REGULATION OF INVERTASE LEVELS IN SUGAR CANE BY AN AUXIN-CARBOHYDRATE MEDIATED CONTROL SYSTEM

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Immature storage tissue of sugar cane contains considerable quantities of reducing sugars derived from sucrose accumulated into the inner space (which must include the vacuole). An invertase which is optimally active at pH 5.5 is present at high levels in this tissue, but only in small amounts in mature tissue which has little reducing sugars (Glasziou, 1961).

This report indicates that the level of invertase is under the regulatory control of auxin, which increases the amount, and carbohydrate, which reduces the amount of the enzyme in storage tissue.

RESULTS AND DISCUSSION

Storage tissue discs which had been incubated 18 hours in a synthetic auxin solution (α-naphthalene acetic acid, NAA) showed increased invertase activity as compared with discs incubated in water. Over the range of NAA concentrations used, 1.42×10^{-5}M was optimal. In subsequent experiments it was demonstrated that incubation of discs in NAA caused up to a 3-fold increase in the amount of enzyme over that of fresh cut discs (Table I). Discs maintained in water sometimes showed an

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increase and at other times a decrease in enzyme level as compared with fresh cut discs. This variability is probably due to variations in the amounts of endogenous auxin.

TABLE I

EFFECT OF INCUBATION TREATMENT ON INVERTASE ACTIVITY

Treatment of Discs	Invertase Activity Expressed as Percent of Controls			
	Exp. 1	Exp. 2	Exp. 3	Exp. 4
Fresh cut (control)	100	100	100	100
18 hours in H_2O	196	238	98	190
18 hours in 1.42×10^{-5}M NAA	244	300	220	270

Immature internodes from a commercial variety of sugar cane (NCO 310) were used. Discs were prepared from the basal 20 mm of internodes, and washed one hour in running tap water. The discs were randomized and distributed into lots of 22 each (1.75 g fresh weight), which were used for assay of total invertase (juice plus cell residue), and for incubation in 20 ml volumes of water or various solutions for 18 hours at room temperature in a shaker. Solutions were changed twice during the incubation period, after which discs were washed for one hour before being assayed for invertase activity.

For assays of invertase discs were ground in a mortar. The juice was expressed through fine muslin and dialyzed to remove sugars. Juice and remaining cell residue were assayed separately in reaction mixtures, containing 1% sucrose-C^{14}, at pH 5.5 and 30° with toluene added. At intervals 5 µl aliquots of reaction mixtures were applied onto Whatman No. 1 paper and chromatographed for subsequent assay of radioactive sugars as described in a previous paper (Glasziou, 1960).

It was observed that incubation of discs in a 1% (w/v) glucose solution caused a considerable diminution in the amount of invertase as compared with the controls (Table II). Further, the stimulating effect of added NAA on invertase levels was completely suppressed by glucose. Similar results were obtained with fructose and sucrose. Galactose and mannitol were less effective. Since sucrose is inverted prior to accumulation by the tissue (Sacher, Hatch and Glasziou, to be

published), the effective suppressor in vivo may be a hexose or close derivative. The lesser effect of galactose and mannitol respectively may reside in the relative efficiency of their conversion to a more active form in the tissue.

TABLE II

EFFECT OF AUXIN AS A STIMULATOR AND GLUCOSE AS A SUPPRESSOR OF INVERTASE LEVELS

Treatment of Discs	Invertase Activity+
Freshcut (controls)	3807
18 hours in H_2O	3743
18 hours in 1.42×10^{-5}M NAA	8315
18 hours in 1% Glucose	1843
18 hours in 1% Glucose + 1.42×10^{-5}M NAA	1872

+ µg of sucrose hydrolyzed/g fresh weight/hour

NAA did not have any effect on invertase activity measured in dialyzed tissue homogenates (juice plus cell residue) from storage tissue. These results indicate that the action of NAA and sugars is on enzyme synthesis.

We conclude that invertase has an important role in cellular mechanisms directing the utilization of carbohydrate for growth or storage, the synthesis of enzyme being regulated by auxin and a feedback control system dependent upon reaction products of invertase activity. Further evidence in support of this hypothesis will be published elsewhere.

REFERENCES

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