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Chemical constituents in some promising genotypes of Indian Mustard [*Brassica juncea* (L.) Czern and Coss.]

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ABSTRACT

Among the oilseed *Brassica* crops, Indian mustard [*Brassica juncea* (L.) Czern & Coss.] is an important source of oil from a nutritional point of view. The nutritional value of oil and cake quality is governed mainly by the composition of its fatty acids, iodine value, saponification, acid value, glucosinolates, crude fibre, protein and limiting amino acids, etc. Seventeen varieties/strains of Indian mustard were taken for saturated and unsaturated fatty acid analysis. The eicosenoic was absent in genotype (NUDBYJ-10) and erucic acid (NUDBYJ-10, LES-46 and Pusa mustard- 21). The fatty acid composition found a variable in different genotypes. Saturated fatty acid, Palmitic + Stearic ranged between 2.3 to 6.5%, Oleic 10.6 to 40.7%, Linoleic 16.1 to 37.7%, Linolenic 13.3 to 26.7%, Eicosenoic 0.00 to 10.30% and Erucic acid 0.00 to 47.50%, respectively. Alternaria blight severity also varied in different genotypes and ranged between 18.75 to 56.25%, maximum being in genotype Kranti and minimum in LES-47. No significant correlation was observed between the fatty acid composition and disease severity. The oil content range from 38.1 to 42.60% and protein content was found highest in variety RGN-73. The amino acid viz. methionine and tryptophan range between 0.41 to 1.81 g/16gN and 0.41 to 1.81 g /16g N, respectively.

Keywords: Indian mustard, Fatty acid, Alternaria blight, Methionine, Tryptophan

INTRODUCTION

Brassicas are members of the Brassicaceae family (Singh *et al.*, 2017a). They occupy a unique position in world agriculture as the source of vegetables, oilseed, forage and fodder, green manure and condiments. *Brassica* seed oil is used for the edible purpose, as industrial lubricants and as a base for polymer synthesis. Oilseed brassica cake is used as a source of protein in animal feeds (Singh *et al.*, 2017b). Among the oilseed *Brassica* crops, Indian mustard [*Brassica juncea* (L.) Czern and Coss.] is an important source of oil from a nutritional point of view (Singh *et al.*, 2014). The nutritional value of oil is governed mainly by the composition of its fatty acids *viz*. Palmitic acid, Stearic acid, Oleic acid, Linoleic acid, Linolenic acid, Eicosenoic acid, Erucic acid along with anti-nutritional factors (Singh *et al.*, 2017a).

Indian mustard has a higher amount of erucic acid and glucosinolates in its oil and meal, respectively (Kewat, 2002). The high amount of glucosinolates in the meal and erucic acid in oil may create health problems *viz.* lipidopsis in young animals, fibrosis in older animals, reduce food intake, causes goitre, stroma and cancer (Singh *et al.*, 2013). In *Brassica* breeding, considerable emphasis is being laid to develop the varieties with low glucosinolates and low erucic acid. The development of rapid and accurate methods for determination of glucosinolate levels in seed in early 1960's led to the identification of *B. napus* cultivar "Bronowski" from Poland. The first double low spring *B. napus* cultivar "Tower" was released in 1974 and afterwards, a number of double zero cultivars were identified in different countries of the world. In India till date, the main emphasis has been to improve the rapeseed-mustard seed yield and oil content. It is necessary that seed oil quality especially the fatty acid composition also be improved wherever possible. Keeping this in view there is a need to screen/develop Indian mustard varieties having low erucic acid (Singh *et al.*, 2013).

MATERIALS AND METHODS

Seeds of seventeen genotypes of Indian mustard were obtained from different coordinating centers of AICRP on Rapeseed-mustard and Department of Genetics and Plant Breeding of ND University of Agriculture and Technology, Kumarganj, Faizabad and were sown at GPB Farm of the University. Five plants of each genotype were bagged for self-pollination and seeds were collected for a further study of chemical composition. Incidence and severity of Alternaria blight disease on each genotype were also recorded. The collected seeds were sun-dried followed by oven drying and were used as the experimental materials in the present investigation for chemical composition analysis. The oil content was determined by Soxhlet Extraction Procedure using petroleum ether (boiling point range 40-60^{o}C) by AOAC (1970).

Oil content

Seed sample was kept in the oven at 70^{0}C for removal of moisture. After removal of moisture, the seed was ground in mortar and pestle for extraction of oil. The conventional

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soxhlate method was used for estimation of foil (AOAC, 1970).

Protein content

In cake was estimated by conventional Kjeldahl method (AOAC, 1970). The nitrogen content was estimated in the cake and multiplied by the factor 6.25 in order to get the protein percentage in the sample.

Tryptophan content

Tryptophan content was estimated by the method of Spies and Chamber, 1949 by using p-dimethyl amino benzaldehyde and sodium nitrite solution and intensity of the colour was measure at 620nm by Spectronic -21. The calculation was done by the standard curve.

Methionine content

Methionine content was analysed as described by using sodium nitro pro side and glycine and metaphosphoric acid. The intensity of the colour was measure at 460 nm. The calculation was done on the basis of standard curve prepare for methionine.

Fatty acid profile of oil

Methyl-esters were prepared and the major fatty acids namely palmitic, stearic, oleic, linoleic, linolenic, eicosenoic and erucic acid were analysed at National Research Centre on Rapeseed-mustard, Bharatpur, Rajasthan using Perkin Elmer Autosystem XL Gas-liquid chromatography (GLC). The carrier gas was used as Industrial grade nitrogen (20 ml/minute flow rate). Hydrogen (ignition gas) and zero Air were also used for help in the ignition. Flame Ionization Detector (FID) was used. The calculations were done using the standard fatty acids. The SP 2300 and SP 2310 stainless steel column were used for fatty acid analysis.

Preparation of Methyl-esters for fatty acid profile

Methyl-esters of fatty acids were prepared from the oil obtained using Soxhlet apparatus. The method used for the preparation of methyl esters is described in "A manual of laboratory technique" published by Nutritional Institute of Nutrition, Hyderabad (AP) India. For preparing methyl-esters of fatty acids, one gram of filtered oil sample was transferred into receiving flask of a capacity of 250 ml. Twenty-five ml methanol and 0.4 g sodium metal were also added to same flask. The flask was then refluxed for two hours at 40-60°C on a water bath. The flask was cooled with running water. The content of flask was transferred into a separatory funnel of 250 ml capacity. Twenty five ml of distilled water and 0.2 ml methyl red indicator were also added in same separatory funnel which resulted in yellow colouration. Now 50% H_2SO_4 (Sulphuric acid) was added in same separatory funnel till pink colour is achieved. 10 ml petroleum ether (B.P. 40-60°C) was also added in the same separatory funnel and shake it well. Two layers were formed in the separatory funnel; lower layer of the separatory funnel was discarded. The upper layer of separatory funnel is called "organic layer". This layer was washed 2-3 times with addition of distilled water. 4-5 g of anhydrous sodium sulphate was added in the separatory funnel. The solution that was obtained after adding anhydrous sodium sulphate is called "Elute". The Elute was then concentrated and subjected for analysis at "Gas-liquid chromatography".

RESULTS AND DISCUSSION

The results indicated that the major saturated fatty acids *viz.*, palmitic and stearic acids along with mono and poly-unsaturated fatty acids like oleic, eicosenoic, erucic, linoleic and linolenic acids were found in the oil. Seventeen

Table 1: Fatty acid composition and *Alternaria* blight severity in different varieties/strains of Indian mustard

varieties/strains	16:0+18:0	18:1	18:2	18:3	20:1	22:1	AB
NUDBYJ -10	3.5	36.6	35.30	25.3	00.0	0.0	37.30
LES -46	6.5	38.5	32.5	26.2	0.5	0.0	25.75
NUDHYJ -6	2.9	18.1	26.7	16.9	3.5	37.5	45.22
JC -210 -541	4.6	17.5	25.35	15.7	10.3	29.5	39.75
Pusa mustard -21	4.5	30.5	37.7	16.8	6.7	0.0	49.00
LES -47	4.3	32.6	32.4	26.7	2.4	1.5	18.75
RLC -2	4.3	33.4	30.2	23.3	2.0	1.3	47.00
Kranti	2.3	16.75	16.1	16.7	7.3	46.0	56.25
RL -1359	3.5	10.6	17.5	15.3	6.5	47.5	50.60
JC -3762	4.5	40.3	30.7	13.3	2.5	0.3	46.56
JC -1359 23 -58	3.5	40.2	31.3	23.5	2.5	0.5	52.33
LES -44	4.4	38.6	32.5	21.3	2.6	0.2	27.56
RGN -73	4.0	13.5	16.3	15.7	7.5	43.6	25.35
ELM -134	5.6	40.7	30.3	14.2	2.1	36.5	47.35
PBR -1205	5.3	18.6	23.5	16.0	6.5	30.3	48.50
PQR -2005 -10	4.6	36.5	30.6	15.7	0.5	10.7	45.6
RH(OE) 0204	4.0	35.0	31.3	16.5	6.6	4.8	55.35

16:0=Palmitic acid, 18:0=Stearic acid, 18:1=Oleic acid, 18:2=Linoleic acid, 18:3=Linoleinic acid, 20:1=Eicosenoic acid, 22:1= Erucic acid, AB= Alternaria blight severity (%)

varieties/strains of Indian mustard were taken for fatty acid analysis. The eicosenoic was absent in genotype was not found in genotypes (NUDBYJ-10) and eruic acid (NUDBYJ-10, LES-46 and Pusa mustard- 21).

The fatty acid composition found a variable in different genotypes. Palmitic + Stearic ranged between 2.3 to 6.5%, Oleic 10.6 to 40.7%, Linoleic 16.1 to 37.7%, Linolenic 13.3 to 26.7%, Eicosenoic 0.00 to 10.30% and Erucic acid 0.00 to 47.50%, respectively.

The variations of different fatty acids showed conformity with those reported by several scientists (Banga and Banga, 2002, Singh *et al.*, 2013). These findings are also strongly supported by Anonymous (2006, 2008) who assessed the all seven fatty acids in 13 strains of *Brassica juncea* and found that it ranged from 2.3 to 5.3% palmitic acid, 1.1 to 2.7% stearic acid, 9.1 to 48.2% oleic acid, 16 to 38.9% linoleic, 9.8 to 15.2% linolenic, 0.9 to 14.1% eicosenoic acid and 0.00 to 51.1% erucic acid. Indian mustard and mustard oils are inferior in quality as they contain a high amount of erucic (28.0-53.0 %) and linolenic (8.5-22.7%) acids although they also contain linoleic (12.0-21.0 %) and oleic (10.0-24.0 %) acids which are nutritionally good (Bhowmic, 2003).

Among the various fatty acids recorded in Indian mustard oil, palmitic, stearic, oleic and linoleic acids are nutritionally desirable whereas, linolenic, eicosenoeic and erucic acids are undesirable. Among the undesirable fatty acids, erucic acid is typified fatty acid whose concentration is comparatively higher as compared to other acids in toria and mustard. Intake of the lower amount of mustard oil having 40% erucic acid would be safe for consumption as reported by Appelquist (1972).

However, several workers have reported that, by and large, due to higher intake of mustard oil having a high concentration of more than 40% erucic acid may cause some health problems like absorption, lipidosis in children and myocardial fibrosis in adults (Banga and Banga, 2002; Kumar, 2005).

Alternaria blight severity also varied in different genotypes and ranged between 18.75 to 56.25%, maximum being in genotype Kranti and minimum in LES-47 (table-1). No significant correlation was observed between the fatty acid composition and disease severity. The variations of the severity of Alternaria blight in different genotypes showed conformity with Singh *et al.* 2017.

Oil Content

The oil content among 17 verities/strain of Indian mustard was observed highest in variety Pusa mustard 21 (42.60) whereas the lowest value was observed in RGN-73 (38.11) (Fig.1). The similar results have been reported by Anonymous (2004), Singh *et al.* (2013).

This reflects the genetic character of the selected varieties. The oil content of Varuna was reported to be higher than Narendra Rai and Vardan by Kumar *et al.* (2001). The increase in oil content was mainly due to an increase in glycoside formation which on hydrolysis produced a higher amount of oil. Considering the degradation of carbohydrate complexes to sucrose formation might have helped in translocation and formation of fatty acids in the seeds. The results are in accordance with Bishnoi and Singh (1979). The oil content is influenced by factors like temperature and moisture during seed development, nitrogen doses and crop species. Generally, cool and moist conditions favour high oil content (Downey, 1983).

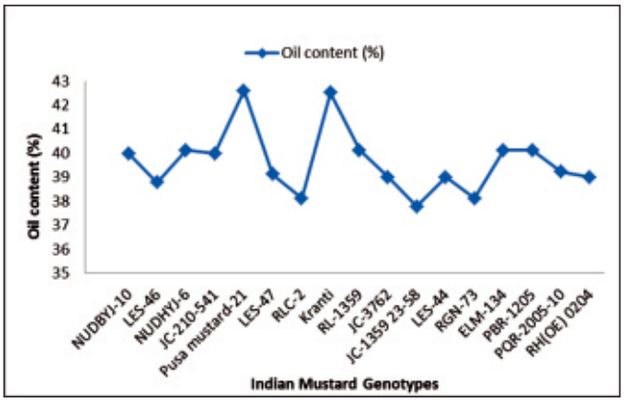

Fig.1 : Oil composition in different varieties/strains of Indian mustard

Protein Content

The protein content in 17 varieties / strain varied from 24-27.82 %. The highest value was recorded in variety Pusa Mustard-21 (28.68%) (Fig. 2). However, the lowest protein was observed in PBR-1205 (24.58%).

Protein synthesis depends upon the number of factors including varieties/strains, environmental factors such as light, temperature and water availability during plant growth and seed maturation. In dicotyledons, seed proteins are stored in the embryo (Duffus and Slaughter, 1980 and Downey and Rimmer, 1993). They exhibited a sequence of increasing trend in protein content in the cake of Indian mustard varieties/strains, where the oil content was reduced. It has also been reported by Kewat (2002) that distribution of protein content varies from seed to seed and it is unequal in distribution.

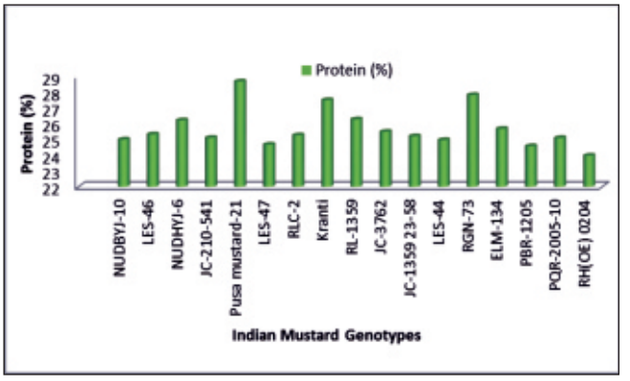

Fig.2: Protein composition in different varieties/strains of Indian mustard

Amino acid composition

Methionine content was found lowest in variety NUDHY- 1-6 (0.41g/16 gN) and highest value were observed in RL-1359 variety (1.81 g/16 gN) (Table 2). However, the Tryptophan

amino acid was recorded highest in variety RL1359 (1.401g/16 gN) followed by lowest in variety LES-46 (0.930g/16gN). These findings are also reported by Kewat and Abidi (2004), who observed that methionine content ranged between 0.77 to 1.34 g/16g N in 15 Indian mustard lines. Besides, one of the essential constituents of methionine is involved in a number of biochemical transmethylation reactions of vital significance in animal and plant (Karlson, 1968). A significant variation in methionine content was observed among the different varieties/strains. It was also confirmed by the observation of and Kewat (2002). In case of Tryptophan content, similar reports were also observed by Kewat (2002) and Kewat and Abidi (2004).

Tryptophan content has got a fundamental role in the biosynthesis of nicotinamide (Vitamin B6) as well as in other metabolic processes (Karlson, 1968). Tryptophan being an essential amino acid is a precursor of auxin (Indoleacetic acid). It acts in the metabolism as an activator of several enzymes such as carbonic anhydrous and alcohol dehydrogenase. The results are in support to Bjerg *et al.* (1987) and Nagraj (1995).

Table 2: Amino acid composition in different varieties/strains of Indian mustard

Variety/strains	Amino Acids composition	
	Methionine (g/16gN)	Tryptophan (g/16gN)
NUDBYJ-10	0.930	0.466
LES-46	1.110	0.495
NUDHYJ-6	1.167	0.411
JC-210-541	1.817	0.872
Pusa mustard-21	1.120	0.911
LES-47	0.932	0.811
RLC-2	0.998	1.110
Kranti	1.710	1.375
RL-1359	1.401	1.811
JC-3762	1.301	1.211
JC-1359 23-58	1.010	0.977
LES-44	0.991	1.017
RGN-73	0.952	1.011
ELM-134	1.227	1.045
PBR-1205	1.400	0.804
PQR-2005-10	1.301	0.971
RH(OE) 0204	1.222	0.979

CONCLUSION

It is concluded that the nutritional status oil and quality of cake is depends upon its fatty acids, limiting amino acids, oil and protein composition. Genotype NUDBYJ-10 contains no eicosenoic as well as no erucic acid was reported in the genotypes NUDBYJ-10, LES-46 and Pusa mustard- 21. The oil content range from 38.1 to 42.60% and protein content was found highest in variety RGN-73. Alternaria blight disease severity was not influenced by fatty acid composition in the tested genotype. There is a need to screen/develop Indian mustard varieties having low erucic acid since, high amount of erucic acid in oil may create health problems viz. lipidopsis in young animals, fibrosis in older animals, reduce food intake,

REFERENCES

Anonymous 2004. Annual progress report of All India Coordinated Research Project on Rapeseed-Mustard, National Research Centre on rapeseed-mustard, Bharatpur. pp. B-l to B-3.

Anonymous 2006. Annual Progress Report, All India Coordinated Research Project on Rapeseed-mustard, National Research Centre on Rapeseed-mustard, ICAR, Bharatpur, Rajasthan.

Anonymous 2008. Annual Progress Report, All India Coordinated Research Project on Rapeseed-mustard, National Research Centre on Rapeseed-mustard, ICAR, Bharatpur, Rajasthan.

AOAC 1970. Association of official Agricultural Chemist. Official Methods of Analysis, 11th edition. Washington DCP. Pp 438.

Appelquist LA. 1972. Chemical constituents of rapeseed (LA Appelquist and R. Ohlson, Editors) Elsevier, Amsterdam, PP-123.

Banga SK and Banga SS. 2002. Erucic acid heredity in oilseed Brassicas: A review. *J. Oilseeds Res.* **19** (1): 1-5.

Bhowmic TP. 2003. Oilseed Brassicas, Constraints and their management. CBS Publishers and Distributors, Darya Ganj, New Delhi, pp-18-21.

Bishnoi KC and Singh K. 1979. Oil yield and quality parameters of three raya varieties as affected by sowing time and nitrogen. *Indian J. Agron.* **24** (3): 255-285.

Bjerg B, Kachlick PW, Harsen LM and Sorensen H. 1987. Metabolism of glucosinolates. *Field Crops* **41**(10):70-75.

Downey RK. 1983. The origin and description of the *Brassica* oilseed crops, pp. 1-20. In: JKG Kramer, FD Sauer and WJ Pidgen eds. *High and low erucic acid rapeseed oils: Production, Usage, Chemistry and Toxicological Evaluation.* Academic Press, Canada.

Kewat R N. 2002. Biochemical studies on triple low contents and fatty acid composition in new varieties/strains of Indian mustard *(Brassic juncea* L. Czern Coss.). Ph. D. *Thesis*, N.D. University of Agriculture and Technology, Kumarganj, Faizabad, U.P. India.

Kewat RN and Abidi AB. 2004. Influence of enzymes on biochemical transformation from flowering to seed development in new strains and varieties of Indian mustard *(Brassica juncea* L. Czern and Coss.). *New Botanist.* **31**(1-4) : 195-202.

Kumar S, Singh B and Rajput AL.2001. Response of Indian mustard *(Brassica juncea)* to source and level of sulphur. *Indian Journal of Agronomy,* **46**(3): 528- 532.

Kumar, PR. 2005. Rapeseed-mustard a high quality oil for health. News circular of All India Coordinated Research Project on Rapeseed-mustard (ICAR), Bharatpur (Rajasthan). **7** (8): 48-51.

Nagraj G. 1995. Quality and utility of oil seeds. Directorate of Oilseed Research, Hyderabad, pp. 9-16.

Singh AK, Singh AK, Choudhary AK, Kumari A and Kumar R. 2017b.Towards oilseeds sufficiency in India: Present status and way forward. *Journal of AgriSearch* **4** (2):80-84.

Singh R, Singh AK and Kumar P. 2014. Performance of Indian mustard (*Brassica juncia* L.) in response to integrated nutrient management. *Journal of AgriSearch* **1**(1): 9-12.

Singh S, Singh HK, Khan NA, Singh RP and Maurya MK. 2017a. Influence of sulphur and zinc on biochemical properties and severity of alternaria blight in Indian mustard. *Journal of AgriSearch,* 4 (3): 212-214.

Singh S, Singh RP, Singh HK and Kumar K. 2013. Oil and cake quality characteristics in new varities/strains of Indian mustard [*Brassica juncea* (L.) Czern and Coss]. *Plant Archives.* **13** (2): 627-632.

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