Indian J. Plant Physiol., Vol. 15, No. 1, (N.S.) pp. 69-72 (Jan.-Mar., 2010)

SHORT COMMUNICATION

PATTERN OF GLUCOSINOLATE CHANGES IN INDIAN MUSTARD (*BRASSICA JUNCEA* L.) DURING DIFFERENT DEVELOPMENTAL STAGES

SATYANSHU KUMAR, J.S. CHAUHAN*, ARUNA ANDY AND M.L. MEENA

Directorate of Rapeseed-Mustard Research, Sewar, Bharatpur-321 303 (Rajasthan)

Received on 23 Oct., 2009, Revised on 10 Jan., 2010

SUMMARY

The changes in total glucosinolate in leaves, roots and stem at 3 developmental stages, pods and seeds of four advanced breeding lines BPR-897-4-11-8-4-91-2, BPR-897-4-11-8-4-91-5, BPR-897-4-11-8-6-93-10, BPR-897-4-11-8-6-93 -11 and Varuna variety of Indian mustard were investigated during 2008-09 cropping season. Highly significant data between developmental stages, plant parts and genotypes interactions suggested differential response of mustrad genotypes to glucosinolates. Glucosinolates in leaves were, in general, higher than that of stem and roots except for roots at 45 days after sowing (DAS). Genotype BPR 897-4-11-8-91-2 accumulated more glucosinolates in stem as compared to leaves and genotype BPR 897-4-11-8-6-93-11 showed more glucosinolates in leaves than roots at 45 DAS. Glucosinolates in stem and roots decreased at higher rate in comparison to that of leaves. Leaf glucosinolates at 45 DAS had positive relationship with that of 60 DAS (r = 0.871). Total glucosinolates in pods and seeds were also positively correlated (r = 0.814) and hence could be a good criterion for identifying low glucosinolates lines at early stage.

***Key words: Brassica juncea*, developmental stages, glucosinolates, leaves, roots, seeds**

Glucosinolates occur in many plants but *Cruciferae* family is the main source of more than 100 different types of glucosinolates (Grubb and Abel 2006). They are relatively non-toxic, however, enzyme myrosinase catalyzed hydrolysis produces a number of bioactive products having adverse as well as beneficial effects on human and animal health (Bones and Rossiter 2006). Their potential usage in the management of soil borne diseases and pests, impact on flavour and acceptance as human and animal feeds have been very well documented (D'Antuno *et al.* 2008). Glucosinolates are biosynthesised from amino acids and comprises amino acid side chain elongation, oxidative decarboxylation conversion in to basic structure and secondary modifications (Texter *et al*. 2007). Distribution of different types of glucosinolates varies with the developmental stage and specific part of the plant. Indolyl glucosinolates are major components in vegetative parts of *Brassica* species but are reported to be lacking in seed (Josefsson 1970). Pod wall has been reported to be the main site of glucosinolates synthesis (Lein 1972). However, its nature in transported form remained inconclusive. Understanding of its transportation mechanism would be very useful as it could be utilized to reduce glucosinolate content in seeds. Translocation has been identified as a major contributor to glucosinolate accumulation in seeds of *Sinapis alba* (Du and Halkier 1998). Further, in *Tropaeolum majus*, leaves have been observed to be the main site of aromatic glucosinolate synthesis (Lykkesfeldt and Moller 1993). Variable

*Corresponding author, E-mail: ms_nrcrm@yahoo.com

response of glucosinolates to seasons in *Brassica oleracea* leaves and different plant parts in *Brassica napus* have been reported (Fieldsend and Milford 1994, Peiwu *et al.* 2007, Yasumoto *et al.* 2007). However, limited information is available for Indian mustard (*Brassica juncea*), a predominantly grown oilseed *Brassica* in India, which has multifarious uses as human (leaves) and animal feed (oil cake) besides edible oil. The aim of the present study was to investigate the changes in total glucosinolates in different plant parts during various developmental stages of Indian mustard to assess their potential for exploitation as fresh vegetables, nutraceutical raw material and also as a bio fumigation agent to control soil borne disease and pests.

The experimental material for the present study comprised five genotypes of Indian mustard with differing glucosinolate contents, *viz*., BPR-897-4-11-8-4-91-2, BPR-897-4-11-8-4-91-5, BPR-897-4-11-8-6-93-10, BPR-897-4-11-8-6-93 -11 and Varuna (Table 1). The experiment was conducted during 2008-2009 cropping season (November-April) at the Experimental Farm, Directorate of Rapeseed-Mustard Research, Sewar, Bharatpur. Each genotype was grown in 6-row plots of 5 m length, spaced 30 cm apart. The plant spacing with in a plot was 10-15 cm, maintained by thinning. There were 3 replications. A fertilizer dose of 40:40:40 kg ha^{-1} (N: P_2O_5: K_2O) was applied at the time of sowing and 40 kg/ha N was top dressed 3-4 days after first irrigation (30 DAS). Second irrigation was given 70 days after sowing (DAS). Plant protection measures were adopted as and when required. Plant samples were collected at 45, 60 and 90 DAS and leaf, stem, roots and pods (90 DAS) were separated. Plant parts were first air-dried followed by hot air drying in an oven at 50 ^{0}C ± 2 ^{0}C. Dried material was finely powdered using an electric grinder. Total glucosinolates content in different plant parts as well as in pod and seeds was estimated by palladium complex method (Kumar *et al*. 2004). The data were analyzed as factorial completely randomized design to know the effects of genotypes, developmental stages, plant parts and their interactions following Gomez and Gomez (1984). Simple correlation coefficients of glucosinolates in various plant parts at different developmental stages were worked out following standard statistical methods

Table 1. Glucosinolate content in pods and seeds of Indian mustard genotypes

Genotype	Total glucosinolates		
	Pod (µmol g^{-1} dw)	Seed (µmol g^{-1} defatted seed meal)	
	2008-09	2007-08	2008-09
BPR 897-4-11-8-4-91-2	27.4	35.8^{c*}	48.9^{b}
BPR 897-4-11-8-4-91-5	16.9	46.3^{b}	41.4^{c}
BPR 897-4-11-8-6-93-10	25.8	47.7^{b}	49.6^{b}
BPR 897-4-11-8-6-93-11	15.3	29.7^{d}	34.1^{d}
Varuna	32.6	94.0^{a}	110.4^{a}

In a column, means followed by a common letter are not significantly different from each other at P = 0.05.

Total glucosinolates differed significantly among genotypes and was also significantly influenced by the developmental stages and plant parts. Highly significant genotype x developmental stages, genotype x plant parts, developmental stages x plant parts and genotype x plant parts x developmental stages interactions (Table 2) suggested differential response of glucosinolates to genotypes, plant parts and developmental stages.

Table 2. Analysis of variance for glucosinolates in Indian mustard

Source of variation	Degrees of freedom	Mean sum of squares
Genotypes	4	116.38**
Developmental stages	2	7133.84**
Genotype x developmental stages	8	54.22**
Plant parts	2	1192.26**
Genotype x plant parts	8	97.78**
Developmental stages x plant parts	4	552.92**
Genotypes x developmental stages x plant parts	16	104.90**
Error	90	1.102

LSD (P = 0.05): Genotypes 0.57; Developmental stages 0.44; Plant parts 0.44; Genotype x developmental stages 0.98; Genotype x plant parts 0.98; developmental stages x plant parts 0.76.

In the present investigation, glucosinolates in leaves varied from 22.4 in BPR 897-4-11-8-91 to 43.9 µmol g^{-1} in BPR 897-4-11-8-6-93 at 45 DAS and increased to 33.5 and 47.3 µmol g^{-1} in the same genotypes at 60 DAS. Thereafter, it showed sharp decline at 90 DAS in all the genotypes (Fig. 1). The leaf glucosinolates reduced substantially by 56 % in BPR 897-4-11-8-4-91-5 and 61.7 % in BPR 897-4-11-8-6-93-11. The reduction in mean glucosinolate content at 90 DAS over 60 DAS was 41.3 %. In stem, all the genotypes had the maximum glucosinolates at 45 DAS, ranging from 28.5 (BPR 897-4-11-8-6-93-11) - 36.3 µmol g^{-1} (BPR 897-4-11-8-4-91-5). It showed continuous decline and reached to a level of 5.4 µmol g^{-1} in Varuna to 15.6 µmol g^{-1} in BPR 897-4-11-8-4-91-2 at 90 DAS. The extent of decrease was 23.5% and 68.1% at 60 and 90 DAS, respectively. Similar trend was observed for root glucosinolates, in general, except genotype BPR 897-4-11-8-4-91-5. It ranged from 30.2-47.3; 35.2-41.8 and 5.8-11.4 3 µmol g^{-1}, respectively, at 45, 60 and 90 DAS. The decrease in glucosinolates was 8.4 % at 60 and 80.8 % at 90 DAS over the maximum glucosinolates at 45 DAS. However, glucosinolates in stem and roots decreased at higher rate in comparison to that of leaves. Glucosinolates in leaves were, in general, higher than that of stem and roots except for roots at 45 DAS. Genotype BPR 897-4-11-8-91-2 accumulated more glucosinolates in stem as compared to leaves and genotype BPR 897-4-11-8-6-93-11 showed more glucosinolaes in leaves than roots at 45 DAS.

Correlation analysis revealed that glucosinolates in leaves at all the three developmental stages did not show any association with glucosinolates in stem, roots, pods and seeds. Total glucosinolates in pods and seeds were positively correlated (r = 0.814). Leaf glucosinolates at 45 DAS had positive relationship with that of 60 DAS (r = 0.871). Similarly, glucosinolates in roots at 60 and 90 DAS also exhibited positive interrelationship (r = 0.813) and root glucosinolates at 45 DAS also showed positive association with seed glucosinolates. On the basis of pooled data over the three developmental stages, respectively, for total glucosinolates in leaves, stem and roots, there was a strong positive relationship between glucosinolates in leaves and roots (r = 0.828). Positive associations of glucosinolates in leaves with that of stem and roots were also recorded. The study revealed that glucosinolates in pods could be a good criterion for identifying low glucosinolates lines and saving time by rejecting high glucosinolate lines in the breeding programme.

Fig. 1. Changes in total glucosinolates in leaves (A), stem (B) and roots (C) at three developmental stages of Indian mustard. Genotype (1) BPR-897-4-11-8-4-91-2; (2) BPR-897-4-11-8-4-91-5; (3) BPR-897-4-11-8-6-93-10; (4) BPR-897-4-11-8-6-93-11; (5) Varuna.

ACKNOWLEDGEMENTS

Authors sincerely thank Director, Directorate of Rapeseed-Mustard Research, Bharatpur for providing necessary field and laboratory facilities for carrying out the present investigations.

REFERENCES

Bones, A.M. and Rossiter, J.T. (2006). The enzymic and chemically induced decomposition of glucosinolates. *Phytochemistry* **67:** 1053-1067.

D'Antuono, L.F., Elementi, S. and Neri, R. (2008). Glucosinolates in *Diplotaxis* and *Eruca leaves*: diversity, taxonomic relations and applied aspects. *Phytochemistry* **69:** 187-199.

Du, L. and Halkeir, B. (1998). Biosynthesis of glucosinolates in the developing siliquae walls and seeds of *Sinapis alba*. *Phytochemistry* **48:** 1148-1150.

Fieldsend, J.K. and Milford, G.F.J. (1994). Changes in glucosinolates during crop development in single and double low genotypes of winter oilseed rape (*Brassica napus*): Production and distribution in vegetative tissues and developing pods during development and potential role in the recycling of sulphur within the crop. *Ann. Appl. Biol.* **124:** 531-542.

Grubb, C.D. and Abel, S. (2006). Glucosinolate metabolism and its control. *Trends Plant Sci*. **11:** 89-100.

Josefsson, E. (1970). Glucosinolates content and amino acid composition of rapeseed (*Brassica napus*) meal as affected by sulphur and nitrogen nutrition. *J. Sci. Food Agric.* **21:** 98-103.

Kumar, S., Yadav, S.K., Chauhan, J.S., Singh, A.K., Khan, N.A. and Kumar, P.R. (2004). Total glucosinolate estimate by complex formation between glucosinolates and tetrachloropalladate (II) using ELISA reader. *J. Food Sci. Technol.* **41:** 63-65.

Lein, K.A. (1972). Genetische und phydsilogisch untersuchungen zur bildung von glucosinolaten in rapssamen: Lokalisierung des hauptbiosyntheseortes durch pfropfungen. *Zeitschrift fur Pflanzenphysiologie* **67:** 333-342.

Lykkesfeldt, J. and Moller, B.L. 1993. Synthesis of benzylglucosinolates in *Tropaeolum majus* L.: Isothyocyanates as potent enzyme inhibitors. *Plant Physiol.* **102:** 609-613.

Peiwu, L.I., Wen, Z., Xiaoxia, D., Xiaomei, C., Yongguo, Z., Yunchang, L.I., Conghua, X.I.E. and Tingdong, F. (2007). A preliminary study on glucosinolates hetrosis in leaf of hybrids in *Brassica napus* L. In: (Sustainable Development in Cruciferous Oilseed Crops Production) 12th International Rapeseed Congress, March 26-30, pp. 15-17. Wuhan, China.

Texter, S., de Kraker, J.W., Hause, B., Gereshenzon, J. and Tokuhisa, J.G. (2007). MAM 3 catalyzes the formation of all aliphatic glucosinolates chain lengths in *Arabidopsis*. *Plant Physiol.* **144:** 60-71.

Yasumoto, S., Matsuzaki, M., Hirokane, H. and Okada (2007). Changes in the contents of glucosinolates during crop development in different parts of rapeseed varieties. In: (Sustainable Development in Cruciferous Oilseed Crops Production) 12th International Rapeseed Congress, March 26-30, pp. 33-35. Wuhan, China.