International Research Journal of Biotechnology Vol. 12(6) pp. 1-6, July, 2022
Available online http://www.interesjournals.org/IRJOB

Research Article

Breeding for Quality Traits Improvement of Mustard (*Brassica Juncea*)

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Received: 01-Jul-2022, Manuscript No. IRJOB-22-63401; **Editor assigned:** 04-Jul-2022, PreQC No. IRJOB-22-63401(PQ); **Reviewed:** 18-Jul-2022, QC No. IRJOB-22-63401; **Revised:** 23-Jul-2022, Manuscript No. IRJOB-22-63401(R); **Published:** 30-Jul-2022, DOI: 10.14303/irjob.2022.12

Abstract

Brassica juncea (2n=36) is an amphidiploid species derived from inter specific cross between *Brassica nigra* (2n=18) and B. rapa (2n=20). *B. juncea* is used as sources of oil, vegetable, condiments and fodder and act as laxative, stimulant to gastric mucosa and increase intestinal secretion. Mustard oil was once considered to be unsuitable for human consumption due to high content of erucic acid. Early maturing, high yielding Australian canola quality *brassica juncea* lines have been developed and are currently being crossed with higher oleic acid sources. In *brassica juncea* the most quality to be considered are erucic content, low glucosinate content, high fatty acid accumulation, and productivity per unit area flavonoids, resistance disease and essential oil. Economically important traits were transferred through hybridization aided with embryo rescue; double low, high oil content, shattering tolerance in *B. napus*, and low erucic/high oleic acid, yellow seed coat color, double low, and resistance/ tolerance to fungal diseases, *Albugo candida* and *Alternaria brassicae* in *B. juncea.* Therefore to boost quality and quality traits in *Brassica Juncea*, heterosis breeding is appropriate method.

Keywords: *Brassica juncea*, Flavonoids, Glucosinate, Quality traits

INTRODUCTION

Brassica juncea is an important oilseed crop in India, China and in south-western areas of the former Soviet Union. India is one of the largest producers of mustard in the world. Crop Brassicaceae are important sources of edible oil with lowest amount of saturated fats and leafy vegetables rich in minerals, anti-oxidants, and tasteful condiments crops (Agnihotri A et al., 2004). Rapeseed-mustard is an important group of oilseed crops accounting about one-fourth of the total oilseeds production in the country (Amy McInnis, 2004).

Indian mustard (*Brassica juncea* L. Czern and Coss) is the second largest oilseed crop in Pakistan after cotton seed oil. It accounts for nearly 30% of the total oilseeds and 27% to edible oil pool of the country (Saeed et al., 2013). On average the local production of edible oil is only 22% and the remaining 78% is being imported from abroad by expending huge foreign exchange (Anonymous, 2013). Fatty acids profile play key role in the use of brassica oil by human beings (Bell J M, 1984). Vegetable oil seed having higher proportion of 16 and 18 carbons unsaturated fatty acids, particularly monounsaturated fatty acids are considered suitable for use as edible oil (Simopoulos, 2008; Ramos et al., 2009; Priyamedha et al*.,* 2014).

The seed of Indian mustard contains 35-45% oil having 92-98% triacylglycerol of fatty acids (C16-C22). It contains lowest saturated fat and possesses more proportion of linoleic (C18:2) and linolenic (C18:3) acid which are not synthesized by the human body (Bhowmik TP et al., 1980). Linolenic acid is an essential dietary fatty acid, but undesirable in edible oil because of being prone to autooxidation resulting in off-

Table 1. Nutritive Value of Mustard Leaves and Seeds (Per 100 gms of Edible Portion).

Food Component	Mustard leaves	Mustard seeds
Moisture	89.8g	8.5 g
Protein (NX 6.25)	4.0 g	20.0 g
Fat	0.6 g	39.7 g
Minerals	1.6 g	4.2 g
Crude fiber	0.8 g	1.8 g
Carbohydrates	3.2 g	23.8 g
Energy	34Kcal	541 Kcal
Calcium	155 mg	490 mg
Phosphorus	26 mg	700 mg
Iron	16.3 mg	7.9mg

Table 2. Enhanced Quality Mustard Strains Registered at ICAR.

TERI GZ-05 - INGR 04078 [TERI-Uphaar]	High oleic and linoleic acid, yellow seeded, double low B. juncea
TERI(OO) R9903 - INGR 04077 [TERI-Uttam]	High oil content, canola quality, early maturing B. napus
TERI (OO) R986-INGR 99007 [TERI-Gaurav]	Early maturing, dwarf double low B. napus
TERI (OO) R985-INGR 99008[TERI-Garima]	High oleic acid, double low B. napus
TERI(OE) R09-INGR 98005[TERI-Shyamali]	Low erucic acid, high oleic B. napus
TERI (OE) R03-INGR 98002 [TERI-Phaguni]	Low erucic-acid, early maturing B. napus
TERI (OE) M21-INGR 98001 [TERI-Swarna]	Low erucic acid, yellow seeded, early maturing B. junce

flavours and reduced shelf life of the oil (Chen S et al., 2013). Erucic acid (C22:1) comprises nearly 50% of total fatty acid which is undesirable for human consumption as they are reported to impair myocardial conductance and increase blood cholesterol. Therefore, it is dire need to reduce glucosinolate and erucic acids in the seed oil of *B. juncea* (Priyamedha et al*.,* 2014).

By product of brassica species which remain after oil extraction is a valuable commodity obtained having sufficient amount of essential amino acids and protein. They also contain some minerals like Ca, Mg and P and vitamin B4 and E as well. Presently, Indian mustard is not only used for livestock feeding, but also play an important role as raw material for the production of various industrial products (Bala et al., 2012). However, in comparison to other popular sources such as soybean, it contains high amount of glucosinolate, which lessen its feed value (Wanasundara, 2011). The amelioration of nutritional qualities of seed meal of Indian mustard by reducing the amount of glucosinolate, therefore, can be of high economic value. Therefore high amount of erucic acid and glucosinolate, are the major hurdle in the production double of zero canola types *B. juncea* varieties.

Although international efforts had been started in the 1950s, for the developing rapeseed-mustard strains with double low characteristics, but these efforts was restricted to *B. napus* and not a single variety has been developed till date in Indian mustard, possessing double low traits (Priyamedha et al., 2014). To boost quality and quality traits in Brassica and other crops heterosis is the most appropriate method (Hassan et al., 2006). Through heterosis we can increase our production in short time by utilization of less input (Pal et al., 1956).

Until recently most of the *B. juncea* types grown in the world were of conventional mustard quality as they contained high levels of glucosinolates in the meal and high levels of erucic acid in the oil fraction. During the past two to three decades, significant attempts have been made to introduce canola quality traits into *B. juncea* in an effort to change its seed quality while retaining its agronomic benefits .The first significant achievement in this direction was the development of low erucic acid *B. juncea* (Kirk et al., 1981). The next breakthrough occurred with the development of a low glucosinolate form of *B. juncea* (Love, et al., 1991) following interspecific hybridization between *B. rapa* and *B. juncea*. The objective of this review is recognizing the traits that have positive contribution to quality of mustard.

LITERATURE REVIEW

Origin and domestication of mustard (*Brassica Juncea*)

B. juncea is described as one of the earliest domesticated plants, with records of its use in Indian agriculture dating back to 2300 BC. Afghanistan is currently described as a primary center of origin for oilseed forms (Chen et al., 2013). China, is where the largest subspecies diversity is considered as a probable primary center for vegetable types (OECD 2012) (Wu et al., 2009).Using Simple Sequence Repeat (SSR), Amplified Fragment Length Polymorphism (AFLP) and Sequence Related Amplified Polymorphism (SRAP), it was demonstrated that oilseed varieties cultivated in China, India, Europe, Australia, Japan and Canada could be divided in two genetically distinct groups (Chen et al., 2013; Srivastava et al., 2004; Wu et al., 2009).

Brassica juncea (2n=36) is an amphidiploid species derived from inter specific cross between *Brassica nigra* (2n=18) and B. rapa (2n=20). Wild forms of *Brassica juncea* have been found in the near East and Southern Iran. There are conflicting views about the origin of *B. juncea* (Bhowmik, 2003). According to Vavilov (1949) Afghanistan and its adjoining regions were the primary center of its origin while central and western China, Eastern India and Asia Minor with Iran comprised the secondary centers of origin.

Middle East has been proposed as the most probable place of origin of *B. juncea* as wild forms of its progenitor species *B. rapa* and *B. nigra* occur together in this region (Olsson, 1960a, b; Mizushima et al., 1967; Prakash et al., 1980). The regions of south western China and North Western Himalayas may constitute two secondary centers where there is enormous diversity in *B. juncea* forms. Biochemical studies support this finding about the diversity in these regions (Vaughan et al., 1963) and further provide evidence for the existence of two geographical races of *B. juncea*, the Chinese pool and the Indian pool (Vaughan et., 1973). This evidence is supported by Song et al., 1988, through RFLP studies which suggest two centers of origin Middle East and China. However Rakow (2004) had opined that China cannot be considered as a center of origin for *B. juncea* because the two parent species *B. nigra* and *B. rapa*, were never found as wild species in that country.

Importance of mustard (*Brassica Juncea*)

Brassica juncea is used as sources of oil, vegetable, condiments and fodder. The oil content of the seeds ranges from 38- 46%. The conventional varieties of *B. juncea* are high in Erucic acid (~40-50%) as well as in glucosinolates (180-200 micro moles). Internationally erucic acid is included as one of the hazardous constituents in food material in the OECD-WHO Food Safety Standards and FAO's Manual on Food Safety Assessment (2008). The seed and oil are used in the preparation of pickles and for flavouring curries and vegetables. Whole seeds, ground or in powdered form, prepared pastes, sauces and oil are all used in cooking. The aroma and pungent flavour of mustards come from the Sulphur containing glucosinolates. Mustard seeds and oil have been traditionally used to relieve muscle pain, rheumatism and arthritic pain. In India, mustard oil is applied over scalp and is believed to stimulate hair growth. Ground mustard seeds act as a laxative, stimulant to gastric mucosa and increase intestinal secretion (Tables 1 and 2).

In Korea, it is used for both food itself and the major ingredient of kimchi, a traditional fermented vegetable food, and kimchi including mustard leaf has recently attracted a lot of attention as a functional food for health maintenance and disease prevention (Kim et al., 2003). The essential oil of *Brassica juncea* seeds, also referred to as mustard oil, has also been used in cosmetics for hair control (Yu et al., 2003).

The major pungent chemical constituent of such commercialized oils is Allyl isothiocyanate which is formed from its precursor during the processing of the seeds (Yu et al., 2003). This isothiocyanate is now considered to be the most important cancer chemo-preventive phytochemical with other potential health benefits (Okulicz 2010) (Zhang et al., 2010) and antimicrobial agent against a variety of organisms (Luciano and Holley, 2009). Structurally diverse glucosinolates and other precursors of isothiocyanates are encountered not only in *Brassica juncea* leaves (Hill et al., 1987), but also in diverse other edible cruciferous vegetables well recognized for their health benefits (Higdon et al., 2007). Amongst many such vegetables, the glucosinolates contents of *Brassica juncea* leaves are reported to be the highest (McNaughton et al., 2003). In general, contents of these Phyto-chemicals in seeds of Brassicacea family grown in tropical environment are higher than of those grown in temperate regions (Tripathi et al., 2007). *Brassica juncea* is known to produce several other classes of bioactive phyto-chemicals including glycosides, flavonoids, phenolic compounds, sterols and triterpene alcohols, proteins and carbohydrates (Appelqvist et al., 1973; Das et al., 2009; Fabre et al., 1997; Jung et al., 2009; Li et al., 2000; Sang et al., 1984; Yokozawa et al., 2002).

Quality traits in mustard (*Brassica juncea)*

Erucic Acid of the total fatty acids, there is predominance of erucic acid fraction (35.7–51.4%) (Chauhan et al., 2007), in mustard cultivars that are sown in India. The effects of erucic acid from edible oils on human health are controversial. Although, no negative health effects of any exposure to erucic acid have ever been reported in human beings in studies based on laboratory animals during early 1970s, (Amy McInnis, 2004) erucic acid appears to have shown toxic effects on the heart at high doses (Food Standards Australia, New Zealand, 2003). Mustard oil was once considered to be unsuitable for human consumption due to high content of erucic acid in United States, Canada and Europian Union. Subsequent studies on rats have shown that they are less able to digest vegetable fats (whether they contain erucic acid or not) than humans and pigs. In Indian *B. juncea*, low erucic acid is controlled by two recessive genes (Kirk et al., 1983).

Glucosinolates

Presence of high levels of Glucosinolates (Chauhan et al. 2002), limits its utilization as livestock and poultry feed. The intact glucosinolates are harmless but the myrosinase enzyme, chemically splits the glucosinolates into glucose and nutritionally undesirable toxic isothiocynates, oxazolidinethione or nitriles. Higher consumption of meal of such seeds in the diet causes enlargement of thyroids in poultry and low palatability for cattle etc. Mustard meal is therefore unusable for non-ruminants like pigs and poultry and can be fed only in a limited way to cattle. Low glucosinolate is controlled by 6-7 recessive genes (Sodhi et al., 2002).

Productivity

To realize further gains in productivity, it is important to utilize new sources of variation which would lead to broadening the genetic base of the existing varieties. Productivity can also be substantially increased by heterosis breeding. It has been shown in the earlier work that there are two diverse gene pools for oilseed *B. juncea*, the east European gene pool and the Indian gene pool and hybrids between lines of the two divergent gene pools are heterotic for yield (Pradhan et al., 1993; Jain et al., 1994; Srivastava et al., 2000). For large scale production of hybrid seeds in mustard, barnase-barstar male sterility and restorer system developed lab through transgenic approaches (Jagannath et al., 2001; Jagannath et al., 2002).

Flavonoids

Colored flavonoids, anthocyanins are the most important group of plant pigments, also considered as multifunctional components of food due to their antioxidant activities and other beneficial biological properties (McDougall et al., 2007; Moreno et al., 2010; Sadilova et al., 2006). The most common anthocyanins are pelargonidin, cyanidin, delphinidin, peonidin, petunidin and malvidin, with cyanidin the most common in Brassica crops (Lo Scalzo et al., 2008; Moreno et al., 2010; Tatsuzawa et al., 2006).

Essential oil

Isothiocyanates are known to be the main group of constituents in the essential oils of *Brassica juncea*. It includes allyl isothiocyanate (54.8-68.8%), 3-butenyl isothiocyanate (4.8-5.9%) and phenethyl isothiocyanate (2.4-3.4%). They represent more than 62.9% of the total essential oil. The sulfides are present in relatively small amounts (14.8-23.4%). They include Diallyl trisulfide (7.8-9.7%), diallyl sulphide (3.2 - 5.5%) and diallyl disulfide (2.7 - 4.1%) (Yu et al., 2003).

Improvement of quality traits in mustard (*brassica juncea*)

Several economically important traits were transferred *via* wide hybridization aided with embryo rescue; double low, high oil content, shattering tolerance in *B. napus*, and low erucic/high oleic acid, yellow seed coat color, double low, and resistance/ tolerance to fungal diseases, *Albugo candida* and *Alternaria brassicae* in *B.juncea.* Seven improved quality genotypes are registered at ICAR (Agnihotri et al., 2004).

Among all the constituents mustard, fibers, tannin, phytic acid, glucosinolate and sinapin lower the feed value of seed meal (cf Chauhan et al., 2002). Glucosinolates are a group of plant thiolucosides found principally among members of family Cruciferae (*Brassicaceae)* principally determines the quality of seed meal. The vegetable tissue and seed of Cruciferous plant contain one or more of over 120 known glucosinolates (Fenewick et al., 1983). Cleavage products from hydrolysis of glucosinolates are detrimental to animal health as they reduce the feed palatability; affect the iodine uptake by the thyroid glands thus reducing feed efficiency and weight gains (Bell 1984; Fenewick et al., 1983), especially in non-ruminants such as pigs and poultry. Thus it is imperative to reduce glucosinolate in Indian mustard cultivars from their present level of very high amount. Glucosinolates and their derived properties have also been reported to have health beneficial effects by reducing the risk of certain cancers in humans (Zhang et al., 1994; Fahey et al., 2001; Shapiro et al., 2001; Mithen et al., 2003), while other glucosinolates are detrimental for human and animal consumption (Rosa et al., 1997). Epidemiological studies indicate that consumption of brassica vegetables is associated with a reduced incidence of cancers at a number of sites including the lung, stomach, colon and rectum (Conaway et al., 2001).

Breeding efforts have been underway since 1970 to reduce glucosinolate content in the seed of rapeseed mustard varieties up to 30 micro moles/g defatted seed meal and erucic acid up to 2% as well as combining both to develop double zero or double low varieties to meet the internationally acceptable standard of oil and seed meal. (Chauhan et al., 2002) reviewed the quality improvement programme of rapeseed-mustard in India. First low erucic acid variety, Pusa Karishma of Indian mustard and first double low variety, GSC 5 of gobhi sarson was released in 2004 and 2005, respectively. Presently, five low erucic varieties have been released in *Brassica juncea*. In gobhi sarson (*B.napus*), 5 double low (low erucic and lo glucosinolate) varieties have been released. The current efforts are to recombine low erucic acid with low glucosinolate content in Indian mustard and refining the agronomic base to improve yield potential of double low gobhi sarson strains.

CONCLUSION AND RECOMMENDATION

Several economically important traits were transferred *via* wide hybridization aided with embryo rescue; double low, high oil content, shattering tolerance in *B. napus*, and low erucic/high oleic acid, yellow seed coat color, double low, and resistance/ tolerance to fungal diseases (Gopalan C et al., 2007). In *brassica juncea* the most quality to be considered are erucic content, low glucosinate content, high fatty acid accumulation, and productivity per unit area, flavonoids, resistance disease and essential oil are the most important parameters that need further breeding. High glucosinate content low acid fatty acid composition resistances to stress are require further research to enhance the crop. In general to improve the consumption status of mustard, breeding intervention is strictly required for enhancing these important traits.

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