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Variation of Thiocyanate Ion Content in Cauliflower and Broccoli Cultivars[1]

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Abstract. **The thiocyanate ion (SCN^-) content in leaves and stems of 2 cauliflower (*Brassica oleracea* L. Botrytis group) and 2 broccoli (*B. oleracea* L. Italica group) cultivars was highest in 15-day seedlings at the cotyledon stage of development, then decreased rapidly and showed little change in 72-day and older plants. The highest quantities of SCN^- in cauliflower curds and broccoli heads generally occurred at the premature stage of development, after which it decreased rapidly. Except for 'Jet Snow' cauliflower, relatively high quantities of SCN^- were found in edible curds of 9 other cauliflower cultivars and heads of 6 broccoli cultivars at the optimum mature stage.**

Plants of the Cruciferae contain glucosinolates (thioglucosides) which undergo enzymic hydrolysis upon tissue disintegration yielding isothiocyanates, thiocyanates, and other related derivatives (5, 16). These glucosinolate-derivatives are responsible for the flavor of cruciferous plants (5, 6), and also for contributing to iodine deficiency and the incidence of goitre (enlargement of the thyroid gland) (5, 7, 14). Many studies have related goitrogenic toxicity of cruciferous plants to their content of thiocyanate ion (SCN^-) (7, 10, 17), and in cultivars of kale *(Brassica oleracea)* SCN^- is largely responsible for their goitrogenicity in rats (13). Thiocyanate occurrence may also indicate the presence of other more potent goitrogens (13). Since there is increasing concern for the presence of toxicants such as goitrogens, glycoalkaloids or nitrates in foods (7, 8, 9, 12), the SCN^- variation in plants during ontogeny and differences between cultivars is of definite interest. This has been reported for radish (3), and is here studied in cauliflower and broccoli.

Materials and Methods

'Jet Snow' and 'Igloo' cauliflower and 'Spartan Early' and 'Waltham 29' broccoli were seeded on April 30, 1974, in 25 × 15 cm fiber flats with turface medium, and grown under greenhouse conditions with mean temperature of 22 ± 4°C. Thirty-day seedlings were transplanted in Block II of the Macdonald College Horticulture fields on a St. Bernard clay loam soil, with 7 seedlings per row, 0.6 m within row spacings, and rows 0.9 m apart, arranged in a randomized block design with 3 replications. Standard recommendations for fertilizer and pesticide applications were followed and plants were irrigated occasionally during dry periods.

Triplicate samples were taken from each cultivar for dry weight determinations and for SCN^- analysis during development (Fig. 1, 2): seed (1 g); 15-day seedlings at the cotyledon stage (50 plants separated into cotyledon and stem); 44-day plants at the 5- or 6-leaf stage (2 plants per replication), and at subsequent 14-day intervals until 114 days after seeding. Representative samples (each 100 g) from combined basal to upper regions of these plants were taken from leaves and stems. Cauliflower curds and broccoli heads were sampled as wedge-shaped pieces. A second investigation was carried out with 10 cauliflower and 6 broccoli cultivars seeded on April 30, 1974 and grown under greenhouse conditions as described above. Date of transplanting, plot design, and field culture were also as described above, although plants were grown in another field (Block IV), and samples were taken only from curds or heads at the optimum mature stage of development of each cultivar (Table 1). Samples for analysis were extracted with twice their volume of distilled water, and colorimetric SCN^- determinations were carried out in triplicate as described previously (3), with values expressed as μg KSCN per g dry wt.

Results and Discussion

Qualitatively, similar changes in SCN^- content were found during development in all plant parts and cultivars studied (Fig. 1, 2). Thus SCN^- content was highest in 15-day seedlings

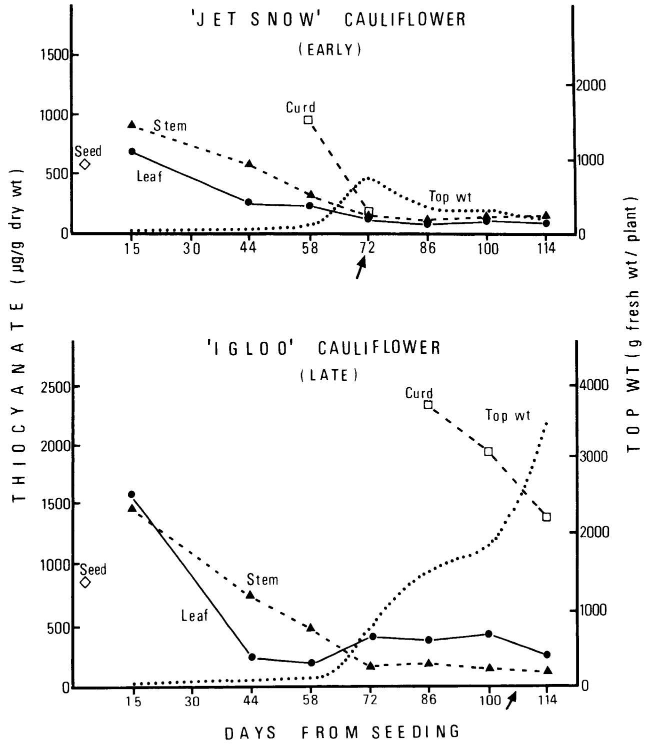

Fig. 1. Variation in top fresh weight and SCN^- content in plant parts of 'Jet Snow' and 'Igloo' cauliflower during development. Arrows indicate approximate date at which curds were at optimum mature stage.

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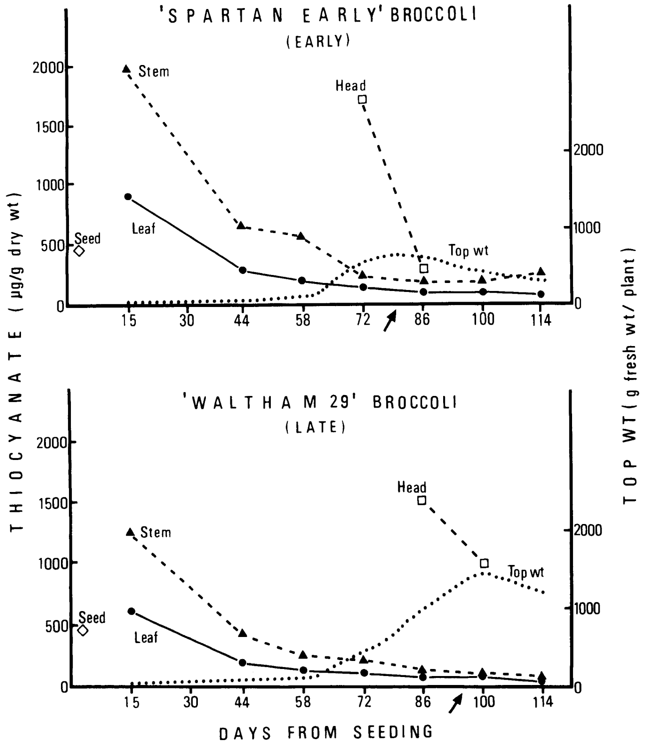

Fig. 2. Variation in top fresh weight and SCN$^-$ content in plant parts of 'Spartan Early' and 'Waltham 29' broccoli during development. Arrows indicate approximate date at which heads were at optimum mature stage.

in both leaves and stems of each cultivar of cauliflower and broccoli, then decreased rapidly with little change in 72-day and older plants. Similarly in flowering parts, highest quanities of SCN$^-$ were found in cauliflower curds and broccoli heads at the premature stage of development (Fig. 1, 2). However, there were also quantitative differences with respect to both cultivars and plant parts. Thus SCN$^-$ content was generally lower in the early cauliflower cultivar 'Jet Snow' than in the later 'Igloo' (Fig. 1), while the reverse was true with broccoli cultivars (Fig. 2).

All of the cauliflower and broccoli cultivars surveyed contained relatively high quantities of SCN$^-$ in mature (edible) tissues except for 'Jet Snow' cauliflower (Table 1). There were significant differences in SCN$^-$ content in mature curds of cauliflower cultivars, but not in mature heads of broccoli cultivars. Similarly, a survey by Mullin and Sahasrabudhe (11) showed relatively high SCN$^-$ content (expressed as equivalents of 3-methylindolylisothiocyanate) in 4 of 5 selected cultivars of both cauliflower and broccoli, with most variation in cauliflower.

The highest occurrence of SCN$^-$ in younger vegetative and reproductive organs in cauliflower and broccoli is similar to that observed in radish tissues (3), for various glucosinolate-derivatives in cabbage heads (15), and also for sulfur distribution in plants in general (1). Thus, unlike other cruciferous vegetables such as radishes, turnips, and cabbages which are relatively low in SCN$^-$ content in their vegetative (edible) parts, the reproductive (edible) parts of cauliflower and broccoli generally can be expected to be higher in SCN$^-$ content. Although the biosynthesis of parent thioglucosides or related enzyme(s) of hydrolysis is not fully understood, reproductive tissues appear to be major sites of synthesis or storage of these compounds (3).

Table 1. SCN$^-$ content in mature curds of cauliflower and heads of broccoli cultivars.

Cultivar	Days from seeding to maturity	SCN$^-$ (µg/g dry wt)
Cauliflower		
Jet Snow	79	268a^{z}
Stokes Early Abundance	85	1025b
Whitehorse	94	948b
Stokes Super Snowball	94	954b
Snowball-Y	99	1172bc
Snowball Imperial	100	1221bc
Snowmound	101	1087b
Clou	101	1089b
Imperial 10-6	101	1084b
Igloo	116	1435c
Broccoli		
Spartan Early	78	950a
Italian Sprouting	85	868a
Cleopatra	91	770a
Waltham 29	92	883a
Green Mountain	101	625a
Early Purple Head	111	871a

^{z}Mean separation by Duncan's multiple-range test at the 5% level.

There is also a trend towards higher SCN$^-$ content in later maturing (associated with increasing curd size) cultivars of cauliflower (Fig. 1; Table 1), similar to that found in cultivars of radishes, turnips (4), and cabbages (2).

Literature Cited

1. Beaton, J. D. 1966. Sulfur requirements of cereals, tree fruits, vegetables and other crops. *Soil Sci.* 101:267-282.
2. Bible, B. B., H. Y. Ju, and C. Chong. 1980. Influence of cultivar, season, irrigation, and date of planting on thiocyanate ion content in cabbages. *J. Amer. Soc. Hort. Sci.* 105:88-91.
3. Chong, C. and B. Bible. 1974. Variation in thiocyanate content of radish plants during ontogeny. *J. Amer. Soc. Hort. Sci.* 99:159-162.
4. ____________ and ____________ . 1974. Relationship between top/root ratio and thiocyanate content in roots of radishes and turnips. *HortScience* 9:230-231.
5. Ettlinger, M. G. and A. Kjaer. 1968. Sulfur compounds in plants. p. 59-144. *In* T. J. Mabry, R. E. Alston, and V. C. Runeckles (eds.) Recent advances in phytochemistry. Appleton-Century-Crofts, N.Y.
6. Freeman, G. C. and N. Mossadeghi. 1972. Influence of sulphate nutrition on flavour components of three cruciferous plants: radish *(Raphanus sativus)*, cabbage *(Brassica oleracea capitata)* and white mustard *(Sinapis alba)*. *J. Sci. Food Agr.* 23:387-402.
7. Greer, M. A., and 1950. Nutrition and goiter. *Physiol. Rev.* 30:513-548.
8. Kehr, A. E. 1973. Naturally-occurring toxicants and nutritive value in food crops: the challenge to plant breeders. *HortScience* 8:4-6.
9. Liener, I. E. 1973. Naturally occurring toxicants of horticultural significance. *HortScience* 8:112-116.
10. Michajlovskij, N. and P. Langer. 1958. Studien über Beziehungen zwischen Rhodanbildung und kropfbildender Eigenschaft von Nahrungsmitteln, I Gehalt einiger Nahrungsmittel an präformiertem Rhodanid. *Z. Physiol. Chem.* 312;26-30.
11. Mullin, W. J. and M. R. Sahasrabudhe. 1977. Glucosinolate content of cruciferous vegetable crops. *Can. J. Plant. Sci.* 57:1227-1230.
12. Nutrition Canada. 1973. Nutrition: a national priority. Information Canada, Ottawa.
13. Paxman, P. S. and R. Hill. 1974. The goitrogenicity of kale and its relations to thiocyanate content. *J. Sci. Food Agr.* 25:329-337.
14. VanEtten, C. H. 1969. Goitrogens. p. 103-142. *In* I. E. Liener (ed.) Toxic constituents of plant foodstuffs. Academic Press, New York.
15. ____________, M. E. Daxenbichler, W. F. Kwolek, and P. H. Williams. 1979. Distribution of glucosinolates in the pith, cambial-cortex, and leaves of the head in cabbage, *Brassica oleracea* L. *J. Agr. Food Chem.* 27:648-650.

16. ———— and I. A. Wolff. 1973. Natural sulfur compounds. p. 210-234. *In* Toxicants occurring naturally in foods. 2nd ed. National Academy of Sciences, National Research Council, Washington, D.C.

17. Wills, J. H., Jr. 1966. Goitrogens in foods. p. 3-17. *In* Toxicants occurring naturally in foods. National Adacemy of Sciences, National Research Council, Washington, D.C., Publ. 1354.

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Application of Trend Analysis to Horticultural Field Trials[1]

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Abstract. **Experimental results from horticultural field trials are obscured by the effect of systematic variation. This variation is directly related to the position of the plot in the field and is referred to as a fertility gradient(s). Trend analysis eliminates the effect of fertility gradients by fitting a polynomial regression equation (response surface model) to the systematic variability in the experimental units. Two cultivar trials of potato (*Solanum tuberosum* L.) conducted to compare results from trend analysis with that using the standard design analysis indicated that fertility gradients existed in the fields and were of a form that could be adequately fitted by a response surface model. A 3-dimensional plot of the response surface model indicated that the fertility gradients formed a very complex surface which could not be eliminated by experimental design. Of the 3 experimental designs used, the Latin square was the most efficient while the completely random was the least efficient. Trend analysis resulted in a large gain in relative efficiency over the standard analyses of completely random and randomized block designs. It also resulted in a substantial gain over that of a Latin square design. Adjusting the means using a response surface model in trend analysis also improved treatment estimates. Tests of significance using adjusted means were more precise and easier to interpret. Trend analysis proved to be the most efficient way to analyze the data, regardless of the experimental design used.**

Experimental results from horticultural field trials are obscured by the effect of 2 types of disturbing factors: A) the effect of factors displaying an unknown, systematic variation and B) the effect of random factors. The systematic variation is directly related to the position of the plot in the field and is referred to as a "fertility gradient(s)." This variation may be attributed to many factors such as weediness, water drainage, direction of winds, and the intrinsic fertility of the soil. The random factor is called pure error and is caused by measurement errors, and genetic variation, plus a large number of small additive effects, e.g., genotype × environment interaction.

The magnitude of experimental error, which comes from both sources of variation, is a question of major importance in horticultural field research. Its proper recognition determines the degree of confidence which may be attached to the results obtained in field work. Not only does the fertility gradient inflate the experimental error, but it also causes lack of independence among elements usually assumed to be normally and independently distributed with population mean $\mu=0$ and variance σ^2 (NID $(0,\sigma^2)$). As pointed out by Yates (7), this lack of independence impairs the entire theoretical basis of the standard analysis, and in particular is liable to invalidate the estimates of error and tests of significance. Logically, given that plot yields are correlated, a continuous function of some form may be fitted to the systematic variation in the experimental units. It follows that such a trend analysis may be applied in which the fitted function eliminates fertility gradient effects from the experimental error.

Since the beginning of the 20th century, uniformly trials have been used to study variability in agricultural fields. Results from these early studies show that neighboring plots are positively correlated, indicating that the plots have certain common tendencies or trends in their productivity. Smith (5) presented an empirical law for the relationship between the size of plot and the variance per unit based on correlation between neighboring plots.

Neyman et al. (4) were among the first investigators to suggest using some form of a functional model to explain systematic soil variation. They proposed using a polynomial regression equation for describing the fertility variation in experiments, where the treatments were arranged in the same order in every block and where the blocks were placed in succession. Van Uven (6) considered the adjustment of yields in field experiments according to the condition of the soil. Federer and Schlottfeldt (2) illustrated the use of covariance to control gradients in a tobacco experiment as a substitute for deliberate stratification in the design. Using uniformity field trials with superimposed treatment effects, Mendez (3) compared 6 alternatives to blocking for the design and analysis of field experiments. He concluded that if the number of treatments is large, say 5 or more, then trend analysis should be considered.

In trend analysis a grid is superimposed on the field by identifying rows and tiers (i.e., latitude and longitude) which define the location of each plot. The systematic variability is removed by fitting a polynomial regression equation to the fertility gradient using rows and tiers as the independent variables in the polynomial function. The resulting function has come to be known as a "response surface" model. A general form of the mathematical model used is:

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