

EFFECT OF Mg^{++} AND Mn^{++} IONS ON SUCROSE SYNTHETASE IN SUGARCANE LEAVES

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Sucrose synthetase is a major enzyme responsible for sucrose synthesis in sugarcane leaves. The enzyme is stimulated by Mg^{++} and Mn^{++} , the former one being the major stimulator. In the present investigation it has been observed that under natural conditions highest sucrose synthetase activity is found when both Mg^{++} and Mn^{++} are present in the metabolic environment. Eventhough Mg^{++} is a major co-factor, it gives highest activity when accompanied by small amounts of Mn^{++} .

INTRODUCTION

Sugarcane is an important plant in which sucrose synthesis is a major metabolic process. Leloir and co-workers (1953 and 1955) showed that in plants sucrose is synthesised by sucrose synthetase as well as sucrose phosphate synthetase. Pandya and Ramkrishnan (1956), Ramkrishnan (1958), and Shukla and Prabhu (1959) reported the presence of an enzyme-sucrose phosphorylase in sugarcane leaves, which catalyses the synthesis of sucrose from glucose-1-phosphate and fructose. However, Frydman and Hassid (1963) showed evidence for the synthesis of sucrose from UDPG and D-fructose in the sugarcane leaves. The results of earlier workers indicate that both the enzymes can synthesise sucrose in sugarcane leaves. However, the studies of Haq and Hassid (1965) indicated that small amount of sucrose is synthesised when fructose-6-phosphate acts as a glucosyl acceptor in place of fructose. This indicates that sucrose synthetase is the major enzyme system responsible for the synthesis of sucrose in sugarcane leaves.

Bean and Hassid (1955), while working with pea preparations, observed that Mg^{++} has a stimulatory effect on sucrose synthetase. Rorem *et al.* (1960), as well as Haq and Hassid (1965) used $MnCl_2$ while assaying the enzyme system. Pressey (1969) observed that Mn^{++} acts as an activator of sucrose synthetase in potato tuber. The main object of this investigation is to study the effect of Mg^{++} and Mn^{++} ions on sucrose synthetase in sugarcane leaves. It has been observed that the soils in Kolhapur region contain appreciable amounts of Mn^{++} and sucrose content of sugarcane is quite high as compared to sugarcane grown in rest of the country. The effect of these ions was studied by adding them at reaction level after extracting the enzyme, as well as supplying to plants as nutrients and as foliar sprays.

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MATERIALS AND METHODS

Sugarcane variety Co-740, which is widely cultivated in this region, was selected for the present investigations. The leaves were thoroughly cleaned and used as the enzyme source. Sucrose synthetase (EC 2.4.1.13) was studied by the method described by Pontis and Leloir (1962) and the amount of sucrose synthesised was estimated by resorcinol method of Roe (1934).

Four-month-old healthy canes were selected for culture studies. Culture experiments were carried out by supplying the plants with $MgSO_4$ $10^{-2}M$ and $MnCl_2$ $10^{-3} M$ once a day for six weeks. The salts were given as nutrient in soil and were also sprayed on leaves for a different set of plants. The plants were properly irrigated whenever necessary. The blades of the third and fourth leaves were washed and used for the analysis. The sampling was done randomly. The enzyme was studied by the method described above. Mg^{++} and Mn^{++} were estimated colorimetrically (Richards 1954 and Horwitz 1965).

RESULTS AND DISCUSSION

In order to study the effect of Mg^{++} and Mn^{++} ions, sucrose synthetase activity of the extracted enzyme was determined in the presence of various concentrations of $MgCl_2$ and $MnCl_2$ and the results are presented in Fig. 1. It is obvious from the figure that both the ions when in contact in individual capacity show stimulation in synthesis of sucrose. When the concentration of $MgCl_2$ increases there is a rapid increase in the enzyme activity and the peak is obtained at the concentration of $4 \mu g/ml$ and the enzyme activity falls down when the concentration increases to $8 \mu g/ml$ onwards. It is interesting to record that when $MnCl_2$ is added to the enzyme system the activity reaches its maximum at $8 \mu g/ml$ after which a sharp fall has been recorded in further concentrations. The work of Pressey (1969) on the effect of Mn^{++} in stimulation of sucrose synthetase indicates that when Mn^{++} alone is present as an activator it is required in high amounts. Eventhough both the ions activate the enzyme system, the maximum activity is more with $MgCl_2$ than with $MnCl_2$. It is obvious that both these ions show stimulation in the enzyme activity at particular concentration above which they have inhibitory effect. These results indicate that Mg^{++} is possibly the main co-factor of the enzyme sucrose synthetase and cannot be completely replaced by Mn^{++} .

Sugarcane plants were supplied with $MgSO_4$ $10^{-2} M$ and $MnCl_2$ $10^{-3} M$ both as nutrient in soil and as foliar spray. Mg and Mn contents and sucrose synthetase activity of control and treated plants are recorded in Table I.

Humbert (1963) reported about 0.1 per cent of Mn and 0.08-0.35 per cent of Mg in sugarcane leaves on dry-weight basis. It is interesting to observe that both the ions show more accumulation when supplied as foliar spray than as a nutrient in soil. It was recorded earlier (Humbert 1963) that Mg levels increase with the increasing Mg applications. In the present investigation it has been found that even in $MnCl_2$ -treated plants Mg accumulation is more than that in control plants which is difficult to explain.

Mn^{++} is one of the essential nutrients for the healthy growth and development of sugarcane (Davis 1931). The highest Mn value is recorded in the leaves

of the plants sprayed with $MnCl_2$. There is no remarkable difference in the Mn content of other treated plants. The values obtained in the present investigation for Mg and Mn are higher than those obtained by Humbert (1963).

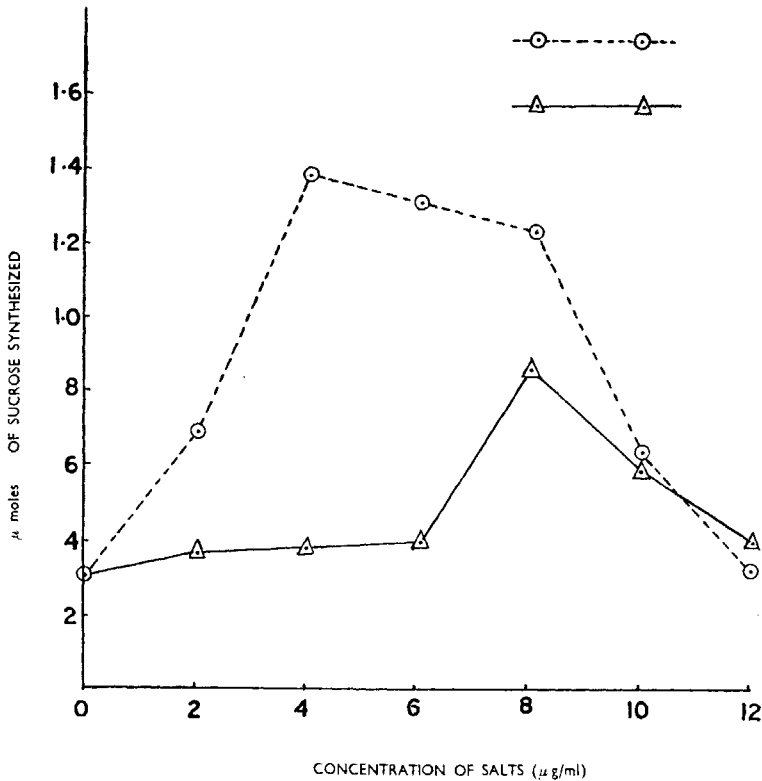


FIG. 1. Effect of various concentrations of $MgCl_2$ and $MnCl_2$ on isolated sucrose synthetase system.

TABLE I

Mg and Mn contents and sucrose synthetase activity in $MgSO_4$ $10^{-2}M$ and $MnCl_2$ $10^{-3}M$ treated plants

Treatment	Mode	Mg*	Mn*	Sucrose synthetase** activity
Control	—	14.39	0.513	0.218
$MgSO_4$ $10^{-2}M$	Spray	56.11	0.611	0.260
$MnCl_2$ $10^{-3}M$	do	29.61	0.79	0.731
$MgSO_4$ $10^{-2}M$	Nutrient	17.11	0.582	0.248
$MnCl_2$ $10^{-3}M$	do	24.64	0.60	0.309

*Values expressed as meq per 100 g dry matter

**Values expressed as μmoles of sucrose synthesised

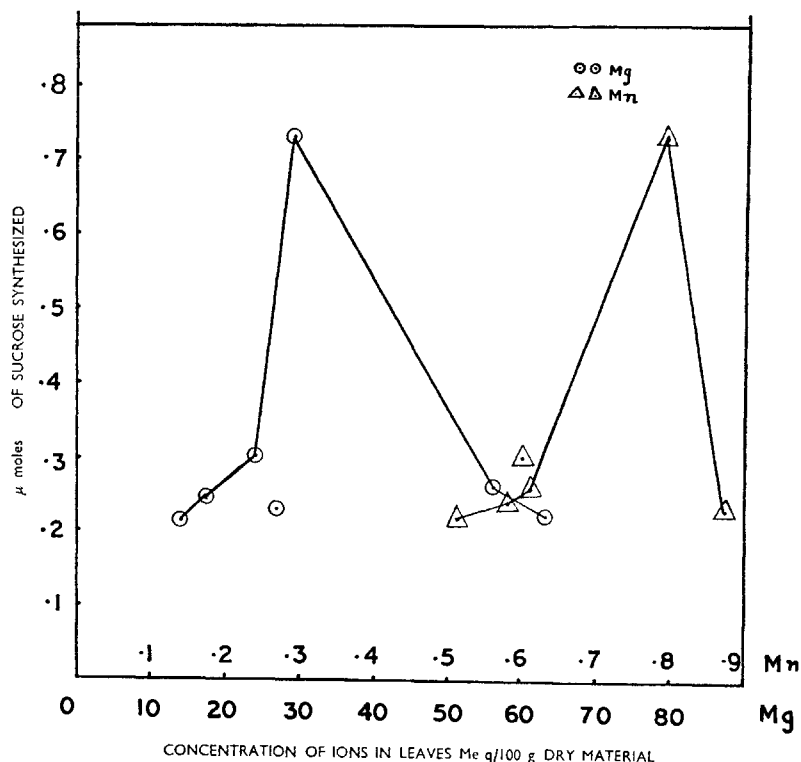


FIG. 2, Effect of Mg^{++} and Mn^{++} concentrations on sucrose synthetase system under natural conditions.

The effect of Mg^{++} and Mn^{++} concentrations on sucrose synthetase system in the treated plants is shown in Fig. 2. From the Figure as well as from the Table it is clear that the maximum sucrose synthetase activity occurs when Mg^{++} ions are accompanied by good amount of Mn^{++} . The results indicate that Mg^{++} alone is not enough to achieve highest sucrose synthetase activity. This can happen even with less Mg^{++} if Mn^{++} is more. It is possible that under natural conditions these two ions with interaction produce a stimulatory range of sucrose synthetase. The range for this is nearly 1:40 ratio of Mn^{++} to Mg^{++} . This can only be provided if isolated enzyme preparation is tested for the effect of these two ions when present together. However, our observations as well as those of other workers indicate that sucrose synthetase under isolated condition behaves differently than when present in cellular environment. Investigations to test the effect of Mg^{++} and Mn^{++} ions together are in progress and will be published elsewhere. The main finding of the present investigation is that Mn-rich soil may produce better sucrose yield than Mn-deficient soil.

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