corresponded to the quantity of oxime nitrogen which had disappeared. I propose to name this enzyme tentatively 'oximase'.

KAZUO YAMAFUJI

Agricultural Chemistry Institute,
Kyushu University,
Fukuoka, Japan. Dec. 20.

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Pectic Enzymes produced by *Bacterium aroideæ*

REID[1] has recently described some properties of the pectic enzymes secreted by the fungus *Byssochlamys fulva*, and compared them with those described earlier by Beavan and Brown[2].

Similar studies are in progress here using *Bacterium aroideæ*. The bacteria are cultured in synthetic media, and solutions of the enzymes are prepared after precipitation from the cell-free culture fluid, dialysis and reprecipitation. Such preparations retain about 50 per cent of the activity of the original culture, macerate disks of potato tuber tissue 500 μ thick in fifteen minutes, have little pectin-esterase activity but rapidly reduce the viscosity of 0·5 per cent solutions of a high-methoxyl (9·15 per cent) citrus pectin at optimum *p*H. For example, one preparation reduced the specific viscosity from an initial value of 3·20 to 0·77 in one minute and to the low value of 0·15 after twenty minutes. These figures are in contrast with those of Beavan and Brown[2], who report their active preparations as macerating disks of potato tuber in eighteen hours and taking approximately ten hours to produce a substantial decrease in the viscosity of 1·0 per cent pectin solutions.

The properties of the viscosity-reducing enzyme secreted by this bacterium have been studied using as a measure of activity the inverse of the time taken for a 0·5 per cent solution of a high-methoxyl (9·15 per cent) pectin to reach a given value as determined from viscosity/time curves. On this basis a linear relation is obtained between enzyme concentration and activity. The enzyme is most active at high *p*H values, having an optimum between 8·5 and 9·0. Inactivation is rapid below *p*H 3·7. Thus, at *p*H 2·7 activity is reduced by 50 per cent after one minute and by 90 per cent after sixty minutes. Inactivation is also rapid at 60° C. and above, only 5 per cent of the original activity remaining after ten minutes at 60° C.

Prolonged dialysis at 5° C. leads to a considerable loss of activity. This is, however, completely restored by the addition of calcium ions. This effect is not produced by sodium, potassium, magnesium, strontium or barium ions. Enzyme preparations are normally prepared from cultures containing excess calcium carbonate; but similar activation by calcium is obtained after culturing the bacterium in media free from calcium ions.

With high-methoxyl pectins, the major viscosity changes take place with little increase in reducing power of the solution. There is little further increase even after treatment for twenty-four hours. Enzyme preparations behaving in this way still rapidly macerate disks of potato tuber. While high-methoxyl pectins have generally been used in these studies, the action of the enzyme on demethoxylated pectins is substantially the same.

The pectinolytic enzyme or enzymes secreted by *Bacterium aroideæ* therefore have properties substantially different from those of polygalacturonase, as described by Lineweaver *et al.*[3], and is peculiar in the following respects: (1) ability to act upon high-methoxyl pectins in the absence of pectin-esterase; (2) a high *p*H optimum; (3) activation by calcium ions; (4) inability to cause substantial increase in the reducing power of pectin solutions while causing very rapid reduction in viscosity.

The above work will be described in detail elsewhere.

R. K. S. WOOD

Imperial College of Science and Technology,
London, S.W.7.
(At present Commonwealth Fund Fellow,
Divisions of Plant Pathology and
Food Technology,
University of California,
Berkeley, California.)
Dec. 20.

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Enzymatic Inactivation of Secretin

THE inactivation of secretin by whole blood, cell-free plasma, serum and urine has been demonstrated by Greengard and Ivy[1,2] to be an enzymatic process. Because of the stability of their secretin preparations towards crystalline pepsin and trypsin and the absence of detectable proteolytic activity of blood, they concluded that 'secretinase' was not a proteolytic enzyme.

In this laboratory, however, it has been possible to show that 'secretinase' activity of plasma is activated by chloroform treatment and inhibited by soya bean trypsin inhibitor. Moreover, our preparations of secretin obtained by the procedure of Greengard and Ivy[3] are, in fact, inactivated by crystalline pepsin.

Secretin assays were carried out on dogs in the usual manner[3], and all *in vitro* experiments were conducted at 37° C. and *p*H 7·4. It was found that 0·4 mgm. secretin (*n*-butanol extracted fraction *S*II[3]) was inactivated by 4 ml. of a 1 : 1 dilution of citrated dog plasma in 0·05 *M* phosphate buffer within 1–3 hr. When chloroform-activated globulin, that is, serum protease prepared according to Christensen and McLeod[4], was tested similarly at eight times the dilution of the original plasma, the rate of inactivation was found to be increased several-fold (see table).

An amorphous trypsin inhibitor was prepared from soya bean by the same procedure as that used by Tauber *et al.*[5] for Lima bean. A concentration of 2 mgm./ml. inhibited maximally 0·4 mgm./ml. B.D.H. trypsin. When 2·5 mgm./ml. of this inhibitor was added to the plasma and chloroform-activated globulin solutions, the 'secretinase' activity was inhibited (see table).

Preliminary experiments in which up to 300 mgm. of amorphous trypsin inhibitor was injected intravenously into dogs showed, however, no effect on the secretin response of the animals. Further experiments are proposed in which crystalline preparations of the inhibitor will be used.

Although the evidence is insufficient to identify the 'secretinase' with the plasma protease, these experiments do suggest that, contrary to previous observations[1,2], the plasma inactivation of secretin proceeds by proteolysis, while the known proteolytic activity of urine[6] could likewise account for its 'secretinase' activity. It may be added that

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