

## Estimation of Protein, Carbohydrate, Starch and Oil Contents of Indigenous Maize (*Zea mays* L.) Germplasm

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### Abstract:

*Analysis of proteins, carbohydrates, starch and oil contents are very important for estimating quality of food. The present study was designed for the identification of promising maize germplasm with important traits for further maize crop improvement. The proximate analysis work was done in Department of Biochemistry, PMAS-Arid Agriculture University Rawalpindi, Pakistan. Twenty (20) accessions of Maize (*Zea mays* L.) belonging to different region of Pakistan were evaluated for diversity on the basis of protein, carbohydrate, starch and oil contents. In all accessions of maize carbohydrates were found*

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*in maximum and oil contents were found in minimum amount. The results showed that the protein contents were ranged from 10.82 to 13.59 %, carbohydrate contents were ranged from 63.03 to 69.36 %, starch contents were ranged from 60.38 to 66.31 % and oil contents were ranged from 4.16 to 5.76 %. These findings would be useful for the quality products form the maize.*

**Key words:** *Zea mays*, Carbohydrates, Oil contents, Proteins, Starch.

## **Introduction**

Maize (*Zea mays* L.) is the third most important cereal crop of the world (Sleper and Poehlman, 2006). In Pakistan it ranked after wheat and rice. It adds 0.5 percent to the GDP and 2.2 percent to the agriculture (Govt. of Pak., 2013). There are five species of genus *Zea* and only 1 specie “mays” is cultivated, remaining species are wild grasses. (Ellenskog-staam *et al.*, 2007)

Maize is consumed directly as food at different growth stages (baby corn to mature grain) (Aus. Gov., 2008). It provides bulk of raw materials for the livestock and many agro-allied industries in the world (Bello *et al.*, 2010; Randjelovic *et al.*, 2011). It contains Vitamins A, B, E and also some important nutrients for metabolism (Orhun, 2013).

In maize seed the carbohydrate is present in high amount than the other chemical components. In carbohydrate the amount of starch is 72 to 73 percent in the maize kernel and the amount of other carbohydrates such as glucose, sucrose and fructose are 1 to 3 percent in the maize kernel (FAO., 1993; Iken *et al.*, 2002; Barikmo *et al.*, 2004; Orhun, 2013). The second largest component in the maize kernel is protein which ranges from 8 to 11 percent (FAO., 1993; Iken *et al.*, 2002; Singh *et al.*, 2004; Orhun, 2013).The third largest component in the maize kernel is oil which ranges from 3 to 18 percent

(Alexander, 1971; FAO., 1993; Heiniger *et al.*, 2001; Chen, 2010; Orhun, 2013).

## Materials and Methods

In the present study twenty (20) accessions of maize (Table 1) were evaluated for protein, carbohydrate, starch and oil contents. Protein contents were estimated by Biuret method of Gornall *et al.* (1949); carbohydrates were estimated by phenol sulphuric acid method of Roberts *et al.* (2011), starch contents were identified by using iodine test of Sullivan, (1935) and total oil contents were determined by ether extract method of Ajayi *et al.* (2004).

**Table1: List of Maize Accession used in current study**

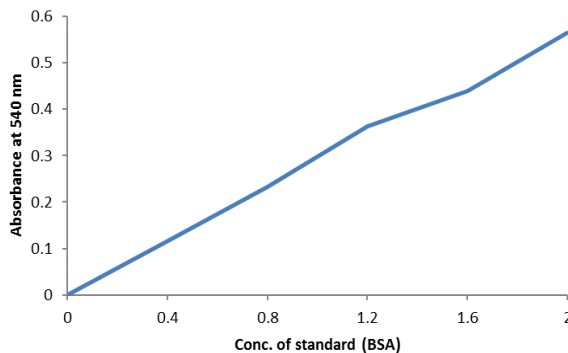
Sr. #	Accession #	Genus & Species	Origin	Province
1	014857	<i>Zea mays</i>	Pakistan	Punjab
2	014861	<i>Zea mays</i>	Pakistan	Punjab
3	014862	<i>Zea mays</i>	Pakistan	Punjab
4	014865	<i>Zea mays</i>	Pakistan	Punjab
5	014868	<i>Zea mays</i>	Pakistan	Punjab
6	014869	<i>Zea mays</i>	Pakistan	KPK
7	014870	<i>Zea mays</i>	Pakistan	Balochistan
8	014874	<i>Zea mays</i>	Pakistan	Balochistan
9	014878	<i>Zea mays</i>	Pakistan	Balochistan
10	014888	<i>Zea mays</i>	Pakistan	Balochistan
11	014919	<i>Zea mays</i>	Pakistan	Balochistan
12	014934	<i>Zea mays</i>	Pakistan	Sindh
13	014935	<i>Zea mays</i>	Pakistan	Sindh
14	014941	<i>Zea mays</i>	Pakistan	Sindh
15	014943	<i>Zea mays</i>	Pakistan	AJK
16	014944	<i>Zea mays</i>	Pakistan	AJK
17	014946	<i>Zea mays</i>	Pakistan	AJK
18	014950	<i>Zea mays</i>	Pakistan	AJK
19	014990	<i>Zea mays</i>	Pakistan	AJK
20	014998	<i>Zea mays</i>	Pakistan	AJK

## Estimation of protein contents

For protein estimation, seeds of maize were taken and ground in to powdered form. Protein was estimated according to Biuret method as described by Gornall *et al.*, 1949, using Bovine Serum Albumin as standard protein. Biuret reagent was prepared by mixing 0.3 g of  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ , 0.5g KI, 0.9g sodium potassium tartrate in distilled water up to 100 mL. Same concentration of reagent was mixed with standards as well as maize accessions OD was noted spectrophotometrically at 540 nm according to procedure. Linear regression equation (Eq.1 and 2) obtained from standard curve was used to find the concentration of proteins in maize accessions (Fig. 1).

$$Y = 0.112 X - 0.1057 \quad (\text{Eq.1})$$

$$X = (Y + 0.1057) / 0.112 \quad (\text{Eq.2})$$



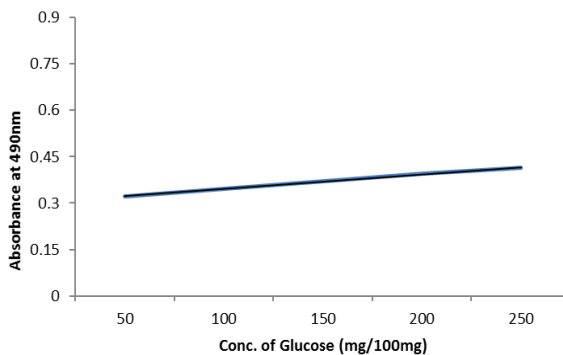
**Fig 1: Standard Curve for Protein Estimation**

## Estimation of carbohydrates

Phenol sulphuric acid method as describe by Roberts *et al.*, 2011 was used to estimate carbohydrates contents in selected maize accessions. Powder of each accession (0.1g) was mixed in 5mL of 2.5 N-HCl and heated in water bath for 3 hours. It was then neutralized by adding sodium carbonate and volume

increase up to 100 mL. Glucose was used as standard for carbohydrates estimation. Each experimental sample (standard and maize accessions) was mixed with 1 ml of 5% phenol solution and 5ml of 96% sulphuric acid solution and kept at 30 °C for 20min. After that absorbance was taken at 490nm and linear regression equation (Eq. 3) obtained from standard curve (Fig.2) was used to estimate carbohydrate in selected maize accessions.

$$X = (Y - 0.2981) / 0.0237 \quad (\text{Eq. 3})$$



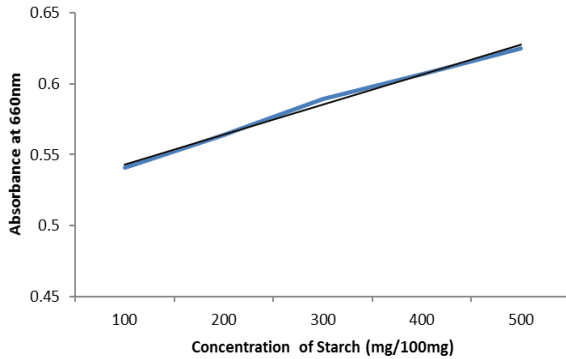
**Fig 2: Standard Curve of glucose for carbohydrate estimation**

### **Estimation of starch in Maize**

Starch was estimated by iodine test in selected maize accessions using glucose as standard by using procedure as describe by Sullivan in 1935. Each experimental sample including standard as well as maize accessions were mixed with 1 ml of iodine solution (4 g potassium iodide and 1.27 g iodine) for 10 min. The absorbance was noted at 660 nm with spectrophotometer. Linear regression equation (Eq.4 & 5) obtained from standard curve was used to estimate starch contents (Fig.3).

$$y = 0.0211x + 0.5219 \quad (\text{Eq. 4})$$

$$X = (Y - 0.5219) / 0.0211 \quad (\text{Eq. 5})$$



**Fig 3: Standard curve of glucose for starch estimation**

### **Estimation of oil by ether extract method**

Oil contents in each maize accession was determined by using Soxhlet apparatus according to method described by Ajayi *et al.*, 2004. 15 g of each accession was placed in thimble and placed in chamber which was 2/3 filled with n-hexane. After running 15-20 cycles of n-hexane, the oil extraction was stopped and oil and n-hexane were separated on rotary evaporator. The oil contents obtained were weight and percentage was found by using following formula.

$$\text{Oil Percentage (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Total weight of sample}} \times 100$$

### **Results and Discussions**

#### **Estimation of protein in Maize accessions of Pakistan**

The result shows that the protein contents ranged from 10.82% to 13.59% (Table 2). The maximum protein percentage (13.59%) were found in the accession 014943 belongs to the Azad Jammu and Kashmir and minimum protein percentage (10.82%) were

found in the accession 014868 belongs to the Punjab area. The result show similarity with the results of Orhun *et al.*,(2013); Sofi *et al.*,(2009) and FAO., (1993) in which the protein amount was ranged 8 to 11%.This result also show similarity with the pervious results of different scientists in which the minimum protein percentage in maize kernel was 5.7% and the maximum protein percentage in maize kernel was 15.8% ( Singh *et al.*, 2004; Iken *et al.*, 2002; Shukla *et al.*, 2001; Hogan *et al.*, 1955; Mitchell *et al.*, 1952). If the amount of protein is more than there is a chance that the amount of zein protein is increased and the amount of non-zein protein (lysine) is decreased Mitchell *et al.* (1952). In the present maize accessions on the basis of protein, diversity were present and this diversity will be helpful for further breeding programs.

### **Estimation of total carbohydrates in Maize accessions**

The results showed that the carbohydrate contents were ranged from 63.03% to 69.36% (Table 2). The maximum carbohydrate percentage (69.36 %) were found in the accession 014874 belongs to Balochistan and minimum carbohydrate percentage (63.03 %) were found in the accession 014878 also belongs to the Balochistan area. This result show similarity with the results of Iken *et al.* (2002) in which the carbohydrate amount was ranged 72 to 73%. In Iken *et al.* (2002) the amount of carbohydrate was more than our result. The difference in the amount of carbohydrates was due to nitrogen application because if the nitrogen application is more than the amount of carbohydrates were decreased and if the application of nitrogen is less than the amount of carbohydrate increased (Singh *et al.*, 2004). On the basis of carbohydrate, diversity were present in the maize accessions and this diversity will be helpful for further breeding programs.

### **Estimation of starch contents in Maize accessions**

The results showed that the starch content was ranged from 60.38 to 66.31% (Table 2). The maximum starch percentage (66.31 %) were found in the accession 014944 belongs to the Azad Jammu and Kashmir and minimum carbohydrate percentage (60.38 %) were found in the accession 014919 belongs to the Balochistan area. The starch content in the maize kernel was reported as the major component in our results as it was reported by Orhun *et al.* (2013); Sofi *et al.* (2009) and FAO., (1993). They reported the starch amount in the ranged of 72 to 73%. There is little bit difference in the amount of starch in our results and in Orhun *et al.* (2013); Sofi *et al.* (2009) and FAO., (1993) results. That difference in the amount of starch contents was due to nitrogen application. When the amount of nitrogen application was increased then the starch content was decreased and when the nitrogen application was decreased then the starch content was increased (Singh *et al.*, 2004). The diversity in the present accessions of maize on the basis of starch were present and this diversity will be helpful for further breeding programs.

### **Estimation of oil contents in Maize accessions**

The results showed that the oil content was ranged from 4.16 % to 5.76 % (Table 2). The maximum oil percentage (5.76 %) were found in the accession 014861 belongs to the Punjab and minimum oil percentage (4.16 %) were found in the accession 014865 also belongs to the Punjab area. The result shows similarity with the results of Orhun *et al.* (2013); Chen, (2010); Heiniger *et al.* (2001); FAO., (1993) and Alexander, (1971). In the present maize accessions on the basis of oil, diversity were present and this diversity will be helpful for further breeding programs.



**Table 2: Estimation of Protein, Carbohydrate, Strach and Oil contents in selected Maize accessions.**

Sr. #	Accession #	Protein (mg/ml)	Carbohydrate (%)	Starch (%)	Oil (%)
1	014857	12.22	66.07	62.37	5.17
2	014861	11.15	66.37	62.56	5.76
3	014862	10.98	66.78	63.23	5.21
4	014865	11.39	67.21	64.46	4.16
5	014868	10.82	69.27	64.17	5.17
6	014869	12.68	65.90	61.24	4.81
7	014870	11.81	65.90	63.18	5.44
8	014874	11.76	69.36	65.83	5.21
9	014878	11.90	63.03	61.18	5.24
10	014888	10.84	67.16	64.32	5.17
11	014919	11.93	63.20	60.38	5.30
12	014934	11.84	67.63	64.51	5.37
13	014935	12.66	63.84	61.52	4.87
14	014941	12.69	63.49	61.71	5.54
15	014943	13.59	68.90	66.02	5.61
16	014944	12.92	68.73	66.31	5.67
17	014946	12.93	63.90	61.05	5.04
18	014950	12.60	66.45	62.23	5.15
19	014990	12.22	64.68	61.52	4.91
20	014998	12.92	64.51	62.23	5.16

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