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Analytical Methods

Variation of glucosinolates in vegetable crops of *Brassica rapa* L. ssp. *pekinensis*

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ABSTRACT

Glucosinolates of Chinese cabbage (*Brassica rapa* L. ssp. *pekinensis*) from Korea were characterised to determine the total glucosinolate content and the diversity amongst glucosinolates; 24 varieties were analysed. The profiles of 14 glucosinolates identified from the leaves were subjected to principal component analysis (PCA) to evaluate the differences among varieties. The Kori, Sandun and e-Norang varieties separated from the others based on glucosinolate concentration. Genetically modified Chinese cabbage containing the *bar* gene could not be separated from non-genetically modified varieties. Glucobrassicanapin, 4-methoxyglucobrassicin, gluconapin and glucobrassicin in Chinese cabbage were confirmed as the main glucosinolate compounds. The Kori, Sandun and e-Norang varieties appear to be good candidates for future breeding programmes since they have a high glucosinolate content. The presence of indolic glucosinolates in all varieties should be studied more extensively because they are the precursor of indole-3-carbinol, a potent cancer chemopreventive agent.

1. Introduction

Glucosinolates are plant secondary metabolites that have long been of toxicological and pharmacological interest. They are an important group of phytochemicals present exclusively in 16 botanical families of the order of Capparales, and they are particularly abundant in Brassicaceae (Fahey, Zalcmann, & Talalay, 2001). They and/or their breakdown products are well known for their fungicidal (Pedras, Chumala, & Suchy, 2003; Pedras, Sarwar, Suchy, & Adio, 2006), bacteriocidal (Hashem & Saleh, 1999; Lin, Kim, Du, & Wei, 2000), nematocidal (Zasada & Ferris, 2004) and allelopathic properties and have recently attracted intense research interest because of their cancer chemoprotective attributes (Fahey et al., 2001).

In vitro studies have demonstrated that sulphoraphane, the isothiocyanate derived from glucoraphanin, inhibits Phase I enzymes responsible for activation of carcinogens and induces Phase II detoxification enzyme systems, thereby increasing the body's cancer defence mechanisms (Mithen et al., 2003; Zhang, Talalay, Cho, & Posner, 1992). Moreover, indole-3-carbinol, the hydrolysis product derived from glucobrassicin, is thought to modulate biotransformation enzyme activity and act as an anticarcinogen (Brew et al., 2009). However, the negative aspects of these compounds have been a major focus of research due to their "antinutritional" or goitrogenic properties. Compounds with these attributes include the glucosinolates found in protein-rich defatted meal from widely grown oilseed crops (Fenwick, Heaney, & Mullin, 1983) and in some domesticated vegetable crops (Griffiths, Birch, & Hillman, 1998; Rosa, Heaney, Fenwick, & Portas, 1997). *Brassica* vegetables therefore play an important role in the diet, and their naturally occurring glucosinolates should be monitored.

Chinese cabbage (*B. rapa* L. ssp. *pekinensis*), the major ingredient in kimchi along with hot pepper and garlic, is an important vegetable in Korea. The widespread popularity of kimchi as a fermented food in other countries has stimulated an increase in the production of Chinese cabbage. Recently, Chen, Zhu, Gerendás, and Zimmermann (2008) compared the composition and content of glucosinolates in five species of Chinese *Brassica campestris* vegetables. Total glucosinolates in Chinese cabbage, choysum and pakchoi were measured to be 36.4–89.6 μmol/100 g fresh weight (FW); the predominant individual glucosinolate was glucobrassicin. However, evaluation of the content or composition of glucosinolates in Korean *B. rapa* L. ssp. *pekinensis* has not been performed. Recently, the National Academy of Agricultural Science, Republic of Korea, developed Chinese cabbage that was genetically modified (GM) to tolerate glufosinate (Baek et al., 2008; Kim et al., 2009). The commercialisation of the GM crop requires a safety assessment for compositional equivalence.

The objectives of this study were to determine total glucosinolate content and profiles in leaves of *B. rapa* L. ssp. *pekinensis* from the Republic of Korea, and evaluate the glucosinolate equivalence

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of a transgenic Chinese cabbage containing the *bar* gene (TS22 line).

2. Materials and methods

2.1. Materials

The National Agrobiodiversity Centre (National Academy of Agricultural Science, Suwon, Republic of Korea) kindly provided 12 cultivars of Chinese cabbage: Kori, IT102910; Tschifu, IT212910; Sandun, IT213905; Taetschenpan, IT213906; Digeson, IT213907; Phoduran, IT213908; Tae Tschong Bang, IT213910; Zibu, IT214688; Fan Hsin Huang, K137045; Tung An Pai Tsai, K137046; Tsao Huang Pai, K137052; and Bp79, K137072. Other varieties were purchased from local seed markets (Gawulmat, Matchum, Buram, Samjin, Jangmi, Hwangseong, Hwiparam, Hukjinju, CR-mat, e-Norang and Noranja). GM Chinese cabbage (cv. Samjin) was developed using *Agrobacterium tumefaciens*-mediated

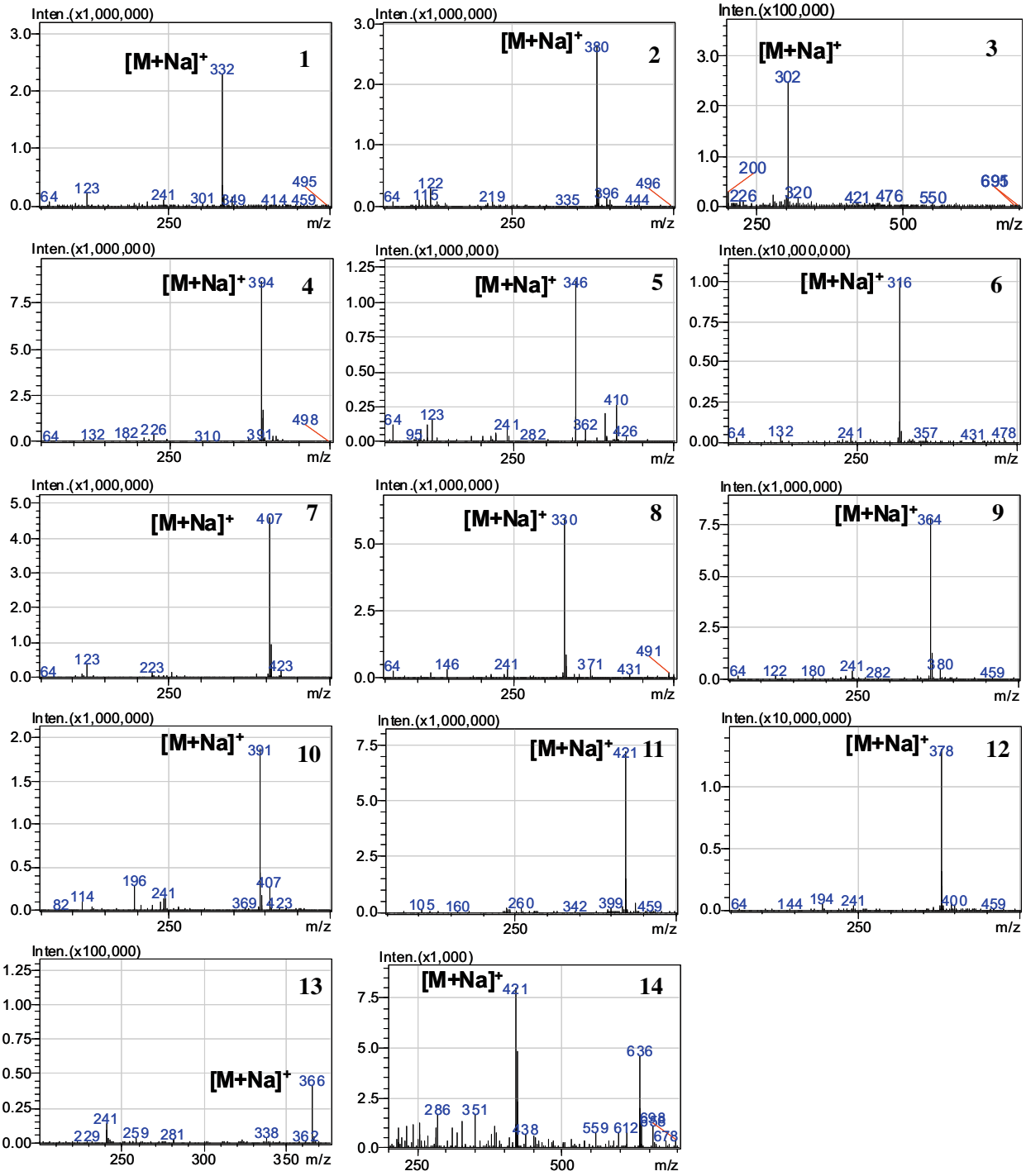

Fig. 1. MS spectra of desulphoglucosinolates extracted from Chinese cabbage. (1) Desulphoprogoitrin (RT: 8.7 min); (2) desulphoglucoraphanin (RT: 9.1 min); (3) desulphosinigrin (RT: 10.0 min); (4) desulphoglucoalysin (RT: 11.1 min); (5) desulphogluconapoleiferin (RT: 11.6 min); (6) desulphogluconapin (RT: 14.1 min); (7) desulpho-4-hydroxyglucobrassicin (RT: 15.4 min); (8) desulphoglucobrassicanapin (RT: 18.4 min); (9) desulphoglucoerucin (RT: 19.0 min); (10) desulphoglucobrassicin (RT: 20.7 min); (11) desulpho-4-methoxyglucobrassicin (RT: 22.8 min); (12) desulphoglucoberteroin (RT: 23.4 min); (13) desulphogluconasturtiin (RT: 23.5 min); and (14) desulphoneoglucobrassicin (RT: 29.8 min).

transformation. Chinese cabbage was genetically modified by the insertion of the *bar* gene which was isolated from genomic DNA of *Streptomyces hygroscopicus*. The T_6 generation of the GM Chinese cabbage was used for analysis.

The seeds were planted in 45-mm pots. After 20 days, the seedlings were transplanted in a glasshouse (National Academy of Agricultural Science, Suwon, Republic of Korea). The youngest leaves produced on each plant were cut after 60 days, immediately frozen in liquid nitrogen and stored at –80 °C. *Helix pomatia* type H-1 sulphatase, sodium acetate and sinigrin were purchased from Sigma (St. Louis, MO, USA). DEAE Sephadex A25 was obtained from GE Healthcare (Uppsala, Sweden).

2.2. Extraction and desulphation of glucosinolates

Samples were freeze-dried and disrupted in liquid nitrogen. Crude glucosinolates were extracted with 70% (v/v) boiling methanol (4.5 ml) from freeze-dried products (100 mg). Desulphation of the glucosinolates was performed on DEAE anion exchange columns. The columns were prepared by adding a slurry of Sephadex A25 activated with 0.5 M sodium acetate to a 1 ml mini-column (Bio-Rad Laboratories, Hercules, CA, USA) to give a final bed volume of 0.5 ml. An aliquot of each methanol extract was loaded onto a pre-equilibrated column and rinsed with 3 ml of distilled water. Next, 75 μl of purified sulphatase was loaded onto each column, and the desulphation reaction was performed overnight (16 h) at room temperature. The desulphated glucosinolates were eluted with 2.4 ml of distilled water and filtered through a 0.20-μm Teflon PTFE syringe filter for analysis. The eluates were analysed immediately by HPLC or stored at −20 °C until analysis.

2.3. Desulphoglucosinolate analysis using HPLC and LC/MS

For qualitative analysis, separation of desulphoglucosinolate was conducted on a C18 column (250 × 2.1 mm, 5 μm, Inertsil ODS-3; GL Sciences, Tokyo, Japan) using a HPLC system equipped with a diode array detector (Shimadzu, Kyoto, Japan). The elution buffers consisted of Buffer A (water) and Buffer B (acetonitrile). The flow rate was 0.2 ml/min. The following elution programme was applied: 0 min, 99% A/1% B; 18 min, 20% A/80% B; 30 min, 20% A/80% B; 32 min, 99% A/1% B; and 42 min, 99% A/1% B. The UV–Visible detector wavelength was set at 227 nm. For MS analysis, the eluate was diverted to a quadrupole mass spectrometer (LC/MS2010A; Shimadzu) equipped with a positive electrospray ionisation source. The spray voltage was set to 4.5 kV and the capillary temperature was set to 250 °C. The scan of the masses ranged from *m/z* 100 to *m/z* 700.

For quantitative analysis, desulphoglucosinolate extracts were separated on a C18 column (250 × 4.6 mm, 5 μm, Inertsil ODS-3; GL Sciences) using a HPLC system equipped with a diode array detector (LC-20A; Shimadzu). The HPLC conditions were the same as described previously, except the flow rate was 1.0 ml/min. Glucosinolate content was calculated using sinigrin as an external standard and the response factor of each compound relative to sinigrin (European Community, 1990).

2.4. Principal component analysis (PCA)

The glucosinolate content in Chinese cabbage varieties was analysed using PCA. The PCA was performed using the SAS 9.1 software package (SAS Institute, Cary, NC, USA) without preprocessing. All data were visualised using the principal components score and loading plots. Each point on the score plot represented an individual sample, and each point on the loading plot represented the contribution of an individual peak to the score.

3. Results and discussion

In this study, glucosinolate from Chinese cabbage growing in Korea was identified by LC/MS. Fig. 1 shows the MS spectra obtained from the Chinese cabbage. The molecular ion and fragmentation patterns matched that found in the literature (Barbieri, Pernice, Maggio, Pascale, & Fogliano, 2008) and allowed for unequivocal identification. According to the chemical structure, different glucosinolates can fall into three principal groups: the aliphatic group, indolic group and aromatic group. Fourteen kinds of glucosinolates, including nine aliphatic, four indolic and one aromatic glucosinolate, were detected in all varieties.

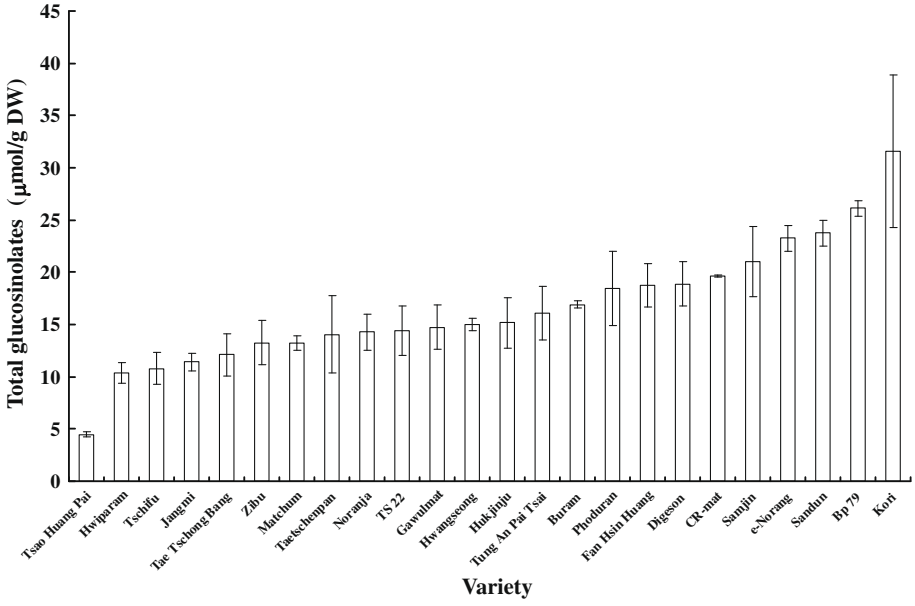

Fig. 2. Total glucosinolate content in 24 Chinese cabbage varieties grown in the Republic of Korea. Data are means of three plants ± SE. TS22, transgenic line; Samjin, conventional counterpart of transformant.

Significant differences in total glucosinolates were not observed amongst Chinese cabbage varieties except for the Tsao Huang Pai and Kori varieties (Fig. 2). The total glucosinolate content in the Chinese cabbage ranged from 4.48 to 31.58 μmol/g dry weight (DW). The glucosinolate content of fresh vegetables varies considerably based on date of planting, environmental conditions and length of growing seasons (Cartea, Velasco, Obregón, Padilla, & De Haro, 2008; Charron, Saxton, & Carl, 2005; Rosa & Heaney, 1996). However, Chen et al. (2008) reported that the total glucosinolate content in Chinese *B. campestris* vegetables ranges from 0.34 to 3.0 μmol/g FW. Previous work has shown that the total glucosinolates content in five groups of *Brassica oleracea* (broccoli, Brussels sprouts, cabbage, cauliflower and kale) ranges from 10.9 to 25.1 μmol/g DW (Kushad et al., 1999).

Identifying the compounds that exhibit the greatest variance within a population and determining closely related compounds are possible using PCA (Kim, Bamba, Harada, Fukusaki, & Kobayashi, 2007). PCA uses an *n*-dimensional vector approach to separate samples on the basis of the cumulative correlation of all component data and then identifies the vector that yields the greatest separation between samples (Kim et al., 2007). The data obtained for the 14 glucosinolates detected were subjected to PCA to outline the glucosinolate profile differences among varieties. The results are indicated by the principal components score plotting (Fig. 3). The abscissa represents the principal component 1 (PC1) score, while the ordinate represents the principal component 2 (PC2) score. Each plot in Fig. 3 implies the corresponding metabolome. PCA revealed that the two highest ranking principal components accounted for 51.7% of the total variance within the data set. The first principal component, accounting for 33.0% of the total variance, resolved the varieties according to the total glucosinolate content. In particular, the Kori variety clearly stood out from the other varieties in PC1, and the Sandun and e-Norang varieties were different from the others in PC2. To investigate the contributors of the principal components further, the metabolic loadings in PC1 and PC2 were compared. In PC1, the corresponding loading was positive for all glucosinolates. Gloconapoleiferin was the predominant glucosinolate in PC1. PCA was also used to determine the distribution of glucosinolates with different chemical structures in PC2. In PC2, the corresponding loading was positive for aliphatic glucosinolates, such as gluconapin, glucoalysin, glucobrassicanapin, glucoraphanin and sinigrin and negative for indolic glucosinolates including 4-hydroxyglucobrassicin, 4-methoxyglucobrassicin, neoglucobrassicin and glucobrassicin. PC2 directly correlated with gluconapin and was inversely related with 4-hydroxyglucobrassicin.

PCA has also been used widely to assess the differences between GM plants and their non-GM counterparts at the metabolome level (Catchpole et al., 2005; Kim et al., 2009). In this study, PCA could not distinguish GM Chinese cabbage (cv. Samjin) from non-GM Chinese cabbage. Special attention in the analysis of substantial equivalence has to be focused on inherent toxic and antinutritive constituents, since genetic modification could affect the expression of gene products not addressed by the genetic modification and thereby alter the content of constituents. OECD has promoted a list of well-defined metabolic constituents for assessment in compositional studies of new rapeseed, with particular emphasis on antinutrients such as glucosinolate. Comparison of glucosinolate content from the GM crop variety with consumer-accepted cultivars demonstrated substantial equivalence; the glucosinolate levels found in GM Chinese cabbage were within the range found in non-GM Chinese cabbage leaves (Tables 1 and 2).

In Chinese cabbage, the predominant aliphatic glucosinolates were glucobrassicanapin (0.49–8.08 μmol/g DW) and gluconapin (0.40–8.99 μmol/g DW) (Table 1). Glucobrassicanapin represented the greatest proportion (21.1%) of total glucosinolates in the leaves (data not shown). Several studies have reported a relationship between bitter taste and the content of some glucosinolate degradation products. Padilla, Cartea, Velasco, De Haro, and Ordás (2007) pointed out that the gluconapin and glucobrassicanapin were responsible for bitterness in *B. rapa*. The hydrolysis products of glucobrassicanapin and gluconapin are isothiocyanates, nitriles and epithionitriles.

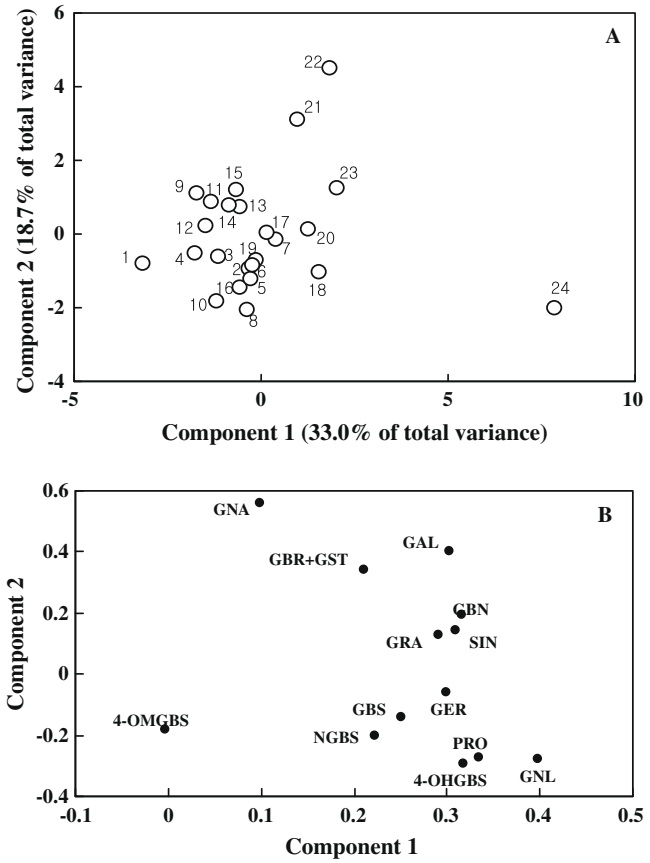

Fig. 3. Scores (A) and loading plots (B) of principal components 1 and 2 of the PCA results obtained from glucosinolate data on 24 Chinese cabbage varieties. (1) Tsao Huang Pai; (2) Hwiparam; (3) Tschifu; (4) Jangmi; (5) Tae Tschong Bang; (6) Zibu; (7) Matchum; (8) Taetschenpan; (9) Noranja; (10) TS22 (Transgenic line); (11) Gawulmat; (12) Hwangseong; (13) Hukjinju; (14) Tung An Pai Tsai; (15) Buram; (16) Phoduran; (17) Fan Hsin Huang; (18) Digeson; (19) CR-mat; (20) Samjin (conventional counterpart of transformant); (21) e-Norang; (22) Sandun; (23) Bp 79; and (24) Kori. PRO, Progoitrin; GRA, Glucoraphanin; SIN, Sinigrin; GAL, Glucoalysin; GNL, Gluconapoleiferin; GNA, Gluconapin; 4-OHGBS, 4-Hydroxyglucobrassicin; GBN, Glucobrassicanapin; GER, Glucoerucin; GBS, Glucobrassicin; 4-OMGBS, 4-Methoxyglucobrassicin; GBR, Glucoberteroin; GST, Gluconasturtiin; NGBS, Neoglucobrassicin.

Isothiocyanates have beneficial effects on human health. Therefore, the most promising varieties for future breeding purposes would be those with the highest total glucosinolate content. Kori and Sandun are two varieties that had the highest total glucosinolate content; the glucobrassicanapin content was 8.08 μmol/g DW and the gluconapin content was 8.99 μmol/g DW, respectively. Note that a previous study showed that these two varieties also had a higher content of flavonoids than other varieties (Kim et al., 2008). However, in the present study, glucoraphanin, the most predominant glucosinolate in broccoli, was very low (0.03–0.56 μmol/g DW).

Glucobrassicin and 4-methoxyglucobrassicin represented the great proportion (11.4% and 20.4%) of total glucosinolates in Chinese cabbage (Table 2). The breakdown products from indolic glucosinolates have been identified as phytoalexins in various *Brassica*

Table 1
Composition and content (μmol/g of DW) of aliphatic glucosinolates (GSs) in *Brassica rapa* L. ssp. *pekinensis*.

Variety	Aliphatic GSs[a]								
	PRO	GRA	SIN	GAL	GNL	GNA	GBN	GER	Total aliphatic GSs
Tsao Huang Pai	0.55 ± 0.17	0.04 ± 0.00	0.06 ± 0.00	0.27 ± 0.00	0.12 ± 0.09	0.58 ± 0.40	0.49 ± 0.34	0.04 ± 0.02	1.94 ± 0.80
Hwiparam	2.10 ± 0.25	0.27 ± 0.01	0.07 ± 0.01	0.69 ± 0.12	0.42 ± 0.15	0.40 ± 0.06	1.15 ± 0.33	0.58 ± 0.21	5.67 ± 0.74
Tschifu	0.43 ± 0.05	0.16 ± 0.07	0.12 ± 0.03	0.91 ± 0.37	0.15 ± 0.09	1.09 ± 0.92	1.50 ± 0.82	0.05 ± 0.01	4.41 ± 2.49
Jangmi	0.60 ± 0.18	0.07 ± 0.02	0.08 ± 0.02	0.37 ± 0.06	0.11 ± 0.04	1.97 ± 0.24	1.76 ± 0.19	0.20 ± 0.02	5.16 ± 0.30
Tae Tschong Bang	2.46 ± 0.53	0.15 ± 0.05	0.07 ± 0.00	1.40 ± 0.68	0.29 ± 0.08	1.25 ± 0.72	1.64 ± 0.80	0.07 ± 0.02	7.33 ± 3.35
Zibu	1.13 ± 0.66	0.56 ± 0.19	0.08 ± 0.00	1.05 ± 0.31	0.18 ± 0.11	0.84 ± 0.42	0.60 ± 0.16	0.27 ± 0.14	4.72 ± 1.14
Matchum	2.49 ± 0.59	0.40 ± 0.10	0.05 ± 0.01	1.00 ± 0.13	0.30 ± 0.05	1.58 ± 0.46	1.73 ± 0.17	0.75 ± 0.24	8.29 ± 0.60
Taetschenpan	1.91 ± 0.94	0.09 ± 0.02	0.08 ± 0.01	0.96 ± 0.62	0.59 ± 0.54	0.66 ± 0.22	2.43 ± 1.40	0.09 ± 0.01	6.81 ± 4.14
Noranja	0.23 ± 0.06	0.09 ± 0.00	0.06 ± 0.00	1.05 ± 0.11	0.04 ± 0.01	3.18 ± 0.46	4.27 ± 1.05	0.15 ± 0.02	9.06 ± 2.41
TS 22[b]	1.00 ± 0.08	0.06 ± 0.00	0.07 ± 0.01	0.33 ± 0.10	0.22 ± 0.03	1.08 ± 0.08	2.36 ± 0.29	0.07 ± 0.03	5.15 ± 0.37
Gawulmat	0.31 ± 0.11	0.10 ± 0.04	0.08 ± 0.03	0.90 ± 0.18	0.06 ± 0.02	3.14 ± 0.90	4.26 ± 0.76	0.17 ± 0.11	9.02 ± 2.36
Hwangseong	0.87 ± 0.10	0.04 ± 0.00	0.05 ± 0.01	0.61 ± 0.16	0.14 ± 0.03	3.51 ± 0.99	4.08 ± 0.39	0.16 ± 0.03	9.45 ± 1.98
Hukjinju	1.18 ± 0.19	0.17 ± 0.10	0.09 ± 0.02	0.79 ± 0.23	0.13 ± 0.03	3.54 ± 1.50	3.30 ± 0.76	0.29 ± 0.15	9.48 ± 3.15
Tung An Pai Tsai	1.15 ± 0.56	0.13 ± 0.04	0.04 ± 0.01	1.49 ± 0.35	0.23 ± 0.12	3.05 ± 1.44	3.54 ± 0.67	0.10 ± 0.08	9.76 ± 3.47
Buram	0.75 ± 0.51	0.10 ± 0.02	0.07 ± 0.00	1.05 ± 0.18	0.14 ± 0.10	3.20 ± 0.43	4.73 ± 0.08	0.27 ± 0.04	10.30 ± 0.13
Phoduran	2.18 ± 1.41	0.03 ± 0.00	0.06 ± 0.01	0.49 ± 0.19	0.50 ± 0.44	2.45 ± 1.41	4.48 ± 2.59	0.08 ± 0.06	10.27 ± 4.53
Fan Hsin Huang	0.70 ± 0.33	0.13 ± 0.04	0.08 ± 0.00	1.59 ± 0.32	0.14 ± 0.04	1.61 ± 0.06	3.77 ± 0.44	0.12 ± 0.02	8.12 ± 0.26
Digeson	1.05 ± 0.44	0.04 ± 0.01	0.12 ± 0.02	1.40 ± 0.09	0.54 ± 0.23	1.44 ± 0.53	6.37 ± 2.29	0.06 ± 0.03	11.01 ± 3.33
CR-mat	1.12 ± 0.21	0.05 ± 0.01	0.05 ± 0.01	1.42 ± 0.23	0.21 ± 0.02	2.00 ± 0.41	3.83 ± 0.17	0.12 ± 0.03	8.80 ± 1.21
Samjin[c]	1.23 ± 0.76	0.32 ± 0.06	0.07 ± 0.01	1.55 ± 0.46	0.17 ± 0.12	4.19 ± 0.79	3.88 ± 1.14	0.46 ± 0.06	11.87 ± 2.24
e-Norang	0.64 ± 0.04	0.24 ± 0.13	0.07 ± 0.00	2.50 ± 0.47	0.08 ± 0.01	4.55 ± 0.47	7.25 ± 0.96	0.41 ± 0.04	15.74 ± 1.29
Sandun	0.88 ± 0.65	0.45 ± 0.04	0.17 ± 0.08	3.66 ± 0.65	0.08 ± 0.03	8.99 ± 0.52	4.27 ± 0.33	0.07 ± 0.02	18.56 ± 1.22
Bp 79	2.22 ± 0.48	0.22 ± 0.05	0.08 ± 0.01	3.39 ± 0.50	0.41 ± 0.05	2.97 ± 1.02	4.26 ± 0.59	0.15 ± 0.03	13.70 ± 2.17
Kori	3.97 ± 2.06	0.49 ± 0.48	0.15 ± 0.03	2.13 ± 0.14	2.34 ± 2.75	1.93 ± 1.77	8.08 ± 6.89	0.94 ± 0.88	19.71 ± 13.07

[a] PRO, progoitrin; GRA, glucoraphanin; SIN, sinigrin; GAL, glucoalysin; GNL, gluconapoleiferin; GNA, gluconapin; GBN, glucobrassicanapin; GER, glucoerucin. Each value is the mean of three replicates ± standard deviation.
[b] TS22, transgenic line (cv. Samjin).
[c] Samjin, conventional counterpart of transformant.

Table 2
Composition and content (μmol/g of DW) of glucoberteroin, indolic and aromatic glucosinolates (GSs) in *Brassica rapa* L. ssp. *pekinensis*.

Variety	Indolic GSs[a]					GBR + Aromatic GSs
	4-OHGBS	GBS	4-OMGBS	NGBS	Total indolic GSs	GBR + GST
Tsao Huang Pai	0.02 ± 0.00	0.13 ± 0.04	2.07 ± 0.48	0.01 ± 0.00	2.22 ± 0.59	0.31 ± 0.12
Hwiparam	0.14 ± 0.03	0.34 ± 0.23	2.61 ± 0.39	0.08 ± 0.07	3.17 ± 0.85	1.51 ± 0.13
Tschifu	0.22 ± 0.05	2.53 ± 0.48	3.00 ± 0.52	0.18 ± 0.13	5.93 ± 0.40	0.43 ± 0.12
Jangmi	0.30 ± 0.06	0.93 ± 0.24	3.96 ± 0.51	0.04 ± 0.03	5.23 ± 0.97	1.02 ± 0.12
Tae Tschong Bang	1.07 ± 0.61	1.01 ± 0.09	2.07 ± 0.77	0.01 ± 0.01	4.17 ± 0.56	0.61 ± 0.03
Zibu	0.24 ± 0.09	2.69 ± 1.26	4.54 ± 0.85	0.11 ± 0.11	7.58 ± 2.73	0.94 ± 0.39
Matchum	0.17 ± 0.03	0.38 ± 0.07	2.32 ± 0.21	0.05 ± 0.02	2.92 ± 0.21	2.03 ± 0.42
Taetschenpan	0.46 ± 0.20	1.88 ± 0.94	4.30 ± 0.61	0.23 ± 0.17	6.79 ± 2.10	0.43 ± 0.18
Noranja	0.04 ± 0.01	0.42 ± 0.05	3.27 ± 0.11	0.02 ± 0.00	3.75 ± 0.08	1.47 ± 0.34
TS 22[b]	0.30 ± 0.13	3.54 ± 1.63	4.04 ± 0.84	0.28 ± 0.21	8.83 ± 3.85	0.41 ± 0.04
Gawulmat	0.13 ± 0.01	0.73 ± 0.31	3.36 ± 0.75	0.06 ± 0.03	4.27 ± 1.00	1.44 ± 0.78
Hwangseong	0.06 ± 0.01	0.53 ± 0.21	3.37 ± 0.68	0.24 ± 0.06	4.19 ± 1.16	1.36 ± 0.16
Hukjinju	0.19 ± 0.11	0.69 ± 0.39	3.03 ± 0.53	0.08 ± 0.05	3.99 ± 1.19	1.69 ± 0.70
Tung An Pai Tsai	0.05 ± 0.02	1.61 ± 0.62	3.16 ± 0.10	0.04 ± 0.02	4.86 ± 0.88	1.43 ± 0.89
Buram	0.14 ± 0.04	0.45 ± 0.08	3.48 ± 0.18	0.07 ± 0.01	4.15 ± 0.32	2.46 ± 0.33
Phoduran	0.48 ± 0.33	2.51 ± 1.54	4.52 ± 0.82	0.03 ± 0.01	7.53 ± 3.04	0.62 ± 0.13
Fan Hsin Huang	0.22 ± 0.09	3.99 ± 1.19	3.84 ± 1.37	0.35 ± 0.21	8.40 ± 3.39	2.19 ± 0.42
Digeson	1.29 ± 0.25	2.75 ± 0.51	2.43 ± 0.38	0.40 ± 0.38	6.87 ± 0.44	1.00 ± 0.25
CR-mat	0.56 ± 0.11	3.40 ± 0.96	4.83 ± 0.62	0.24 ± 0.10	9.02 ± 1.72	1.76 ± 0.34
Samjin[c]	0.45 ± 0.26	1.42 ± 0.75	4.77 ± 1.14	0.55 ± 0.71	7.19 ± 3.45	1.93 ± 0.38
e-Norang	0.05 ± 0.04	0.67 ± 0.26	3.38 ± 0.32	0.11 ± 0.04	4.20 ± 0.61	3.31 ± 0.51
Sandun	0.32 ± 0.12	1.42 ± 0.12	2.14 ± 0.77	0.03 ± 0.01	3.94 ± 1.00	1.26 ± 0.34
Bp 79	0.09 ± 0.04	6.81 ± 1.72	3.03 ± 0.08	0.11 ± 0.06	10.04 ± 2.16	2.36 ± 0.26
Kori	1.47 ± 0.84	4.47 ± 0.76	3.42 ± 1.29	0.38 ± 0.15	9.74 ± 2.63	2.13 ± 1.52

[a] 4-OHGBS, 4-hydroxyglucobrassicin; GBS, glucobrassicin; 4-OMGBS, 4-methoxyglucobrassicin; GBR, glucoberteroin; GST, gluconasturtiin; NGBS, neoglucobrassicin. Each value is the mean of three replications ± standard deviation.
[b] TS22, transgenic line (cv. Samjin).
[c] Samjin, conventional counterpart of transformant.

species, suggesting that they have a role in plant defence (Pedras et al., 2003, 2006). Furthermore, indole-3-carbinol is a potential chemopreventive agent (Brew et al., 2009; Tolonen et al., 2002) and thus Chinese cabbage could be a good candidate for future breeding programmes.

In conclusion, the glucosinolate content of several varieties of *B. rapa* L. ssp. *pekinensis* from Korea differed greatly. PCA serves to align, visualise and differentiate the components in large data sets. PCA in the present study allowed easy visualisation of complex data; the three varieties, Kori, Sandun and e-Norang, were

separated from the others in PC1 and PC2. The distribution of glucosinolates with different chemical structures was shown by the PC2 loading results and demonstrates the robustness of the present experimental system. In addition, the glucosinolate content of the GM Chinese cabbage was compositionally equivalent to that of several cultivars.

Consumers are aware of the need for a constant supply of phytochemical-containing plants for antioxidant support and disease prevention. This study provides valuable information regarding future breeding programmes for glucosinolate-containing plants. The breakdown products from glucosinolates have potential beneficial effects on human health. Glucobrassicanapin, 4-methoxyglucobrassicin, gluconapin and glucobrassicin were the major glucosinolates found in Chinese cabbage. Chinese cabbage containing a relatively high content of indolic glucosinolates should be of high dietary value.

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