

Genetic and Environmental Effects on Levels of Glycoalkaloids in Cultivars of Potato (*Solanum tuberosum* L.)

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ABSTRACT

Glycoalkaloids were assayed in fifty-five potato cultivars, fifty-four breeding lines and one other species (*S. stolonifer*). The total glycoalkaloid content ranged from 16.3 µg/g FW for Alpha to 317.0 µg/g FW for Berita, with most values lying between 35 and 65 µg/g. The α -solanine content, as a percentage of total glycoalkaloids, ranged from 28.3% for Avenir to 57.0% for H42, with the majority of values lying between 38% and 46% α -solanine. There was a highly significant correlation between high total glycoalkaloid content and per cent α -solanine ($P < 1\%$). The presence of β_2 -chaconine was also related in a highly significant way to high total glycoalkaloid content. Potatoes grown at Yanco (hot, dry, inland climate) contained more glycoalkaloids (~60%) than when grown at Glen Innes (cooler, high altitude climate). However, there was no significant difference between total and relative glycoalkaloid levels of cultivars grown at Glen Innes and Healesville (coastal, temperate climate). A significantly higher per cent α -solanine content, but not total glycoalkaloid content, was observed for potatoes grown in the second year at Glen Innes.

INTRODUCTION

All potato cultivars and breeding lines contain glycoalkaloids. At low levels (<100 µg/g FW) they are a normal constituent of potatoes and

contribute to the typical flavour of a potato (Ross *et al.*, 1978). However, when present at higher concentrations ($> 100 \mu\text{g/g}$ FW), they may cause a bitter, burning taste (Sinden & Deahl, 1976; Ross *et al.*, 1978). Potatoes with more than $200 \mu\text{g/g}$ FW are regarded as unsafe to eat (Bomer & Mattis, 1924; Sinden & Webb, 1972). Cases of poisonings and deaths are reported by Jadhav & Salunkhe (1975) and Morris & Lee (1984).

Glycoalkaloids can accumulate to high levels in potato tubers because of exposure to light, wounding, or chilling (Jadhav & Salunkhe, 1975; Maga, 1980). Previous studies, however, have shown that the base level of glycoalkaloids in potato tubers can vary considerably, because of genetic differences (Sinden & Webb, 1972; Haard, 1977; Butcher, 1978; Ross *et al.*, 1978; Lepper, 1949) and environmental effects such as climate, season and locality (Sinden & Webb, 1978; Ross *et al.*, 1978; Lepper, 1949). Dangerously high levels of glycoalkaloids were found (Zitnak & Johnston, 1970) in the recently released cultivar 'Lenape' and this cultivar had to be withdrawn.

Glycoalkaloids have been assayed in only a limited number of potato cultivars, due to the lengthy analytical methods required. Recently, rapid and accurate HPLC methods have been developed which can measure levels of individual glycoalkaloids (Morris & Lee, 1981). This is the first report of levels of individual glycoalkaloids in a wide range of cultivars, including over forty cultivars and fifty breeding lines not previously studied. These cultivars were examined over a period of two years and at several localities.

MATERIALS AND METHODS

Plant material

Cultivars and breeding lines from the two major potato breeding programmes in Australia were examined over two seasons (1980 and 1981). These programmes are located at the Victorian Department of Agriculture Research Station, Healesville, Victoria (coastal temperate climate) and at the New South Wales Department of Agriculture Research Station, Glen Innes, New South Wales (cooler high altitude climate). Some cultivars and breeding lines were also grown at Yanco, New South Wales (hot dry inland climate). A minimum of twenty tubers of each cultivar, wrapped in light-proof calico and enclosed in hessian sacks, were received from each location.

Glycoalkaloid analysis

Each cultivar or breeding line was analysed in triplicate. Longitudinal sections from six or more potato tubers were blended in a food processor and subsamples of 20 g then blended with 5% acetic acid (3 min, setting 7; Sorvall Omnimixer, Dupont Newton, Con. 06470, USA), and the filtrate made up to 100 ml. In the 1980 samples alkaloids were precipitated by adjusting the pH to 10.5 ± 0.5 with ammonium hydroxide and holding at 70°C for 1 h followed by > 4 h at ambient temperature. The precipitate was washed once with 0.1 M NH₄OH, dried, redissolved in 5% acetic acid and filtered through a 2 µm filter tip (Supelco, Bellefonte, PA, USA). The acid extracts from the 1981 samples were purified by passing 10 ml through a C₁₈ SepPak cartridge (Waters Assoc., Milford, MA 01757, USA), rinsing with 3 ml of 20% ethanol and finally eluting the glycoalkaloids in 1.0 ml ethanol. Between samples the cartridge was rinsed with 4 ml of ethanol and reconditioned with 10 ml of 5% acetic acid.

HPLC quantification was with a C₁₈ Radial Pak column (Morris & Lee, 1981). This method was rapid (3–6 min per sample) and allowed quantification of individual glycoalkaloids. Concentrations of α-solanine and α-chaconine were calculated on peak height using α-solanine (Sigma Chem., St. Louis, MI 63178, USA) as a standard. The concentration of β₂-chaconine was calculated on peak area using a β₂-chaconine standard prepared from α-chaconine (Sigma Chem.) according to Morris & Lee (1981).

RESULTS

Total and individual glycoalkaloid levels for Healesville

The results in Table 1 for potatoes grown at Healesville indicate that total glycoalkaloid levels of tubers can vary greatly, ranging from 16.3 µg/g FW (Alpha) up to a very high and unsafe level of 317.0 µg/g FW (Berita). The majority of the cultivars and breeding lines, 43 out of 84 (this total includes breeding lines not listed in the Table), had glycoalkaloid levels within a narrow range of 45 ± 12 µg/g. The only cultivars which exceeded the maximum limit of 200 µg/g FW (as recommended by Sinden & Webb, 1972) were Berita and Lenape. However, several cultivars and breeding

TABLE 1

The Total Relative Glycoalkaloid Contents of Potato Cultivars and Breeding Lines Grown at Healesville, Victoria, during 1980 and 1981

<i>Cultivar (Year)</i>	<i>Glyco-alkaloid content (µg/g FW)</i>	<i>Percent α-solanine</i>	<i>Cultivar (Year)</i>	<i>Glyco-alkaloid content (µg/g FW)</i>	<i>Percent α-solanine</i>
Abnaki (81)	53.8	31.0	Norchief (81)	96.7	35.5
Alpha (80)	16.3	46.0	Ontario (80)	23.1	44.0
Arran Victory (81)	63.2	32.7	Patrones (81)	77.5	28.9
Arka (81)	33.4	32.8	Pentland Beauty (81)	131.7	40.2
Avenir (80)	48.3	28.3	Pentland Crown (80)	99.8	52.3
Avon (80)	38.7	36.7	Pentland Falcon (80) ^a	36.8	46.3
Berita (80)	317.0	52.7	Pentland Glory (80)	58.6	46.7
Bintje (81)	59.3	34.4	Red La Soda (81)	53.1	38.0
Bison (80)	55.0	44.0	Red Pontiac (80)	36.2	40.7
Brownell (81)	36.5	42.0	Rima (81)	34.2	37.5
Calrose (80)	145.3	45.0	Sebago (80)	43.4	43.7
Coliban (80)	53.3	45.3	Sequoia (81)	50.3	41.6
Conchita (80)	36.7	41.3	Suttons Foremost (80)	50.4	33.0
Cortland (80)	34.8	46.7	Tasman (80)	80.4	44.7
Craig Royal (81)	54.0	37.7	Ulster Sceptre (80)	18.1	40.7
Crispa (81)	42.3	39.9	Ulster Supreme (81)	44.3	38.7
Dalco (81)	32.8	43.5	Up to Date (81)	69.4	42.2
Exton (80)	60.2	44.0	Venus (80)	46.4	46.7
Furore (81)	35.4	30.3	Wauson (80) ^a	52.7	39.7
Green Mountain (81)	52.6	41.2	<i>Solanum stolonifera</i> (81)	94.2	49.8
Huron (81)	22.7	29.0	73-18-11 (80) ^a	147.3	37.7
Katahdin (80)	49.2	43.0	73-21-1 (81)	49.3	29.0
Kennebec (81)	128.6	48.7	73-21-10 (81)	70.0	50.0
King Edward (81)	67.2	33.9	75-9-2 (80)	21.2	46.3
Kurrell (81)	55.4	46.1	77-10 (81) ^a	71.9	29.3
Lenape (81) ^a	253.7	44.2	8288-542 (80)	101.3	39.3
Mauris Peer (81)	159.5	38.4	B4159-2 (80)	29.7	38.7
Mauris Piper (80)	23.0	41.7	H42 (80) ^a	180.8	57.0
Menomee (80)	49.1	37.7	PF 6/1 (80)	192.4	48.7

^a Significant content of β₂-chaconine (> 1%)

lines had levels in the 100–200 µg/g range, which is considered too high by Ross *et al* (1978). The cultivars in this range were Calrose, Kennebec, Mauris Peer and Pentland Beauty; breeding lines were 73-18-11, 8288-542, H42 and PF6/1.

The relative content of these individual glycoalkaloids varied considerably ranging from 28 ± 3% α-solanine in Avenir to 57.0% in H42

(with the balance being α -chaconine except where stated). Most of the cultivars had similar contents of α -solanine, $40 \pm 5\%$, as a proportion of the total glycoalkaloids (47/84). Several cultivars also had significant amounts ($> 1\%$) of β_2 -chaconine present; these ranged from 2.5% in Lenape up to 23.4% β_2 -chaconine in 73-21-1 and also included Pentland Crown (7.8%), Wauson (2.8%), 77-10 (14.8%) and H42 (7.3%). The levels of β_2 -chaconine recorded for 77-10 (14.8%) and for 73-21-1 (23.4%) are the highest levels of this glycoalkaloid reported in potato tubers.

Total and individual glycoalkaloid levels for Glen Innes

The results for potatoes grown at Glen Innes are given in Table 2. Glycoalkaloid contents again varied considerably, with Crana having the lowest level (32.8 $\mu\text{g/g}$) and the breeding line S3050 (grown in 1980) having the highest level (163.8 $\mu\text{g/g}$). The majority of cultivars and breeding lines (21/38) again had values within a narrow range of $57 \pm 12 \mu\text{g/g}$. This range is slightly higher than observed at Healesville. Although several breeding lines had values lying in the range of 100–200 $\mu\text{g/g}$ —namely, S3041 (1980), S3050, S3172 and S3217—no cultivars or breeding lines had levels greater than 200 $\mu\text{g/g}$.

Relative α -solanine content ranged from 32.6% (S3081) to 51.3% (S3050 in 1980), and for the majority of cultivars (25/38) was again within a narrow range of $41 \pm 4\%$. This range is similar to that found at Healesville. Three cultivars had significant β_2 -chaconine contents, S3064 (2.8%), S3050 (3.5%) and Exton (4.4%).

Cultivar and seasonal effects at Glen Innes

Four cultivars and four breeding lines were grown at Glen Innes in both 1980 and 1981, and while the average glycoalkaloid level was lower in 1981, the difference between years was not significant. There was a difference between years for some cultivars ($P < 1\%$), with Kennebec having a significantly higher level in 1981 than in 1980 and S3041, S3050 and S3064 having significantly lower levels in 1981. The relative content of α -solanine did, however, vary significantly ($P < 0.1\%$) between 1980 (45.3%) and 1981 (42.0%). There was also a difference between years for various cultivars ($P < 1\%$), with all cultivars except Exton and Sebago having significantly less per cent α -solanine in 1981. The different

TABLE 2
The Total and Relative Glycoalkaloid Contents of Potato Cultivars and Breeding Lines Grown at Glen Innes, New South Wales, During 1980 and 1981

<i>Cultivar (Year)</i>	<i>Glycoalkaloid content ($\mu\text{g/g FW}$)</i>	<i>Per cent α-solanine</i>
Bintje (81)	63.2	39.1
Burgama (81)	61.2	37.7
Coliban (80)	60.0	43.3
Crana (81)	32.8	43.5
Exton (80, 81) ^a	62.5/68.6	38.0/39.7
Katahdin (81)	84.4	42.1
Kennebec (80, 81)	51.6/85.9	52.3/44.4
Kurrell (81)	48.7	45.7
Maramba (80, 81)	48.0/61.4	53.3/38.6
Sebago (80, 81)	40.3/46.4	43.0/40.5
Tasman (81)	73.8	39.2
S3041 (80, 81)	127.5/97.7	45.0/35.7
S3050 (80, 81) ^a	163.8/105.6	51.3/44.3
S3064 (80, 81) ^a	88.1/54.8	50.3/42.9
S3081 (81)	60.7	32.6
S3083 (81)	55.7	37.3
S3112 (81)	58.3	44.4
S3141 (80, 81)	39.8/58.1	41.5/35.6
S3172 (81)	111.1	48.4
S3217 (81)	120.2	41.6

^a Significant content of β_2 -chaconine ($> 1\%$)

preparatory technique used in 1981 gave per cent α -solanine values which averaged 1.4% lower than did the technique for 1980. However, even allowing for this change, the per cent α -solanine difference between the two years is significant.

Differences between Glen Innes and Healesville

Eight cultivars were grown both at Healesville and at Glen Innes. There was no significant difference in either average glycoalkaloid content (Healesville, 65.3 $\mu\text{g/g}$; Glen Innes, 64.3 $\mu\text{g/g}$) or in average per cent α -solanine (Healesville, 43.0%, Glen Innes, 41.0%) between the two districts. There were significant individual differences between Healesville

TABLE 3
The Total and Relative Glycoalkaloid Contents
of Potato Cultivars and Breeding Lines Grown
at Yanco, New South Wales, During 1981

<i>Cultivar</i>	<i>Glycoalkaloid content</i>	<i>Percent α-solanine</i>
Crana	64.3	45.4
Exton	112.2	41.2
Katahdin	116.5	39.9
Sebago	88.3	39.4
S3041	125.6	40.5
S3081	72.8	34.1
S3083	76.5	39.1
S3112	109.0	42.3

and Glen Innes in total glycoalkaloid content (Katahdin and Kennebec) and in per cent α -solanine (Exton, Kurrell). In neither case, however, were the differences consistent.

Total and individual glycoalkaloid levels at Yanco

Eight of the cultivars and breeding lines grown at Glen Innes in 1981 were also grown at Yanco (Table 3), with glycoalkaloid contents ranging from 64.3 $\mu\text{g/g}$ (Crana) to 125.6 $\mu\text{g/g}$ (S3041). The average glycoalkaloid content was significantly higher ($P < 0.1\%$) at Yanco (95.7 $\mu\text{g/g}$) than at Glen Innes (59.9 $\mu\text{g/g}$). However, the average per cent α -solanine content at Yanco (40.2%) was not found to be significantly different from that at Glen Innes (39.5%).

Relationship of total glycoalkaloids with the ratio between glycoalkaloids

There was a tendency for cultivars or breeding lines with high glycoalkaloid content also to have a high per cent of α -solanine. Figure 1 shows the relationship between total glycoalkaloid content and per cent α -solanine; it is described by the equation: per cent α -solanine = $37.23 + 0.0515$ (glycoalkaloid content $\mu\text{g/g}$ FW) and was significant at the 1% level ($R = 0.33$). This relationship was significant for the data from Healesville ($P < 1\%$) and Glen Innes ($P < 5\%$) considered alone, but not for Yanco—because of the small sample size from this district.

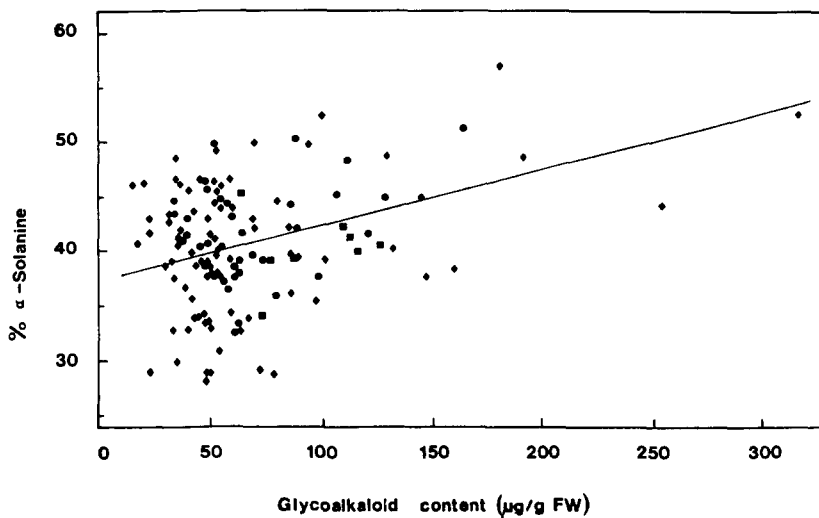


Fig. 1. Correlation of per cent α -solanine and glycoalkaloid levels for all tested potato lines grown at Healesville (\blacklozenge), Glen Innes (\bullet) and Yanco (\blacksquare) during 1980 and 1981.

Cultivars or breeding lines which contained β_2 -chaconine had a significantly higher average glycoalkaloid content ($114.3 \mu\text{g/g}$) than did those without significant β_2 -chaconine ($69.1 \mu\text{g/g}$). When analysed by Student's *t* test (Steel & Torrie, 1960), this difference was significant at the 1% level.

DISCUSSION

There is a good agreement between the glycoalkaloid levels obtained in this work and those reported previously for these cultivars. Only three have been widely studied—namely Lenape, Katahdin and Kennebec. Levels of glycoalkaloids in Lenape of 266, 290, 227 and $310 \mu\text{g/g}$ have been reported by Zitnak & Johnson (1970), Sinden & Webb (1972), Cronk *et al.* (1975) and Bushway *et al.* (1980), respectively; such levels compare well with the $253.7 \mu\text{g/g}$ obtained here for Lenape in the current study. Levels of glycoalkaloids in Katahdin of 55, 80, 53, 56, 60 and $116 \mu\text{g/g}$ are reported by Wolf & Duggar (1940), Sinden & Webb (1972), Deahl *et al.* (1973), Cronk *et al.* (1975), Fitzpatrick *et al.* (1977), Butcher (1978) and Bushway *et al.* (1980), respectively; in this study values of 49.2, 89.4 and $116.5 \mu\text{g/g}$ were obtained which compare favourably. Zitnak & Johnston

(1970), Sinden & Webb (1972), Deahl *et al.* (1973), Fitzpatrick *et al.* (1977) and Bushway *et al.* (1980), respectively, have reported levels of 90, 100, 47, 94 and 75 $\mu\text{g/g}$ for Kennebec; again, these are similar to the 51.6, 85.9 and 128.6 $\mu\text{g/g}$ reported here.

Cultivars studied by two groups include Sebago and Green Mountain with 9 $\mu\text{g/g}$ (Haard, 1977) and 40 $\mu\text{g/g}$ (Wolf & Duggar, 1946), compared with 52.6 $\mu\text{g/g}$ in this work. Previously reported values for Sebago of 14 $\mu\text{g/g}$ (Patchett, 1977) and 21 $\mu\text{g/g}$ (Wolf & Duggar, 1946), are both considerably lower than the range of 40.3–88.3 $\mu\text{g/g}$ obtained in this work. All other previously reported glycoalkaloid levels for the cultivars studied here (with current values in brackets) are: Red Pontiac, 40 $\mu\text{g/g}$ —Sinden & Webb (1972) [36.2 $\mu\text{g/g}$]; Wauson, 32 $\mu\text{g/g}$ —Fitzpatrick *et al.* (1977) [52.7 $\mu\text{g/g}$]; Rima, 54 $\mu\text{g/g}$ —Butcher (1978) [34.2 $\mu\text{g/g}$]; Bison, 34 $\mu\text{g/g}$ —Ross *et al.* (1978) [55.0 $\mu\text{g/g}$]; Patrones, 36 $\mu\text{g/g}$ —Ross *et al.* (1978) [77.5 $\mu\text{g/g}$]; Arran Victory, 67 $\mu\text{g/g}$ and Bintje, 23 $\mu\text{g/g}$ —Ross *et al.* (1978) [63.2 and 59.3 $\mu\text{g/g}$, respectively].

This study is the first in which the relative amounts of α -solanine and α -chaconine in potato tubers have been examined over a wide genetic base. Besides being of theoretical interest, the relative proportions are also of practical interest since, in some studies, α -chaconine has been found to be more toxic than other glycoalkaloids to the foetus of several animal species (Nishie *et al.*, 1975; Chaube & Swinyard, 1976).

Virtually all previous studies of the relative contents of α -solanine and α -chaconine have used either TLC (Ahmed & Muller, 1978; Cadle *et al.*, 1978; Bushway *et al.*, 1980), or GC after chemical modification (Seigfried, 1976; Gregory *et al.*, 1981). Unfortunately, both these methods can result in substantial inaccuracies in quantifying the level of each glycoalkaloid present. All previously reported values lie in the range 27–54% α -solanine, except for two cultivars, Hansa (12% α -solanine) and Amigo (15% α -solanine) (Seigfried, 1976).

Of the cultivars examined in this work, only three (Lenape, Katahdin and Kennebec) have been previously studied for the levels of individual glycoalkaloids. The level of α -solanine in Lenape has been reported as 38% of the total glycoalkaloids (Cadle *et al.*, 1978) as determined by TLC and 35% (Bushway *et al.*, 1980) by HPLC estimation, compared to 44.2% in this work. Katahdin has been found to contain 27% α -solanine (Gregory *et al.*, 1981) as determined by permethylation and GC and 36% (Bushway *et al.*, 1980) by HPLC, compared to 39.9–43.0% α -solanine found in this work. Finally, the α -solanine content of Kennebec has been

reported as 34% (Gregory *et al.*, 1981), compared to levels between 44.4 and 51.6 reported in this work. The values for per cent α -solanine reported by these workers tend to be slightly lower than those found in this work.

CONCLUSION

Two of the cultivars examined in this paper (Lenape and Berita) had glycoalkaloid levels greater than the upper safety limit of 200 $\mu\text{g/g}$. However, Lenape is the only cultivar that has previously been reported as exceeding this level in non-greened, uninjured tubers (Zitnak & Johnson, 1970; Sinden & Webb, 1972; Cronk *et al.*, 1974). Berita was found to have a level of glycoalkaloids even higher than Lenape; therefore, it should similarly be withdrawn as a commercial potato cultivar. Several cultivars and breeding lines had glycoalkaloid levels in the region of 100–200 $\mu\text{g/g}$, a level regarded as unsafe by Lepper (1949) and Ross *et al.* (1978) and which can impart an undesirable bitter taste (Lepper, 1949; Sinden & Deahl, 1976; Ross *et al.*, 1978). There are three cultivars (Calrose, Mauris Peer and Pentland Beauty) and six breeding lines (73-18-11, H42, PF6/1, S3041, S3050 and S3217) that consistently have glycoalkaloids in this area of concern. Caution should be exercised when growing these genetic lines, particularly when environmental factors that can increase glycoalkaloid levels are likely to occur. These factors include cool weather, rough handling (Sinden & Webb, 1972; Maga, 1980) and even high levels of magnesium in the soil (Evans & Mondy, 1984).

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