heat did not drive off all the acid, and more alkali was needed in the indicator than was used previously. 0.44 per cent