

**PHYSICOCHEMICAL PROPERTIES AND MINERAL PROFILE
STUDY OF DIFFERENT RAPESEED AND MUSTARD (*Brassica spp.*)
GENOTYPES**

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**PHYSICOCHEMICAL PROPERTIES AND MINERAL PROFILE
STUDY OF DIFFERENT RAPESEED AND MUSTARD (*Brassica spp.*)
GENOTYPES**

BY

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A Thesis

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CERTIFICATE

*This is to certify that the thesis entitled “**PHYSICO-CHEMICAL PROPERTIES AND MINERAL PROFILE STUDY OF DIFFERENT RAPESEED AND MUSTARD (Brassica spp.) GENOTYPES**” submitted to the Department of Biochemistry, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE in BIOCHEMISTRY**, embodies the result of a piece of bona fide research work carried out by **SHARMIN SULTANA**, Registration No. **13-05320** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged.

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DEDICATED TO-

***My Beloved Parents and
Respected Research Supervisor***

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PHYSICOCHEMICAL PROPERTIES AND MINERAL PROFILE STUDY OF DIFFERENT RAPESEED AND MUSTARD (*Brassica* *spp.*) GENOTYPES

ABSTRACT

To evaluate the physicochemical characteristics of five varieties of rapeseed and mustard were taken to study their seed weight, moisture, ash, carbohydrate, protein, fat, total energy of the whole seed and chemical constant and fatty acid composition of oil and finally minerals composition of oilcake. Among the varieties Tori-7 contained the highest thousand grain weight (4.53 ± 0.10 g) and moisture percentage ($4.68 \pm 0.02\%$) whereas Sampad contained the highest amount of oil content ($43.23 \pm 1.06\%$) and gross energy (553.51 ± 5.20 kcal/g) but the lowest amount of ash ($11.05 \pm 0.03\%$). Significantly the highest amount of protein ($26.80 \pm 0.07\%$) and carbohydrate ($22.29 \pm 0.81\%$) was observed in BARI Sarisha-15. In case of chemical constant the highest saponification value (168.25 ± 0.15) and iodine value (106.27 ± 1.03) and the lowest acid value (1.20 ± 0.02) were observed in BARI Sarisha-15. Significantly the highest amount of linoleic acid was observed in Sampad where the highest amount of palmitic acid ($3.23 \pm 0.05\%$) and erucic acid ($51.43 \pm 0.02\%$) were found in BARI Sarisha-17 and the highest amount of behenic acid ($1.31 \pm 0.03\%$), total saturated fatty acid (TSFA) ($11.30 \pm 0.19\%$) was found in BARI Sarisha-14. The concentration of stearic acid varied from $1.20 \pm 0.02\%$ to $1.56 \pm 0.03\%$; whereas arachidic acid contents ranged from $4.04 \pm 0.07\%$ to $5.65 \pm 0.09\%$. On the other hand, BARI Sarisha-15 contained the highest amount of oleic acid ($18.44 \pm 0.03\%$) and total unsaturated fatty acid (TUFA) ($89.11 \pm 0.03\%$). BARI Sarisha-14 contained the highest amount of calcium ($0.44 \pm 0.01\%$), potassium ($0.90 \pm 0.01\%$), copper (13.00 ± 0.86 ppm), and manganese (31.25 ± 3.25 ppm). BARI Sarisha-17 contained the highest amounts of magnesium ($0.45 \pm 0.01\%$), phosphorous ($1.09 \pm 0.03\%$) and Tori-7 contained the highest amounts of sulfur ($0.43 \pm 0.01\%$), iron (248.25 ± 3.25 ppm), and zinc (48.37 ± 1.8 ppm). However, this study suggested that although BARI Sarisha-14 contained notable mineral composition but in terms of qualitative aspect, BARI Sarisha-15 performed better over Sampad and Tori-7.

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LIST OF ABBREVIATIONS AND ACRONYMS

Full Word Abbreviation

And others	<i>et al.</i>
Coefficient of variation	CV
Poly Unsaturated Fatty Acid	PUFA
Days after flowering	DAF
World Health Organization	WHO
Non-Protein Nitrogen	NPN
Hours	h
Cultivars	cv.
Fatty acid	FA
Liter	L
Titer Value	TV
Saponification Value	SV
Iodine value	IV
Acid value	AV
Normality	N
Analysis of variance	ANOVA
Association of Official Analytical Chemist	AOAC
Bangladesh Agriculture Research Institute	BARI
Degree Celsius (Centigrade)	°C
Least significant difference	LSD
Gram	g
Miligram	mg
Milliliter	ml
Parts per Million	ppm
Food and Agriculture Organization	FAO
Microgram	µg
Microliter	µl

CHAPTER I

INTRODUCTION

Bangladesh has cultivated many oil seeds crops like mustard, sesame, groundnut, linseed, niger, safflower, sunflower and soybean. Among these, mustard is counted as the major oil crop belongs to the genus *Brassica* of the family *Brassicaceae*. It is one of the main cultivable edible oil seed crops of Bangladesh. It occupies first position of the list in respect of area and production among the oilseed crops grown in this country (BBS, 2019). In the year of 2018-2019 it covered 2.7 lakhs hectare (ha) land and the production was 3.11 lakhs metric ton (Mt), whereas the total oilseed production was 5.19 lakhs Mt and total area covered by oilseed crops was 4.03 lakhs ha (BBS, 2019).

It is the foremost important popular oil crop which is grown in Rabi season in Bangladesh. Only a couple of decades ago, in Bangladesh mustard oil was the exclusive vegetable oil, medicinal ingredient and supplied fat in our daily diet. However, the yield of this crop in Bangladesh is far lower than compared to other countries. The typical yield of rapeseed-mustard in Bangladesh is extremely low (1.15 t ha⁻¹) that was 50% of the globe average (BBS, 2019).

Recently, BARI has released some mustard varieties, which have high yield potential under the circumstance the farmers have gotten lower yield of mustard with their local varieties with poor crop management practices. The tender leaves of these cultivars serve as vegetable, while the seeds as a source of lubrication and vegetable oil. The residue left after oil extraction (i.e., feed or meal) being rich in protein (Durrani and Khalil, 1990) are often used as livestock feed. Oil is one

among the required nutrients for the human body, which is supplied by animal and a couple of plant sources (Nabipour *et al.*, 2007). Herbal oils are the most sources of fats and fat-soluble vitamins, which have a considerable role within the human diet (Stuchlik and Zac, 2002). After cereals, oil seeds are the second food sources throughout the globe, whose oil is rich of fatty acid types (Siavash *et al.*, 2005). It produces 9 kcal energy from 1 gm of oil per unit as compared with other diets (carbohydrate and Protein). During a diet for human health 20-25% of calories should come from fats and oils. The mustard oil isn't used just for cooking purpose but is also used for hair dressing, body massaging and for various sorts of pickles preparation. It is also used as medicinal purposes. In traditional medicines, it is used to relieve the pain associated with arthritis, muscle sprains and strains. Seed paste applied on wounds whereas paste of leaf said to heal cattle wounds (Sahedi, 2015). Rapeseed mustard oil quality is determined by the constituent fatty acids including palmitic, stearic, oleic, linoleic, linolenic, eicosenoic and erucic acids; and is highly affected by the variety type (Nasr *et al.*, 2006; Javidfar *et al.*, 2007). One of the main breeding objectives regarding rapeseed besides the oil quantity is to increase its oil quality (Azizi *et al.*, 1999).

Rapeseed and mustard are common names used for different species of the family Cruciferae (*Brassicaceae*). Rapeseed includes *Brassica compestris* and *B. napus* and mustard specifically refers to *Brassica juncea* and *Eruca sativa*. There are considerable differences in agronomic characteristics, yield, and fatty acid (FA) composition of seed oil between species and between varieties (Bauer *et al.*, 2015). From the nutritional perspective, linoleic acid is the most important unsaturated fatty acid due to its ability to reduce the risk of cardiovascular diseases (Farvid *et al.*, 2014). Linoleic and linolenic acids are essential fatty acids not

synthesized by our body; it has to be supplied through meals. In addition, oleic acid is among unsaturated fatty acids whose antioxidant effects have been proved (Berry and Rivlin, 1997). Erucic acid, although, anti-nutritional and should be <2% in the edible oil, higher erucic acid is of considerable industrial importance. The yield of mustard can be augmented by adopting modern and recommended technologies along with the use of high yielding varieties. Fertilizer is the depending source of nutrient that can be used to boost up growth and yield of rapeseed (Sinha *et al.*, 2003).

In addition there are some cultivars viz. BARI Sarisha-14, BARI Sarisha-15, BARI Sarisha-17 along with old cultivar sampad and Tori-7, are now cultivating in different regions of our country. The nutritional quality of all these released cultivar is not yet analyzed. If the nutritional quality of rapeseed and mustard is known, its consumption as well as its multipurpose uses will be increased which play a vital role in improving the nutritional status of the people of our country. Now a burning question arises about the physico-chemical quality of BARI cultivars rapeseed and mustard. Therefore the present study was undertaken with a view to determine physico-chemical properties of some popular selected released cultivars to ensure their nutritional status.

Objectives

1. To compare the physico-chemical parameters and nutritional quality of different varieties of rapeseed and mustard.
2. To compare the gross energy obtain from different varieties of rapeseed and mustard varieties.
3. To determine fatty acid composition of different genotype of mustard and rapeseed.
4. To determine the mineral composition of different varieties of rapessed and mustard oilcake.

CHAPTER II

REVIEW OF LITERATURES

Vegetable oils and fats constitute an important component of human diet, ranking third after cereals and animal products. *Brassica* is considered to be the most important source of vegetable and protein rich meat worldwide (Arif, *et al.*, 2012). Mustard and rapeseed is important oil crop in human and animal nutrition because of its high level of fats/oil, fatty acid, protein and other nutritionally valuable substances. As a high energy component of food, edible oil is important for meeting the caloric requirement. Physical properties, chemical analysis, estimation of minerals and total energy of different types of rapeseed and mustard observed by other researcher are described in this section.

Grain Weight

Thousand seed weight is a very important character of rapeseed and mustard, where highest consideration is one of the seed yields. A good number of literatures are available on the variability of this trait.

Hossain *et al.* (2015) found that seed weight varied with their size and shape. They found that thousand grain weights were determined at 13% moisture level. The highest thousand grain weight was found in BARI Sarisha-12 (4.38 g), this was significantly higher than all others released variety and the lowest thousand grain weight was found in BARI Sarisha-9 (3.06 g). Statistically similar results were shown by BARI Sarisha-9 (3.56 g), BARI Sarisha-12 (3.75 g) and Sarisha-6 (3.75 g).

According to Damian (2014), 1000 seed weight of the mustard seeds increased from 4.30 g to 5.10 g, as the moisture content increased from 7.00% to 15.99%.

A study was carried out by Banga *et al.* (2013) to observe the seed yield in different genotypes. According to the study 1000-seed weight was the maximum in RL-1359 (5.15 g).

An experiment was carried out by Chowdhury *et al.* (2010) on evaluation of rapeseed and mustard varieties. The research work indicated that the highest 1000 grain weight was found in BARI Sarisha-9 (4.90 g) and lowest was shown by BARI Sarisha-12 (2.50 g).

Some characteristics were evaluated by Siddiqui and firoz (2004), namely *Brassica carinata* cultivars (IGC-01 and Pusa Gaurav), *B. juncea* cultivars (Jagannath, Kranti, Rohini, and TERI (OE) M21-Swarna) and *B. napus* cultivars (Hyola PAC-401) in case of grain weight. The highest grain weight observed was from TERI (OE) M21-Swarna (3.95 g) among other cultivars.

According to Mondal and Wahhab (2001) weight of 1000 seeds varied from variety to variety and species to species. They found that thousand seed weight of 2.50-2.65 g in case of improved Tori-7 (*B. campestris*) and 1.50-2.80 g in Rai-5 (*B. napus*).

Karim *et al.* (2000) performed a research with released variety and advanced line cultivars that showed significant difference in weight of thousand seeds. They found higher weight of 1000 seed in J-3023 (3.43 g), J-3018 (3.42 g) and J-4008 (3.50 g).

An experiment was conducted by Kaul and Das (1986) to evaluate thousand grain weights of *Brassica campestris* seeds. Result indicates that grain weight of *Brassica* seeds ranges from 2.60 g - 3.10 g for brown-seeded form and 2.50 - 4.50 g for yellow-seeded form, respectively.

Moisture

In storage condition, the permeable moisture level of different oil seeds ranged from 10-12% (Sahedi, 2015). Moisture content is important factor than other nutrients as they vary with it. It is also important for insect infestation and disease.

According to Hossain *et al.* (2015) moisture content of different released and line cultivars of mustard and rapeseed was ranged from 4.00% to 5.20%. They found the highest moisture content (5.20%) was observed from BARI Sarisha-12; while the lowest moisture content (4%) was found in BARI Sarisha-9.

Sarker *et al.* (2015) conducted an experiment in the matter of moisture content of black and yellow mustard cakes. Moisture content of black and yellow mustard cake were about $9.20 \pm 0.5\%$ and $9.73 \pm 0.6\%$ respectively.

Barrozo *et al.* (2008) carried out an experiment on mustard storage moisture. They found out that for long-term storage mustard seed should be kept at less than 9% moisture. If the mustard needs to be dried for safe storage, the drying air temperature and seed temperature should not exceed 65°C and 45°C, respectively.

Huda (2001) reported about the moisture content of rapeseed mustard varieties and advanced lines which indicates that seeds having moisture content from 8-10% stored well and safe moisture content for sealed storage is 4-8%.

A research was carried out by BARI (1987-88) to determine the moisture content of rapeseed-mustard varieties and advanced lines. According to the research, moisture content of different released and line cultivars are about 7.06%, 8.36%, 7.66%, 7.98%, 7.41%, 8.38%, 7.81%, and 7.82% for SS-75, Krisna, PT-303, Varuna, TS-72, Kranti, S-5 and Tori-7 respectively.

Oil

Fats and oil are important to maintain body temperature. It provides 9 kcal energy from each gram of oil.

An experiment on composition and physical properties of different oil seeds conducted by Hossain *et al.* (2015). They observed the oil content of different varieties of mustard and rapeseed varied from 38.75% to 42.25%.

Arif *et al.* (2012) carried out an experiment to find out the potential nutrients rich of oil seed varieties. They reported that the highest amount of crude oil was observed in *B. juncea* and *B. napus* i.e. 45.67% and 43.87%, respectively.

The composition and physical properties of different oil seeds were examined by Gadei *et al.* (2012). They observed that oil seeds have high oil content, ranged from 28% – 32% with relatively high protein content (28% – 36%). Moser *et al.* (2009) also found the similar result.

Some features of *B. carinata* cultivars (IGC-01 and Pusa Gaurav), *B. juncea* cultivars (Jagannath, Kranti, Rohini and TERI (OE) M 21-swarna) and *B. napus* cv. (Hyola PAC-401) were introduced by Siddiqui and firoz (2004). They reported that cultivars Pusa Gaurav gave the highest oil content (40%) among these.

According to Sengupta and Das (2003) oil content of mustard seeds ranges from 40% - 41.5% and kernel 45% - 47.1%.

An experiment was conducted by Bhowmik (2003) on some released rapeseed and mustard varieties and advanced line cultivars and their oil content. Results revealed that oil content of brown sarson, yellow sarson and toria ranges from 44-45%, 42-46% and 42.44%, respectively.

According to Mandal *et al.* (2002) one of the most remarkable characteristics of *Brassica* species is the high oil content in the seeds,

ranging in wild types from about 21.50% - 46.70%; similar result were found by Kumar and Tsunoda (1980).

Tyagi and Singhal (2001) analyzed proximate composition of oil and crude protein. The highest amount of oil was found about 38.96% from Toria kranti and crude protein was about 46.23% from Toria shgamgarh.

An experiment was conducted by Niraj *et al.* (2000) to compare 21 genotypes of Indian mustard. Results showed that considerable variation in oil content was ranged from 36.49% to 40.19% at per with vardan (39.14%) and varuna (38.13%).

According to Rathore (2000) oil content of sarson, toria and rai ranges from 43-45%, 30-35% and 31-35%, respectively.

A study was conducted by Novoselov *et al.* (1997) to find out the oil content of different rapeseed oil. Results revealed that oil content of rapeseed ranges from 45-46%.

An experiment was carried out by Mazzoncini *et al.* (1993) on seed oil content of *Brassica carinata* which ranged from 32.50% to 40.60% and higher oil content found in rapeseed that ranged from 40.50% to 47.30%.

BARI (1992-93) determined the oil content of eight brown seeds of *B. campestris* lines. Oil contented of different advanced lines was about 41.77%, 42.40%, 42.43%, 41.37%, 41.75% and 41.88% for BC-2192, BC-2493, BC-2592, BC-2693, BC-2892 and BC-2093 respectively. No significant difference was shown by the results among the advanced lines with respect to its oil content.

BARI (1992-93) reported the oil content of seven yellow seeds of *B. campestris* lines including two check varieties and advanced lines. Oil content was ranged from (41.99% to 42.52%).

Vijay *et al.* (1992) evaluated an experiment in terms of the oil contents in 65 released varieties and advanced line of rapeseed and mustard. The result showed that many released varieties and advanced line cultivars

had high oil content (more than 40%) and particularly RH, RK and DIRA had low oil content, mostly ranged from of 35% - 38%.

An experiment was carried out by BARI (1987-88) on oil content of fourteen *Brassica campestris* genotypes. Result indicates that highest oil content i.e., 42.87% and 42.69% for the lines OTBC-0893 and OTBC-1493 and others advanced lines were not significantly different from the first lines.

Oil cake

Oil cake is the nutritious feed items for cattle and fish. It is also used as a good organic matter.

Hossain *et al.* (2015) carried out an experiment to evaluate the percentage of oil cake of different varieties of mustard and rapeseed where the BARI Sarisha-12 contained significantly highest amount of oil cake (61.25%). The lowest value was found in BARI Sarisha-9 (57.75%).

Chowdhury *et al.* (2010) conducted an experiment to evaluate the oil cake percentage of different released varieties and line cultivars of mustard and rapeseeds. They reported that, BARI Sarisha-12 contained significantly highest amount of oil cake (59.95%), followed by BARI Sarisha-9 (59.47%) and BARI Sarisha-6 (59.25%). The lowest value was found in BARI Sarisha-11 (57.75%), followed by advanced lines Din-2 (58.14%).

Appelqvist and Ohlson (1992) stated that typically rapeseed oil (Kind of mustard seed) contain 58% cake.

Ash

Ash is the residue that remains after the complete combustion of the organic compound of a food product.

Hossain *et al.* (2015) reported that, ash content of different released varieties of mustard and rapeseed were variable and ranged from 9.60% to 12.50%. BARI Sarisha-12 contained the highest amount (9.60%) of ash.

According to Sarker *et al.* (2015) ash content of black and yellow mustard cakes was about $7.10 \pm 0.3\%$ and $5.90 \pm 0.3\%$, respectively.

According to Abul-Fadl *et al.* (2011) both yellow and brown mustard varieties containing an adequate percentage of ash, dietary fiber and total carbohydrates which were found to be as 4.08, 5.87 and 16.60% in yellow variety and 3.88, 6.34 and 16.49% in brown variety, respectively.

An experiment was conducted by Sosulski and Bakal (1991) to evaluate ash content of different rapeseed-mustard varieties and advanced lines. According to the experiment ash content of *B. campestris* and *B. rapa* ranges from 7.00 - 8.40% and 7.30 - 8.00%, respectively.

Nehrins and fettu (1990) reported that the ash content of Canadian wild mustard and rapeseed were found to be 4.38% and 7.30%, respectively.

A study was carried out by Kaul and Das (1986) to evaluate partial analysis of India cruciferous oilseeds. Result indicates that the ash content of *Brassica campestris* variety dichotoma BS-2 and Toria T-36 were 3.74% and 3.22%, respectively.

Protein

Around 28-32% of the mustard seed total weight is composed of proteins. Proteins are polymers of amino acids. Proteins form the structural elements of cells and tissue in the human body and are considered as the basis of life, but they are also essential components in different food systems.

Sarker *et al.* (2015) conducted an experiment on the protein contents in black and yellow mustard cakes. They reported that protein content of that two varieties were about 38.17% and 28.80% respectively.

According to Marnoch and Diosady (2006) the Crude protein content of mustard cakes obtained were 38.17% and 28.80% which were lower (45.0% and 34.0%) than those reported by many other authors (Prapakornwiriya and Diosady, 2004). However, Chowdhury *et al.* (2010)

and Kumar *et al.* (2002) reported comparatively equal amount of crude protein in mustard cakes.

According to Al-Jasass *et al.* (2012) mustard seeds contained about 28-32% protein.

An experiment was carried out by Abul-Fadl *et al.* (2011) to evaluate the protein content of mustard seeds of yellow and brown varieties. According to the experiment yellow mustard seed had a higher content of protein (36.73%) than in brown mustard seed (32.48%).

Sengupta and Das (2003) reported that protein content of rapeseeds was ranged from 44.2% - 44.7%.

Mirza *et al.* (1998) carried out an experiment to evaluate the protein content of rapeseed oil where significant differences were found among protein contents which were negatively correlated with oil content.

A research was performed by Sosulski and Bakal (1991) on proximate composition of different released and line cultivars of rapeseed and turnip rape. The results showed that protein content of different released varieties and line cultivars were variable i.e., *B. campestris* content 40.8% and *B. napus* content 45.5% protein in seed oil.

Carbohydrate

Rapeseed and mustard contain relatively lower amount carbohydrate.

A research was conducted by Hossain *et al.* (2015) to evaluate the percentage of carbohydrate of different varieties of mustard and rapeseeds and they found that, carbohydrate contained in the seeds ranged between 12.75% - 17.02%.

According to Bachheti *et al.* (2012) mustard seeds contained about 23.8% carbohydrate.

Gopalan *et al.* (1981) examined the nutritive values of Indian foods found out that dry mustard seeds contained 20% - 23% carbohydrate.

Chemical properties of Oil

The chemical characteristics of oil determine the quality and stability of oil.

Khan *et al.* (2013) carried out an experiment to analyze the chemical properties of the mustard oil showed that the saponification value, iodine number, acid value is >170, >100 and <0.5 respectively.

Chemical constant of mustard and rapeseed oil was determined by Chowdhury *et al.* (2010). They observed that, saponification values of different released variety and advanced lines were ranges from 154-168.3; iodine values were ranges from 93.45-110.2 and acid values were ranges from 1.31-1.61.

A study was conducted by Richet *et al.* (1987) by using fifteen different solvents for the extraction of oil from rapeseeds under identical condition. Acid value, saponification value and iodine value of the extracted oils were also recorded. The values were 2.7 and 7.6 in benzene and methyl ester for acid value, 170.4 and 182.4 in propyl alcohol and toluene for saponification value and 82.1 and 98.5 in butyl oxide and acetone in CCl₄ for iodine value, respectively.

Fatty acid composition

Mustard and rapeseed contain different types of saturated and unsaturated fatty acid. Oleic acid, linoleic acid, linolenic acid, palmitic acid and stearic acid are the most important and essential fatty acids in rapeseed oil.

Gas liquid chromatography (GLC) analysis of oil seeds was conducted by Bachheti *et al.* (2012) which revealed that it contained oleic acid (73.58%), linoleic acid (19.26%), palmitic acid (3.31%), myristic acid (1.18%) and stearic acid (2.68%). Mustard oil possess 60% monounsaturated fatty acids of which 42% Erucic acid and 12% Oleic acid, it had also 21% polyunsaturated of which 6% was the omega-3

alpha-Linolenic acid and 15% omega-6 linoleic acid along with 12% saturated fats were conducted. The concentration of oleic acid (18:1), a beneficial monounsaturated fatty acid, ranges from 3.6-32.2% in rapeseed-mustard oil.

Abul-fadl *et al.* (2011) reported that, erucic acid was predominant fatty acid in yellow (37.89%) and brown mustard seeds oils (23.90%). Both yellow and brown mustard seeds oils contained a little amount (8.45% to 8.94%) of saturated fatty acids as compared to the other edible oils. Oleic acid was the prevalent unsaturated fatty acids, which was ranged between 19.08% to 20.24% of total fatty acid profiles in both yellow and brown mustard seed oils, respectively. Moreover, linoleic acid was the second dominant unsaturated fatty acid recorded about from 12.37% to 21.36% in both yellow and brown mustard seed oils, respectively.

According to Moser *et al.* (2009) mustard oil has a special fatty acid composition contained about 20–28% oleic acid, 10–12% linoleic, 9.0–9.5% linolenic acid, and 30–40% erucic acid, which is indigestible for human and animal organisms.

An experiment was carried out by Nasr *et al.* (2006) on five important fatty acids, i.e. oleic acid, linoleic acid, linolenic acid, stearic acid and palmitic acid were commonly found in ten rapeseed released varieties and advanced line. Oleic acid levels in different rapeseed released varieties and advanced line were 51% to 62%, while there was 18-32% linoleic acid, 2% - 16% linolenic acid, 0.15% - 2.2% stearic acid and 4% - 8% palmitic acid.

Siddiqui and firoz (2004) conducted an experiment on nutritional analysis of different rapeseed varieties such as *B. carinata* cultivars (IGC-01 and Pusa Gaurav), *B. juncea* cultivars (Jagannath, Kranti, Rohini and TERI (OE) M 21-Swarna) and *B. napus* cv. (Hyola PAC-401). They found that

Jagannath, Kranti and Rohini contain highest amount of linolenic acid (22.76%), erucic acid (43.30%) and palmitic acid (5.63%).

An experiment was carried out by Sengupta and Das (2003) on fatty acid composition in seven edible oils. They reported that amount of linoleic acid (18:2) was ranges from 11% - 22%.

Niraj *et al.* (2000) evaluated the 21 genotypes of Indian mustard and found out considerable variation in fatty acid profile. Low amount of erucic acid was observed in many genotypes ranging from 40.12% to 49.7%.

An experiment was conducted by Ullah *et al.* (1997) on twelve released and line cultivars of rapeseed mustard including three species namely *B. campestris*, *B. juncea* and *B. napus*. Significant differences were observed among these released and line cultivars in respect of fatty acid composition. They reported that newly introduced variety Nap-8509 (*B. napus*) contained lowest (36.4%) erucic acid and tradition Tori-7 contained lowest amount of linoleic acid.

An experiment on different released and line cultivars of rapeseed was carried out by Mazzoncini *et al.* (1993). They reported that seed oil contained a high percentage of erucic acid (34% - 35%), linoleic acid (21% - 22%) and linolenic acid (18% - 19%).

According to Sanches and Raquot (1990) lipid content of different rapeseed varieties and line cultivars ranged from 36.8% to 41.35% with significant differences between cultivars. Palmitic acid comprised 50% - 70% of total saturated fatty acid; oleic acid comprised 64.7% to 72.1% of mono-unsaturated fatty acid and linolenic acid 13.0% to 17.5% of total poly unsaturated fatty acid. Traces of erucic acid was found that ranged from non-detectable to 1.08%.

Wahhab and Caesa (1980) introduced some features with low content of erucic acid in rapeseed-mustard varieties and advanced lines. They

identified one seed containing as low as 29.61% of erucic acid. Either oleic or linoleic acid has replaced erucic acid content as a major constituent.

According to Bhowmik (2003) Indian rapeseed and mustard oils are inferior in quality as they contain high amount of erucic acid (28.0-53.0%) and linolenic acids (8.5-22.7%). They also reported seed oil contain nutritionally good fatty acid i.e. linoleic (12.0-21.0%) and oleic acid (10.0-24.0%).

An experiment was evaluated by Appelqvist (1980) to determine the fatty acid composition of mustard released varieties and advanced line cultivars. He reported that fatty acid composition i.e., for palmitic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, eicosenoic acid and erucic acid were about 3.0%, 0.8%, 9.9%, 13.5%, 9.8%, 6.3% and 52.3%, respectively.

Rollet and Raquet (1995) determined the composition of French rape oil (*Brassica sp.*). Result indicates that fatty acid content in rape seed oil was about 51%, 29%, 16%, 1%, 1.5% and 1.5% for erucic acid, oleic acid, linoleic acid, lenolenic acid, palmitic acid and lignoceric acid, respectively.

Mineral

Living organism cannot synthesize minerals in the body unlike other nutrient elements. Animal body require inorganic elements for maintenance of vital processes essential for life. The general functions of minerals are structural component of body organ and tissues, constituents of body fluids and tissues, electrolytes, and catalysts in enzyme and hormone system. The major minerals are calcium, magnesium, phosphorus, potassium and sulphur and trace minerals are iron, manganese, zinc and boron.

Sarker *et al.* (2015) reported that total minerals content of black mustard cake was about 7.10% and yellow mustard cake was about 5.90%.

An experiment was conducted by Bachheti *et al.* (2012) in the matter of physico-chemical properties of some conventional food oils and they found that mineral content of mustard seed oil (*Brassica Compestris*) i.e., 694.3(g/100g), 4.86(g/100 g), 492.1(g/100g), 0.034(g/100g), 0.019 (g/100g), 0.007(g/100 g),8.11(g/100g) and 0.84(g/100g) for P, Zn, Ca ,Mg ,K, Na, Fe and Cu, respectively.

Sengupta and Das (2003) conducted an experiment to determine the minerals content of rapeseed and mustard varieties and advanced lines. According to the report mineral contents of rapeseed in 100 g edible portion was 4.2 g (calcium 490 mg, phosphorus 700 mg and Iron 17.9 mg and other minerals were present in negligible quantity).

An experiment on the mineral content of defatted rapeseed meals was conducted by (Josefson, 1988). They found that mineral content of rapeseed released and line cultivars are as follows: 1.72%, 1.10%, 0.70%, 1.61%, 68 ppm and 18 ppm for sulphur, phosphorus, calcium, potassium, zinc and iron, respectively.

Kaul and Das (1986) examined the minerals content of different oilseed varieties and advanced line cultivars. They found out that 18 different released varieties and advanced line cultivars content variable quantity of minerals i.e., 5.76%, 0.49%, 0.93%, 0.82% and 0.25% for N, P, K, Ca and Mg respectively.

CHAPTER III

MATERIALS AND METHODS

3.1 Materials

Five released varieties of rapeseed and mustard (*Brassica spp.*) namely Sampad, Tori-7, BARI sarisha-17, BARI sarisha-14 and BARI sarisha-15 were selected for the study. The seeds were collected from the Oilseeds Research Centre of BARI, Gazipur. Seed were cleaned, sun dried and stored into plastic container in a cool place until used for the chemical analysis.

3.2 Brief description of varieties

Sampad: This is a composite variety evolved by BAU. Its grain color is yellow and round in shape. The grain is medium in size.

Tori-7: This is a composite variety evolved by BARI. Its grain colour is blackish and round in shape. The grain is large in size.

BARI Sarisha-17: This is a composite variety evolved by BARI. It was developed by hybridization between BARI Sarisha-15 and Sonali Sarisha. Its grain color is yellow and round in shape. The grain is medium in size.

BARI Sarisha-14: This is a composite variety evolved by BARI. Its grain colour is yellow and round in shape. The grain is large in size.

BARI Sarisha-15: This is a composite variety evolved by BARI. Its grain colour is yellow and oval in shape. The grain is small in size.



Sampad



Tori-7



BARI Sarisha-17



BARI Sarisha-14



BARI Sarisha-15

Plate 1. Photograph of rapeseed and mustard seeds.

3.3 Physicochemical Properties

The investigation was conducted at the Biochemistry laboratory of the Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh and the laboratory of Soil Resource Development Institute (SRDI), Khamarbari, Dhaka, Bangladesh. The period from October, 2019 to February, 2020. The degree to which the chemical analysis indicates the nutritive value is dependent on the constituents determined. Some constituents are determined fairly easily and rapidly, while others require much more time and analytical ability. Before undertaking an analysis, the results of which will represent the composition of a crop or a consignment of a food-grain, it is necessary to ensure that the sample taken is randomly selected so as to be truly representative of the original bulk, and is sufficient in amount.

3.4 Determination of 1000 grain weight

The mass was determined by randomly selecting 100 seed samples and weighing in an electronic balance of 0.0001 g sensitivity. The weight was then converted into 1000 seed mass.

3.5 Determination of moisture

Moisture content of sunflower sample was determined by conventional method i.e., drying in an oven at 100^o C for overnight.

Materials

1. Aluminum moisture dish
2. Electrical weighing balance
3. Oven
4. Desiccator

Procedure

Empty aluminum moisture dish was weighted (W1) and 2.5 g sample (dried at normal temperature after harvest) was taken in a moisture dish and weighted (W2). The sample was spread evenly and placed without lid in oven and dried samples overnight at 100⁰ C. The dishes were transferred to desiccators to cool. Aluminum dish was weighed after cooling (W3) (Aziz *et al.*, 2018).

Calculation:

$$\% \text{ Moisture} = \frac{W2 - W3}{W2 - W1} \times 100$$

3.6 Determination of ash

In this method, destruction of organic matter is accomplished by high temperature oxidation. The critical factors are (i) nature of the ashing vessel, (ii) placement in the muffle furnace and (iii) ashing temperature and time. The vessels used for ashing ranges from silica to platinum crucible or dishes. The sample size can vary from 0.5 to 2.0 g, depending on expected concentration of elements to be determined. The temperature used for ashing vary from 475 to 600⁰C or even more and time required for digestion can be from 4 to 12 hours (depending on the sample weight and type of sample) (Paul *et al.*, 2017).

Materials:

1. Silica crucible
2. Balance
3. Muffle furnace
4. Desiccators.

Procedure

First take the weight of silica crucible and take 2.0 g of dried ground sample in the silica crucible. The crucibles are placed in a cool muffle furnace (Protech, Model: PT-1700M). Care should be taken to see that the crucibles are away by at least 2 cm from the walls of the furnace. This is to avoid localized over-heating. Ashing is usually done by heating the furnace at 600⁰C for about 5 hours (temperature and duration of heating need to be standardized as these two factors vary from sample to sample). The crucibles were transferred into the desiccator and cooled them to room temperature and weighted. It was done immediately to prevent moisture absorption. The incineration repeated until constant weight was obtained.

Calculation:

$$\% \text{ Ash} = \frac{\text{Weight of the ash}}{\text{Weight of the sample taken}} \times 100$$

3.7 Chemical Analyses

3.7.1 Oil extraction and cake preparation

Materials:

1. Electrical weighing balance
2. Soxhlet apparatus
3. Menthol heater
4. Petroleum ether (40-60⁰C)

Procedure

The mustard oil was extracted using Soxhlet method (Aziz *et al.*, 2018) with some modifications. First, 50g sample was taken in aluminum dish

and dried for 1 hour at 100°C temperature. Sample flour was prepared by using blender. Then sample was taken into 25x100mm cellulose soxhlet extraction thimble (Whatman 2800-250). The thimble was put into the soxhlet and 250 ml petroleum ether (AR grade) was poured into the soxhlet flask. Then it was connected to round bottom flask followed with condenser and total apparatus was placed on menthol heater (40-60°C temperature) for 16-20 hours. When extraction of oil was completed menthol heater turned off and permitted to cool. Allow the round bottom flask open for 1 days to remove the solvent from the extracted oil. Thimble was removed from the soxhlet and dried the sample in normal air. Finally, rapeseed and mustard oilcake (a byproduct after getting oil) was collected. The yield of rapeseed and mustard oil was calculated by following formula.

$$\text{Oil percentage of seeds} = \frac{\text{Weight of extracted oil}}{\text{Weight of seeds}} \times 100$$



Plate 1. Photograph showing Extraction of rapeseed and mustard oil by soxhlet aparatus.

3.7.2 Estimation of total protein content by Microkjeldhal method

The protein content of food stuff is obtained by estimating the nitrogen content of the material and multiplying the nitrogen value by 6.25 (according to the fact that nitrogen constitutes on average 16% of a protein molecule). This is referred to as crude protein content, since the non-protein nitrogen (NPN) present in the material is not taken in consideration. The estimation of nitrogen is done by Kjeldhal method (AOAC, 2010) which depends upon the fact that organic nitrogen when

digested with sulphuric acid in the presence of catalyst selenium oxide, mercury or copper sulfate is converted into ammonium sulphate. Ammonia liberated by making the solution alkaline is distilled into a known volume of a standard acid which is then back titrated.

The nitrogen present in the sample is converted to ammonium sulphate by digestion at (380°C) with sulphate acid in presence of a catalyst, potassium sulphate and mercuric oxide. Ammonia liberated by distilling the digest with sodium hydroxide solution is absorbed by boric acid and is titrated for quantitative estimation.

Equipments

1. Balance
2. Microkjeldhal (Mkj) digestion set
3. Mkj distillation set.

Reagents

1. Digestion mixture: 100 g of potassium sulphate (K_2SO_4) was thoroughly mixed with 20 g of copper sulfate ($CuSO_4 \cdot 5H_2O$) and 2.5 g selenium dioxide (SeO_2) was added with it.
2. 60% Sodium hydroxide solution: 600 g sodium hydroxide and 50 g sodium thiosulphate were dissolved in distilled water, cooled and made the volume up to 1 liter.
3. Boric acid: 40 g of boric acid was dissolved in water and made up to 1 liter.
4. Double indicator: 200 mg each methyl red and bromocresol green was dissolved separately in 100 ml of 70% ethanol. One part of methyl red and five parts of bromocresol green were mixed before use.

5. Hydrochloric acid (0.02 N HCl): 8.5 ml concentrated hydrochloric acid was added to 5 liter of distilled water. Standardized to 0.02 N acids by titrating it against standard sodium carbonate (0.02 N) solution.

Procedure

A known quantity of the finely mustard cake sample (100 mg) weighted out in an Mkj digestion flask. About 2 g digestion mixture was added with it 2 ml of concentrated sulphuric acid was dispensed into the flask. Then it was digested for about 2 hrs in Mkj digestion set and was cooled the clear digest. The digest was dissolved in minimum amount of distilled water and carefully transferred to an Mkj distillation set. 10 ml of sodium hydroxide solution was added and distilled it. The distillate was collected for 5 min into 5 ml boric acid containing 2 drops of mixed indicator in a 50 ml conical flask, till the color of solution was changed. The distillate was titrated against a standard hydrochloric acid and noted the titer value (TV).

Calculation:

$$N\% = \frac{(14.007) \times (\text{normality of the acid, } 0.02) \times (\text{TV})}{\text{Weight of sample (mg)}} \times 100$$

Where 14.007 is the equivalent weight of nitrogen.

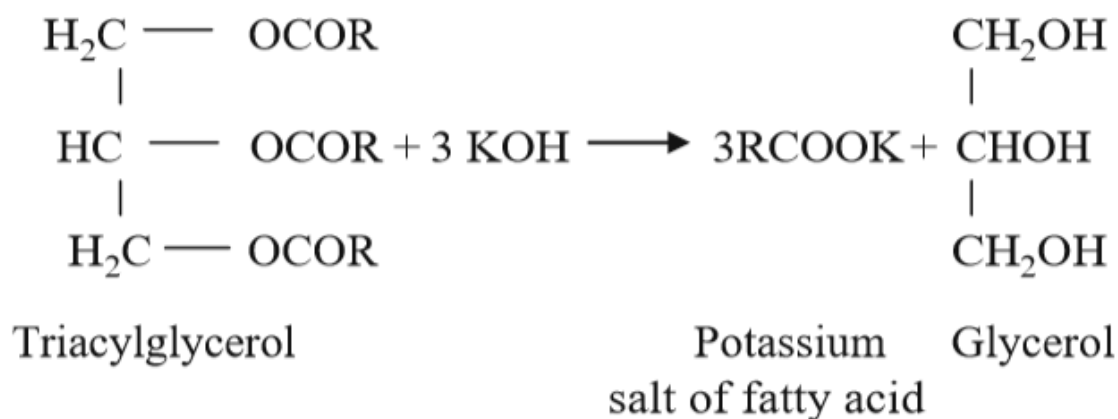
Nitrogen % is converted into protein by multiplying with a factor 6.25 for cereals and pulses.

3.7.3 Chemical constant

3.7.3.1 Determination of Saponification value:

Saponification value is the number of milligrams of KOH required to completely saponify 1 g of oil. The method is based upon the principle

that fat, on treatment with excess of alcoholic KOH is used up. The excess of KOH left unused may then be found by titrating it against a standard acid.



The Saponification equivalent (groups of saponified per equivalent of base) is proportional to the average molecular weight of the fatty acids present in the lipid.

Reagents

1. Hydrochloric acid 0.5N: 20.9ml 37% HCl mixed with water for 500ml solution.
2. Alcoholic solution of Potassium hydroxide. Take 14.02 g potassium hydroxide and dissolve it in very little water. Make up to 500ml by adding alcohol (C₂H₅OH) of specific gravity 0.81.
3. Phenolphthelin solution, 1% in alcohol: 0.2g phenolphthelin mixed with ethanol to make up to 20ml.

Procedure

1-2 g of oil taken in bottom flask. Added 25 ml of 0.5 N alcoholic potash solutions and fit the flask with a cork and a long air condenser. Reflux the contents of the flask for about 30 minutes by heating on boiling water

bath. Cool the flask and add 1 ml of 1% solution of phenolphthalein and titrate the excess of the alkali against standard N/2 acid (A). At the same time and under similar conditions carry out a blank experiment (B) without fat (25 ml of the same alcoholic KOH heated in a similar way is titrated, against .05 N acid) (Sharif *et al.*, 2017).

Calculation:

$$\text{Saponification value} = \frac{56.1 \times (B - A) \times \text{strength of acid}}{\text{Weight of substance in g}}$$

3.7.3.2 Iodine value

Iodine value or Iodine absorption number is the percentage of iodine monochloride (ICI) in terms of iodine absorbed by the oil. Some oils and fats contain many unsaturated fatty acid constituents such as oleic and linoleic acids which take up halogen to form saturated compounds. The extent of this combination however, depends on the degree of unsaturation (Sharif *et al.*, 2017).

Reagents required

1. 0.1N sodium thiosulphate: 12.41g sodium thiosulphate pentahydrate mixed with water to make up to 500 ml.
2. Starch solution (indicator): Starch (1g) was dissolved in 50 ml hot water and diluted to 100 ml with cold water.
3. Hanus solution: Iodine (13g) was dissolved in glacial acetic acid. Bromine (3 ml) was added to it and the solution was diluted with glacial acetic acid to 1 liter.
4. 15% Potassium iodide solution: Dissolve 75 g KI crystals in water and make up the volume to 500 ml.

Procedure

1. In a 500ml dry glass stoppered bottle, added 5ml of given oil sample (the oil sample was dissolved in 10 ml chloroform (the concentration of the oil sample is 5%).
2. To the content of the bottle 25 ml of Hanus solution was added and the mixture was allowed to stand in the dark for exactly 30 minutes with occasional shaking.
3. 10 ml Potassium iodine solution was mixed to it and the mixture was shaken well.
4. 100 ml freshly boiling cooled water was added to the mixture and the content of the bottle was titrated with 0.1N sodium thiosulphate solution, using starch solution as indicator.
5. A blank experiment (without the oil) was performed exactly in the same manner as described above.

Calculations:

$$\text{Iodine value} = \frac{S \times (X - Y) \times 0.127 \times 100}{W}$$

Where, S=Strength of the sodium thiosulphate solution.

X= ml of sodium thiosulphate solution required in the blank test.

Y= ml of sodium thiosulphate solution required in true test.

W=Weight of the oil taken in grams.

3.7.3.3 Acid value

The acid number or value of a fat or oil tells the amount of free fatty acid present in it. The acid number is expressed as the Number of milligrams of potassium hydroxide required to neutralize the free fatty acids present in one gram of fat. The acid number of a fat can give the extent of

rancidity in a stored sample. A fat which has been both processed and stored properly has a very low acid number. During storage fats may become rancid as a result of peroxide formation at the double bonds by atmospheric oxygen and hydrolysis by micro-organisms with the liberation of free fatty acid. The amount of free fatty acid present therefore gives an indication of the age and quality of the fat.

Reagents

1. Phenolphthalein: 1 percent solution in alcohol neutralized with 0.1 N NaOH.
2. Denatured alcohol (Neutral): Mix 10 volumes of ethyl alcohol with 1 volume of methyl alcohol and neutralize with N/4 NaOH using phenolphthalein as indicator.
3. 0.25N Sodium hydroxide and 0.1N Sodium hydroxide.
4. 0.05N potassium hydroxide solution

Procedure

Weigh 5-7 g of oil in 250 ml conical flask and add 50 ml denatured alcohol (neutral) and shake well. Now add 2 ml of phenolphthalein as indicator and titrate against 0.05N potassium hydroxide with vigorous shaking after each addition till a permanent light pink color is produced which persists for at least 1 minute (Sharif *et al.*, 2017).

Calculation:

$$\text{Acid value} = \frac{56.1 \times N \times V}{W}$$

Where, N= Strength of potassium hydroxide

V= Number of ml of aqueous potassium hydroxide required for titration

W= Weight of the oil taken in the gram

3.7.4 Estimation of fatty acid composition

Fatty acid composition was determined by Gas-liquid chromatographic method (Uppstrom and Johansson, 1978).

Reagent

- Ethylate reagent (Petroleum ether / 0.02M sodium hydroxide in ethanol (2/3))
- A Salt solution (80 g NaCl and 3g Sodium hydrogen Sulphate in 1 litre water)

Procedure

1. About 12 mg of oil or equivalent amount of oil seeds was taken (seed was crushed in an oil paper and then transferred into a test tube).
2. The sample was extracted and transesterified at the same time with 5 ml ethylated reagent and shaken.
3. The samples were kept for overnight at room temperature.
4. 10 ml salt solution was added and shaken. As soon as the two layers were separated, the benzene phase was transferred to small test tubes.
5. A Philips PU 4500 chromatograph instrument was used with flame ionization detector (FID).
6. A glass column (1.5m x 4mm) was packed with BDS. With this column the injection post, column and detector temperature was set at 220° C, 185° C and 240° C, respectively.
7. Nitrogen flow (used as carrier gas) rate was 22 ml/min, the injection volume was 2µl.
8. Peak areas were measured with an electronic digital integrator (Shinadzu C-R6A chromatopac).

3.7.5 Estimation of Carbohydrate: Total carbohydrate Estimation

The method was described by Raghuramula *et al.* (2003). The content of the available carbohydrate was determined by the following equation:

$$\text{Carbohydrate} = 100 - [(\text{Moisture} + \text{Fat} + \text{Protein} + \text{Ash} + \text{Oil/Fats}) \text{ g/100g}]$$

3.8 Estimation of minerals

3.8.1 Preparation of reagents

3.8.1.1 Reagents for P determination

Reagent A

1. Antimony trioxide (45 g) was added in 400ml water then added 150ml conc. H₂SO₄ and allowed to cool.
2. Ammonium molybdate (7.5 g) was dissolved in 300ml water Cool antimony solution and molybdate solution was mixed by adding 1 liter of water.

Reagent B

1. Gelatin (1 g) was dissolved in 100 ml hot water.
2. Reagent A (150 ml) dissolved to about 500 ml water and dissolved gelatins were mixed and finally, 1 g of ascorbic acid was dissolved with it to make volume 1 liter.

3.8.1.2 Reagent for Ca and Mg determination

1% Lanthanum solution: Lanthanum oxide (La₂O₃) (59 g) were added in 50 ml of water then added 250 ml conc. H₂SO₄ gradually and carefully. It was made up to 5 liters by adding water.

3.8.1.3 Reagents for S determination

Mixed acid seed solution: 65 ml of conc. HNO_3 and 250 ml glacial acetic acid were added to about 500 ml of water. 3 ml of 1000 ppm S standard solution was added and made volume to liter with water.

Turbidimetric reagent

Polyvinyl pyrrolidone (PVP K30) was dissolved (10 g) in about 100 ml of hot water along with 150 g of $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ was dissolved in about 500 ml of water. The PVP and barium chloride solutions were mixed and were made to liter with water

3.8.2 Preparation of standards

1. For standard calibration Cu, Fe, Mn and Zn were prepared together in water following concentration as $2\mu\text{g Cu/ml}$, $10\mu\text{g Fe/ml}$, $4\mu\text{g Mn/ml}$, $2\mu\text{g Zn/ml}$.
2. The P, K and S were also prepared together in water as $20\mu\text{g P/ml}$, $100\mu\text{g K/ml}$, $20\mu\text{g S/ml}$.
3. Finally, Ca and Mg as $100\mu\text{g Ca/ml}$, $40\mu\text{g Mg/ml}$ were prepared in the same solution.

3.8.3 Digestion solution

Nitric-perchloric solution: Conc. Perchloric acid (100 ml) was added to 500 ml concentrated HNO_3 to prepare nitric-perchloric solution.

3.8.3.1 Digestion of sample for determination of P, K, Ca, Mg, S, Cu, Fe, Zn and Mn

Digestion procedure

500 mg oil cake was taken into a 50 ml boiling flask. 5 ml of nitric-perchloric acid (5:1) solution was added and placed in digestion chamber. Started digestion for 1 hour and 30 minutes at 370°C temperature. After completely digestion removed from digestion chamber and was cooled

and 15 ml water was added. For collection of aliquot the flask was slowly agitated and heated to suspend the ash and filter.

3.8.4 Analytical procedure

By using a combination diluter-dispenser, 1 ml aliquot was taken from filtrate and 19 ml water (dilution 1) was added. The other dilutions were made in the following order. For S determination, 7 ml of aliquot from dilution 1, 9 ml of acid seed solution and 4ml of turbid metric solution were mixed together thoroughly. It was allowed to stand 20 minutes and not longer than one hour. The reading was taken in turbid meter or in colorimeter at 535 nm using a cuvette with 2 cm light path. For P and K determination, 1 ml aliquot from dilution 1, 9 ml of water and 10 ml of color reagent were mixed together. It was allowed to stand about 20 minutes and reading was taken by using spectrophotometer at 680 nm for P and by using a flame photometer (Model AnA-135, OSK, Japan) at 766.5 nm for K. For Ca and Mg determination, 1 ml aliquot from dilution 1, 9 ml of water and 10 ml of 1% lanthanum solution were mixed together. It was analyzed by Atomic Absorption Spectrometer contraAA 700(ANALYTIK JENA). For Fe, Mn and Zn determination, the original filtrate was used to analyze these elements by Atomic Absorption Spectrometer (Sharif *et al.* 2017).

3.9 Statistical Analysis

The recorded data for each character from the experiments was analyzed statistically with a one way ANOVA to find out the variation resulting from experimental treatments using Statistix 10 program. The mean for all the treatments were calculated and analysis of variance of characters under the study was performed by F variance test. The mean differences were evaluated by (LSD) Least Significance Difference test (Gomez, *et al.* 1984).

CHAPTER IV

RESULT AND DISCUSSION

Five released cultivars of rapeseed and mustard (*Brassica spp.*) were taken for the determination of physical and chemical characteristics. The seeds were stored in the refrigerator under a suitable storage condition. The proximate composition and some other nutrients compositions of mustard seeds are also testified.

Analytical studies of the whole seeds

The proximate composition of whole rapeseed and mustard seeds of different released cultivars is presented in different tables. The data have also been estimated on a moisture-free basis in order to allow for a better comparison of the different fractions. The data stated are the average of three replication and have been presented and discussed.

4.1 Physical characteristics of rapeseed and mustard released cultivars

4.1.1 1000 grain weight

The significant variations were observed among the different varieties in terms of the effect on the thousand-grain weight (Table 1). The highest weight of thousands grains was found in Tori-7(4.53 ± 0.10 g), which was followed by BARI Sarisha-17(4.07 ± 0.15 g) and Sampad (3.69 ± 0.13 g). The lowest weight of thousands of grains was found in BARI Sarisha-14 (2.93 ± 0.22 g). The present values were consisted with the results reported by (Banga *et al.*, 2013; Siddiqui and firoz, 2004; Chowdhury *et al.*, 1987 and Kaul and Das, 1986). Chowdhury *et al.* (2010) reported range of weight of thousand seed 2.5 g to 4.9 g, among different Bangladeshi mustard varieties and advanced lines. Banga *et al.*

(2013) and Siddiqui and firoz (2004) found that the highest amount of 1000 seed weight were 5.15 g and 3.95 g.

4.1.2 Moisture

The moisture percentage of different mustard and rapeseed genotypes was ranged from $4.45 \pm 0.02\%$ to $4.68 \pm 0.02\%$ (Table 1). The highest value of moisture percentage was found in Tori-7 ($4.68 \pm 0.02\%$), which was followed by Sampad ($4.60 \pm 0.02\%$) and BARI Sarisha-14 ($4.56 \pm 0.03\%$). The lowest value of moisture content was found in BARI Sarisha-17 ($4.40 \pm 0.02\%$), which was also followed by BARI Sarisha-15 ($4.45 \pm 0.02\%$). The results of the moisture content were significantly lower than that of Sarker *et al.* (2015), BARI annual report (1987-88). According to Sarker *et al.* (2015) moisture content of mustard cakes were $9.20 \pm 0.5\%$ and $9.73 \pm 0.6\%$. BARI (1987-88) reported that moisture content ranges from 7.41% to 8.38%. These may be influenced by different level of sun drying after harvesting.

4.1.3 Dry matter

A statistically significant variation was observed for dry matter content of different released cultivar mustard and rapeseed that have been presented in (Table 1). Significantly the highest amount of dry matter contained was recorded in BARI Sarisha-17 ($95.60 \pm 0.02\%$), followed by BARI Sarisha-15 ($95.55 \pm 0.02\%$) and BARI Sarisha-14 ($95.44 \pm 0.03\%$). The lowest amount of dry matter contained was found in Tori-7 ($95.32 \pm 0.02\%$) which was significantly the lowest among all the variety and also followed by Sampad ($95.39 \pm 0.02\%$).

Table 1. Weight of thousand seed, Moisture and Dry matter of different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Weight of 1000 seeds (g)	Moisture percentage	Dry matter percentage
	Mean \pm SD (n=3)		
Sampad	3.69 ^c \pm 0.13	4.60 ^b \pm 0.02	95.39 ^d \pm 0.02
Tori-7	4.53 ^a \pm 0.10	4.68 ^a \pm 0.02	95.32 ^e \pm 0.02
BARI Sarisha-17	4.07 ^b \pm 0.15	4.40 ^e \pm 0.02	95.60 ^a \pm 0.02
BARI Sarisha-14	2.93 ^d \pm 0.22	4.56 ^c \pm 0.03	95.44 ^c \pm 0.03
BARI Sarisha-15	3.68 ^c \pm 0.17	4.45 ^d \pm 0.02	95.55 ^b \pm 0.02
LSD (0.05)	0.2861	0.0339	0.0339
CV (%)	4.16	0.41	0.02

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

4.1.4 Oil content

The oil content of the mustard and rapeseed depends on many factors like genetic factor; agro-ecological conditions including cultivation sites and crop management system etc. The oil content of different released cultivar of mustard and rapeseed were extracted by petrolium ether (40-60°C) varied from 35.27 \pm 1.01% to 43.23 \pm 1.06% (Table 2). The variety BARI Sarisha-14 had the lowest amount of oil content (35.27 \pm 1.01%), while the variety Sampad contained significantly the highest amount of oil (43.23 \pm 1.06%), followed by Tori-7 (40.26 \pm 0.98%), BARI Sarisha-17 (40.26 \pm 0.98%) and BARI Sarisha-15 (37.22 \pm 0.99). The results

clearly showed that variety Sampad, Tori-7, BARI Sarisha-17 can be measured as better source of oil. Present values were higher than the reported value of Gadei *et al.*, (2012) and Moser *et al.* (2009), who found that oil content of mustard seed ranges from 28–32%; whereas Arif *et al.* (2012), Bhowmik (2003), Novoselov *et al.* (1997) reported that oil content of rapeseed ranges from 45.67%-43.87%, 42-46% and 45-46% respectively, which are slightly higher than present results. On the other hand, the present investigations were more or less similar the reported values of (Sengupta and Das, 2003; Mandal *et al.*, 2002; Rathore, 2000; Niraj *et al.*, 2000; Vijay *et al.*, 1992). These variations might be due to biological factor, environmental factor, soil and crop management practices.

4.1.5 Oil cake

Oil cake/meals are used for diverse purposes. Oil cake is a nutritious food items for cattle and fish. It is also used as a good organic fertilizer and ingredient of composts. The BARI Sarisha-14 contained the highest amount of oil cake ($64.72 \pm 1.01\%$), followed by BARI Sarisha-15 ($62.78 \pm 0.99\%$). The lowest value was found in Sampad ($56.77 \pm 1.06\%$), followed by BARI Sarisha-17($59.76 \pm 0.98\%$) and Tori-7(61.03 ± 0.67) (Table 2). Chowdhury *et al.* (2014) and Appelqvist and Ohlson (1992) found the similar result. Chowdhury *et al.* (2014) showed that oil cake percentage vary from 58.14 to 59.95% and Appelqvist and Ohlson (1992), reported that typically rape seed oil (kind of mustard seed) contain 58% cake.

4.1.6 Dry weight of cake

Dry cakes are used to estimate the content of different nutrient which are crucial for our poultry feed, organic fertilizer and other different purposes. The dry weights of cake had been presented in (Table 2). The

highest value was obtained from BARI Sarisha-14 (60.17 ± 1.03), followed by BARI Sarisha-15 ($58.33 \pm 1.00\%$). The lowest value was found in Sampad ($52.17 \pm 1.05\%$), followed by BARI Sarisha-17 ($55.36 \pm 0.98\%$) and Tori-7 (56.35 ± 0.68).

Table 2. Proximate analysis of oil content, oil cake and Dry wt. of cake of different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Oil percentage	Oil cake	Dry oil cake
	Mean \pm SD (n=3)		
Sampad	$43.23^a \pm 1.06$	$56.77^d \pm 1.06$	$52.17^d \pm 1.05$
Tori-7	$38.97^b \pm 0.67$	$61.03^c \pm 0.67$	$56.35^c \pm 0.68$
BARI Sarisha-17	$40.26^b \pm 0.98$	$59.76^c \pm 0.98$	$55.36^c \pm 0.98$
BARI Sarisha-14	$35.27^d \pm 1.01$	$64.72^a \pm 1.01$	$60.17^a \pm 1.03$
BARI Sarisha-15	$37.22^c \pm 0.99$	$62.78^b \pm 0.99$	$58.33^b \pm 1.00$
LSD (0.05)	1.7366	1.7366	1.7483
CV (%)	2.45	1.56	1.70

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

4.1.7 Ash

Ash content of different released cultivars of mustard and rapeseed were variable and ranged from $11.05 \pm 0.03\%$ to $12.49 \pm .03\%$ (Table 3). Significantly the highest amount of ash contained was recorded in BARI Sarisha-17 ($12.49 \pm .03\%$), and followed BARI Sarisha-15 ($11.90 \pm 0.02\%$) and BARI Sarisha-14 ($11.59 \pm 0.04\%$). On the other hand Sampad ($11.05 \pm 0.03\%$) contained the lowest amount of ash content that was also

followed by Tori-7($11.31 \pm 0.02\%$). The present values were significantly higher than the reported value of (Sarker *et al.*, 2015; Abul-Fadl *et al.*, 2011; Sosulski and Bakal, 1991; Nehrins and fettu, 1990 and Kaul and Das, 1986).

4.1.8 Protein

Protein is the major nutrient of different genotypes of rapeseed and mustard. Protein content is genetically controlled. It is also influenced by nitrogen fertilizer application and agronomics practices. The protein content was determined on moisture free basis. Protein content of different genotypes of rapeseed and mustard have been presented in (Table 3). The statistically the highest amount of protein was obtained from BARI Sarisha-15 ($26.80 \pm 0.07\%$) and followed by BARI Sarisha-14 ($26.28 \pm 0.25\%$) and BARI Sarisha-17 ($26.01 \pm 0.20\%$). On the other hand, Sampad ($23.08 \pm 0.20\%$) was contained the lowest amount of protein and also followed by Tori-7($24.70 \pm 0.38\%$). The present values were more or less similar with the reported values of (Sarker *et al.*, 2015; Chowdhury *et al.*, 2010; Sosulki *et al.*, 1991 and Mirza *et al.*, 1998). However these result are lower than those reported by many other authors: Prapakornwiriya and Diosady (2004) determined the protein 45.0%, 34.0% respectively and Sengupta and Das (2003) revealed that protein content of rapeseed were ranges from 44.2-44.7%. This might be due to the nitrogen fertilizer application, ecology and agronomics practices.

4.1.9 Carbohydrate

Carbohydrate content of different released genotypes of rapeseed and mustard were determined moisture free basis. The data had been presented in (Table 3). The highest amount of carbohydrate found in BARI Sarisha-15 ($22.29 \pm 0.81\%$) but statistically identical with BARI Sarisha-14 ($21.72 \pm 1.12\%$). The lowest amount of carbohydrate was

obtained from BARI Sarisha-17 ($16.85 \pm 0.77\%$) whereas it was also statistically identical with Sampad ($18.03 \pm 0.87\%$) and Tori-7 ($18.23 \pm 0.61\%$). Agronomics practices, environmental factors as well as variation among the released cultivars might be influenced the carbohydrate content. The present values were slightly lower than the reported values of (Bachheti *et al.*, 2012 and Gopalan *et al.*, 1981). Bachheti *et al.* (2012) found that mustard seeds contain 23.8% carbohydrate and Gopalan *et al.* (1981) stated that dry mustard seeds contained 20-23% carbohydrate.

Table 3. Proximate analysis of Protein percentage, Carbohydrate percentage and Ash percentage of different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Ash (%)	Protein (%)	Carbohydrate (%)
	Mean \pm SD (n=3)		
Sampad	11.05 ^e \pm 0.03	23.08 ^d \pm 0.20	18.03 ^b \pm 0.87
Tori-7	11.31 ^d \pm 0.02	24.70 ^c \pm 0.38	18.23 ^b \pm 0.61
BARI Sarisha-17	12.49 ^a \pm 0.03	26.01 ^b \pm 0.20	16.85 ^b \pm 0.77
BARI Sarisha-14	11.59 ^c \pm 0.04	26.28 ^b \pm 0.25	21.72 ^a \pm 1.12
BARI Sarisha-15	11.90 ^b \pm 0.02	26.80 ^a \pm 0.07	22.29 ^a \pm 0.81
LSD (0.05)	0.0562	0.4477	1.5588
CV (%)	0.26	0.97	4.41

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

4.2 Chemical characteristics of rapeseed and mustard released cultivars.

4.2.1 Chemical constant of oil

4.2.1.1 Saponification value

Saponification value of oil/fats defined as the number of mg of KOH required to saponify one gram of fats /oil. It is inversely proportionate to the molecular weight or chain length of the fatty acids present in the

fats/oil. Saponification values of different released cultivars were ranges from 161.34 ± 1.00 to 168.25 ± 0.15 and had been presented in (Figure 1 and Appendix I). The highest Saponification value was found in BARI Sarisha-15 ($168.25 \pm 0.15\%$), which was statistically identical with Tori-7 ($166.77 \pm 0.51\%$) and BARI Sarisha-17 ($166.16 \pm 2.37\%$). But Sampad contained the lowest amount of Saponification value ($161.34 \pm 1.00\%$), which was also statistically identical with BARI Sarisha-14. The present values were lower than the reported values of (Khan *et al.*, 2013 and Richet *et al.*, 1987).

4.2.1.2 Iodine value

Iodine value is defined as grams of iodine absorbed by 100 g fats/oil. It helps to estimate the degree of unsaturation. The iodine values of different mustard and rapeseed genotypes have been presented in (Figure 1 and Appendix I). The highest amount of iodine value were observed in BARI Sarisha-15 (106.27 ± 1.03), followed by BARI Sarisha-14 (103.10 ± 0.16) and BARI Sarisha-17 (101.20 ± 0.38). The lowest amount of iodine value recorded in Sampad (98.09 ± 0.77), followed by Tori-7 (99.74 ± 0.35). The observed values were supported by the reported values of (Chowdhury *et al.*, 2010; Khan *et al.*, 2013 and Richet *et al.*, 1987).

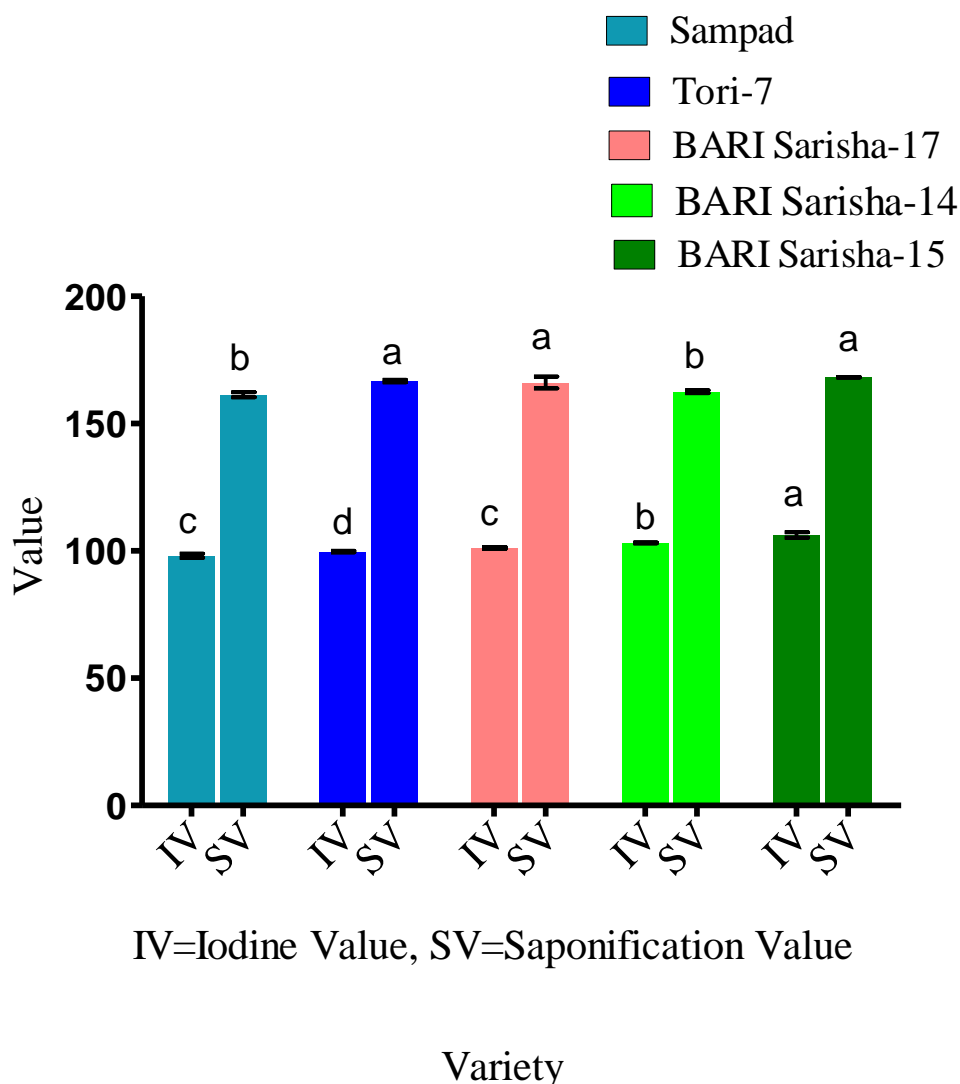


Figure 1. The Chemical constant (Saponification Value and Iodine Value) of oil of the different released cultivars of rapeseed and mustard (*Brassica ssp.*)

4.2.1.3 Acid value

It is defined as the milligrams of KOH required to neutralize the free fatty acids present in 1 g of fats/oil. This value is used in determining the rancidity due to free fatty acids. Acid values of different released cultivar of mustard and rapeseed had been presented in (Figure 2 and Appendix I). The highest acid value was found from Sampad (1.72 ± 0.05), followed by BARI Sarisha-17 (1.60 ± 0.04); whereas the lowest acid value was found from BARI Sarisha-15 (1.20 ± 0.02), followed by Tori-7

(1.27 ± 0.03) and BARI Sarisha-14 (1.35 ± 0.04). BARI Sarisha-15 was statistically similar with Tori -7; whereas Tori-7 also statistically similar with BARI Sarisha-14. Chowdhury *et al.* (2010) and Khan *et al.* (2013) found the more or less similar result. Although the present values were lower than the reported values of (Richet *et al.*, 1987).

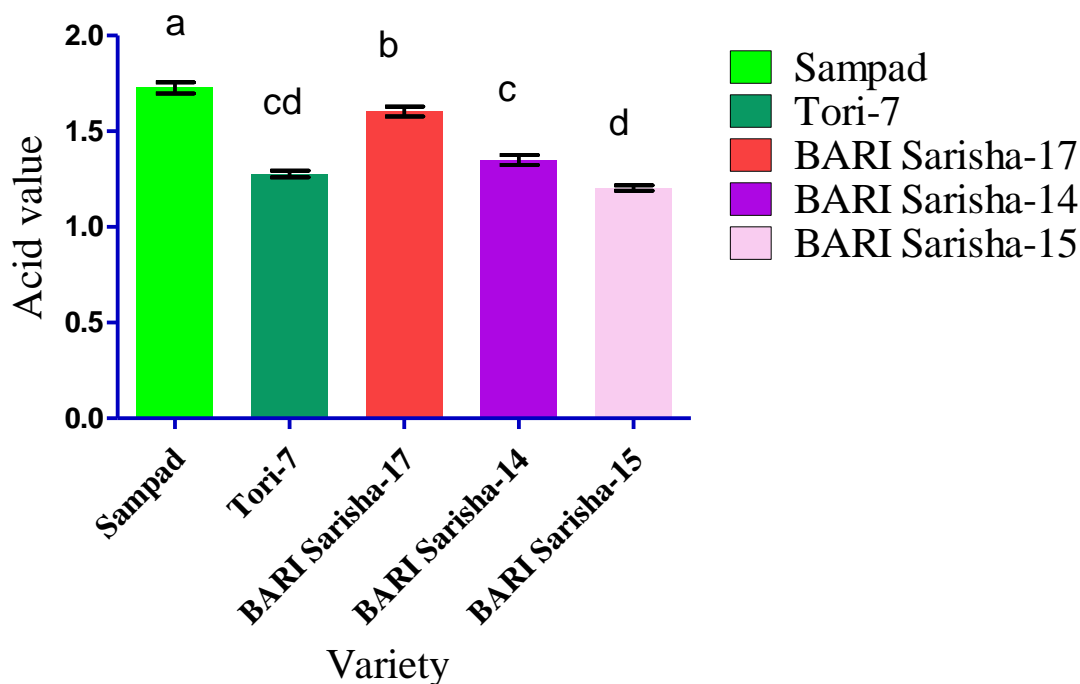


Figure 2. The Chemical constant (Acid Value) of oil of the different released cultivars of mustard (*Brassica ssp*)

4.2.2 Fatty acid composition

Comparison of fatty acid composition determined through gas chromatography are demonstrated in (Table 4). According to results, there was a significant difference between the studied rapeseed and mustard released cultivars in terms of their fatty acid compositions. Significantly the highest amount of palmitic acid was observed in BARI Sarisha-17 ($3.23 \pm 0.05\%$); followed by BARI Sarisha-14 ($2.87 \pm 0.07\%$) and BARI Sarisha-15 ($2.86 \pm 0.06\%$). The lowest amount of palmitic acid content was observed in Sampad ($2.15 \pm 0.03\%$). The concentration of

stearic acid varied from $1.20 \pm 0.02\%$ to $1.56 \pm 0.03\%$; whereas arachidic acid contents ranged from $4.04 \pm 0.07\%$ to $5.65 \pm 0.09\%$. BARI Sarisha-14 contained the highest amount ($1.31 \pm 0.03\%$) of behenic acid. BARI Sarisha-15 contained the highest amount ($18.44 \pm 0.03\%$) of oleic acid; followed by BARI Sarisha-17 ($18.34 \pm 0.02\%$) and the lowest amount was found in Sampad ($14.34 \pm 0.02\%$) which was significantly lowest among all the varieties. Linoleic acid content of the released genotypes ranged from $13.07 \pm 0.02\%$ to $15.55 \pm 0.02\%$. The highest amount of linoleic acid contents was found in Sampad ($15.55 \pm 0.02\%$) which was significantly highest among all the released cultivars and the lowest amount ($13.07 \pm 0.02\%$) was found in BARI Sarisha-17. The linoleic acid content is important from the stand point of utilization of oil for food products. Linolenic acid, erucic acids were also present in these varieties. The concentration of linolenic acid varied from $5.88 \pm 0.04\%$ to $7.89 \pm 0.01\%$; whereas erucic acid contents ranged from $47.67 \pm 0.02\%$ to $51.43 \pm 0.02\%$. Differences were found among the released cultivars of rapeseed and mustard in respect of erucic acid content. BARI Sarisha-17 contained the highest amount of erucic acid ($51.43 \pm 0.02\%$), followed by Tori-7 ($50.22 \pm 0.04\%$) and lowest amount was found in BARI Sarisha-14 ($47.67 \pm 0.02\%$); which was significantly lowest among all the varieties. GLC analytical data indicated that the major fatty acid composition of the five genotypes of mustard and rapeseed oils included unsaturated fatty acid ranging from $85.27 \pm 0.03\%$ to $89.11 \pm 0.03\%$. The highest amount of total unsaturated fatty acid contained BARI Sarisha-15 ($89.11 \pm 0.03\%$). The lowest amount of total unsaturated fatty acid contained Tori-7 ($85.27 \pm 0.03\%$), while only a minor fraction by saturated fatty acids ($8.74 \pm 0.09\%$ to $11.30 \pm 0.19\%$) (Figure 3 and Appendix II).

Table 4. Fatty acid composition of different released cultivars of rapeseed and mustard (*Brassica spp.*).

Name of the released cultivars(Treatments)	Palmitic acid (C _{16:0})	Stearic acid (C _{18:0})	Arachidic acid (C _{20:0})	Behenic acid (C _{22:0})	Oleic acid (C _{18:1})	Linoleic acid (C _{18:2})	Linolenic acid(C _{18:3})	Erucic acid (C _{22:1})
mean±SD								
Sampad	2.15 ^c ±0.03	1.34 ^c ±0.02	4.21 ^c ±0.06	1.04 ^c ±0.01	14.34 ^e ±0.02	15.55 ^a ±0.02	6.84 ^e ±0.03	48.53 ^d ±0.02
Tori-7	2.24 ^c ±0.04	1.24 ^d ±0.01	4.04 ^d ±0.07	1.22 ^b ±0.01	16.15 ^d ±0.03	15.45 ^b ±0.02	6.22 ^d ±0.04	50.22 ^b ±0.04
BARI Sarisha-17	3.23 ^a ±0.05	1.20 ^d ±0.02	4.05 ^d ±0.06	0.89 ^e ±0.01	18.34 ^b ±0.02	13.07 ^e ±0.02	5.88 ^e ±0.04	51.43 ^a ±0.02
BARI Sarisha-14	2.87 ^b ±0.07	1.46 ^b ±0.01	5.65 ^a ±0.09	1.31 ^a ±0.03	16.53 ^c ±0.03	15.34 ^c ±0.02	7.13 ^b ±0.03	47.67 ^e ±0.02
BARI Sarisha-15	2.86 ^b ±0.06	1.56 ^a ±0.03	4.73 ^b ±0.08	0.97 ^d ±0.01	18.44 ^a ±0.03	14.03 ^d ±0.02	7.89 ^a ±0.01	48.74 ^c ±0.02
LSD (0.05)	0.1032	0.0387	0.1419	0.03	0.0499	0.0428	0.0666	0.0453
CV (%)	2.12	1.56	1.72	1.66	0.16	0.16	0.54	0.05

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

From the present data, it might be suggested that all the *Brassica* oil seeds are suitable for edible purpose as they contained higher amount of unsaturated fatty acid. These findings were in conformity with the results by (Abul-fadl *et al.*, 2011; Moser *et al.*, 2009; Niraj *et al.*, 2000 and Appelqvist, 1980). Abul-fadl *et al.* (2011) reported that, erucic acid was in yellow and brown mustard seeds oils was represented about 37.89% and 23.90%, respectively. Oleic acid ranged between 19.08% to 20.24% of total fatty acid profiles in both yellow and brown mustard seed oils respectively. Moreover linoleic acid was recorded from 12.37 to 21.36 in both yellow and brown mustard seed oil.

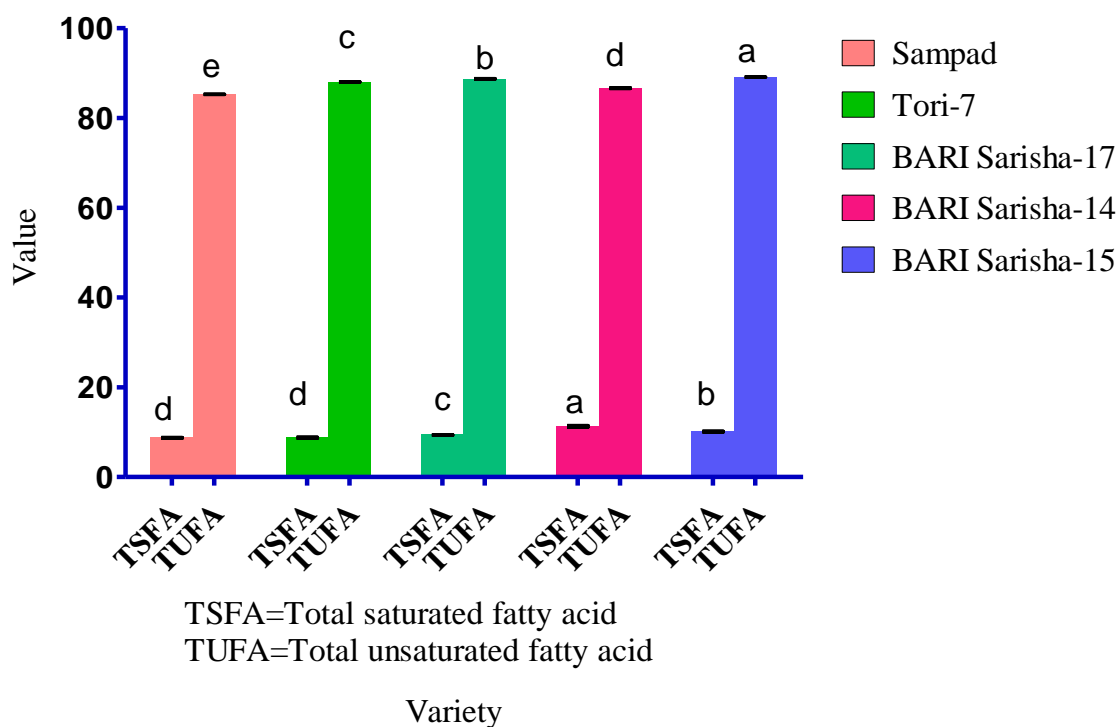


Figure 3. Percentage of total saturated and unsaturated fatty acid of oil of the different released cultivars of rapeseed and mustard (*Brassica* spp.)

4.3 Minerals

Different major and minor minerals were analyzed in this work. The amounts of major minerals content of rapeseed and mustard had been illustrated in (Table 5 and Figure 4). It was well known that rapeseed and mustard contained a small amount of micro minerals (Table 6).

4.3.1 Major Minerals

4.3.1.1 Calcium (Ca)

In case of calcium content of different released cultivars of rapeseed and mustard was ranged from $0.24 \pm 0.01\%$ to $0.44 \pm 0.01\%$ (Table 5). Significantly the highest amount of calcium (Ca) content was observed in BARI Sarisha-14 ($0.44 \pm 0.01\%$), followed by BARI Sarisha-17 ($0.37 \pm 0.01\%$), Tori-7 ($0.32 \pm 0.01\%$). The lowest amount of calcium content was obtained from BARI Sarisha-15 ($0.24 \pm 0.01\%$), followed by Sampad ($0.25 \pm 0.01\%$). The present investigations were supported by reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Josefson, 1988; Sengupta and Das, 2003). Bachheti *et al.* (2012), Sengupta and Das (2003) reported that mustard contained 492.1 mg and 490 mg respectively, while Josefson (1988) stated that mustard contain 0.7% Ca.

4.3.1.2 Magnesium (Mg)

Magnesium is the major minerals for human nutrition. Magnesium content of different genotypes of rapeseed and mustard had been presented in (Table 5). Magnesium content of different released cultivars was ranged from $0.33 \pm 0.01\%$ to $0.45 \pm 0.01\%$. The highest amount of Magnesium content was found in BARI Sarisha-17 ($0.45 \pm 0.01\%$); followed by Tori-7 ($0.43 \pm 0.01\%$) and the lowest amount in BARI Sarisha-15 ($0.33 \pm 0.01\%$); followed by BARI Sarisha-14 ($0.39 \pm 0.01\%$) and Sampad ($0.40 \pm 0.01\%$). BARI Sarisha-17 was significantly higher than all other varieties. The present investigations were supported by

reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Sengupta and Das, 2003; Josefson, 1988).

Table 5. Proximate analysis of major minerals (Calcium, Magnesium and Sulfur) content of different genotypes of rapeseed and mustard (*Brassica* ssp.) oil cake.

Name of the released cultivars (Treatments)	Ca%	Mg%	S%
	Mean \pm SD (n=3)		
Sampad	0.25 ^d \pm 0.01	0.40 ^c \pm 0.01	0.41 ^b \pm 0.001
Tori-7	0.32 ^c \pm 0.01	0.43 ^b \pm 0.01	0.43 ^a \pm 0.01
BARI Sarisha-17	0.37 ^b \pm 0.01	0.45 ^a \pm 0.01	0.41 ^b \pm 0.001
BARI Sarisha-14	0.44 ^a \pm 0.01	0.39 ^c \pm 0.01	0.34 ^c \pm 0.001
BARI Sarisha-15	0.24 ^d \pm 0.01	0.33 ^d \pm 0.01	0.33 ^c \pm 0.01
LSD (0.05)	0.0182	0.0182	0.0182
CV (%)	3.09	2.50	2.60

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

4.3.1.3 Sulfur (S)

Statistically, significant variation was recorded in Sulfur content due to different mustard and rapeseed genotypes (Table 5). The statistically significant highest Sulfur content was recorded from Tori-7 (0.43 \pm 0.01%), which was followed BARI Sarisha-17 (0.41 \pm 0.001%) and Sampad (0.41 \pm 0.001%). The lowest Sulfur content was observed BARI Sarisha-15 (0.33 \pm 0.01%), which was statistically identical to BARI Sarisha-15 (0.34 \pm 0.001%). The present investigations were supported by reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Sengupta and Das, 2003; Josefson, 1988).

4.3.1.4 Phosphorus (P)

In case of Phosphorus content of different released genotypes of rapeseed and mustard was ranged from $0.70 \pm 0.01\%$ to $1.09 \pm 0.03\%$ (Figure 4 and Appendix III). The highest amount of Phosphorus (P) content was observed in BARI Sarisha-17 ($1.09 \pm 0.03\%$), followed by BARI Sarisha-14 ($1.02 \pm 0.01\%$), Tori-7 ($0.99 \pm 0.01\%$). The lowest amount of phosphorus content was obtained from BARI Sarisha-15 ($0.70 \pm 0.01\%$), which was also followed by Sampad ($0.87 \pm 0.02\%$). The present investigations were supported by reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Sengupta and Das, 2003; Josefson, 1988).

4.3.1.5 Potassium (K)

Statistically, significant variation was recorded in terms of Potassium content due to different rapessed and mustard genotypes (Figure 4 and Appendix III). The highest Potassium content was recorded from BARI Sarisha-14 ($0.90 \pm 0.01\%$), which was followed by Tori-7 ($0.84 \pm 0.03\%$). The lowest Potassium content was observed from BARI Sarisha-17 ($0.48 \pm 0.02\%$), which was also followed by BARI Sarisha-15 ($0.78 \pm 0.01\%$) and Sampad ($0.78 \pm 0.01\%$). The present investigations were supported by reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Sengupta and Das, 2003; Josefson, 1988).

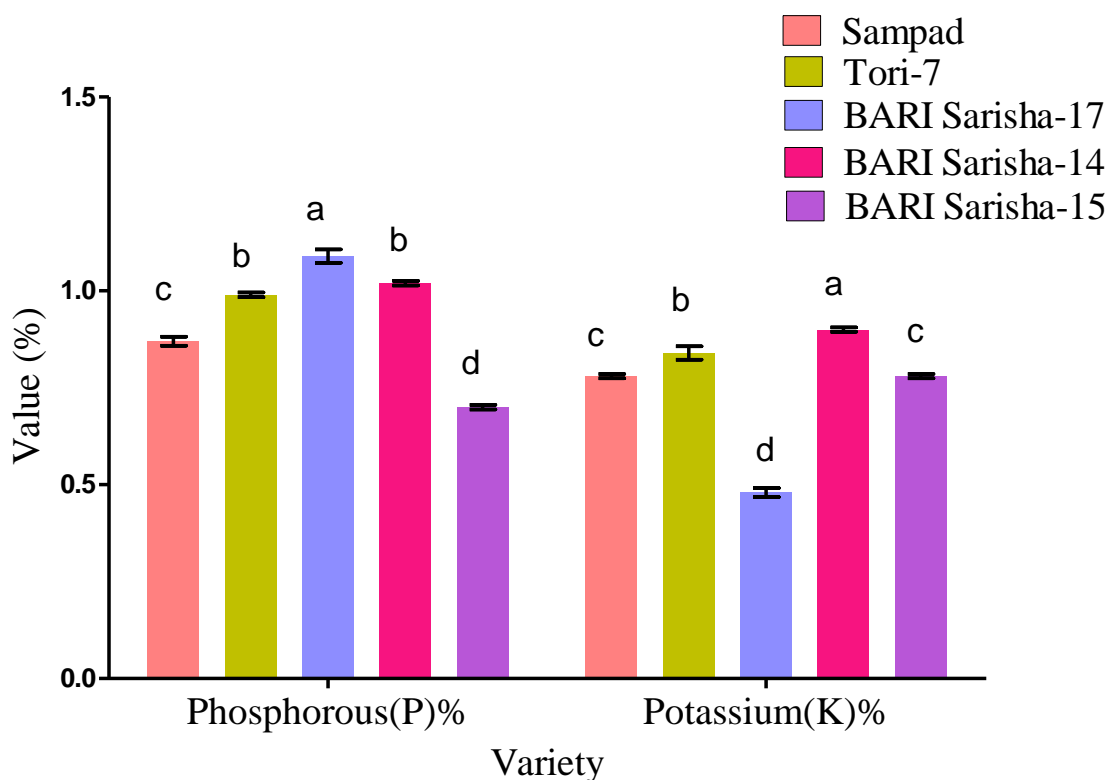


Figure 4. Proximate analysis of major minerals (Potassium and phosphorous) content of different released cultivars of rapeseed and mustard (*Brassica* spp.) oil cake

4.3.2 Minor minerals

4.3.2.1 Copper (Cu)

Copper contained of different varieties of rapeseed and mustard was ranged from 9.00 ± 0.28 ppm to 13.00 ± 0.86 ppm (Table 6). Significantly the highest amount of Cu contained observed in BARI Sarisha-14 (13.00 ± 0.86 ppm) which was followed by BARI Sarisha-17 (10.25 ± 0.13 ppm), whereas BARI Sarisha-17 was statistically similar to BARI Sarisha-15 (9.50 ± 0.16 ppm). The lowest amount of Cu contained observed in Tori-7(8.25 ± 0.16 ppm) which was also statistically similar to Sampad (9.00 ± 0.28 ppm). The present investigations were supported by reported value of (Sarker *et al.*, 2015; Bachheti *et al.*, 2012; Sengupta and Das, 2003; Josefson, 1988).

Table 6. Proximate analysis of minor minerals content of different genotypes of rapeseed and mustard (*Brassica* spp.) oil cake.

Name of the released cultivars (Treatments)	Zn (ppm)	Cu (ppm)	Mn (ppm)	Fe (ppm)
	Mean \pm SD			
Sampad	17.05 ^c \pm 1.32	9.00 ^{cd} \pm 0.28	28.25 ^b \pm 3.10	69.50 ^d \pm 3.10
Tori-7	48.37 ^a \pm 1.8	8.25 ^d \pm 0.16	27.25 ^c \pm 3.25	248.25 ^a \pm 3.25
BARI Sarisha-17	21.32 ^b \pm 0.84	10.25 ^b \pm 0.13	30.75 ^a \pm 3.00	103.00 ^c \pm 3.00
BARI Sarisha-14	23.45 ^b \pm 2.27	13.00 ^a \pm 0.86	31.25 ^a \pm 3.25	126.50 ^b \pm 3.25
BARI Sarisha-15	14.21 ^d \pm 1.05	9.50 ^{bc} \pm 0.16	26.25 ^d \pm 3.25	72.25 ^d \pm 3.25
LSD (0.05)	2.8203	0.7666	0.5729	5.7701
CV (%)	6.23	4.21	1.10	2.56

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

4.3.2.2 Iron (Fe)

Statistically, significant variation was recorded in terms of Iron content due to different rapeseed and mustard genotypes (Table 6). The highest iron content was recorded from Tori-7 (248.25 \pm 3.25ppm) which was followed by BARI Sarisha-14 (126.50 \pm 3.25ppm) and BARI Sarisha-17 (103.00 \pm 3.00ppm). The variety Sampad showed the lowest amount of iron (69.50 \pm 3.10ppm) which was statistically identical with BARI Sarisha- 15 (72.25 \pm 3.25ppm). These might be influenced the different levels of Fe in soil, Fertilizer and variation among the varieties. The present values were higher than the reported values of (Bachheti *et al.*, 2012 and Josefson, 1988) who found that Fe content of mustard seed oil were 8.11 (g/100g) and 18 ppm respectively.

4.3.2.3 Manganese (Mn)

The Manganese content of different varieties of rapeseed and mustard were ranges from 26.25 ± 3.25 ppm to 31.25 ± 3.25 ppm in (Table 6). Significantly the highest amount of manganese contained was found in BARI Sarisha-14 (31.25 ± 3.25 ppm) which was statistically identical with BARI Sarisha-17 (30.75 ± 3.00 ppm) and followed by Sampad (28.25 ± 3.10 ppm). The lowest amount was found in BARI Sarisha-15 (26.25 ± 3.25 ppm) which was followed by Tori-7 (27.25 ± 3.25 ppm). The present values were supported by the reported value of (Bachheti *et al.*, 2012 and Josefson, 1988).

4.3.2.4 Zinc (Zn)

The zinc content of different rapeseed and mustard oil cake was ranged from 14.21 ± 1.05 ppm to 48.37 ± 1.8 ppm (Table 6). Significantly the highest amount of zinc was found in Tori-7 (48.37 ± 1.8 ppm) and followed by BARI Sarisha-14 (23.45 ± 2.27 ppm), BARI Sarisha-17 (21.32 ± 0.84 ppm). The lowest amount of zinc was found in BARI Sarisha-15 (14.21 ± 1.05 ppm) which was also followed by Sampad (17.05 ± 1.32 ppm). The present values were supported by the reported value of (Bachheti *et al.*, 2012 and Josefson, 1988).

4.4 Gross energy

Energy from carbohydrate of mustard and rapeseed varied significantly due to different released genotypes (Table 7). Significantly the highest amount of energy from carbohydrate found in two cultivars BARI Sarisha-14 (89.17 ± 3.27 kcal/g) and BARI Sarisha-15 (86.91 ± 4.48 kcal/g); while significantly the lowest amount found in BARI Sarisha-17 (67.42 ± 3.09 kcal/g) which was statistically identical with Tori-7 (72.92 ± 2.44 kcal/g) and Sampad (72.12 ± 3.51 kcal/g). The significantly highest energy from protein observed from Tori-7 (107.21 ± 0.29 kcal/g),

followed by BARI Sarisha-14 (105.13 ± 1.00 kcal/g) and BARI Sarisha-17 (104.05 ± 0.80 kcal/g); whereas significantly the lowest amount of energy (92.34 ± 0.82 kcal/g) from protein observed from Sampad. The highest amount of energy from fat was observed in Sampad (389.04 ± 9.54 kcal/g), which was significantly higher than all other released cultivars, followed by BARI Sarisha-17 (362.13 ± 8.87 kcal/g) and Tori-7 (350.73 ± 6.03 kcal/g); whereas the lowest amount counted from BARI Sarisha-14 (317.49 ± 9.09 kca/g).

Table 7. Proximate analysis of Gross energy from carbohydrates, Proteins and oils of different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Energy from protein	Energy from fat	Energy from carbohydrate
	mean \pm SD (n=3)		
Sampad	92.34 ^d \pm 0.82	389.04 ^a \pm 9.54	72.12 ^b \pm 3.51
Tori-7	107.21 ^a \pm 0.29	350.73 ^b \pm 6.03	72.92 ^b \pm 2.44
BARI Sarisha-17	104.05 ^b \pm 0.80	362.13 ^b \pm 8.87	67.42 ^b \pm 3.09
BARI Sarisha-14	105.13 ^b \pm 1.00	317.49 ^d \pm 9.09	89.17 ^a \pm 3.27
BARI Sarisha-15	98.82 ^c \pm 1.55	334.98 ^c \pm 8.95	86.91 ^a \pm 4.48
LSD (0.05)	1.7906	15.630	6.2350
CV (%)	0.97	2.45	4.41

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

The study found that gross energy of different released cultivars of rapeseed and mustard ranged from 511.80 ± 4.87 to 553.51 ± 5.20 kcal/g. The statistically highest amount of gross energy found from Sampad (553.51 ± 5.20 kcal/g), followed by BARI Sarisha-17 (533.61 ± 5.00

kcal/g) and Tori-7 (530.86 ± 3.37 kcal/g); while lowest amount of gross energy recorded from BARI Sarisha-14 (511.80 ± 4.87 kcal/g) followed by BARI Sarisha-15 (520.71 ± 4.87 kcal/g).

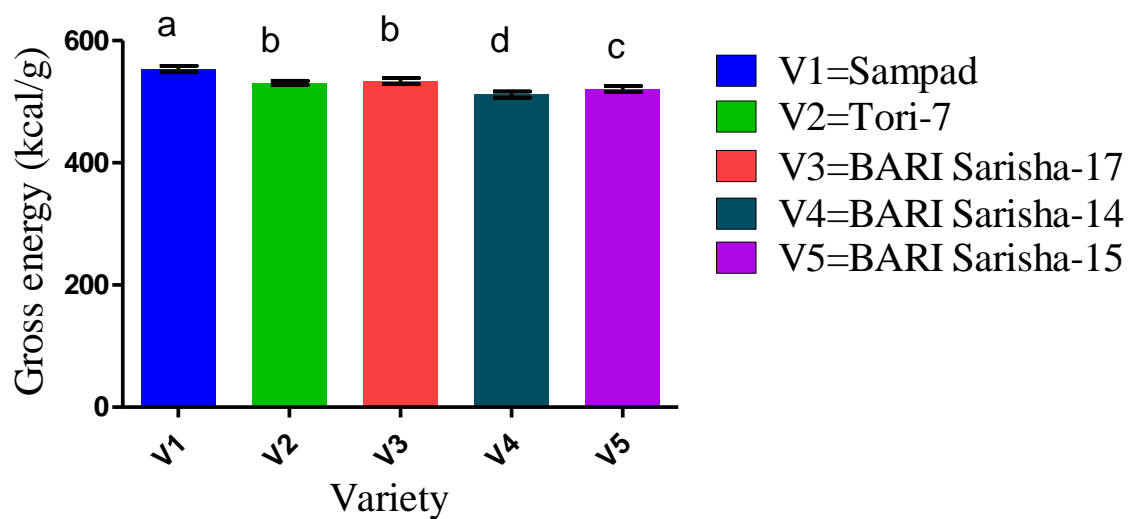


Figure 5. Gross Energy of the different released cultivars of rapeseed and mustard (*Brassica* spp.)

CHAPTER V

SUMMARY AND CONCLUSION

From my research work we found that, the highest weight of thousands grains was found in Tori-7 (4.53 ± 0.10 g) and the lowest thousand grains weight was found in BARI Sarisha-14 ($2.93d \pm 0.22$ g). The highest value of moisture percentage was found in Tori-7 ($4.68 \pm 0.02\%$); whereas the lowest moisture content was found in BARI Sarisha-17 ($4.40 \pm 0.02\%$). Significantly highest amount of dry matter contained was recorded in BARI Sarisha-17 ($95.60 \pm 0.02\%$); while the lowest amount of dry matter contained was found in Tori-7 ($95.32 \pm 0.02\%$). The oil content of different released cultivar of mustard and rapeseed were extracted by petroleum ether (40-600C) varied from $35.27 \pm 1.01\%$ to $43.23 \pm 1.06\%$. Significantly highest amount of oil content was found in Sampad ($43.23 \pm 1.06\%$) while the lowest amount of oil content was observed in BARI Sarisha-14 ($35.27 \pm 1.01\%$). The BARI Sarisha-14 contained highest amount of oil cake ($64.72 \pm 1.01\%$), and the lowest value was found in Sampad ($56.77 \pm 1.06\%$). The highest dry weights of cake were obtained from BARI Sarisha-14 ($60.17 \pm 1.03\%$) and the lowest value obtained from Sampad ($52.17 \pm 1.05\%$).

Saponification values of different released cultivars were ranges from (161.34 ± 1.00 to 168.25 ± 0.15). The statistically highest saponification value was found in BARI Sarisha-15 ($168.25 \pm 0.15\%$) whereas Sampad contained lowest amount of saponification value ($161.34 \pm 1.00\%$). The highest amount of iodine value were observed in BARI Sarisha-15 (106.27 ± 1.03) and the lowest amount of iodine value recorded in Sampad (98.09 ± 0.77). The highest acid value was found from Sampad (1.72 ± 0.05); whereas the lowest acid value was found from BARI Sarisha-15 (1.20 ± 0.02).

Significantly the highest amount of palmitic acid was observed in BARI Sarisha-17 ($3.23 \pm 0.05\%$) and the lowest amount of palmitic acid content was observed in Sampad ($2.15 \pm 0.03\%$).

The concentration of stearic acid varied from $1.20 \pm 0.02\%$ to $1.56 \pm 0.03\%$; whereas arachidic acid contents ranged from $4.04 \pm 0.07\%$ to $5.65 \pm 0.09\%$. BARI Sarisha-14 contained the highest amount ($1.31 \pm 0.03\%$) of behenic acid.

BARI Sarisha-15 contained the highest amount ($18.44 \pm 0.03\%$) of oleic acid and the lowest amount was found in Sampad ($14.34 \pm 0.02\%$) which was significantly the lowest among all the varieties. The highest amount of lenoleic acid contained was found in Sampad ($15.55 \pm 0.02\%$) and the lowest amount ($13.07 \pm 0.02\%$) was found in BARI Sarisha-17. The concentration of linolenic acid varied from $5.88 \pm 0.04\%$ to $7.89 \pm 0.01\%$; whereas erucic acid contents ranged from $47.67 \pm 0.02\%$ to $51.43 \pm 0.02\%$. BARI Sarisha-17 contained the highest amount of erucic acid ($51.43 \pm 0.02\%$) and the lowest amount was found in BARI Sarisha-14 ($47.67 \pm 0.02\%$); which was significantly the lowest among all the varieties. Mustard and rapeseed oils included unsaturated fatty acid ranging from $85.27 \pm 0.03\%$ to $89.11 \pm 0.03\%$; while only a minor fraction by saturated fatty acids ($8.74 \pm 0.09\%$ to $11.30 \pm 0.19\%$).

Ash content of different released genotypes of mustard and rapeseed were variable and ranged from $11.05 \pm 0.03\%$ to $12.49 \pm 0.03\%$. Significantly the highest amount of ash contained was recorded in BARI Sarisha-17 ($12.49 \pm 0.03\%$). On the other hand Sampad ($11.05 \pm 0.03\%$) contained the lowest amount of ash content. The highest amount of protein was obtained from BARI Sarisha-15 ($26.80 \pm 0.07\%$), the lowest amount of protein found in Sampad ($23.08 \pm 0.20\%$). The highest amount of carbohydrate found in BARI Sarisha-15 ($22.29 \pm 0.81\%$). The lowest

amount of carbohydrate was obtained from BARI Sarisha-17 ($16.85 \pm 0.77\%$). Significantly the highest amount of calcium (Ca) content was observed in BARI Sarisha-14 ($0.44 \pm 0.01\%$), while the lowest amount of calcium content was obtained from BARI Sarisha-15 ($0.24 \pm 0.01\%$). The highest amount of magnesium content was found in BARI Sarisha-17 ($0.45 \pm 0.01\%$) and the lowest amount in BARI Sarisha-15 ($0.33 \pm 0.01\%$). The statistically significant highest Sulfur content was recorded from Tori-7 ($0.43 \pm 0.01\%$) and the lowest Sulfur content was observed BARI Sarisha-15 ($0.33 \pm 0.01\%$). Significantly highest amount of Phosphorus (P) content was observed in BARI Sarisha-17 ($1.09 \pm 0.03\%$) and the lowest amount of phosphorus content was obtained from BARI Sarisha-15 ($0.70 \pm 0.01\%$). The statistically significant highest Potassium content was recorded from BARI Sarisha-14 ($0.90 \pm 0.01\%$) and the lowest Potassium content was observed from BARI Sarisha-17 ($0.48 \pm 0.02\%$). Significantly highest amount of Cu contained observed in BARI Sarisha-14 ($13.00 \pm 0.86\text{ppm}$), and the lowest amount observed in Tori-7 ($8.25 \pm 0.16\text{ppm}$). Significantly highest amount of Fe contained was observed in Tori-7 ($248.25 \pm 3.25\text{ppm}$). The variety Sampad showed lowest amount of Fe ($69.50 \pm 3.10\text{ppm}$). Significantly highest amount of Mn contained was found in BARI Sarisha-14 ($31.25 \pm 3.25\text{ppm}$), while the lowest amount was found in BARI Sarisha-15 ($26.25 \pm 3.25\text{ppm}$). Significantly highest amount of zinc was found in Tori-7 ($48.37 \pm 1.8\text{ppm}$) and the lowest amount of zinc was found in BARI Sarisha-15 ($14.21 \pm 1.05\text{ppm}$). The highest amount of gross energy found from Sampad ($553.51 \pm 5.20\text{kcal/g}$); while lowest amount of gross energy recorded from BARI Sarisha-14 ($511.80 \pm 4.87\text{kcal/g}$).

From my experiment, it was observed that none of the released cultivars of rapeseed and mustard performed the best by all nutrient parameters.

But the Sampad performed the best considering the oil contents and gross energy. In case of fatty acid composition, BARI Sarisha-15 performed good results and contained highest oleic acid fraction. BARI Sarisha-14 showed the good performance for the most mineral contents. BARI Sarisha-15 contained highest amount of protein, carbohydrate, saponification value, iodine value and total unsaturated fatty acid. Based on the information mentioned above, it may be concluded that, BARI Sarisha-15 with appropriate qualitative and quantitative properties in their seed oil contents, can be grown in large scale as they contained the highest amount of different nutrient contents.

RECOMMENDATION

- From the experiment we can recommend that BARI Sarisha-15 is the best.
- Further analysis of different mustard variety should be done to know their nutrient content.
- Nutritional analysis is also important for breeders to evolve more nutrients rich mustard variety.
- Chemical composition and nutritional traits suggests the future strategy for the nutritionist, health advisors and dieticians as to how to make best use of the rapeseed and mustard.

CHAPTER VI

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APPENDICES

Appendix I. The chemical constant (Saponification value, Iodine value and Acid value) of oil of the different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Iodine value	Saponification value	Acid value
		Mean \pm SD (n=3)	
Sampad	98.09 ^e \pm 0.77	161.34 ^b \pm 1.00	1.72 ^a \pm 0.05
Tori-7	99.74 ^d \pm 0.35	166.77 ^a \pm 0.51	1.27 ^{cd} \pm 0.03
BARI Sarisha-17	101.20 ^c \pm 0.38	166.16 ^a \pm 2.37	1.60 ^b \pm 0.04
BARI Sarisha-14	103.10 ^b \pm 0.16	162.52 ^b \pm 0.54	1.35 ^c \pm 0.04
BARI Sarisha-15	106.27 ^a \pm 1.03	168.25 ^a \pm 0.15	1.20 ^d \pm 0.02
LSD (0.05)	1.1433	2.1874	0.0738
CV (%)	0.62	0.73	2.83

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

Appendix II. Percentage of total saturated and unsaturated fatty acid of oil of the different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Total saturated fatty acid(TSFA) %	Total unsaturated fatty acid(TUFA) %
	Mean \pm SD (n=3)	
Sampad	8.74 ^d \pm 0.09	85.27 ^e \pm 0.03
Tori-7	8.75 ^d \pm 0.12	88.05 ^c \pm 0.04
BARI Sarisha-17	9.38 ^c \pm 0.08	88.72 ^b \pm 0.07
BARI Sarisha-14	11.30 ^a \pm 0.19	86.67 ^d \pm 0.10
BARI Sarisha-15	10.12 ^b \pm 0.11	89.11 ^a \pm 0.03
LSD (0.05)	0.2355	0.1145
CV (%)	1.34	0.07

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

Appendix III. Proximate analysis of major minerals (Potassium and phosphorous) content of different released cultivars of rapeseed and mustard (*Brassica* spp.) oil cake

Name of the released cultivars (Treatments)	P%	K%
	Mean \pm SD (n=3)	
Sampad	0.87 ^c \pm 0.02	0.78 ^c \pm 0.01
Tori-7	0.99 ^b \pm 0.01	0.84 ^b \pm 0.03
BARI Sarisha-17	1.09 ^a \pm 0.03	0.48 ^d \pm 0.02
BARI Sarisha-14	1.02 ^b \pm 0.01	0.90 ^a \pm 0.01
BARI Sarisha-15	0.70 ^d \pm 0.01	0.78 ^c \pm 0.01
LSD (0.05)	0.0325	0.0325
CV (%)	1.92	2.37

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.

Appendix IV. Gross energy of the different released cultivars of rapeseed and mustard (*Brassica* spp.)

Name of the released cultivars (Treatments)	Gross energy
	Mean \pm SD (n=3)
Sampad	553.51 ^a \pm 5.20
Tori-7	530.86 ^b \pm 3.37
BARI Sarisha-17	533.61 ^b \pm 5.00
BARI Sarisha-14	511.80 ^d \pm 4.87
BARI Sarisha-15	520.71 ^c \pm 4.87
LSD (0.05)	8.5744
CV (%)	0.89

In a column, means with a similar letter (s) are not significantly different and those having a dissimilar letter (s) are significantly different by LSD at 5% level of significance. Where SD = standard deviation; CV = Coefficient of Variation; LSD = Least significant difference were determined.