Pak. J. Bot., 47(6): 2239-2246, 2015.

ASSESSMENT OF AMYLOSE AND AMYLOPECTIN VARIABILITY IN BARLEY GERMPLASM

BAKHT NISA MANGAN[1,2], CUI LICAO[1], LIU HUI[1], ABDUL WAHID BALOCH[2], MUHARAM ALI[2], MUHAMMAD SIDDIQUE LASHARI[2] AND SONG WEINING[1]*

[1]*State Key Laboratory of Crop Stress Biology in Arid Areas, College of Agronomy & Yangling Branch of China Wheat Improvement Center, Northwest A & F University, Yangling Shaanxi ,712100, China*
[2]*Department of Agronomy & Soil Science, Plant Breeding & Biotechnology, Sindh Agriculture University, Tandojam, Pakistan*
**Corresponding author e-mail: sweining2002@yahoo.com; Tel: +86-29-87082984; Cell: +86-13468930678*

Abstract

Barley grain is composed of carbohydrates, proteins, dietary fiber, vitamins and minerals. The present study was conducted to investigate the variability for amylose and amylopectin content in different accessions of wild and cultivated barley from different regions. Our results showed that apparent amylose content ranged from 14.1 to 35.8%, 5.7 to 26.8% and 13.9 to 36.2% for wild barley, barley landraces and varieties, respectively. The highest range for amylopectin content was observed in barley landraces from 33.7 to 81.7% with the highest mean average value and the lowest range from 39.9 to 63.7% with 52.5% mean average value was observed in wild barley for amylopectin content. Furthermore, we found that out of 157 accessions, 52 had an average content of amylose (20-30%), whereas six accessions had more than 30% of amylose content. Our results indicated that the wild barley and barley varieties had considerable variation for amylose and amylopectin ratio compared to barley landraces, which not only provided some useful information about the difference in the amount of amylose and amylopectin content among these barley accessions, but also offered some prospects of using selected germplasm for barley quality improvement in respect of preferred amylose and amylopectin content.

Key words: Amylose, Amylopectin, Barley.

Introduction

Cereal grains are composed of carbohydrates, proteins, vitamins, minerals and fats, which are required for good human growth and health. Barley was first assumed as human food but developed gradually into a feed, malting and brewing grain. Barley is most widely adapted cereal grain species with production from fertile to deserts than any other cereal crop. It is still a major food source for some cultures in Asia (Himalayan nations) and northern Africa like Morocco and Ethiopia (Newman & Newman, 2006).

Barely is considered as a nutritionally dense food, with low calories and it is not as starchy as the wheat and/or rice. It also contains the dietary fiber, iron, copper, manganese and selenium. Starch is found in the considerable amount in cereal and the most important polysaccharide for human diet, serving more than 70% of its dry weight. Starch is composed of two different glucan chains i.e., amylose and amylopectin. The differences between amylose and amylopectin depend on the number of sided branches and series of polymerization. Amylose contains low series of polymerization (< 104 units) with a linear chain of D-glucose, on the other hand amylopectin demonstrate with a great number of series for polymerization (105-106 units). The variation in amounts of amylose and amylopectin, are responsible for its unique physical and chemical properties with strong influences on functional properties of flour or semolina and on its specific uses in the food (Zeng *et al*., 1997; Yoo & Jane, 2002; Yuan *et al*., 1993; Kobayashi *et al*., 1986). Some starch physicochemical properties are very important for the end-use of product as its gelatinization; pasting and gelation depend on the ratio of amylose to amylopectin.

The products made from high amylose starch are described as environment-friendly because they will be almost completely degraded within a very short time. Numerous reports have shown that amylose helps to make lower blood glucose and insulin levels in humans by digesting more slowly as compare to amylopectin, therefore, need of next meal will be felt after long period (Heijnen *et al*., 1995; Holt & Miller, 1995). Increasing the amylose content of diet is thus likely to be beneficial for many members of society, particularly those with obesity or hyper insulinemia (Behall & Howe, 1995). Recent studies indicate that amylose is important in reducing the glycemic and insulin impact of foods (Behall & Scholfied, 2005) and in increasing the body's fat burning ability which may help to maintaining a healthy weight (Higgins *et al*., 2004).

Improvement of amylose content in barley requires a better understanding of genetic mechanism controlling the amylose metabolism. Amylose concentration in barley is controlled by amylose (*amo1*) and waxy (*wax*) genes. The single recessive gene *amo1* is responsible for amylose content of up to 45% (Schondelmaier *et al*., 1992; Swanston *et al*., 1995). The *amo1* gene is located on chromosome 1H (Schondelmaier *et al*., 1992) and the *wax* gene is located on chromosome 7H (Lundqvist *et al*., 1997). The interaction between these two genes results in different levels of amylose in grain. Different variation for amylase content has been found by different authors, Salomonsson & Sundberg (1994), and Bjorck *et al*. (1990) found normal barley starch with 25-30% amylose, the high with 35-40% amylose content and the waxy starch with 8-9% amylose content. Both the scientists studied six American and Swedish varieties. Variation in amylose content is affected by both genetic and environmental factors. Temperature is notably responsible

for to reducing the starch accumulation, smaller A- and B-granules, and in barley fewer B-granules lead to increase the total amylose content (Tester *et al*., 1991). Therefore mentioned synthesis rate of starch is also affected by temperature at different growth stages. The ratio of amylose and amylopectin in total starch increases by increasing the age of endosperm (Merritt & Walker, 1969; Kang *et al*., 1985).

In case of wheat, enzyme GBSSI (*Granule-bound starch synthase* I), also known as waxy protein, is responsible for amylose synthesis. Reduction in the content of amylose in starch has been associated with the lack of waxy protein(s). Different species of wheat respond differently for amylose content like 1.3 to 28.5% in *Triticum monococcum* (einkorn), 7.2 to 38.0% in *T. turgidum* (emmer) and 0.0 to 52.3% in *S.cereale* (rye), while in another study the *T. durum* and *T. polonicum* accumulate apparent amylose contents that are significantly greater than that of *T. dicoccum* (Rodriguze-Quijano *et al*., 2003; Ali *et al*., 1999).

Our present study aimed to conduct the surveys of amylose and amylopectin ratio in grain of barley species, with the objectives to analyze the variation of starch properties in barley and identify the most outstanding barley genotypes for amylose and amylopectin for further study.

Materials and Methods

Seed accession and experimental site: In this study, sixty wild barley accessions from Israel and Jordan, forty eight barley landraces from Jordan and forty nine barley varieties from different parts of world were evaluated to determine availability and variability of starch traits, such as, amylose and amylopectin. All materials were grown in different rows by hand drilling after conventional tillage operation, at the experimental site of Northwest A&F University, Yangling, China. Field management and timing of management practices including fertilization was generally followed by local commercial production practices. Field was irrigated equally with flooding irrigation system under managed system of irrigation and equally look after was done for weed management and disease control.

Flour sample preparation: Grain samples were processed after grinding by using Tekpa Laboratory milling system JFS-13A (with sieve 0.5 mm). The mill was cleaned between samples.

Amylose and amylopectin measurement methods: Nearly 100 mg were used to analyze apparent amylose and amylopectin ratio. The iodine-potassium iodide (I:KI) protocol was adapted for standard testing (Washington *et al.*, 2000). Iodine-potassium iodide staining was first reported for amylose measurements in potato (Hovenkamp-Hermelink *et al.*, 1988). All the tests were replicated at least twice. Absorbance were measured at 620 and 443nm for amylose 525 and 725nm for amylopectin by using Shimadzu UV-1800, Spectrophotomer, China, in order to estimate amylose and amylopectin content.

Statistical analysis: Starch properties were evaluated by analysis of variance (ANOVA) using the general linear model of the Statistical Analysis System (SAS Institute, Cary, NC). Multiple comparisons were made by least significant difference (LSD). The compression between population mean followed by student t-test at (<0.05%). Analysis of variance was employed to test the genetic diversity between accessions, using a nested block design model.

Results

A conventional Iodine-potassium Iodide (I:KI) method for the estimation of the apparent amylose and amylopectin content of barley accessions was adapted. Duplicate samples of each accession were evaluated for properties of Starch: amylose and amylopectin content and all of the lines analyzed are shown in Tables 1 and 2. Starch properties differences between certain lines were significant. As shown in Table 3, barley landraces showed a considerable high variation (CV, 38.5%) for apparent amylose content followed by wild barely and barley varieties 26.4 and 24.3%, respectively. It is noted that the mean and range of apparent amylose content in barley varieties and wild barley accessions was almost similar (Table 3). The range of wild barley was 14.1-35.8% with the mean value of 20.6% whereas, barley varieties ranged between 13.9-36.2% with the mean value of 21.3%. On the other hand, barley landraces ranged between 5.7-26.8% with the average value of 14.2% of amylose. Among the wild barley accessions, the wild barley accession Karak 2 Muth HS-27 from Jordan produced the lowest (14.1%) apparent amylose content with 53.7% amylopectin, while from TBBS population (Israel), the wild barley accession TBBS-54 produced the highest (35.8%) amount of apparent amylose content with 54% of amylopectin (Table 1).

Among the barley landraces, Jarash-11 had the lowest amylose content 5.7% with 53.7% amylopectin, while Karak Muth-12 produced the highest (26.8%) amount of apparent amylose with 61% of amylopectin. Among the forty barley varieties, the lowest 13.98% apparent amylose content was observed in variety Xiu 81-7 from China with 40.7% amylopectin whereas the variety Prohilise produced maximum (36.2%) of apparent amylose content with 43.2% amylopectin (Table 2). In the case of starch trait amylopectin, again landraces showed high coefficient of variance i.e., 19.1% as compared to wild barley and barley varieties (Table 3). Wild barley and barley varieties exhibited very close CV value, which is 11.9 and 12.7%, respectively. As shown in Table 3, the much different was not also found in range of wild barley (39.9-63.7%) and barley varieties (39.7 to 64.2%), the mean value of the both germplasm was also very close to each other i.e., 52.7 and 50.2% for amylopectin content, respectively. Barley landraces produced the highest average mean amount of 57.4% with range of 33.7-81.7% of amylopectin content as compared to other sets of barley. Mt Giloba3-13 from Israel produced the lowest (39.9%) of apparent amylopectin content while TBBS-75 from Israel showed best performance (63.7%) for amylopectin, however, both the highest and the lowest accessions similar had ratio for apparent amylose content which is 18.7 and 18.8%, respectively. Among the varieties, European variety Eu Optic produced maximum (64.2%) amount of amylopectin while Plana was with lowest (39.7%) amount, apparent amylose content in both the highest and the lowest varieties was 16.2 and 19.3%, respectively (Table 2).

Table 1. Mean values of apparent amylose and amylopectin content % of wild and landraces barley.

Accessions	Origin	Amylopectin %	Amylose %	Accessions	Origin	Amylopectin %	Amylose %
			Wild barley				
Mt Gilboa barley 3-3	Israel	52.7	15.8	Karak Faqo HS 3	Jordan	49.7	27.9
Mt Gilboa barley 3-4	Israel	58.8	19.0	Karak Faqo HS 8	Jordan	55.7	24.9
Mt Gilboa barley 3-9	Israel	42.8	17.2	Karak Faqo HS 13	Jordan	51.1	19.8
Mt Gilboa barley 3-12	Israel	60.0	18.9	Karak Faqo HS 18	Jordan	58.8	20.9
Mt Gilboa barley 3-13	Israel	39.9	18.7	Karak Faqo HS 23	Jordan	48.5	20.8
Mt Gilboa barley 3-19	Israel	44.9	17.2	Karak Faqo HS 30	Jordan	57.8	30.2
Mt Gilboa barley 3-22	Israel	56.7	16.4	Karak Faqo HS 33	Jordan	54.7	30.4
Mt Gilboa barley 3-25	Israel	61.2	26.5	Karak Faqo HS 36	Jordan	60.3	29.0
Mt Gilboa barley 3-26	Israel	58.1	26.1	Karak Faqo HS 38	Jordan	44.7	24.7
Mt Gilboa barley 3-27	Israel	52.1	19.2	Karak 2 Mutah HS 4	Jordan	52.2	25.9
Mt Gilboa barley 3-34	Israel	60.1	15.5	Karak 2 Mutah HS 7	Jordan	63.2	29.1
Mt Gilboa barley 3-37	Israel	51.2	17.1	Karak 2 Mutah HS 8	Jordan	58.5	16.9
Mahola 22-5	Israel	57.9	18.7	Karak 2 Mutah HS 9	Jordan	43.5	15.3
Mahola 22-15	Israel	53.7	17.2	Karak 2 Mutah HS 10	Jordan	54.6	15.0
Mahola 22-16	Israel	50.1	16.4	Karak 2 Mutah HS 12	Jordan	45.0	15.2
Mahola 22-18	Israel	55.8	28.2	Karak 2 Mutah HS 13	Jordan	39.7	19.8
Mahola 22-20	Israel	42.8	29.1	Karak 2 Mutah HS 14	Jordan	54.1	16.2
Mahola 22-22	Israel	62.3	26.0	Karak 2 Mutah HS 22	Jordan	45.9	18.0
Mahola 22-23	Israel	51.6	22.0	Karak 2 Mutah HS 24	Jordan	49.9	15.5
Mahola 22-24	Israel	56.7	19.3	Karak 2 Mutah HS 27	Jordan	53.7	14.1
Mahola 22-25	Israel	58.7	19.1	Karak 2 Mutah HS 28	Jordan	48.8	15.8
Mahola 22-28	Israel	60.5	15.5	Iribid sal Hs 1	Jordan	57.0	16.0
TBBS 54	Israel	54.8	35.8	Iribid sal Hs 4	Jordan	49.9	16.0
TBBS 55	Israel	55.1	29.1	Iribid sal Hs 5	Jordan	42.0	15.9
TBBS 56	Israel	56.0	26.7	Iribid sal Hs 10	Jordan	44.7	14.3
TBBS 57	Israel	55.5	31.2	Iribid sal Hs 18	Jordan	46.9	17.9
TBBS 65	Israel	40.0	25.7	Iribid sal Hs 19	Jordan	48.2	18.0
TBBS 73	Israel	49.6	19.2	Iribid sal Hs 21	Jordan	48.8	16.6
TBBS 74	Israel	57.8	19.1	Iribid sal Hs 23	Jordan	50.8	16.04
TBBS 75	Israel	63.7	18.8	Iribid sal Hs 25	Jordan	48.1	14.6
			Barley landraces				
Aman-20	Jordan	45.7	19.9	Karak faqo-3	Jordan	65.7	12.7
Aman-21	Jordan	53.0	21.4	Karak faqo-4	Jordan	70.0	13.8
Aman-22	Jordan	44.5	20.0	Karak faqo-9	Jordan	36.2	8.7
Aman-23	Jordan	47.7	23.1	Karak faqo-10	Jordan	54.0	7.42
Aman-24	Jordan	49.5	18.8	Karak faqo-19	Jordan	71.2	11.2
Aman-25	Jordan	58.0	17.7	Karak faqo-26	Jordan	33.7	8.7
Aman-26	Jordan	55.5	19.8	Karak faqo-29	Jordan	81.7	12.1
Aman-31	Jordan	45.0	21.3	Karak faqo-30	Jordan	50.7	8.0
Jarash-3	Jordan	42.0	17.9	Karak Muth-3	Jordan	71.5	22.5
Jarash-4	Jordan	67.2	11.6	Karak Muth-4	Jordan	68.0	22.9
Jarash-5	Jordan	43.2	9.8	Karak Muth-10	Jordan	60.0	24.5
Jarash-6	Jordan	56.5	8.4	Karak Muth-12	Jordan	61.0	26.8
Jarash-10	Jordan	42.0	16.4	Karak Muth-13	Jordan	59.5	20.9
Jarash-11	Jordan	53.7	5.7	Karak Muth-15	Jordan	68.7	11.0
Jarash-12	Jordan	54.7	7.89	Karak Muth-16	Jordan	72.2	20.2
Jarash-13	Jordan	55.7	11.9	Karak Muth-17	Jordan	72.0	19.1
Shoubak Ghair HS 6	Jordan	52.2	9.9	Irbid sal-1	Jordan	59.5	10.7
Shoubak Ghair HS 7	Jordan	59.0	13.3	Irbid sal-3	Jordan	56.5	9.1
Shoubak Ghair HS 9	Jordan	72.5	12.7	Irbid sal-4	Jordan	59.5	12.2
Shoubak Ghair HS 10	Jordan	59.5	9.9	Irbid sal-5	Jordan	76.0	13.8
Shoubak Ghair HS 11	Jordan	57.2	6.1	Irbid sal-7	Jordan	45.2	13.3
Shoubak Ghair HS 12	Jordan	59.7	12.2	Irbid sal-8	Jordan	49.5	13.0
Shoubak Ghair HS 16	Jordan	70.2	9.4	Irbid sal-11	Jordan	52.2	13.1
Shoubak Ghair HS 19	Jordan	68.5	8.5	Irbid sal-12	Jordan	49.7	11.6

Table 2. Mean values of apparent amylose and amylopectin content% of barley varieties

Accessions	Origin	Amylopectin %	Amylose %	Accessions	Origin	Amylopectin %	Amylose %
Stopetoe	USA	45.50	26.08	Eu century	Europe	51.50	24.76
Morex	USA	55.75	25.20	Prohilise	unknown	43.25	36.27
Harrington	Canada	44.50	27.71	EuRolfi	Europe	54.75	24.71
Schooners	Australia	51.75	16.75	Alexis	Europe	61.00	18.85
Stirling	Australia	44.00	16.19	Farm Vug ton	USA	42.75	19.82
CHOPAIS	Canada	42.50	17.20	Chariot	Europe	59.25	19.14
ColterUsa	USA	55.25	15.45	Nikingett	Europe	56.50	18.83
Eu Inari	Europe	56.75	18.02	Noga	Norway	51.50	19.24
Eu Optic	Europe	64.25	16.12	IONA	unknown	47.75	21.61
Clipper	Australia	57.75	18.67	Mona	Israel	46.25	25.65
Garent	Australia	48.00	21.22	Kino Nij07	unknown	55.50	17.58
Eubarke	Europe	42.50	23.90	Barbican	unknown	51.50	18.77
Gairdner	Australia	40.50	28.43	Bob	USA	49.00	15.47
Khrahya	Europe	49.25	21.59	Eu Annabel	Europe	46.75	16.74
B.Kapter	Europe	48.00	25.63	Atahualpa	ICARDA	51.50	15.44
Pallas	Sweden	45.25	24.45	Yunyin Barely 1	China	55.25	25.49
Sirivs	unknown	55.00	31.67	Yunyin Barley 5	China	48.50	28.70
B.Tallon	Austraila	63.00	15.41	Yong 257	China	45.50	17.19
Plana	unknown	39.75	19.35	Shanghai Barley	China	45.00	15.34
Prior	Europe	51.75	22.39	Xiuda 10	China	54.50	19.81
Triumph	Germany	45.75	18.77	Czekh Barley	China	57.00	16.87
Baronesse	USA	56.50	29.56	Ganpi 3	China	41.75	26.30
Ta pgolbori	Korea	48.75	31.82	Xiu 81-7	China	40.75	13.98
Korv	Europe	43.75	23.70	Nong 83-133	China	47.00	16.01
Khemus	Bulgaria	61.50	19.84				

Table 3. Amylose and amylopectin content% of wild barley (Israel and Jordan), barley landraces (Jordan) and barley varieties from China and different countries.

		Wild Barley	Barley Landraces	Barley Varieties
Samples		**60**	**48**	**49**
Origin		**Israel and Jordan**	**Jordan**	**China and Diff. Count.**
Amylose %	Mean ± SD	20.6 ± 5.4	14.2 ± 5.4	21.3 ± 5.2
	Range	14.1-35.8	5.7-26.8	13.9-36.2
	CV %	26.4	38.5	24.3
Amylopectin %	Mean ± SD	52.5 ± 6.2	57.4 ± 10.9	50.2 ± 6.4
	Range	39.9-63.7	33.7-81.7	39.7-64.2
	CV%	11.9	19.0	12.7

Variations among six wild barley populations from Israel and Jordan and landraces population from Jordan was observed for both characters of starch i.e. amylose and amylopectin and is presented in Tables 4 and 5. Our findings indicate that large variations exited between wild barley (TBBS, Mt Giloba, Mahola, Karak Faqo, Karak Muth, Irbid) and barley landraces (Aman, Jarash and Shoubak Ghair, Karak Faqo, Kark Muth, Irbid) populations. Highly significant variation at $p<0.001$ levels for apparent amylose content between six wild barley populations was observed, while no any significant difference was observed between wild barley populations for starch character amylopectin. In the case of variation within populations for amylose and amylopectin content, all populations of wild barley showed highly significant variation except Irbid population that showed only significant variation at 0.05 levels for amylose (Table 4). Variation within population of landrace (Aman, Jarash, Shoubak Ghair, Karak Faqo, Kark Muth, Irbid) from Jordan were highly significant at $p<0.001$ level for both amylose and amylopectin content, while variation between landrace population was highly significant for amylose and slightly significant for amylopectin at $p<0.001$ and $p<0.05$ levels, respectively (Table 5).

Table 3. Summary statistics of apparent amylose and amylopectin content % of various barley germplasm.

Samples		Wild barley	Barley landraces	Barley varieties
		60	48	49
Origin		Israel and Jordan	Jordan	China and Diff. Count.
Amylose %	Mean ± SD	20.6 ± 5.4	14.2 ± 5.4	21.3 ± 5.2
	Range	14.1-35.8	5.7-26.8	13.9-36.2
	CV %	26.4	38.5	24.3
Amylopectin %	Mean ± SD	52.5 ± 6.2	57.4 ± 10.9	50.2 ± 6.4
	Range	39.9-63.7	33.7-81.7	39.7-64.2
	CV %	11.9	19.0	12.7

Table 4. Variation between and within wild barley populations for amylose and amylopectin content%.

Populations	Amylose% and Amylopectin% Variation between population				Amylose% variation within populations	Amylopectin% variation within populations
	Amylose%		Amylopectin%		ANOVA	ANOVA
	Mean	ANOVA	Mean	ANOVA		
TBBS	25.6a	7.19***	54.0a	1.46NS	51.0***	56.6***
Mt Giloba	19.02b		53.2a		10.9***	31.2***
Mahola	25.4a		55.6a		42.3***	29.3***
KarakFaqo	21.2ab		53.5a		39.2***	30.0***
Karak 2 Muth	18.02b		50.7a		8.6***	33.5***
Iribid	16.1b		48.5a		4.3*	11.1***

Significant at 0.05 levels, ***Significant at 0.001 levels, NS = Non-significant

Table 5. Variation between and within populations of barley landraces for amylose and amylopectin content%.

Populations	Amylose% and Amylopectin% Variation between population				Amylose% variation within populations	Amylopectin% variation within populations
	Amylose		Amylopectin		ANOVA	ANOVA
	Mean	ANOVA	Mean	ANOVA		
KarakFaqo	11.2b	21.8***	57.9ab	3.32*	31.7***	355.4***
Karak 2 Muth	21.1a		66.6a		51.9***	31.8***
Iribid	12.1b		56.0ab		25.8***	180.3***
Aman	20.3a		49.8b		25.2***	13.9***
JArash	11.2b		51.9b		251.4***	130.2***
ShoubakGahir	10.2b		62.3ab		23.7***	98.1***

*Significant at 0.05 levels, ***Significant at 0.001 levels

Mean squares from one-way ANOVA showed highly significant differences in amylose and amylopectin content among six populations of wild and landraces barley from Israel and Jordan at $p \leq 0.001$ level (Fig. 1). As mentioned in Fig. 1(A), six populations of wild barley ranged from16-25% for mean of amylose, among these populations, TBBS and Mahola from Israel showed 25% average mean for amylose content followed by Karak Faqo (21%) from Jordan. For the trait of amylopectin, these populations ranged from 48 to 55% content, again Mahola from Israel had the highest (55%) average mean for amylopectin content followed by TBBS from Israel (54%). Variation for six populations of barley landraces are presented in Fig. 1(B) which ranged from 10 to 21%, Karak Muth population produced maximum (21%) mean range of amylose content followed by Aman (20%). With respect to amylopectin, again K. Muth produced the highest (66%) amount of amylopectin content followed by S. Ghair (62%). The mean range of six landraces populations were 49-66% Fig. 1(B).

Discussion

Apparent amylose and amylopectin ratio is an efficacious character for starch synthesis and its functionality (Yue *et al*., 1999; Sissons & Batey, 2003). According to the reports that enriched amylose food is responsible to increase resistance against diseases and it improves health (Samaan *et al*., 2006) by producing lower glycemic index (Soh *et al*., 2006; Bird *et al*., 2008). The variation and availability of amylose content in cereals depends on the genetic background and is influenced by the environment (Hallstrom *et al*., 2011). This investigation brings out a considerable number of accessions carrying sufficient amount of amylose and amylopectin content. There was a clear evidence for the functionality of major genes which promote amylose content in barley.

Out of 157 analyzed accessions, 52 accessions produced normal content of amylose (20–30%), while six accessions had more than 30% of apparent amylose content. According to the reports, the normal cereal contains 18-33% amylose and 72-82% amylopectin, respectively. We found no any

waxy or amylose free genotype among wild, landraces and barley varieties. Table 1 also describes the variation of apparent amylose and amylopectin content in 108 accessions of wild barley and barley landraces from Israel and Jordan, meanwhile Table 2 expresses variations of amylose and amylopectin content in 49 barley varieties from different countries. Among wild, landraces and barley varieties, the wild accessions from Israel performed better with a wide range and highest mean average of amylose as compared to those in the wild barley and barley landraces from Jordan. There were slight differences between wild barley and barley varieties for amylose content (Table 3). This comprehensive availability and variability of starch traits is due to a wide variability in genetics of accessions. Another hypothetical explanation for this variability can be that, in diploid species many genes are functional to promote the synthesis of amylose content. It has been proved that the gene *amo1* is authoritative in any barley accession containing amylose above 45% (Watanabe *et al*., 1998; Merritt, 1967). Along with genetic background, environmental factors also affect the amylose content (Bultosa, 2003); there is a clear evidence that higher temperature enhances total amylose content in a cultivar depending on its genetic background (Tester *et al*., 1991; Nakamura *et al*., 1993a; Mohammad *et al*., 1999; Asaoka *et al*., 1984; Asaoka *et al*., 1985; Asaoka *et al*., 1989). Similarly, seed size is also an important character which plays a vital role in amylose synthesis along with genetic background and environmental factors (Lu *et al*., 1996; Shi *et al*., 1994; Fergason *et al*., 1966; Bewley *et al*., 1985).

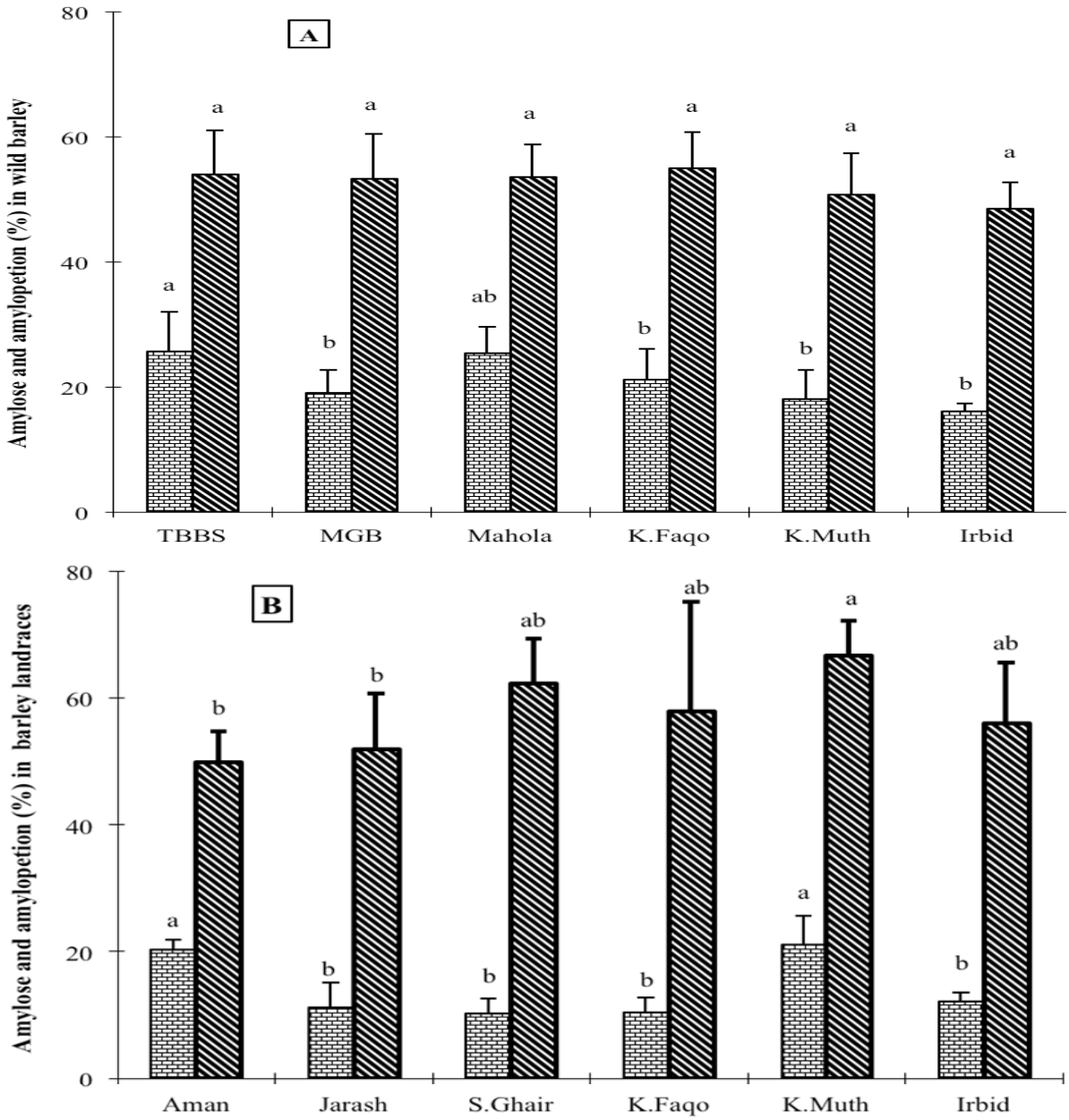

Fig. 1. Comparison of amylase % and amylopectin % in six populations of wild barley (A) and six populations of barley landraces (B). Different letters above bars indicates significant differences at 0.05 level by t-tests. Symbols and bars represent the mean ± SD (n=3).

It is concluded that the amylose and amylopectin ratio in different barley genotypes had only a marginal difference of variation. The present results have shown means and ranges of amylose and amylopectin content that were more or less as expected from the ploidy of the species and their relationships to each other. The ranges are broad enough that it is possible to increase them through a targeted breeding program.

Acknowledgement

The authors wish to express their thanks to the Chinese Ministry of Agriculture, Key Scientific and Technical Innovation, Chinese Ministry of Education (707054) and Natural Science Foundation of China (31271705) for their financial support.

References

Ali, M.K., L. Frederick, Stoddard and Donald and R. Marshall. 1999. Amylose Content in Segregating Populations of Einkorn, Emmer and Rye. *Starch/Stärke*. 51: Nr. 2-3: S. 66-73.

Asaoka, M., K. Okuno and H. Fuwa. 1985. Effect of environmental temperature at the milky stage on amylose content and fine structure of amylopectin of waxy and non waxy endosperm starches of rice (*Oryza sativa* L.). *Agric. & Biol. Chem*., 49: 373-379.

Asaoka, M., K. Okuno, K. Hara, M. Oba and H. Fuwa. 1989. Effects of environmental temperature at the early developmental stage of seeds on the characterization of endosperm starches of rice (*Oryza sativa* L.). *J. Japanese Soc. Starch Sci*., (Denpun Kagaku). 36: 1-8.

Asaoka, M., K. Okuno, T. Sugimoto, J. Kawakami and H. Fuwa. 1984. Effect of environmental temperature during development of rice plants on some properties of endosperm starch. *Starch*, 36: 189-193.

Behall, K.M. and D.J. Scholfield. 2005. Food amylose content affects postprandial glucose and insulin responses. *Cereal Chem*., 82: 654-659.

Behall, K.M. and J.C. Howe. 1995. Effect of long-term consumption of amylose vs amylopectin starch on metabolicvariables in human subjects. *Amer. J. Clin. Nutr*., 61: 334-340.

Bewley, J.D. and M. Black. 1985. Seeds: Physiology of Development and Germination. Plenum Press, New York.

Bird, A.R., M.S. Vuaran, R.A. King, M. Noakes, J. Keogh, M.K. Morell and D.L. Topping. 2008. Wholegrain foods made from a novel high-amylose barley variety (Himalaya 292) improve indices of bowel health in human subjects. *Brit. J. Nut*., 99: 1032-1040.

Bjorck, A., C. Eliasson, A. Drews, M. Gudmudsson and R. Karlsson. 1990. Some nutritional properties of starch and dietary fiber in barley genotypes containing different levels of amylose. *Cereal Chem*., 67(4): 327-333.

Bultosa, G. 2003. Physico-chemical and functional properties of grain tefEragrostistef (Zucc.) Totter] starch. A Ph.D. Thesis submitted to the Department of Food Science, Faculty of Natural and Agricultural Sciences, University of Pretoria. 157p.

Fergason, V.L., J.L. Helm and M.S. Zuber. 1966. Effect of kernel position on amylose starch content; distribution of amylose within corn endosperm (*Zea mays* L.). *Crop Sci*., 6: 273-275.

Hallstrom, E., F .Sestili, D. Lafiandra, I. Bjorck and E. Ostman. 2011. A novel wheat variety with elevated content of amylose increases resistant starch formation and may beneficially influence glycaemia in healthy subjects. *Food & Nut. Res*., 55: 70-74.

Heijnen, M.L.A., J.M.M. Van-Amelsvoort and J.A. Westrate. 1995. Interaction between physical structure and amylose: amylopectin ratio of food on postprandial glucose and insulin responses in healthy subjects. *Eur. J. Clin. Nut*., 49(6): 446-57.

Higgins, J.A., D.R. Higbee, W.T. Donahoo, I.L. Brown, M.L. Bell and D.H. Bessesen. 2004. Resistant starch consumption promotes lipid oxidation. *Nut. & Metabolism*, 1-8.

Holt, S.A. and J.B. Miller. 1995. Increased insulin responses to ingested foods are associated with lessened satiety. *Appetite*, 24:43-54.

Hovenkamp-Hermelink, J.H.M., J.N. DeVries, P. Adamse, E. Jacobsen, B. Witholt and W.J. Feenstra. 1988. Rapid estimation of the amylose/amylopectin ratio in small amounts of tuber and leaf tissue of the potato. *Potato Res*., 31: 241-246.

Kang, M.Y., Y. Sugimoto, S. Sakamoto and H. Fuwa. 1985. Developmental changes in the amylose content of endosperm starch of barley (*Hordeumvulgare* L.) during the grain filling period after anthesis. *Agric. & Biol. Chem*,, 49(12): 3463-3466.

Kobayashi, S., S.J. Schwartz and D.R.L. Back. 1986. Comparison of the structures of amylopectin from different wheat varieties. *Cereal Chem*., 63: 71-74.

Lu, T.J., J.L. Jane, P.L. Keeling and G.W. Singletary. 1996. Maize starch fine structure affected by ear developmental temperature. *Carbohydrate Res*., 282: 157-170.

Lundqvist, U., J.D. Franckowiak and T. Konishi. 1997. New and revised descriptions of barley genes. *Barley Genet. Newsletter*, 26: 22-516.

Merritt, N.R. 1967. A new strain of barley with starch of high amylose content. *J. Inst. Brewing*, 73: 583-585.

Merritt, N.R. and J.T. Walker. 1969. Development of starch and other components in normal and high amylose barley. *J. Inst. Brewing*, 75: 156-163.

Mohammad, K.A., F.L. Stoddard, D.R. Marshall, M.N. Uddin and X. Zao. 1999. Starch extraction and amylose analysis from half seeds. *Starch/Stärke*, 51: 62-66.

Nakamura, T., M. Yamamori, H. Hirano and S. Hidaka. 1993a. Identification of three Wx proteins in wheat (*Triticum aestivum* L.). *Bioch. Genet*., 31: 75-86.

Newman, C.W. and R.K. Newman. 2006. A brief history of barley foods. *Cereal Foods World*, 51: 4-7.

Rodr´ıguez-Quijano, M., R. Lucas and J.M. Carrillo. 2003. Waxy proteins and amylose content in tetraploid wheats *Triticumdicoccum* Schulb, *Triticum durum* L. and *Triticum polonicum* L. *Euphytica*, 134: 97-101.

Salomonsson, A.C. and B. Sundberg. 1994. Amylose content and chain profile of amylopectin from normal, high amylose and waxy barleys. *Starch/Stiarke*, 46: 325-328.

Samaan, J., G.H. El-Khayat, F.A. Manthey, M.P. Fuller and C.S. Brennan. 2006. Durum wheat quality: II. The relationship of kernel physicochemical composition to semolina quality and end product utilization. *Inter. J. Food Sci. & Tech.*, 41: 47-55.

Schondelmaier, J. A. Jacobi, G. Fischbeck and A. Jahoor. 1992. Genetical studies on the mode on inheritance and localization of the amo1 (high amylose) gene in barley. *Pl. Breeding*, 109: 274-280.

Shi, Y.C., P.A. Seib and J.E. Bernardin. 1994. Effects of temperature during grain-filling on starches from six wheat cultivars. *Cereal Chem*., 71: 369-383.

Sissons, M.J. and I.L. Batey. 2003. Protein and starch properties of some tetraploid wheat. *Cereal Chem*., 80: 468-475.

Soh, H.N., M.J. Sissons and M.A. Turner. 2006. Effect of starch granule size distribution and elevated amylose content of durum dough rheology and spaghetti cooking quality. *Cereal Chem*., 83: 513-519.

Swanston, J.S., R.P. Ellis and J.R. Stark. 1995. Effects on grain and malting quality of genes altering barley starch composition. *J. Cereal Sci*., 22: 265-273.

Tester, R.F., J.B. South, W.R. Morrison and R.P. Ellis. 1991. The effects of ambient temperature during the grain-filling period on the composition and properties of starch from four barley genotypes. *J. Cereal Sci*., 13: 113-127.

Washington, J.M., A. Box, A. Karakousis and A.R. Barr. 2000. Developing waxy Wild barley for food, feed and malt. *Barley Genet*., 8: 303-306.

Watanabe, N., Y. Noda, N. Goto and H. Miura. 1998. Variation for apparent amylose content of endosperm starch in *Triticum durum* and *Aegilopssquarrosa*. *Euphytica*, 101: 283-286.

Yoo, S.H. and J.L. Jane. 2002. Structural and physical characteristics of waxy and other wheat starches. *Carbohydrate Polymers*, 49: 297-305.

Yuan, R.C., D.B. Thompson and C.D. Boyer. 1993. Fine structure of amylopectin in relation to gelatinization and retro gradation behavior of maize starches from three wax containing genotypes in two inbred lines. *Cereal Chem*., 70: 81-89.

Yue, P., P. Rayas-Duarte and E. Elias. 1999. Effect of drying temperature on physicochemical properties of starch isolated from pasta. *Cereal Chem*., 76(4):541-547.

Zeng, M., C.F. Morris, I.L. Batty and C.W. Wrigley. 1997. Sources of variation for starch gelatinization, pasting and gelation properties in wheat. *Cereal Chem*., 74: 63-71.

(Received for publication 26 March 2014)