# Effect of Nitrogen fertilizer on Cold-Induced Sweetening in potato tubers during storage

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## Summary

Nitrogen (N) fertilizer is used routinely in potato cultivation to optimize yield. However, the role of N fertilization on potato post-harvest storage and reducing sugar accumulation is less conclusive. Data collected from a greenhouse study have shown increased levels of leaf chlorophyll, higher biomass, increased soluble proteins and reduced partitioning of fresh weight to tubers in response to increasing N rate. Results from a field study conducted in 2018 involving six cultivars grown under three N fertilizer regimes showed altered cell N status and enzymes expression at harvest. Altered key enzymes expression may impact long term storage and processing quality of tubers.

## Introduction

Balancing economic yield with environmental concerns associated with N fertilization is often challenging. Commercial potato production is especially prone to environmental contamination when N fertilizer irrigation and unpredictable rainfall results in nitrate leaching (1). Excessive loss of nitrate from the potato root zone is a serious environmental problem (2). The consequences of nitrate leaching have prompted policy makers and society to search for mitigating options. In addition to environmental concerns, excessive available N stimulates top growth and delays tuber formation and maturity. Nitrogen use efficiency has been shown to decrease in a curvilinear manner with increasing crop N supply (3).

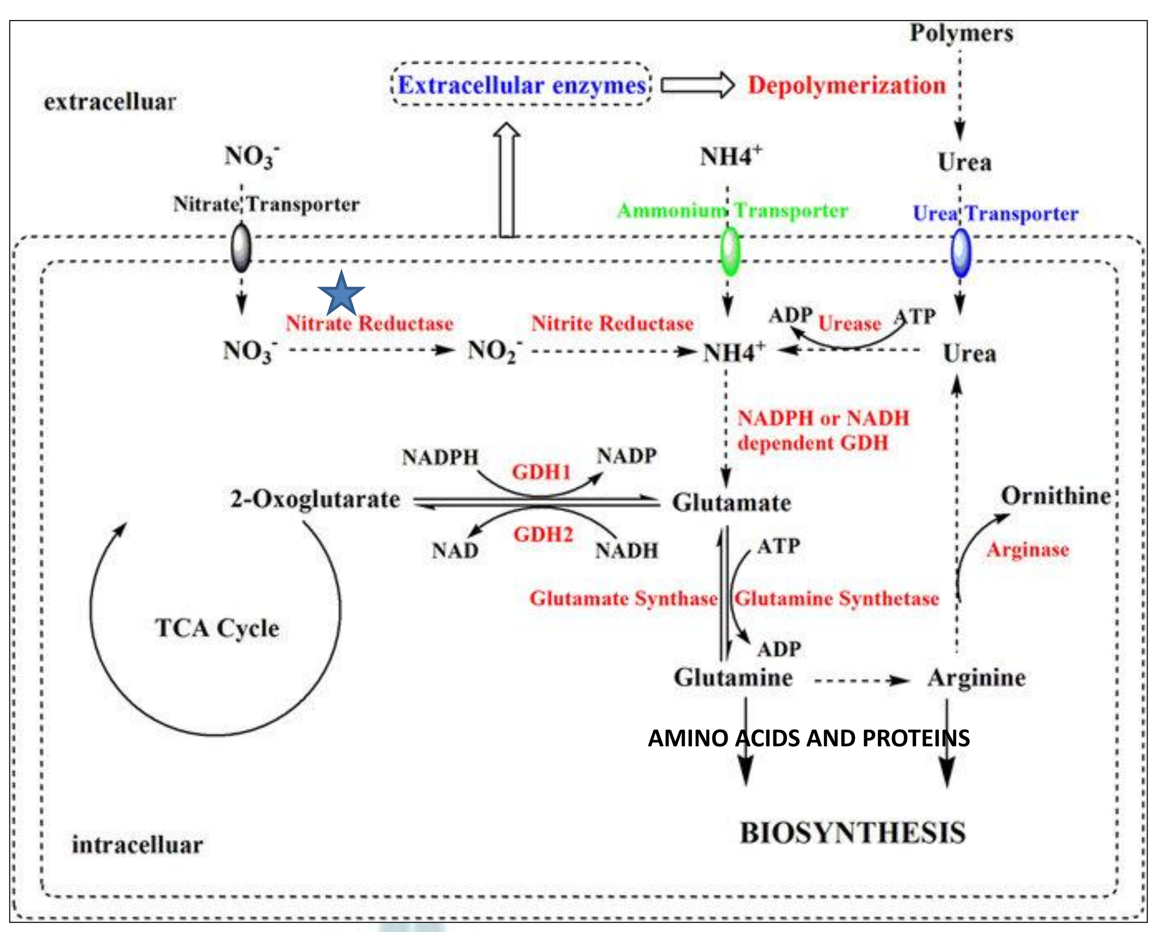
N fertilization influences tuber sugar content and fry color by interfering with tuber chemical maturation (4,5). Systematic studies are lacking on the effect of N fertilization on expression of various enzymes related to carbohydrate metabolism in potato tubers. Studies have shown a close association of key enzymes with reducing sugar (RS) accumulation. Changes in carbohydrate metabolizing enzyme expression in response to N status may have significant effects on tuber RS accumulation during storage.

The objectives of this study are to:

1. Explore the effect of N fertilization on concentrations of RS accumulation and the underlying cellular mechanisms and

2. Screen potato cultivars that can perform well under low N input conditions.

Figure 1. $NO_3^-$ absorption by the roots and assimilation into soluble proteins. The first two key enzymes are Nitrate Reductase (NR) and Nitrite Reductase (NiR).

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## Materials & Methods

To gain a better understanding of nitrogen use efficiency, six potato cultivars and clones (Russet Burbank, Umatilla, Clearwater, Proprietary Var, Lamoka and MN13142) having a wide variation in their Cold-Induced Sweetening (CIS) resistance were selected.

In 2018, the cultivars were planted at Sand Plain Research Farm, Becker, MN in a Hubbard loamy sand soil. A randomized complete block design with three replication was used. Three N rates (120, 240, and 360 lbs/Ac) were evaluated. All potatoes were harvested in early October and suberized for three weeks at room temperature.

Five tubers from each plot were analyzed for sugars, fry color and other traits at the bud end and the stem end. For biochemical analysis, 5 grams tissue was ground with liquid nitrogen and extracted with HEPES extraction buffer pH 7.5. All steps were conducted on ice.

Soluble protein content was determined using the dye-binding method of Bradford (6) and expressed as mg per g FW. A microplate based method was used for basal and total acid invertase activity determination (7). Enzyme activity was expressed as units (µmols Glc formed per hour) per mg protein.

Sugars, glucose and sucrose were analyzed using a YSI model 2000 Industrial Analyzer (Yellow Springs Instruments Co., Inc., Yellow Springs, OH). The concentration of sugar was expressed in mg $g^{-1}$ FW.

## Results

Figure 2. Soluble protein concentration at harvest. Data represents means of 3 reps ± SE

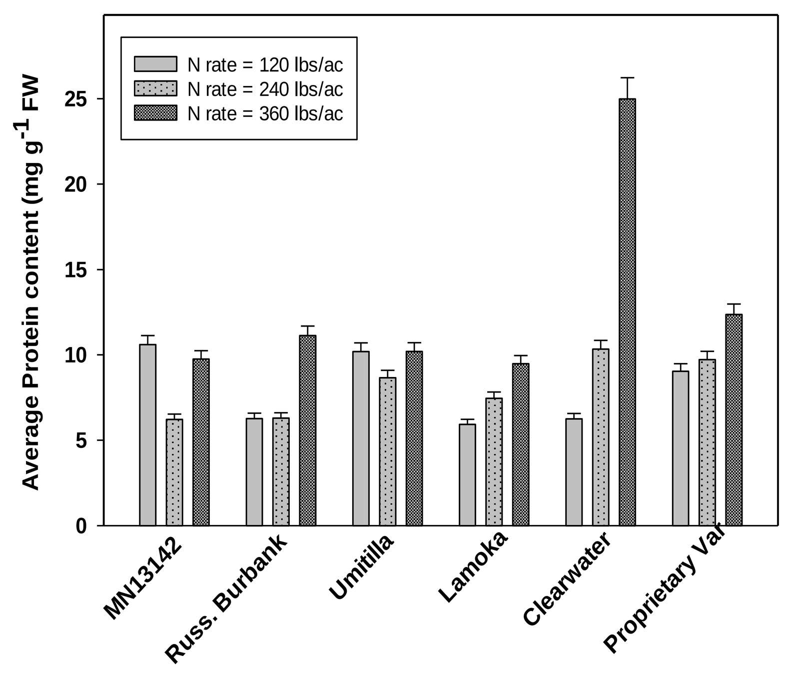



Figure 3. Basal acid invertase enzyme activity at harvest. Data represents means of 3 reps ± SE

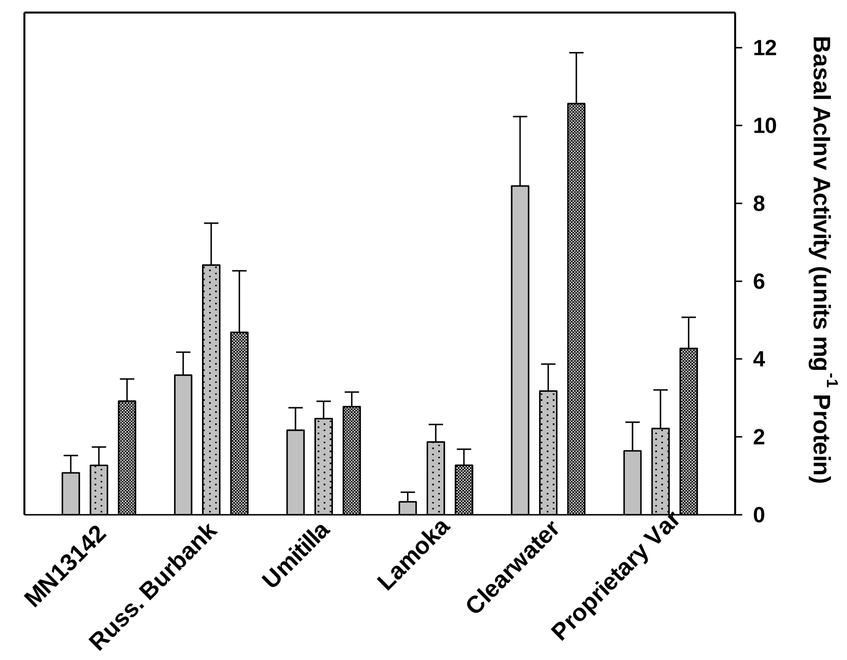



Figure 4. Average glucose conc. at stem and bud end of tubers at harvest. Data represents means of 3 reps ± SE

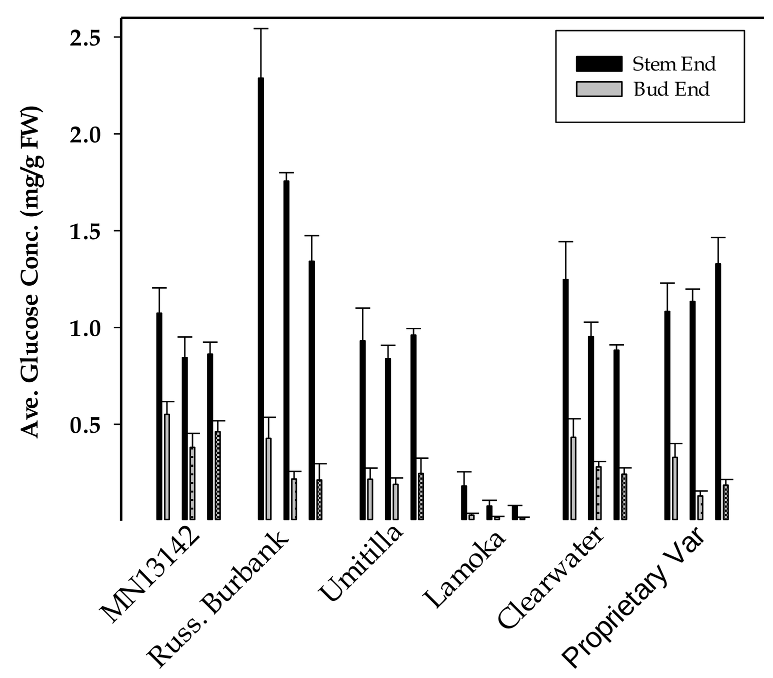



Figure 5. Average reflectance at stem and bud end of tubers at harvest. Data represents means of 3 reps ± SE

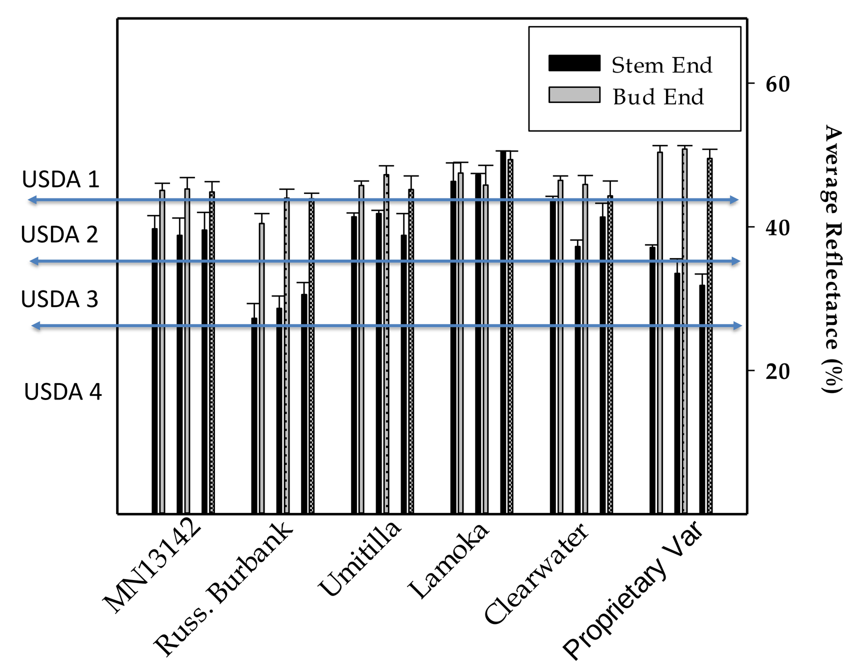



Figure 6. Average sucrose conc. at stem and bud end of the tubers at harvest. Data represents means of 3 reps ± SE

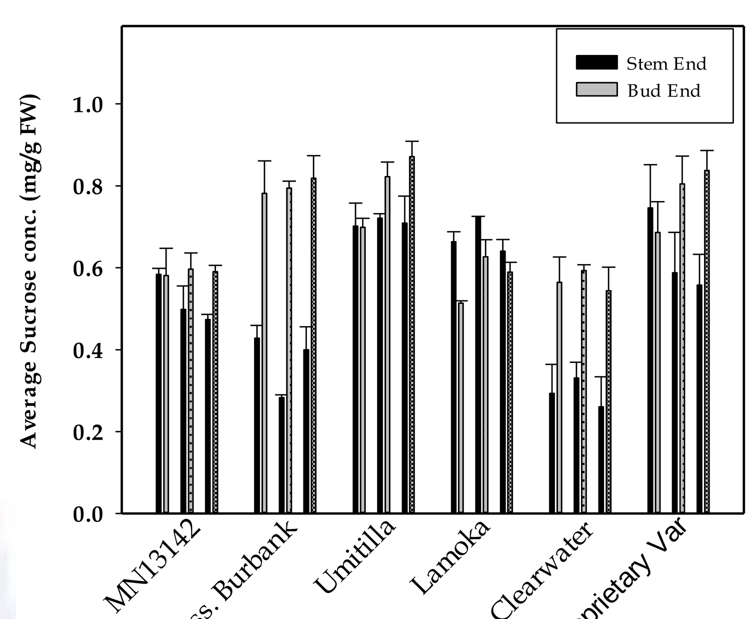
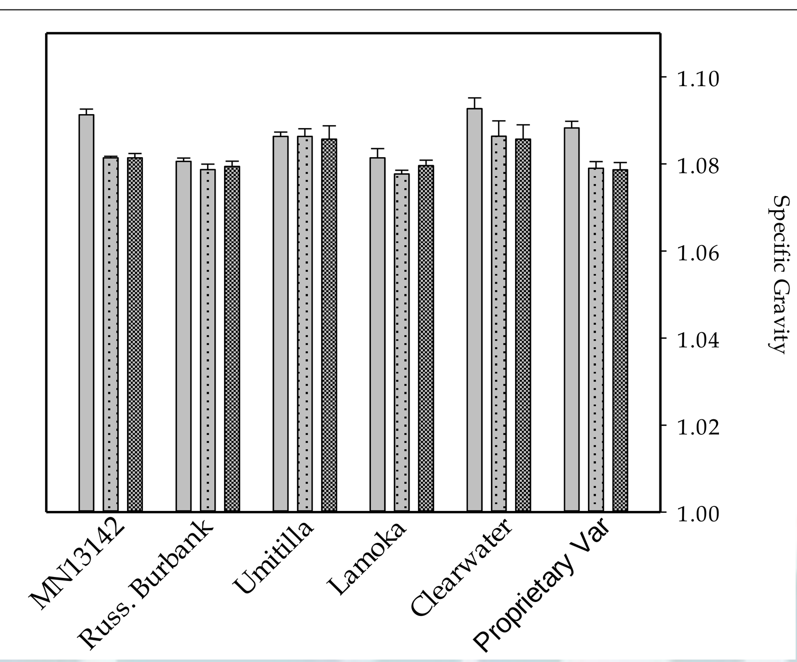



Figure 7. Average specific gravity of the tubers at harvest. Data represents means of 3 reps ± SE .




N rates = 120, 240 and 360 lbs/ac

## Discussion

The effects of N fertilizer on soluble protein content, RS and enzyme activity was cultivar-specific. Soluble protein content increased with increasing N rates in four of the six cultivars (Fig 2). High N status in the cell can result in high metabolic activity of key starch synthesizing enzymes (8–10).

The increase in soluble protein content resulted in higher expression of starch metabolizing and other enzymes. Muttucumaru et al. (11) reported a substantial increase in asparagine and total free amino acid in response to increasing N fertilization.

N supply has been reported to affect the sugar concentration and interconversion of simple sugars and complex carbohydrates such as fructans (12). Russet Burbank and Lamoka had higher AcInv at 240 lbs/ac than 120 and 360 lba/ac N fertilizer rate (Fig 3). Altered AcInv activity did not result in proportional sugar contents change at harvest (Fig 4 & 6) due to differential expression of AcInv inhibitor protein (data not presented).

RS decreased in Russet Burbank, MN13142 and Clearwater as N rate increased. Proprietary var and Umitilla showed a reverse trend. The difference in glucose levels between stem and bud end was very low in Lamoka and MN13142, indicative of low or no stem end defects. All the cultivars except Russet Burbank had acceptable fry color (Fig 5). MN13142 and Proprietary var showed significant decrease in specific gravity as N rate increased (Fig 7).

## Conclusions & Future Research

- High metabolic activity was recorded at the bud end of the tubers in all cultivars. Higher N levels increased expression of enzymes including AcInv that is related to RS accumulation during cold storage.
- Higher levels of AcInv enzyme activity at harvest may lead to higher RS accumulation during storage.
- Altered expression of invertase inhibitor in response to N fertilizer rate needs to be further explored.
- Tuber proteome analysis will be used to identify differentially expressed key enzymes involved in N uptake and carbohydrate metabolism (Fig. 1).

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