

IMPACT OF ROOT DISEASES ON STORAGE: CARBOHYDRATE IMPURITY FORMATION

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Introduction

Sugarbeet roots contain a number of nonsucrose carbohydrates that co-extract with sucrose during processing. These carbohydrate impurities are present at low concentrations relative to sucrose, but have a significant impact on sugarbeet processing quality and sucrose yield. Carbohydrate impurities form during the production and postharvest storage of sugarbeet roots with the largest accumulation of impurities occurring during storage. The impurities are products of sucrose metabolism and are formed either by the action of endogenous sugarbeet enzymes or enzymes from pathogenic organisms living on or within the root. As metabolic derivatives of sucrose, carbohydrate impurities are directly responsible for sucrose loss in sugarbeet roots. Their impact on sucrose yield is compounded by their ability to interfere with processing. Carbohydrate impurities cause color, crystallization and filtration problems during sugarbeet root processing and increase the loss of sucrose to molasses.

Carbohydrate impurities include monosaccharides, oligosaccharides and polysaccharides. The major monosaccharide impurities in sugarbeet root are the invert sugars, fructose and glucose. These two sugars are formed by the enzymatic degradation of sucrose, either by the endogenous sugarbeet root enzymes, invertase and sucrose synthase, or by invertases of sugarbeet root pathogens. Invert sugars co-extract with sucrose, but degrade during processing to organic acids and colored compounds. Their degradation products require the addition of reagents to increase the alkalinity of the extraction juice and reduce the yield of white sugar. The major oligosaccharides in sugarbeet roots are the trisaccharides, raffinose, 1-kestose, 6-kestose and *neo*-kestose. Raffinose is formed by the addition of a galactosyl residue to sucrose by raffinose synthase, an endogenous sugarbeet root enzyme. Raffinose is typically present at harvest and generally increases during storage. The kestoses form by the addition of a fructosyl residue to sucrose and differ from each other by the linkage position of the fructose moiety. Kestose formation is a byproduct of invertase activity which may be of plant or pathogen origin. In sugarbeet processing, raffinose and the kestoses co-extract with sucrose without degradation. Their presence significantly reduces the rate of sucrose crystallization and alters sucrose crystal morphology causing an increase in sucrose loss during crystal filtration. The major polysaccharide impurities in sugar beet root are the gums, dextran and levan. Dextran and levan are high molecular weight polymers of glucose and fructose that are enzymatically produced by bacteria that colonize sugarbeet roots after cells are disrupted by injury. To date, gum formation has only been reported to occur in frost damaged roots. Gums are particularly problematic impurities in sugarbeet processing since their presence can cause serious filtration problems.

The formation of carbohydrate impurities during sugarbeet root storage has been examined in healthy roots, frost damaged roots and roots after infection by storage pathogens. To our knowledge, no studies have examined the type and quantity of carbohydrates impurities that accumulate during storage of roots exhibiting root rot symptoms at harvest. As root rots, especially *Aphanomyces* root rot, become more prevalent in the Red River Valley of Minnesota and North Dakota, information regarding the storage and processing characteristics of diseased roots is needed to assess the value and feasibility of harvesting and storing diseased roots.

Materials and Methods

Mature sugarbeet roots of three varieties (Hilleshög Horizon, Beta X709 and Maribo 9363) were harvested from three commercial fields on September 29, 2000 and separated into four groups based on severity of visible disease symptoms. The four groups of disease severity were 1) no rot: roots with no visible symptoms of root rot, 2) russeted: roots exhibiting small areas of russetting due to root infection, 3) moderate: roots exhibiting a moderate degree of infection affecting less than 25% of the surface of the root and 4) severe: roots exhibiting severe symptoms of root rot affecting more than 50% of the root surface. Diseased roots exhibited primarily symptoms of *Aphanomyces* root rot, although other root diseases were also likely to be present, especially in roots exhibiting severe rot symptoms. Roots were hand harvested, washed and placed in storage at 4°C (40° F) and 95% relative humidity. Roots were sampled after 18 and

85 days in storage by removal of a 1.1 cm core section through the widest portion of the root. Samples were rapidly frozen with $N_2(liq)$, lyophilized, ground to a fine powder and analyzed for carbohydrate impurities by gas chromatography as described by Long and Chism (1987). Representative samples of roots exhibiting no rot and severe rot symptoms were also analyzed by high performance liquid chromatography by N. Jerry Chatterton, USDA/ARS, Forage and Range Research Laboratory, Logan, UT to determine relative quantities of raffinose, 1-kestose, 6-kestose and *neo*-kestose (Chatterton *et al.* 1993). Data was analyzed with Duncan's multiple range test using a 95% confidence interval to determine significance of differences.

Results

Changes in carbohydrate impurities during storage of sugarbeet roots affected with root rot at time of harvest were investigated in a small pilot study. Specifically, changes in the major sugarbeet root monosaccharide and oligosaccharide impurities were determined. The concentrations of polysaccharide impurities were not determined in this study, but will be examined in future studies.

Monosaccharide impurities: The concentrations of the two major monosaccharide impurities, fructose and glucose, were significantly elevated in sugarbeet roots exhibiting severe symptoms of root rot (Figure 1A and B). Fructose and glucose concentrations, however, were not different in roots exhibiting symptoms of no rot, russetting or moderate rotting. Severely rotted roots contained 2.5-fold more fructose and 1.9-fold more glucose than roots with no rot symptoms after 18 days in storage on a fresh weight basis. With additional time in storage, the fructose and glucose content of severely rotted roots increased, and after 85 days, severely rotted roots contained 4.5-fold more fructose and 2.9-fold more glucose than roots exhibiting no rot symptoms. In roots exhibiting no rot, russetting or moderate rot symptoms, fructose and glucose content decreased with storage to 85 days, although the decline was not statistically significant.

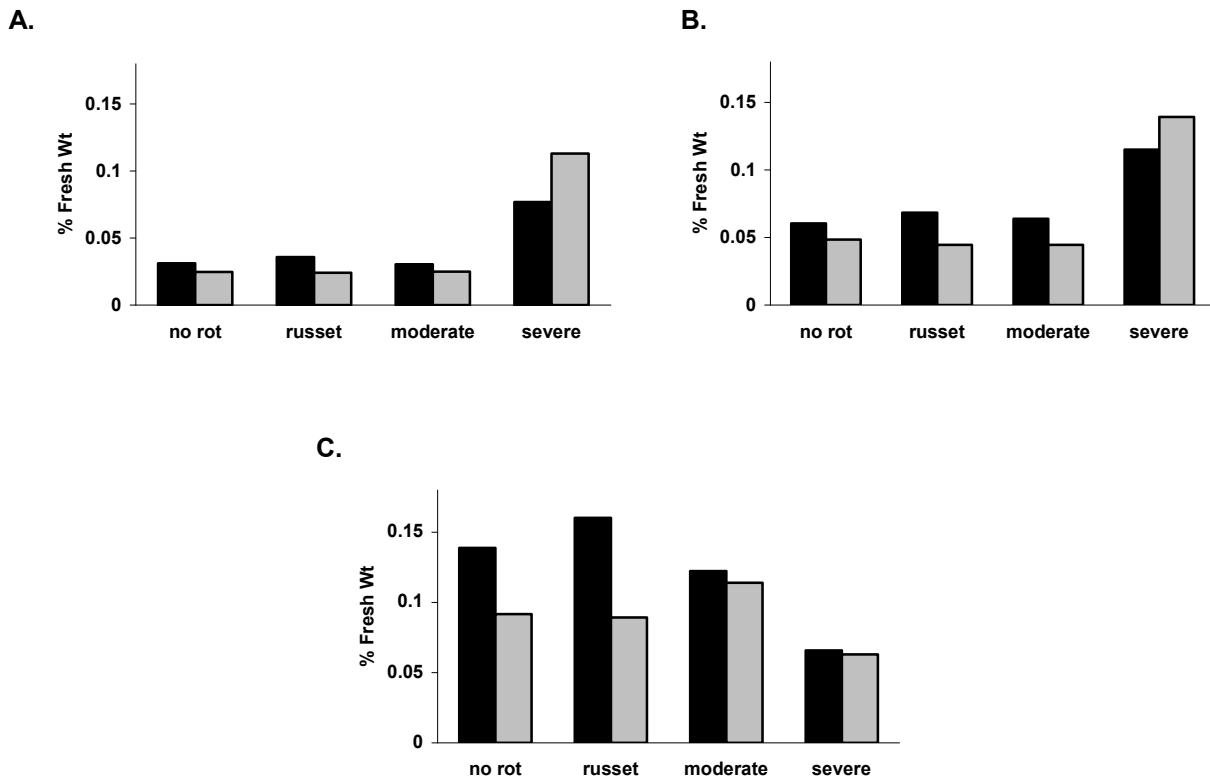


Figure 1: Change in carbohydrate impurities in sugarbeet roots with varying degrees of root rot severity after: \surd 18 days, and \surd 85 days in storage at 4°C and 95% relative humidity. **A.** Change in fructose content. **B.** Change in glucose content. **C.** Change in trisaccharide content.

Oligosaccharide impurities: The GC assay used in this study was unable to separate raffinose, 1-kestose, 6-kestose and *neo*-kestose. These trisaccharide impurities, therefore, were quantified together. Generally, the trisaccharide content declined with an increase in root rot severity, although this decline was only statistically significant in severely rotted roots ([Figure 1C](#)). Severely rotted roots contained approximately half the trisaccharide content of healthy roots. The trisaccharide content of roots of all disease severities declined with storage from 18 to 85 days. The greatest decline in trisaccharide content occurred in roots exhibiting symptoms of no rot or russeting. To ascertain the relative contribution of raffinose and the kestoses to the total concentration of trisaccharides, representative samples of roots with no rot and severe rot symptoms were subjected to HPLC analysis. Raffinose was the only trisaccharide detected in roots exhibiting no rot symptoms. In roots with severe rot symptoms, small quantities of 1-kestose and 6-kestose were also found. *Neo*-kestose was not detected in any of the samples analyzed.

Conclusions

The results presented are the product of a small pilot study on the impact of root disease on sugarbeet root storage properties. Any conclusions drawn from these studies, therefore, should be tempered with the knowledge that this study was conducted with a limited number of roots from a single harvest year. Nevertheless, in these initial studies, the following were observed:

1. Invert sugars were elevated in sugarbeet roots exhibiting severe root rot symptoms. No increase in invert sugar concentration was observed in russeted or moderately rotted roots. In severely rotted roots, invert sugars were present at sufficient levels to cause severe processing problems.
2. Prolonged storage increased invert sugar concentration in severely rotted roots, although no accumulation of invert sugars was observed in roots with no rot, russeting or moderate rot symptoms. The lack of accumulation of invert sugars in roots with no rot, russeting and moderate rot symptoms suggests that any sucrose degraded during storage (see accompanying report: Impact of root diseases on storage: Extractable sucrose and respiration, Campbell and Klotz) was completely utilized either by the sugarbeet root or its accompanying pathogenic organisms.
3. Raffinose was the major trisaccharide impurity in sugarbeet root, regardless of the severity of root rot disease or time in storage. Kestoses were not detected in healthy roots and were evident in severely diseased roots at low concentrations.
4. Trisaccharide content was lower in sugarbeet roots exhibiting severe rot symptoms. The decline in trisaccharide content with disease severity was unexpected and suggests that raffinose accumulation was impaired in diseased roots, or alternatively, raffinose degradation was accelerated in diseased roots.
5. Trisaccharide content of sugarbeet roots declined during storage, regardless of the severity of disease symptoms. This decline suggests that raffinose was metabolized during storage.

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Literature Cited

Chatterton NJ, Harrison PA, Thornley WR, Bennett JH. 1993. *New Phytologist* 124: 389-396.
Long AR, Chism GW. 1987. *J. Food Sci.* 52: 150-154.