From the evidence above it appears that the substituents on position 4(5) of the mercaptoimidazole ring must have, or be capable of giving, a system of double bonds conjugated with the ring for the magenta colour to develop. Ergothioneine is the only substance known to occur in biological fluids which meets these requirements[3].

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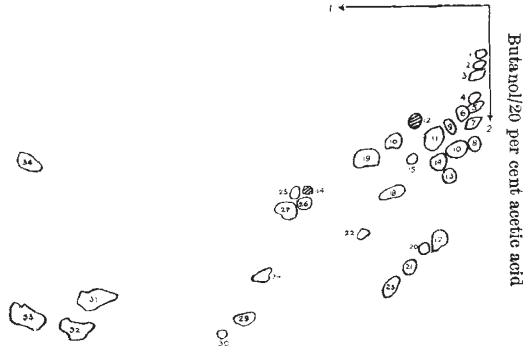
Autolysis Products of Pepsin

DURING the recrystallization of pepsin by the method of Northrop[1], which involves heating a solution of the enzyme to 45° C. at *p*H 4, followed by cooling, it was noticed that the warm solution deposited a considerable quantity of needle crystals totally different from crystalline pepsin; these were identified as tyrosine. Continued autolysis under these conditions yielded 35 mgm. of tyrosine in 24 hr. from 6 c.c. of a solution containing 1·0 gm. of dry pepsin. This represents approximately 40 per cent of the total tyrosine present. Investigation of the dialysed mother liquor by two-dimensional paper chromatography[2,3] revealed the presence of at least thirty-four ninhydrin-positive substances (see diagram), many of which disappeared on acid hydrolysis, indicating that they are peptides. It is suspected that some of the spots are simple amino-acids, as indicated by their position and speed of development. Samples of the autolysing solution taken at 5-min. intervals during the first hour showed on paper chromatography with butanol – acetic acid in one dimension that a very complex pattern of spots is present from the beginning, though extremely faint at first. Analysis in one dimension could not, of course, decide whether the initial pattern contains all the thirty-four spots finally detected; but it is provisionally concluded that the formation of the final fragments are not rate-determining steps.

It was possible to isolate reasonably pure specimens of the fragments Nos. 1, 2 and 3 (see diagram) by multiple analysis in one dimension using butanol – acetic acid; the areas containing the peptides were detected under ultra-violet light and eluted with water. Hydrolysis with 5 *N* hydrochloric acid at 100° C. for 24 hr. followed by two-dimensional analysis showed that the amino-acids in the accompanying list were present. 'Under-cystine' gives rise to a strong blue-mauve ninhydrin spot; it travels in butanol – 20 per cent acetic acid with the same speed as cystine, but is faster in butanol – 25 per cent pyridine, appearing between lysine and arginine. 'Under-cystine' has not yet been identified; the speed with which it develops suggests that it is an amino-acid, though a resistant peptide cannot be excluded.

Butanol/25 per cent pyridine



Peptide No.	1	2	3
'Under-cystine' (m.w.120 ?)	(1)	(2)	(2)
Aspartic acid	(2)	(3)	(4)
Glutamic acid	(1)	(1)	(3)
Serine	(3)	(3)	(4)
Glycine	(2)	(2)	(2)
Alanine	(1)	(1)	(1)
Histidine	(1)	—	—
Arginine	—	(1)	—
Threonine	—	(1)	(2)
No. of amino-acid residues	11	14	18
Provisional mol. wt., approx.	1,080	1,430	1,850

Careful examination of the size and intensities of the spots gave the provisional molecular ratios shown in the above table. To confirm the identity of these constituents, the two-dimensional analysis was repeated with the addition of authentic samples of those amino-acids the presence of which was suspected and also cystine; with the exception of 'under-cystine', no extra spots were obtained.

Treatment of ice-cold weakly acid solutions of the peptides with 0·01 *N* sodium nitrite followed by urea, hydrolysis and paper chromatography indicated that in all three cases alanine is the end amino-acid with the free amino-group, and that this occurs only once in these peptide molecules. The provisional molecular weights given in the table follow from this result.

The publication of this note is prompted by the temporary interruption of this work; it is hoped to continue and extend these experiments shortly. I wish to acknowledge a grant from the Research Fund of the Chemical Society.

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Mesomorphism of some Alkoxynaphthoic Acids

THE existence of mesophases in the melts of *p-n*-alkoxybenzoic and *p-n*-alkoxycinnamic acids has already been established[1,2]. In these acids the length of the rod-shaped molecules is enhanced by hydrogen-bond formation, and the mesomorphism arises from the association of the acids in double molecules. In this way, *p-n*-propoxybenzoic acid, which has the simplest molecular structure yet found to give a mesomorphic form, acquires a structure which is similar in length and shape to that of a typical nematic compound, such as azoxyanisole.