

Investigation of the medium for barley (*Hordeum vulgare* L.) immature embryo culture

Abdullah Hassn MOHAMMED^{1,2}

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Abstract: Germination of immature embryos of interspecific crosses *in vitro* is the most important problem in barley breeding programs. The effect of four media types (MS, ½ MS, B-5, ½ B-5) on immature embryo culture of nine barley varieties (Sameer, Buraq, Aksad, Shuaa, Arevat, Alwarkaa, Alhadher, Amel, and Rihan) was studied by using completely randomized design (C.R.D.) with six replicates. Analysis of variance showed highly significant effects due to varieties, media, and interaction for all studied characteristics, indicated high variation among varieties and media. Aksad variety was significantly superior to all varieties for all studied traits except root number (RN). Medium of ½ B-5 was significantly superior to all media for all studied traits. Aksad interactions with media except B-5 media gave full germination with highly significant superiority over all interactions. Aksad x ½ B-5 interaction showed a highly significant superiority for shoot length (SL) and RL. As a result, ½ B-5 medium was the efficient medium for germination of immature embryos of most varieties. These varieties could be used in breeding program of interspecific crosses with wild species by using ½ B-5 medium.

Key words: barley varieties, MS medium, Gamborg's B-5 medium, final germination percentage, shoot length, root length, root number

Preučevanje gojišč za gojenje nezrelih embrijev ječmena (*Hordeum vulgare* L.)

Izvleček: Kalitev nezrelih embrijev *in vitro* je pri medvrstnih križanjih največji problem pri žlahtnjenju ječmena. Preučevan je bil učinek štirih vrst gojišč (MS, ½ MS, B-5, ½ B-5) na kulture nezrelih embrijev devetih sort ječmena (Sameer, Buraq, Aksad, Shuaa, Arevat, Alwarkaa, Alhadher, Amel, in Rihan) v popolnem naključnem poskusu s šestimi ponovitvami. Analiza variance je pokazala visoko značilne učinke sort, gojišč in njihovih interakcij za vse preučevane lastnosti, kar kaže na velike razlike med sortami in gojišči. Sorta Aksad je bila značilno superiorna napram vsem ostalim sortam za vse preučevane lastnosti, razen za število korenin. Gojišče ½ B-5 je bilo značilno najboljšo za vse preučevane lastnosti. Sorta Aksad je imela popolno kalitev na vseh gojiščih, razen na gojišču B-5. Sorta Aksad je imela na gojišču ½ B-5 značilno najdaljše poganjke in korenine. Izkazalo se je, da je gojišče ½ B-5 najbolj učinkovito za kalitev nezrelih embrijev večine obravnavanih sort. Te sorte bi lahko bile uporabljene v žlahtniteljskih programih za medvrstna križanja z divjimi vrstami ječmena z uporabo gojišča ½ B-5.

Ključne besede: sorte ječmena, MS gojišče, Gamborg's B-5 gojišče, odstotek končne kalitve, dolžina poganjka, dolžina korenine, število korenin

¹ Field Crops Department, College of Agriculture, Tikrit University, Tikrit, Iraq

² Corresponding author, e-mail: abdullah@tu.edu.iq

1 INTRODUCTION

Barley (*Hordeum vulgare* L.) is one of the most important cereal crops in the world. It is ranking as the fourth major crop in production and area next to wheat, rice and maize (Goedeke et al., 2007; Blake et al., 2011). Barley is one of the earliest, the most domesticated food crop that humans have known since prehistoric times. It was the main source for making bread, soup and porridge dishes in the countries of the ancient world. Currently, barley has many uses, whether in human nutrition or animal feed. Barley flour is used alone or mixed with wheat flour in the industry of various types of pastries, especially bread. Barley is also included in many food industries, such as baby food, breakfast cereals, biscuits, beer, other alcoholic and non-alcoholic beverages, malt syrup, malted milk, and chemical industries. Barley is also a rich source of vitamins, minerals and dietary fiber. Its high content of soluble dietary fiber has reinforced the importance of barley and its place as an important food ingredient. Many studies have shown that regular consumption of barley reduces the risk of certain diseases such as colon cancer, gallstones, high blood pressure and chronic heart disease (Idehen et al., 2017; Biel et al., 2020). In India, most of the barley produced (90 %) is used for human consumption. Barley is also used as animal feed. Its grain (rich in carbohydrates) and the resulting straw are used to feed animals. It is also grown in some countries for use as green fodder or as pasture for animals. Some countries, including Iraq, use almost 80 % of grown barley as fodder (FAO, 2009). The use of barley as feed has a special value by giving desirable parts of visceral fat (Kumar et al., 2012). Barley is also grown as a cover crop to prevent soil erosion (water and air erosion) and curb weeds (Mohammed, 2018). Abiotic and biotic stresses lead to a decrease in crop production and nutritional value of crop grains over the world. Drought, salinity, and disease are among the most important stresses from an economic point of view and the most important in biology (Hussain, 2006; Mohammed et al., 2021). Due to the limited genetic variance in the cultivated varieties of barley, the use of large genetic variance that available in the wild species is necessary in the breeding programs (Ellis et al., 2000; Sreenivasulu et al., 2008). Therefore, interspecific crosses between domestic barley and wild species such as *H. bulbosum* L. and *H. murinum* L. are a major source of genetic differences and gives plant breeders a great opportunity to recombine the chromosomes set in order to take advantage of the positive aspects found in several individuals and collect them in one organism (Araus et al., 2003; Nevo and Chen, 2010). Many researchers have pointed that the widespread of wild species of barley indicates the high potential for great genet-

ic diversity and adaptation to biotic and abiotic stresses that serve as an important genetic resource for breeding programs. Where the different genotypes of wild species constitute an important source for plant breeders in improving the adaptive traits that contribute to increasing and stabilizing production under conditions of biotic and abiotic stresses, and can be an entry point for improving varieties (Suprunova et al., 2007; Alassaf, 2018). However, interspecific crosses between domestic and wild species of barley has many sexual barriers such as very low seed set and abort endosperm due to sexual incompatibilities. Therefore, plant tissue culture plays an important role in solving these problems that faced by plant breeders in breeding programs to improve barley varieties (Houben et al., 2011). Immature embryo rescue is one of the most important applications of tissue culture to solve the problem of sexual incompatibilities. Embryo culture technology also has several uses such as producing rare species, haploid plants that use to produce homozygous diploid plants, testing seed viability, and breaking seed dormancy. In addition, embryo culture is used to produce large number of plants from one embryo by the callus method. However, mature embryos can grow in a simple nutrient medium, but immature embryos need a more complex nutrient medium. As a result, immature embryos need to be grown in efficient culture media to mature and give a complete and healthy natural plant (Burun and Poyrazoglu, 2002; Pickering and Johnston, 2005; Chahal and Gosal, 2006). Crosses also differ significantly in the type of medium that suitable for each cross (Mohammed et al., 2020). Therefore, there is a need to determine the most suitable culture media for the largest number of genotypes that provide the regular growth of embryo in order to obtain successful breeding programs to improve barley varieties.

2 MATERIALS AND METHODS

Experiments were carried out in the field and laboratory of College of Agriculture/ Tikrit University in order to study the effect of different types of media on immature seed germination of nine varieties of barley. Seed of varieties used in this study were obtained from Seed Technology Center/ Office of Agricultural Research/ Iraqi Ministry of Science and Technology. Barley varieties were: (Sameer, Buraq, Aksad, Shuaa, Arevat, Alwarkaa, Alhadher, Amel, Rihan). Seed of each of the nine varieties were planted in the field during the winter session of 2020/2021 to obtain immature embryos for use in the media assessment experiment. All crop service operations such as irrigation, fertilization and weed control were carried out according to researchers' recommendations

for the barley crop. Spikes from each variety were harvested 12-14 days after pollination which embryo length was approximately 1.5 mm. The harvested spikes (with peduncle) were placed vertically in a beaker containing a little amount of water which cover spike peduncle and then placed in the refrigerator until dissection (Devaux 2003). Seed were removed from spikes of each of the nine varieties then were sterilized in 70 % ethanol for a minute. After that seeds were disinfested with 1 % sodium hypochlorite solution (NaClO) for 6 minutes then rinsed three times with sterile distilled water then dried on sterile paper napkin. After that, immature embryos of each variety were dissected from their caryopses and placed in 100 ml jars (five embryos per jar) containing either full concentration of Murashige and Skoog medium (Sigma-Aldrich Co., M-5519, St. Louis, MO), half concentration of MS medium, full concentration of Gamborg B-5 medium (Sigma-Aldrich Co., G-5893, St. Louis, MO), or half concentration of Gamborg B-5 medium. All types of media were without plant growth regulators. Planted jars were labeled and wrapped with Parafilm^M PM-996 (Bemis Company Inc., Neenah, WI). Process was conducted under a laminar flow hood (*ENVIRCO, Environmental Air Control, Inc.*, Hatfield, PA) to control contamination. All types of media were supplemented with 0.8 % agar (Sigma-Aldrich Co., A-1296, St. Louis, MO); 3 % and 2 % sucrose (Fisher Scientific, S5-500, Fair Lawn, NJ) for MS and Gamborg B-5 media, respectively; pH of MS and Gamborg B-5 media was adjusted to 5.7 and 5.5, respectively by using 0.1 N NaOH (Burun and Poyrazoglu, 2002; Houben et al., 2011). Germination process was carried out in the growth chamber (Percival, Mod. 135LLVL, Controlled Environments, Boone, IA) in the darkness at a temperature of 25 °C ± 2 for ten days. The number of normal germinated embryos was only counted daily to determine final germination percentage by using the following equation described by Scott et al. (1984):

$$FG \% = \frac{\text{Final number of germinated embryos}}{\text{Total number of embryos planted}} \times 100$$

Table 1: Analysis of variance of nine varieties of barley (Sameer, Buraq, Aksad, Shuaa, Arevat, Alwarkaa, Alhadher, Amel, Rihan), four types of media (MS, ½ MS, B-5, ½ B-5), and their interaction for final germination percentage (FGP), shoot length (SL), root length (RL), and root number (RN)

Source of variation	Degrees of freedom	FGP	SL	RL	RN
Varieties (V)	8	***	***	***	***
Media (M)	3	***	***	***	***
V x M	24	***	***	***	***
Error	180				

*** Significant at the 0.001 probability level
NS, significance level $p > 0.05$

Comparisons were made among the types of media for final germination percentage, shoot length (cm), root length (cm), and root number for the nine varieties of barley. Experiment was arranged as a completely randomized design (C. R. D.) with six replications (jars).

Data were statistically analyzed as a CRD by using PROC MEANS and PROC GLM in SAS (Version 9.4, SAS Institute, 2011, Cary, NC). Significant mean separation among types of media, varieties, and the interactions was determined using Fisher's least significant difference (LSD) at $\alpha = 0.05$ and 0.01 . Data were graphed using SigmaPlot version 13 (Systat Software Inc., San Jose, CA).

3 RESULTS AND DISCUSSION

Analysis of variance showed highly significant differences among barley varieties, media types, and barley varieties x media types of interaction for all studied characteristics (Table 1).

The significant effects among barley varieties, and among media types indicated that there was a high genetic variation among varieties, and high variation among media. Furthermore, the significant interaction between varieties x media indicated that each variety responded to each medium type independently. Previous studies have also found significant differences among varieties, media types, and barley varieties x media interaction (Burun and Poyrazoglu, 2002; Hayes et al. 2003; Han et al. 2011; Houben et al. 2011; Kahani et al. 2012; Mohammed, 2018; Mohammed et al. 2020).

Germination of immature embryos is the most important problem that plant breeder faces in breeding program of interspecific crosses between domestic barley and wild barley to improve barley varieties. The final germination percentage (FGP) of immature embryos of nine barley varieties were evaluated based on four types of media (Table 2). The results showed a high significant differences among barley varieties for FGP trait. The

mean of FGP of immature embryos of barley varieties ranged from 36.11 to 99.17 % at four types of media. Aksad variety gave the highest FGP value (99.17 %) and was highly significant superior to all varieties; it followed by Arevat variety with 86.67 % of FGP. While Rihan variety had the lowest value of FGP (36.11 %). Table 2 also showed highly significant differences among media types for this trait and ranged from 64.33 to 77.69 % of immature embryos. The half concentration of Gamborg's B-5 medium (1/2 B-5) showed the highest value of FGP (77.69 %) and was significant superior on all media types; while the half concentration of Murashige and Skoog (1/2 MS) medium gave the lowest value (64.33 %).

Interaction between barley varieties x media types revealed highly significant difference for FGP trait. The value of FGP of combinations ranged from 23.33 to 100.00 % (Table 2). The interactions between Aksad variety with MS, ½ MS, and ½ B-5 media were superior by giving the full and highest value of FGP (100.00 %). While the interaction between (Rihan x ½ MS), (Rihan x B-5), and (Shuaa x ½ MS) showed the lowest average of FGP which amounted to 23.33 %, 26.67 %, and 28.33 %, respectively. These results confirmed the high significant differences among varieties as well as media types. Also, these data confirm that individual varieties of barley responded differently to each type of media. For example, Shuaa, Alwarkaa, Alhadher, and Rihan varieties gave the highest value of FGP of immature embryos when they cultured on ½ B-5 medium. While Buraq, Amel, and Arevat varieties had the highest value in ½ MS, B-5, and ½ MS & B-5 media, respectively. On the other hand, Arevat

variety was superior with complete FGP in MS, ½ MS, and ½ B-5 media. In conclusion, Aksad variety and half concentration of B-5 medium were the best for final germination trait. Many researchers have found that MS, ½ MS, B-5 and other media can be used for culturing immature and mature embryo. These results were confirmed by previous studies. Mihailescu and Giura (1996) reported that germination of barley embryo was better in B-5 medium than MS. While Burun and Poyrazoglu (2002) found that MS and ½ MS media were better than B-5 medium when they evaluated the FGP of Kaya 7794 variety on MS, ½ MS, B-5, and RC (Randolph and Cox) media. Han et al. (2011) reported that FGP of barley varieties in B-5 medium was higher by 10 % than MS medium. Also, Chen et al. (2011) reported that is difficult to draw a conclusion about the most suitable medium for different barley genotypes. Chen's result confirmed by Mohammed (2018) and Mohammed et al. (2020) found that each genotype of barley responded to each culture medium independently of germinated immature embryo of many crosses that obtained from interspecific crosses between domestic and wild barley. This result may be due to component of media, genotypes, embryo size, sucrose rate, and pH. As a result, these varieties could be used in breeding program of interspecific crosses with wild barley species by using the ½ B-5 media to improve some quality and quantity traits of barley varieties.

Table 3 showed the shoot length (SL) of germinated immature embryos of nine barley varieties that cultured at four types of media. The results showed highly significant differences among barley varieties for SL

Table 2: Mean of final germination percentage (FGP) of nine barley varieties in four types of media

Varieties of Barley	Types of Media				Mean (%)
	MS	½ MS	B-5	½ B-5	
Sameer	67.67	62.22	74.17	82.22	71.57
Buraq	62.98	74.60	56.11	70.63	66.08
Aksad	100.00	100.00	96.67	100.00	99.17
Shuaa	58.89	28.33	52.67	73.33	53.31
Arevat	87.78	93.33	93.33	72.22	86.67
Alwarkaa	68.89	63.81	67.78	84.78	71.31
Alhadher	59.33	56.67	60.00	76.00	63.00
Amel	58.89	73.33	93.33	86.67	78.06
Rihan	41.11	26.67	23.33	53.33	36.11
L.S.D. (0.05)	8.212				4.106
L.S.D. (0.01)	10.900				5.450
Mean (%)	67.28	64.33	68.60	77.69	
L.S.D. (0.05)	2.737				
L.S.D. (0.01)	3.633				

Table 3: Mean of shoot length (SL) of nine barley varieties in four types of media

Varieties of Barley	Types of Media				Mean (cm)
	MS	½ MS	B-5	½ B-5	
Sameer	5.73	1.29	1.97	3.94	3.23
Buraq	5.07	4.58	4.92	5.30	4.96
Aksad	3.50	2.23	8.97	11.98	6.67
Shuaa	2.35	1.38	2.43	4.18	2.59
Arevat	6.97	6.97	3.06	5.58	5.65
Alwarkaa	2.06	2.95	2.99	3.17	2.79
Alhadher	1.87	1.53	5.18	5.18	3.44
Amel	2.43	5.38	7.50	8.00	5.83
Rihan	4.07	1.80	1.98	5.98	3.46
L.S.D. (0.05)	1.670				0.835
L.S.D. (0.01)	2.217				1.108
Mean (cm)	3.78	3.12	4.33	5.92	
L.S.D. (0.05)	0.556				
L.S.D. (0.01)	0.739				

trait. The mean of SL of germinated immature embryos of barley varieties ranged from 2.59 to 6.67 cm at four types of media.

Aksad variety gave the highest SL value (6.67 cm) and was highly significant superior to all varieties; it followed by Amel and Arevat varieties with values of 5.83 and 5.65 cm of SL, respectively. Whilst, Shuaa and Alwarkaa varieties gave the lowest value 2.59 and 2.79 cm of SL. Table 3 also revealed highly significant differences among media types for SL trait and ranged from 3.12 to 5.92 cm. The type ½ B-5 medium gave the highest value of SL (5.92 cm) and was significant superior on all media types; while the type 1/2 MS medium had the lowest value of SL (3.12 cm). The barley varieties x media types interaction showed highly significant difference for the trait of SL. The shoot length value of combinations ranged from 1.29 to 11.98 cm (Table 3). The interaction of Aksad variety x ½ B-5 medium was a high significant superior by giving the highest value of SL which amounted to 11.98 cm; it followed by Aksad x B-5 and Amel x ½ B-5 with 8.97 and 8.00 cm of SL, respectively. Whereas the interactions of Sameer, Shuaa, Alhadher, and Rihan varieties with ½ MS media gave the lowest values of SL. These results confirmed the high significant differences among varieties and among media types for the SL trait. These data confirm that individual varieties of barley responded differently to each type of media for this trait. For instance, MS medium was suitable for Sameer and Arevat varieties as well as ½ MS medium for Arevat by giving the highest value of SL of germinated immature embryos. While ½ B-5 medium was suitable medium

for Aksad, Shuaa, Rihan, Alhadher, and Amel varieties as well as B-5 medium for 'Alhadher' and 'Amel'. On the other hand, Buraq and Alwarkaa varieties did not show significant differences for SL trait in the four types of media. For this trait, Aksad variety was also the best variety when its immature embryos cultured on 1/2 B-5 medium. The same result was found by Mohammed (2018) that some crosses were superior when cultured in MS, while other crosses were superior in B-5 medium for SL trait. These data are consistent with previous findings which reported that genotypes responded to culture medium independently (Burun and Poyrazoglu, 2002; Russowski et al., 2006; Chen et al., 2011; Mohammed et al., 2020).

The root length (RL) of immature embryos of nine barley varieties were evaluated based on four types of media (Table 4).

The results showed a highly significant difference among barley varieties for RL trait. The mean of RL of immature embryos of barley varieties ranged from 1.45 to 4.68 cm at four types of media. Aksad variety had the highest value (4.68 cm) of RL and was highly significant superior to all studied varieties of barley for RL trait; Arevat and Amel varieties followed Aksad variety in terms of superiority of RL trait (4.02 and 4.01, respectively). While Shuaa and Sameer varieties had the lowest RL values (1.45 and 1.79, respectively). Table 4 also showed highly significant differences among media types for RL trait and ranged from 2.34 to 3.66 cm of immature embryos of barley varieties. The medium of 1/2 B-5 had the highest value (3.66) of RL trait and was significantly superior on all types of media. Whilst the 1/2 MS medium showed

Table 4: Mean of root length (RL) of nine barley varieties in four types of media

Varieties of Barley	Types of Media				Mean (cm)
	MS	½ MS	B-5	½ B-5	
Sameer	3.07	0.38	1.40	2.30	1.79
Buraq	4.37	2.96	4.00	3.63	3.74
Aksad	3.62	1.66	5.48	7.97	4.68
Shuaa	1.75	1.30	1.00	1.76	1.45
Arevat	4.79	4.83	2.76	3.72	4.02
Alwarkaa	1.62	3.05	2.76	2.29	2.43
Alhadher	2.06	1.77	3.95	4.12	2.97
Amel	1.71	3.90	5.61	4.83	4.01
Rihan	4.25	1.22	2.27	2.37	2.53
L.S.D. (0.05)	1.197				0.598
L.S.D. (0.01)	1.590				0.795
Mean (cm)	3.03	2.34	3.25	3.66	
L.S.D. (0.05)	0.399				
L.S.D. (0.01)	0.530				

the lowest RL value (2.34 cm). The combination between barley varieties x media types showed highly significant difference for RL trait. The value of RL of interactions ranged from 0.38 to 7.97 cm (Table 4). The combination between Aksad variety x 1/2 B-5 media were superior by giving the highest RL value (7.97 cm); it followed by the interaction of Amel and Aksad with B-5 medium which amounted to 5.61 and 5.48 cm, respectively. While Sameer variety x ½ MS interaction gave the lowest average of RL trait (0.38 cm). These results confirmed the high significant differences among varieties and among media types. These data also confirm that individual barley varieties responded differently to each type of media. For example, Sameer, Rihan, and Buraq varieties were superior in RL when cultured on MS medium as well as B-5 medium for Buraq variety. While Alwarkaa and Arevat varieties had the highest value in ½ MS medium as well as MS medium for Arevat variety. On the other hand, ½ B-5 was suitable medium for Aksad variety as well as B-5 medium for Alhadher and Amel varieties. Whilst four types of media had no significant effect on RL of Shuaa variety. For this trait, Aksad variety was also the best variety when its immature embryos cultured on 1/2 B-5 medium. These results were confirmed by previous studies (Burun and Poyrazoglu, 2002; Russowski et al., 2006; Han et al., 2011; Mohammed et al., 2020) that is difficult to draw a conclusion about the most suitable medium for different barley genotypes.

Table 5 revealed the root number (RN) of germinated immature embryos of nine barley varieties that cultured at four types of media. The results showed a highly

significant differences among varieties of barley for RN trait. The mean of RN of germinated immature embryos of varieties of barley ranged from 2.88 to 4.32 at four media types. Aksad, Amel, Arevat, and Buraq varieties had the highest values of RN and were highly significant superior to all other studied of barley varieties with values of 4.32, 4.29, 3.98, and 3.94 of RN trait, respectively. While the other five varieties gave the lowest values of RN. Table 5 showed highly significant differences among types of media for RN trait and ranged from 2.99 to 4.12 roots. The medium type of ½ B-5 gave the highest value of RN which amounted to 4.12 roots and was significant superior on all media types; whereas the medium type of 1/2 MS had the lowest RN value (2.99). The interaction between barley varieties x media types showed highly significant difference for the trait of RN. The root number value of combinations ranged from 1.67 to 5.58 roots (Table 5). The interaction of Aksad and Amel varieties with ½ B-5 medium, Arevat x ½ MS medium, and Aksad x B-5 medium had the highest values of RN which amounted to 5.58, 5.27, 5.20, and 5.11 roots, respectively. While the interactions of Rihan and Sameer varieties with ½ MS medium, 'Alhadher' x MS medium, and 'Shuaa' x B-5 medium gave the lowest values of RN. These results confirmed the high significant differences among varieties and among media types for the RN trait. Also, these data confirm that individual varieties of barley responded differently to each type of media for this trait.

For instance, MS medium was suitable for Sameer variety by giving the highest value of SL of germinated immature embryos. While ½ MS medium was suitable

Table 5: Mean of root number (RN) of nine barley varieties in four types of media

Varieties of Barley	Types of Media				Mean (cm)
	MS	½ MS	B-5	½ B-5	
Sameer	4.67	1.69	2.44	3.78	3.15
Buraq	4.06	3.33	3.81	4.55	3.94
Aksad	4.17	2.42	5.11	5.58	4.32
Shuaa	3.33	2.84	1.94	4.03	3.04
Arevat	3.78	5.20	3.21	3.72	3.98
Alwarkaa	3.07	2.44	3.50	2.50	2.88
Alhadher	1.89	3.00	3.94	3.44	3.07
Amel	3.11	4.33	4.45	5.27	4.29
Rihan	3.58	1.67	2.33	4.17	2.94
L.S.D. (0.05)	1.045				0.522
L.S.D. (0.01)	1.387				0.693
Mean (cm)	3.52	2.99	3.42	4.12	
L.S.D. (0.05)	0.348				
L.S.D. (0.01)	0.462				

medium for Arevat variety. Aksad, Buraq, Shuaa, Amel, and Rihan varieties were superior in RN when cultured on ½ B-5 medium, while Alwarkaa and Alhadher varieties gave the highest value in B-5 medium. For this trait, Aksad variety was also the best variety when its immature embryos cultured on 1/2 B-5 medium. These results are consistent with previous studies which reported that genotypes responded to culture medium independently (Burun and Poyrazoglu, 2002; Russowski et al., 2006; Chen et al., 2011; Han et al., 2011; Mohammed et al., 2020).

4 CONCLUSION

There were highly significant differences among barley varieties, media types, and interactions for all studied characteristics except the leaf number, which indicated that there was a high genetic variation among varieties, and high variation among media. Aksad variety, ½ B-5 medium, and Aksad x ½ B-5 gave the highest values for all studied traits. Varieties of barley responded to each type of media independently. As the FGP is the most important trait, ½ B-5 medium was suitable for most varieties, while some varieties were superior either in B-5, MS, or ½ MS media. As a result, it is difficult to draw a conclusion about the efficient medium for all varieties. These varieties could be used in breeding program of interspecific crosses with wild barley species by using the efficient type of media for each variety to improve some quality and quantity traits of barley varieties.

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