Title:

Influence of high latitude light conditions on sensory quality and contents of health and sensory-related compounds in swede roots (*Brassica napus* L. ssp. *rapifera* Metzg.)

Running title:

Influence of high latitude light on eating quality of swede roots

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ABSTRACT

BACKGROUND: Swede roots (rutabaga) were produced in controlled climate with natural light (69° 39' N) to investigate the effect of photoperiod and photosynthetic light period on sensory attributes and health/sensory-related compounds.

RESULTS: The root size, sensory quality and concentration of glucosinolates (GLS) were significantly affected when altering the natural 24 h photoperiod with darkness or fluorescent lights. Shortening the photosynthetic light period to 12 h produced smaller roots with lowest scores for sweetness, acidic taste and fibrousness, and highest scores for bitter and sulphur taste. The photosynthetic light period and duration of twilight/darkness of the photoperiod had a combined influence on GLS, with highest contents under short 12 h photoperiod/photosynthetic light period.

CONCLUSION: Our results suggest that the photoperiod has little influence *per se* on sensory quality and phytochemicals in swede roots. However, high latitude photoperiods with long photosynthetic light periods may reduce GLS contents and influence some sensory attributes, in contrast to shorter photoperiods.

Keywords: swede roots, rutabaga, photoperiod, photosynthetic light period, sensory quality, glucosinolates.

INTRODUCTION

Swede (*Brassica napus* L. ssp. *rapifera* Metzg.) is a root vegetable crop, popular for fresh and processed food in northern regions of Europe and North America. It has historically been an important source of health-related compounds such as vitamin C, due to its suitability for long-term winter storage. The vegetable is an enlarged part of the stem base (hypocotyl) including the upper part of the root, which functions as a vegetative winter storage organ. This bulbous organ is commonly referred to as swede root or hypocotyl, rutabaga, swedish turnip or simply swede.[1] With origins from Fennoscandia, it is suitable for growth in temperate/cool climates and can be successfully grown in Scandinavia at latitudes above the Arctic Circle (66° 33' N) in summer.[2] In southern temperate zones, it is also grown in autumn, spring, and through winter in coastal areas without prolonged frosts.[3,4] Thus, being produced over a wide range of photoperiods, from 24 h above the Arctic Circle to below 12 h between autumn-spring equinoxes. The photoperiod can be subdivided into a photosynthetic light period with enough irradiance for net photosynthesis, and periods of lower light intensities/twilight in the morning and evening. The length of these periods are dependent on the cloud cover, solar elevation and time of season. The lengths increase with latitude in summer. Locations above the Arctic circle, such as Tromsø (69° 39' N), has 24 h photoperiod from May to August, and up to 20 h daily photosynthetic light periods.

Swede roots, like many other crucifer vegetables, are rich in vitamin C and glucosinolates (GLS), and normally contains over fifty percent soluble sugars per dry matter.[4-6] GLS are sulphur containing glucosides involved in plant defense in Brassicas, which upon hydrolysis (i.e. by the enzyme myrosinase) can produce isothiocyanates, nitriles, thiocyanates, epithionitriles and oxazolidines.[7] Some of these breakdown-products confer bioactivity with potential health benefits for humans.[8,9] Other GLS such as sinigrin and progoitrin contribute to *Brassica* specific flavors, pungency and bitter taste.[10] Progoitrin is one of the major glucosinolates found in swede.[11] Both light and temperature affect the accumulation of GLS in a cultivar dependent manner in Brassicas,[12] and GLS biosynthesis is under diurnal regulation.[13] In swede, GLS contents decrease with progressing season under both long day length in summer and short day length in winter.[14,15] L-ascorbic acid (AA) is an important antioxidant in photosynthesis and signaling,[16] and AA (and its oxidized form dehydroascorbic acid, DHA) is an essential nutrient vitamin C in the human diet.[17] AA usually accumulates in leaves under high irradiance and low temperature,[18] and light/darkness-cycles result in diurnal variations of AA pools,[19] Field studies in Scandinavia of swede roots indicate no differences in vitamin C content between northern and southern latitudinal sites, or in a few cases higher contents at northern sites.[2,5]

Climatic factors such as light, temperature and precipitation can all influence the sensory quality of crucifer vegetables.[20-23] For swede roots there are some sensory results from field trials using an untrained sensory panels. Hårdh *et al.*[2] reported better taste of roots in Scandinavia from northern (65-69° N) versus southern (55-60° N) latitudes, with 2-6 h longer day lengths and 2-5 °C lower mean temperatures. Production on Prince Edward Island, Canada (46° N) under shortening days and decreasing temperatures in Autumn, had higher sugar content, sweeter taste and overall better acceptability in November compared to in October.[3] A study from Dundee, Scotland (56° N) also showed accumulating content of soluble sugars during the autumn and early winter, together with increasing hardness.[4] There are currently no studies where the effects of day length conditions is separated from the effects of temperature on sensory attributes and phytochemical contents in swede.

The main aim of this study is therefore to examine the influence of contrasting (1) day lengths and (2) photosynthetic light periods on the contents of glucosinolate, vitamin C, sugars and sensory quality in swede roots under controlled climate.

EXPERIMENTAL

Plant material and growth conditions

Swede seeds 'Vigod' were germinated on a mixture of standard fertilized peat soil and perlite (70/30 volume) at 21 ºC in climate-regulated chambers, and moved to 15 ºC after germination. After 4 weeks, plants were transplanted and grown in 7.5 litre pots. Growth temperature was set to constant 15 ºC (±0.5 °C) and humidity was standardized to give a water vapour pressure deficit of 0.5 kPa. After the growth period, roots were harvested and stored as described by Johansen et al.[22]

Day length treatments

The experiment was carried out under natural light in the phytotron of the University of Tromsø (69º 39' N, 18° 55' E) from May 27 to August 15 in 2011. The influence of day length and photosynthetic light period was investigated in parallel with a separate study on the influence of temperature with common control treatment.[22] Photoperiods and photosynthetic light periods were modified by moving plants from natural light at 20:00 to dark chambers with different fluorescent light sources, and returned to natural light at 08:00 (Figure 1). The 12 h period of natural light thus included the core of the daily photosynthetic light period, with maximum solar elevation between 12:41-12:51 during the experiment. The control treatment of 24 h natural light included low intensity light or twilight at night. The average daily photosynthetic light period within the experimental period was estimated from global radiation data (>100 µmol m^{-2} s^{-1}) to be 15.7 h. The maximum and minimum total daily hours were respectively 20 h and 9 h, depending on the cloud cover. The irradiance at 400-700 nm within the glass chamber of the phytotron was measured with maxima of 400-500 µmol m^{-2} s^{-1} on sunny days. Treatments with artificial growth light were given in dark chambers equipped with fluorescent light (Philips TLD 840) at 100-150 µmol m^{-2} s^{-1}. Low irradiance light bulbs (Energy Saver Osram DUlux 41-827) at 12-15 µmol m^{-2} s^{-1} were used in treatments with photoperiodic extension. Treatments with 18 h photosynthetic light period was arranged as 12 h natural light (08:00 -20:00) and 3 h + 3h artificial growth light in dark chambers preceding and following the 12 h natural light period.

Sensory and chemical analyses

The sensory analysis was performed by a trained panel of 12 members in an accredited sensory laboratory at Nofima, and GLS, soluble sugars and vitamin C were extracted and quantified as described by Johansen *et al.*[22]

Statistical analyses

The sensory data were analyzed using analysis of variance (ANOVA) as described by Johansen *et al.*[22] One-way ANOVA for the growth data and phytochemical contents, and analyses of linear correlations, were performed with Minitab Version 16.1.0 (Microsoft, State College, PA, USA). All ANOVA analyses were supplemented by pairwise comparisons using Tukey's multiple comparisons test (α=0.05).

RESULTS

Effects on swede growth

The length of photosynthetic light period significantly affected the swede root size and root fresh weight (Table 1). Shortening the natural photosynthetic light period to 12 h produced smaller roots of less fresh weight, compared to treatments with longer 15.7-18 h periods. Extending the photoperiod with low intensity fluorescent light versus darkness at night, had no significant effects on root size or root fresh weight. Leaf dry matter content, root dry matter percentage and root shape (diameter versus height ratio) were not significantly influenced by any of the treatments.

Phytochemical contents of peeled roots

Twelve different GLS were detected by HPLC, of which 10 were quantified (Table2). Glucoraphanin and epiprogoitrin were not quantified as they were detected in trace amounts (peak areas less than 1% of the sum of GLS) and not in all samples. Gluconasturtiin was also detected in trace amounts by LC/Q-TOF/MS and may contribute minimally to the glucoberteroin HPLC peak area due to overlapping retention times. The quantified aliphatic GLS were in decreasing order of concentration progoitrin, glucoberteroin, gluconapoleiferin and glucobrassicanapin, and the indolic GLS were glucobrassicin, 4-hydroxy-glucobrassicin and 4-methoxy-glucobrassicin. Altering the natural 24 h photoperiod with darkness or fluorescent light sources, significantly affected eight of ten quantified glucosinolate types (GLS) (Table 2). Photoperiod had no significant effect by itself when comparing glucosinolate levels between treatments with identical photosynthetic light periods of 12 h and 18 h. There was however a trend of higher concentrations in treatments with a dark period at night versus photoperiodic extension, for seven of the influenced GLS. Reducing the photoperiod and photosynthetic light period to 12 h resulted in highest concentrations for these seven GLS, in contrast to lowest concentrations in the 24 h control. The treatments with artificial growth light or photoperiod extension with low intensity light all had concentrations in the range between these two contrasts, with 18 h photosynthetic light period + 6 h extension being most similar to the 24 h control. There were no significant differences in concentrations of AA and DHA for any of the light treatments (Table 3), nor in concentrations of soluble sugars: glucose, fructose and sucrose (Table 4).

Sensory quality of peeled roots

Thirteen of twenty-five sensory attributes were significantly influenced by altering the 24 h natural light (Table 5). Extending the photoperiod did not have a major influence on the sensory quality, as only one attribute; colour strength, was influenced by being lower at 12 h photoperiod versus 12 h + 12 h extension. The photosynthetic light period on the other hand had more influence. Sweet taste and fibrous texture scored higher for all treatments with long 15.7-18 h period than the treatments with 12 h period. In addition, there were also some differences between 12 h and 18 h photosynthetic light period, depending on darkness or low intensity light at night. Both treatments with 12 h PAR-period (with and without low intensity night light) scored highest for bitter taste and lowest for acidic taste, while the 18 h + 6 h darkness in contrast scored respectively lower and higher for these attributes. Furthermore, the treatment with 12 h + 12 h darkness scored higher than 18h + 6 h darkness for pungent flavour, sulphur and metallic taste.

There were no significant correlations between sweet or bitter taste and the measured concentrations of sugars or glucosinolates.

DISCUSSION

Altering the 24 h natural day length at (69° 38’ N) in the current study, significantly affected the size of swede roots, their sensory quality and GLS concentrations. The photosynthetic light period appeared to be more important than photoperiod of the experimental factors. Especially as the length of photoperiod had no significant effects by itself, and any effects of photoperiod were also dependent on the length of the photosynthetic light period. Only one sensory attribute; colour strength, was influenced by photoperiod under 12 h photosynthetic light period, scoring higher in long 24h versus short 12 h photoperiod. This lack of influence of photoperiod *per se* suggests that the tested cultivar is day neutral for swede root development and quality. To our knowledge, there are no reported studies of photoperiodic control of root development in swede or other root bulb forming crucifers, e.g. turnip or kohlrabi.

Contrary to photoperiod, the length of the photosynthetic light period did have a significant influence *per se* on both the size and sensory quality of swede roots. Shortening the solar photosynthetic light period from an average of 15.7 h to 12 h resulted in significantly smaller roots, with consistently less sweet taste and fibrousness compared to treatments with 15.7-18 h period. In addition, these smaller roots scored highest for bitter and sulphur taste, and lowest for acidic flavour, all attributes considered important for consumer preferences in *Brassicas* species.[24] Our results are in agreement with fast growth and better taste of swede roots produced at northern

Scandinavian sites versus southern sites.[2] Longer photosynthetic light periods at high latitudes may compensate for the inhibitory effect of low temperatures on growth, and further enhance the positive influence of low growth temperature on sensory quality in swede.[22] The combination of natural day lengths in Tromsø and low growth temperature also appears to produce sweeter/less bitter taste in broccoli florets.[23] It would therefore be interesting also to test the interaction between day length and temperature on sensory quality in swede.

The photosynthetic light period and the twilight/dark conditions of the photoperiod appears to have a combined influence on the concentrations of GLS in swede roots. The two most contrasting treatments consistently had GLS concentrations at opposite ends of the concentration ranges. It was highest under short photoperiod 12 h and lowest under the control treatment of 24 h natural light. For most of the GLS there was also a consistent trend of lower concentrations when photoperiods were extended by 6 h or 12 h low intensity light compared to darkness. This suggests that long day lengths with prolonged periods of light/twilight accumulate less GLS in swede than short day lengths with a distinct dark period at night. These results are consistent with some studies on broccoli and curly kale involving 24 h photoperiods versus short 12 h days, although results for influence of photoperiod in the literature varies greatly depending on species/variety, tissue type and temperature.[12,23,25-27] Lowest contents under 24 h light is at odds with the diurnal regulation of reduced GLS biosynthesis in darkness in leaves of *Arabidopsis*.[13] It is possible that long photoperiods such as 24 h without a distinct dark period at night, disrupts the circadian rhythm for GLS accumulation in sweede roots.

The total and individual concentrations of glucose, fructose and sucrose were not significantly different between any of the treatments in our experiment. The levels detected in our study were similar to those of other field grown swede cultivars.[28,29] The similar sugar concentration in larger roots from 15.7-18 h and smaller roots from 12 h photosynthetic light periods diverge from results of field-grown swede. In a study by Hårdh et al.,[2] northern latitudinal sites with longer photoperiod/photosynthetic light period also produced larger roots, but with higher sugar concentrations than southern sites. This may be due to lower growth temperatures at northern latitudes, as low temperatures increases concentrations of soluble sugars in swede.[3,4,22] The levels of soluble sugars interact in sensory perception with goitrin, the break-down product of progoitrin, determining sweet versus bitter taste in brassicas.[30] In light of the similar sugar concentrations between our treatments, it may be possible to predict bitter/sweet taste based on GLS concentrations. However, within our measured ranges of aliphatic GLS-alkenols and other GLS it was not possible to find correlations between bitter or sweet taste.

The concentrations of L-ascorbic acid and total vitamin C were not significantly affected by any of our treatments with different day length, or PAR periods. Similarly, two studies have reported no difference in vitamin C contents of swede (and cabbage) grown in northern and southern parts of Norway over two seasons,[2,5] although there were slightly higher vitamin C concentrations at northern sites in Sweden.[2] When harvesting at different dates, Dragland[5] found a negative relation between contents of vitamin C and increasing root fresh weights, whereas in our study no such relation could be observed. The similar concentrations and similar dry matter content in our study could therefore indicate similar developmental stage between treatments. In floral tissues of broccoli grown under fluorescent lights, Steindal *et al*.[26] reported lower vitamin C contents under long day length 24 h than at short day length 12 h. However, a similar study under long versus short photosynthetic light period did not reveal any differences in vitamin C contents of broccoli florets,[23] as in the current study.

CONCLUSIONS

Day lengths with long photosynthetic light periods can give faster growth in swede compared to shorter photosynthetic light periods at similar temperatures. It may also produce vegetables with a positively perceived sensory quality by consumers. Twilight/darkness of the photoperiod may act together with the photosynthetic light period reducing GLS concentrations under high latitude long day lengths. Our study demonstrate that the day length/photosynthetic light period can influence quality attributes in swede roots, which may be benefit marketing of regional production.

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Table 1. Growth and development of swede under controlled photoperiods and photosynthetic light periods in Tromsø (69° 39’ N, 18° 55’ E). The photosynthetic light period consisted of natural light (nat) and artificial growth light (art). Photoperiods were modified by extensions of low intensity light (ext) or darkness at night. Mean values, n = 7-8.

Treatment	Leaf dry matter (g)	Root fresh weight (g)	Root dry matter (%)	Root diameter (cm)	Root shape index[a]
12h nat	20.8	579 b	11.1	11.4 b	1.5
12h nat+12h ext	19.0	628 b	11.0	11.7 b	1.6
18h nat/art[b]	20.0	993 a	11.6	13.7 a	1.6
18h nat/art+6 ext	19.2	936 a	11.8	13.3 a	1.6
24h nat (control)	20.6	899 a	11.3	13.2 a	1.5
p-value	0.365	<0.001	0.091	<0.001	0.904

[a]Root diameter/root height

[b]18h nat/art: 3 h artificial growth light preceding and 3 h following 12 h natural light

Different letters within columns indicate significant difference by Tukey’s multiple comparisons test ($\alpha = 0.05$).

Table 2. Mean concentration of glucosinolates (µmol g^{-1} dry matter, n = 5) in swede roots grown under controlled photoperiods and photosynthetic light periods in Tromsø (69° 39’ N, 18° 55’ E). The photosynthetic light periods consisted of natural light (nat) and artificial growth light (art), and photoperiods were modified by extensions of low intensity light (ext) or darkness at night.

	12h nat	12h nat +12h ext	18h nat/art[a]	18h nat/art +6h ext	24h nat (control)	p-value
GER[b]	1.14	0.99	1.08	0.93	0.86	0.081
PRO	8.42 a	7.34 ab	7.61 ab	6.20 ab	5.50 b	0.035
GBE	3.33 a	3.03 ab	2.99 ab	2.48 b	2.42 b	0.014
GAL	0.38 a	0.38 a	0.29 a	0.26 a	0.28 a	0.038
GBN	0.39 a	0.32 ab	0.29 ab	0.20 b	0.18 b	0.003
GNL	1.23 a	1.19 a	0.94 ab	0.80 b	0.80 b	0.001
GBS	0.62 a	0.54 ab	0.39 bc	0.32 bc	0.31 c	0.001
4HO-GBS	0.65 a	0.52 ab	0.46 bc	0.39 bc	0.33 c	<0.001
4Me-GBS	0.28 a	0.24 ab	0.20 b	0.17 b	0.16 b	0.002
Neo-GBS	0.19	0.22	0.17	0.18	0.25	0.393
Alifatic	14.9 a	13.7 ab	13.3 ab	10.9 ab	10.1 b	0.015
Indolic	1.8 a	1.5 ab	1.2 b	1.1 b	1.1 b	0.002
Total	16.7 a	15.2 ab	14.5 ab	12.0 b	11.2 b	0.011

[a]18h nat/art: 3h artificial growth light preceding and 3h following 12h natural light

[b]GER, glucoerucin; PRO, progoitrin; GBE, glucoberteroin; GAL, glucoallysin; GBN, glucobrassicanapin; GNL, gluconapoleiferin, GBS, glucobrassicin; 4HO-GBS, 4-hydroxyglucobrassicin; 4Me-GBS, 4-methoxyglucobrassicin; Neo-GBS, neoglucobrassicin.

Different letters within rows indicate significant difference by Tukey’s multiple comparisons test (α = 0.05).

Table 3. Mean concentration of ascorbic acid (AA) and total vitamin C (AA+dehydro AA) (mg $100g^{-1}$ FM, n = 8) in swede roots grown under controlled photoperiods and photosynthetic light periods in Tromsø (69° 39’ N, 18° 55’ E). The photosynthetic light periods consisted of different periods of natural light (nat) and artificial growth light (art), and photoperiods were modified by extensions of low intensity light (ext) or darkness at night.

	AA	Total vitamin C
12h nat	21.2	21.7
12h nat+ 12h ext	20.6	21.8
18h nat/art†	21.6	22.9
18h nat/art† + 6h ext	23.5	23.8
24h nat (control)	21.6	22.6
p-value	0.194	0.421

†18h nat/art: 3h + 3h fluorescent growth light preceding and following 12h natural light

Table 4. Mean concentration of soluble sugars (g $100g^{-1}$ DM, n = 5) in swede roots (*Brassica napus* L. ssp. *rapifera* Metzg.) grown under controlled photoperiod and photosynthetic light period at 15 °C in Tromsø (69° 39’ N, 18° 55’ E). The photosynthetic light periods consisted of different periods of natural light (nat) and artificial growth light (art), and photoperiods were modified by extensions of low intensity light (ext) or darkness at night.

	Glucose	Fructose	Sucrose	Total
12h nat	32.2	24.4	2.2	58.8
12h nat + 12h ext	31.0	23.4	3.6	58.0
18h nat/art[a]	34.2	25.9	3.2	63.2
18h nat/art + 6h ext	32.3	25.3	3.6	61.2
24h nat (control)	31.2	23.6	3.3	58.1
p-value	0.631	0.597	0.889	0.621

[a]18h nat/art: fluorescent growth light 3h preceding and 3h following 12h natural light

Table 5. Mean sensory scores (n = 24, 12 panelists and two replicates) of colour, odour, taste/flavour and texture attributes in swede roots grown under controlled photoperiods and photosynthetic light periods at 15 °C in Tromsø (69° 39’ N, 18° 55’ E). The photosynthetic light periods consisted of natural light (nat) and artificial growth light (art). Photoperiods were modified by extensions of low intensity light (ext) or darkness at night.

		12h nat	12h nat +12h ext	18h nat/art[a]	18h nat/art +6h ext	24h nat (control)	p-value
Colour	Hue	4.75	4.83	5.11	5.28	4.93	0.094
	Strength	4.35 b	4.93 a	4.97 a	5.08 a	4.98 a	<0.001
	Whiteness	5.47 a	5.20 ab	4.93 ab	4.88 b	5.08 ab	0.026
Odour	Acidic	3.52	3.25	3.63	3.08	3.90	0.100
	Green	2.97 ab	2.60 b	3.02 ab	2.85 b	3.59 a	0.006
	Sulphur	4.76	4.67	4.44	4.71	4.63	0.854
	Metallic	2.95	2.92	2.80	3.03	3.04	0.589
	Stale	1.98	2.35	1.98	2.26	2.06	0.395
	Soil	1.79	2.07	1.95	1.96	1.78	0.263
	Pungent	3.55	3.09	2.74	2.83	3.16	0.051
	Cellar	2.37	2.68	1.86	1.86	2.26	0.055
Taste/ flavour	Acidic	2.89 b	2.72 b	3.64 a	3.21 ab	3.89 a	<0.001
	Sweet	3.18 b	3.14 b	4.46 a	4.44 a	4.25 a	<0.001
	Bitter	4.97 a	4.96 a	3.52 b	4.17 ab	3.60 b	<0.001
	Green	2.83	2.76	2.99	2.84	3.04	0.575
	Soil	2.02	2.50	2.00	2.21	1.80	0.104
	Cellar	2.77 ab	3.46 a	2.08 b	2.35 ab	2.37 ab	0.024
	Pungency	4.38 a	3.77 ab	3.12 b	3.43 b	3.59 ab	0.006
	Sulphur	4.83 a	4.87 a	4.10 b	4.55 ab	4.57 ab	0.019
	Metallic	3.30 a	3.17 ab	2.79 b	2.87ab	2.95 ab	0.009
	Stale	2.48 ab	3.38 a	2.28 b	2.47 ab	2.44 ab	0.042
	Aftertaste	5.14 a	5.10 a	4.55 a	4.85 a	4.66 a	0.038
Texture	Firmness	5.15	5.30	4.75	5.07	5.08	0.138
	Crispiness	4.15	4.31	3.87	4.10	4.06	0.410
	Juiciness	4.45	4.63	4.53	4.49	4.83	0.508
	Astringency	3.00 ab	3.20 a	2.28 b	2.61 ab	2.53 ab	0.016
	Fibrousness	2.38 b	2.15 b	4.34a	4.36 a	4.21 a	<0.001

[a]18h nat/art: 3h fluorescent growth light preceding and 3h following 12h natural light

Different letters within rows indicate significant difference by Tukey’s multiple comparisons test (α = 0.05).