The Influence of Stage of Maturity, Level of Nitrogen Fertilization and Storage on the Concentration of Solanine in Tubers of Three Potato Cultivars¹

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The glycoalkaloid solanine, glycoside of solanidine, is the principal, naturally occurring toxicant (acetlycholinesterase inhibitor) found in white potatoes. Numerous accounts of solanine causing intestinal discomfort and occasional illness in humans and livestock have been reviewed by BOMER and MATTIS (1924) and GULL (1960). The cause of poisoning has been generally associated with consumption of immature, green, small or well-sprouted tubers containing excessive concentrations of solanine.

The concentrations of solanine found in commercial potatoes are normally less than 12 mg per 100 g of fresh tuber. However, during 1969 a new processing cultivar, Lenape (USDA seedling number B5141-6), was shown by ZITNAK and JOHNSTON (1970) to contain approximately twice the concentration of solanine present in commonly grown commercial cultivars. The concentration of solanine in Lenape was mostly in excess of 20 mg per 100 g of fresh tuber weight. While the actual level of toxicity of solanine has not yet been established, a persistently acrid taste becomes quite distinct upon ingestion of tubers containing 20 or more mg of solanine per 100 g of fresh tuber.

Released specifically in 1967 as a chipping potato with a high solids and low sugar content, Lenape potatoes were quickly accepted by processors (AKELEY et al. 1968). Although it was thought that chips made from Lenape posed no health hazard, the USDA withdrew the cultivar name and thus terminated seed certification, primarily on the basis that consumption of whole boiled or baked potatoes might produce discomfort or even illness (ANONYMOUS 1970). Hence, Lenape is referred to in the remainder of this report by its seedling number (B5141-6).

This study was initiated to determine the influence of stage of maturity, level of nitrogen fertilization and storage on the accumulation of solanine in B5141-6 tubers and two common varieties, Russet Burbank (Netted Gem), and Katahdin. Russet Burbank tubers occasionally contain high concentrations of solanine (ZITNAK 1961), while the solanine in Katahdin is typical of most other commercial cultivars.

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Experimental

A split-plot field design with four replications was employed. Main plots provided three stages of maturity. Maturity-1 tubers were harvested 118 days after planting from a main plot which had been sprayed 12 days before harvest with Dow Premerge, a vine killer. Maturity-2 tubers were harvested 118 days after planting from a second main plot. Maturity-3 tubers were harvested 146 days after planting from a third main plot.

Each main plot was subdivided into two levels of nitrogen fertilization, i.e.: Nitrogen 1, consisting of an application of 135.3 kg per ha (a normal level) and Nitrogen 2 consisting of an application of 202.9 kg of nitrogen per ha (a level in excess of that required for optimum production). Phosphorus and potassium applications remained constant for both nitrogen treatments of 112.2 kg per ha. Each of the three cultivars was randomized within each subplot.

Freshly harvested tubers were removed immediately from the field and graded. Size grades retained for fresh analyses ranged in diameter from 6.0 to 7.6 cm for B5141-6 and Katahdin and 3.8 to 6.0 cm for Russet Burbank. B5141-6, Katahdin and Russet Burbank tubers selected for analysis averaged 245, 260, and 275 g, respectively. As seen in Table 1, no significant difference in solanine concentrations occurred in B5141-6 tubers ranging in weight from 200-300 g, whereas the concentration in 100 g tubers was significantly higher. After selection of the fresh samples for laboratory analyses, all remaining tubers were maintained in commerical storage at 10°C and 95% relative humidity for 45 days.

TABLE 1.

The influence of tuber size on the concentration of solanine in B5141-6

Solanine Concentration (mg per 100 g fresh tuber)	
34.9 a*	
28.5 b	
26.4 b	

^{*} Means followed by the same letter are not significantly different from each other at the 1% probability level.

Sample Preparation: Each sample consisted of six randomly selected tubers. Each tuber was divided into eight equal sections parallel to the stem axis, one section of which was retained from each tuber for a composite sample. The composite sample was diced, weighed, combined with an equal weight of 95% ethanol and blended for 1.5 minutes. While maintain-

ing a uniform suspension with the use of a magnetic stirrer, 200 g of the macerate were transferred to sample bottles, sealed and stored at -35° C until analyzed.

Solanine Analysis: Solanine was extracted from duplicate 40 g subsamples according to the procedure of BAKER et al. (1955). Briefly, this included a Soxhlet extraction for 16 hours with 120 ml of 95% ethanol and 3 ml of glacial acetic acid. The Soxhlet extracted residue was rinsed with 10 ml of 3% acetic acid, followed by 10 ml of 95% ethanol. The extract and rinsings were transferred to a 400 ml beaker and evaporated slowly to a 3 to 4 ml volume to which 5 ml of 5% H₂SO₄ were added. The extract was filtered through a 5.5 cm glass fiber filter disc* and rinsed with 10 ml of 5% H₂SO₄ to remove flocculent lipid material. The combined filtrates to which 8 ml of concentrated NH₄OH were added, were transferred to conical centrifuge tubes. After 20 minutes the tubes containing the alkaline suspension were placed in an 80°C water bath for 20 minutes to flocculate the solanine. Subsequently, the tubes were held overnight at 4°C.

The solanine fraction was separated by centrifugation at 32,000 rpm for 20 minutes and the supernatant discarded. The precipitate was suspended in 15 ml 1% NH $_4$ OH and centrifuged as above. The supernatant was discarded.

The colorimetric procedure of WIERZCHOWSKI and WIERZCHOWSKA (1961) was used in determining the concentration of solanine. By this method the washed precipitate was dissolved in 10 ml of 1% H₂SO₄ and centrifuged to remove any undissolved material. A fresh solution of SbCl₃ was prepared daily, approximately two hours in advance of use from reagent grade SbCl₃ and concentrated HCl (3:2 w/v). A 1.0 ml aliquot of acidified extract, appropriately diluted to obtain a solanine concentration of 0 to 0.6 mg per ml was combined and mixed thoroughly with 4 ml of SbCl₃. After 1 hour, optical density was determined using a Beckman Model B spectrophotometer at a wavelength of 525 Mµ. A regression coefficient was calculated using purified solanine**standards.

Results and Discussion

The solanine concentration was 108 to 282% and 173 to 404% higher in B5141-6 than in Russet Burbank and Katahdin, respectively (Table 2). The means of all treatments for Russet Burbank and Katahdin coincide well with the 2 to 15 mg per 100 g fresh tuber concentration considered as normal for commercial potatoes. Except for one treatment, solanine in B5141-6 exceeded 20 mg per 100 g which is in agreement with ZITNAK and JOHNSTON (1970).

Reeve Angel, grade 934AH

Analytical solanine, Lot No. 82155, obtained from K & K Laboratories, Inc. Plainview, New York.

TABLE 2

The influence of stage of maturity, level of nitrogen fertilization and storage on Solanine⁸ in B5141-6, Russet Burbank and Katahdin.

Variety	Nitrogen		Maturity ^C		
	Application	1	2	3	
		Fresh Tubers			
B5141-6	1 2	20.8ac ^{1/} 17.5a	23.5b 28.9b	20.5c 20.7c	
Russet Burbank	1 2	8.0a 8.4a	7.4ab 7.3ab	6.9b 6.5b	
Katahdin	1 2	4.8a 6.4a	5.8b 6.3a	17.8c 8.5b	
	, , , , , , , , , , , , , , , , , , ,	Stored Tubers			
B5141-6	1 2	30.4a 33.1a	27.4b 41.3b	37.7c 40.8b	
Russet Burbank	1 2	11.8a 12.6ac	11.8a 10.8b	13.0b 12.7c	
Katahdin	1 2	7.5a 9.1a	8.7a 8.2a	11.4b 11.1b	

Each entry is a mean for four replications and duplicate analyses expressed as as mg per 100 g of fresh tuber.

Nitrogen means within a variety and maturity level prefaced by a common vertical line are significantly different at the 5% probability level.

Nitrogen 1: Normal rate, 135.3 kg per ha Nitrogen 2: Excessive rate, 202.9 kg per ha

Maturity 1: Vine killed 106 days; harvested 118 days after planting

Maturity 2: Harvested 118 days after planting Maturity 3: Harvested 146 days after planting

Maturity means within a variety and nitrogen treatment followed by the same letter are not significantly different at the 5% probability level.

The solanine in fresh tubers of B5141-6, was significantly higher in maturity 2 (28.9 mg) than in maturity 1 (17.5 mg) and 3 (20.7 mg). In this study the first harvest of B5141-6 may have occurred during a phase of active synthesis or translocation, wherein maturity 2 compared to 1 (vine killed) was permitted to accumulate solanine an additional 12 days, and thus would have exhibited a higher concentration. Maturity 3 may have been harvested during a phase of additional tuber growth lacking further accumulation of solanine, which would explain, by a "dilution effect", the lower solanine concentration. Solanine declined linearly throughout the maturity stages observed in fresh Russet Burbank tubers. Whereas in Katahdin, the solanine increased gradually throughout the same intervals. Similar patterns of variation in solanine content of tubers during the growing season were reported by WOLF and DUGGER (1946). Excessive nitrogen fertilization resulted in significantly more solanine in fresh B5141-6 tubers at maturity 2 and Katahdin tubers of maturities 1 and 3.

Stored tubers contained signficantly more solanine than fresh tubers for all comparisons (CRONK 1971). Compared to fresh tubers, stored samples of B5141-6 from plots fertilized with excessive nitrogen increased in solanine 89, 43 and 97% for maturities 1, 2 and 3, respectively. Similarly, solanine in stored B5141-6 tubers from normal nitrogen plots increased 46, 17 and 84% for maturities 1, 2 and 3, respectively. However, stored B5141-6 tubers from high nitrogen plots were significantly higher in solanine than those from normal nitrogen plots. Solanine increased in the range of 48 to 60% in stored Russet Burbank tubers of maturites 1 and 2. Tubers of maturity 3 contained 88 and 95% more solanine for the normal and excessive nitrogen treatments, respectively. A similar increase in solanine was observed in stored Katahdin tubers.

The increase of solanine in stored tubers compared with fresh tubers is without precedent. The 45-day period of storage in this study was considerably less than the three months initial sampling reported by other investigators (HILTON 1951, HILTON and GAMBORG 1957, LAMPITT et al. 1943). WANG (1970) also noted an increase in glycoalkaloid* content, but only as a result of a rise in storage temperature; however, he also observed that the glycoalkaloid levels of potato tubers of four cultivars generally decreased during controlled storage at 4.5 and 10°C. Most of the decrease occurred during the first two months of storage. A plausible explanation for the increase in solanine concentration observed in this study may be related to the time of sampling as well as temperature. The 45-day period may represent a point at which solanine is at its highest level and subsequently decreases with additional time in storage.

Solanine is the predominant, but not the exculsive glycoside present in white potatoes.

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Summary

Results indicated that the accumulation pattern of solanine during the harvesting intervals observed differed markedly between cultivars and was influenced by the stage of maturity. The influence of nitrogen fertilization on solanine was inconsistent. All stored tubers of all varieties at 10°C for 45 days exhibited a highly significant increase in solanine.