

PHENOLOXIDASE FROM THE LEAVES OF THE COTTON PLANT

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There is information in the literature on the presence of phenoloxidase in the leaves of the cotton plant [1].

We extracted the proteins from freshly frozen leaves with a tenfold amount of 0.1 M citrate-phosphate buffer, pH 7.2, in the presence of 1% of ascorbic acid and of Kapron powder — 0.5 g of powder per g of leaves. The extract was filtered and centrifuged at 18,000 rpm for 15 min, and the supernatant liquid was dialyzed against distilled water for a day. The phenoloxidase activity in the dialyzate was determined by Drawert's method [2], using as the substrate mixture 2 ml of a 10% solution of catechol, 2 ml of 2% proline, and 1 ml of enzyme solution. After the lapse of 20 min, the reaction was fixed by the addition of 0.5 ml of a 5% solution of trichloroacetic acid. A change in the absorption (violet filter) by 0.001 in 1 min was taken as the unit of activity.

To determine the highest specific activity of the phenoloxidase in the leaves of the cotton plant, we studied the dynamics of the accumulation of this enzyme in young cotton shoots. The experiments were performed on cotton-plant leaves collected in the early periods of vegetation in the Tashkent oblast in 1972. No phenoloxidase was found in the seeds and the cotyledonous leaflets. The specific activity of the phenoloxidase was 7 in May, 14 in June, 50 in July (chopping period), 40 in August, and 10 in September. Consequently, the greatest phenoloxidase activity is observed in the period of vigorous growth of the cotton plant.

In addition to the soluble phenoloxidase, we found in the leaves of the cotton plant a latent phenoloxidase which was extracted by the buffer solution mentioned but only in the presence of the detergent Triton X-100. All the operations were performed at a temperature of +4–6°C.

Thus, the optimum conditions have been selected for the extraction of phenoloxidase from the leaves of the cotton plant of variety 108-F, and the vegetation period with the greatest specific activity of this enzyme has been determined.

LITERATURE CITED

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2. F. Drawert and H. Gebbing, *Naturwiss.*, **50**, 522 (1963).

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