CHARACTERIZATION OF *ZEA MAYS* L. THROUGH MORPHOLOGICAL, BIOCHEMICAL AND MOLECULAR MARKERS

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Abstract. 18 open-pollinated maize accessions were assessed for variability under randomized complete block design with 3 replications. Each plot comprised of two rows each 5 m in length with a plant to plant distance of 30 cm and a row to row distance of 75 cm. Data on morpho-biochemical traits were collected and subjected to statistical analysis to find out the variability among the maize accessions and association of increasing protein and oil content with the agronomic traits. From the source population 5 varieties with a high percentage of oil and protein, were selected and analyzed for total seed protein profile using Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis. Significant results of variance and covariance were observed for all morpho-biochemical traits, which revealed significant variation among the maize accessions. SDS-PAGE showed considerable variation in protein banding among the maize accessions. Correlation results showed an independent association of morphological traits with oil and protein contents in almost all the source population. This indicated that oil and protein content of maize grain can be increased without negatively affecting the important agronomic traits. Based on the results, the variability could be utilized to develop commercial maize varieties with improved quality of grain utilizing the conventional techniques of selection and hybridization.
Keywords: *biodiversity, correlation, oil content, protein content, SDS-PAGE*

Introduction

Maize is the 3rd most important crop in Pakistan after wheat and rice with an average yield of 4.3 ton ha^{-1} (Farooq, 2013) below the worldwide maize yield of 5.2 ton ha^{-1} (FAOSTAT, 2012). Therefore, it is very important to screen the new indigenous and exotic lines with an improved yield to be used in maize breeding programs (Iqbal et al., 2015). Genetic diversity is a basic tool for crop improvement (Iqbal et al., 2014) which could be managed, maintained and explained by morphological, biochemical and molecular markers (Vojka et al., 2016; Iqbal et al., 2015).

Pakistan imports 75% of edible oil (Zaidi, 2014) and could save billions by promoting local edible oil. Besides carbohydrates corn grain also has a considerable percentage of oil (3.77 to 11.56%) and protein (5.7 to 11.60%) (Deif et al., 2012; Singh et al., 2005; Mittelmann et al., 2003; Okporie et al., 2002) and annually gives almost 15% of the proteins obtained from food-crops throughout the world (Li and Vassal, 2004). Oil content and protein content of maize grain are positively associated with yield and its components (Sreckov, 2011; Obi and Onysishi, 1994), therefore, could be increased without adversely affecting yield (Okporie et al., 2007). Oil and protein of corn grain can be increased by different breeding techniques, e.g. recurrent selection method, which has been one of the most effective and successful methods used in breeding programs for maize improvement (Berner, et al., 2006; Uguru, 2005; Obi, 1991).

Many studies investigated variability among maize germplasm through morphological (Kariuki et al., 2016; Iqbal et al., 2015; Shrestha, 2014) and biochemical markers (Deif et al., 2012; Mittelmann et al., 2003). These experiments determined variability through morphological and biochemical characterization of maize varieties under RCBD with 3 replications and reported a wide range of variation for different morpho-biochemical characters investigated. Vojka et al. (2016) used morphological and molecular markers to study the variation among 29 inbred lines and reported that morphological and molecular markers are equally effective and accurate practices to determine the pedigree information of tested maize lines.

Other studies (Mittelmann et al., 2003) evaluated oil content, protein content and grain yield in a group of maize test-crosses under RCBD with three replications. Results showed week association and significant variation for oil content, protein content and grain yield. Deif et al. (2012) set an experiment to study the variability of 14 maize inbred lines and 15 crosses for oil and protein content. Results showed significant differences for protein content and oil content ranging from 8.34% to 11.60% and 7.67 to 11.56%, respectively. On the other hand, in crosses protein and oil content ranges from 8.22% to 13.94% and 9.27% to 11.29%, respectively. The highest values for protein content (13.94%), oil content (10.65%) and grain yield per plant (128.73 g) were reported by a cross.

Previous studies suggested that plant height is strongly associated with maize yield (Hegy et al., 2002; Sangoi et al., 1998; Aldrich et al., 1986), while 100-kernel weight and ear length predict yield components, whereas days to 50% silking predicts the maturity time of maize plants (Okporie et al., 2007). Furthermore, maize forage yield is preferred by farmers because it contains sufficient quantities of protein and minerals and possesses high digestibility as compared to other non-legume fodders (Chaudhary et al., 2014). Therefore, Plant height, grain yield, ear length, days to 50% silking and dry forage yield are among the most important agronomic traits to be assessed and should not be adversely affected in the maize breeding programs. Okporie et al. (2007) investigated 8 maize varieties to find out the correlation of increasing oil and protein content with five important agronomic parameters viz., plant height, ear height, 100-kernel weight, kernel density and days to 50% silking, under RCBD with 3 replications. They reported an independent association among the traits investigated and concluded that oil and protein content of maize can be increased without adversely affecting these agronomic traits.

Pakistan is not only facing the problem of having low maize yield ha^{-1} but also the poor quality of corn grain. Therefore, to achieve the goal efforts need to develop high

yielding maize varieties with an improved percentage of oil and protein which can be obtained by the determination of variability among the local and exotic germplasm to be used in future breeding programs.

The objectives of this study are: (1) to determine variation among selected maize varieties through morphological, biochemical and molecular markers; (2) to determine the association of oil and protein content with selected agronomic markers.

Materials and methods

The experimental material comprised of 10 open pollinated maize varieties viz., Sadaf, Islamabad Yellow, Agati-2002, SG-2002, Soan-3, Sahiwal-2002, Margala, Rakaposhi, Islamabad white and Kashmir Gold and 8 advance maize lines viz., BS-1, EV-7004 Q, EV-5098, EV-1097, BS-2, EV-6089, EV-6098, and EV-1098. The Seed of 8 open pollinated varieties and 9 advance maize lines were collected from National Agriculture Research Council (NARC) Islamabad and one local variety 'Kashmir Gold' was used as a check. The experiment was carried out using randomized complete block design with three replications at the research area of Faculty of Agriculture, Rawalakot, Department of Plant Breeding and Molecular Genetics. Each plot comprised of two rows each 5 m in length with a plant to plant distance of 20 cm and a row to row distance of 75 cm. All recommended agronomic practices were followed to raise an ideal crop. Data on 5 competitive plants of the selected one-meter square area from each plot was recorded on maturity. Nitrogen and phosphorus fertilizers were applied @ 60 and 90 kg ha^{-1}, respectively. Data was recorded before and after harvesting of the crop for five morphological traits viz., days to 50% silking, plant height, 1000-grain weight, ear length and dry forage yield and two biochemical traits viz., oil content and protein content. Morphological parameters were recorded by using the standard descriptors formulated by IBPGR (1980). The protein content was determined by the Micro-Kjeldahl method as described by Pearson (1976). While the oil content was obtained with the help of Soxhlet's extractor as described by Anon (1978).

From the source population two advance maize lines viz., EV-5098, EV-1097, EV-6089, two commercial varieties viz., Agaiti-2002, Rakaposhi and a Local Check (Kashmir Gold) with a high percentage of oil and protein were selected and analyzed for total seed protein profile using Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis (SDS-PAGE) as described by Laemmli (1970). The gel picture was taken, uploaded to ImageJ software and the distance covered by protein bands was calculated and scored by assigning + or – sign for the presence or absence of bands, respectively.

Data for agronomic and biochemical traits were submitted to analysis of variance using the GLM procedure of the SAS system v9.2 (SAS Institute, 2007) and the treatment means were compared by the Tukey's test at 5% level of significance. Correlation associations of oil content and protein content of maize kernel with plant height, days to silking, ear length, 1000-grain weight, and dry forage yield and correlation coefficients were calculated as given by Okpori (2006).

Results

The mean values for days to silking, plant height, ear length, 1000-grain weight, and dry forage yield are given in *Table 2*. Data for these morphological characters were

analyzed statistically and genotypic and phenotypic variance and covariance were estimated (*Table 1*). Significant differences were observed among maize accessions for days to silking, plant height, ear length, 1000-grain weight and dry forage weight (*Table 1*). The genotypic and phenotypic variances were significantly different for all the traits (*Table 1*). Genotypic and phenotypic variability values for days to silking (0.22% and 0.23%, respectively), plant height (1.4% and 1.4%, respectively), ear length (0.09% and 0.10%, respectively), 1000-grain weight (7.4% and 7.9%, respectively) and dry forage yield (281.25% and 326%, respectively) showed less environmental influence on these tested characters.

Table 1. *Estimates of parameters of variability for yield and its components*

Characters	Mean	Range	Genotypic variance	Phenotypic variance	GCV	PCV
Days to 50% silking	81.9	71.5-87.8	17.9	18.9	0.2	0.2
Plant height (cm)	199.9	170-241	274	278	1.4	1.4
Ear length (cm)	15.35	12.8-18	1.3	1.6	0.1	0.1
1000 grain weight (g)	232.8	180.83-345	1717	1839	7.4	7.9
Dry matter yield (kg ha^{-1})	12640.3	8564-16295	3555051	4120678	281.3	326.0
Oil content (%)	5.2	2.10 – 8.40	3.72	3.73	36.85	36.88
Protein content (%)	4.80	2.07 – 7.80	3.19	3.20	37.13	37.15

The minimum days to silking were taken by Rakaposhi with an average value of 73, whereas variety EV-1097 took maximum 86.9 days, similarly, Aguiti-2002 attained maximum plant height of 238.0 cm, whereas variety EV-1097 was short stature variety with 172.6 cm plant height. Agaiti-2002 and local check attained the maximum ear length with an average of 17.8 cm and 17.65 cm, respectively, and EV-7004Q attained minimum ear length of 13.10 cm. Maximum grain weight with an average value of 308.8 g was obtained from Agaiti-2002, whereas the minimum weight of 100-grains was presented by Sahiwal-2002 (181.1 g). EV-1098 produced maximum dry matter yield (16190 kg ha^{-1}), whereas the variety EV-7004Q yielded less dry matter ha^{-1} with an average value of 9342.0 kg.

The mean values for oil content and protein content are given in *Table 2*. Data for these biochemical characters were analyzed statistically and significant differences were observed among maize varieties for oil content and protein content and genotypic and phenotypic variance and covariance were estimated (*Table 1*) for oil content (36.85% and 36.88%, respectively) and protein content (37.13% and 37.15%, respectively). Results showed less environmental influence on the traits of oil content and protein content. Maximum values for oil content and protein content were obtained by EV-6089 (8.40%) and Agaiti-2002 (11.80%), respectively. Meanwhile, minimum values were obtained by EV-1098 (2.10%, 6.06%, respectively). The coefficient of variation (CV %) for days to silking (1.15), plant height (1.5), ear length (3.15), 1000-grain weight (4.53), dry forage yield (5.95) oil content (2.83) and protein content (1.78) indicates the excellent experimental precision.

From the photograph of the protein gel (photograph not presented) banding pattern drawn and the distance covered by protein bands measured (*Table 3*). The protein gel expressed nine bands at the marginally variable distance. At the distance of 1 cm (10 kDa) and 1.6 cm (15 kDa), all the varieties showed a common band. EV-6089, Rakaposhi and Local check (Kashmir Gold) recorded one band each at the distance of 2.7 cm (25 kDa). EV-6089 and Local check shared one common band at the distance of 3.1 cm (30 kDa) in

the protein profile. Agaiti-2002, EV-5098 and EV-1097 gave one band each travelled the distance of 3.4 cm (40 kDa). Rakaposhi had two isolated bands at the distance of 4 cm (70 kDa) and 6.4 cm (130 kDa). Similarly, EV-5098 expressed a solitary band at the distance of 4.8 cm (100 kDa) in the protein profile. EV-5098 and EV-1097 both had one band each at a distance of 7.3 cm (170 kDa).

Table 2. Mean values of different morphological traits of maize (Zea mays L.) varieties

SN	Variety	Days to silking	Plant height (cm)	Ear length (cm)	1000-grain weight (g)	Dry forage yield (g)	Protein content (%)	Oil content (%)
1	Rakaposhi	86.90 A	201.5 EF	14.85 BCDEF	206.5 CD	14010.0 ABCDE	10.11 C	3.10 EF
2	Islamabad white	86.70 AB	178.0 H	15.20 BCDEF	204.7 CD	14880.0 AB	7. 97 G	1.8 G
3	Sadaf	85.70 ABC	197.1 F	15.40 BCDE	234.0 C	10750.0 FGH	8.66 F	3.30 EF
4	Islamabad yellow	85.05 ABC	196.5 F	16.50 AB	214.3 CD	9884.0 GH	9.26 DE	4.50 C
5	EV-7004 Q	85.00 ABC	203.3 DE	13.10 G	274.5 AB	9342.0 H	8.10 G	4.00 D
6	BS-2	84.85 ABC	177.3 H	13.80 EFG	202.7 CD	12260.0 CDEFG	9.47 D	3.97 D
7	BS-1	84.45 ABCD	189.0 G	15.20 BCDEF	208.8 CD	12140.0 DEFG	6.47 I	1.40 GH
8	Local check (XY)	83.90 ABCD	219.9 B	17.65 A	298.8 AB	14200.0 ABCDE	11.27 B	6.20 AB
9	EV-6089	83.50 BCD	212.7 C	16.30 ABC	300.9 AB	11190.0 FGH	11.17 B	6.37 A
10	SG-2002	83.40 BCD	220.1 B	13.70 FG	278.8 AB	9984.0 GH	6.70 I	3.50 E
11	Sahiwal-2002	82.80 CDE	204.4 DE	16.45 AB	181.1 D	11040.0 FGH	10.20 C	0.47 I
12	Margala	82.65 CDE	188.3 G	14.85 BCDEF	198.5 CD	12700.0 BCDEF	6.17 J	1.17 H
13	EV-6098	81.30 DE	208.3 CD	14.50 DEFG	269.5 B	14440.0 ABCD	7.30 H	2.87 F
14	EV-5098	79.80 E	187.4 G	15.40 BCDE	208.4 CD	14700.0 ABC	10.10 C	3.97 D
15	EV-1097	75.00 F	172.6 H	15.85 BCD	186.0 D	11750.0 EFGH	9.17 E	4.80 C
16	EV-1098	75.00 F	199.6 EF	15.00 BCDEF	214.4 CD	16190.0 A	6.07 J	0.10 I
17	Agaiti-2002	74.80 F	238.0 A	17.75 A	308.3 A	14980.0 AB	11.76 A	5.80 B
18	Soan-3	73.95 F	204.9 DE	14.75 CDEF	200.4 CD	13090.0 BCDEF	8.70 F	3.00 H
	LSD$_{0.05}$	2.89	9.21	1.48	5.11	2311	0.26	0.45
	CV (%)	1.15	1.50	3.15	4.53	5.95	1.78	2.83

*Means with similar letters are statistically non-significant

Table 3. Tabulated representations of SDS-PAGE results of six maize varieties

Distance (cm)	kDa	Agaiti-02	EV-5098	EV-1097	EV-6089	Rakaposhi	Local check
1	10	+	+	+	+	+	+
1.6	15	+	+	+	+	+	+
2.7	25	-	-	-	+	+	+
3.1	30	-	-	-	+	-	+
3.4	40	+	+	+	-	-	-
4	70	-	-	-	-	+	-
4.8	100	-	+	-	-	-	-
6.4	130	-	-	-	-	+	-
7.3	170	-	+	+	-	-	-

+ Presence of protein band. - Absence of protein band

Almost all the maize varieties from the source population expressed negative or positive but non-significant correlation of oil and protein with agronomic traits viz., days to silking, plant height, ear length, 1000-grain weight and dry forage yield (*Table 4a* and *4b*). For some traits maize varieties viz., BS-2, Agaiti-2002 and EV-6089, Sahiwal-2002 for days to silking, EV-6089, Islamabad white, EV-1098, Rakaposhi,

Agaiti-2002, BS-2 and EV-6089 for plant height, Margala, Soan-3, EV-704 Q, SG-2002, Soan-3 and Kashmir Gold for ear length, Sadaf, EV-5098, Islamabad yellow, Agaiti-2002 and Sahiwal-2002 for 1000-grain weight, EV-7004 Q and Soan-3 for forage yield, showed positive or negative and significant results.

Table 4a. *Correlation (r) between oil content and five morphological traits*

Varieties	Dry matter	Ear length (cm)	Plant height (cm)	Days to silking	1000-grain weight
Sadaf	-0.705	-0.592	0.461	-0.165	-0.982*
BS-1	0.279	0.277	-0.277	0.867	0.287
Islamabad yellow	-0.260	-0.397	0.705	0.962	0.986*
Agaiti-2002	0.608	0.719	0.798	-0.984*	0.602
EV-7004 Q	1.000**	-0.348	0.721	0.327	0.146
EV-5098	-0.321	0.619	0.823	-0.741	-1.000*
EV-1097	-0.849	0.645	-0.549	0.902	-0.189
BS-2	-0.371	0.037	0.915	1.000**	0.655
Margala	0.191	0.973*	0.856	0.886	-0.756
EV-6089	-0.602	-0.786	0.975*	-0.990*	-0.735
Islamabad white	-0.710	-0.803	0.999**	0.560	-0.350
EV-6098	0.951	0.189	1.000**	0.911	-0.800
Rakaposhi	-0.716	-0.209	0.999**	-0.795	-0.867
SG-2002	0.405	0.803	-0.674	0.444	-0.884
EV-1098	-0.965	-0.866	-0.995**	0.972	0.822
Soan-3	-0.416	-0.998**	-0.873	0.406	-0.956
Sahiwal-2002	0.294	-0.397	-0.669	0.842	0.979*
Local check (Kashmir Gold)	-0.472	0.914	-0.511	0.812	-0.588

Table 4b. *Correlation (r) between protein content and morphological traits*

Varieties	Dry matter	Ear length	Plant height	Days to silking	1000-grain weight
Sadaf	-0.256	-0.916	0.843	0.350	-0.945
BS-1	-0.627	-0.629	0.629	0.905	0.949
Islamabad yellow	0.612	0.721	-0.376	-0.782	-0.846
Agaiti-2002	0.988*	0.041	0.993*	-0.836	-0.116
EV-7004 Q	-0.409	0.999**	0.350	0.737	-0.966
EV-5098	-0.659	0.371	-0.904	0.952	0.500
EV-1097	0.807	-0.700	0.485	-0.932	0.115
BS-2	-0.664	0.374	0.998**	0.931	0.359
Margala	0.477	0.998**	0.661	0.706	-0.918
EV-6089	0.481	0.866	-0.933	1.000**	0.824
Islamabad white	-0.954	-0.439	0.865	0.878	-0.742
EV-6098	-0.919	-0.277	-0.995**	-0.945	0.742
Rakaposhi	-0.519	-0.447	0.956	-0.922	-0.964
SG-2002	-0.343	0.992*	0.027	0.941	-0961
EV-1098	-0.641	0.091	-0.329	0.620	-0.172
Soan-3	-0.309	-0.985*	-0.923	0.298	-0.983*
Sahiwal-2002	-0.110	0.000	-0.319	0.987*	0.818
Local check (Kashmir Gold)	0.158	-0.997**	0.764	-0.576	0.820

Discussion

Morphological comparisons were made to know the extent of variation among maize varieties under investigation to estimate the genetic diversity. Statistical analysis of the data reflected a high level of variation for all the morphological traits. This is in agreement with the results reported by Hussain et al. (2011) who have screened the same maize varieties and reported the wide range of variation for all the morphological markers used and reported values ranged from 69 to 84 and 145 to 205 for days to silking and plant height, respectively. Present results for days to silking, plant height and ear length also showed resemblance with the findings of Koirala and Gurung (2002), who reported a significant amount of variability for days to silking, plant height and ear length. Koirala and Gurung (2002) recorded days to silking varied from 72 to 77 days, 198 to 230.0 cm plant height and a similar range of values for ear length. Similar findings were also reported by Borras et al. (2007), Baqa et al. (2014) and Ghimire and Timsina (2015) for days to silking. Present investigation showed that 1000-grain weight results were closer to the findings of Koirala and Gurung (2002) and Sabeena and Dar (2005), who reported a wide range of variability for 1000 grain weight, values ranged from 340 to 325 g. Similarly, significant variations for grain yield were reported by other investigations (Mubeen et al., 2015; Charles et al., 2013; Adebayo and Menkir, 2015). High variability for important agronomic traits, i.e. days to silk, plant height, ear length, 1000-kernel weight and grain yield in our investigation was also supported by the results of previous studies (Sonmez, 2018; Iqbal et al., 2015; Shrestha, 2013; Ranatunga et al., 2009; Miguel et al., 2008; Ihsan et al., 2005; Beyene et al., 2005; Dijak et al., 1999). Results for dry matter yield in this study do not resemble with the findings of Sabeena and Dar (2005), who reported the values ranged from 8150 to 12000 kg ha^{-1} for this trait. These findings also show deviation with the findings of Koirala and Gurung (2002), who observed the maximum dry forage yield of 14000 kg ha^{-1}. Genotypic and phenotypic variances are presented in *Table 1*. For all the traits studied, genotypic and phenotypic variances were significant, which is a piece of evidence that variability within the maize varieties is sufficiently divergent and constitutes potential candidate varieties on which improvement program can be initiated.

The values for oil content and protein content were significantly different and varied between 0.10% - 6.40% and 6.06% - 11.80%, respectively. The results were in clear agreement with the previous findings reported significantly different percentage of oil and protein values ranged from 3.77% to 11.56% and 5.7% to 12.08%, respectively (Sonmez, 2018; Deif et al., 2012; Seiam and Khalifa, 2007; Mittelmann et al., 2003; Singh et al., 2005). Varieties in the present study showed less percentage of oil content as compared to the previous studies. This difference may be due to the environmental factors and plant growth conditions which could modify the percentage of oil percentage (Villalobos et al., 1996).

Use of SDS-PAGE markers for the determination of variability and identification of varieties by extensive study of seed storage protein in many crop species became an efficient tool (Shah et al., 2003; Tawab, 2004; Anjali and Sanjay, 2012; Iqbal et al., 2014). In this study, the variability within selected varieties was confirmed by molecular investigations on the basis of gene product extracted from the seed of six maize accessions using SDS-PAGE markers. At the distance of 1 cm (10 kDa) and 1.6 cm (15 kDa) similar varieties could be an indication of their evolutionary interrelationship. EV-6089, Rakaposhi and Local check (Kashmir Gold) recorded one

band each at the distance of 2.7 cm (25 kDa) in the protein gel which indicated their similar genetic makeup. EV-6089 and Local check were different from the others as they shared one common band at the distance of 3.1 cm (30 kDa) in the protein profile. Agaiti-2002, EV-5098 and EV-1097 gave one band each travelled the distance of 3.4 cm (40 kDa) in the protein gel which showed their genetic resemblance as well as evidence that these varieties could have some common heritage. Rakaposhi had two isolated bands at the distance of 4 cm (70 kDa) and 6.4 cm (30 kDa) that showed Rakaposhi is genetically different from all other varieties studied so far. Similarly, EV-5098 expressed a solitary band at a distance of 4.8 cm (100 kDa) in the protein profile and showed that EV-5098 has a different genetic background as compared to other varieties studied so far. EV-5098 and EV-1097 both had one band each at a distance of 7.3 cm (170 kDa) which indicated that these maize accessions have some common genes and genetically they are different from other varieties. Similar results were reported in other investigations which exploited high level of genetic variability among studied germplasm and revealed that some bands are scored by all varieties and some bands shared by one or more than one variety (Khan et al., 2014; Osman et al., 2013; Verma, 2006; Laura, 2006; Haider, 2002; Tubajika, 2001; Gorinstein, 1999). In the present study, 22% of total visible protein bands were monomorphic and 78% were polymorphic. Results are in clear agreement with other findings (Vivodik et al., 2016; Iqbal et al., 2014; Rashed et al., 2010) which reported a wide range of variability among maize germplasm and recorded 31% to 39% monomorphic and 61% to 65% polymorphic protein bands.

The positive or negative and significant results of correlation suggest that an increase in oil content of maize can adversely affect the important agronomic traits. But these results are negligible as not even a single genotype has significant results for all the traits studied and only a few lines showed significant results for few traits. Since no significant relationship existed in the majority of remaining varieties of the source population, it indicates that high oil maize can be bred without adversely affecting the studied agronomic traits. Similar results were reported by Okporie and Oselebe (2007), Okporie and Obi (2002), Obi and Onish (1994) and Rajni et al. (1983). For 1000-grain weight, the present finding was contrary to the work reported by Okporie and Obi (2002) and Obi and Onish (1994), who found oil content to be positively and significantly correlated with 1000-grain weight.

Results of some morphological and biochemical traits deviate from the previous findings. This departure may be due to the environmental factors, variable sample size, and source population. For reliable detection of contrasted varieties usually experiments were performed in two or more consecutive years but the present experiment has not been repeated in consecutive years because Hussain et al. (2011) have previously evaluated the performance of the same varieties which have been screened in the present study and they also recorded significant differences for all the morphological traits studied and reported that these varieties had a wide genetic background, therefore, the results of the present investigation have sufficient and the real scientific interest.

Conclusion

In this study, morphological and biochemical markers confirmed a high level of variability among all the maize varieties investigated. Similarly, molecular markers (SDS-PAGE) also confirmed the wide range of variation among selected maize

varieties. The variability can be used in future breeding programs to improve maize varieties. Further, results showed non-significant association between oil and protein content with days to 50% silking, plant height, ear length, 1000-grain weight and dry forage yield, which indicated that oil and protein content that are among the most important and desirable traits of maize grain could be increased by using effective breeding methods without negatively affecting the important agronomic traits studied. Finally, recurrent selection and hybrid breeding might be recommended for developing high yielding maize varieties with the improved percentage of oil content and protein content.

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