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A brief overview: Present status on utilization of mustard oil and cake

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Edible oil consumption in India has nearly doubled to reach 13.4 kg (2010 – 2011) per capita in the last five years, and the value is expected to grow further to 24 kg by 2020. Traditionally, mustard oil is popular edible oil in certain regions of India; its consumption accounts 18% of the total oil need of the country. Due to balanced amount of monounsaturated and polyunsaturated fatty acids, doctors and consumers are inkling towards incorporation of mustard oil in domestic cooking. In recent past, mustard accounted for 26% of total oilseeds produced in India, and the production is expected to rise. The cake/meal, i.e. the residue/by-product obtained after extraction of oil is, therefore, anticipated to increase proportionately. Mustard cake is a rich source of protein, but contains some anti-nutritional factors also. Several research papers, patents and assessment reports are available on mustard oil including the utilization of the cake, viz. for agriculture, animal livestock and as a proteinaceous resource for human consumption. This overview is an attempt to integrate the above information on a common platform.

Keywords: Mustard seed, Mustard cake, Mustard oil, Utilization of mustard cake

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Global oilseed production is projected to rise by 4% during 2013/14[1]. By the year 2020, the production is expected to expand further by 23%, and will be led by United States of America (US), Brazil, China, India, Argentina and European Union (EU). While the biodiesel industry is projected to represent a significant source of demand in the EU and US, in China and India growth occurs primarily in food use[2]. Edible oil consumption in India has nearly doubled to reach 13.4 kg (2010 – 2011) per capita in the last 5 yrs, making India the second largest consumer (after China); the value is expected to grow further to 24 kg by 2020[2].

Mustard or rapeseed is among the major oilseeds in the world, and belongs to the genus '*Brassica*'. In India, the majorly grown species are *B. juncea* (L.) Czern. (brown mustard) and *B. campestris* L. syn. *B. rapa* L.(yellow mustard) along with *B. nigra* (L.) Koch (black mustard) and *B. napus* L. subsp. *oleifera* DC. The other species grown worldwide include *Sinapis alba /B. hirta* (white/pale yellow mustard). *B. hirta*, *B juncea* and *B. nigra* are also known for their condiment value. Species like *Sinapis alba and B. napus* are being increasingly used as cover crops in temperate region. Though the plant is cultivated as oilseed crop, the leaves of young plant are used as vegetable, and are good source of vitamin A, vitamin C, calcium and iron.

Mustard seed contain about 24 – 40% oil, 17 – 26% protein and 19% hull. Mustard seeds are processed for oil extraction and the residue obtained is called mustard cake. Mustard oil accounts for 18% of Indian edible oil consumption and has characteristic pungent taste. The proportionate increase in per-capita edible oil consumption and awareness on health benefits of mustard oil has lead to increase in demand of later. These conditions are bound to produce increased by-product, mustard cake which is a rich source of protein. The need of utilizing the cake has driven research in various foci. This review attempts to present a compendium on this aspect highlighting mustard seed production, its products and by-product utility.

Production of mustard seed

Table 1 showcases the worldwide increasing trend in mustard production leading to 67.6 million tonne (Mt) in 2013/14 from the current level of 64.1 Mt and 61.7 Mt in 2012/13 and 2011/12, [1,3,4]. As estimated in 2009/10, India held rd rank after China and USA sharing 10.7% (Table 2) of world production[3]. Within

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India, mustard is an important edible oilseed crop (nd after soybean) and accounts for 26% (Tables 3 & 4) of total oilseeds produced assessed for the year 2005 – 2010%[5,6,7].

Mustard oil

Mustard oil is a primary product of mustard seed. The oil can be extracted using cold pressing (*kachi ghani*) or mechanical expelling, and solvent extraction. For the later, seeds are cracked, cooked and mechanically pressed to expel oil, followed by solvent extraction form residual meal (defined below). The cooking step liberates oil from the complex lipoprotein molecules by destroying the bonds between lipid and protein, and also inactivates enzyme resulting in high yield and less pungent oil. The characteristic pungent odor of the oil is due to action of myrosinase on glucosinolate leading to formation of allyl isothiocyanates. Moreover, the enzyme produce different intensity of pungency depending on type of glusoside, for instance, hydrolysis of sinalbin (white mustard) produces less irritating oil than that of sinigrin (black mustard). In case of cold pressing, the absence of heat leads to myrosinase activity and yields dark yellow pungent mustard oil. Apart from its signature odour (allyl isothiocyanate), the oil is perceived to have better retention of micronutrients and antioxidants.

Fatty acid composition determines the quality and stability of oil. Mustard oil is considered among the healthy edible oil with fatty acids composition including 4.51 ± 3.83% of palmitic acid, 2.78 ± 0.59% of stearic acid, 38.21 ± 2.88% of oleic acid, 25.31 ± 5.74% of linoleic acid, 11.30 ± 6.09% of linolenic acid, 10.86 ± 3.29% of archidonic acid and 11.35 ± 13.83% of erucic acid[8]. Linolenic (Omega-3) and linoleic (Omega-6) are essential fatty acids. The composition revealed that mustard oil is having low amount of saturated fatty acid and equal proportion of monounsaturated fatty acid (MUFA) and polyunsaturated fatty acid (PUFA). Almost equal proportion of MUFA and PUFA (Table 5) relates to the health beneficial property of the oil as it raises the good HDL cholesterol ratio and omega-3 fatty acid reduces the risk of chemically induced cancer. These findings have changed the consumers' perception regarding mustard oil consumption and have increased its demand at domestic end. Mustard oil also contains natural antioxidant, phytosterols, tocopherol, Vitamin K and some polyphenols which has anti-bacterial, anti-fungal and anti-carcinogenic properties. However, presence of ant-nutritional factor, erucic acid, is also a concern. Erucic acid is long monounsaturated chain fatty acid and its high intake (> permissible 2% of total fatty acid content regulated by FSSA, India and CODEX) can cause severe health problems related to heart. The *in-vivo* studies have revealed the deposition of fat on heart tissue and eventually myocardial lesions in experimental animals fed on erucic acid diet. Although reports of such experiments on human subjects are lacking, results from animal trials have discouraged its large scale use in human diets. Chowdhury *et al*.[8] categorized mustard oil as low erucic variety (up to 5% of the acid) and high erucic variety (>14% of the acid). On the contrary, erucic acid at low levels (about 1% in beef fat) reduces the accumulation of very long chain saturated fatty acids (such as C26:0) responsible for demyelination in brain[9].

Table 3—Total Oilseeds and Rapeseed/Mustard production in India during 2006-2012

Year	Oilseed (Mt)[5]	Rapeseed/Mustard (Mt)[6]
2011-12	29.80	6.80
2010-11	32.48	8.18
2009-10	24.88	6.61
2008-09	27.72	7.20
2007-08	29.76	5.83
2006-07	24.29	7.44

Table 1—Mustard production trend in world[3]

Year	2003-04	2004-05	2005-06	2006-07	2007-08	2008-09	2009-10
Area (m.ha)	25.53	26.79	27.32	26.79	28.08	31.09	30.74
Production (Mt)	39.42	46.14	48.57	46.27	48.29	57.84	59.93
Yield (kg/ha)	1540	1720	1780	1730	1720	1860	1950

Table 2—Contribution of different countries in Mustard production 2009-10[3]

Countries	USA	China	India	Germany	France	Australia	UK	Others
Share (%)	20.8	22.9	10.7	10.5	9.3	3.2	3.3	19.3

Table 4—Contribution of different oilseed crop in total production under oilseeds, Average (2005 -10), India[7].

Oilseed	Soybean	Mustard	Groundnut	Linseed	Sunflower	Castor	Safflower	Sesame
Share (%)	35.9	26.0	25.6	0.6	4.6	3.7	0.8	2.4

Table 5—Percentage of saturated (SFA), monounsaturated (MUFA), polyunsaturated (PUFA) and total unsaturated (MUFA+ PUFA) fatty acid in mustard oil[8]

	SFA	MUFA	PUFA	MUFA + PUFA
Mustard oil	15.94±2.58	49.57±8.56	36.62±6.42	86.18±3.07

Mustard oil composition attributes heat stability, high smoke point (204°C) and oxidative stability. Thus, mustard oil is suitable for all types of cooking including frying, but should be used along with other cooking oils to reduce the erucic acid content.

Mustard cake

Mustard cake, about 60% of the seed, is generated as by-product during extraction of the oil. India holds third position in world for production of mustard cake (5.7 Mt every year)[10]. Recently, India has started exporting mustard cake to countries like Japan, Italy, Thailand and few Middle East countries but only in small amount of 0.4 Mt, and almost 2.7 Mt of meal is wasted[11].

Composition

The composition of mustard cake varies with the variety, growing conditions and processing methods. The crude protein content varies from 33 – 40% of which 80 – 83% is true protein with appreciable proportion of albumin, glutelin[12] and globulin. The protein is rich in lysine and sulphur containing amino acid which are limiting in cereal protein, making it excellent complementary to cereals in completing biological value of protein. Moreover, the composition of amino acids is well balanced for application as protein supplement for human nutrition. Some properties of the mustard protein have been reported to be comparable with casein and even better than other plant proteins including that of soybean, pea, and wheat[13]. In addition, the *in-vivo* studies on rat revealed that the protein efficiency ratio of mustard (2.64) is higher than soybean (2.19). Mustard protein consists of two fractions: a high molecular weight fraction (11S/12S), cruciferin, constituting about 25% and a low molecular weight protein fraction (2S), napin, constituting about 70% of the total proteins. Wu & Muir[14] explained the cruciferin structure which consisted of more than 10 polypeptides and non-covalent interactions. These interactions are more important than disulphide bonds in stabilizing the structural conformation. On the other hand, napin consisted of 2 polypeptides and stabilized primarily by disulphide bonds. However, low molecular weight protein fraction, napin like Bra j 1, Bra n 1 and Sin a 1 of some variety namely ***B. juncea***, ***B. napus*** and ***S. alba***, respectively, exhibit allergenicity due to stable structure arising from high disulphide linkages[15].

Mustard cake contains 21% carbohydrate, 21% lipid, 8.5% crude fibre, and 8% ash[16]. Carbohydrate constitutes mainly soluble sugars (≈10%), cellulose (4 – 5%), pectins (4 – 5%), hemicellulose (3%) and starches (< 1%). It is also a good source of bioactive components including phenolics (77 – 81 mg/kg), glucosinolates and phytates; phenolics can be present in free or esterified (upto 80%[17]) forms or it can be conjugated with other insoluble compounds. In white mustard, p-hydroxybenzoic acid is the major phenolic present, followed by sinapic acid, which together represent 36% of the total phenolics. Phenolics impart bitter taste and dark colour to the protein and its products. Residual glucosinolate that has not been acted upon by enzyme 'myrosinase' and thereafter extracted in oil remain in the cake. Glucosinolates in mustard cake may go up to 200 mg/gm, whereas the permissible amount is up to 30 mg/gm for international standards. It is worth mentioning that in animal system, glucosinolates undergo hydrolysis of thioglucosidic bond yielding glucose and unstable aglucone which are toxic to target tissue including liver, kidney and thyroid functioning. However, presently, glucosinolates have come in lime light as anti-cancerous substances through Hydroxy Radical Scavenging Activity and or by inducing Apoptosis of cancerous cells. Type of glucosinolates is species specific. For example, yellow mustard (*Sinapis alba*) contains hydroxybenzyl glucosinolate and oriental mustard (*Brassica juncea*) contains sinigrin which is belong to benzyl and allyl class of glucosinolates. Phytates are strong chelating agents which bind to several metal ions in the body fluid thus, rendering them unavailable for cellular uptake.

Utilization

Several attempts have been made for utilization of mustard cake. Few of them are discussed below with

special emphasis on protein for human consumption depicting the upcoming potential for product development.

Animal livestock

Mustard cake is used as a feed for cattle, poultry and aquatic animal; however, the information on percentage of feed to be given is scanty. In case of sheep and goats, mustard cake do not affect feed intake, feed efficiency, nitrogen balance, mineral balance and growth performance of growing lambs[18]. According to Tripathy *et al*.[19], partial replacement of soybean meal by mustard cake amounting 80 gm/kg diet had no adverse effects on growth and heath of growing rabbits. The average metabolizable energy of mustard cake was reported to be 9·62 and 8·75 MJ/kg for meat-type and egg-type chicks. While investigating the effect of feeding mustard cake in layer type chicken, egg weight, shell thickness and yolk quality were affected adversely only upon 15% mustard cake inclusion, while albumen quality started showing deterioration at 10% inclusion itself[20]. Moreover, the cake is less palatable to ruminant due to its bitter taste resulting from the presence of tannin therein. According to Brown & Morra[21], mammalian systems metabolize and eliminate these products rapidly so they do not pose any problem to either humans or other mammals; consequently, mustard cake can be used as a feed supplement. Interestingly, data on carry-over of glucosinolates into milk, meat and eggs are limited. The carry-over into milk is estimated to remain below 0.1% of the total concentration of glucosinolates in the diet. However, glucosinolates in mustard cake is toxic (due to development of toxic substances on enzymatic hydrolysis) especially to non-ruminants.

The effects of feeding mustard cake to fish have not been extensively investigated but a few reports are available. Abnormalities in the liver, congestion and cytoplasmolysis were found in Indian major carp (*Labio rohita*) after feeding 20 and 30% mustard cake, an inclusion rate up to 10% was found suitable[22]. The tolerance limit of dietary glucosinolate (allylisothiocyanate) in carp is less than 0.4 mg/gm of diet. Hossain *et al*.[23] observed that the growth of silver carp (*Hypophthalmichthys molitrix*) and bata (*Cirrhinus reba*) is faster by using feed containing rice bran and mustard cake.

Fertilizer

Organic fertilizers can play vital role in sustaining soil quality and crop production. Mustard cake is rich in nitrogen (4.8%), phosphorous (2% as P_2O_5) and potassium (1.3% as K_2O) which are essential requirement to maintain fertility of soil and proper growth of plant[24]. They opined, the oil cakes are slow in mineralizing but gaseous nitrogen losses are much smaller in comparison to NO_3 fertilizers. While studying the effect of organic fertilizers including mustard cake, cow dung and poultry manure with inorganic ones, Ullah *et al*.[25] reported that although organic fertilizers maintained good health of soil but they were slow to release adequate nutrients timely; thus, recommended combined application. Ibrahim & Mumtaz[26] in their research observed that fungal inoculation with mustard cake increased the plant yield.

Natural weedicide

Inorganic chemical weedicides can solve the problem of weeds, but pose health problems to human and other mammals. To combat this problem, use of organic agricultural by-product is being promoted. In addition to isothiocyanates, the enzymatic action on glucosinolates also produces thiocyanate, nitriles, and other compounds when added to moist soil[27]. Several breakdown products of glucosinolate in mustard cake have pesticidal properties, including weed suppression capacity[28]. These researchers have documented the efficacy of mustard cake as biopesticide to control weeds.

Fermentation medium

Mustard cake has been studied in solid state fermentation media for enzyme, mushroom and lactic acid production. For enzymes the microbial cultures were *Aspergillus ficuum* NRRL 3135[29] for phytase, and *Bacillus subtilis* OCS 02[30] and *Yarrowia lipolytica* NCIM 3589[31] for lipase. Mustard cake together with Mg^{2+} ions supplemented in whey medium improved the lactic acid production by immobilized *Lactobacillus casei*[32]. According to Bano *et al*.[33], mustard cake as nitrogen source added to rice straw substrate increased the yield of oyster mushroom by 50–100%.

Biodegradable films

Use of low-value agricultural by-product such as mustard cake as a viable source of biopolymer was evaluated by Hendrix *et al.*[34] and Kim *et al.*[35]. Both of them used high pressure processing for homogenizing defatted mustard cake in the film forming solution. The ranges for the properties of water vapour permeability, water solubility, tensile strength, and

elongation of the films were 3.4–5.0 gm.mm/m^2.kPa.h, 30.3–34.4%, 1.3–5.5 MPa, and 0.9–18.1%, respectively[34]. Kim *et al*.[35] reported the values as 1.5–2.4 gm.mm/m^2.kPa.h, 1.1–1.6%, 5.1–8.8MPa, and 2.9–5.0%, in that order. Kim *et al*.[35] also observed that mustard cake as a component in coating effectively reduced lipid oxidation in smoked salmon during storage.

Natural antioxidants

After extraction of oil, polar antioxidants like phenolic acids, lignans or flavonoids remain in cake, and subsequently can be extracted with polar organic solvents to obtain in concentrated form to act as primary antioxidants, synergists and chelators. They possess moderate antioxidant activity; nevertheless, they could be applied with success in special foods. Shukla *et al*.[36] and Shahidi & Wanasundara[37] reported the antioxidant potential of methanolic extract of low-pungency mustard flour in meat product and correlated the effect with total phenolic content. In further studies, Shahidi *et al*.[38] reported total phenolic content of extract as 919 mg sinapic acid equivalent/g extract and the antioxidant potential was comparable with butylated hydroxyl anisole.

Protein source for human consumption

Mustard protein is comparable with soy protein in terms of FAO pattern of essential amino acid requirements for human adults. To minimize prevailing malnutrition in highly populated developing countries, mustard cake could be a promising alternative. Sustainable availability of cake supports this anticipation. The use of *Brassica* seeds as a protein source, however, is limited by the presence of undesirable toxic and anti-nutritional non-protein components, including glucosinolates and phytates, as mentioned above. The concentration of these undesirable components must be substantially reduced in isolated protein in regards with suitable human consumption.

Several attempts have been under taken to extract protein from mustard cake through initial dissolution of the protein fraction in water or alkaline/salt solution, followed by salting out or isolectric precipitation techniques. Within the concentration range of 0.01–0.1M NaC1/NaH_2PO_4 at *p*H 5.5–6.5 used for dissolution of canola protein, Ismond & Welsh[39] achieved optimal removal of antinutritoinal factor at 0.1 M of either the salt at *p*H 5.5. Klockeman *et al.*[12] obtained 87.5% protein by dissolving defatted canola cake in alkaline medium followed by precipitation at *p*H 3.5. Although the isolated canola protein had poor solubility at *p*H 2 to 10, it was nutritionally balanced for use in products for 10−12 yrs-olds and adults. It is worth mentioning that hydrolysate of canola protein isolate reduces the hypertension and congestive heart failure by inhibiting angiotensin-converting-enzyme[14]. Sadeghi *et al*.[40] prepared 95% pure protein isolate with reduced anti nutritional factors like isothiocyanates and oxazolidine thione (hydrolysed product of glucosinolates), phenolics and phytates, using mustard cake from dehulled seed. Also, the *in vitro* digestibility of the isolate was 92.4% as compared to 80.6% of the cake. Diosady *et al*.[41] provides a method for processing defatted oil seeds to form bland and nontoxic protein isolates. The researchers recommended food drink products containing the isolated protein. Mejia[42] reported 'Generally Recognised as Safe (GRAS)' status for canola protein isolates like cruciferin-rich (Puratein) representing globulin fraction and napin-rich (Supertein) as albumin fraction. The basic steps in production included solubilization in NaCl solution, followed by several clarifications and filtrations, and then concentration using ultra-filtration with suitable membranes. It was stated that foods containing the canola protein isolates (puratein and/or cruciferin) could be targeted to the general population.

Conclusion

Studies pointing towards health benefit of mustard oil have inclined consumers towards the use of mustard oil in domestic cooking. This can be feasible supported by continuous production of mustard seed, concomitantly increasing the production of mustard cake. A versatile approach has been taken for utilization of this proteinaceous resource. An overview of undertaken researches will propel utilization of mustard cake in a fruitful manner with economic feasibility.

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