

## **Chemical and Enzymatic Changes in Strongly Damaged Beets**

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### *ABSTRACT*

*Damage caused by skips during loading and unloading may influence beet quality during the first few hours following the damage. Sucrose content decreases sharply during the first 24 h and (together with acid and neutral invertase) sucrose synthetase seems to be metabolically active in sucrose hydrolysis after harvest. These data are important since, following factory processing, losses could be considerable by the end of the campaign.*

### INTRODUCTION

Damage to sugar-beet roots is a possible source of sugar loss, either by the formation of fragments which cannot be recovered or by metabolic and enzymatic processes that increase damage to the roots when they are not

immediately processed (McCready & Goodwin, 1966; Wyse & Dexter, 1971; Cole, 1977; Wyse, 1978; Akeson & Stout, 1978; Wyse & Peterson, 1979; Peterson *et al.*, 1980).

One of the causes of injury to roots, before their arrival in the sugar factory, is to be found in the use of the skip. This machine, which picks up the roots from the pile and puts them on trucks, can cause cracks and bruises which are reflected in the industrial value of raw material, particularly during short or long periods of storage. After preliminary experiments carried out in 1982 (Baraldi *et al.*, 1983), the following year (Baraldi *et al.*, 1984) we evaluated the amount of damage, and various chemical and enzymatic parameters, related to the variation, during storage, of the technological value of the roots loaded by skips. From this investigation we assessed that the most important consequences of the damage were found during the first hours following picking.

This result could assume particular importance since the interval between the time of loading on trucks by the skips, and the time of processing of the roots in the sugar factory, is unlikely to be less than 24–48 h.

The purpose of this study was to quantify the harmful effect of the damage on sugar-beet harvesting, and also to examine acid and neutral invertase and sucrose synthetase, key enzymes of carbon metabolism in plants.

## MATERIALS AND METHODS

We used drawn skips, type FALCO B-14 (Fig. 1), as in previous experimentation (Baraldi *et al.*, 1984). About 30 tons of beet just harvested and piled up were shifted using skips.

The damaged roots, corresponding to types 3, 4, 5 and 6 of Fig. 2, and representing about 10–15% of all the roots, were selected for the preparation of samples. Fifteen samples, corresponding to five treatments and three replications, of about 50 kg of roots each were made up.

The roots utilized for the preparation of the samples were carefully topped and selected from 12 to 16 cm in diameter. At the end of the sampling we analyzed the first sample while the others, left on the ground, were analyzed in the sequence shown in Fig. 3, with a maximum time of storage of about 60 h.

While the polarization and the dry substance analysis were carried out in the 'brei' (fine beet particles), the other analytical determinations and enzymatic investigation were carried out on the extraction juice of the same brei, stored at  $-20^{\circ}\text{C}$ . (ICUMSA, 1979), and data related to the initial dry substance; reducing



Fig. 1. Loading skips.

sugars were determined by the Berlin Institute method using Muller's solution (ICUMSA, 1979) and expressed in % polarization.

The raffinose and lactic acid amounts were enzymatically evaluated (ICUMSA, 1979).

The specific activities of acid and neutral invertase (EC. 3.2.1.26) and sucrose synthetase (EC. 2.4.1.13) were determined as described by Silvius & Snyder (1979).

The maximum and minimum temperature and rainfall are shown, for the time of experimentation, in Table 1.

## RESULTS AND DISCUSSION

The observed decrease of dry substance of the roots (Fig. 3) may be related to particularly damp and rainy weather. It is well known, as reported by Vukov & Hangyal (1985), that the changes in the composition of the roots, particularly regarding the dry substance, are dependent on the relative humidity (RH) of the air: below 85% there is a loss, over 90% there is a condensation of water.

Although this water can penetrate into the roots even if these latter are uninjured, this phenomenon is more important at positions corresponding

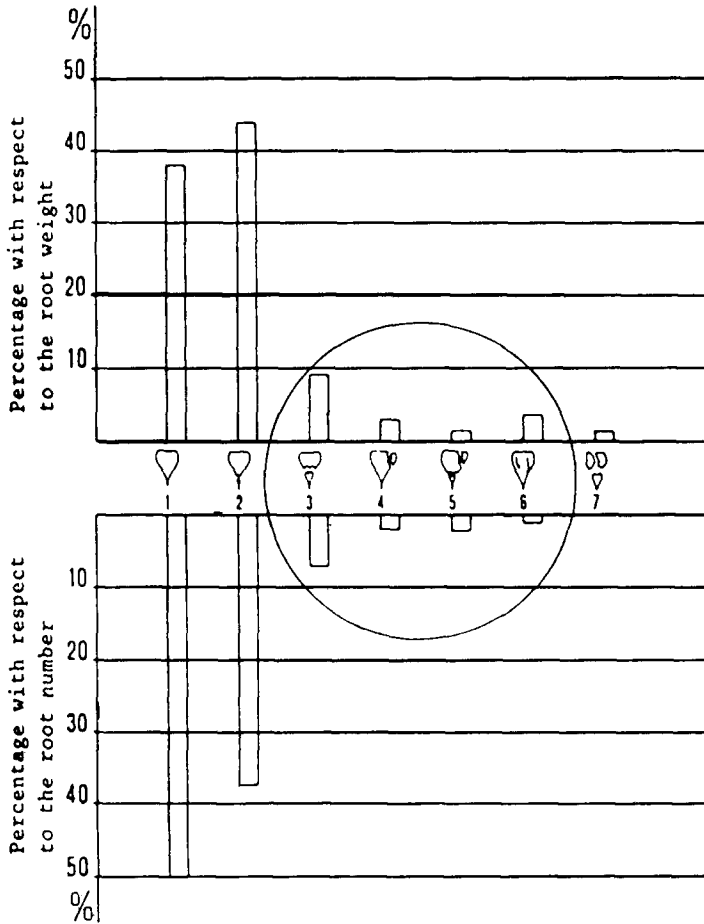


Fig. 2. Damage caused by skips. The experiment was carried out on the types of roots included within the circle. 1, Sound beet; 2, beet showing a tail cutting  $\phi < 6$  cm; 3, beet showing a tail cutting  $\phi > 6$  cm; 4, beet showing side damage; 5, beet showing side damage and tail cutting; 6, beet showing cracks; 7, fragments  $\phi > 3$  cm.

TABLE 1  
Maximum and Minimum Temperatures and Rainfalls during the Experiments

Date	Temperature ( $^{\circ}$ C)		Rainfall (mm)
	Min	Max	
September 18	14.8	27.1	—
September 19	16.0	24.8	8
September 20	14.2	19.1	52
September 21	15.5	23.8	—

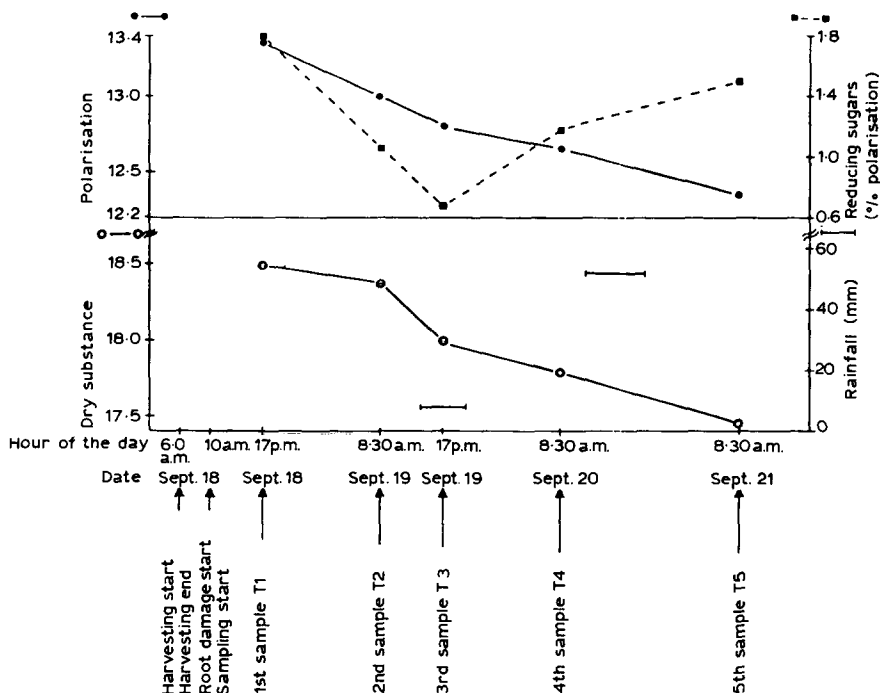


Fig. 3. Variation of polarization, reducing sugars, dry substance and rainfall during the experiment. On the abscissa are shown the data and hours in which the harvesting, damage and analysis took place.

to the damage. So the injury makes the adsorption and desorption of water easier and consequently the change of the dry substance.

Regarding the variation of polarization during the first period of storage (Fig. 3), the sucrose content sharply decreases in the first few hours. The regression analysis allowed us to calculate, in the first 24 h, a  $0.023^{\circ}\text{Pol/h}$  decrease ( $0.55 \text{ g sucrose}/100 \text{ g root/day}$ ). Whereas successively the decrease was about half ( $0.013^{\circ}\text{Pol/h}$  corresponding to about  $0.31 \text{ g sucrose}/100 \text{ g root/day}$ ). These data are in agreement with our values obtained on roots damaged by skips in 1982 (Baraldi *et al.*, 1983) ( $0.30 \text{ g of sucrose}/100 \text{ g root/day}$  over 3 days) and in similar experiments carried out in 1983 (Baraldi *et al.*, 1984) ( $0.32 \text{ g of sucrose}/100 \text{ g root/day}$  over 3 days and  $0.26 \text{ g of sucrose}/100 \text{ g root/day}$  over 6 days).

Christodoulou *et al.* (1983) found that the sucrose losses on Greek beets, normally harvested and stored for 10 days, ranged from  $0.075\%$  to  $0.166\%$ . After 24 h following the damage, the sucrose loss progressively declines as a result, probably, of the healing phenomena.

These conclusions agree very well with the laboratory results obtained by Dilley *et al.* (1970).

**TABLE 2**  
Raffinose and Lactic Acid Contents in the Extraction Juice obtained from Damaged Sugar Beet Roots Stored on the Ground

<i>Sample</i>	<i>Raffinose</i> (g % Pol.)	<i>Lactic acid</i> (mg % Pol.)
1	0.33	2.3
2	0.34	9.3
3	0.32	14.0
4	0.30	15.5
5	0.31	17.4

These authors checked the respiration rate on beet previously cleaned and cut artificially; they calculated losses of sucrose of about 0.24 g/100 g root/day over the first 24 h and then a drastic decrease during the following days to the value of 0.06 g/100 g root/day.

Table 2 shows that the raffinose content does not change during brief storage, in accordance with literature data (Vukov & Hangyal, 1985), while there is a very small increase in lactic acid even if it does not exceed 20 mg% Pol in the extraction juice at the end of the storage.

The levels of enzyme specific activity are shown in Table 3.

**TABLE 3**  
Variation of Enzymatic Activities in Damaged Sugar Beet Roots Stored on the Ground. The Activities of Acid and Neutral Invertase and Sucrose Synthetase are Expressed in Terms of  $\mu$ moles of Sucrose Hydrolyzed  $.1/2\text{ h}^{-1} . \text{mg}^{-1}$  Protein. Differences between Storage Periods on the Ground in Hours were Measured by Analysis of Variance and Neuman-Keuls Test on the Minimum Significant Differences

<i>Storage period on the ground (h)</i>	<i>Acid invertase</i>	<i>Neutral invertase</i>	<i>Sucrose synthetase</i>
0 (T1)	4.48 <sup>ab</sup>	3.76 <sup>a</sup>	2.43
15.30 (T2)	4.16 <sup>a</sup>	3.93 <sup>ab</sup>	2.45
24 (T3)	5.78 <sup>c</sup>	5.19 <sup>ab</sup>	2.53
39.30 (T4)	5.20 <sup>bc</sup>	5.27 <sup>b</sup>	2.04
63.30 (T5)	5.43 <sup>bc</sup>	4.98 <sup>ab</sup>	2.54
Residual standard deviation	0.8949	1.2366	0.8175
Degrees of freedom	43	43	48

<sup>a,b,c</sup> Means in the same row with unlike superscripts differ ( $p < 0.05$ ).

The specific activity of sucrose synthetase remained unchanged as previously determined on undamaged beets, beets damaged due to dropping on a concrete surface (Vaccari *et al.*, 1983) and beets damaged by use of skips (Baraldi *et al.*, 1983).

On the other hand, the specific activities of acid and neutral invertase behave differently.

Acid invertase of time  $T_2$  presents a significantly lower value ( $p < 0.05$ ) than that of times  $T_3$ ,  $T_4$ ,  $T_5$ , and is of the same order of magnitude as time  $T_1$ .

The specific activity of neutral invertase has, on the other hand, a significantly lower value ( $p < 0.05$ ) at time  $T_1$  than at time  $T_4$ , while this latter does not differ from  $T_2$ ,  $T_3$ ,  $T_5$ .

These results indicate that, during the first hours of beet storage, in addition to acid and neutral invertases, sucrose synthetase is also metabolically active in sucrose hydrolysis in the harvested beets in agreement with Turner & Turner (1975) and Gander (1976). According to these authors, the enzyme is mainly involved (despite its name) in sucrose breakdown rather than in synthesis.

The initial decrease of reducing sugars could be justified by lower observed invertase activity and especially by changes of respiration rates of sugar beet roots that are increased by the level of mechanical damage (Cole, 1977; Wyse, 1978) because the beet had to heal its bruises (Devillers, 1981).

Moreover, taking into account the high increase in respiration phenomena during the first hours following the damage (Dilley *et al.*, 1970), we could deduce that the consumption of reducing sugars during this time is higher than their production by enzymes; consequently, the root utilizes part of its reserve of reducing sugars.

When the respiration declines, because the process of healing is finished, the production of invert sugars exceeds their consumption and consequently their content increases.

Vukov & Hangyal (1985) reported that the reducing sugar content in harvested beets is the result of two processes: enzymatic sucrose hydrolysis and invert sugar oxidation, so that the content of this latter depends on the rates of the two processes.

## CONCLUSIONS

The results obtained indicate that the consequence of damage after a short storage seems limited; however, if we extrapolate the whole of the data into daily processed beets for all of the sugar campaign, the sugar loss is significant.

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