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Review Article

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Establishing core collections for enhanced use of germplasm in crop improvement

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ABSTRACT

Plant genetic resources are the basic raw materials and their use in breeding is the most sustainable way to conserve biodiversity. Low use of germplasm in crop improvement programs has resulted large gap between the number of germplasm preserved in genebanks and the number of germplasm used in crop breeding across the globe. Breeders are reluctant to use germplasm largely either due to lack of reliable information on economic traits besides linkage drag or due to breakdown of co-adapted gene complexes, which may prolong cultivar development time. The reduced subsets, representing diversity in the germplasm collection of a given species preserved in genebank, in the form of core or mini core collections are the ideal genetic resources for discovering new sources of variations for use in crop improvement programs. Two decades of research at ICRISAT has led to the establishment of core and mini core collections and their subsequent evaluations has resulted in identification of new sources of variations, for example, resistance to abiotic and/or biotic stresses in chickpea, groundnut, pigeonpea, pearl millet, sorghum, finger millet and foxtail millet. Likewise, a number of nutritionally dense (high protein, Ca, Fe and Zn) germplasm have been identified in finger millet, foxtail millet, groundnut, pearl millet, and sorghum. A few groundnut germplasm with improved oil quality, as determined by variation in oleic and linoleic fatty acids, were also identified. Many of these germplasm were agronomically at par or even superior over controls and showed specific and wide adaptation. The identified sources may be used in genomics and breeding to broaden the cultigen's genepool in these crops.

Keywords**:** association mapping, cereals, core and mini core collections, legumes, germplasm, population structure and diversity

Introduction

Plant genetic resources (PGR) are the basic raw materials for use in crop improvement programmes. The use of PGR in crop improvement is one of the most sustainable ways to conserve valuable genetic resources for the future, and simultaneously to increase food and nutritional security (McCouch et al. 2013). Key to successful crop improvement is a continued supply of genetic diversity in breeding programs, including new or improved variability for target traits. Globally, 7.4 million accessions are preserved in over 1750 genebanks, with cereals and food legumes, respectively, constituting 45% and 15% collection (Figure 1). Eleven percent of these accessions are preserved in CGIAR Centers' genebanks. ICRISAT has the largest collections of its mandate crops (chickpea, groundnut, pigeonpea, pearl millet, and sorghum) and of small millets, totaling 120,454 accessions from 144 countries (Table 1), with mandate crops germplasm constituting 91.4% and small millets 8.6% of its collections. Of these, 2.3% constitute wild and weedy relatives of the mandate crops and of small millets. Managing and utilizing such large diversity in germplasm collections are greatest challenge to germplasm curators and crop breeders.

Breeders are often reluctant to use germplasm and prefer to use their own working collections,

Table 1. Current status of germplasm collections preserved at ICRISAT genebank (accessed on Jan 01, 2014) and safety duplicates preserved at Svalbard Global Seed Vault, Norway.

Crop	ICRISAT		Svalbard Global Seed Vault
	Accessions (#)	Countries (#)	Accessions (#)
Barnyard millet	749	10	726
Chickpea	20,268	60	16,931
Finger millet	6,084	24	5,828
Foxtail millet	1,542	26	1,458
Groundnut	15,446	92	13,900
Kodo millet	665	2	653
Little millet	473	5	460
Pearl millet	22,658	50	19,685
Pigeonpea	13,771	74	9,519
Proso millet	849	30	813
Sorghum	37,949	92	34,027
Total	**120,454**	**144**	**104,000**

largely due to non-availability of reliable information on traits of economic importance (Upadhyaya et al. 2011e), fear of carrying linkage drag, and possibly due to breakdown of co-adapted gene complexes (Ortiz et al. 1998). The other reasons for the underutilization of germplasm include i) lack of accurate and precise large-scale multi-location evaluation of germplasm, ii) lack of rational systematic entry points into the vast international collections, and iii) lack of robust, cost-effective tools to facilitate the efficient utilization of exotic germplasm in crop breeding (Dwivedi et al. 2009; Upadhyaya et al. 2011e). Clearly, there is a need to increase the use of genetically diverse germplasm with beneficial traits to meet the emerging challenges in agricultural production. Reduced subsets in the form of core (~10% of entire collection) or mini core (~1% of entire collection or ~10% of core collection) collections, representing diversity of the entire collection of a given species in the genebank, have been suggested as a resource to study population structure and diversity, discover new sources of variation and identify agronomically beneficial and genetically diverse germplasm for use in crop improvement programs.

This short article provides the current status of PGR preserved in ICRISAT genebank and the formation of reduced subsets (core or mini core collections) for assessing population structure and diversity and discovering new sources of variations, which may be used in cultivar development.

Forming reduced subsets to promote use of germplasm in crop breeding

Reduced subsets such as core (Frankel 1984) and mini core (Upadhyaya and Ortiz 2001) collections, representing diversity of the entire collection of a given species, has been suggested as a gateway to enhance utilization of germplasm in crop improvement programs.

A. Methodology for forming core and mini core germplasm collections

Upadhyaya et al. (2009a) elaborated the standard procedure that they used for developing core and mini core collections. Essentially, this includes stratifying entire collection by taxonomic groups and country of origin, with accessions from smaller and adjacent countries with similar agro-climates grouped together. The standardized data (to eliminate scale differences) is then subjected to hierarchical cluster algorithm of Ward (1963), which optimizes an objective function because it minimizes the sums of squares within groups (clusters) and maximizes the sums of squares among groups. Following this, from each cluster, ~10% of the accessions are randomly selected for inclusion into the core collection. In situation, where the accessions are less than ten in a cluster, at least one accession is included. The means, the variances, the Shannon-Weaver diversity index (H´), and the frequency distribution of traits between the entire collection and core collection is used to test the validity of the latter. To know whether phenotypic associations, which may be under genetic control, were conserved in the core collection, the phenotypic correlations in the entire collection and core collection is estimated and compared. Similar procedure is used to develop mini core collection using the core collection which is further evaluated

for agronomic and/or nutritional traits and data from such evaluations is subjected to statistical analysis as detailed above (Figure 2).

B. Core and mini core collections

Both core and mini core collections are available in chickpea, finger millet, foxtail millet, groundnut, pigeonpea, pearl millet and sorghum, and core collections in barnyard-, kodo-, little- and proso millets (Tables 2 and 3). The core collection constituted about 10% of the accessions of the entire collection of a given species preserved in genebank, while the mini core collection constituted 1% of the accessions of entire collection or 10% of the accessions of the core collection representing diversity of the core collection and entire collection of a given species preserved in a genebank.

C. Genotype-based reference sets

ICRISAT in collaboration with the Generation Challenge Programme (GCP) developed global composite collections of its mandate crops and of finger and foxtail millets, which were subsequently SSR-genotyped to form genotype-based reference sets (Table 4). The reference sets in sorghum consists of 383 accessions, 200 accessions in foxtail millet, while in chickpea, groundnut, pigeonpea, pearl millet and finger millet 300 accessions each.

Core and mini core collections as resource to discovering new sources of variations

Research to date suggests that core and mini core collections or genotype-based reference sets (Glaszmann et al. 2010) have been found useful in extracting germplasm with agronomically beneficial traits for use in crop improvement programs. The researchers at ICRISAT and elsewhere have extensively evaluated these subsets for resistance to abiotic and biotic stresses and for agronomic and nutritional traits, and reported a number of germplasm

Table 2. Core collections of ICRISAT mandate crops and small millets.

Crop	Accessions used (#)	Traits used (#)	Core collection accessions (#)	Reference
Chickpea	16,991	13	1,956	Upadhyaya et al. 2001
Finger millet	5,940	14	622	Upadhyaya et al. 2006b
Foxtail millet	1,474	23	155	Upadhyaya et al. 2008b
Groundnut	14,310	14	1,704	Upadhyaya et al. 2003
Pearl millet	20,844	22	2,094	Upadhyaya et al. 2009b
Pigeonpea	12,153	14	1,290	Reddy et al. 2005
Proso millet	833	20	106	Upadhyaya et al. 2011c
Sorghum	22,474	21	2,247	Grenier et al. 2001
Barnyard millet	736	21	89	Upadhyaya et al. 2014c
Kodo millet	656	20	75	
Little millet	460	20	56	

Table 3. Mini core collections of ICRISAT mandate crops and small millets.

Crop	Entire collection (#)	Mini core collection accession (#)	% of entire collection	Traits used (#)	Reference
Chickpea	16,991	211	1.24	16	Upadhyaya and Ortiz 2001
Finger millet	5,940	80	1.34	18	Upadhyaya et al. 2010
Foxtail millet	1,474	35	2.37	21	Upadhyaya et al. 2011b
Groundnut	14,310	184	1.28	34	Upadhyaya et al. 2002
Pearl millet	20,844	238	1.14	12	Upadhyaya et al. 2011a
Pigeonpea	12,153	146	1.2	16	Upadhyaya et al. 2006a
Sorghum	22,473	242	1.08	21	Upadhyaya et al. 2009c

accessions with agronomically beneficial traits, as detailed below.

A. Resistance to abiotic and biotic stresses

A number of accessions resistance to abiotic and/or biotic stresses have been reported in chickpea (Gaur et al. 2013; Krishnamurthy et al. 2013a, b; Upadhyaya et al. 2013a), finger millet (Kiran Babu et al. 2013; Krishnamurthy et al. 2014a), foxtail millet (Sharma et al. 2013a; Krishnamurthy et al. 2014b), groundnut (Upadhyaya et al. 2014a), pigeonpea (Krishnamurthy et al. 2012; Sharma et al. 2012a), pearl millet (Sharma et al. 2013b), and sorghum (Vadez et al. 2011; Sharma et al. 2010, 2012b). Some germplasm accessions, for example in chickpea and groundnut, showed multiple resistance to abiotic and biotic stresses, with a few having good agronomic and seed quality traits (Upadhyaya et al. 2013a, 2014a). In addition, a number of agronomically superior but susceptible to abiotic and/or biotic stresses have also been reported in chickpea and groundnut (Upadhyaya et al. 2013a, 2014a) which may be used as resource for agronomic traits in resistance breeding programs.

B. Agronomic and nutritional traits

A number of chickpea and groundnut mini core accessions had no resistance to abiotic or biotic stresses, however, some of these were agronomically superior and adapted to diverse environments (groundnut: rainy and /or postrainy environments; chickpea: irrigated and /or rainfed environments), which form the good source for use as parents in resistance breeding programs (Upadhyaya et al. 2013a, 2014a). Likewise, a number of seed-nutrient dense (Oil and protein, Fe and Zn) accessions have been identified in finger millet, foxtail millet, groundnut, and sorghum (Kumar et al. 2009; Upadhyaya et al. 2011b,d, 2012a,b). Early maturity and seed size are agronomically important traits, and accessions with early maturity and large seed size have been reported both in chickpea (Kabuli types) and groundnut (Spanish type) (Upadhyaya et al. 2006c, 2007; Gowda et al. 2011). Small millet grains are rich in Ca, and some accessions with high Ca content have been reported both in finger and foxtail millets (Upadhyaya et al. 2011b,d). The legume kernel-derived fibers stimulate the growth of colonic bifidobacteria, which contributes to improved colon health (Smith et al. 2006; Fernando et al. 2010). More recently, large variability was observed among chickpea mini core accessions for oligosaccharides such as sucrose (3.6 to 54.1 mg g^{-1}), raffinose (0.2 to 15.1 mg g^{-1}), stachyose (2.8 to 59.4 mg g^{-1}), and ciceritol (4.8 to 90.7 mg g^{-1}) (ICRISAT unpublished data). Clearly, there is a need to screen mini core collections to identify germplasm with nutraceuticals properties for use as ingredients in functional foods for improving human health.

C. Bioenergy trait

Of recent, sweet sorghum has gained importance as smart bioenergy (Ethanol) crop. Sweet sorghum stalks are rich in sugar. When sorghum mini core collection accessions were evaluated for stalk-sugar content, as measured by Brix (%) for two seasons, a number of accessions with high Brix (%) and some with good agronomic traits were identified. For example, IS# 13294, 13549, 23216, 23684, 24139, 24939, and 24953 with significantly greater mean brix (14.0% to 15.2%) than the best control, IS 33844 (12.4%) across environments; however, these were found low yielder and had lower 100-seed weight. On contrary, IS# 1004, 4698, 23891, and 28141 significantly outyielded IS 33844 by 11.7% to 22.7%, but were comparable to controls for Brix (~13%) (Upadhyaya et al. 2014b). These accessions are therefore ideal genetic resource for use in breeding programs to develop sweet sorghum as bioenergy crop.

Core and mini core collections as diversity panel for assessing population structure, diversity, allelic richness and association genetics

The reduced subsets (both conventional mini core collections and reference sets) are ideal genetic resource for use as diversity panels for studying the population structure and diversity, and conduct association genetics for agronomically beneficial traits.

A. Dissecting population structure, diversity, and allelic richness

The analysis of allelic richness revealed that these reference sets captured between 78 to 95% allelic diversity of the global composite collections (Table 4), which sufficiently discriminated the majority of the accessions (with few exceptions) clustered based on biological and/or geographical diversity (Figure 3). For examples, the sorghum accessions structured according to geographic regions and race within region (Billot et al. 2013), the chickpea accessions separated based on seed types, with desi types differentiating from kabuli types (Upadhyaya et al. 2008a), the groundnut accessions largely clustered at subspecies level, and most of the wild relatives of chickpea, groundnut and pigeonpea grouped separately from cultivated types (Generation Challenge Program Project abstracts 2008; www.generationcp.org/component/docman/doc_download/281-2008?Itemid=15). More importantly, a

Table 4. Global composite collections and reference sets formed for ICRISAT mandate crops and small millets.

Crop	SSRs used (#)	Composite collection		Reference set		Reference
		accession (#)	Allele (#)	accession (#)	Allele (#)	
Chickpea	48	2915	1683	300	1315 (78.1)	Upadhyaya et al. 2008a
Finger millet	20	959	231	300	206 (89.2)	GCP Project Report 2008 (abstract)
Foxtail millet	19	452	362	200	315 (87)	GCP Project Report 2008 (abstract)
Groundnut	21	852	490	300	466 (95.1)	GCP Project Report 2008 (abstract)
Pearl millet	19	1021	230	300	218 (94.8)	GCP Project Report 2008 (abstract)
Pigeonpea	20	952	197	300	164 (83.2)	GCP Project Report 2008 (abstract)
Sorghum	41	3367	783	383	613 (78.3)	Billot et al. 2013

Figures in parenthesis represent proportion of reference set alleles representing composite collection alleles.

number of rare (frequency ≤1%), common (frequency from 1% to 20%), most frequent (frequency >20%) and unique (present in one or group of accessions but absent in other accessions or group of accessions) alleles were detected in all the crops investigated. Unique alleles together with DUS (distinct, uniform and stable) (Rathinavel et al. 2005) test may be used for identifying for propriety germplasm. Rare alleles are rich-resource to associate these allelic differences with new variations arising as a result of mutations or environment-induced variations in germplasm.

B. Association mapping

Advances in genomics have provided greater insights to the germplasm users into the structure of the germplasm diversity and mine allelic variation associated with beneficial traits for use in crop breeding. Understanding how diversity is structured to unlock its potential for crop improvement is an emerging area worldwide made possible by the rapid advances in scale, robustness and reliability and the sharp fall in unit costs of deploying marker technology. The genomes of several food crops including chickpea, foxtail millet, pigeonpea and sorghum have been sequenced (Patterson et al. 2009; Zhang et al. 2012; Varshney et al. 2012, 2013). More importantly, the resequencing of diverse germplasm with agronomically beneficial traits may provide researcher opportunities to associate these sequence differences with trait variation, as evidenced in maize and sorghum (Lai et al. 2010; Zheng et al. 2011). The deployment of new generation sequencing technologies will further accelerate efficient and precise identification and tracking of thousands of genetic variants in the populations at much reduced cost (Hamilton and Buell 2012).

The mini core collection of sorghum has been extensively genotyped using large number of SSRs and SNPs markers and together with phenotyping data, a number of significant marker-trait associations have been discovered. For example, six and ten SNPs linked to plant height and maturity, with some having close proximity to previously mapped height/maturity QTL or candidate genes for plant height and inflorescence architecture (Upadhyaya et al. 2013b; Morris et al. 2013); eight SNPs associated with resistance to anthracnose (*Colletotrichum sublineolum*) with candidate genes for disease resistance found in most of the detected SNPs (Upadhyaya et al. 2013c); two to five SNPs linked to leaf rust (*Puccinia purpurea*) and grain mold (*Fusarium* sps.) (Upadhyaya et al. 2013d); one to two SSR alleles linked to kernel weight and tiller numbers localized with previously mapped QTL (Upadhyaya et al. 2012c).

Multi-trait genetically diverse germplasm for use in crop improvement

The genetically diverse germplasm with multiple resistant traits offer breeders opportunity to develop breeding and genetic mapping populations combining multiple resistances into an agronomically improved genetic background for cultivar development. A number of genetically diverse germplasm pairs with resistance to abiotic and/or biotic stresses, those possessing good seed quality and resistance to abiotic and/or biotic stresses, or those that are agronomically

desirable but susceptible have been identified both in chickpea and groundnut (Upadhyaya et al. 2013a, 2014a) for enhancing trait values. Clearly, more research is needed to identify genetically diverse germplasm pairs with agronomically beneficial traits to support crop breeding.

Conclusions

Core and mini core collections representing diversity in the entire collection of the germplasm of a given species preserved in the genebank are ideal resource for efficient conservation and utilization of plant genetic resources in crop improvement programs. Core and mini core collections or genotype-based reference sets are available in chickpea, finger millet, foxtail millet, groundnut, pigeonpea, pearl millet, and sorghum. New sources of variations for resistance to abiotic and/or biotic stresses and of agronomic and nutritional (oil and protein, Ca, Fe and Zn, O/L ratio) traits have been reported for use in crop breeding. More importantly, a number of accessions with multiple resistance and nutrient dense types, some with specific adaptation (rainy and/or postrainy seasons), are available in ICRISAT genebank, which can be accessed after signing with ICRISAT the ***Standard Material Transfer Agreement*** (www.icrisat.org/icrisat-ip-mta-htm). With the availability of abundant genomic resources on these crops, it is visualized that there will be increased use of genomics-based germplasm analysis to enhance use of germplasm and make impact in breeding programmes in near future.

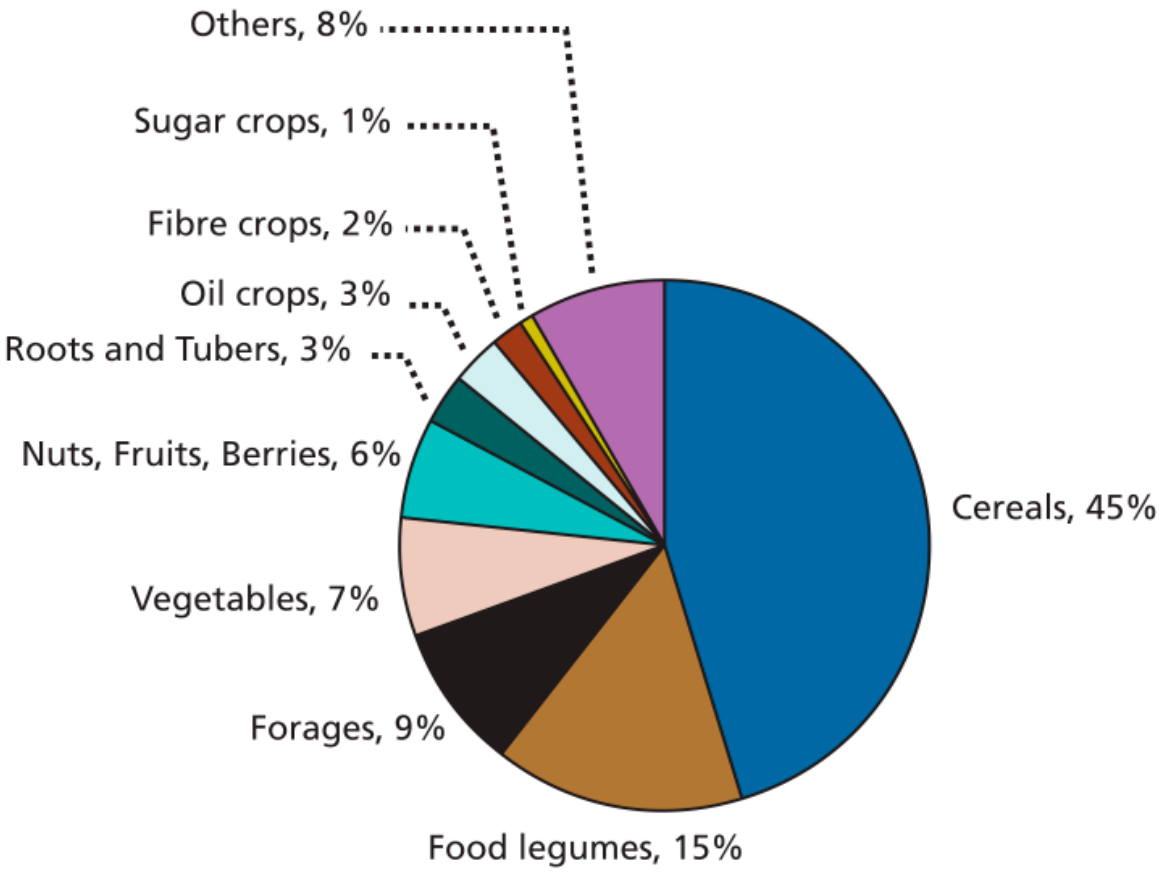


Figure 1. Plant genetic resources conserved over 1750 genebanks globally (FAO 2010; http://www.fao.org/docrep/013/i1500e/i1500e.pdf).

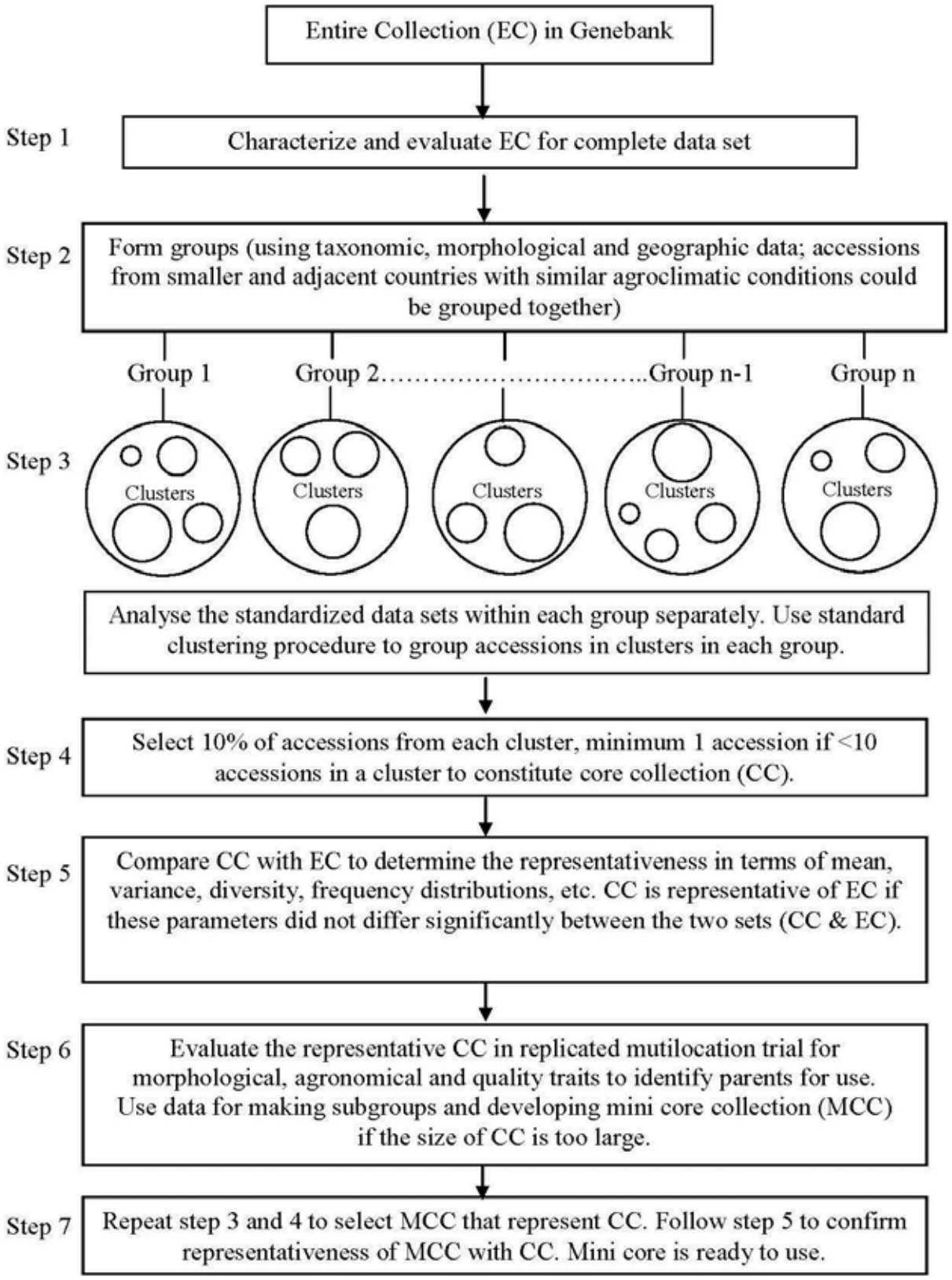
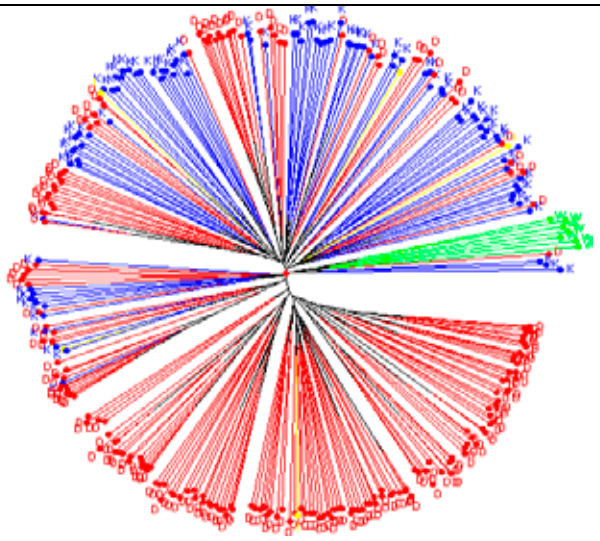
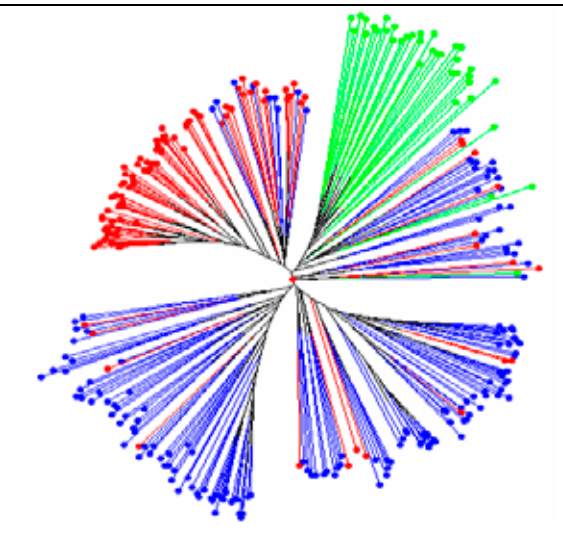


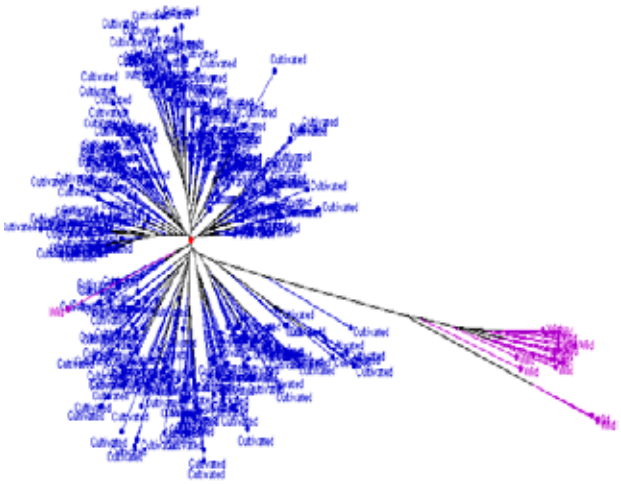
Figure 2. Flow diagram to establish core and mini core collections in a crop (Upadhyaya et al. 2009a).



Chickpea (Upadhyaya et al. 2008a)



Groundnut (Generation Challenge Program Project abstracts 2008)



Pigeonpea (Generation Challenge Program Project abstracts 2008)

Sorghum (Billot et al. 2013)

Figure 3: Tree diagram depicting population structure and diversity in reference sets of chickpea, groundnut, pigeonpea and sorghum.

References

Billot C, Ramu P, Bouchet S, Chantereau J, Deu M, Gardes L, Noyer J-L, Rami J-F, Rivallan R, Li Y, Lu P, Wang T, Folkertsma RT, Arnaud E, Upadhyaya HD, Glaszmann J-C and Hash CT (2013). Massive sorghum collection genotyped with SSR markers to enhance use of global genetic resources. PLoS ONE 8: e59714.

Dwivedi SL, Upadhyaya HD and Gowda CLL (2009). Approaches to enhance the value of genetic resources in crop improvement. The FAO Regional Project "Capacity building and enhanced regional collaboration for the conservation and sustainable use of plant genetic resources in Asia". (http://www.fao.org/docrep/014/am265e00.pdf).

FAO (2010). The second report on the state of the world's Plant Genetic Resources for food and agriculture. http://www.fao.org/docrep/013/i1500e/i1500e.pdf.

Fernando WMU, Hill JE, Zello GA, Tyler RT, Dahl WJ and Van Kessel AG (2010). Diets supplemented with chickpea or its main oligosaccharide component raffinose modify fecal microbial composition in healthy adults. Benef. Microbes 1:197-207.

Frankel OH (1984). Genetic perspective of germplasm collection. In: Genetic manipulations: impact on man and society, Arber W, Limensee K, Peacock WJ and Stralinger P (eds.), Cambridge University Press, Cambridge, pp: 61-170.

Gaur PM, Jukanti AK, Samineni S, Chaturvedi SK, Singh S, Tripathi S, Singh I, Singh G, Das TK, Aski M, Mishra N, Nadarajan N and Gowda CLL (2013). Large variability in chickpea for tolerance to herbicides imazethapyr and metribuzin. Agronomy 3:524-536.

Generation Challenge Program Project abstracts (2008). Generation Challenge Program: Cultivating plant diversity for the resource poor. (Abstract). pp. 47-50 and 64-66.

Glaszmann JC, Kilian B, Upadhyaya HD and Varshney RK (2010). Accessing genetic diversity for crop improvement. Curr. Opin. Plant Biol. 13:167-173.

Gowda CLL, Upadhyaya HD, Dronavalli N and Singh S (2011).Identification of large-seeded high-yielding stable kabuli chickpea germplasm lines for use in crop improvement. Crop Sci. 51:198-209.

Grenier C, Hamon P and Bramel-Cox PJ (2001). Core collection of sorghum: II. Comparison of three random sampling strategies. Crop Sci. 41:241-246.

Hamilton JP and Buell CR (2012). Advances in plant genome sequencing. Plant J. 70:177-190.

Kiran Babu T, Thakur RP, Upadhyaya HD, Reddy PN, Sharma R, Girish A and Sarma NDRK (2013). Resistance to blast (*Magnaporthe grisea*) in a mini core collection of finger millet germplasm. Eur. J. Plant Pathol. 135:299-311.

Krishnamurthy L, Kashiwagi J, Tobita S, Osamu I, Upadhyaya HD, Gowda CLL, Gaur PM, Sheshshayee MS, Singh S, Vadez V and Varshney RK (2013a). Variation in carbon isotope discrimination and its relationship with harvest index in the reference collection of chickpea germplasm. Funct. Plant Biol. 40:1350-1361.

Krishnamurthy L, Kashiwagi J, Upadhyaya HD, Gowda CLL, Gaur PM, Singh S, Purushothaman R and Varshney RK (2013b). Partitioning coefficient – a trait that contributes to drought tolerance in chickpea. Field Crops Res. 149:354-365.

Krishnamurthy L, Upadhyaya HD, Saxena KB and Vadez V (2012). Variation for temporary water logging response within the mini core pigeonpea germplasm. J. Agric. Sci. 150: 357-364.

Krishnamurthy L, Upadhyaya HD, Purushothaman R, Gowda CLL, Kashiwagi J, Dwivedi SL, Singh S and Vadez V (2014a). The extent of variation in salinity tolerance of the mini core collection of finger millet (*Eleusine coracana* L. Gaertn.) germplasm. Plant Sci. 227:51-59.

Krishnamurthy L, Upadhyaya HD, Gowda CLL, Kashiwagi J, Purushothaman R, Singh S and Vadez V (2014b). Large variation for salinity tolerance in the core collection of foxtail millet (*Setaria italica* (L.) P. Beauv.) germplasm. Crop Pasture Sci. 65:353-361.

Kumar AA, Reddy BVS, Ramaian B, Reddy PS, Sahrawat KL and Upadhyaya HD (2009). Genetic variability and plant character association of grain Fe and Zn in selected core collection accessions of sorghum germplasm and breeding lines. eSAT J. 7.

Lai J, Li R, Xu X, Jin W, Xu M, Zhao H, Xiang Z, Song W, Ying K, Zhang M, Jiao Y, Ni P, Zhang J, Li D, Guo X, Ye K, Jian M, Wang B, Zheng H, Liang H, Zhang X, Wang S, Chen S, Li J, Fu Y, Springer NM, Yang H, Wang J, Dai J, Schnable PS and Wang J (2010). Genome-wide patterns of genetic variation among elite maize inbred lines. Nature Genetics 42:1027-1030.

McCouch S, Baute GJ, Bradeen J, Bramel P, Bretting PK, Buckler E, Burke J M, Charest D, Cloutier S, Cole G, Dempewolf H, Dingkuhn M, Feuillet C, Gepts P, Grattapaglia D, Guarino L, Jackson S, Knapp S, Langridge P, Lawton-Rauh A, Lijua Q, Lusty C, Michael T, Myles S, Naito K, Nelson RL, Pontarollo R, Richards CM, Rieseberg L, Ross-Ibarra J, Rounsley S, Hamilton RS, Schurr U, Stein N, Tomooka N, van der Knaap E, van Tassel

D, Toll J, Valls J, Varshney RK, Ward J, Waugh R, Wenzl P and Zamir D (2013). Agriculture: Feeding the future. Nature 499:23-24.

Morris GP, Ramu P, Deshpande SP, Hash CT, Shah T, Upadhyaya HD, Riera-Lizarazu O, Brown PJ, Acharya CB, Mitchell SE, Harriman J, Glaubitz JC, Buckler ES and Krecovich S (2013). Population genomic and genome-wide association studies of agroclimatic traits in sorghum. Proc. Natl. Acad. Sci. (USA) 110:453-458.

Ortiz R, Ruiz-Tapia EN and Mujica-Sanchez A (1998). Sampling strategy for a core collection of Peruvian quinova germplasm. Theor. Appl. Genet. 96:475-483.

Paterson AH, Bowers JE, Bruggmann R, Dubchak I, Grimwood J, Gundlach H, Haberer G, Hellsten, U, Mitros T, Poliakov A, Schmutz J, Spannagl M, Tang H, Wang X, Wicker T, Bharti AK, Chapman J, Feltus FA, Gowik U, Grigoriev IV, Lyons E, Maher CA, Martis M, Narechania A, Otillar RP, Penning BW, Salamov AA, Wang Y, Zhang L, Carpita NC, Freeling M, Gingle AR, Hash CT, Beller B, Klein P, Kresovich S, McCann MC, Ming R, Peterson DG, Rahman M, Ware D, Westhoff P, Mayer KFL, Messing J and Rokhsar DS (2009). The *Sorghum bicolor* genome and the diversification of grasses. Nature 457:551- 556.

Rathinavel K, Manickam S and Sabesh M (2005). Training manual on DUS test in cotton with reference to PVV and FR legislation.

Reddy LJ, Upadhyaya HD, Gowda CLL and Singh S (2005). Development of core collection in pigeonpea [*Cajanus cajan* (L.) Millspaugh] using geographic and qualitative morphological descriptors. Genet. Resour. Crop Evol. 52:1049-1056.

Sharma R, Rao VP, Upadhyaya HD, Reddy VG and Thakur RP (2010). Resistance to grain mold and downy mildew in a minicore collection of sorghum germplasm. Plant Dis. 94: 439-444.

Sharma M, Rathore A, Mangala UN, Ghosh R, Sharma S, Upadhyaya HD and Pande S (2012a). New sources of resistance to fusarium wilt and sterility mosaic in a mini core collection of pigeonpea germplasm. Eur. J. Plant Pathol. 133:707-714.

Sharma R, Upadhyaya HD, Manjunatha SV, Rao VP and Thakur RP (2012b). Resistance to foliar diseases in a mini core collection of sorghum germplasm. Plant Dis. 96:1629-1633.

Sharma R, Girish AG, Upadhyaya HD, Humayun P, Talluri KB, Panduranga Rao V and Thakur RP (2013a). Identification of blast resistance in a core collection of foxtail millet germplasm. Plant Dis. 98:519-524.

Sharma R, Upadhyaya HD, Manjunatha SV, Rai KN, Gupta SK and Thankur RP (2013b). Pathogenic variation in the pearl millet blast pathogen *Magnaporthe grisea* and identification of resistance to diverse pathotypes. Plant Dis. 97:189-195.

Smith SC, Choy R, Johnson SK, Hall RS, Wildeboer-Veloo ACM and Welling GW (2006). Lupin kernel fiber consumption modifies fecal microbiota in healthy men as determined by rRNA gene fluorescent *in situ* hybridization. Eur. J. Nutr. 45:335-341.

Upadhyaya HD, Bramel PJ, Ortiz R and Singh S (2002). Developing a mini core of peanut for utilization of genetic resources. Crop Sci. 42:2150-2156.

Upadhyaya HD, Bramel PJ and Singh S (2001). Development of a chickpea core subset using geographic distribution and quantitative traits. Crop Sci. 41:206-210.

Upadhyaya HD, Dronavalli N, Singh S and Dwivedi SL (2012a). Variability and stability for kernel iron and zinc contents in the ICRISAT mini core collection of peanut. Crop Sci. 52:2628-2637.

Upadhyaya HD, Mukri G, Nadaf HL and Singh S (2012b). Variability and stability analysis for nutritional traits in the mini core collection of peanut. Crop Sci. 52:168–178.

Upadhyaya HD, Wang Y-H, Sharma S, Singh S and Hasentstein KH (2012c). SSR markers linked to kernel weight and tiller number in sorghum identified by association mapping. Euphytica 187:401-410.

Upadhyaya HD, Dronavalli N, Dwivedi SL, Kashiwagi K, Krishnamurthy L, Pande S, Sharma HC, Vadez V, Singh S, Varshney RK and Gowda CLL (2013a). Mini core collection as a resource to identifying new sources of variation for use in chickpea breeding and genomics. Crop Sci. 53:2506-2517.

Upadhyaya HD, Wang Y-H, Gowda CLL and Sharma S (2013b) Association mapping of maturity and plant height using SNP markers with the sorghum mini core collection. Theor. Appl. Genet. 126:2003-2015.

Upadhyaya HD, Wang Y-H, Sharma R and Sharma S (2013c). Identification of genetic markers linked to anthracnose resistance in sorghum using association analysis. Theor. Appl. Genet. 126:1649-1657.

Upadhyaya HD, Wang Y-H, Sharma R and Sharma S (2013d). SNP markers linked to leaf rust and grain mold resistance in sorghum. Mol. Breed. 32:451-462.

Upadhyaya HD, Dwivedi SL, Vadez V, Hamidou F, Singh S, Varshney RK and Liao B (2014a). Multiple resistance and nutritionally dense germplasm identified from mini core collection in groundnut. Crop Sci. 54:679-693.

Upadhyaya HD, Dwivedi SL, Ramu P, Singh SK and Singh S (2014b). Effect of post - flowering drought on genetic variability for stalk sugar content in sorghum mini core collection. Crop Sci. 54:2120-2130.

Upadhyaya HD, Dwivedi SL, Singh SK, Singh S, Vetriventhan M and Sharma S (2014c). Forming core collections in barnyard, kodo, and little millets using morphoagronomic descriptors. Crop Sci. 54:2673-2682.

Upadhyaya HD, Dwivedi SL, Baun M, Varshney RK, Udupa SM, Gowda CLL, Hoisington D and Singh S (2008a). Genetic structure, diversity, and allelic richness in composite collection and reference set in chickpea (*Cicer arietinum* L.). BMC Plant Biol. 8:106.

Upadhyaya HD, Pundir RPS, Gowda CLL, Reddy VG and Singh S (2008b). Establishing a core collection of foxtail millet to enhance the utilization of germplasm of an underutilized crop. Plant Genet. Resour. 7:177-184.

Upadhyaya HD, Reddy LJ, Gowda CLL, Reddy KN and Singh S (2006a). Development of a mini core subset for enhanced and diversified utilization of pigeonpea germplasm resources. Crop Sci. 46:2127-2132.

Upadhyaya HD, Gowda CLL, Pundir RPS, Reddy VG and Singh S (2006b). Development of core subset of finger millet germplasm using geographical origin and data on 14 quantitative traits. Genet. Resour. Crop Evol. 53:679-685.

Upadhyaya HD, Reddy LJ, Gowda CLL, Singh S (2006c). Identification of diverse groundnut germplasm: Sources of early maturity in a core collection. Field Crops Res. 97:261-271.

Upadhyaya HD, Pundir RPS, Dwivedi SL, Gowda CLL (2009a). Mini core collections for efficient utilization of plant genetic resources in crop improvement programs. Inf. Bull. no. 78. pp. 52.

Upadhyaya HD, Gowda CLL, Reddy KN and Singh S (2009b). Augmenting the pearl millet core collection for enhancing germplasm utilization in crop improvement. Crop Sci. 49: 573-580.

Upadhyaya HD, Pundir RPS, Dwivedi SL, Gowda CLL, Reddy VG and Singh S (2009c). Developing a mini core collection of sorghum for diversified utilization of germplasm. Crop Sci. 49:1769-1780.

Upadhyaya HD and R Ortiz (2001). A mini core subset for capturing diversity and promoting utilization of chickpea genetic resources in crop improvement. Theor. Appl. Genet. 102: 1292-1298.

Upadhyaya HD, R Ortiz, Bramel PJ and Singh S (2003). Development of a groundnut core collection using taxonomical, geographical and morphological descriptors. Genet. Resour. Crop Evol. 50:139-148.

Upadhyaya HD, Salimath PM, Gowda CLL and Singh S (2007). New early-maturing germplasm lines for utilization in chickpea improvement. Euphytica 157:195-208.

Upadhyaya HD, Sarma NDRK, Ravishankar CR, Albrecht T, Narsimhudu Y, Singh SK, Varshney SK, Reddy VG, Singh S, Dwivedi SL, Wanyera N, Oduori COA, Mgonja MA, Kisandu DB, Parzies HK and Gowda CLL (2010). Developing a mini core collection in finger millet using multilocation data. Crop Sci. 50:1924-1931.

Upadhyaya, HD, Yadav D, Reddy KN, Gowda CLL and Singh S (2011a). Development of pearl millet mini core collection for enhanced utilization of germplasm. Crop Sci. 51:227-223.

Upadhyaya HD, Ravishankar CR, Narsimhudu Y, Sarma NDRK, Singh SK, Varshney SK, Reddy VG, Singh S, Parzies HK, Dwivedi SL, Nadaf HL, Sahrawat KL and Gowda CLL (2011b). Identification of trait-specific germplasm and developing a mini core collection for efficient use of foxtail millet genetic resources in crop improvement. Field Crops Res. 124:459-467.

Upadhyaya HD, Sharma S, Gowda CLL, Reddy VG and Singh S (2011c). Developing proso millet (*Panicum miliaceum* L.) core collection using geographic and morpho-agronomic data. Crop Pasture Sci. 62:383-389.

Upadhyaya HD, Ramesh S, Sharma S, Singh SK, Varshney SK, Sarma N D R K, Ravishankar C R, Narasimhudu Y, Reddy VG, Sahrawat KL, Dhalalakshmi TN, Mgonja MA, Parzies HK, Gowda CLL and Singh S (2011d). Genetic diversity for grain nutrients in a core collection of finger millet (*Eleusine coracana* (L.) Gaertn.) germplasm. Field Crops Res. 121: 42-52.

Upadhyaya HD, Thudi M, Dronavali N, Gujaria N, Singh S, Sharma S and Varshney RK (2011e). Genomic tools and germplasm diversity for chickpea improvement. Plant Genetic Resources 9: 45-58.

Vadez V, Krishnamurthy L, Hash CT, Upadhyaya HD and Borrell AK (2011). Yield, transpiration efficiency, and water-use variations and their relationships in the sorghum reference collection. Crop Pasture Sci. 62:645-655.

Varshney RK, Chen W, Li Y, Bharti AK, Saxena RK, Schlueter, JA, Donoghue MTA, Azam S, Fan G, Whaley AM, Farmer AD, Sheridan J, Iwata A, Tuteja R, Penmetsa RV, Wu W, Upadhyaya HD, Yang S-P, Shah T, Saxena KB, Michael T, McCombie WR, Yang B, Zhang G, Yang H, Wang J, Spillane C, Cook DR, May GD, Xu X and Jackson SA (2012). Draft genome sequence

of pigeonpea (*Cajanus cajan*), an orphan legume crop of resource-poor farmers. *Nature Biotechnology* 30: 83-89.

Varshney RK, Song C, Saxena RK, Azam S, Yu S, Sharpe AG, Cannon S, Baek J, Rosen BD, Tar'an B, Millan T, Zhang X, Ramsay LD, Iwata A, Wang Y, Nelson W, Farmer AD, Gaur PM, Soderlund C, Penmetsa RV, Xu C, Bharti AK, He W, Winter P, Zhao S, Hane JK, Carrasquilla-Garcia N, Condie JA, Upadhyaya HD, Luo M-C, Thudi M, Gowda CLL, Singh NP, Lichtenzveig J, Gali KK, Rubio J, Nadarajan N, Dolezel J, Bansal KC, Xu X, Edwards D, Zhang G, Kahl G, Gil J, Singh KB, Datta SK, Jackson SA, Wang J and Cook DR (2013). Draft genome sequence of chickpea (*Cicer arietinum*) provides a resource for trait improvement. Nat. Biotechnol. 31: 240-246.

Ward J (1963). Hierarchical grouping to optimize an objective function. J. Am. Stat. Assoc. 38:236-244.

Zhang G, Liu X, Quan Z, Cheng S, Xu X, Pan S, Xie M, Zeng P, Yue Z, Wang W, Tao Y, Bian C, Han C, Xia Q, Peng X, Cao R, Yang X, Zhan D, Hu J, Zhang Y, Li H, Li H, Li N, Wang J, Wang C, Wang R, Guo T, Cai Y, Liu C, Xiang H, Shi Q, Huang P, Chen Q, Li Y, Wang J, Zhao Z and Wang J (2012). Genome sequence of foxtail millet (*Setaria italica*) provides insights into grass evolution and biofuel potential. Nat. Biotechnol. 30:549-554.

Zheng LY, Guo SX, He B, Sun LJ, Peng Y, Dong SS, Liu TF, Jiang S, Ramchandran S, Liu CM and Jing HC (2011). Genome wide patterns of genetic variation in sweet and grain sorghum (*Sorghum bicolor*). Genome Biology 12:R114.

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Analysis of Ukrainian Polissya and Forest-steppe winter wheat (*Triticum aestivum* L.) cultivars for the presence of "resistant" allelic state of non-race-specific disease resistance locus *Lr34/Yr18/Pm38*

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ABSTRACT

Resistance against rusts and powdery mildew is one of the most important traits of modern wheat cultivars, because in some years these diseases can significantly reduce crop yields. The genes that confer durable non-race-specific resistance are of interest to many breeding programs. One of them is *Lr34/Yr18/Pm38*. This multiple resistance locus is associated with partial and durable resistance to rust diseases, powdery mildew and tolerance to barley yellow dwarf virus. We used marker *caISBP1* (insertion site-based polymorphism marker) specific for the resistance, associated with the *Lr34/Yr18/Pm38* gene. It is a co-dominant marker between loci coding ABC-transporter and cytochrome *P450* in the region involved in resistance expression. We investigated a collection of 28 winter wheat cultivars of NSC ''Institute of Agriculture NAAS Ukraine'' developed in different periods. The "resistant" allelic state of the marker *caISBP1* was identified in 11 cultivars (39% of the total number). The "susceptible" allelic state (the Lr34- allele) was identified in 9 wheat cultivars (32%) and 8 (29%) cultivars were heterozygous. Our investigation confirmed the presence of the Lr34+ allele in Polissya and Forest-Steppe winter wheat cultivars.

***Keywords*:** rust diseases, adult plant resistance, winter wheat

Introduction

A search for cultivars can be donors of resistance genes is a time-consuming but important task of wheat breeding. For effective selection, a breeder should have more or less solid data about the resistance genes and the opportunity for their quick identification during the breeding process. Determining of resistance associated with some genes by the phenotype may be inexact and time-consuming. Detection of resistance gene accelerates with the use of molecular DNA markers which can rapidly identify allelic state of a gene in laboratory with high precision. Using DNA markers in research of resistance gene reduces the time and increases efficiency and accuracy of the investigation in the most cases. The rust diseases of wheat are caused by fungi of the order *Uredinales*. There are three species of wheat rusts: leaf rust (*Puccinia triticina* f. sp. *tritici*), stem rust (*P.graminis* f. sp. *tritici*) and yellow rust (*P. striiformis* f. sp. *tritici*). In some years yield losses due leaf rust could reach 15-20%, stem rust – 60-70%, yellow rust – 20% and more (Peresypkin et al. 1990; Bublyk et al. 1999).

Rust fungi suppress the process of synthesis of HMW glutenin components in kernels which affects bread-making properties. Also they decline the rate of synthesis and deposition of starch and proteins in endosperm causing puny grain (Kolyuchyy et al. 2007).

Breeders can use wide non-race-specific adult plant resistance (APR) genes which stay effective for a long period. APR is associated with reduction of disease development, slowdown of pathogen haustorium penetration in cells of host plant (Krattinger et al. 2009).

Lr34 is one of the most important APRs genes in wheat breeding due to its wide spectrum resistance and stability. It is present in about 50% of cultivars worldwide and was a component of resistance of highly productive cultivars at the start of green revolution (Hoisington et al. 1999). The *Lr34* gene has been staying effective against rusts more than 100 years (Krattinger et al. 2009; Dakouri et al. 2010).

This multiple resistance locus is associated with partial and durable resistance to rust diseases (Dyck et al. 1987; McIntosh et al. 1992; Singh 1992; Kolmer et al. 2011), powdery mildew (*Blumeria graminis* (DC.) Speer) and tolerance to barley yellow dwarf virus (Spielmeyer et al. 2005; Singh 1993). The gene was localized at the short arm of the *7D* chromosome of wheat (Dyck et al. 1987). The *Lr34* locus putatively codes the ATP-binding cassette (ABC)-transporter, a protein localized at the cytoplasmic membrane (Krattinger et al. 2009). A number of molecular marker linked to this gene have been developed (Bossolini et al. 2006, Lagudah et al. 2009). Among them there are the markers localized in the *Lr34* locus or the regulated ones (Dakouri et al. 2010). We used the marker *caISBP1* (insertion site-based polymorphism marker) specific for the resistance, associated with the *Lr34/Yr18/Pm38* gene. It is a co-dominant marker between the ABC-transporter and cytochrome *P450* in region involved in resistance expression (Dakouri et al. 2010).

The aim of our investigation was detecting the allelic state of the *Lr34/Yr18/Pm38* locus using DNA markers because cultivars of NSC "Institute of Agriculture NAAS" have never been investigated for the presence of this resistance. The resistance-associated allele of the *Lr34/Yr18/Pm38* gene can be involved in breeding process to obtain cultivars with resistance to multiple pathogens in Polissya and Forest-Steppe due the positive influence of its climatic conditions on *Lr34* resistance expression.

Materials and methods

The DNA was extracted from kernels of 28 winter wheat cultivars developed in periods from 1971 to 1990 and 1992 – 2012 in Cereal Breeding and Seed Growing Department of the National Science Centre "Institute of Agriculture NAAS Ukraine": Polesskaya-90, Polesskaya-95, Kolectivnaya-77, Shchedraya-Polesya, Polesskaya-71, Polesskaya-70, Polesskaya-80, Polesskaya-87, Polesskaya-107, Polesskaya-bezostaya, Kievskaya-73, Polesskaya-29, Polesskaya-1259, Polesskaya-92, Miryutinka, Zhuravka, Stolichna, Epilog, Gnom, Krayevyd, Artemida, Kopilivchanka, Analog, Benefis. The cultivars Olgana, IZ49-12 (Kesaria-Poliska) and IZ15-12 (Pamyati-Girka) are under field trials. The cultivar Kievskaya-polukarlikovaya was developed in cooperation with the Institute of Plant Physiology and Genetics of NAS (Kiev, Ukraine). Seeds were obtained from the collection of NSC "Institute of Agriculture NAAS" and the National Centre of Plant Genetic Resources (Kharkov, Ukraine). These cultivars were released for growing condition of Forest-Steppe and Polissya agroclimatic zones.

DNA was extracted using Diatom™ DNA Prep 100 (NEOGENE®, Ukraine) kit by the standard protocol from the bulk of 7 kernels. We used allele-specific marker *caISBP1* (insertion site based polymorphism marker) for the identification of the allelic state of the *Lr34/Yr18/Pm38* locus (Dakouri et al. 2010). This codominant marker is located between the *Lr34* locus which encodes the ABC-transporter and first cytochrome *P450* in the site effecting the resistance expression. The GenPak® PCR Core (NEOGENE®, Ukraine) was used for PCR. The results were visualized by electrophoresis in 2-2,5% agarose gel with 1xTBE buffer and stained with ethidium bromide. The amplified DNA fragments of 509 b.p. in length were obtained in case of "resistance" allelic state of the marker *caISBP1* (the *Lr34*+ allele). In case of the "susceptible" allelic state of the marker (the *Lr34*- allele) fragments of 391 b.p. in length were observed (Dakouri et al. 2010). The cultivars Chinese Spring (the *Lr34*+ allele, *St*+) and Renan (the *Lr34*- allele, *St*-) were used as standards. Marker of molecular mass was GeneRulerTM 50 bp DNA Ladder ready-to-use (Fermentas, Lithuania).

Results and discussion

Among 28 cultivars developed in the Institute of Agriculture 11 showed the *Lr34*+ allele of the marker *caISBP1*. They are Stolichna, Artemida, Analog, Benefis, Polesskaya- 70, Polesskaya-bezostaya, Kievskaya-73, Kievskaya - polukarlikovaya, Polesskaya - 80, Zhuravka, IZ15-12, (Table 1). On the Figure 1 represented the electropforegrams of several cultivars showed different results.

In 9 cultivars we detected the "susceptible" allelic state of the marker *caISBP1*: Polesskaya-90, Kolectivnaya-77, Shchedraya-Polesya, Epilog, Gnom, Krayevyd, Kopilivchanka, Olgana and Polesskaya-1259. Heterozygosity was detected in 8 samples: Polesskaya-95, Polesskaya-71, Polesskaya-87, Polesskaya-107, Polesskaya-29, Polesskaya-92, Miryutinka, IZ49-12. Resistance associated with *Lr34* is common in cultivars developed in period before 1990 and 1992-2012 equally.

Our results are in agreement with the previous publication of Karelov et al. (2011) showing that the

Lr34+ allele was widely spread among Ukrainian bread wheat cultivars. A sufficiently high resistance conferred by *Lr34* in cultivars adapted for the Polissya and Forest-Steppe gives evidence of its broad adaptive value, which does not lose the value nowadays. The specificity of breeding process in the Institute of Agriculture (using moderate infectious backgrounds) may account for selection of breeding material with the *Lr34*+ allele. It is known, that main genetic source of *Lr34* resistance was cultivar Bezostaya1, used as a parental component by breeders. This cultivar has a unique combination of adaptability, disease resistance and high bread making quality (Morgounov et al. 2011). Analysis of seed storage proteins loci of investigated cultivars showed the presence of gliadin and HMW glutenin alleles that possibly derived from Bezostaya1, which could be selected due to high bread-making quality (Zaika et al. 2012, Labuschagne et al. 2002).

Thus we performed the investigation of 28 winter wheat cultivars of NSC ‘’Institute of Agriculture NAAS Ukraine” developed in different periods. For molecular diagnostic we used the loci-specific marker *caSNBP1* of the *Lr34/Yr18/Pm38* gene conferring APR against several phytopathogens. Presence of the resistance-associated allele of the marker was identified in 11 cultivars (39% of the total number). These cultivars can be used in breeding programs as donors of the resistance conferred by the *Lr34/Yr18/Pm38* gene.

Table 1 Allelic state of *Lr34/Yr18/Pm38* loci in winter bread wheat cultivars of NSC “Institute of Agriculture NAAS”.

Cultivar	Year of release	Allelic state	Cultivar	Year of release	Allelic state
Polesskaya-71	1971	+/-	Polesskaya-95	1996	+/-
Polesskaya-70	1974	+	Kopilivchanka	2003	-
Kievskaya-73	1974	+	Stolichna	2005	+
Kolectivnaya-77	1974	-	Gnom	2007	-
Kievskaya-Polukarlikovaya	1977	+	Artemida	2008	+
Miryutinka	1978	+/-	Analog	2008	+
Polesskaya-Bezostaya	1981	+	Benefis	2008	+
Polesskaya-80	1982	+	Epilog	2009	-
Shchedraya-Polesya	1986	-	Krayevyd	2012	-
Polesskaya-87	1990	+/-	Olgana		-
Polesskaya-92	1992	+/-	Zhuravka		+
Polesskaya-90	1994	-	Polesskaya-107		+/-
Polesskaya-1259	1995	-	IZ15-12		+
Polesskaya-29	1996	+/-	IZ49-12		+/-

«+» – “resistant” allelic state, «-» – “susceptible” allelic state, «+/-» – heterozygous for the Lr34+ allele) with the marker *caISBP1*

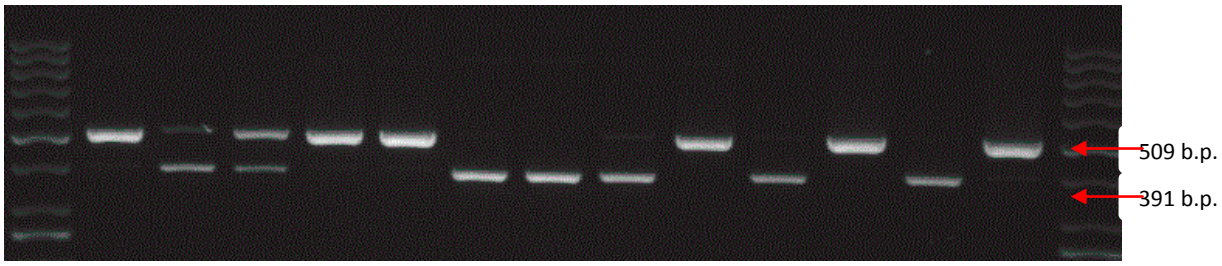

Figure 1 Electrophoregram of PCR products, obtained with DNA samples of bread winter wheat and primers that flank marker *caISBP1*: *1*– Polesskaya-80; *2* – Polesskaya-87; *3* – Polesskaya-107; *4* – Zhuravka; *5* – Stolichna; *6* – Epilog; *7* – Gnom; *8* – Krayevyd; *9* – Artemida; *10* – Kopilivchanka; *11* – Analog; *12* – Renan (St-); *13* – Chinese Spring (St+); L – marker of molecular weight (50 bp DNA Ladder)

References

Bossolini E, Krattinger SG, Keller B (2006). Development of simple sequence repeat markers specific for the *Lr34* resistance region of wheat using sequence information from rice and Aegilops tauschii. Theoretical Applied Genetics 113:1049–1062.

Bublyk LI, Vasechko GI, Vasiliev VP, Lesovoy MP (editor) (1999). Handbook of plant protection. Urogay, Kiev, [book in Russian].

Dakouri A, McCallum BD, Walichnowski AZ, Cloutier S (2010). Fine-mapping of the leaf rust *Lr34* locus in *Triticum aestivum* (L.) and characterization of large germplasm collections support the ABC transporter as essential for gene function. Theoretical Applied Genetics 121: 373–384.

Dyck PL (1987). The association of a gene for leaf rust resistance with the chromosome *7D* suppressor of stem rust resistance in common wheat. Genome 29:467–469.

Hoisington D, Khairallah M, Reeves T, Ribaut J, Skovmand B, Taba S, Warburton M (1999). Plant genetic resources: what can they contribute toward increased crop productivity? Proceedings of National Academy of Sciences of USA 96: 5937–5943.

Karelov AV, Pirko IaV, Kozub NA, Sozinov IA, Pirko NN, Litvinenko NA, Lyfenko SF, Koliuchiĭ VT, Blium IaB and Sozinov AA (2011). Identification of allelic state of leaf rust-resistance *Lr34* gene in the cultivars of soft winter wheat of Ukrainian breeding. Cytology and Genetics. 45(5):3-10, [article in Russian].

Kolmer JA, Garvin DF, Jin Y (2011). Expression of a Thatcher Wheat Adult Plant Stem Rust Resistance QTL on Chromosome Arm 2BL is Enhanced by *Lr34*. Crop Science 51:526–533.

Kolyuchyy VT, Vlasenko VA, Borsuk GU (2007). Wheat breeding, seed production and growing technology (2007), Agrarna nauka, Kiev [book in Ukrainian].

Krattinger SG, Lagudah ES, Spielmeyer W, Singh R, Huerta-Espino J, McFadden H, Bossolini E, Selter LL, Keller B (2009). A Putative ABC Transporter Confers Durable Resistance to Multiple Fungal Pathogens in Wheat. Science 323: 1360 – 1363.

Labuschagne MT, Pretorius ZA, Grobelaar B (2002). The influence of leaf rust resistance genes Lr29, *Lr34*, Lr35 and Lr37 on breadmaking quality in wheat. Euphytica 124: 65–70.

Lagudah ES, Krattinger SG, Herrera-Foessel S, Singh RP, Huerta-Espino J, Spielmeyer W, Brown-Guedira G, Selter LL, Keller B (2009). Gene-specific markers for the wheat gene *Lr34/Yr18/Pm38* which confers resistance to multiple fungal pathogens. Theoretical Applied Genetics 119:889–898.

McIntosh RA, Wellings CR, Park RF (1995). Wheat Rusts: An Atlas of Resistance Genes.CSIRO, Australia, pp: 74–76.

Morgounov A, Tufan HA, Sharma R, Akin B, Bagci A, Braun H-J, Kaya Y, Keser M, Payne TS, Sonder K, McIntosh R (2011). Global incidence of wheat rusts and powdery mildew during 1969–2010 and durability of resistance of winter wheat variety Bezostaya 1. European Journal of Plant Pathology 132:323–340.

Peresypkin VF, Kirik NN, Lesovoy MP (1990). Diseases of agriculture crops. Urogay, Kiev, V.1 [book in Russian].

Singh RP (1992). Genetic Association of Leaf Rust Resistance Gene *Lr34* with Adult Plant Resistance to Stripe Rust Resistance in Bread Wheat. Phytopathology V.82 (№8): 835–838.

Singh RP (1993). Genetic association of gene Bdv1 for tolerance to Barley Yellow Dwarf Virus with genes *Lr34* and Yr18 for adult plant resistance to rusts in bread wheat. Plant Diseases 77: 1103–1106.

Spielmeyer W, McIntosh RA, Kolmer J, Lagudah ES (2005). Powdery mildew resistance and *Lr34*/Yr18 genes for durable resistance to leaf and stripe rust co segregate at a locus on the short arm of chromosome *7D* of wheat. Theoretical Applied Genetics 111: 731–735.

Zaika EV, Kozub NA, Sozinov IA, Girko VS, Golik LN (2012). Characteristic of winter common wheat cultivars for storage protein loci of the NSC "Institute of Agriculture NAAS Ukraine" // International science conference "Breeding and genetics of agriculture crops: tradition and perspectives"(100 years of SGI), Odessa, pp: 152-153, [thesis in Ukrainian].

Research Article

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Effect of GA_3 concentrations in basal medium on embryos germination of Cleopatra mandarin x carrizo citrange and Cleopatra mandarin x Flying Dragon

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ABSTRACT

The gametic sterility, the long duration of the young stage, species incompatibilities and especially the high rate of polyembryony are common difficulties in citrus breeding. Polyembryonic cultivars usually contain a zygotic embryo at the earlier stages of seeds. By embryo rescue techniques, the zygotic embryos further development can be secured. In this study, suitable GA_3 concentration (0; 1; 2 mg/L) and embryo development stages (days after pollination 105, 115, 125) for embryo germination at Cleopatra mandarin x Carrizo citrange and Cleopatra mandarin x 'Flying Dragon' trifoliate orange were examined. The results indicated that the highest germination rate was observed from 2 mg/L GA_3 containing M&T media which were harvested 115 days after pollination. The highest germination rates of embryos taken 115 days after pollination and germinated on 2 mg/L GA_3 containing media in Cleopatra mandarin x Carrizo citrange and Cleopatra mandarin x Flying Dragon were obtained with 65% and 30%, respectively.

Keywords: *citrus*, embryo rescue, GA_3, germination, hybridization, immature embryo

Introduction

Citrus, having major importance in the world and Turkey, is propagated by vegetative and generative methods. However, rootstocks in citrus have to be used because of particular diseases and the some soil and climatic conditions.

One of the major problems in citrus breeding is competition between zygotic and nucellar embryos (Soost and Roose 1996). This challenge is eliminated by in vitro embryo rescue techniques of developing embryos. The success of embryo rescue depends on ingredient of medium and embryo developing stages (Jaskani et al. 2005). The germination capacity of citrus embryos can be affected by embryo's genetic structure and embryo developing stage (Viloria et al. 2005).

Various studies have reported that the addition of 0.01 mg/L GA_3 (Chagas et al. 2005), 1 mg/L GA_3 (Ollitrault et al. 2007), and 2 mg/L GA_3 (Gmitter et al. 1990) in growing media for embryos is important in developing citrus plants.

In this study, we aimed to obtain hybrid individuals from Cleopatra mandarin (*Citrus reshni* hort. ex Tanaka) x Carrizo citrange [*Citrus sinensis* (L.) Osb. × *Poncirus trifoliata* (L.) Raf.], and Cleopatra mandarin (*Citrus reshni* hort. ex Tanaka) x 'Flying Dragon' trifoliate orange (*Poncirus trifoliata* (L.) Raf.).

Material and methods

Cleopatra mandarin, 'Flying Dragon' and Carrizo citrange were used as materials. Carrizo citrange and 'Flying Dragon' trifoliate orange were used as father plant and Cleopatra mandarin used as mother plant in the crossing combinations.

Murashige and Tucker (1969) medium was used as basic culture medium and 50 g/L sucrose, 25 mg/L adenine sulfate, 500 mg/L malt extract were put in medium. Then, 0, 1 and 2 mg/L GA_3 were supplemented to the prepared medium and 8 g/L agar was added. The fruits were taken 105, 115 and 125 days after pollination (DAP), were

surface sterilized (Ollitrault et al. 2007). Then, the seeds were removed from the fruit by forceps and immature embryos were taken from the microphyl parts of seeds under binocular. Two embryos were placed into Petri dishes separately containing culture medium. And then, the Petri dishes were incubated in growth chamber. Germinated embryos were counted and rate of germination of embryos was determined. Later, embryos were sub-cultured in Murashige and Skoog (1962) medium containing 0.02 mg/L NAA and 20 g/L sucrose in culture tubes to provide seedling growing (Perez-Tornero and Porras 2008), and then they were incubated. The plantlets in sub-culture were transferred to pots. The experiment was conducted as randomized plot design with 10 replications and each replication have two embryos. Data were subjected to analysis of variance with mean separation by LSD's test.

Results

Embryo development stages, GA_3 concentrations in the medium and their interactions were significant on germination rate of Cleopatra mandarin x Carrizo citrange (Table 1) and Cleopatra mandarin x 'Flying Dragon' trifoliate orange (Table 2) hybrid embryos ($p \leq 0.05$).

Table 1. Germination rate (%) of Cleopatra mandarin x Carrizo citrange

Embryo development stage	GA_3 Concentration			
	Control 0 mg/L GA_3	1mg/LGA_3	2mg/L GA_3	Means (DAP)
105 DAP	15.00 e*	35.00 c	40.00 bc	45.00
115 DAP	25.00 d	45.00 b	65.00 a	30.00
125 DAP	10.00 e	45.00 b	25.00 d	26.67
Means (GA_3)	16.17	41.67	43.33	

* Different letters indicate significant differences (P<0.05) according to the LSD test
* (LSD: 5.1462)

In Table 1, the highest germination rate of Cleopatra mandarin x Carrizo citrange were obtained from embryos taken 115 days after pollination and germinated on 2 mg/L GA3 containing media with 65.00%. In Table 2, the highest germination rates of Cleopatra mandarin x Flying Dragon were obtained from embryos taken 115 days after pollination and germinated on 2 mg/L GA_3 containing media with 30.00%.

Table 2.Germination rate (%) of Cleopatra mandarin x Flying Dragon

Embryo development stage	GA_3 Concentration			
	Control 0 mg/L GA_3	1mg/LGA_3	2mg/L GA_3	Means (DAP)
105 DAP	5.00 d *	10.00 cd	20.00 b	11.67
115 DAP	5.00 d	5.00 d	30.00 a	13.33
125 DAP	5.00 d	10.00 cd	15.00 bc	10.00
Means (GA3)	5.00	8.33	21.67	

* Different letters indicate significant differences (P<0.05) according to the LSD test
* (LSD: 5.1462)

Discussion

According to our results, the highest germination rate in embryo development stages were obtained 115 days after pollination in two hybridization combinations. Similarly, 118 days after pollination for embryo germination has been reported by Chagas et al. (2005). On the other hand, there were reports indicating good embryo germinations after 50 days (Wang et al. 1999), 80 days (Tan et al. 2007), 100 days (Deng et al. 1996), 105 days (Scarano et al. 2003) and 120 days (Carimi et al. 1998; Das et al. 2000; Kurt and Ulger 2014).

2 mg/L GA_3 dose was appropriated for embryo germination. Gmitter et al. (1990) studied different citrus species and cultivars, and they indicated that adding 2 mg/L GA_3 to the medium gave positive effects in increasing citrus embryo germination. Some reports also showed the addition of 0.01 mg/L GA_3 (Ribeiro et al. 2000), 0.1 mg/L GA_3 (Pasqual et al. 1990), 1 mg/L GA_3 (Kurt and Ulger 2014) and to growing on the medium alone resulted in good embryo germination of citrus. These differences may be due to the growing location and cultivars used.

As a result, it was determined that the best embryo rescue stage was 115 days after pollination. In addition, 2 mg/L GA_3 was determined the appropriate dose in media for embryo rescue.

References

Carimi F, Pasquale F and Puglia AM (1998). *In vitro* rescue of zygotic embryos of sour orange (*Citrus aurantium* L.) and their detection based on RFLP analysis. Plant Breeding 117:261–266.

Chagas EA, Pasqual M, Ramos JD, Pio LAS, Dutra LF and Cazetta JO (2005). Activated charcoal and gibberellic acid concentrations on immature embryos culture. Ciencia e Agrotecnologia 29(6):1125–1131.

Das A, Paul AK and Chaudhuris S (2000). Micropropagation of sweet orange (*Citrus sinensis* Osbeck) for the development of nucellar seedling. Indian J. Expt. Bot.38(3):269–272.

Deng XX, Yi HL, Li F and Guo WW (1996). Triploid plants regenerated from crossing diploid pummel and tangerine with allotetraploid somatic hybrid of citrus. Proc. Int. Soc. Citricult. 1:189–192.

Gmitter FG, Ling XB and Deng XX (1990). Induction of triploid Citrus plants from endosperm cali in vitro. Theor. Appl. Genet. 80:785–790.

Jaskani MJ, Khan IA and Khan MM (2005). Fruit set, seed development and embryo germination in interploid crosses of Citrus. Sci. Hort. 107:51–57.

Kurt S and Ulger S (2014). Production of Common Sour Orange × Carrizo Citrange Hybrids Using Embryo Rescue. *International Journal of Fruit Science*, 14:42–48.

Murashige T and Skoog F (1962). A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant. 15:473–497.

Murashige T and Tucker PH (1969). Growth factor requirements of citrus tissue culture. Proc. 1st Intl. Citrus Symp. 3:1155–1161.

Ollitrault P, Guo W and Grosser JW (2007). Somatic hybridization, p. 235–260. In:I. Khan (ed.). Citrus genetics, breeding and biotechnology. CABI, New York.

Pasqual M, Riberio VG and Ramos JD (1990). Influencia do GA_3 e do carvao ativado sobre o enraizamento in vitro de embrioes de laranja 'Natal'. Pesquisa Agropeciaria Brasileira, Brasilia 25(10):1477–1482.

Perez-Tornero O and Porras I (2008). Assessment of polyembryony in lemon: rescue and in vitro culture of immature embryos. Plant Cell Tissue Organ Cult.93:173–180.

Ribeiro VG, Sanábio D, Souza CN de, Lopes PSN, Bocardo MR and Pasqual M (2000). Efeitos de ácido giberélico e carvão ativado no cultivo in vitro de *Citrus limonia* Osbeck x *Poncirus trifoliata* (L.) Raf. Pesquisa Agropecuária Brasileira, Brasília 35:27–30.

Scarano MT, Tusa N, Abbate L, Lucretti S, Nardi L and Ferrante S (2003). Flowcytometry, SSR and modified AFLP markers for the identification of zygotic plantlets in backcrosses between 'Femminello' lemon cybrids (2n and 4n) and a diploid clone of 'Femminello' lemon (*Citrus limon* L. Burm. F.) tolerant to mal secco disease. Plant Sci. 164(6):1009–1017.

Soost RK and Roose M (1996). Citrus, In: Fruit breeding: Tree and tropical fruits, Janick J and Moore JN (eds.), Wiley, New York, pp: 257–323.

Tan M, Song J and Deng X (2007). Production of two mandarin x trifoliate orange hybrid populations via embryo rescue with verification by SSR analysis. Euphytica 157:155–160.

Viloria Z, Grosser JW and Bracho B (2005). Immature embryo rescue, culture and seedling development of acid citrus fruit derived from interploid hybridization. Plant Cell, Tissue and Organ Cult. 82:159–167.

Wang JF, Chen ZG and Lin TX (1999). Observation on the embryonic development in citrus after cross pollination. Chin. Dev. Reprod. Biol. Soc. 8(2):57–63.

Research Article

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Safflower *(Carthamus tinctorius L.)* breeding activities at Trakya Agricultural Research Institute

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ABSTRACT

There is a vegetable oil deficit in Turkey. Oil crops production in Turkey does not meet the requirements of vegetable oil. About 60 % of vegetable oil consumed in Turkey is imported every year paying about 3 billion dollars. Safflower plant being resistant to drought may be one of the alternative oil crops in Turkey. Some of the main obstructions in safflower production in Turkey are low seed yield, low oil content, diseases and insects. The varieties in production have an oil content of about 28-32 %. New superior safflower varieties have to be developed by breeding and selection methods among the existing genetic variation. The Safflower breeding program at Trakya Agricultural Research Institute was initiated in 2000 with two different oil types, oleic and linoleic to develop new varieties which have high yielding capacity, yield stability, high oil content, wide adaptation ability, resistance to diseases and insects prevalent in the region and all over the Turkey. About 60 materials in oleic types and 250 materials in linoleic types collected around the World were tested in observation nurseries and crossing blocks every year. All the materials were planted in the middle of March. Pedigree method was utilized in segregating populations developed after hybridization. Single plant selection was started in F_2 population and ended in F_7. Each year about 500-600 single plants were selected and replanted next year as a new generation.

***Keywords*:** trakya, safflower, oil, oleic, linoleic, oil yield.

Introduction

Safflower (*Carthamus tinctorius* L.) is one of the alternative oil crops in Turkey. First research activities on safflower in Turkey were initiated in 1929-1930 in Eskisehir province. These activities lasted about 10 years; untill the beginning of World War II in 1939. During this period, a composite variety, "Yenice 1813", was developed and released to the farmers in the region. Due to the increase in population, vegetable oil deficit appeared. That's why research on safflower was reinitiated in 1958, after about a 20-year interruption (Dincer, 1964). Three varieties had been developed until 2011. Only two of them, Dincer and Remzibey-05, are still in production. The performances of these two varieties are acceptable but, oil contents are very low.

Safflower production is supported by Turkish Ministry of Food Agriculture and Livestock. Each farmer receives about 0.2 dollar for per kilo of safflower crop produced after selling in 2013. This amount was about 0.1 dollar in 2006 when the support was started (Anonymous, 2013a). The total safflower planted area was 150-170 hectares in 2005. Due to these supports, safflower planted areas have been increased. Safflower planted area in Turkey reached about 50.000-60.000 ha for the time being. The average yield per hectare is about 1300 kg (Anonymous, 2013b). Some of the main obstructions in safflower production in Turkey are low seed yield, low oil content, diseases and insects. The varieties in production have an oil content of about 28-32 %. They also

are susceptible to safflower rust (*Puccinia carthami*), Alternaria leaf blight (*Alternaria* spp.) and safflower fly (*Acanthiophilus helianthi* Rossi.).

There is a vegetable oil deficit in Turkey because oil crops production in Turkey does not meet the domestic requirements of vegetable oil. About 60 % of vegetable oil consumed in Turkey is imported every year paying about 3 billion dollars. Safflower plant being resistant to drought may be one of the alternative oil crops in Turkey. New superior safflower varieties have to be developed by breeding and selection methods among the existing genetic variation.

Successful results were obtained in genetic improvement in safflower such as seed yield and oil content (Ramachandran 1985) and combining high yield with high oil content (Harishbabu et.al. 2005). All the works must be emphasized on indirect selection of higher head number per plant and 1000 seed weight and lower number of branches along with thinner seed pericarp to improve seed and oil yield in safflower (Zheng, et al. 1993). Traits, such as 1000 seed weight and seed number per plant are the best selection criteria to improve oil yield genetically in drought stress conditions. On the other hand, in non-drought conditions, traits 1000 seed weight, days to physiological maturity and seed number per plant are the most important components for oil yield and must be improved (Golparvar, 2011). Head numbers per plant, head weight and thinner seed pericarp are the important traits for improving seed and oil yield in safflower cultivars (Rao et.al 1997; Corleto et.al 1997; Mozaffari et.al 2006). Seed yield per plant is positively and significantly correlated with heads per plant (r = 0.65**), seeds per head (r = 0.76**) and primary branches per plant (r = 0.38*). According to path analysis, days to maturity, primary branches per plant and seeds per head have the highest positive direct effect on seed yield, respectively. Seeds per head and head per plant are the most important selection criteria to improve seed yield in safflower (Golkar et.al 2011). There is a negative but significant correlation between dry matter and seed yield and between dry matter and oil yield in safflower (Jamshi-Moghadam and Pordad, 2006). The inheritance of the spininess trait was studied by many researchers. It was found out that spininess is dominant over spinelessness and four genes (namely, *Sa, Sb, Sc* and *Sd*) are involved in determining the level of spininess (Narkhede and Deokar 1990; Pahlavani et al. 2004). Grain yield is positively correlated with seed weight and plant height (Johnson 2001). The number of head per plant is the most important character determining grain yield per plant and the number of head has the highest positive correlation with grain yield (Bagawan and Ravikumar, 2001).

The safflower breeding program at Trakya Agricultural Research Institute was initiated in 2000 with two different oil types, oleic and linoleic. The main objectives of the safflower breeding program at Trakya Agricultural Research Institute (TARI) are to develop new varieties which have high yielding capacity, yield stability, high oil content, wide adaptation ability, resistance to diseases and insects prevalent in the region and all over the Turkey. Since TARI is the National Safflower Research Coordinator, all the research activities are conducted not only for Trakya region but also for throughout Turkey.

Materials and methods

Both oleic and linoleic type materials were collected from around the World. About 60 genotypes in oleic types and 250 genotypes in linoleic types were tested in observation nurseries and crossing blocks every year. After evaluation of the domestic and foreign materials in terms of earliness, single plot yield, oil content, reaction to prevalent diseases, etc, with the materials found promising, about 15 crossing combinations in total were made each year both in oleic and linoleic type safflowers. In crossing, emasculations were made in the late afternoon and pollinations were made in the next morning for the best results. Pedigree method was utilized in segregating populations getting after hybridization. Single plant selection was started in F_2 population. The selection was ended in F_7. Each year about 500-600 single plants were selected and replanted next year as a new generation. When the single plant selection reached to F_6 or/and F_7, the plots were harvested in bulk and put in the preliminary, and regional yield trials, respectively, for testing with present varieties in terms of mainly seed yield, oil content and oil yield. The performance of each new line was tested in each trial and calculated whether they were above or below the trial's average yield. The preliminary yield and yield trials were set up only in Edirne (headquarters) location. Three different locations (Edirne, Kırklareli and Tekirdag) were utilized for safflower regional yield trials.

All the materials were planted in the middle of March. Crossing blocks were planted as 3 rows in 2 m length. For yield trials, Randomized Complete Block Design (RCBD) with 4 reps was utilized. The six-row plots were 5.0 m long with the 0.17 m x 0.05 m plant spacing. Two rows in the middle were harvested and the border two rows were discarded as side effects. Harvested plot size was 3.4 m². Harvest was made in

the first week of August with combine in some years, by hand in some years. Some observations, such as seed yield (kg ha^{-1}), 1000 seed weight (g), flowering and physiological maturity (days), plant height (cm), oil content (%) and oil yield (kg ha^{-1}) were taken. All the data taken were analyzed statistically using JUMP program.

Results

The yield of various varieties in 2012 varied between 3550-4240 kg/ha. One of the check (C) varieties, Dincer, gave the highest seed yield with 4240 kg/ha. One of our new lines (TRE-ASL09/14) was the second after Dincer in terms of seed yield with 4160 kg/ha. Our other line, TRE-ASO12/08, was third with 4000 kg/ha in terms of seed yield (Table 1). The two new lines had higher oil content than two check varieties, Dincer and Remzibey-05. One of our lines, TRE-ASO12/08, ranked first among all check varieties with 42.0 % oil content. The same results are also valid for 2013 growing season. According to the 2-year (2012 and 2013) average; the check variety, Dincer, was the first among five entries with 4665 kg/ha seed yield. The line, TRE-ASL09/14, ranked second with 4365 kg/ha seed yield after the check variety, Dincer. In terms of oil content, the line, TRE-ASO12/08, ranked first among the five entries with a 42,1 % oil content. It means that the new line had higher oil content than all check varieties (Table 1). The line, TRE-ASL09/14, had been in state registration trial since 2009. The other line, TRE-ASO12/08, has also been in state registration trial since 2012. This line has been given a production permit untill it is registered. This is an oleic type variety which has an oil content of 40-42 % on dry matter basis. It also has an oleic fatty acid content of 73-75 %. One of the line, TRE-ASL09/14, was registered and named as “LINAS” on April 10, 2013. This variety is linoleic type and has an oil content of 37-38 % at a dry basis (at 0 % moisture).

Besides these two lines, many oleic and linoleic type new lines bulked in F_7 in 2013 were tested in preliminary yield trials (Table 2 and 3). In oleic type yield trial, seed yields varied between 2930 kg/ha and 5220 kg/ha. Five new oleic lines gave higher seed yield than the lowest check variety, Balci. In terms of oil content, twelve lines had higher oil content than all check varieites. One line had the highest oil content with 48,7 %. Oil yield (seed yield x oil content) of the entries varied between1391 kg/ha and 2019 kg/ha (Table 2). In linoleic type yield trial, seed yields varied between 3000 kg/ha and 5410 kg/ha. The newly registered variety, LINAS as a check, gave the highest seed yield with 5410 kg/ha. Four new linoleic lines gave higher seed yield than the lowest check variety, Balci. In terms of oil content, three lines had higher oil content than all check varieties. Oil yield varied between1072 kg/ha and 2159 kg/ha. Our newly registered variety, LINAS, had the highest oil yield with 2159 kg/ha among twenty entries.

Discussion

As a result of safflower breeding program at Trakya Agricultural Research Institute, two lines (TRE-ASL09/14 and TRE-ASO12/08) have been developed so far. One of them, TRE-ASL09/14, was registered and named as “LINAS” on 10.04.2013. This is a linoleic type safflower variety. The seeds of this variety will be multiplied in 2014 and sold to the farmers for 2014 autumn planting and 2015 spring planting. Other line, TRE-ASO12/08, is still at the state registration trial and will be registered in 2015. This line will be named as “OLAS” in 2015 when it is registered.

The other new lines bulked in F_7 in 2012 were at preliminary yield trials in 2013 growing season. Some of them will be offered for registration if their performances are better than the check varieties in the coming years (2014 and 2015 growing seasons). These two varieties, LINAS and OLAS, having high yielding and high oil capacity, will contribute to lessen the vegetable oil deficit in Turkey in coming years.

Table 1. Performance of Two New Safflower Varieties in 2012 and 2013

VARIETIES	2012		2013		2 Year Average Yield (kg/ha)*	2 Year Average Oil Content (%)**
	Yield (kg/ha)*	Oil Content (%)**	Yield (kg/ha)*	Oil Content (%)**		
DINCER (C)	4240	32,6	5090	29,8	4665	31,2
LINAS (TRE-ASL09/14)	4160	39,3	4570	40,3	4365	39,8
TRE-ASO12/08	4000	42,0	3670	42,2	3835	42,1
REMZIBEY-05 (C)	3790	34,4	4090	32,3	3940	33,4
BALCI (C)	3550	41,0	3690	39,2	3620	40,1
L.S.D (0.05)	353	0,5	369	0,4	235	0,5
C.V (%)	17,8	1,6	11,8	1,5	12,3	1,5

*- Based on 10 % moisture
**- At dry matter basis (0 % moisture)

Table 2. Performance of New Oleic Safflower Lines in 2013

OLEIC LINES	Flowering Period (day)*	Physiological Maturity Period (day)*	Plant Height (cm)	1000 Seed Weight (g)**	Oil Content (%)***	Oil Yield (kg/ha)	Seed Yield (kg/ha)**	
DINCER (C)	76	109	87	49	29,2	1524	5220	a
TRE-OA05-02-212110T	78	110	90	37	37,3	1846	4950	ab
TRE-OA05-05-113110T	73	105	77	42	39,4	1915	4860	ab
REMZIBEY-05 (C)	73	106	80	44	32,1	1554	4840	ab
TRE-OA05-02-252110T	76	109	91	40	40,6	1827	4500	abc
TRE-OA05-06-121110T	76	111	78	38	42,6	1891	4440	a-d
TRE-OA05-05-251110T	77	111	89	34	46,1	2019	4380	a-e
BALCI (C)	73	106	78	43	39,7	1731	4360	a-e
TRE-OA05-02-242110T	76	109	85	35	39,6	1719	4340	a-e
TRE-OA05-02-122110T	74	107	79	36	40,5	1737	4290	b-f
TRE-OA05-06-162120T	77	111	84	40	40,4	1729	4280	b-f
TRE-OA05-06-122110T	77	111	88	45	39,7	1695	4270	b-f
TRE-OA05-06-173110T	75	109	73	45	41,7	1747	4190	b-f
TRE-OA05-05-151110T	75	110	85	38	40,6	1547	3810	c-g
TRE-OA05-05-231110T	75	108	86	34	39,5	1454	3680	c-g
TRE-OA05-01-211110T	76	109	86	40	38,0	1391	3660	c-g
TRE-OA05-05-231130T	74	107	85	43	48,7	1748	3590	d-g
TRE-OA05-02-231110T	73	106	77	31	42,5	1500	3530	efg
TRE-OA05-05-231120T	76	109	85	42	41,7	1430	3430	fg
TRE-OA05-04-141110T	73	106	75	39	48,5	1421	2930	g

CV (%) = 12,9 LSD = 891 kg/ha for seed yield.
*- Calculated as days starting from planting date
**- Based on 10 % moisture
***- At dry matter basis (0 % moisture)

Table 3. Performance of New Linoleic Safflower Lines in 2013

LINOLEIC LINES	Flowering Period (day)*	Physiological Maturity Period (day)*	Plant Height (cm)	1000 Seed Weight (g)**	Oil Content (%)***	Oil Yield (kg/ha)	Seed Yield (kg/ha)**	
LINAS (C)	76	109	94	49	39,9	2159	5410	a
DINCER (C)	76	108	88	50	29,2	1577	5400	a
Seledas-162	79	112	92	41	34,0	1717	5050	ab
REMZIBEY- 05 (C)	73	106	85	44	31,9	1544	4840	abc
Seledas-153	78	110	87	41	39,7	1910	4810	abc
TRE-LA05-02-211130T	76	108	81	46	34,0	1612	4740	abc
TRE-LA05-04-221110T	74	107	87	45	37,8	1788	4730	abc
BALCI (C)	75	109	80	45	39,5	1845	4670	bc
TRE-LA05-04-131110T	77	110	89	48	38,4	1747	4550	bcd
TRE-LA05-03-222110T	76	108	87	44	39,4	1690	4290	cde
TRE-LA05-03-111110T	75	107	85	47	36,7	1563	4260	cde
TRE-LA05-07-131110T	76	111	96	47	38,1	1501	3940	def
TRE-LA05-06-141110T	75	108	90	54	36,2	1390	3840	defg
TRE-LA05-08-241210T	75	109	84	43	38,4	1421	3700	efgh
TRE-LA05-01-321110T	76	108	84	39	35,5	1306	3680	efgh
TRE-LA05-05-121110T	76	109	90	50	38,8	1354	3490	fgh
TRE-LA05-04-111110T	76	108	83	49	39,1	1357	3470	fgh
TRE-LA05-05-121120T	74	106	91	50	33,5	1112	3320	fgh
TRE-LA05-01-111210T	74	106	79	55	33,4	1072	3210	gh
Seledas-160	76	108	76	35	48,1	1443	3000	h

CV (%) = 10,3 LSD = 716 kg/ha for seed yield.

*- Calculated as days starting from planting date

**- Based on 10 % moisture

***- At dry matter basis (0 % moisture)

References

Anonymous (2013a). Ministry of Food Agriculture and Livestock (www.tarim.gov.tr)

Anonymous (2013b). Turkish Statistical Institute, Annual Reports. (www.tuik.gov.tr)

Bagavan I and R L Ravikumar 2001. Strong undesirable linkages between seed yield and oil components-A problem in safflower improvement. Proceeding of the V International Safflower Conference, USA, July 23-27, pp: 103-107.

Corleto A, Cazzato E and Vetricelli P (1997). Performance of hybrid and O.P. safflower in two different Mediterranean environments. *Proceedings of the Fourth International Safflower Conference*. Italy, Bari, 2-7 June, pp. 276-278.

Dincer N 1964. Aspir (Safflower) Tarım Bakanlığı, Ziraat İşleri Genel Müdürlüğü Yayınları, D-102. 8 sayfa

Golkar P, Arzani A and Rezaei A M (2011). Determining relationships among seed yield, yield components and morpho-phenological traits using multivariate analyses in safflower (*Carthamus tinctorious* L.) Annals of Biological Research . Vol. 2 Issue 3, p. 162-169

Golparvar A R (2011). Genetic Improvement of Oil Yield in Spring Safflower Cultivars under Drought and Non-drought Stress Conditions *Electronic Journal of Biology*, Vol.7(2): 40-43

Harishbabu A N, Rudna Naik, V, Hanumantharaya L, Raju S G and Yaragoppa S D (2005). Evaluation of Promising Breeding Lines of Safflower for Alternaria Tolerance, Seed Yield and its Components. Karnataka Journal of Agricultural Sciences: 18 (3) p. 803-806.

Jamshi-Moghadam M, Pourdad S S 2006. Determination of safflower genotypes under moisture stress in controlled conditions and field. *Journal of Agricultural and Natural Resources* 10(2), 155-167.

Johnson R C, Ghorpade P B and Bradley V C 2001. Evaluation of the USDA core safflower collection for seven quantitative traits. V. International Safflower Conference. USA. pp. 143-149.

Mozaffari K and Asadi A A (2006). Relationships among traits using correlation, principal components and path analysis in safflower mutants sown in irrigated and drought stress condition. Asian. J. Plant. Sci, 5(6): 977-983.

Narkhede B N and Deokar A B 1990. Inheritance of spininess and pericarp types in safflower. J. *Maharashtra Agric. Univ*. 15, 279–281.

Pahlavani M H, Saeidi G and Mirlohi A F 2004. Inheritance of flower color and spininess in safflower (*Carthamus tinctorius* L.). J. Hered. 95, 265–267.

Ramachandran M 1985. Genetic improvement of yield in safflower – problems and prospects. *Journal of Oilseeds Research*, 2: 1-9

Rao V and Ramachandram M (1997). An analysis of association of yield and oil in safflower. *Fourth International Safflower Conference*. Italy, Bari, 2-7 June, pp. 185-191.

Zheng N, Futang C and Xinchun S, et al. (1993). Path analysis of correlated characters on flower yield of safflower, *Third Int. Safflower Conf.* Nijing, China, 582-588.

Review Article

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A promising fruit: Cherry laurel (*Prunus Laurocerasus* L.) and steps on breeding

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ABSTRACT

Cherry laurel (*Prunus laurocerasus* L.), is a fruit native to the regions bordering the Black Sea in Southwestern Asia and Southeastern Europe and widely spread out in the North part of Turkey. There are many cultivars which show different characteristics in Turkey and germplasm provides a rich parental material for crossing opportunities. The cherry laurel tree has pleasant fruits when fully ripe and also is a valuable fruit for industrial uses. Production of this fruit has increased over time and evaluation possibilities are varied. The detailed information about characteristics and nutritional value of cherry laurel was obtained from earlier studies and served for breeding of new cultivars. Studies on breeding of this species are focused on selection of superior types, resistance to disease, environmental adaptability, and molecular mapping of these types. This review aims to create a perspective to results of undergoing studies and discusses an overview of breeding opportunities of cherry laurel.

Keywords**:** *prunus laurocerasus*, breeding, selection, moleculer studies, biological studies, propagation

Introduction

Cherry laurel (*Prunus laurocerasus* L.) is an evergreen shrub or small tree up to 6 m; the leaves are dark green 5-15 cm in length and 4-10 cm in width and is naturally grown around the Black Sea Region, especially in North and South Anatolia (Kolayli et al. 2003). Cherry laurel has the chromosome number 2n = 22x = 176 (Contreras and Meneghelli 2013). Seeds of cherry laurel easily are spreading by birds which have led to concern regarding its increasing potential. The fruit is a small cherry, turning red to black when ripe. It is mostly consumed as fresh fruit in local markets, and the fruit of the cherry laurel is used in making jam, pickle, and cake; it is also eaten as dried and become more popular in the recent years (Sulusoglu and Cavusoglu 2011a). As a result of increasing market value, the demand for the fruit by producers and processing industry has increased which is an opportunity for the developing commercial cultivars for cherry laurel. Tapping germplasm resources to improve cultivated plants depends on introducing natural variability through traditional and biotechnological breeding methods (Bridgen 1994). Selection is basic of the fruit breeding and is the corner-stone upon. Studies have focused on selection of high productive cultivars of cherry laurel and determining of characteristics of the selected types, as well as propagation methods economically too. Herewith follows a brief overview of the main topics regarding cherry laurel breeding.

The first step of the breeding studies

Selection of superior cultivars

Efforts of characterization studies of cherry laurel can be traced into the late of 1900s, in which the incredible advances have been achieved for the last 20 years in terms of cultivar development. Studies have been conducted to determine promising cherry laurel types in the different regions of Turkey and trees with different ages at different locations under various growing conditions were identified and some of their pomological and chemical traits were determined

(Table 1). Studies on the selection of superior types have provided valuable information to help select the appropriate types in breeding programs for targeted cultivar development.

The first study reported in the literature was conducted by Özbek (1952) in Giresun forest area. Özbek made a travel in the Black Sea Region and investigated tree and fruit characteristics of this specie. Especially 'Fındık' cultivar grow in Giresun was evaluated briefly. Other studies were conducted in Trabzon, Rize, Ordu, Samsun, Sakarya and Kocaeli cities of Turkey (İslam 1996; Karadeniz and Kalkışım 1996; Islam and Odabas 1996; Bostan and İslam 2003; Akbulut et al. 2007; Islam and Vardal 2009; Beyhan 2010; Sulusoglu 2011b; Macit and Demirsoy 2012; İslam and Deligöz 2012) that all of the sites has Black Sea Region characterized by clay-loam soil tex ture with temperate climate in which winters are not very cold and summers are warm-hot with rare spring frosts.

Fruit quality in general has been a driving force in selection studies efforts. Fruit size and weight is important criteria to select of commercial types. The acceptable cherry laurel is sweet, with little astringent taste and rich color when ripe (Sulusoglu and Cavusoglu 2011a). The fruit has a seed inside and flesh to seed ratio is important criteria for the selection of types to industrial use in the future studies.

The modified weight rank method with rates from satisfactory to undesirable with a relative scale for every character (Macit, 2008; Beyhan, 2010; Sulusoglu, 2011b; İslam and Deligöz 2012; Macit and Demirsoy, 2012) has been used to determine the superior types. These selection criteria used for cherry laurel is tree productivity criteria (yield per trunk diameter area, yield per tree, fruit number per cluster, fruit harvesting time), fruit characteristics (fruit weight, width or length of fruit, fruit firmness, flesh/seed ratio, appearance of fruit, fruit uniformity) and fruit chemical characteristics (soluble solid contents, astringency, fruit taste, titratable acidity).

Pomological studies indicated that genotypes selected as promising had showed variability in the different areas of Turkey. Most of the studies were carried out in the different ecological conditions and under different ages of tree that were managed under different cultural conditions. All of these factors negatively affected the productivity of the trees and fruit quality. Only one experiment was conducted under the controlled conditions, in Samsun (Macit and Demirsoy 2012). Selected cherry laurel types from different place of the Black Sea region were planted in an orchard in Samsun and phenological and pomological analysis completed and some of these genotypes were suggested as a raw material for the breeding programs. Adaptation studies need to be organized in the other potential areas which are the second phase of the selection studies. Orchard performance of the selected types will give better results for selection of promise types as a cultivar.

Less known fruit species received much attention for their health benefit substances including antioxidans, total phenolics and fatty acids. Cherry laurel fruit serve as a good source of natural

Table 1. Pomological and chemical characteristics used in the selection of Cherry laurel types

Study area	Pomological and chemical character									Literature
	Cluster length (cm)	Fruit number/ cluster	Fruit weight (g)	Fruit width (mm)	Soluble solids (%)	Titratable acidity	Flesh seed ratio	Fruit taste	Astringency	
Samsun	-	+	+	+	+	+	+	+	+	Macit and Demirsoy, (2012)
Ordu	+	-	-	+	+	+	-	+	+	İslam and Deligöz (2012)
Kocaeli	+	+	+	+	+	+	+	+	+	Sulusoglu (2011b)
Sakarya	-	+	+	+	+	-	-	-	-	Beyhan (2010)
Rize	+	+	+	+	+	+	-	+	+	Islam and Vardal (2009)
Samsun	+	+	+	+	+	+	+	+	+	Macit (2008)
Samsun	-	+	+	-	+	+	+	-	-	Akbulut et al. (2007)
Trabzon	+	+	+	+	+	+	-	+	+	Bostan and İslam (2003)
Trabzon	-	+	+	-	-	-	+	-	-	Islam and Odabaş (1996)

antioxidant (Alasalvar et al. 2005; Kolayli et al. 2003; Halilova and Ercişli 2010).

Much interest has been developed in the last ten years on the antioxidant content of cherry laurel (Halilova and Ercişli 2010; Kasım et al. 2011). Fresh fruit samples of cherry laurel were examined for their total anthocyanin content by using a spectrophotometric differential pH method and cherry laurel genotypes showed differences in their total anthocyanins levels (Kasım et al. 2011). Yaylacı-Karahalil and Şahin (2011), investigated different phenolic constituents and total antioxidant properties of cherry laurel in Trabzon. Phenolic constituents were measured by reverse phase-high performance liquid chromatography (RP-HPLC). Among the chemical characteristics, soluble solid contents and titratable acidity as a malic acid, were used as criteria for selection of the superior types (Table 1).

The nutritional properties of selected types are important for the breeding studies. Gas chromatography-mass spectrometry studies (Ayaz 1997) and HPLC method (Alasalvar et al. 2005) were used to determine the sugar content of the cherry laurel and six sugar forms were identified in fruits; these included monosaccharides (xylose, arabinose, fructose and glucose), trace amounts of disaccharide sucrose, and sorbitol and mannitol as sugar alcohols (Ayaz 1997; Alasalvar et al. 2005). Cherry laurel fruits contain high amount of fatty acids (Ayaz et al. 1997; Halilova and Ercişli 2010). Mineral composition content of cherry laurel fruits was investigated by Kalyoncu et al. (2013) and demonstrated that cherry laurel fruit was rich in potassium.

Studies to serve the breeding of cherry laurel

In general, cherry laurel is considered resistant to disease and is recognized as a species can be grown easily (Sulusoglu and Cavusoglu 2011a). Quaglia et al (2013) reported a new pathogen for cherry laurel that is named "*Diplodia seriata*" found in Italy, and they suggested that this pathogen is potentially dangerous for cherry laurel. Although usage of the cherry laurel is common in Turkey, there have been limited studies including some of cherry laurel demonstration orchards planted in Kocaeli University Arslanbey Vocational School, Black-Sea Agricultural Research Institution, and Atatürk Horticultural Central Research Institution to investigate the cultural practice and performance of cherry laurel.

The second step of the breeding studies

***Ex-vitro* propagation of cherry laurel plants**

Obtaining seedlings of fruit tree species in traditional breeding programs depends on stratification of seeds at low temperatures for several months and germinating the seed the following spring. Information on germination of cherry laurel seed was limited to germination stage (Young and Young 1992; Norman 1993). The study of pre-treatments and stratification time on seed germination of cherry laurel seeds indicated that seeds soaked in to hot water 90 days after stratification resulted highest germination percentage while application of GA_3 shortened the stratification time of seeds without endocarp (Sulusoglu and Cavusoglu 2014).

After selection, economical mass propagation of cherry laurel is the second step of the breeding program. Vegetative propagation is important in propagation of valuable material, because the genotype of cultivar and varieties is usually highly heterozygote, and the characteristics which distinguish them are often lost by seed propagation (Hartmann et al. 2002). Cherry laurel rooting depends on concentrations of indole-3-butiric acid (IBA) talc or solution application, rooting media, genotype and cutting collection seasons (Riberio et al. 2010; Yazici et al. 2009; Posta 2009; Sulusoglu and Cavusoglu 2009). Semi-hardwood cuttings of sixteen cherry laurel types, selected following pomological studies (Sulusoglu, 2011b) rooted successfully by applying IBA and hence superior rooting genotypes were identified for economic commercial orchard (Sulusoglu and Cavusoglu 2010).

***In Vitro* Studies**

Successful results of micropropagation are very important for the future of breeding studies. The micropropagation approach could be used for genetic transformation, for self-rooted or grafted viral free material production in nurseries that *in vitro* based meristem culture/heat therapy protocol was effective for virus elimination (Kalinina and Brown 2007). *In vitro* proliferation, which can be carried out throughout the year, could provide an alternative, rapid means to produce more clonal material in a shorter period. Initiation cultures were prepared in Murashige and Skoog (MS) medium supplemented with different concentrations of BAP and NAA (Ponchia, 1991) and the maximum shoot proliferation was achieved with 0.5 mg/l BAP and 0.01 mg/l NAA hormone combination. Rooting of micro shoots was better when supplied with naphthalene acetic acid (NAA) than IBA in GA_3 including rooting medium.

Kalinina and Brown (2007) used axillary shoot meristem of cherry laurel to establish *in vitro* cultures. The best Meristem proliferation media was MS consisted of MES (2-(N-Morpholino)-ethanesulphonic acid) and supplemented with 0.5 mg/l BA+0,5 mg/l IBA+2,0 mg/l GA_3. Rooting of micro shoot were achieved in two stage; firstly rooting was

inducted in media including IBA for 4 days and then transferred to IBA-free media for next elongation period. Micro plantlets showed high survival rate in soil. In another study, micropropagation of cherry laurel was achieved with shoot-tip culture (Sulusoglu and Cavusoglu 2013a) in which 2.0 mg/l BAP gave the maximum shoot induction with 4.0 explant in the establishment culture and shoot elongation was better too. Shoot proliferation rate and length of shoots increased when IBA used together with BAP that provided extensive shoot growth and resulted in better rooting in the next step again with IBA. This micropropagation protocol can be used for maintenance of clonal propagation of cherry laurel.

One biotechnological technique that has been beneficial is embryo culture. Embryo culture is a valuable *in vitro* technique for breeding. It is most often used to rescue embryos from interspecific and intergeneric crosses for the material that has importance for breeders. The method also can be used to rescue seedless triploid embryos, produce haploids, overcome seed dormancy, or determine seed viability; useful for understanding embryo morphogenesis and precocious germination (Bridgen 1994). The culture of mature embryos from ripened seeds is used to eliminate seed germination inhibitors or to shorten the breeding cycle, to understand the physical and nutritional requirements for embryonic development (Hu and Wang 1986; Dunwell 1986). Embryo culture can shorten the breeding cycle by overcoming dormancy in seeds. Seed dormancy factors may be localized in the seed coat, the endosperm, or both. Isolating and culturing the embryos in aseptic conditions allows germination and fast growth of embryo that in turn shortens the breeding cycle. Isolated embryos can also be vernalized and this reduces the generation time (Sharma and Gill 1983).

So far the studies have been conducted on embryo culture of cherry laurel are scanty. In one study, mature embryos of cherry and black types of cherry laurel were cultured on MS media supplemented with BAP and IBA hormone combinations. Shoot formation was not observed in cherry type embryos while black type embryos produced seedlings with rudimentary roots which did not show any growth even after transfer to the fresh media. Cold stratification played an important role on the growth of embryos in the same hormone combinations in MS media. Embryo germination and seedlings growth both increased in embryos stratified for 60 days while embryos stratified for 30 days required high hormone concentration to continue to next growth and this protocol would be useful in future breeding program of cherry laurel. Embryo culture of cherry laurel prevented loss of material during germination stage and shortened the time to obtain seedlings in the same season the seed is collected (Sulusoglu 2012).

Molecular studies

The taxonomic classification within the genus *Prunus* which is mainly based on fruit morphology has been controversial. The revised classification by Rehder (1940), which describes five subgenera; *Amygdalus, Cerasus, Laurocerasus, Padus* and *Prunus* to accommodate variation within the genus, is widely accepted taxonomic group. The subgenera *Padus* and *Laurocerasus* in which cherry laurel is placed in the group is more isolated within the genus *Prunus* (Aradhya et al. 2004). Knowledge of the genetic diversity and relationships among the cultivated species of *Prunus* is important in recognizing the gene pools. Traditional taxonomic classifications provide rough guidelines to species relationships, but molecular evaluations provide detail information about the genetic structure and differentiation within and among taxa useful for plant breeders and geneticist.

Conservation of plant material and truly selection of superior genotypes before cultivation are highly important when used in breeding and gene transfer. If the selected superior types show genetic similarity due to climatic and environmental differences, then repetition will be avoided at the beginning of the breeding studies.

Optimization of SSR fingerprinting technique for species and type level is very important. The first genetic fingerprinting analysis among the 40 cherry laurel genotypes distributed in 6 location of the East Marmara Region of Turkey were reported by Hajyzadeh et al (2013). In this study, genetic similarities and differences in naturally growing 40 cherry laurel genotypes, two sweet cherry cultivars and one laurel type were determined. A total of 12 SSR primers developed from peach, sweet and sour cherry, plum or apricot were used. The eight primer pairs were found to be polymorphic among the cherry laurel genotypes. Cluster analysis showed that assessed genotypes were divided into two major group and several sup-groups while the resolution within the groups remained unsolved.

Before SSR technique, different researchers studied the genetics of cherry laurel using RAPD technique (Sandalli et al, 2005; Lee and Wen, 2001; Bortiri et al, 2001; Turkoglu et al, 2010). Microsatellites have been extensively used in *Prunus* genetics investigations for germplasm characterization (Lacis et al. 2009), determination of genetic diversity (Wünsch 2009), germplasm

management (Cheng and Huang 2009), cultivar identification (Xuan et al. 2009), and mapping genetic linkage (Lalli et al. 2008) in the last decade. If a microsatellites determined in a one species of *Prunus* could be used in the other species that has a transferability and ability to detect polymorphism (Wünsch 2009). Turkoglu et al. (2010) analyzed 10 SSR loci, previously identified for *Prunus* to examine genetic relationship among 23 rootstock candidates belong to different *Prunus* species including five genotypes of *P. laurocerasus* which were grouped in the second sub-cluster of first cluster together with *P. avium* and *P. cerasus* rootstocks. Unweighted pair-group method of arithmetic mean analysis demonstrated that *P. laureocerasus* genotypes had less genetic variation.

A sterile form of common cherry laurel would be useful in curbing its escape from cultivation. Contreras and Meneghelli (2013) attempted to induce polyploidy using *in vitro* exposure of "Otto Luyken" shoots to oryzalin with the objective of developing sterile form of cherry laurel. Shoots were treated for 1, 2, 14, or 28 days with 0, 6.25, 12.5, 25, 50, 100, or 150 µM oryzalin. Ploidy level of surviving shoots was determined using flow cytometer analysis of DAPI stained nuclei. As expected, when each meristem was analyzed individually, there was a reduced number of mixoploids, as more of the separated meristems were 22x and 44x. Here the target of developing sterile forms should be clearly discussed using those polyploids.

Biological studies

In the fruit trees pollen quality consists of viability, morphological homogeneity, pollen germination and pollen tube growth rate which are very important component of fertilization and fruit setting; therefore, study of pollen traits is one of the most important approaches for growers and breeders. *P. laurocerasus* L. is specie that needs a pollinator for fruit setting and pollination is very important for quality of fruits (unpublished data, Sulusoglu and Cavusoglu). The study was carried out to determine *in vitro* pollen viability and pollen germination of cherry laurel and the result showed that the viability was changed according to types and tests used. IKI test gave more clear and well-pointed results considerable. Fifteen percent sucrose gave the best pollen germination rate for most of the types (Sulusoglu and Cavusoglu 2013b).

Conclusion

Cherry laurel is a promising fruit for the future and breeding step is going well to selection of superior types and to improve the genotypes for production quality. The physical and chemical characters of cherry laurel types have underlined an interesting variability. Molecular studies could support truly selection of superior genotypes before cultivation and conservation of plant material. Selected types will serve as source of new cultivars of cherry laurel and propagation studies will enable to production of saplings for the new plantation areas. Biological studies will assist to make orchard design, selection and placement of the pollinator but studies at an early stage and needs to be improved. The particular interest given to this Turkey's natural fruit will improve the production diversification and food security, as well as sustainable crop production. As a result of the breeding studies, an alternative fruit crop will be obtained for the growers and the abundant high-quality curative fruit will be supported to the customers.

References

Akbulut M, Macit İ, Ercişli S and Koc A (2007). Evaluation of 28 cherry laurel (Laurocerasus officinalis) genotypes in the Balck Sea region, Turkey. New Zealand J. of Crop and Hort. Sci. 35:463-465.

Alasalvar C, Al-Farsi M and Shahidi F (2005). Compositional characteristics and antioxidant components of cherry laurel varieties and pekmez. J. of Food Science 70(1):47-52.

Aradhya M K, Weeks C and Simon CJ (2004). Molecular characterization of variability and relationships among seven cultivated and selected wild species of Prunus L. using amplified fragment length polymorphism. Scientia Hort. 103:131-144.

Ayaz FA (1997). Studies on water soluble sugar and sugar alcohol in cultivars and wild forms of *Laurocerasus officinalis* Roem. Pak. J. Botany 29(2):331-336.

Ayaz FA, Kadioğlu A, Reunanen M and Var M (1997). Phenolic acid and fatty acid composition in the fruits of *Laurocerasus officinalis* Roem. and its cultivars. J. of Food Composition and Analysis 10:350-357.

Beyhan O (2010). A study selection of promising native cherry laurel (*Prunus laurocerasus* L.) genotypes from Sakarya, Turkey. The J. of Animal and Plant Science 20(4): 231-233.

Bortiri E, Oh SH, Jiang J, Baggett S, Granger A,Weeks C, Buckingham M, Potter D and Parfitt DE (2001). Phylogeny and systematics of *Prunus* (Rosaceae) as determined by sequence analysis of ITS and the chloroplast *trn*L-*trn*F spacer DNA, Syst. Bot. 26 (4):797-807.

Bostan SZ and İslam A (2003). Pomological and phenological traits of local cherry laurel (*Prunus laurocerasus* L.) types grown in Trabzon province of Turkey. Journal of Faculty of Agriculture (OMU) 18:27-31.

Bridgen, P.B. (1994). A Review of Plant Embryo Culture. Hortscience, 29(11):1243-1245.

Cheng Z and Huang H (2009). SSR fingerprinting Chinese peach cultivars and landraces (*Prunus persica*) and analysis of their genetic relationships. Scientia Hort. 120:188-193.

Contreras R and Meneghelli, L. 2013. *In vitro* chromosome doubling of *Prunus laurocerasus* 'Otto Luyken'. ashs.org/abstracts, 13715. (http://archives.ashs.org/abstracts/2013/abstracts13/abstract_id_13715.html)

Dunwell JM (1986). Pollen, ovule and embryo culture as tools in plant breeding, p.375-404. (In: LA Withers and PG Alderson, eds, Plant Tissue Culture and Its Agricultural Applications Butterworths, London).

Hajyzadeh M, Cavusoglu, A, Sulusoglu M and Unver T (2013). DNA SSR fingerprinting analysis among cherry laurel (*Prunus laurocerasus* L.) types. J. of Food, Agriculture & Environment 11(2):630-638.

Halilova H and Ercişli S (2010). Several Physico-chemical characteristics of Cherry Laurel (*Laurocerasus officinalis* Roem.) Fruits. Bıotechnol. and Biotechnol.EQ.24(3).1970-1973.

Hartmann HT, Kester DE, Davis Jr FT and Geneve, RL (2002). Plant propagation: principles and practices. New Jersey:Prentice Hall.

Hu C and Wang P (1986). Embryo culture: Technique and applications 43-96. (In: DA Evans, WR Sharp and PV Ammirato (eds.). Handbook of Plant Cell Culture vol.4. Macmillan,New York.).

İslam A (1996). Improvement by selection of cherry laurel (*Prunus laurocerasus* L.) grown in Vakfikebir and its surroundings. Yuzuncu Yıl University, The Journal of Agricultural Faculty (Turkey) 6(4):147-158.

Islam A and Odabaş F (1996). Improvement by selection of cherry-laurel (*Prunus laurocerasus* L.) grown in Vakfikebir and its surroundings-I. Yuzuncu Yıl University, The Journal of Agricultural Faculty (Turkey) 6(4) p.147-158.

Islam A and Vardal E (2009). Pomological characteristics of cherry laurel (*Prunus laurocerasus* L.) Grown in Rize. Proc.1st IS on Pomogranate. Acta Hort. 818, ISHS 2009. p:133-136.

Islam A and Deligöz H (2012). Ordu İlinde karayemiş (*Laurocerasus officinalis* L.) seleksiyonu. Akademik Ziraat Dergisi 1(1):37-44.

Kalinina A and Brown D (2007). Micropropagation of ornamental prunus spp. and GF305 peach, a *prunus* viral indicator. Plant Cell Rep 26(7):927-935.

Kalyoncu İ, Ersoy N, Yalcın-Elidemir A and Dolek C (2013). Mineral and some physico-chemical composition of "Karayemiş" (*Prunus laurocerasus* L.) fruits grown in Northeast Turkey. World Academy of Science, Engineering and Techn. 78:1393-1395.

Karadeniz T and Kalkişim Ö (1996). Investigation on selection of cherry laurel (*Prunus laurocerasus* L.) grown in Akçaabat. Yuzuncu Yıl University, J. Agric. Fac. (Turkey) 6(1):147-153.

Kasım R, Sulusoglu M and Kasım U (2011). Relationship between total anthocyanin level and colour of natural cherry laurel (*Prunus laurocerasus* L.) fruits. African Journal of Plant science 5(5):323-328.

Kolayli S, Küçük M, Duran C, Candan F and Dinçer B (2003). Chemical and antioxidant properties of *Laurocerasus officinalis* Roem (Cherry laurel) fruit grown in the Blacksea Region. J. Agric. Food Chem. 51(25): 7489-7494.

Lacis G, Rashal I, Ruisa S, Trajkovski V and Iezzoni AF 2009. Assessment of genetic diversity of Latvian and swedish sweet cherry (*Prunus avium* L.) genetic resources collections by using SSR (microsatellite) markers. Scientia Hort. 121:451-457.

Lalli DA, Abbott AG, Zhebentyayeva TN, Badenes ML, Darmsteegt V, Poljk J, Krska B and Salava J (2008). A genetic linkage map for an apricot (*Prunus armeniaca* L.) BC1 population mapping Plum Pox virus resistance. Tree Genetics and Genomes 4:481-493.

Lee S and Wen J (2001). A phylogenetic analysis of *Prunus* and the *Amygdaloideae* (*Rosaceae*) using ITS sequences of nuclear ribosomal DNA. Am. J. of Bot. 88(1):150-160.

Macit İ (2008). Karadeniz Bölgesi Karayemiş (*Prunus laurocerasus* L.) Seleksiyonu II. Aşama, Yüksek Lisans Tezi (Master Thesis) (basılmamış). Ondokuz Mayıs Üniversitesi Fen Bilimleri Enstitüsü Bahçe Bitkileri Anabilim Dalı, Samsun p: 48.

Macit I and Demirsoy H (2012). New promising cherry laurel (*Prunus laurocerasus* L.) genotypes in Turkey. Bulgarian Journal of Agricultural Science 18(1):77-82.

Norman CD (1993). Seed Germination Theory and Practice, 2nd Ed. Baltimore p: 242

Özbek S (1952). Karayemiş (*Prunus laurocerasus* L.) A.Ü. Ziraat Fakültesi Yıllığı, 4, p.8

Ponchia G (1991). Research on *in vitro* propagation of *Prunus laurocerasus* cv. Otto Luyken. Acta Horticulturae 300:177-180.

Posta DS (2009). Vegetative planting material producing at the *Prunus laurocerasus* L. Bulletin UASVM Horticulture 661(1-2):204-208.

Quaglia M, Moretti C and Buonaurio R (2013) Molecular characterization of *Diplodia seriata*, a new pathogen of *Prunus laurocerasus* in Italy. Phytoparasitica,doi:10.1007/s12600-013-0350-9.

Rehder A (1940). Manual of cultivated trees and shrubs, 2nd. ed. MacMillan, New York, p. 452-481.

Riberio MM, Collado LM and Antunes MA (2010) The influence of indole-3-butyric-acid in *Prunus laurocerasus* vegetative propagation. Acta Hort. (ISHS) 885:277-283
(http://www.actahort.org/books/885/885_38.htm)

Sandall C, Beris F S, Canakci S, Demirbag Z and Beldüz AO (2005). RAPD analysis of three cultivars and a wild form in *Prunus laurocerasus* (Rosaceae). Biol. Brat. 60(1):83-87.

Sharma HC and Gill BS (1983) New Hybrids between *Agropyron* and wheat. 2. Production, morphology and cytogenetic analysis of F1 hybrids and backcross derivatives. Theor. Appl. Genet. 66:111-121.

Sulusoglu M and Cavusoglu A (2009). Cutting propagation possibilities of natural cherry laurel (*Prunus laurocerasus* L.) of Turkey. American Eurasian Journal of Sustainable Agriculturae 3(2):234-237.

Sulusoglu M and Cavusoglu A (2010). Vegetative propagation of Cherry laurel (*Prunus laurocerasus* L.) using semi-hardwood cuttings. African Journal of Agricultural Research 5(23): 3196-3202

Sulusoglu M and Cavusoglu A (2011a). Karayemiş Yetiştiriciliğinin Mevcut Durumu ve Kullanım Alanlarının Ortaya Konması. Türkiye 6. Ulusal Bahçe Bitkileri Kongresi, 4-8 Ekim 2011, Şanlıurfa.

Sulusoglu M (2011b). The cherry laurel (*Prunus laurocerasus* L.) tree selection. African Journal of Agricultural Research 6(15): 3574-3582.

Sulusoglu (2012). Development of embryo culture protocol for cherry laurel (*Prunus laurocerasus* L.)", J. of Food Agriculture & Environment 10 (3&4), 347-352.

Sulusoglu M and Cavusoglu A (2013a). Micropropagation of cherry laurel *Prunus laurocerasus* L. J. of Food, Agriculture & Environment 11 (1): 576-579.

Sulusoglu M and Cavusoglu A (2013b). *In vitro* pollen viability and pollen germination in *Prunus laurocerasus* L. (Cherry laurel). International Plant Breeding Congress, Abstract Book, International Plant Breeding Congress, Antalya-Turkey p.521.

Sulusoglu M and Cavusoglu A (2014). Effect of pre-treatments on seed germination of *Prunus laurocerasus* L. (Cherry laurel). 49th Croatian & 9th International Symposium on Agriculture, Dubrovnik-Croatia. Pomology, Viticulture and Enology p:722-726.

Turkoglu Z, Bilgener S, Ercisli S, Bakir M, Koc A, Akbulut M, Gercekcioglu R, Gunes M and Esitken A (2010). Simple sequence repeat-based assessment of genetic relationships among Prunus rootstocks. Genet. Mol. Res. 9(4):2156-2165.

Wünsch A (2009). SSR markers for fingerprinting Prunus species. Acta Hort. 814:689-694.

Xuan H, Wang R, Büchele M, Möller O and Hartmann W (2009). Microsatellite markers (SSR) as a tool to assist in identification of sweet (*Prunus avium*) and sour cherry (*Prunus cerasus*). Acta Hort. (ISHS) 839:507-514.

Yaylacı-Karahalil F, Şahin H (2011). Phenolic composition and antioxidant capacity of Cherry laurel (*Laurocerasus officinalis* Roem.) sampled from Trabzon region, Turkey. African Journal of Biotechnology 10(72):16293-16299.(DOI: 10.5897/AJB11.1929)

Yazici K, Dal B, Gozlekci S, Kaynak L and Ersoy N (2009). Effects of cutting type and duration time on rooting of three cherry laurel (*Prunus laurocerasus* L.) genotypes. Acta Hort. (ISHS) 818:199-204.

Young J A and Young CG (1992). Seeds of Woody Plants in North America. Dioscorides Press, Oregon, Portland.

Research Article

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"BATEM Göral": New mandarin cultivar

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ABSTRACT

Citrus is the most widely produced fruit group in the world. Approximately 3 million tons of citrus fruit are produced in Turkey. With total export of one million tons, our country is ranking as the 4th Citrus exporter in the world and ranking as the 2nd among the Mediterranean countries. Mandarin production is 874.832 tons in Turkey and 8,72% of total mandarin production was Clementine mandarin in 2012. From different citrus ecological regions top selection of qualified individuals at "Citrus Budwood Selection-Certification and Variety Development Project" 7 promising types were selected from the Clementine mandarin types and BATEM Göral has been registered as new mandarin cultivar in 2011. BATEM Göral have been compared with Clementine mandarin in distinctness, uniformity and stability tests (DUS). In study results showed that 'BATEM Göral' has larger fruit weight and diameter.

***Keywords*:** mandarin, Clementine, variety, register, selection

Introduction

The citrus have major importance in the world and Turkey. Citrus is the most extensively produced tree fruit crop in the world. The increase of citrus world annual production reached more than 130 million tons (FAO 2012).

Natural mutation is very common in citrus. Many of world's most important cultivars have arisen through somatic mutation. The citrus industry of the world is highly dependent on few varieties such as 'Washington Navel', 'Valencia', 'Shamouti', 'Pera', 'Hamlin' oranges, 'Marsh Seedless' grapefruit, easy peeling mandarins such as 'Satsuma', 'Clementine' (Spiegel-Roy and Goldschmidt 2003). Most of present scion and rootstock cultivars of citrus are the progeny of chance seedlings or a mutant branch of a tree, called 'budsport'. The commercially successful cultivars now grown have resulted from the selection, propagation and advanced testing of thousands of such superior chance seedling (Khan and Kender 2007). The first formal citrus breeding programme was started by USDA in Florida in 1893 (Cooper et al. 1962). Today, there are numerous citrus breeding programmes spread in all major citrus-producing countries (Roose and Williams 2007).

Clementine first appeared at the beginning of the last century in the garden of an orphanage in Algeria as a natural cross between mandarin and sweet orange. Clementines are now the main mandarins in the Mediterranean area and also are being grown in several countries of the Americas and South Africa. Clementine (*Citrus clementina* Hort. ex Tan.) was classified as a Citrus species (Tanaka 1977). Currently, this species is one of the most important mandarin hybrid especially in the Mediterranean countries due to its good fruit quality and flavour, high yield, easy peeling (Uzun and Yesiloglu 2012). Many Clementine clones with high quality and different maturity time were obtained from clonal selection and most of them registered as new cultivars. Bud mutations often arise in Clementine, as it is the case also for orange and Satsuma mandarin, which are generally detected by the growers in branches of trees showing altered horticultural traits, such as maturity

and flowering time, or fruit characteristics (Breto et al. 2001).

Material and methods

DUS tests (UPOV 2003) were carried out at the Fruit Department in Bati Akdeniz Agricultural Research Institute. Clementine (Algerian) and KLA 69 clone were used as material. The following yield and fruit quality characteristics were studied on the cultivars: yield (kg/tree), fruit weight (g), fruit length (mm), fruit diameter (mm), rind thickness (mm), number of seeds, number of segments, Juice (%), total soluble solids % (TSS), titratable acidity % (TA) and ratio of TSS/TA.

The study was conducted in a randomized plot design. All data were analyzed statistically using analysis of variance techniques and the means were separated by LSD's test.

Results

Results regarding the fruit weight and fruit diameter revealed a significant difference between Clementine and KLA 69 clone mandarin ($p \leq 0.05$). Means for other fruit quality characteristics were not found significant (Table 1).

Table 1.Yield and pomological analysis results in mandarin cultivars

Characteristics	'BATEM Göral' (KLA 69)	Clementine (Algerian)	Significance
Fruit weight (g)	104.68 a*	85.13 b	LSD: 9,73
Fruit lenght (mm)	53.32	50.20	ns
Fruit diameter (mm)	62.18 a	56.74 b	LSD:1,24
Rind thickness (mm)	2.40	2.06	ns
Number of segments	9.30	9.10	ns
Number of seeds	6.93	6.80	ns
Juice (%)	52.21	53.20	ns
TSS %	11.70	11.80	ns
Titratable acidity(TA)%	0.82	0.88	ns
TSS/TA	14.36	13.53	ns
Yield (kg/tree)	130.10	108.00	ns

* Different letters indicate significant differences ($P<0.05$) according to the LSD tests

Discussion

Top selection of qualified individuals at "Citrus Budwood Selection - Certification and Variety Development Project" that carried out between 1979 and 1984 were selected 7 promising types from the Clementine mandarin types from different citrus ecological regions in Turkey. KLA 69 clone was compared to Clementine mandarin. Clementine mandarins of various types and varieties are grown in worldwide. Based upon several years of observation and DUS testing, it was registered as 'BATEM Göral' and released as a superior mandarin cultivar with fruit weight and fruit diameter in 2011. 7 promising types from the Clementine mandarin selections were evaluated by Turgutoglu et al. (2011). KLA 69 clone was selected using weight-ranked method. This cultivar is recommended for commercial cultivation throughout the citrus-growing areas in Turkey. Wright (2007), Uzun et al. (2011) and Georgiou (2002) studied in different Clementine mandarin varieties.

References

Breto MP, Ruiz C, Pina JA and Asins MJ (2001). The diversification of Citrus clementina Hort. ex. Tan, A vegetatively propagated crop species. Mol. Phylo. Evol.21: 285-293.

Cooper WC, Reece PC and Furr JR (1962). Citrus breeding in Florida- past, present and future. Proceedings of the Florida State Horticultural Society75:5-13.

FAOSTAT (2012) http://faostat.fao.org/site/567/default.aspx#ancor Accessed 15 September 2013.

Georgiou A (2002). Evaluation of rootstocks for 'Clementine' mandarin in Cyprus. Scientia Horticulturae ; 93(1):29-38. DOI: 10.1016/S0304-4238(01)00311-9.

Khan IA and Kender WJ (2007). Citrus Genetics, Breeding and Biotechnology. In: Citrus Breeding, Khan, I (ed.), CAB Int. Wallingford, pp:1-5. http://books.google.com.tr/

Roose ML and Williams TE (2007). Citrus Genetics, Breeding and Biotechnology. In: Mutation Breeding, Khan, I (ed.), CAB Int. Wallingford, pp: 345-350. http://books.google.com.tr/

Spiegel-Roy P and Goldschmidt EE (2003). Biology of Citrus. In: Genetic Improvement in Citrus. Cambridge University Press, pp: 190. http://books.google.com.tr/

Tanaka T (1977). Fundamental discussion of Citrus classification. Stud. Citrol. 14: 1–6.

Turgutoglu E, Tuncay M, Demirel H, Apaydın Y, Kurt S and Demir G (2011). Clementine Mandarin Variety Selection for Bati Akdeniz Region. VI. National Horticultural Congress in Turkey, Abstract Book:2.

UPOV (2003) http://www.upov.int/edocs/tgdocs/en/tg201.pdf. Accessed 20 October 2008.

Uzun A, Gulsen O, Kafa G and Seday U (2011). Determination of field performance and molecular discrimination of clementine mandarin selections. 2nd Balkan Symposium On Fruit Growing.

Uzun A and Yesiloglu T (2012). Genetic Diversity in Citrus, Genetic Diversity in Plants, Caliskan M (ed.), ISBN: 978-953-51-0185-7, http://www.intechopen.com/download/ get/type/pdfs/id/31479

Wright GC (2007). Mandarin Selection Trials in Arizona – 2006-07. Citrus rootstock and cultivar breeding and evaluation for the Arizona citrus industry – 2006-07. http://extension.arizona.edu/sites/extension.arizona.edu/files/pubs/az1441-08.pdf

Review Article

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Biotechnological approaches in strawberry tree (*Arbutus unedo* L.) breeding

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ABSTRACT

Arbutus unedo L. (Strawberry tree) belongs to *Ericaceae* family, is an evergreen shrub or tree, mostly known around Mediterranean region in natural habitat, has valuable medicinal and aromatic properties. The plant is mainly used for its edible fruits. In addition, *A. unedo* have increasing importance in afforestration programmes, beekeeping facilities, ornamental purposes and elucidating plant physiology. Therefore, the species is seemingly a promising fruit plant. Orchards are very limited but increasing demand to the plant will result in initiations of establishment new fruit orchards with superior genotypes viz. tolerant to the abiotic stress, resistant to pests and diseases, rich contents in terms of valuable compounds, convenience to postharvest operations, being visually preferable and suitable to target climate. Plant breeding is an important component to overcome elimination of unwanted features and to reach admirable characters. The main steps in breeding are effective selection, succesfully adaptation and inheritance via transferring the features to subsequent generations. Studies on *A. unedo* breeding via biotechnological approaches were reviewed. In *A. unedo* breeding, only few biotechnological techniques have used in all known for the purposes especially for selection and propagation of the determined best type. The mostly used biotechniques for *A. unedo* were consantrated on *in vitro* propagation and fingerprint analyses to determine genetic diversity and the favourite genotypes wherein the population with compared with pomological features for help in breeding programme.

Keywords**:** *arbutus unedo*, strawberry tree, breeding, biotechnology

Introduction

Breeding better cultivar has become a highly efficient way to improve plant production for yield, quality and reduced input (Andersen, 2011). The major objective of plant breeding programs is to develop new genotypes that are genetically superior to those currently available for a specific target environment or a target populaton of environments (Wang, 2011). In the first step, conventional plant breeding depends on phenotypic selection. For the purpose, plant breeders deal with certain selection methods. After the process, in the second step; fertilization, hybridization, measurements of inheritance, chromosomal threads, mutation, cell and tissue culture methods, genetic sources, develop new varieties, plant propagation, viz. methods are used to reach the artificial trade varieties. Biotechnological methods sometimes used to either ensure progress of breeding or continuity of the breeding programme. Most of the breeding studies have carried out in the field crop plants. Especially studies on cereals (Korkut et al, 2001; Akar et al, 2008), legumes (Altınbaş, 2003; Toker et al, 2012; Güllüoğlu et al, 2010), oilseed plants (Kaya et al, 2004; Uzun et al, 2003), fiber plants (Karademir et al, 2009), and sugar-starch plants (Barzen et al, 1992) share in the first place. Studies about breeding of horticultural plants (Bolat and Güleryüz, 1992; Pırlak et al, 1997), ornamental

and forestry plants (van Eijk and Leegwater, 1975; Hubert and Lee, 2005) and medicinal-aromatic plants (Arslan et al, 2002; Kumar and Gupta, 2008) always have come from behind although everyday we have used them as food, beverages, medicine, environment and ornamenal purposes.

Different geographical regions of Turkey allow fruit to be grown in almost every part of the country, and Turkey has become one of the main fruit production centers of the world (Küden and Küden, 2008). Because of being gene center of apricot, hazelnut, pistachios, fig and grape, the first studies have focalized on the plants in Turkey (Balkaya and Yanmaz, 2001). Scientific studies on fruit breeding (Bolat and Güleryüz, 1992; Pırlak et al, 1997; Uzun et al, 2008; Acar et al, 2013; Aksu, 2013; Ozdemir et al, 2013), especially on new promising fruit species have performed for a while.

Arbutus unedo, is one of the new promising fruits species, that is newly introduced and studies. The strawberry tree is an evergreen shrub or small tree (Figure 1a). In autumn, the white or pinkish flower clusters, along with the 20-25 mm orange fruits from the previous year (Figure 1b), contrast to the dark foliage (Gratani and Ghia, 2002). The plant natively takes place in Northern Africa (Algeria, Morocco,Tunisia); in Western Asia (Turkey, Lebanon, Syria, Cyprus); in Northern Europe (Ireland); in East Europe (Ukraine); in Southeastern Europe (Albania, Bulgaria, Croatia, Greece, Italy) and in Southwestern Europe (France, Portugal, Spain) (USDA-ARS-GRIN, 2013). According to our personal observation, the species is well-adapted in Turkey.

Although the main usage of the plant is fruit consumption, the fruits or leaves have valuable food content and medicinal and aromatic properties (Ayaz et al,2000; Pawlowska et al, 2006; Males et al, 2006; Fortalezas et al, 2010; Ruiz-Rodriguez et al, 2011; Özcan and Hacıseferoğulları, 2007; Pabuçcuoğlu et al, 2003). The plant can be used for recovery after forest damage or burning via lignotuber (Canadell and Lopez-Soria, 1998; Konstantinidis et al, 2006) for ornamental purpose (Metaxas et al, 2008) via leafy branch in cut-flowers or via habitus and flowers in plantation in gardens and for beekeping facilities (Dalla Serra et al, 1999; Rasmont et al, 2005). At the same time the evergreen *A. unedo* was subjected to plant physiology studies (Panicucci et al, 1998; Navarro et al, 2007) to reach conclusions on photosynthetic response or water usage under different treatments.

Firstly, on the way of conventional plant breeding, *A. unedo* selections were performed depending on pomology, phenology and morphological features of selected different plants or selected different populations (Karadeniz et al, 2003; Şeker et al, 2004; Yarılgaç and İslam, 2007; Celikel et al, 2008; Sulusoglu et al, 2011; Molina et al, 2011). Unfortunately other conventional plant breeding methods could not be reached in this plant.

The purpose of this review was to put out the biotechnological steps conducted to help *A. unedo* breeding that may be progressed.

In vitro tissue culture techniques

The techniques include studies on *in vitro* fertilization, embryo culture and protoplast fusion containing wide hybridization method, haploids method, somaclonal variation method and studies on propagation of plants, synthetic seed, pathogen eradication and germplasm preservation containing micropropagation topic (Brown and Thorpe, 1995). For this purpose very few studies have been reached about A. unedo.

One of the firsts was about *in vitro* breaking of dormancy of axillary buds (Rodrigues et al, 2001). The researchers used nodal segments from adult plant in November and February considered for experiment, corresponding to bud dormancy and breaking bud dormancy period respectively. At the same time they studied the effect of cutting method (surgical blade or laser cut). For the study, determined medium was used containing zeatin and dithiothreitol under certain time light/dark photoperiod. It was reported that, the laser cut method considered for overcoming problems with *in vitro* micropropagation of *A. unedo* in winter which enables higher productivity through the year.

In another study, micropropagation of *A. unedo* was achieved (Mereti et al, 2002) and the plantlets were succesfully acclimatized. For this purpose, nodal segments from defoliated actively growing shoots were cultured in medium containing benzyladenine for shoot culture and indole-3-butiric acid or indole-3-acetic acid for root culture under photoperiod. In the rooting experiment medium with or without peat:perlite (1:4 v:v) mixture to addition at a 1:1 (v:v) ratio were also tested. Apart from the mentioned study, a very similar study on *in vitro* rooting was published in 2003 (Mereti et al, 2003).

On the other hand Canhoto et al. (2007) obtained somatic embryos in all stages from *A. unedo* leaf explants cultured in a modified medium with benzyladenine and naphthalene acetic acid.

Gomes and Canhoto (2009) studied on micropropagation of six selected adult strawberry trees. The study was started with shoot apices and nodal segment under dark condition for a week, and after then photoperiod condition. Different

compounds added into the main medium for plant formation were used in the study. Several factors of the experiment were detected. One of the important result was observed is that shoot multiplication might be influenced by the genotypes. Some clones were found difficult to propagate and this indicated being related with the levels of endogenous growth regulators in plant genotypes.

Gomes et al, (2009) studied on micropropagation of selected *A. unedo* trees and somatic embryogenesis. The importance and difference of the study was achievement of somatic embryo conversion.

According to another study (Gomes et al, 2010), effect of different combinations of benzyladenine or other cytokinins or naphthalene acetic acid with benzyladenine interaction with genotypes were subjected. The results indicated that multiplication rate depends on the *A. unedo* genotypes and plant growth regulators types.

Gomes (2011) performed another *in vitro* study on testing mycorrhizae inoculation that can improve plant adaptation and tolerance to stressful conditions. Results and implications of the study on *A. unedo* breeding program were discussed.

El-Sayed El-Mahrouk et al. (2010) aimed at their study to develop an efficient method for propagation of *A. unedo* through adventitious shoots and somatic embryogenesis using segments of *in vitro* obtained axillary shoots. Axillary shoots induction were induced in plant growth regulator free medium. When thidiazuron added, medium gave the best for shoot multiplication. Embryogenic callus and somatic embryos on the calli were obtained from internodal segments at relatively higher concentration of benzyladenine and naphthalene acetic acid combination. After then plantlets continued to next development in hormone-free media.

Plant genetics and molecular biology techniques

The analysis of the genetic diversity of *A. unedo* takes the first place among the researches in this topic. To evaluate genetic diversity or genetic similarity -in association with phenology, morphology, physiology, geograpy and/or ecology-, molecular markers are useful instruments in plant improvement and breeding via determination certain genotypes.

Genetic diversity among the nine Tunusian *A. unedo* populations were assessed using sixty five polymorphic RAPD loci (Takrouni and Boussaid, 2010). According the results of the study, three groups of the populations analysed and the groups were not related with geographical or bioclimatical origin.

In another genetic variability study, DNA extraction from leaves of *A. unedo* was optimised first (Sa et al, 2011) because concentration, purity and quality of the extracted DNA is important. After the procedure, the DNA samples were used in molecular analysis based on both RAPD and ISSR to indicate diversity (Lopes et al, 2012). Results proved that there were high levels of gene flow among populations so this caused low differentiation. The cluster analysis showed that only one population formed a distinctive cluster, remaining formed a second cluster due to their geographical approach.

Gomes et al. (2013) have analysed the genetic relationship among twenty seven *A. unedo* genotypes from a broad geographic range using twenty RAPD and eleven SSR markers. Results indicated that no correlation was found between the markers or geographical origin.

As distinct from the above experiments, Zamboni et al. (2008) extracted the total RNA from *Arbutus unedo* and several other woody-plants for gene expression analysis as represented non-model species for molecular biology. Expression of the agricultural important gene will lead to the progress in *Arbutus unedo* breeding.

Conclusion

On behalf of biotechnology, significant progress has been made in plant breeding using cell or tissue culture *in vitro* and studies or manupulation in genetics and molecular techniques of plants, in particular in crop plants. *A. unedo* is a newly promising horticultural species so few studies have conducted on the way of its breeding. For the present, conventional selection with the help of studies on genetic similarities, and micropropagation of the choosen superior types came to the fore. In addition to these before studies in breeding of A. unedo, next studies can be expected to be subject and/or progress about haploids in crosses, fertilization, changes in ploidy, screening of diseases, stress etc. conditions, rootstock breeding, breaking seed or bud dormancy, obtaining variation via cell, meristem, anther, callus and protoplast cultures, somatic and zygotic embryogenesis and cryopreservation etc. in *in vitro*. At the same time breeding in *A. unedo* can cause development new cultivar in pest, disease, weed, pesticides-resistance, bringing in physiological behavior as photosynthesis, nutritional storage, nitrogen fixation, postharvest resistance positively and gene silecing etc. topics in genetics and molecular biology as achieved previosly in certain plants. This point should be emphasized again that studies on *A. unedo* breeding (conventional or high-tech applications) are in the spring of its study life when compared with numerous field and horticultural plants

Figure 1. (a) *Arbutus unedo* L. in natural habitat, (b) One of the *Arbutus unedo* L. genotype studied by Sulusoglu et al, (2011) selection studies.

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References

Acar I, Arpaci S, Yilmaz A, Atli HS, Kafkas S, Eti S, Caglar, S 2013. A new almond breeding program in Turkey, Acta Hort. (ISHS) 976:63-68.

Akar T, Sayim I, Ergun N, Aydogan S, Sipahi H, Avci M, Dusunceli F, Guler S, Sanal T, Ayranci T, Kahraman R, Ince T, Aydin N, Kendal E, Engin A 2008. Improvement of candidate malting barley cultivars for diverse agro-ecologies of Turkey, Proceedings of 10th International Barley Genetics Symposium, Egypt (Eds.Salvatore Ceccarelli and Stefania Grando) 443-446.

Aksu M (2013). Red Flesh Apples and Breeding Studies, International Plant Breeding Congress, 10-14 November, Antalya, TURKEY(Abstract book), p. 535.

Altınbaş M (2003). Possibilities of using biological and harvest index in winter sown chickpea (Cicer arietinum L.) breeding, Anadolu J. of AARI, 13(1): 46-57. (in Turkish)

Andersen SB (2011). Virtual plant breeding, (In Plant Breeding, Eds. I.Y. Abdurakhmonov, Intech Open Access Publisher, Rijeka, Crotia) 3-18.

Arslan N, Gürbüz B, İpek A (2002). The Cultivation and Breeding Studies on Some Medicinal and Aromatic Plants in Ankara Conditions, Proceedings of the Workshop on Agricultural and Quality Aspects of Medicinal and Aromatic Plants: 17-27p, May 29-June 1 (2001), Adana / Turkey.

Ayaz FA, Kucukislamoglu M, Reunanen M (2000). Sugar, Non-volatile and Phenolic Acids Composition of Strawberry Tree (*Arbutus unedo* L. var. ellipsoidea) Fruits, Journal of food Composition and Analysis, 13(2): 171-177.

Balkaya A, Yanmaz R (2001). Conservation facilities of plant genetic resources and working systems of seed gene banks, Ekoloji, 39: 25-30. (in Turkish)

Barzen E, Mechelke W, Ritter E, Seitzer JF, Salamini F (1992). RFLP markers for sugar beet breeding: chromosomal linkage maps and location of major genes for rhizomania resistance, monogermy and hypocotyl colour, The Plant Journal, 2(4): 601-611.

Bolat S, Güleryüz M (1992). Konya İlinde Kaliteli Yazlık Elma Tiplerinin Seleksiyon Yolu İle Islahı Üzerinde Bir Araştırma, Türkiye I. Ulusal Bahçe Bitkileri Kongresi, Cilt 1:523-526. (in Turkish)

Brown D C, Thorpe T A, (1995). Crop improvement through tissue culture, World Journal of Microbiology and Biotechnology, 11: 409-415.

Canadell J, Lopez-Soria L (1998). Lignotuber reserves support regrowth following clipping of two Mediterranean shrubs, Functional Ecology, 12: 31-38.

Canhoto JM, Lopez ML, Sequeira J, Gomes F (2007). Somatic embryogenesis and organogenesis

induction from adult trees of *Arbutus unedo* and *Arbutus canariensis*, IUFRO Tree Biotechnology, Azores, p.12.

Celikel G, Demirsoy L, Demirsoy H (2008). The strawberry tree (*Arbutus unedo* L.) selection in Turkey, Scientia Horticulturae, 118: 115-119.

El-Sayed El-Mahrouk M, Dewir YH, Omar AMK (2010). *In vitro* propagation of adult strawberry tree (*Arbutus unedo* L.) through adventitious shoots and somatic embryogenesis, Propagation of Ornamental Plants, 10(2): 93-98.

Dalla Serra A, Franco MA, Mattivi F, Ramponi M, Vacca V, Versini G (1999). Aroma characterization of Sardinian strawberry tree (*Arbutus unedo* L.) honey, Italian Journal of Food Science, 11 (1): 47-56.

Fortalezas S, Tavares L, Pimpao R, Tyagi M, Pontes V, Alves PM, McDougall G, Stewart D, Ferreira RB, Santos CN (2010). Antioxidant properties and neuroprotective capacity of strawberry tree fruit (*Arbutus unedo*), Nutrients, 2: 214-229.

Gomes F, Canhoto JM (2009). Micropropagation of strawberry tree (*Arbutus unedo* L.) from adult plants, *In Vitro* Cell. Dev. Biol.-Plant, 45:72-82.

Gomes F, Lopez ML, Santos T, Canhoto JM, 2009. Micropropagation of selected strawberry trees of *Arbutus unedo* L. through axillary shoot proliferation and somatic embryogenesis, Acta Horticulturae, 839:111-116.

Gomes F, Simoes M, Lopez ML, Canhoto JM (2010). Effect of plant growth regulators and genotype on the micropropagation of adult trees of *Arbutus unedo* L. (strawberry tree), New Biotechnology, 27(6): 882-892.

Gomes MFFN (2011). Strategies for the improvement of *Arbutus unedo* L. (strawberry tree): *in vitro* propagation, mycorrhization and diversity analysis, Departamento de Ciencias da Vida, Universidade de Coimbra, pp. 208

Gomes F, Costa R, Ribeiro MM, Figueiredo E, Canhoto JM (2013). Analysis of genetic relationship among *Arbutus unedo* L. genotypes using RAPD and SSR markers, Journal of Forestry Research, 24(2): 227-236.

Gratani L, Ghia E (2002). Changes in morphological and phisiological traits during leaf expansion of *Arbutus unedo*, Environmental and Experimental Botany, 48:51-60.

Güllüoğlu L, Kurt C, Arıoğlu H, Zaimoğlu B, Aslan M (2010). The researches on soybean (*Glycine max* Merr.) variety breeding for resistance to whitefly in Turkey, Turkish Journal of Field Crops, 15(2):123-127.

Hubert J, Lee S (2005). A review of the relative roles of silviculture and tree breeding in tree improvement: the example of Sitka spruce in Britain and possible lessons for hardwood breeding, Forestry, 78(2): 109-120.

Karademir E, Gençer O, Karademir Ç (2009). Determination of heterotic effects in breeding of multiadversity resistance in cotton (*Gossypium hirsutum* L.), Tarım Bilimleri Dergisi, 15(3): 209-216. (in Turkish)

Karadeniz T, Kalkışım Ö, Şişman T (2003). The fruit characteristics of strawberry tree types grown in Trabzon region and their propagation by cutting, Ulusal Kivi ve Üzümsü Meyveler Sempozyumu, Türkiye, 476-480. (in Turkish)

Kaya Y, Demirci M, Evci G (2004). Sunflower (*Helianthus annuus* L.) breeding in Turkey for broomrape (*Orobanche cernua* Loeffl.) and herbicide resistance, Helia, 27(40): 199-210.

Konstantinidis P, Tsiourlis G, Xofis P (2006). Effect of fire season, aspect and pre-fire plant size on the growth of *Arbutus unedo* L. (strawberry tree) resprouts, Forest Ecology and Management, 225: 359-367.

Korkut KZ, Başer I, Bilgin O (2001). Genotypic and phenotypic variability, heritability and phenotypic correlation for yield and yield components in bread wheat varieties, Acta Agronomica Hungarica, 49(3): 237–242.

Kumar J, Gupta PK, (2008). Molecular aproaches for improvement of medicinal and aromatic plants, Plant Biotechnol. Rep, 2: 93-112.

Küden AB, Küden A (2008). Germplasm collection and breeding studies of low chilling cultivars, Acta Hort. 772: 503-506.

Lopes L, Sa O, Pereira JA, Baptista P (2012). Genetic diversity of Portuguese *Arbutus unedo* L. populations using leaf traits and molecular markers: An approach for conservation purposes, Scientia Horticulturae, 142: 57-67.

Males Z, Plazibat M, Vundac VB, Zuntar I (2006). Qualitative and quatitative analysis of flavonoids of the strawberry tree-*Arbutus unedo* L. (*Ericaceae*), Acta Pharm. 56: 245-250.

Mereti M, Grigoriadou K, Nanos GD (2002). Micropropagation of the strawberry tree, *Arbutus unedo* L, Scientia Horticulturae, 93: 143-148.

Mereti M, Grigoriadou K, Leventakis N (2003). *In vitro* rooting of strawberry tree (*Arbutus unedo* L.) in medium solidified by peat-perlite mixture in combination with agar, Acta Horticulturae, 616: 207-210.

Metaxas D, Syros T, Economou A (2008). Factor affecting vegetative propagation of *Arbutus unedo* L. by stem cuttings, Propagation of Ornamental Plants, 8(4): 190-197.

Molina M, Pardo-De-Santayana M, Aceituno L, Morales R, Tardio J (2011). Fruit production of strawberry tree (*Arbutus unedo* L.) in two Spanish forests, Forestry, 84(4): 419-429.

Navarro A, Sanches-Blanco MJ, Banon S (2007). Influence of paclobutrazol on water consumption and plant performance of *Arbutus unedo* seedlings, Scientia Horticulturae, 111: 133-139.

Ozdemir Y, Aktepe-Tangu N, Akcay ME (2013). Generating omega-3 rich olive oil by cross breeding, Eur. J. Lipid Sci. Technol, 115: 977–981.

Özcan MM, Hacıseferoğulları H (2007). The strawberry (*Arbutus unedo* L.)fruits: Chemical composition, physical properties and mineral contents, Journal of Food Engineering, 78: 1022-1028.

Pabuçcuoğlu A, Kıvçak B, Baş M, Mert T (2003). Antioxidant activity of *Arbutus unedo* leaves, Fitoterapia, 74(6): 597-599.

Panicucci A, Nali C, Lorenzini G (1998). Differential photosynthetic response of two mediterranean species (*Arbutus unedo* and *Viburnum tinus*) to sulphur dioxide, Chemosphere, 36(4-5): 703-708.

Pawlowska AM, De Leo M, Braca A (2006). Phenolics of *Arbutus unedo* L. (*Ericaceae*) fruits: Identification of anthocyanins and gallic acid derivatives, J. Agric. Food Chem. 54, 10234-10238.

Pırlak L, Güleryüz M, Aslantaş R, ve Eşitken A (1997). Erzurum İlinin Tortum ve Uzundere İlçelerinde Kaliteli Yazlık Elma Tiplerinin Seleksiyon Yoluyla Islahı Üzerinde Bir Araştırma, Yumuşak Çekirdekli Meyveler Sempozyumu, Yalova. s: 21-28. (in Turkish)

Rasmont P, Regali A, Ings TC, Lognay G, Baudart E, Marlier M, Delcarte E, Viville P, Marot C, Falmagne P, Verhaeghe J-C, Chittka L (2005). Analysis of Pollen and Nectar of *Arbutus unedo* as a Food Source for *Bombus terrestris* (Hymenoptera: Apidae), Journal of Economic Entomology, 98(3): 656-663.

Rodrigues AP, Sergio PM, Teixeria MR, Pais MS (2001). *In vitro* break of dormancy of axillary buds from woody species (Persea indica and Arbutus unedo) by sectioning with a laser beam, Plant Science, 161: 173-178.

Ruiz-Rodríguez BM, Morales P, Fernández-Ruiz V, Sánchez-Mata MC, Cámara M, Díez-Marqués, C, Pardo-de-Santayana M, Molina M, Tardío J (2011). Valorization of wild strawberry-tree fruits (*Arbutus unedo* L.) through nutritional assessment and natural production data, Food Research International, 44: 1244-1253.

Sa O, Pereira JA, Baptista P (2011). Optimization of DNA extraction for RAPD and ISSR analysis of *Arbutus unedo* L. leaves, Int. J. Mol. Sci. 12: 4156-4165.

Sulusoglu M, Cavusoglu A, Erkal S (2011). *Arbutus unedo* L. (Strawberry tree) selection in Turkey Samanli Mountain locations, Journal of Medicinal Plants Research, 5(15): 3545-3551.

Şeker M, Yücel Z, Nurdan E (2004). Investigation of morphological and pomological characteristics of strawberry tree (*Arbutus unedo* L.) population in the natural flora of Çanakkale district, Tarım Bilimleri Dergisi, 10(4):422-427. (in Turkish)

Takrouni MM, Boussaid M (2010). Genetic diversity and population's structure in Tunusian strawberry tree (*Arbutus unedo* L.), Scientia Horticulturae, 126: 330-337.

Toker C, Canci H, Inci NE, Ceylan FO (2012). Improvement in imidazolinone resistance in cicer species by induced mutation, Plant Breeding, 131(4):535-539.

USDA-ARS-GRIN (2013) National Genetic Resources Program, Germplasm Resources Information Network - (GRIN) [Online Database], National Germplasm Resources Laboratory, Beltsville, Maryland, URL: http://www.ars-grin.gov/cgi-bin/npgs/html/taxon.pl?3849 (02 November 2013)

Uzun B, Lee D, Donini P, Çağirgan ML (2003). Identification of a molecular marker linked to the closed capsule mutant trait in sesame using AFLP, Plant Breeding, 122(1): 95-97.

Uzun A, Gulsen O, Kafa G, Seday U (2008). 'Alata', 'Gulsen', and 'Uzun' Seedless Lemons and 'Eylul' Early-maturing Lemon, HORTSCIENCE, 43(6):1920–1921.

van Eijk JP, Leegwater J (1975). Criteria for early selection in tulip breeding, Acta Hort. 47:179-186.

Wang J 2011. Modelling and simulation of plant breeding strategies, (In Plant Breeding, Eds. I.Y. Abdurakhmonov, Intech Open Access Publisher, Rijeka, Crotia) 19-40.

Yarılgaç T, İslam A (2007). Pomological characteristics of the strawberry tree fruit (*Arbutus unedo* L.) of Ünye (Ordu) region, 5. Ulusal Bahçe Bitkileri Kongresi, Türkiye, 556-560. (in Turkish)

Zamboni A, Pierantoni L, De Franceschi P (2008). Total RNA extraction from strawberry tree (Arbutus unedo) and several other woody-plants, iForest, 1: 122-125.

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Yield and yield components, morphological and quality characteristics of Aromatik-1 rice variety: the first aromatic rice in Turkey

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ABSTRACT

Aromatic rice (*Oryza sativa* L.) has high quality aroma with cooking quality. This study was carried out in order to select aromatic rice with introduction of nurseries from IRRI (International Rice Research Institute) at Trakya Agricultural Research Institute between 1999 to 2004. The materials were tested for primary yield, yield and regional yield trials. As a result of these trials, YRF-204 aromatic rice line was evaluated for yield, yield components, morphological and quality characteristics in registration trials in 2005 and registered as Aromatik-1 in 2007. Aromatik-1 variety has high yield and aromatic characteristics. Aromatik-1 had 13 (104 days after sowing) and 12 days (149 days after sowing) later to flowering and maturity than that of Baldo (91 days after sowing) and (137 days after sowing), respectively. Aromatik-1 had 5.11 % more yield than the check cultivar Baldo over means of 5 year experiments from 2000 and 2004, while it had less (52 %) head rice yield than that of Baldo (55 %). Although Aromatik-1 rice cultivar is indica type rice, 52 % head rice is very good for this type of rice. Aromatik-1 variety was the first aromatic rice registered in Turkey for yield, some morphological and quality characteristics The present study shows that yield and quality parameters in rice as Aromatic-1 variety could be improved via traditional breeding methods.

Keywords: *aromatic rice, Oryza sativa, quality, yield*

Introduction

Aroma represents high quality in rice (Oryza sativa L.), and cooking quality and head rice yield have close relations with aroma (Nagaraju et.al. 1975, Tripathi and Rao 1979). On the other hand, aromatic rices have less yield and more susceptibility to diseases and pests than the other rice (Berner and Hoff 1986). Although they had lower yield, researchers in Europe studies to improve aromatic rice for Europe (Lorieux et. al. 1997). There are various ideas about inheritance of aroma in rice. Some researchers reported that it was governed by a single gene, some researchers reported that it was under more genes than one gene inheritance. Main component responsible for aroma in rice is 2-acetyl-pyroline (AcPy), and this component was found in every part of rice plant except in roots (Buttury et al. 1983).

Rice production area in Turkey ranged from 40 000 ha to 100 000 ha during last 50 years. Annual total milled rice production were between 150 000 and 500 000 Metric Tons and this production is not sufficient for domestic consumption in Turkey. Turkey self sufficiency for rice was 43% in 2000, it increased to more than 90,7 % in 2010. There is a need to improve rice production nearly 10 % to be self sufficient in Turkey. Turkey can be 100 % self sufficient for rice in near future. But there was no any aromatic or basmati type rice production until 2007. Despite import of small amount of aromatic rice each year, it will be continued to import if aromatic rice is not registered for Turkey. .

Table 1. Paddy yield of rice genotypes for 2000, 2001 and 2002

Primary yield trial, 2000		Yield trial, 2001		Yield trial, 2002	
Genotypes	T/ Ha	Genotypes	T/ Ha	Genotypes	T/ Ha
Rocca (st)	5.82	Demir (st)	8.84	BALDO (St)	627
YRF-204	5.69	Rocca (st)	7.89	YRF-204	569
Osmancık-97 (st)	7.19	YRF-204	7.44	YAR-96-V14	537
IR 70444-87-2-1	4.18	Kıral (st)	6.84	YRF-203	526
YAR-96-v14	4.26	Osmancık-97 (st)	6.62	IR0445-229-4-1	523
IR70422-51-3-2	4.96	IR 70444-87-2-1	6.53	JJ92(ADT41)	471
IR-70422-66-5-2	5.33	YAR-96-v14	6.24	RAJBH0G	426
IR-70422-105-3-3	4.34	IR70422-51-3-2	6.08	IR66233-151-1-1	415
Baldo (st)	6.67	IR-70422-66-5-2	6.07	IR70422-51-3-2	414
YRF-203	5.01	IR-70422-105-3-3	5.82	IR70445-5-2-2	403
IR70416-53-2-2	3.88	Baldo (st)	5.72	IR0422-105-3-3	385
IR70086-3-3-1-3	3.33	YRF-203	5.71	IR70422-66-5-2	385
IR70086-19-1-2-2	4.20	IR70416-53-2-2	5.45	IR70416-53-2-2	384
IR66696-55-2-2-3	3.58	IR70086-3-3-1-3	5.23	IR70422-152-1-1	330
RAJBHOB	4.11	IR70086-19-1-2-2	4.87	IR70086-19-1-2-25	329
IR70423-170-2-3	2.71	IR66696-55-2-2-3	4.76	IR70086-3-3-1-2	304
RP3138-42-11-7-1	1.26	RAJBHOB	4.75	IR66696-55-2-2-3	282
IR670-228-1-5	2.38	IR70423-170-2-3	4.57		
IR67420-228-1-5	1.89	RP3138-42-11-7-1	4.44		
IR70418-112-1-2	1.10	IR670-228-1-5	4.12		
IR67414-216-3-4-2-3	1.61	IR67420-228-1-5	4.09		
IR70421-188-2-1	1.24	IR70418-112-1-2	3.93		
		IR67414-216-3-4-2-3	2.96		
		IR70421-188-2-1	2.89		
C.V(%)	16	C.V(%)	17.3	C.V(%)	12.5
LSD (5%)	1.36	LSD (5%)	3	LSD (5%)	1.61
			1.56		

All rice varieties grown in Turkey are non aromatic japonica type rice. Aromatic and basmati type varieties were not produced in Turkey (Beşer et al. 2007). Aromatic type rice is more expensive than japonica rice. For these reasons aromatic rice breeding project has been started in Turkey in 2007. In this paper, results pertaining to performance of Aromatik-1 rice cultivar, which is the first registered aromatic rice in Turkey, for yield, morphological and quality characteristics are presented.

Materials and methods

Preliminary studies were started in 1999 with introduction of International Rice Fine Grain Aromatic Rice Observation Nursery (IRFAON -1999) from IRRI (International Rice Research Institute). In 1999, IRFAON 1999 nursery was raised in augmented design. Each line was planted at 5 m2. More materials were continued to introduce during later years from IRRI. Introduced materials were tested at screening nurseries, primary yield, yield and regional yield trials between 2000 and 2004. Primary yield trial was conducted with three replications and each line was planted at 8 m2 plots. Yield and regional yield trials were conducted at 4 x 5 m (20 m2) plots with three replications. Since there was not any registered aromatic type and indica type rice variety in Turkey during this research, japonica type best local checks for grain quality such as Baldo, Kıral and high yielding varieties such as Osmancık-97, Rocca and Demir were used as checks for different years in yield trials. Except the mentioned local checks, a japonica type aromatic rice, Rajbhob was also used as check. Experiments were fertilized with P2O5 of 80 kg/ ha and N of 150 kg/ha each year. All phosphorus was applied before sowing, nitrogen was applied at three times 1/3 part was applied at before sowing, while 1/3 part was applied at tillering stage and other 1/3 part was applied at panicle initiation.

Screenings were taken according to Standard Evaluation System for Rice (Anonymous 1996). Yield and other characters such as plant height, panicle length, days to flowering, days to maturity, panicle sterility, 1000-kernel weight, brown rice yield (%), total milled rice yield (%), and head rice yield were recorded.

Results and discussion

Experimental results for yield are given for five years from 2000 to 2004 in Table 1 and Table 2.

YRF-204 (Aromatik-1) had 2nd, 3rd, 2nd, 2nd, 2nd and 4th place at primary yield trial in 2000, yield trial in 2001, yield trial in 2002, regional yield trial in 2002, yield trial in 2003 and yield trial in 2004, respectively. YRF-204 was adapted very well for Turkey climatic conditions. Baldo or this type rice is accepted the highest quality rice in Turkish rice market and it is sold at the highest price. As can be seen from Table 3, YRF-204 (Aromatik-1) had 5.11 % more yield than the Baldo rice for 5 years mean from 2000 and 2004. From these results, Aromatik-1 rice cultivar can be grown competitively by Turkish rice producer.

Table 2. Paddy yield of rice genotypes for 2002, 2003 and 2004

Regional yield trial, 2002		Yield trial, 2003		Yield trial, 2004	
Genotypes	T/ Ha	Genotypes	T/ Ha	Genotypes	T/ Ha
OSMANCIK-97	7.93	YAR-96-V14	5.67	Baldo (st)	5.53
YRF-204	7.29	YRF-204	5.04	IR 70422-152-1-1	5.46
YAR-96-V14	6.73	Baldo (st)	4.04	YAR-96-V14	5.07
BALDO (st)	5.82	RAJBHOG	3.20	YRF-204	4.73
YRF-203	4.61	IR 70422-51-3-2	3.07	IR 0445-229-4-1	4.72
IR70422-51-3-2	4.33	IR 66696-55-2-2-3	2.94	IR 70445-5-2-2	4.29
IRO422-105-3-3	4.13	IR 0445-229-4-1	2.92	IR 66232-274-1-3-2	4.24
IR70086-19-1-2-2	4.00	IR 70416-53-2-2	2.91	IR 71743-32-2-1	4.08
IR70422-66-5-2	3.38	YRF-203	2.88	YRF-203	3.58
IR70086-3-3-1-3	3.21	IR 70086-3-3-1-2	2.72	IR 66233-151-1-1	3.49
IR70416-53-2-2	3.00	JJ92 (ADT41)	2.45	IR 70422-51-3-2	3.40
IR70444-87-2-1	2.96	IR 70422-152-1-1	2.28	IR 70086-3-3-1-2	3.04
IR66696-55-2-2-3	2.19	IR 70086-19-1-2-25	2.23	JJ92 (ADT41)	2.97
		IR 70445-5-2-2	2.05	IR 70416-53-2-2	2.96
		IR 71743-32-2-1	1.73	RAJBHOG	2.90
		IR 67014-138-3-1	1.70	IR 70086-19-1-2-25	2.20
		IR 67013-76-2-3-3	1.32	IR 66696-55-2-2-3	1.75
		IR 66233-151-1-1	1.14	DR 31	1.58
				IR 67014-138-3-1	1.46
				IR 67013-76-2-3-3	1.10
				Azucena	0.76
C.V(%)	12.5	C.V(%)	31.78	C.V(%)	12.62
LSD (5%)	0.97	LSD (5%)	1.48	LSD (5%)	0.69

Table 3. Paddy yields of Aromatik-1 and Baldo varieties between 2000 and 2004, and mean yield differences of Aromatik-1 variety from check variety Baldo

	Paddy yield T/ ha, between 2000 and 2004						Mean Yield T/ Ha	Mean yield differences from check (%)
	2000	2001	2002	2002	2003	2004		
Baldo (Check)	6.67	5.72	6.27	5.82	4.04	5.53	5.67	-
Aromatik-1	5.69	7.44	5.69	7.2	5.04	4.73	5.96	5.11

Table 4. Some quality characteristics of Aroamtik-1 and Baldo varieties

Genotype	Paddy kernel length (L)and width (W) (mm)		Polished rice kernel length (L) and width (W) (mm)		Milling yield (%)			Paddy 1000 kernel weight (g)
	L	W	L	W	Brown	Total	Head	
Baldo	9.3	3.7	6.9	3.2	79	70.3	55	38.8
Aromatik-1	9.4	2.4	6.9	2.1	82.1	70.2	52	23.7

Table 5. Some morphological and quality characteristic of rice genotypes in 2004

Lines/ cultivars	Days to flowering	Days to maturity	Plant height (cm)	Panicle length (cm)	Panicle sterilty (%)	Paddy rice		Polished rice		1000 paddy kernel weight (g)
						Length (mm)	Width (mm)	Length (mm)	Width (mm)	
Baldo (st)	91	137	96.7	16.1	13.7	9.3	3.7	6.9	3.2	38.8
IR 66696-55-2-2-3	108	151	81.3	19.6	48.1	11.4	2.3	7.5	2.0	22.6
IR 70086-19-1-2-25	111	152	90.8	20.5	56.1	11.9	2.4	7.4	1.9	24.9
IR 70086-3-3-1-2	105	153	90.3	19.9	50.6	11.0	2.3	7.6	1.9	23.4
IR 70416-53-2-2	99	149	82.7	19.3	40.2	10.4	2.2	7.4	1.9	22.3
IR 70422-51-3-2	104	151	92.1	20.1	38.5	9.9	2.4	7.3	2.0	23.5
RAJBHOG	90	144	91.9	15.1	40.5	8.2	3.2	5.9	2.8	19.9
YAR-96-V14	94	151	69.9	16.0	27.6	9.9	2.7	6.9	2.3	26.0
YRF-203	104	155	88.9	17.7	47.0	9.6	2.6	7.0	2.2	23.6
YRF-204 (Aromatik-1)	100	149	79.7	17.9	30.7	9.4	2.4	6.9	2.1	23.7
IR 0445-229-4-1	105	155	80.5	18.7	46.1	9.3	2.3	7.5	2.0	23.6
IR 66233-151-1-1	104	152	90.4	21.1	36.1	11.3	2.2	8.1	1.8	25.2
IR 70422-152-1-1	100	151	83.7	19.2	48.6	9.2	2.1	6.6	1.9	20.5
IR 67014-138-3-1	113	156	74.4	19.9	67.8	11.2	2.3	7.7	1.9	22.7
JJ92 (ADT41)	112	155	90.3	23.2	61.0	10.8	2.3	7.3	1.9	25.2
IR 67013-76-2-3-3	122	158	72.7	15.9	83.8	10.2	2.2	7.0	1.9	20.8
IR 71743-32-2-1	112	156	79.6	16.9	63.1	11.0	2.2	7.1	1.8	21.3
IR 70445-5-2-2	103	156	84.8	21.5	41.4	10.4	2.3	7.5	1.9	21.1
Azucena	-	-	107.8	18.7	89.2	9.8	2.8	7.1	2.5	28.0
DR 31	116	159	73.0	19.3	62.3	9.7	2.5	6.8	2.1	22.3
IR 66232-274-1-3-2	116	155	76.3	19.9	61.4	11.4	2.2	7.4	1.9	24.5

Head rice yield is very important for Turkish market. As can be seen in Table 4, Aromatik-1 had 52 % head rice, while Baldo had 55 % head rice yield. Although Aromatik-1 rice cultivar is indica type rice, 52 % head rice is very good for this type of rice. It is nearly japonica type check Baldo cultivar for head rice rate. Results for some of the morphological and quality characteristics for 2004 are given in Table 5. Days to flowering (DTF) of YRF-204 (Aromatik-1) rice was recorded as 104 days, and its flowering time was later than that of Baldo rice with 91 days. Days to maturity (DTM) of YRF-204 (Aromatik-1) was 149 days and its days to maturity was 12 days later than that of Baldo. On the other hand, YRF-204 (Aromatik-1) had short plant height and lodging resistant than the Baldo variety. 1000-kernel weigh of YRF-204 (Aromatik-1) was smaller than that of Baldo.

As a result of five years experiments from 2000 to 2004, YRF-204 was identified to nominate for registration trial. After registration trials for two years, YRF-204 was registered as Aromatik-1 and started producing foundation seeds. Aromatik-1 has been produced by farmers as a first registered aromatic rice cultivars in Turkey. This study suggested that yield and related components and quality characteristics in rice could be improved via conventional breeding approaches.

References

Anonymous (1996). Standard Evaluation System for Rice. 4th ed. International Rice Research Institute, Manila, Philippines.

Berner DK, Hoff BJ (1986). Inheritance of scent in amerikan long grain rice. Crop Science 26:876-878

Beşer N, Sürek H, Kaya R (2007). Trakya bölgesi şartlarına uygun aromatik çeltik (Oryza sativa L.) geliştirme çalışmaları. Türkiye 7. Tarla Bitkileri Kongresi 25-27 Haziran 2007, Erzurum

Buttery RG, Ling LC, Juliano OB, Turnbaugh JG (1983). Cooked rice aroma and 2-acetyl-1-pyrroline. J. Agric Food Chem 31:823-826

Lorieux M, Petrov M, Pons B, Clement G, Faure J, Ghesquiere A (1997). Genetic and biochemical analysis of aroma in rice. In Communications of International Symposium on rice quality

Nagaraju M, Chaudhary D, Rao MJB (1975). A simple technique to identify scent in rice and inheritance pattern of scent. Current Science 44:599

Tripathi RS, Rao MJBK (1979). Inheritance and linkage relationship of scent in rice. Euphytica 28:319-323.

Research Article

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Assessment of pollen viability and germination in seven varieties of lemon

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ABSTRACT

We investigated the viability and *in vitro* germination of pollen in seven lemon varieties (*Citrus limon* (L.) Burm f.) ('BATEM Sarısı', 'BATEM Pınarı', 'Interdonato', 'Kütdiken', 'İtalyan Memeli', 'Meyer' and 'Lamas'). The viability was tested with TTC (2, 3, 5–triphenly tetrazolium chloride) and *in vitro* germination was tested with agar–plate to estimate pollen viability and germination in these varieties. Additionally, effect of different sucrose concentration levels (5%, 10%, 15%, 20% and 25%) for *in vitro* pollen germination was observed. The results indicated that pollen of 'Meyer' had the highest viability with 86.74%. The pollen of 'Lamas' and 'Meyer' varieties had the highest *in vitro* germination with 39.77% and 39.04%, respectively. This study results showed that 20% and 25% sucrose concentrations in the agar plates had the highest germination with 36.16% and 32.12%, respectively.

***Keywords*:** lemon, pollen, viability, germination, sucrose

Introduction

Citrus is the most widely produced fruit group in the world. There has been a rapid increase in the production both in the world and in Turkey. Approximately 759.711 tons of lemon fruits are produced in Turkey (FAO 2014). With total export of 374.734 tons lemons are ranking as the first within the citrus fruits in Turkey (AKIB 2014).

Information about the ability of pollen grains to germinate when they reach the stigmas of flowers of their own species is valuable both for horticultural purposes and general botanical research. Viability tests provide a means of assessing the potential of pollen to germinate on the stigma (Firmage and Dafni 2001). Several different methods have been suggested to determine the viability of pollen (Dafni 1992; Kearns and Inouye 1993; Marcucci et al. 1984; Demirkeser et al. 2001).

The objective of this study was to determine the pollen viability and *in vitro* germination percentage of different lemon cultivars.

Material and methods

Studies were carried out on pollen grains of seven cultivars of lemon 'BATEM Sarısı', 'BATEM Pınarı', 'Interdonato', 'İtalyan Memeli', 'Kütdiken' , 'Lamas' and 'Meyer'. Pollens collected during the spring of 2011 and 2012 were used in these experiments.

In order to determine the pollen viability and germination capacity, well-grown flower from each variety were picked about ten o'clock in the balloon stage flowers. They were put in paper bags and were brought to the laboratory. The anthers were left to dehisce for 24 h at room temperature at about 23°C, and the fresh pollen was immediately used for pollination (Distefano et al. 2009).

Effects of various sucrose concentrations on the pollen germination and pollen viability rates with Tetrazolium (TTC) test were determined. TTC (2, 3, 5–triphenlytetrazoliumchloride), stain tests were used to determine pollen viability. One or two drops of TTC solution was put on a clean micro slide and kept for 3-4 h at ambient conditions (Norton 1966). For this assay, two lamella for each cultivars and four regions of each lamella were investigated; viable, semi-viable and dead pollen numbers and their percentages were determined. Pollen viability was scored based on the staining level as pollen with red color viable, with

light red semi-viable and with colorless nonviable.

Five different sucrose concentrations (5, 10, 15, 20 and 25%) were used in the agar–plate tests to determine pollen germination and were added to 1% agar and 0.1% boric acid. In tests, the pollen was incubated at 21°C for 24 h under dark conditions. The two plates were scanned for each pollen source. In the germination test, five microscopic areas from each replication were counted randomly at the end of the 24 h incubation period. The values for viability and germination of pollens were subjected to square–root transformation for statistical analysis. The experiment carried out completely randomized plot design. Duncan's test was used to compare the means.

Results

The pollen viability in lemon varieties was found to be significantly different in the TTC solution (Table 1). The highest pollen viability was obtained from Meyer variety (86.74%) followed by Kütdiken (69.22%) in the TTC stain test.

Table 1.The rate of pollen viability in the TTC

Cultivars	The rate of viable pollen(%)
BATEM Pınarı	52.66 cd *
BATEM Sarısı	40.62 d
Interdonato	59.48 bc
İtalyan Memeli	54.57 c
Kütdiken	69.22 b
Lamas	52.29 cd
Meyer	**86.74 a**

*Different letters indicated significant differences (P<0.05) according to the Duncan test

The differences in pollen germination among cultivars were significant in all the sucrose concentrations in agar–plate test. The sucrose concentrations in the agar–plate methods were found to have different effects on pollen germination of all cultivars. The highest pollen germination was obtained in Lamas (39.77%) and Meyer (39.04%). The optimum sucrose concentrations in the agar-plate method for pollen germination of all cultivars were 20% and 25% (Table 2).

Table 2.The rate of pollen germination in the agar-plate test

Cultivars	Sucrose concentrations (SC)					Means of cultivars
	5%	10%	15%	20%	25%	
BATEM Pınarı	20.10	22.04	12.72	19.52	28.98	20.67 bc*
BATEM Sarısı	15.29	15.08	17.38	28.65	20.65	19.41 c
Interdonato	11.80	13.38	13.28	31.53	22.09	18.42 c
İtalyan Memeli	31.36	34.38	31.63	37.31	40.60	35.06 a
Kütdiken	25.51	23.34	18.93	23.30	41.54	26.52 b
Lamas	36.11	29.65	42.09	44.09	46.90	39.77 a
Meyer	23.89	38.19	40.32	40.47	52.34	39.04 a
Means of SC	23.44 B	25.15 B	25.19 B	32.12 A	36.16 A	

*Different letters indicate significant differences (P<0.05) according to the Duncan test

Discussion

Eti (1991); Parfitt and Almedhi (1984); Seilheimer and Stösser (1982) have indicated that germination percentage vary significantly according to fruit species or cultivars. Sucrose concentrations affected the pollen germination of lemon varieties. Our study, the optimum sucrose concentrations for pollen germination of all cultivars were 20% and 25%. Ateyyeh (2005) found that pollen of *Citrus maxima* showed the highest germination percentage when placed in media containing 20% sucrose. In the literature, sucrose concentration has been determined as % 10 (Seilheimer and Stösser 1982) and % 15 (Werner and Chang 1981; Parfitt and Almehdi 1984) in the pollen germination percentage of different fruit species. As a result, these stain tests may be used to determine pollen viability in these species to provide only a rough estimate of viability. However, the exact amount of viable pollen may be determined *in vitro* by pollen germination.

It is clear that no one test is suitable for testing viability in all species. It is also apparent that some of the stains that have been suggested most often over estimate viability.

References

AKIB (2014). http://www.akib.org.tr/files/downloads/arastirmaraporlari/ysm/yms_ocak-aralik. 2012.pdf Accessed 10 November 2014.

Ateyyeh AF (2005). Improving *in vitro* Pollen Germination of Five Species of Fruit Trees. Dirasat, Agricultural Sciences, Volume 32 (2):189-194.

Dafni A (1992). Pollination Ecology: A practical approach. New York, New York: Oxford University Press.

Demirkeser TH, Eti S and Kaplankiran M (2001). The effect of self and cross-pollination on the fruit set and quality of Nova mandarin. 6th International Congress of Citrus Nurserymen, 9-13 July 2001, Ribeirao Preto, SP-Brazil. Proceedings, 305-308.

Eti S (1991). Bazı meyve tür ve çeşitlerinde değişik *in vitro* testler yardımıyla çiçek tozu canlılık ve çimlenme yeteneklerinin belirlenmesi. Çukurova Üniversitesi Ziraat Fakültesi Dergisi, 1(6): 69-80.

FAOSTAT (2014). http://faostat.fao.org/site/567/default.aspx#ancor Accessed 10 November 2014.

Firmage DH and Dafni A (2001). Field tests for pollen viability; a comparative approach. Proc. 8th Pollination Symp, Benedek P and Richards KW (eds.), Acta Hort. 561.

Kearns CA and Inouye DW (1993). Techniques for pollination biologists. Niwot, Colorado: University Press of Colorado.

Marcucci MC, Ragazzini D and Sansavani S (1984). The effects of gamma and laser rays on the functioning of apple pollen in pollination and mentor pollen experiments. Journal of Hort. Sci. 9: 57-61.

Norton JD (1966). Testing of Plum pollen viability with tetrazolium salts. American Society of Horticultural Science, 89: 132-134.

Parfitt DE and Almehdi A (1984). Liquid nitrogen storage of pollen from five cultivated *Prunus* species. HortScience 19:69–70.

Seilheimer M and Stösser R (1982). Die Eignung verschiedener Apfelsorten als Pollenspender. Erwerbsobstbau, 24: 62–65.

Werner DJ and Chang S (1981). Stain testing viability in stored peach pollen. HortScience, 16 (4): 522–523.

Research Article

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Effect of drought stress on some physiological parameters, yield, yield components of durum (*Triticum durum* desf.) and bread (*Triticum aestivum* L.) wheat genotypes

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ABSTRACT

Drought is the most important limiting factor for growth and productivity of crop plants. The aim of this research was to study the effect of soil water deficit on gas exchange parameters, photosynthetic pigments content, relative water content, area, dry weight, leaf specific mass of flag leaves from durum and bread wheat genotypes. Gas exchange parameters of leaves measured by using LI-COR 6400-XT Portable Photosynthesis System. Drought caused reduction in photosynthesis rate, stomatal conductance, transpiration rate, mesophyll conductance, pigments content, area, dry weight, relative water content of flag leaves. Leaf specific mass increased under rain-fed condition. Strong relationships were detected between stomatal conductance and transpiration rate, between mesophyll conductance and photosynthesis rate. Photosynthesis is less inhibited than transpiration rate under water stress. Under influence of water stress the content of photosynthetic pigments, also the ratio of chlorophyll to carotenoids decreases. Drought led decrease in yield and yield components of wheat genotypes.

Keywords**:** wheat, soil water deficit, gas exchange parameters, yield

Introduction

Drought stress is one of the most widespread environmental stresses when the available water in the soil is reduced and atmospheric conditions cause continuous loss of water by transpiration and evaporation (Kramer 1980). Up to 26% from the usable areas of the Earth is subjected to drought (Blum 1986). Drought is considered as the main factor limiting plant growth and yield worldwide. Wheat the major crop plant in the daily diet of 35% of world population, is a sources of energy from carbohydrates and proteins. Important stages of wheat development (stem elongation, heading-flowering, grain filling) occurs during the time when the water deficit in the soil increases in rain fed regions. Wheat is one of the widely cultivated crops in Azerbaijan, where drought is the main limiting factor for its production (Aliyev 2001). Up to 35% of the 650,000 hectare wheat grown areas is under rain-fed conditions.

The response of plants to water stress depends on several factors such as development stage, severity and duration of stress and cultivar genetics (Beltrano and Marta 2008). The adaptation strategies of the plants to drought stress include drought escape, drought avoidance and drought tolerance (Levitt 1980). Photosynthesis, the most significant process which influence crop production, is also inhibited by drought stress. The effects can be direct, as the decreased CO_2 availability caused by diffusion limitations through the stomata and the mesophyll (Flexas et al. 2004) or the alterations of photochemical reactions (Tang et al. 2002) and photosynthetic metabolism (Lawlor and Cornic 2002). Under field conditions, stomatal regulation of transpiration was shown as a primary event in plant response to water deficit leading to decrease of CO_2 uptake by the leaves (Chaves 1991, 2002; Cornic and Massacci 1996). Stomatal responses are more closely linked to soil moisture

content than to leaf water status. Reduced plant size, leaf area, and leaf area index are a major mechanism for moderating water use and reducing injury under drought stress (Mitchell et al. 1998). Drought causes decrease in grain yield and yield components of field grown wheat genotypes (Veesar et al. 2007; Moayedi et al. 2010; Akram 2011).

The purpose of this research was to study the effect of soil water deficit on some physiological parameters in leaves of durum wheat and bread wheat genotypes and to determine physiological traits which can be used for identification tolerant wheat genotypes under water stress conditions.

Materials and methods

Field experiment was carried out in the research area of Plant Physiology and Biotechnology Department of Research Institute of Crop Husbandry located in Absheron peninsula, Baku, during the 2012-2013 growing season. Six durum wheat genotypes (Garagylchyg 2, Vugar, Shiraslan 23, Barakatli-95, Alinja- 84, Tartar) seven bread wheat genotypes (Gobustan, Giymatli-2/17, Gyrmyzygul 1, Azamatli-95, Tale-38, 12nd FAWWON№97, 4th FEFWSN№50) were used for this study. Sowing was done at an average density 400 seeds m^{-2} with self-propelled mechanical planter in 1 mx10 m plots, consisting of 7 rows placed 15 cm apart. Each genotype was sown with three replications both in irrigated and rain-fed conditions. Irrigated plots were watered at stem elongation, flowering and grain filling stage. Fertilization was applied as N_{120} , P_{60} , K_{60} per hectare. Thirty per cent of the nitrogen applied at planting and the rest at the beginning of stem elongation.Net photosynthesis rate (P_n), stomatal conductance (g_s), intercellular CO_2 concentration (C_i), transpiration rate (T_r) were measured with a Portable Photosynthesis System LI-6400 XT (LI-COR Biosciences, Lincoln, NE, USA). Light intensity was measured by using Light-Meter LI-250A (LI-COR Biosciences) equipped with Pyranometer PY 71968 (LI-COR Biosciences). Figure 1 shows the daily variation of photosynthetically active radiation (PAR). PAR reaches maximum in the 13th hour of the day. Flag leaf photosynthetic pigments content (mg g^{-1} DW) was determined following the method of Lichtenthaler (1987). About 0,1 g fresh leaves were ground in 96% ethanol for the extraction of chlorophyll and carotenoids. Absorbance of the supernatant was recorded at 664,2, 648,6 and 470 nm spectrophotometrically (Genesys 20, Thermo Scientific, USA). Pigments content calculated by the following formulas.

Chl $a=(13{,}36\cdot A_{664,2}-5{,}19\cdot A_{648,6})\cdot 25/DW$ Chl $b=(27{,}43\cdot A_{648,6}-8{,}12\cdot A_{664,2})\cdot 25/DW$

Chl $(a+b)=(5{,}24\cdot A_{664,2}+22{,}24\cdot A_{648,6})\cdot 25/DW$

$Car(x+c)=(4{,}785\cdot A_{470}+3{,}657\cdot A_{664,2}-12{,}76\cdot A_{648,6})\cdot 25/DW$

The flag leaf area (LA, sm^2) was measured with an area meter (AAC-400, Hayashi Denkon Co, LTD, Japan).Leaf dry weight was then determined, and Leaf Specific Mass (LSM, leaf dry matter per unit leaf area, mg mm^{-2}) was calculated. The relative water content (RWC) of the flag leaf was determined gravimetrically. Immediately after cutting at the base of lamina, leaves were preserved within plastic bags and in time transferred to the laboratory. Fresh weight (FW) was determined after removal and turgid weight (TW) was measured after saturating leaves in distilled water for 24 h at room temperature. After saturating, leaves were carefully blotted dried with tissue paper. Dry weight (DW) was measured after oven drying the leaves samples at 105°C for 24 h. RWC was calculated by using the following formula: RWC(%)= (FW-DW)/(TW-DW)x100.

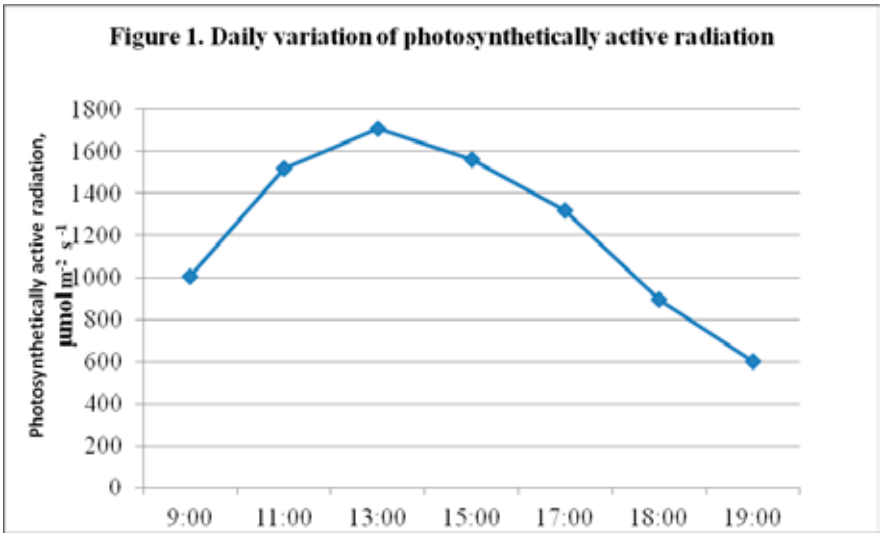


Figure 1. Daily variation of photosynthetically active radiation

Plant height and exposed peduncle length (the distance from the flag leaf ligule to the base of spike) were determined from 30 plants per plot. Spike weight, spike length and width, number of spikelet per spike, number and weight of grain per spike were determined from five plants per plot. Soil moisture content was determined in the 0-20, 20-40, 40-60 cm depths and expressed as percentage of the field moisture capacity (Table 1).

Correlations among parameters, standard errors of means were calculated by SPSS software.

Table 1. Soil moisture content (% of the field capacity)

Soil layer, cm	Irrigated	Rain-fed
Heading stage		
0-20	69,43±1,2	32,47±1,83
20-40	52,83±2,76	37,35±1,44
40-60	58,94±3,64	29,33±1,42
Grain formation		
0-20	61,04±0,84	24,18±0,85
20-40	59,94±1,23	32,13±1,16
40-60	60,72±0,63	15,94±1,18

Results and discussion

There were differences between genotypes for heading time (Table 2). Water stress affected heading time. The genotypes Garagylchyg 2, Gobustan, Azamatli 95 were early-heading. The genotypes Tale 38, 4thFWFWSN№50, Gyrmyzygul 1 were late-heading. Early- heading has been known as a major drought escaping mechanism, particularly in terminal drought stresses (Levitt 1980), allows plants to finish the life cycle before deeper water deficit. Early-heading genotypes have much time for the accumulation of assimilates in the grain.

Effect of drought stress on gas exchange parameters. Photosynthesis is the primary source of dry matter production and grain yield of crop plants (Shao et al. 2005). Leaf photosynthesis may vary with leaf age, position, leaf surface, and general plant and development stage (Richards 2000). Variations in daily time course of weather parameters such as light intensity, temperature, relative humidity, etc. also affect leaf gas exchange. Water deficit significantly affected the leaf gas exchange parameters (Table 3). In the heading stage, we did not observe a sharp decrease of flag leaf gas exchange parameters. Reduction in g_s during grain formation more affected on T_r, than P_n. This trend continued in the grain filling stage (date not shown).

Table 2. Number of days to 50% heading of wheat genotypes (days calculated from sowing time-1st November)

Wheat genotypes	Irrigated	Rain-fed
Garagylchyg 2	174	172
Vugar	183	180
Shiraslan 23	182	177
Barakatli 95	179	174
Alinja 84	178	172
Tartar	182	180
Gobustan	175	170
Giymatli 2/17	179	174
Gyrmyzygul 1	184	181
Azamatli 95	172	169
Tale 38	188	182
12ndFAWWON№97	182	180
4thFEFWSN№50	188	185

A strong reduction of P_n, g_s, T_r during grain formation were observed in durum wheat genotypes Vugar (42%, 79%, 60%), Alinja 84 (36%, 71%, 56%), Shiraslan 23 (34%, 85%, 69%), Barakatli 95 (35%, 69%, 50%), Tartar (31%, 72%, 52%), in bread wheat genotypes Gobustan (37%, 88%, 74%), Gyrmyzygul 1 (40%, 76%, 65%), Azamatli-95 (45%, 37%, 41%), 12ndFAWWONN№97 (35%, 64%, 49%). Relatively smaller reduction of gas exchange parameters were found in genotypes Giymatli-2/17, Tale-38, 4thFEFWSNN№50. Gas exchange parameters of genotypes Barakatli 95, Alinja 84, Gyrmyzygul 1, Azamatli 95 were strongly affected by water stress in both stages. In the heading stage, we found an increase in C_i, due to decreased conductance of mesophyll cells to CO_2. However, in the grain formation stage we found a reduction in C_i. Perhaps this was due to stronger decrease in gs. The mesophyll conductance (g_m) is determined by the rate of electron transport (Q-quenching) from photosystem II to photosystem I over the thylakoid membranes and by the rate of CO_2 assimilation by the Calvin cycle (E-quenching) (Schapendonk et al. 1989). The mesophyll conductance (g_m) was calculated as the ratio of P_n to C_i, water use efficiency (WUE) was calculated as the ratio of P_n to T_r (Table 4). The gm decreased, but the water use efficiency (WUE) increased under the influence of water stress.

Table 3.Gas exchange parameters of T.durum Desf.and T.aestivum L. genotypes in response to drought stress.

Wheat genotypes	Experiment condition	Pn, µmolCO_2 m^{-2} s^{-1}		g_s,molH_2O m^{-2} s^{-1}		C_i, µmolCO_2 mol^{-1}		T_r,mmolH_2O m^{-2} s^{-1}	
		Heading stage	Grain formation	Heading stage	Grain formation	Heading stage	Grain formation	Heading stage	Grain formation
Triticum durum Desf.									
Garagylchyg 2	Irrigated	14,2	18,1	0,892	0,529	348	303	4,36	6,13
	Rain-fed	11,3	16,6	0,688	0,223	355	246	3,47	3,81
Vugar	Irrigated	14,8	21,6	0,589	0,647	319	299	4,14	7,93
	Rain-fed	13,3	12,5	0,445	0,135	318	226	3,64	3,24
Shiraslan 23	Irrigated	17,2	16,3	0,584	0,568	322	310	4,84	7,25
	Rain-fed	13,8	10,8	0,559	0,087	345	291	4,50	2,24
Barakatli 95	Irrigated	18,2	22,0	0,540	0,555	315	302	4,85	8,13
	Rain-fed	13,0	14,3	0,410	0,173	318	225	4,01	4,10
Alinja 84	Irrigated	15,3	21,5	0,498	0,492	318	273	4,61	6,94
	Rain-fed	11,2	13,8	0,380	0,144	334	214	3,82	3,04
Tartar	Irrigated	18,4	23,5	0,501	0,640	302	282	4,55	8,84
	Rain-fed	13,5	16,2	0,426	0,173	309	195	4,37	4,21
Triticum aestivum L.									
Gobustan	Irrigated	14,8	16,5	0,642	0,717	335	338	5,05	6,57
	Rain-fed	11,9	10,4	0,524	0,086	338	314	4,49	1,71
Giymat-li-2/17	Irrigated	15,8	19,4	0,421	0,364	302	279	4,65	4,78
	Rain-fed	10,4	16,2	0,289	0,209	318	286	3,02	3,33
Gyrmyzy gul1	Irrigated	18,0	14,3	0,481	0,366	304	306	4,28	5,33
	Rain-fed	12,7	8,6	0,229	0,088	281	254	3,18	1,89
Azamatli 95	Irrigated	15,9	17,1	0,570	0,325	313	273	5,95	5,69
	Rain-fed	13,6	9,38	0,221	0,206	268	276	3,61	3,35
Tale-38	Irrigated	20,8	20,7	0,728	0,598	300	313	8,24	6,82
	Rain-fed	12,2	17,6	0,399	0,308	317	256	5,13	5,36
12 nd FAW-WON N97	Irrigated	13,2	16,2	0,260	0,312	279	278	4,59	5,32
	Rain-fed	10,7	10,5	0,238	0,113	288	254	3,37	2,69
4th FEFWSN N50	Irrigated	20,8	24,3	0,525	0,485	284	279	6,65	6,95
	Rain-fed	16,8	17,8	0,474	0,298	296	236	5,39	6,47

Table 4. Effect of drought stress on mesophyll conductance (gm) and water use efficiency (WUE)

Wheat genotypes	Experiment condition	gm molCO$_2$ m^{-2} s^{-1}		WUE µmolCO$_2$ mmol^{-1}H$_2$O	
		Heading stage	grain formation	Heading stage	grain formation
T.durum Desf.					
Garagylchyg 2	Irrigated	0,040	0,060	3,26	2,95
	Rain-fed	0,032	0,067	3,26	4,36
Vugar	Irrigated	0,046	0,072	3,57	2,72
	Rain-fed	0,042	0,055	3,65	3,86
Shiraslan 23	Irrigated	0,054	0,053	3,55	2,25
	Rain-fed	0,040	0,037	3,07	4,82
Barakatli 95	Irrigated	0,058	0,073	3,75	2,71
	Rain-fed	0,041	0,064	3,24	3,49
Alinja 84	Irrigated	0,048	0,079	3,32	3,10
	Rain-fed	0,034	0,064	2,93	4,54
Tartar	Irrigated	0,060	0,083	4,04	2,66
	Rain-fed	0,043	0,083	3,09	3,85
T.aestivum L.					
Gobustan	Irrigated	0,044	0,049	2,93	2,51
	Rain-fed	0,035	0,033	2,65	6,08
Giymatli 2/17	Irrigated	0,052	0,070	3,40	4,06
	Rain-fed	0,033	0,057	3,44	4,86
Gyrmyzygul 1	Irrigated	0,059	0,047	4,21	2,68
	Rain-fed	0,045	0,034	3,99	4,55
Azamatli 95	Irrigated	0,051	0,063	2,67	3,00
	Rain-fed	0,051	0,034	3,77	2,80
Tale 38	Irrigated	0,069	0,066	2,52	3,03
	Rain-fed	0,039	0,068	2,38	3,28
12ndFAWWON №97	Irrigated	0,047	0,058	2,88	3,04
	Rain-fed	0,037	0,041	3,17	3,90
4thFEFWSN №4	Irrigated	0,073	0,087	3,13	3,50
	Rain-fed	0,057	0,076	3,12	2,75

An increase in WUE could be due to more reduction in T_r than P_n by water deficit. More reduction of gm was observed in genotypes Shiraslan 23, Barakatli 95, Alinja 84, Tartar, Giymatli 2/17, Gyrmyzygul 1, Tale 38 during heading stage, in genotypes Vugar, Shiraslan 23, Alinja 84, Gobustan, Gyrmyzygul 1, Azamatli 95, 12ndFAWWON№97 during grain formation. This could be due to more decrease in Pn, than in C_i. A sharp increase in the WUE of genotypes Garagylchyg 2, Shiraslan 23, Gobustan, Gyrmyzygul 1 indicates a strong decrease in the T_r. Table 5 shows correlation between gas exchange parameters and calculated g_m and WUE under irrigated and rain-fed conditions. Positive and significant correlations were found between the P_n and g_s, T_r, g_m. Correlation between the P_n and gm, was more strong than between the P_n and g_s, indicating the dominance of g_m in reducing of P_n (Siddique et al. 1999). Negative correlation was observed between Pn and C_i. Positive correlations were observed between gs and C_i, T_r. Correlation between g_s and WUE was negative and significant under rain-fed condition. Negative and significant correlations were found between C_i and g_m. Correlation between T_r and gm was positive and significant. Negative and significant correlation was observed between T_r and WUE. Correlation between g_m and WUE was positive and significant under rain-fed condition.

Table 5. Correlation coefficients between gas exchange parameters, gm and WUE.

Irrigated	Parameters	P_n	g_s	C_i	T_r	g_m	WUE	Rain-fed
	Pn	1	0,433**	-0,070	0,819**	0,778**	0,058	
	gs	0,341**	1	0,592**	0,592**	0,019	-0,271*	
	Ci	-0,459**	0,500**	1	0,156	-0,594**	-0,399**	
	Tr	0,800**	0,366**	-0,305*	1	0,535**	-0,445**	
	gm	0,975**	0,196	-0,622*	0,766**	1	0,244*	
	WUE	0,130	-0,161	-0,228	-0,458**	0,163	1	

**, Correlation is significant at the 0,01 level; *, Correlation is significant at the 0,05 level

Effect of water deficit on RWC. During drought stress, the water balance of plants is disrupted, as result of which the RWC and water potential of leaves decreases (Bajjii et al.2001). Although RWC was higher in non-stressed plants than stressed ones, there were no significant differences between cultivars at these levels of RWC (Figure 2). Higher RWC was observed in genotypes Barakatli 95, Alinja 84, Tartar, Gyrmyzygul 1, Tale38, 12ndFAWWONN№97, and 4thFEFWSNN№50. The genotypes Tartar, Gyrmyzygul 1, Tale 38, 12ndFAWWONN№97, and 4thFEFWSN №50 were late heading, and their younger flag leaves contained relatively more water. Lower RWC was observed in genotypes Shiraslan 23, Gobustan, Giymatli 2/17, Azamatli95. The genotypes Azamatli 95 and Gobustan were the earliest heading. Under the influence of water stress significant reduction of RWC was found in genotypes Garagylchyg 2 (12%), and Giymatli 2/17(14%). A slight decrease of RWC was observed in genotypes Vugar, Alinja

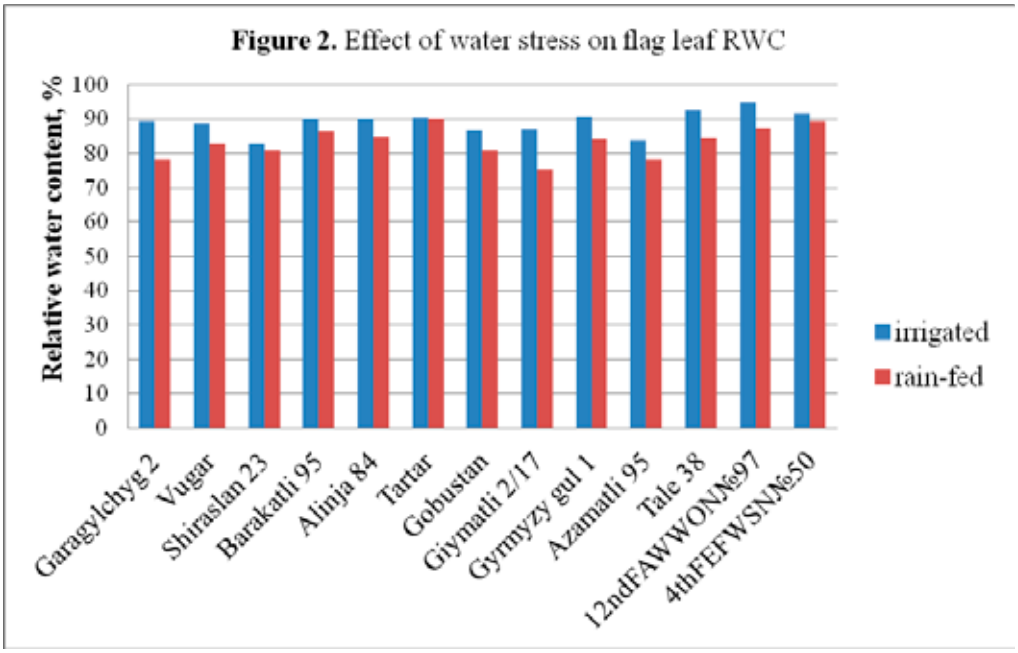


Figure 2. Effect of water stress on flag leaf RWC

84, Gobustan, Gyrmyzygul 1, Azamatli 95, Tale 38, 12ndFAWWONN№97, non-significant reduction in genotypes Shiraslan 23, Barakatli 95, and 4thFEFWSNN№50. The difference in RWC of irrigated and rain-fed plants was almost imperceptible in genotype Tartar. In the field, strengthening of water stress occurs gradually, it allows plants to develop various mechanisms of adaptation to resist to water scarcity.

Effect of water stress on flag leaf area. Water stress limits the growth of assimilating surface area of flag leaf of tested wheat genotypes (Figure 3). The reduction in leaf size which results in smaller transpiring area, is an adaptive response to water deficit (Tardieu 2005). A significant decrease in the flag leaf area was observed in all genotypes. More profound reduction of flag leaf area was observed in genotypes Shiraslan 23 (44%) and Vugar (35%), Gyrmyzy gul 1(37%), Tale 38 (34%), Garagylchyg 2 (31%), Barakatli 95 (31%), 4thFEFWSNN№50 (30%), 12ndFAWWONN№97 (28%), Tartar (28%). Relatively little reduction of flag leaf area under water stress was observed in genotypes Azamatli 95(18%), Alinja 84 (20%), Gobustan (23%), Giymatli 2/17 (23%). Deep reduction can be explained to the fact that the formation of the flag leaf of late- heading wheat genotypes

(Vugar, Shiraslan 23, Tartar, Gyrmyzy gul1, Tale 38, 4thFEFWSN№50, and 12ndFAWWON№97) occurs at a severe water shortage. A more profound reduction of flag leaf area in these genotypes was compensated with conservation of RWC at high level.

Effect of water stress on flag leaf dry biomass. A common adverse effect of water stress on crop plants is the reduction in fresh and dry biomass production (Zhao et al. 2006). Water scarcity causes a decrease in dry biomass of flag leaf (Figure 4).

As in the case of leaf area, a strong reduction

Figure 3. Effect of water stress on flag leaf area

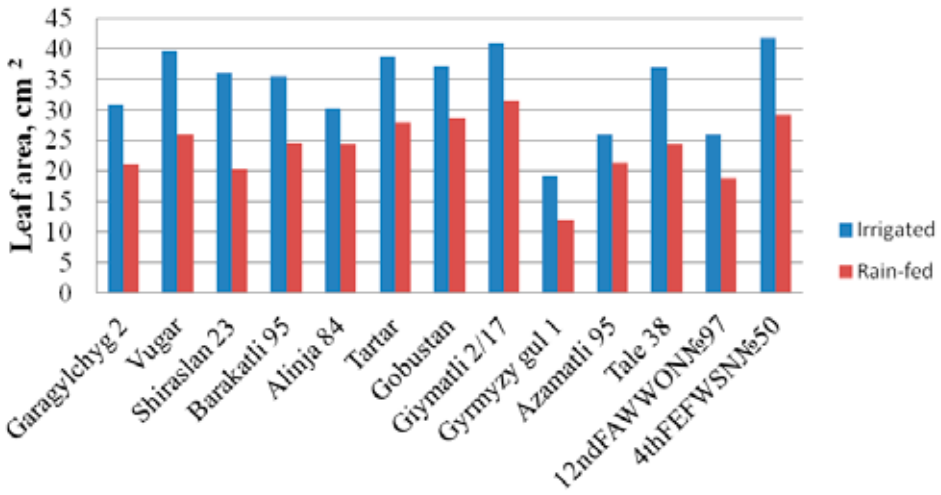
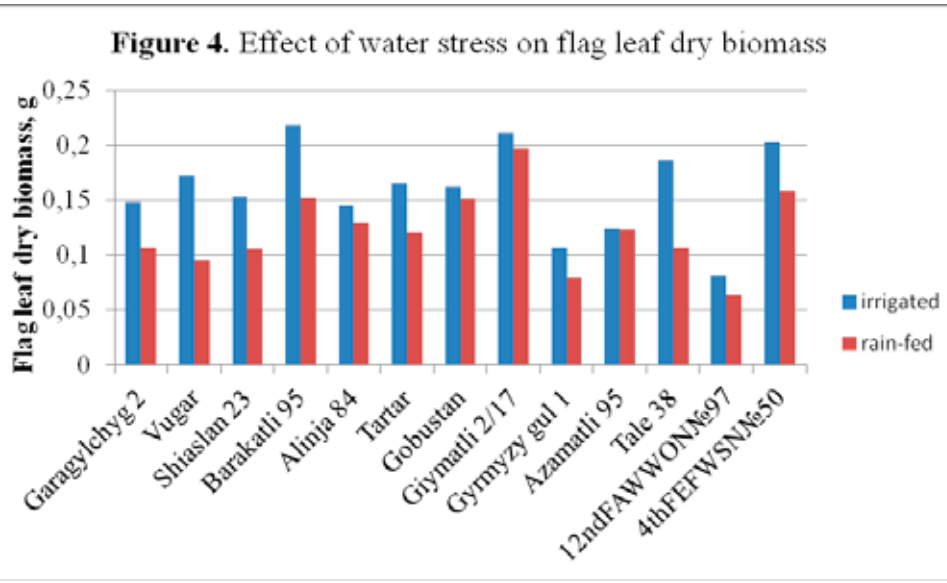


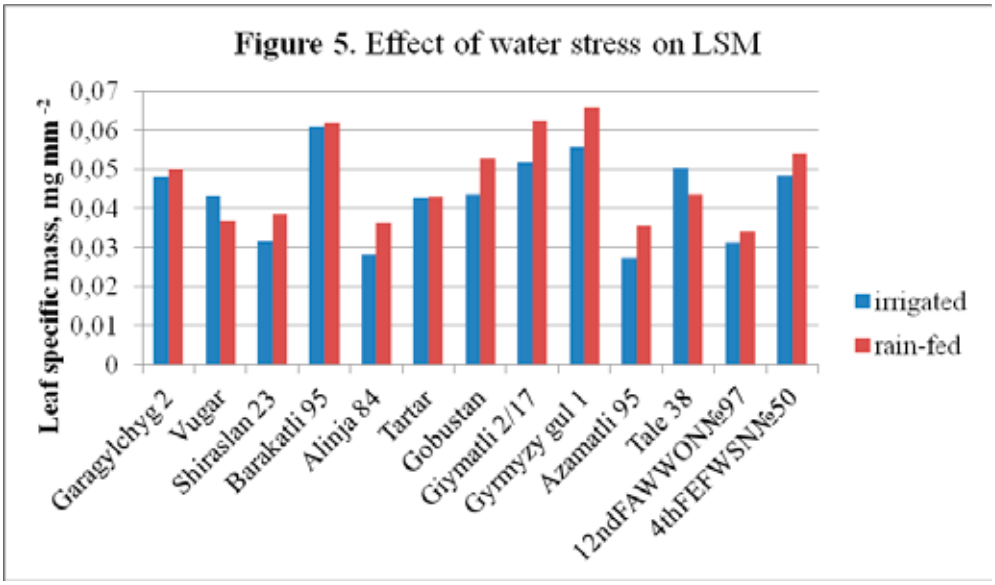
Figure 4. Effect of water stress on flag leaf dry biomass



of dry biomass was observed in all genotypes of durum wheat, with exception of Alinja 84, in bread wheat genotypes Gyrmyzygul 1, Tale 38, 12ndFAWWON№97, 4thFEFWSN№50. A smaller reduction of flag leaf dry biomass under water stress was observed in genotypes Azamatli 95, Gobustan, Giymatli 2/17, Alinja 84. A more profound reduction of flag leaf dry mass was detected in genotypes Vugar (44%) and Tale 38 (43%).

Effect of water stress on Leaf Specific Mass (LSM). LSM calculated from the ratio of flag leaf dry mass to flag leaf area and it is inverse leaf specific area. LSM is considered to reflect relative carbon accumulation, at lower nutrient or moisture availabilities or at higher light irradiances, leaves tended to be smaller, with higher LSM, density and thickness (Witkowski and Byron 1991). It was revealed an increase of LSM under water stress in most wheat genotypes (Figure 5).Such an increase in the LSM is probably adaptive response to drought and is due to the relatively greater reduction in leaf area than the dry mass. A reduction of LSM was observed in genotypes Vugar and Tale 38, because of the greater reduction in dry mass. A higher LSM was observed in genotypes Barakatli 95, Gyrmyzygul 1, Giymatli 2/17, Tale 38, 4thFEFWSN№50,

Garagylchyg 2, lower LSM was observed in



genotypes Azamatli 95, Alinja 84, 12ndFAWWON№97, Shiraslan 23. A slight increase in LSM was observed in genotypes Garagylchyg 2, Barakatli 95, Tartar, more profound increase was observed in genotypes Azamatli 95, Alinja 84, Shiraslan 23, Giymatli 2/17, Gobustan, Gyrmyzygul 1.

Effect of water stress on photosynthetic pigments content. Photosynthetic pigments are important to plants mainly for harvesting light and production of reducing powers (Anjum et al. 2011). In general, water stress caused significant declines in photosynthetic pigments content, in the ratio of Chl(a+b)/Car(x+c) and increase in the ratio of Chla/b (Table 6).The decrease in chlorophyll content under drought stress may be the result of pigment photo-oxidation and chlorophyll degradation. Lower values of the ratio Chl(a+b)/Car(x+c) indicates water stress damage to the photosynthetic apparatus, which is expressed by faster breakdown of chlorophylls than carotenoids. Photosynthetic pigments were higher among bread wheat genotypes than durum wheat ones. Higher decrease of chlorophyll content was observed in genotypes Vugar (35%), Shiraslan 23 (29%), Barakatli 95 (21%), Gobustan (29%), Giymatli 2/17 (31%), Azamatli 95 (37%), and 4thFEFWSN№50 (28%). A slight decrease was observed in genotypes Gyrmyzy gul 1, 12ndFAWWON№97, Alinja 84, Tale 38 and Garagylchyg 2. An increase in Chl a/b could be due to more reduction in Chlb than Chla by water deficit.

Correlations between physiological parameters. Table 7 shows correlations between studied physiological parameters. The Pn was positively and significantly correlated with RWC, LA, and DW. The relationship between P_n and C_{hl} content was positive, but non-significant. Because the LSM is characteristics for water stress condition, the correlation between the Pn and LSM was negative, but non-significant. The RWC positively and significantly correlated with C_{hl} content, positively but non-significantly correlated with LA and DW. Correlation between LA and DW was positive and significant, correlation between LA and Chl was positive but non-significant. The DW was positively, non-significantly correlated with LSM.

Effect of water stress on plant height, exposed peduncle length, spike components: Plant height and number of spikelet per spike, spike length and width were not reduced significantly under the influence of soil drought (Table 8). However, spike weight, number and weight of grains per spike were severely, as well as the exposed peduncle length was significantly reduced under the influence of soil drought.The decrease in the height of cultivars was more expressed among bread wheat genotypes. A significant reduction in plant height was observed in durum wheat genotype Tartar, and in all genotypes of bread wheat with the exception of Gobustan. The exposed peduncle has been identified as one of the photosynthetically active organs in wheat, contributes about 9-12% of grain dry mass (Wang et al. 2001). Long exposed peduncle was detected in genotypes Vugar, Shiraslan 23, Gobustan, Azamatli 95, 4thFEFWSN№50, short exposed peduncle was detected in genotypes Giymatli 2/17, Gyrmyzygul 1. Spike weight, grain number and grain weight per spike were more reduced in durum wheat genotypes Garagylchyg 2, Vugar, Barakatli 95, Alinja 84, Tartar, in bread wheat genotypes Gobustan, Giymatli 2/17, Gyrmyzygul 1, Azamatli 95, Tale 38, 12ndFAWWON№97, 4thFEFWSN№50. Large aboveground biomass was formed in genotypes Shiraslan 23, Gobustan, Tale 38, Gyrmyzygul 1 less in genotypes 12ndFAWWON№97, 4thFEFWSN№50. More reduction of aboveground biomass was observed in genotypes Vugar, Shiraslan 23, Alinja 84, Gobustan, Tale 38 less reduction in

Table 6. Changes of Chl a, b and Chl (a+b) contents, Car (x+c) content, Chl a/b and Chl (a+b)/Car (x+c) of wheat genotypes under water stress condition.

Wheat genotypes		Chl a mg g^{-1}dw	Chl b mg g^{-1}dw	Chl (a+b) mg g^{-1}dw	Car (x+c) mg g^{-1}dw	Chl a/b	Chl (a+b)/ Car (x+c)
T. durum Desf.							
Garagylchyg 2	irr.	7,14	3,34	10,48	1,76	2,14	5,96
	r-f	5,50	3,06	8,56	1,18	1,80	7,25
Vugar	irr.	6,02	2,93	8,95	1,45	2,06	6,16
	r-f	4,00	1,86	5,86	0,98	2,15	5,97
Shiraslan 23	irr.	5,68	2,68	8,36	1,41	2,12	5,93
	r-f	4,08	1,89	5,97	1,02	2,15	5,84
Barakatli 95	irr.	6,08	2,81	8,89	1,54	2,16	5,76
	r-f	4,83	2,19	7,02	1,15	2,21	6,09
Alinja 84	irr.	5,10	2,66	7,76	1,24	1,92	6,26
	r-f	4,46	2,01	6,47	1,16	2,22	5,57
Tartar	irr.	4,90	2,51	7,41	1,17	1,96	6,34
	r-f	6,23	2,69	8,92	1,58	2,32	5,66
T.aestivum L.							
Gobustan	irr.	6,78	3,30	10,08	1,58	2,06	6,37
	r-f	5,08	2,57	7,65	1,20	1,98	6,35
Giymatli 2/17	irr.	5,85	2,68	8,53	1,38	2,18	6,17
	r-f	4,07	1,84	5,91	1,12	2,21	5,26
Gyrmyzygul 1	irr.	7,19	3,22	10,41	1,86	2,23	5,60
	r-f	7,17	3,06	10,24	1,93	2,34	5,31
Azamatli 95	irr.	6,68	3,70	10,38	1,38	1,81	7,50
	r-f	4,43	2,06	6,49	1,12	2,15	5,82
Tale 38	irr.	7,68	3,54	11,22	1,84	2,17	6,08
	r-f	6,44	3,13	9,57	1,60	2,06	5,99
12^{nd}FAWWON №97	irr.	6,80	3,57	10,37	1,67	1,98	6,21
	r-f	6,68	3,29	9,97	1,65	2,03	5,98
4^{th}FEFWSN №50	irr.	7,14	3,49	10,63	1,80	2,04	5,92
	r-f	5,20	2,49	7,69	1,34	2,08	5,75

Note: irr.-irrigated; r-f.-rain-fed

Table7. Correlations between different physiological parameters

Parameters	P_n	RWC	LA	DW	LSM	Chl
Pn	1					
RWC	0,527**	1				
LA	0,798**	0,321	1			
DW	0,674**	0,116	0,845**	1		
LSM	-0,171	-0,327	-0,201	0,330	1	
Chl	0,274	0,623**	0,113	-0,043	-0,235	1

**. Correlation is significant at the 0, 01 level

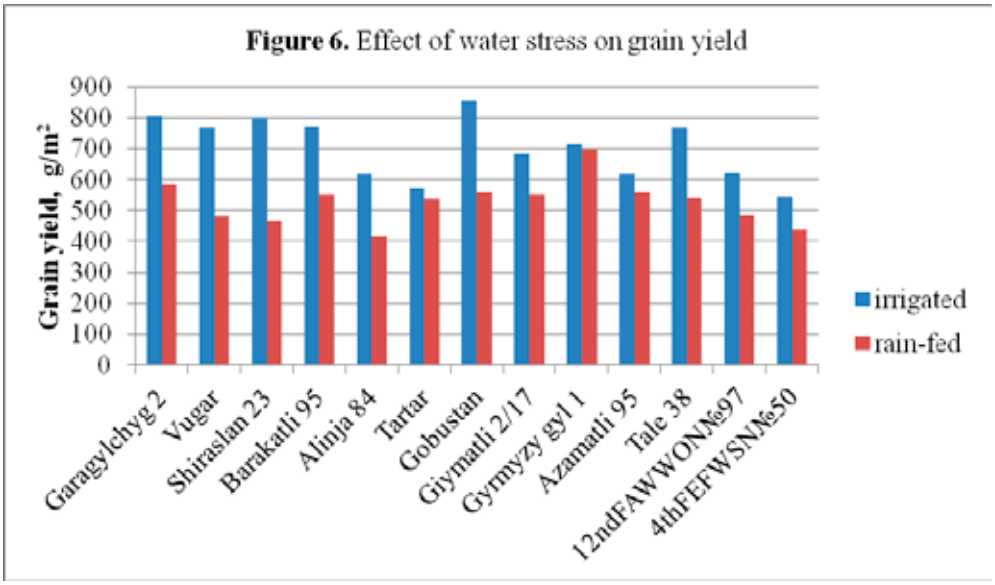
genotypes Tartar, 4thFEFWSN№50, Gyrmyzygul 1, Giymatli 2/17. 1000 kernel weight (TKW) is a major yield component determining final yield, it may be a form of compensation for the spike reduction under water deficit condition (Moayedi et al. 2010). TKW was higher in genotypes Alinja 84, Tartar, Giymatli 2/17, was lower in genotypes Gyrmyzygul 1, 12ndFAWWON№97 and 4thFEFWSN№50. Profound decrease in the TKW observed in genotypes Tale 38, 12ndFAWWON№97 and 4thFEFWSN №50. The harvest index (HI) is the proportion of grain yield to biological yield and it shows the ability of the plants to translocate physiological matters to grains. We found an increase in HI in all genotypes with the exception of Gobustan.

In rain-fed condition the ratio of grain yield to

Table 8. Effect of drought stress on yield and yield components

	Genotypes	Plant height, cm	Exposed peduncle, cm	AGB, g	Spike weight, g	Spike length, cm	Spike width, cm	Spikelet per spike	Grain number per spike	Grain weight per spike,g	TKW g	HI
Irrigated condition	Garagilchig 2	100,00±0,49	18,57±0,24	1924	4,77±0,35	7,8±0,16	1,62±0,06	22,2±0,97	68,2±2,82	3,65±0,29	49,56	0,42
	Vugar	103,13±0,72	22,42±0,30	1971	4,42±0,26	7,8±0,15	1,64±0,02	22,2±0,58	68,0±3,56	3,43±0,18	45,35	0,39
	Shiraslan 23	102,64±0,42	21,32±0,39	2053	3,50±0,18	8,0±0,21	1,56±0,05	19,8±0,70	50,0±3,65	2,72±0,16	47,51	0,39
	Barakatli 95	101,97±0,72	18,29±0,21	1880	3,53±0,12	7,86±0,29	1,54±0,06	21,4±0,24	60,4±0,81	2,62±0,13	49,41	0,41
	Alinja 84	102,97±0,50	18,26±0,25	1584	4,31±0,31	7,90±0,10	1,48±0,05	18,2±0,49	59,2±2,03	3,37±0,19	53,80	0,39
	Tartar	104,81±0,68	16,51±0,21	1587	4,77±0,48	8,63±0,16	1,78±0,07	20,0±0,57	57,6±6,28	3,40±0,36	57,63	0,36
	Gobustan	109,83±0,58	22,24±0,33	2021	3,41±0,14	10,86±0,07	1,28±0,03	17,8±0,2	58,6±2,5	2,72±0,12	44,45	0,42
	Giymatli 2/17	104,07±0,65	13,07±0,27	1657	4,04±0,26	8,98±0,15	1,56±0,04	21,4±0,51	65,8±2,71	3,30±0,24	51,38	0,41
	Gyrmyzygul 1	91,54±0,42	9,66±0,09	1949	2,11±0,10	7,78±0,16	1,14±0,05	16,0±0,31	49,4±2,23	1,71±0,09	36,07	0,37
	Azamatli 95	108,61±0,48	24,49±0,29	1788	3,28±0,08	11,90±0,42	1,42±0,03	18,4±0,24	60,0±2,21	2,55±0,10	43,56	0,35
	Tale 38	106,5±0,47	17,58±0,22	2039	4,11±0,14	11,16±0,14	1,22±0,05	21,0±0,31	64,2±2,81	3,02±0,10	44,03	0,38
	12ndFAWWON№97	95,75±0,48	16,53±0,18	1498	1,88±0,15	8,10±0,26	1,12±0,04	14,3±0,49	39,0±2,39	1,49±0,12	37,22	0,42
	4thFEFWSN№50	104,35±0,42	21,48±0,22	1334	3,65±0,11	10,38±0,08	1,46±0,02	19,2±0,37	62,2±0,91	2,73±0,09	42,66	0,41
Rain-fed condition	Garagylchyg 2	93,91±0,57	15,29±0,16	1318	3,17±0,06	7,6±0,22	1,16±0,05	20,0±0,71	44,2±1,39	2,42±0,03	48,64	0,44
	Vugar	97,87±0,35	16,42±0,29	1186	3,17±0,18	7,0±0,17	1,48±0,05	20,2±0,58	50,8±2,54	2,59±0,16	44,74	0,41
	Shiraslan 23	98,48±0,47	16,88±0,25	1146	2,72±0,20	6,88±0,09	1,40±0,03	18,7±0,42	45,0±2,31	2,17±0,20	43,91	0,41
	Barakatli 95	96,12±0,54	15,13±0,14	1311	2,70±0,19	6,82±0,27	1,36±0,07	17,8±0,58	41,8±2,15	2,12±0,18	45,94	0,42
	Alinja 84	98,12±0,48	14,72±0,23	1046	2,75±0,14	7,44±0,42	1,32±0,04	18,2±0,58	42,0±2,49	2,12±0,17	47,66	0,40
	Tartar	93,59±0,41	12,72±0,20	1290	3,25±0,40	8,12±0,38	1,38±0,05	18,8±0,8	44,6±4,69	2,46±0,31	54,12	0,42
	Gobustan	101,93±0,45	18,67±0,29	1353	2,17±0,27	9,62±0,24	1,00±0,03	16,2±0,37	43,6±3,53	1,75±0,20	41,73	0,41
	Giymatli 2/17	89,22±0,72	10,89±0,25	1279	2,65±0,15	8,44±0,11	1,24±0,04	18,6±0,24	47,0±1,84	2,11±0,13	46,43	0,43
	Gyrmyzygul 1	81,56±0,46	7,61±0,10	1518	1,30±0,04	7,72±0,03	0,98±0,02	15,5±0,22	36,0±1,87	1,07±0,07	31,72	0,46
	Azamatli 95	97,10±0,46	19,82±0,33	1346	1,85±0,14	9,84±0,34	1,10±0,04	14,4±0,50	33,8±2,88	1,39±0,09	38,80	0,42
	Tale 38	91,24±0,48	11,22±0,17	1391	1,72±0,08	8,60±0,07	1,08±0,03	16,6±0,24	36,8±1,15	1,14±0,06	35,94	0,39
	12ndFAWWON№97	84,90±0,45	14,67±0,20	1103	1,26±0,04	7,04±0,14	0,96±0,02	12,4±0,24	29,6±0,40	1,01±0,04	30,50	0,44
	4thFEFWSN№50	94,06±0,59	18,76±0,17	1053	2,05±0,13	8,96±0,33	1,12±0,03	16,2±0,73	41,4±2,80	1,51±0,09	30,33	0,42

Note: AGB-aboveground biomass, TKW-thousand kernel weight, HI- harvest index



AGM significantly increased in genotypes Tartar, Gyrmyzygul 1, Azamatli 95.

The grain yield is the total out-put of all the yield components. The average yield of all genotypes dropped considerably under water deficit condition (Figure 6). More reduction of grain yield was observed in genotypes Vugar (37%), Shiraslan 23(42%), Barakatli 95 (29%), Alinja 84 (33%), Gobustan (35%), and Tale 38 (29%). We consider these genotypes as drought susceptible. Less reduction of grain yield was observed in genotypes Gyrmyzygul 1(2%), Tartar (6%), Azamatli 95(9%). We consider these genotypes as drought tolerant.

In the present study, it was observed that leaf gas exchange parameters (P_n, g_s, T_r) were positively correlated with DH, PH, AGB, SW, spike width, spikelet per spike, GNS, GWS (Table 9). Correlation between g_s and EPL, g_s and GY, also T_r and EPL, T_r and GY were significant. The lack of significant correlation between P_n and grain yield suggests that selection for higher rates of leaf photosynthesis has not improved yield most probably because the source is less limiting than the sink (Bogale et al. 2011). LA was strongly correlated with PH and SW, SGN and SGW, which suggests that large leaf area contributes formation of more assimilates that is transported to the spike. RWC was only significantly correlated with DH. LDW was positively and significantly correlated with PH, AGB, SW, spike width, spikelet per spike, SGN and SGW. Chl content was positively and significantly correlated with PH, spike/m^2, AGB, SL and GY. PH, SW, spikelet per spike, SGN, SGW positively and significantly correlated with most physiological parameters. Therefore, these traits may deem a good criterion for selection.

Note: DH- days to heading, PH-plant height,

Table 9. Correlations between physiological parameters and plant height, exposed peduncle length, yield and yield components

Traits	Pn	gs	Tr	LA	RWC	LDW	LSM	Chl
DH	0,471*	0,390*	0,496**	0,321	0,688**	0,130	-0,282	0,444*
PH	0,566**	0,665**	0,588**	0,742**	0,170	0,587**	-0,199	0,080
EPL	0,325	0,484*	0,458*	0,504**	0,00	0,322	-0,300	0,067
Spike/m2	-0,056	0,132	0,034	-0,073	0,384	-0,136	-0,086	0,656**
AGB	0,435*	0,775**	0,612**	0,498**	0,324	0,413*	-0,024	0,545**
GY	0,329	0,723**	0,515**	0,416*	0,283	0,361	0,033	0,563**
SW	0,683**	0,696**	0,659**	0,635**	0,261	0,496*	-0,160	0,059
SL	0,271	0,383	0,312	0,360	0,027	0,329	0,001	0,412*
SWH	0,600**	0,578**	0,641**	0,555**	0,218	0,414*	-0,166	-0,145
Spikelet/Spike	0,584**	0,534**	0,524**	0,529**	0,121	0,511**	0,036	-0,028
SGN	0,745**	0,788**	0,720**	0,715**	0,378	0,621**	-0,074	0,297
SGW	0,669**	0,695**	0,652**	0,634**	0,257	0,495*	-0,162	0,040
TKW	0,456*	0,371	0,375	0,408*	0,020	0,345	-0,066	-0,263
HI	-0,493*	-0,463*	-0,525**	-0,451*	-0,223	-0,327	0,173	-0,094

**, Correlation is significant at the 0, 01 level *, Correlation is significant at the 0, 05 level

EPL-exposed peduncle length, S/m^2-spike per m^2, AGB-aboveground biomass, GY-grain yield, SW-spike weight, SL-spike length, SWH- spike width, SGN-spike grain number, SGW-spike grain weight, TKW- thousand kernel weight, HI- harvest index

Conclusion

Soil water deficit caused a decrease in gas exchange parameters, area, dry weight, photosynthetic pigments of flag leaf from durum and bread wheat genotypes. Stomatal conductance regulates photosynthesis rate and transpiration rate. Strong relationships were detected between stomatal conductance and transpiration rate, between mesophyll conductance and photosynthesis rate. Photosynthesis is less inhibited than transpiration rate under water stress. Despite the fact that the gas exchange parameters, leaf area and dry mass strongly influenced by drought, relative water content in the flag leaf remained relatively high. Water scarcity leads to a greater reduction in leaf area than the dry weight, as a result of the specific leaf mass increases. Under influence of water stress the content of photosynthetic pigments, also the ratio of chlorophyll to carotenoids decreases. Water stress significantly reduces spike weight, grain number and grain weight per spike. Grain yield non-significantly correlated with photosynthesis rate, but significantly correlated with chlorophyll content. Most decrease in the area and dry mass of flag leaf was observed in durum wheat genotypes. The chlorophyll content was highest in the flag leaf of bread wheat genotypes. The greatest reduction in plant height was observed in genotypes of bread wheat. The largest decrease in the weight of the spike, in the number and weight of grain per spike were observed in genotypes of bread wheat. Durum wheat genotype Vugar, and bread wheat genotype Tale 38 were more sensitive to soil drought.

References

Akram M (2011). Growth and yield components of wheat under water stress of different growth stages. Bangladesh J.Agril.Res. 36(3) :455- 468.

Aliyev J.A (2001). Physiological bases of wheat breeding tolerant to water stress. Proceedins of the 6th International Wheat Conference, Budapest, Hungary, 2000. In: Wheat in a Global Environment (Bedo Z, Lang L, eds.), Kluwer Academic Publishers, Dordrecht, Boston, London 9:693-698.

AnjumSA, Xie X, Wang L, Salem MF, Man Cand Lei W (2011). Morphological, physiological and biochemical responses of plants to drought stress. African Journal of Agricultural Research 6(9): 2026-2032.

Bajjii M, Lutts S, Kinet KM (2001). Water deficit effects on solute contribution to osmotic adjustment as a function of leaf ageing in three durum wheat (*Triticum durum* Desf) cultivars performing in arid conditions. Plant Science 60: 669-681.

Beltrano J, Marta GR (2008). Improved tolerance of wheat plants (*Triticumaestivum* L.) to drought stress and rewatering by the arbuscularmycorrhizal fungus Glomusclaroideum: Effect on growth and cell membrane stability. Braz.J.Plant Physiol 20(1): 29-37.

Blum A (1986). Breeding crop varieties for stress environments.Critical Reviews in Plant Sciences 2: 199-237.

Bogale A, Tesfaye K, Geleto T (2011). Morphological and physiological attributes associated to drought toleranxe of Ethiopian durum wheat genotypes under water deficit condition. Journal of Biodiversity and Environmental Sciences 1(2):22-36.

Chaves MM (1991). Effects of water deficits on carbon assimilation. Journal of Experimental Botany 42: 1-16.

Chaves MM, Pereira JS, Maroco J, Rodrigues ML, Ricardo CP, Osorio ML, CarvalhoJ, Faria T and Pinheiro C (2002). How Plants Cope with Water Stress in the Field. Photosynthesis and Growth. Annals of Botany 89:907-916.

Cornic G, Massacci A (1996). Leaf photosynthesis under drought stress. In: Baker NR, ed. Photosynthesis and Environment. The Netherlands: Kluwer Academic Publishers.

Flexas J, Bota J, Loreto F, Cornic Gand Sharkey TD (2004). Diffusive and metabolic limitations to photosynthesis under drought and salinity in C3 plants. Plant Biology 6: 269-279.

Kramer PJ (1980). Drought, stress, and the origin of adaptation. In Adaptation of Plants to Water and High Temperature Stress (eds N. C. Turner and P.J.Kramer) pp. 7-20. John Wiley and Sons, New York, NY, USA.

Lawlor DW, Cornic G. (2002). Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. Plant, Cell & Environment 25: 275-294.

Levitt J(1980). Stress terminology. In: Turner NC, Kramer PJ, editors. Adaptation of plants to water and high temperature stress. New York: Wiley pp.439-473.

Lichtenthaler HK (1987). Chlorophylls and carotenoids: Pigments of photosynthetic biomembranes.- Methods in Enzymology 148: 350-382.

Mitchell JH, Siamhan D, Wamala MH, Risimeri JB, Chinyamakobvu E, Henderson SA, Fukai S (1998). The use of seedling leaf death score for evaluation

of drought resistance of rice. Field Crops Research 55: 129-139.

Moayedi AA, Boyce AN, Barakbah SS (2010). The Performance of Durum and Bread Wheat Genotypes Associated with Yield and Yield Component under Different Water Deficit Conditions. Australian Journal of Basic and Applied Sciences 4(1): 106-113.

Richards A (2000). Selectable traits to increase crop photosynthesis and yield of grain crops. Journal of Experimental Botany 51:447-458.

Schapendonk AHCM, Spitters CJT, Groot PJ (1989) .Effects of water stress on photosynthesis and chlorophyll fluorescence of five potato cultivars. Potato Research 32: 17-32.

Shao HB, Liang ZS, Shao MA, Wang BC (2005). Changes of some physiological and biochemical indices for soil water deficits among 10 wheat genotypes at seedling stage.Colloids Surf. B: Biointerfaces 42(2): 107-211.

Siddique MRB, Hamid A, Islam MS (1999). Drougth stress effects on photosynthetic rate and leaf gas exchange of wheat. Bot.Bull.Acad.Sin. 40: 141-145.

Tang AC, Kawamitsa Y, Kanechi M and Boyer JS (2002). Photosynthesis at low water potentials in leaf discs lacking epidermis. Annals of Botany 89(7):861-870.

Tardieu F (2005). Plant tolerance to water deficit: physical limits and possibilities for progress. Comptes Rendus Geoscience 337: 57-67.

Veesar NF, Channa AN, Rind MJ, Larik AS (2007). Influence of water stress imposed at different stages on growth and yield attributes in bread wheat genotypes (*Triticum aestivum* L.) Wheat Inf. Serv. 104: 15-19.

Wang ZM, Wei AI, Zheng DM (2001). Photosynthetic characteristics of non-leaf organs of winter wheat cultivars differing in ear type and their relationship with grain mass per ear. Photosynthetica 39(2):239-244.

Witkowski, ETF and Lamont BB (1991). Leaf specific mass confounds leaf density and thickness. Oecologia 88(4): 486-493.

Zhao TJ, Sun S, Liu Y, Liu JM, Yan YB, Zhou HM (2006). Regulating the drought-responsive element (DRE)-mediated signaling pathway by synergic functions of transactive and transinactive DRE binding factors in Brassica napus. J.Biol.Chem. 281: 10752-10759.

Research Article

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Reaction of wheat germplasm to stem rust in Georgia

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ABSTRACT

Stem rust had represented a major threat to wheat production in the world including Georgia. Breeding for resistance to rusts is a major strategy for most wheat improvement programs. The wild and domestic relatives of wheat are important sources for disease resistance. Therefore, the objective of this study was identification of resistant genotypes to Georgian races of stem rust.

A collection of fifty wheat accessions including endemic wheat species and subspecies, domestic varieties, new advanced cultivars and introduced entries from different international nurseries were evaluated under the artificial infection of stem rust in the field and greenhouse conditions. The wheat germplasm was screened using the predominant stem rust races mixture of Georgia. Resistance was detected in majority of the tested entries. The endemic species: *Triticum monoccoccum* (var.*laetissimum* Korn), *Triticum timopheevi* (var.*tipicum* Zhuk -var.*viticulosum* Zhuk), *Triticum georgicum* (var.*chvamlicum* Supat), *Triticum dicoccum* (var. *farrum*), *Triticum carthlicum* Men (var. *fuliginosum* Zhuk), *Triticum carthlicum* Men (Var. *stramineum* Zhuk), *Triticum macha* Dek et Men (var. *megrelicum*), *Triticum macha* Dek et Men (var. *colchicum*), *Triticum macha* Dek et Men (var. *palaeo-imereticum*), *Triticum spelta* (var.*dekaprelevichi Dorof*) and old Georgian varieties: Khulugo, Tetri ipkli were resistant and moderate resistant to stem rust. Also, some introduced accessions (DBDI-2WWSRRN-34, Dorade/altay2000/4Bez/Nad//LZM (es85.24)
3/F900k, Haurani / aegtaushi / cham6-6 / mz / cno67 / 3Ifn / 4 / ant / 5 / Attila-19FAWWON, Sunco / pastor-19FAWWON, SRMA / tui / / babax / 3JGR-11LR-Res-132, BTZ-18FAWWON-IRR-149) showed moderate resistance.

***Keywords*:** wheat, landraces, species, resistance, rust

Introduction

Georgia is a country of origin of wheat and presents an ancient center of agriculture. At the same time it is distinguished by a diversity of a number of unique endemic species and old local varieties of different crops. Therefore, the genetic resources spread in Georgia are significant not only from historic point of view, but also for selection of the valuable varieties for cultivation.

The wheat is very old crop for Georgia; it is presented with very large diversity which is stipulated by variation of soil and climate condition of Georgia. There has been described 14 species of wheat out of which 5 are endemic and there are numerous Georgian landraces of wheat. The local wheat has many unique features such as high immunity, and high biochemical

and technological quality of grain. They also were adapted well to the local conditions (Menabde, 1948). The bread and other products made from this wheat were highly appreciated among local population and they were deeply connected with national traditions and custom. However, local varieties have been disappearing from agriculture. The new commercial varieties replaced land races. Most of these varieties are susceptible to the local races of major diseases and produce low yield. The low yield of wheat in region can be explained by stable development of diseases, low-yielding and poorly-adapted varieties etc. Breeding for disease resistance remains the major way for control of diseases.

Rusts are recognized as a widely occurring and most damaging disease of wheat in the world. Wheat stem rust is a most feared disease due to its ability to inflict substantial losses. The last major stem rust epidemics occurred in various countries (Kenya, Ethiopia, Uganda, Yemen) caused grain losses of up to 70% in farmer's fields (Wanuera, 2008; Pretorius et al, 2000; Bariana, 2008). FAO estimates that 30 countries are either affected by Ug99 or at potential risk. There is a chance that Ug99 will reach Georgia where barberry is widespread (Anonymous, 2008). However, stem rust incidence and severity recorded during 2011-2013 cereal rust surveillance surveys were mainly moderate and low, respectively (Sikharulidze et al, 2013).

Because a large proportion of the world's commercial wheat varieties are susceptible to Ug99, new sources of rust resistance are required for breeding improved varieties. Growing of resistant varieties is well-known to be the cheapest and ecologically safe method of the crop protection. Creation of such varieties requires constant search of the trait donors i.e. new sources with high level of resistance expression. The wild and domestic relatives of common wheat are important sources for resistance to diseases. In addition, as rust pathogens are highly specialized and evolution of new virulence is more frequent in pathogen population, breeding for resistance to this disease should always be more systematic and dynamic.

The objective of this study was to evaluate the stem rust responses of a range of wild relatives, local varieties and introduced germplasm from international nurseries.

Materials and methods

A collection of fifty wheat accessions including twelve endemic wheat species and subspecies, nine domestic varieties, seven advanced varieties and nineteen introduced entries from different international nurseries were evaluated under the artificial infection of stem rust both in the field and in the greenhouse conditions. Experimental material has been received from Lomouri Farming Institute of Georgia. The wheat germplasm was evaluated for reaction to mixture of four predominant stem rust races (PHCQF, CFHC, PHCMF,NHPGF) in Georgia. These races contained the virulence on Sr 5, Sr 6, Sr 7b, Sr 9e, Sr 9a, Sr 9g, Sr9d, Sr 17, Sr 30, Sr 38, Sr McNair and SrTmP. The races structure of the stem rust population was determined according to the identification system (Roelfs and Martens, 1988) based on inoculation of isogenic Sr-lines with *Puccinia graminis* spores. The spores of prevalent stem rust races identified from Georgia regions were multiplied on cultivar Morocco and collected in separate test tubes to inoculate wheat cultivars.

Seedlings consisted of eight to ten plants per wheat entries were tested under the greenhouse conditions. Seven-day-old seedlings were inoculated in the second leaf stage with the water-spores mixture (approximately 3-5mg of freshly collected spores per 1ml of distilled water suspension) and incubated during 24 hours in a dew chamber in dark condition at 20-220C and 100% humidity. After that plant were transferred to a glasshouse where the temperature varied between 22-250C and relative humidity 60-70%. For each seedling, infection types (IT) were recorded 12-14 days after inoculation, based on a 0-4 scale of Stakman et al, (1962). ITs "0" to 3- were regarded as low IT and ITs "3" and "4" as a high IT.

Field trails were established on autumn (25 October) 2012. Each entry was sown in three 1-m long rows spaced 15 cm apart. Cultivar Bezostaya 1 was used as susceptible check. In early May, 2013 the plants were inoculated at the flag leaf stage with the mixture of same races of stem rust by spraying the spore-water suspension. Inoculated plants were covered by polyethylene film for a moist chamber. Data of infection types were recorded 12-14 days after inoculation according to the host response (Roelfs et al, 1992). Two types of scoring were combined: a) the host response to infection in the field was scored using 'R' to indicate resistance or miniature uredinia; 'MR' to indicate moderate resistance, expressed as small uredinia; "MS' to indicate moderate susceptible, expressed as moderate size uredinia somewhat smaller than the fully compatible type, and "S' to indicate full susceptibility. b) The modified Cobb's scale (Peterson et al, 1948) was used to determine the percentage of possible tissue (100%) rusted. The disease scorings were performed with the appearing of the first symptoms with ten day intervals three times.

Results

The responses of fifty wheat accessions including wheat species, domestic landraces and introduced entries from different international nurseries to stem rust races at seedlings and adult plant stages are presented in Tables 1 and 2. High and moderate level (R, MR) of juvenile and adult resistance to the disease was detected in nearly all tested species: *Triticum monoccoccum* (var.*laetissimum* Korn), *Triticum timopheevi* (var. *tipicum* Zhuk-var.*viticulosum* Zhuk), *Triticum georgicum* (var.*chvamlicum* Supat), *Triticum dicoccum* (var.*farrum*), *Triticum carthlicum* Men (var. *fuliginosum* Zhuk), *Triticum carthlicum* Men (var. *stramineum* Zhuk), *Triticum macha* Dek et Men (var. *megrelicum* Men), *Triticum macha* Dek et Men (var. *colchicum*), *Triticum macha* Dek et Men (var. *palaeo-imereticum*) and *Triticum spelta* (var. *dekaprelevichi Dorof*.) Exception was *Triticum durum* Desf. and *Triticum compactum* Host (var. *icterinum*) which showed the susceptibility at seedling stage. Only two old domestic varieties (Khulugo and Tetri ipkli) out of nine under study were moderate resistant to stem rust in both stages. cv. Vardzia showed MR reaction in seedling stage but it was susceptible in adult stage.

Effective adult resistance was found in seven varieties: Sauli 9, Lomtagora 123, Lomtagora 126, Lomtagora 109, Lomtagora 107, Lomtagora 149, Lomtagora 155, which were selected from international nurseries developed by ICARDA and CIMMYT during years and accepted for release in Georgia. These introduced entries were highly or moderately resistant at seedling stage too. Turkish variety Somnez was moderate resistant in seedling stage but in adult stage it showed susceptibility.

A consistent resistant reaction both in the seedling and adult plant stages was confirmed in six introduced accessions out of twenty: Haurani/aegtaushi/cham6-6/mz/cno67/3Ifn/4/ant/5/Attila-19FAWWON, Dorade/altay2000/4Bez/Nad//LZM-3/F900k-alres5, SRMA/tui//babax/3JGR-11LR-Res-132, DBDI-2WWSRRN-17, Sunco/pastor-19FAWWON, BTZ-18FAWWON-IRR-149. All tested entries were resistant in seedling stage but twelve introduced entries showed susceptible reaction in the field.

Discussion

The Georgian wheat landraces have been widely used in breeding of wheat as they represent rich sources of genes conferring resistance to diseases. Over the years, breeding for rust resistance has been based on Georgian endemic wheat species: *Triticum timopheevi, Triticum zhukovski, Triticum carthlicum* in the world. Much experience has been gained using wild wheat relatives for identification of new resistance sources (Tyryshkin et al, 2011; Knott and Zang, 1990; McIntosh and Gyarfas, 1971; Dekaprelevich, 1961) For instance, *T. timopheevi* and *T. monococcum* are known as valuable sources of resistance to the main fungal diseases, which have been incorporated into some improved varieties (Tomerlin et al, 1984, Brown-Guedira et al, 1996; Beteselassie et al. 2006). *Triticum carthlicum* was found to have also resistance to leaf and stripe rusts (Dekaprelevitch and Naskidashvili, 1976).

The results of our research showed that a majority of tested accessions had high and moderate resistance to Georgian population of stem rust where effective resistance genes were the followings: Sr11, Sr21, Sr24, Sr36, SrTmp and Sr31. The results of this study also support this fact that the wild relatives could be valuable sources of resistance to the stem rust races in the area to this day. This research results could be useful for the national and inter breeding programs in either further evaluation the stem rust resistant lines for varietal identification or using them as parents in the crossing. The many Georgian varieties (Vardzia, Bagrationi, Deda, Mukhrani, Motsinave) were developed from local landraces: Dika, Khulugo, Dolis puri (Naskidashvili et al, 1983; Naskidashvili et al, 1993).

Over last 15 years new wheat cultivars were introduced into the country via different ways including genotypes developed by international breeding programs. In this case, it is necessary to evaluate them to the existing stem rust races. To improve productivity of winter wheat in the development countries, the ICARDA and CIMMYT in collaboration with national partners have been working in framework of International Winter Wheat Improvement Program. In accordance with the results obtained from present study seven already released new varieties selected from international nurseries showed high and moderate level of resistance to prevalent Georgian races of stem rust. Also, eight advanced lines with adult resistant were identified.

As resistance genes Sr11, Sr21, Sr24, Sr36, SrTmp and Sr31 are mainly effective in Georgian population of stem rust, it is possible that these genes could be components of stem rust resistance in the tested entries.

Thus, presented research allowed the identification of stem rust resistant germplasm of wheat. These effective rust resistance sources could be widely used for breeding programs for producing new resistant cultivars all over the world.

Table 1. Infection types and field responses of wheat species and old Georgian varieties to stem rust

#	Accessions	Origin/Coutry/	Seedling Infection Types	Adult plant Reaction Types
1	*Tr.monococcum* L. var. *laetissimum* Korn	GEO	1	R
2	*Tr. timopfeevii* Zhuk. var. *tipicum* Zhuk	GEO	1.2	R
3	*Tr. georgicum* Dekap. var. *chvamlicum* Supat.	GEO	2	R
4	*Tr. Dicoccum* var. *farrum*	GEO	1,2	R
5	*Tr.ibericum* Men. var. *fuliginosum* Zhuk.	GEO	2+	R
6	*Tr.ibericum* Men. var. *stramineum* zhuk.	GEO	0-1	MR
7	*Tr.macha* Dek et Men. var. *colchicum*	GEO	2+	MR
8	*Tr.macha* Dek et Men. var. *megrelicum* Men	GEO	2;	MR
9	*Tr.macha* Dek et Men. var. *palaeo-imereticum*	GEO	2	MR20
10	*Tr. spelta* L. var. dekaprelevichii Dorof	GEO	2	MR
11	*Tr.compactum* Host var. *icterinum* Al.	GEO	3	MS
12	*Tr.durum* Desf. (Tavtukhi)	GEO	2+	MS
13	Tbilisuri 5	*T.aestivum* var. *aestivum*	3	80MS
14	Dolis puri 35-4	*T.aestivum* var. *aestivum*	3	80MS
15	Khulugo	*Tr. aestivum* var. *lutescens*	1/2	R-MR
16	Lagodekhis grzeltavtava	*Tr. aestivum* var. *lutescens*	3	MS-MR30
17	Korboulis dolis puri	*T.aestivum* var. *aestivum*	3	80MS
18	Akhaltsikis tsiteli doli	*T.aestivum* var. *ferrugineum*	3	80MS
19	Tetri ipkli	*Triticum aestivum* L.	2+	60MR
20	Vardzia	*T.aestivum* var. *Ferrugineum*	2	50MR
21	Almasi	*T.aestivum* var. *aestivum*	3	80MS
22	Bezostaya 1(susceptible check)		3	90MS

Table 2. Infection types ad field responses of introduced accessions of wheat to stem rust

#	Accessions	Origin	Seedling Infection Types	Adult plant Reaction Types
1	Sauli 9 (*Tr. aestivum*)		2	60MR
2	Lomtagora 123 (*Tr. aestivum*)		1.2	20MR
3	Lomtagora 109 (*Tr. aestivum*)		2	40MR
4	Lomtagora 149 (*Tr. aestivum*)		1,2	20MR
5	Lomtagora 126 (*Tr. aestivum* var. *lutescens*)	Pehlivan/Jagger	2+	20MR
6	Lomtagora 155 (*Tr. aestivum*)	*BEZ/SDV1/5/338-K1-1// TJB368.251*	0-1	30MR
7	Lomtagora 107(*Tr. aestivum*)	CUPRA-1/3/CROC1/AE SQUARROSA	2+	50MR
8	Somnez (*Tr.aestivum* var. *lutescence*)	Turkey	3	80MS
9	Lukillus- *Tr. aestivum*	18FAWWON-IRR-53	2; 2+	50MR
10	BTZ- *Tr. aestivum* var. *lutescens*1	8FAWWON-IRR-149	1,2	5MR
11	Unknown pedigree-2- *Tr. aestivum*	18FAWWON-SA-49	2	60MR
12	Stetatus- *Tr. aestivum*	2WWSRRN – 17	2+	40MS
13	DBDI- *Tr. aestivum*	2wWWSRRN-34	2+	5MR
14	Attila*2/pastor//Orkinos-2-11lr-res-3	11LR-RES-3	2+	70MS
15	Dorade/altay2000/4/Bez/Nad// LZM(es85.24)3/ F900k –aalr-res-5	11LR-RES-5	2	10MR
16	Haurani/aeg taushi/cham6/6/mz/ / cno67/3/lfn/4/ant/5/Attila-	19FAWWON	1/2	20MR
17	Sunco/pastor-	19FAWWON	0	40MR
18	Kupava/burbot-6//Jagger-	11LR-RES-18	2,2+	80MS
19	Demir-	11lr-RES-125	2+	60MS
20	SRMA/tui//babax/3JGR-	11LR-Res-132	1,2-	20MR
21	Mina/4/pmf/maya/yaco/3/ CO693591/CTK-	11LR-Res-207	2+	80S-MS
22	Saulesku 44/tr510222	8EYT-SA -9908	2+	80MS
23	#293	*Tr. aestivum* var. *erythrospermum* Korn.)		80S
24	#2216	*Tr. aestivum* var. *lutescens*	2+	60S
25	#288	*Tr. aestivum*	2+	70S
26	#211	*Tr. aestivum* var. *lutescens*	2	40MS
27	#302	*Tr. aestivum*	2	20MS
28	#202	*Tr. aestivum* var . *ferrugineum*	2	20MS
	Bezostaya 1		3+	90S-MS

References

Anonymous, (2008). Wheat Rust Disease Global Programme. Food and Agriculture Organization of the United Nations. Rome, 45 p.

Bariana H S (2008). Stem rust resistance in wheat. Proceedings of the International Conference on wheat stem rust Ug99. pp.20-24

Beteselassie N, Fininsa C and Badebo A. (2006). Sources of stem rust resistance in Ethiopian tetraploid wheat accessions. African Crop Science Journal, Vol. 15, No. 1, pp. 51 -57

Brown-Guerda G L, Jil B S, Bockus W W, Cox T S Hatchett J H, Leath S, Peterson C J, Thomas J B, Zwer P K (1996). Evaluation of a collection of wild timopheevi wheat for resistance to disease and arthropod pests. Plant diseases 80 :928-933

Dekaprelevich L L (1961). Die Art Triticum macha Dek et Men. im Lichte neuster Untersuchungen uber die Herkunft der Hexaploiden Weizen. Z. Pflanzenzuchtg. 45:17-30.

Dekaprelevich L L and Naskidashvili P P (1976). Triticum persicum v. stramineum, genetic source of resistance to yellow and brown rust [Georgian]. Soobsccheniya Akademii Nauk Gruzinskoi Ssr. 82:689-691.

Knott D R and Zang H T (1990). Leaf rust resistance in durum wheat and its relatives. In: Wheat Genetic Resources: Meeting Diverse Needs, Srivastava, J.P. and Damania, A.B. (eds). John Wileyand Sons, Chichester, UK, pp. 311-316.

McIntosh R A and Guarfas J (1971). Triticun timopheevi as a source of resistance to wheat stem rust. Z.Pfazenzucht, 66:240-248

Menabde V L (1948). Wheats of Georgia. Georgian Academy of Sciences Press, Tbilisi, 267p.

Naskidashvili P, Sikharulidze M, Chernish E (1983). Breeding of wheat in Georgia. Tbilisi,

Naskidashvili P, Iashagashvili G, Kevkhishvili V (1993). Valuable contribution of Georgian endemics and landraces in the breeding.(In Georgian). Tbilisi, Georgia, 40p.

Peterson R F, Campbell A B, and Hannah A E (1948). A diagrammatic scale for estimating rust intensity on leaves and stems of cereals. Canadian Journal of Research 26: 496-500.

Pretorius Z A, Singh R P, Wagoire W and Peyne T (2000). Detection of virulence to wheat stem rust resistance gene Sr31in Puccinia graminis in Uganda. Plant Disease, 84:203

Roelfs A P and Martens J W (1988). An international system of nomenclature for Puccinia graminis f.sp. tritici. Phytopathology, 78:526-533.

Roelfs A P, Singh R P, Saari E E (1992). Rust Diseases of Wheat. Concepts and methods of Disease Management, Mexico, P.F.CIMMYT, pp.81.

Sikharulidze Z, Natsarishvili K, Mgeladze L and R Dumbadze (2013). Wheat stem rust research in Georgia. BGRI Technical Workshop, 8 - 21, August, 2013, India, New Dehli, Abstract book, p.94

Stakman E C, Stewart D M and Loeggering W Q (1962). Identification of physiologica races of Puccinia graminis var. tritici. US.Dept. Agri. ASR E617: 53pp.

Tyryshkin L G, Klesova M A, Kovaleva M A, Lebedeva T V, Zuev E V, Brykova N, Gashimov M E (2011). Current status of bread wheat and its relatives from VIR collection study for effective resistance to fungal diseases Proceedings of 8th wheat conference, 1-4 June. Petersburg, Russia

Tomerlin J R, El-Morshidy M A, Moseman J G, Baenziger P S and Kimber G (1984). Resistance to Erysiphe graminis f. sp. tritici, Puccinia recondita f. sp. tritici, and Septoria nodorum in wild Triticum species . Plant Dis. 68:10-13.

Wanuera R (2008). Status and impact of Ug99 in Kenya. Procc. International Conference on wheat stem rust Ug99. p 9-11

Research Article

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Comparison of chemical composition of two durum wheat (*Triticum durum* L.) and bread wheat (*Triticum aestivum* L.) germ oils.

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ABSTRACT

Wheat belong to the genus Triticum, as annual plants of the family Gramineae or Poaceae, grown in many countries, including Tunisia with one million hectares annually. Wheat grain is a particular fruit, caryopsis and the outer envelope is adherent to plant seed material. During milling, envelopes (hulls) are separated from the grain (endosperm + embryo). The embryo or germ is the essential part of the seed to plant reproduction and is containing a lot of fat (about 15%) or oils. The Soxhlet technique is used for the extraction of wheat germ oil. Normal hexane (n-hexane) is commonly used for edible oil extraction. Comparison of the extracted oil of durum wheat germ and soft wheat germ showed a marked difference in their chemical composition. The basic chemical composition analyses revealed low values of dry matter (14.77g /100g of durum wheat germ and 19.87g /100g of Soft wheat germ), low amounts of total ash content (5.3g/100g of durum wheat germ and 4.99g /100g of Soft wheat germ) and high fat contents (17.12g /100g of durum wheat germ and 15.96g /100g of Soft wheat germ). The yield of extraction by Soxhlet was about 13.12% for durum wheat germ and 11.22% for soft wheat germ. The fatty acid composition of these two wheat germ oils indicates the presence of C18:2, C16:0 and C18:1. The major one is C18:2 with 56.68% for Soft wheat germ oil and 53.43% for durum wheat germ oil.

***Keywords*:** germ oil, durum & soft wheats, soxhlet extraction, fatty acid composition, chemical composition

Introduction

Cereals have an important role in human nutrition, either for cooking or as raw material for obtaining flour for baking. Botanically they belong to the grass family (Gramineae), that include wheat, rice, barley, oats, rye, maize, sorghum, and millets (Belderok,2000). Wheat is one of the most important crops in the world with the largest production of any crop. This is because it is highly adaptative to environmental conditions and because of its unique characteristics where it can be processed into various types of edible products (Shewry and Tathmn, 1997). The different attributes of Triticum durum and Triticum aestivum are due to the differences in kernel physiochemical properties. Durum wheat grains are harder, larger and more vitreous than bread wheat grains. Durum wheat is tetraploid (AABB), While bread wheat is hexaploid (AABBDD), and consequently, the absence of D genome is to some extent- responsible for the

reduction in durum wheat baking performance (Kerber and Tipples 1969; Ceoloni et al, 1996; Pogna et al, 1996; Redaelli et al, 1997; Joppa et al, 1998; Lafiandra et al, 2000). The main components of the wheat kernel are barn, germ and endosperm (Dexter and Wood, 1996). Wheat germ is a by-product of the wheat milling industry. Germ constitutes about 2–3% of the wheat grain and can be separated in a fairly pure form from the grain during the milling process. Wheat germ contains about 11% oil (Sonntag, 1979). α-Tocopherol, polyunsaturated lipids, protein, threonine, methionine, lysine, raffinose, sucrose, thiamin and riboflavin were chosen since these are the components regarded as most important in commercial wheat germ. Compared to wheat, Tunisian barley varieties have also a high level of antioxidant capacity compared to that of other cereal (wheat bran 0.042 lmol of trolox equivalent/g of wheat bran) based on TEAC assay.

Solvent extraction is a common method of extraction of oils from vegetable matter. Normal hexane (n-hexane) is commonly used for edible oil extraction. Wheat germ oil is used in products such as foods, biological insect control agents, pharmaceuticals and cosmetic formulations (Kahlon, 1989). This valuable product is not only used in the food industry as various food additives, but also in various areas of medicine for the treatment of many diseases. Wheat germ oil is different from many vegetable oils. A distinctive feature of the wheat germ oil compared to most vegetable oils is its high content of "vitamin of youth" E (tocopherol) and polyunsaturated fatty acids (from 45 to 60%), linolenic (up to 11%) and oleic acid (12 to 30%). Both of which are of great importance in human metabolism and cannot be synthesized by the organism. Furthermore, linoleic acid helps to eliminate cholesterol and is a precursor of cell membrane phospholipids (Salinas R, 1993). Also in wheat germ oil in much smaller amounts are saturated fatty acids (14 to 17% palmitic, stearic 0.5 to 2.3%, and so one) (Nechaev A.P, 1975).

This work aim to study the chemical composition of two wheat germ oils, durum and bread wheat produced in Tunisia.

Materials and methods

Wheat germ material

Bread and Durum wheat germ samples, by-products used in the present study, were generated by one of the Tunisian milling [GMT], located in Tunis. Germ was obtained from milling of bread and durum wheat. Wheat germs were directly stored in a freezer at (-18 °C) until extraction and analysis.

Separation and clearing of wheat germ

Wheat germ contains significant amounts of organic impurities not oiled: mealy particulate grain (up to 4%), husks (5-6%), etc. The presence of impurities in processed embryos increases the losses of oil in cake or meal. Purification of wheat germ from husks and impurities was done by sifting wheat germ with a double sieve comprises a sieve of the order of 1000 µm and a sieve of the order of 630 µm. The sieving time was 3 to 4 minutes.

Oil extraction

Wheat germ oil extraction was carried out according to the AOCS Official methods using n-hexane (AOCS, 1998). Oil content of wheat germ samples was determined by using a Soxhlet apparatus with n-hexane as a solvent for 6 h. Solvent used for oil extraction is n-hexane, with a highest available purity. The n-hexane solvent was done from SIGMA - ALDRICH Company (USA). All other chemicals used in this study were of analytical grade.

The extraction procedure was repeated twice and the solvent was evaporated from the extract solvent mixtures at 40°C under vacuum using a rotavapor (Rotavapor R-210/215, 230V, 50/60 Hz) until constant weight was attained. The wheat germ oils obtained was drained under a nitrogen stream (N2) and was then stored in a freezer at (-18 °C) until analysis. The amount of oil extracted by solvent was gravimetrically determined. Extraction yield (Y) was defined by the following equation:

$$\mathbf{Y}\ (\%) = \frac{\text{amount of extract collected (g)}}{\text{amount of sample used for extraction (g)}} * 100$$

Analytical methods

Fatty acid composition

Fatty acid composition of the extracted oil was analyzed by gas chromatography (GC-SM) (Agilent 19091S-433). The GC unit was a HP-5MS equipped with a flame ionization detector (FID) and a polar phénylméthyl-siloxane capillary column (60m×25mm×0.25µm film thickness), was used for fatty acid analysis. Methylation of the fatty acids was carried out according to the AOCS Official Method Ce 2-66 (AOCS, 1998). The helium carrier gas flow rate was 1ml/min. The injector temperature was maintained at 230°C. A temperature program with total run time of 82 min was used. The column temperature, after an initial isothermal period of 2 min at 50°C, was increased to 220°C at a rate of 4°C/min, and maintained at this temperature for 37.5 min. The detector conditions were as follows: temperature 250°C, N2 flow 40 ml/min, air flow 450 ml/min and make-up gas (He) 45 ml/min. Germ

oil samples (1 ml) were injected by an auto sampler (HP-5MS, HP Company, Wilmington, DE). Peak areas were calculated and data collection was managed using an HP Chemstation.

Organic matter and ash contents quantification

Wheat germ samples (2.5g) induplicate for each sample were analyzed for residual water content and ash content using previously validated method (De Vasconcelos et al, 2009). Wheat germ samples were submitted to a drying processing in an oven at 105◦C for 12 h, and then the samples were weighed. Dried samples are incinerated at 550◦C for 3 h, and the ash content was obtained.

Chemical analysis

Official methods of American oil chemist's society (AOCS, 1998) were used for the determination of the refractive index, density, acid value and iodine value of the wheat germ oils. The antioxidant activity of the wheat germ oils was determined by a β-carotene/ linoleic acid system, as described by Matthus (2002).

Results and discussion

Chemical composition of wheat germs

Modern technologies of grain into flour can get the germ of up to 10-35% cuts that affect the composition and biological value of the finished product (Butkovsky et al, 2006). The extraction rate of oil through soxhlet was found 13.12% for durum wheat germ and 11.22% for bread wheat germ, which was in close agreement with the results found in the study of Dunford and Zhang (2003). Variation in oil yield could be attributed to differences in plant variety, cultivation climate, ripening stage and the extraction method used (Nyam et al, 2009). As illustrated in Table 1, the nutritional and biological value of durum and bread wheat germ were studied and found that the total ash, moisture, fat and protein levels were 4.99%, 19.87%, 15.96 % and 23.3% for bread wheat while they were 5.3%, 14.77%, 17.12% and 25.3% for durum wheat, respectively. Thus the difference in chemical composition of the two wheat germs depends significantly on the genetic characteristics of raw materials, climate, cultivation of grain, as well as its productivity. Such a chemical composition reveals the valuable potencies of such a wheat germ.

Chemical analysis of wheat germ oils

The results of physico-chemical parameters in Table 2 indicate that the characteristics of extracted wheat germ oils are in agreement with recent published values for these indices. The density or specific gravity of oil at any given temperature compared to water at a specified temperature is known to increase as the degree of unsaturation increases (i.e. with higher iodine value) (Muhammad, 2008). The densities values (g/ml) found for extracted durum wheat germ oil and bread wheat germ are 0.92 and 0.86. However, the iodine value for durum wheat germ oil was found to be 192.6 and 191.4 for bread wheat germ, which is also within the limits of literature (O'Brien, 2004 and Przybylski, 2004). It can be seen also that the acid values were 91.5 (g/100g) for bread wheat germ oil and 89.5(g/100g) for durum wheat germ oil. The antioxidant activity of wheat germ oils was measured by the bleaching of ß-carotene. The comparison of durum wheat germ oil and bread wheat germ oil showed an appreciable antioxidant activity. In addition, studying the data in table 2 shows that beta-carotene content from durum wheat germ oil (17.12 mg/g) is greater than bread wheat germ oil (15.96 mg/g). It is indicated by the results of this work that wheat germ oils were established, could serve as a source of natural antioxidants or nutraceuticals.

Fatty acid composition

Fatty acids contents of wheat germ oils were obtained by GC analyses. Table 3 shows the fatty acids composition, expressed as percentage of total fatty acids, of oil samples obtained by using organic solvents and Soxhlet extraction. Total unsaturated and polyunsaturated fatty acid content of bread wheat germ oil was about 81.64 and 18.51%, respectively. While for durum wheat germ oil it was about 82.08% and 17.91% of total unsaturated and polyunsaturated fatty acid content. The most abundant saturated fatty acid was palmitic acid with more than 94% of the saturated fatty acids for bread wheat germ oil and 94.6% for durum wheat germ oil. These results are in a good agreement with the previous results reported by Lancas et al, 1994, Michael et al 2006, Michael and Nurhan 2007 and Yuldasheva et al. 2010. Wheat germ n-hexane extracts consisted of about 69% linoleic acid (18:2 n6) for bread wheat germ oil and 53.9% for durum wheat germ oil, which is an essential fatty acid. It is worth mentioning that the high amount of linoleic acid makes wheat germ oil specifically prone to oxidation and degradation under the conditions used for conventional edible oil extraction and refining methods (Krings et al, 2000). It has been also suggested that unsaturated fatty acid, especially polyunsaturated fatty acid intake reduces cardiovascular heart disease (CHD) (Simopoulos, 1999). Also this fatty acid may have favorable nutritional implications and beneficial physiological effects in the prevention of cancer (Oomah et al, 2000).

Conclusion

The current study has revealed that the used technology for the production of wheat germ oil, as confirmed by the literature, showed a good composition and extraction yield. The Tunisian wheat germ is not used for human feeds, which allows us to think about industrial applications. The quantity of triturated wheat in Tunisia is about 9000 tons per day resulting in 6 tons of wheat germ, and the amount of oil that can be obtained is approximately 650 liters per day. Thus the present study can serve as an opportunity for the industrial to invest in this field. Yield and quality improvement of wheat germ oils needs other process as cold pressing and supercritical CO_2 extraction. The production of oil from wheat germ provides the use of renewable resource, and at the same time adding value to food products. Breeding for improved genetic material within quality parameters required somewhere phenotyping toward fonctionnal foods. Thus, incorporation of such materials into bakery products would enhance their nutritional and physiological properties, but their functionality and acceptability should be taken into consideration.

Table 1. Chemical composition (dry basis) of durum and bread wheat erms.

Component	Bread wheat germ oil	Durum wheat germ oil
Moisture content [a]	19.87	14.77
Crude oil [a]	15.96	17.12
Crude protein [a]	23.8	25.3
Total ash [a]	4.99	5.3

(w /w)[a]

Table 2. Physicochemical characterization of durum and bread wheat germ oils.

Parameter	Bread wheat germ oil	Durum wheat germ oil
Refractive index	1.48	1.46
Density (g/ml)	0.86	0.92
Acid value (g/100g)	91.5	89.5
Iodine value	191.4	192.6
β-carotene (mg/g)	15.96	17.12

Values are means of three determinations. (w /w)

Table 3. Fatty acids composition (expressed as % of total fatty acids), measured by a gas chromatography-flame ionization detection (GC-FID) method, of durum and bread wheat germ oils.

Fatty acids (%)	Bread wheat germ oil	Durum wheat germ oil
Myristic acid C14:0	0.186	0.12
Palmitic acid C16:0	17.47	16.95
Palmitoleic acid C16 :1	0.18	0.26
Stearic acid C18:0	0.64	0.71
Oleic acid C18 :1	15.24	20.47
Linoleic acid C18 :2	56.68	53.43
Linolenic acid C18 :3	8.11	6.67
Arachidic acid C20:0	0.22	0.13
Gadoleic acid C20 :1	1.25	1.25

References

AOCS (1998). The Official and Tentative Methods of the American Oil Chemist's Society, 3rd Ed. American Oil Chemist's Society, 508 South Sixth Street, Champaign, Illinois.

Belderok B, Mesdag H and Donner DA (2000). Bread-Making Quality of Wheat. Springer, New York.

Butkovsky VA, Galkin GE, Ptushkina HP (2006). Modern techniques and technology of flour / M. Delhi print. – p. 319.

Ceoloni C, Biaggestti M, Ciuffi M, Forte P and Pasquini M (1996). Wheat chromosome engineering at the 4 x leve. The potential of different alien gene transfers into durum wheat. Euphytica, 89,87-97.

Dexter J E and Wood PJ (1996). Recent applications of debranning of wheat before milling. Trends Food Sci. Technol, 7, 35-41.

De Vasconcelos,R Bennetta Castroc, P Cardosoe , M.J Saavedrab and E A Rosaa (2009). Study of composition, stabilization and processing of wheat germ and maize industrial by-products M.C.B.M.

Dunford N T and Zhang M (2003). Pressurized solvent extraction of wheat germ oils. Food Research International, 36, 905– 909.

Joppa L R, Klindworth D L and Hareland G A (1998) Transfer of high molecular weight gluteninsfrom spring wheat to durum wheat. Slinkard, A. E, Ed. Proceedings of the Ninth International Wheat genetics Symposium, Saskatoon, SK, Canada. University of Saskatchewan E xtension Press.2 57-260.

Kahlon T S (1989). Nutritional implications and uses of wheat and oat kernel oil. Cereal Food Worls.34, 872-875.

Kerber E R and Tipples K H (1969). Effects of the D genome on milling and baking properties of wheat. Canadian Journal of Plant Science, 49,255-263.

Krings U El-Saharty, Y S El-Zeany, B A Pabel and B Berger R G (2000). Antioxidant activity of extracts from roasted wheat germ. Food Chem. 71, 91-95.

Lancas FM, M E C Queiroz and I C E da Silva (1994). Seed oil extraction with supercritical carbon dioxide modified with pentane. J. Chromatographia, 39:687-692.

Laflandra D, Margiotta B, Colaprico G, Masci S, Roth M R and Mac Ritchie F (2000). Introduction of the D-genome related high and low molecular glutenin subunits into durum wheat and their effect on technological properties. Shewry P R and Tatham A S, Eds. Gluten Proteins 2000. Proceedings of the Seventh International Gluten Workshop, Cambridge, UK. Royal Society of Chemistry. 51-54.

Michael Eisenmenger, Nurhan T Dunford, Fred Eller, Scott Taylor and Jose Martinez (2006). Pilot-scale supercritical carbon dioxide extraction and fractionation of wheat germ oil. J. Am Oil Chem. Soc. 83:863-868.

Michael Eisemmenger and Nurhan Turgut Dunford (2007). Bioactive components of commercial and supercritical carbon dioxide processed wheat germ oil. J. Am Oil Chem. Soc. 85:55-61.

Matthus B (2002). Antioxidant activity of extracts obtained from residues of different oilseeds. J. Agric. Food. Chem,50, pp 3444-3452

Muhammad Umair Arshad, Surayia Zakir, Faqir Muhammad Anjum, Tahir Zahoor and Haq Nawaz (2008). Nutritive Value of Cookies Containing Wheat Germ Oil, Pakistan Journal of Life and Social SciencesPak. j. life soc. sci. , 6(2): 127-134.

Nechaev AP and Sandler Z Y(1975). Lipids grain. - M.: Kolos, – P. 157.

Nyam K L,Tan C P, Lai O M, Long K and Che Man Y B (2009) Physicochemical properties and bioactive compounds of selected seed oils. Food Sci. Technol. 42, 1396-1403.

O'Brien R D (2004). Fats and Oils. Formulating and Processing for Applications (2nd edition), CRC Press, Washington, DC.

Oomah D B, Ladet S, Godfrey D V, Liang J and Girard B (2000). Characteristics of raspberry (Rubus idaeus L) seed oil. Food Chem. 69,187-193.

Pogna N E, Mazza L, Redaelli R and Ng P K W (1996). Gluten quality and storage protein composition of durum wheat lines containing Gil-DY Glu-D3 loci. Wrigley, C.W, Ed. Proceedings of the 46th Australian Cereal Chemistry Conference in Association with the Sixth International Gluten Workshop, Melbourne, Australia. Royal Australian Chemical Institute. 18-22.

Przybylski R (2004). Canola oil: physical and chemical properties. http://www.canola-council.org, accessed on: April 23, 2007.

Redaelli R, Pogna N E and Ng P K W (1997). Effects of prolamins encoded by chromosomes IB and ID on the rheological properties of dough in near-isogenic lines of bread wheat. Cereal Chemistry, 74,102-107.

Salinas R (1993). Alimentos Grasos y Nutrición, Bromatología Aplicada a la Salud; El Ateneo: Buenos Aires, Argentina, 1993; Chapter 7.

Shewry P R and Tatham A S (1997). Biotechnology of wheat quality. Journal of the Science of Food and Agriculture, 73,397-406.

Simopoulos A P (1999) Essential fatty acids in health and chronic disease. American journal of Clinical Nutrition,70(3), 560–569.

Sonntage NOV (1979). Composition and characteristics of individual fats and oils, in Bailey's Industrial Oil and Fat Products, Vol. 1, 4th edn, edited by D. Swern, John Wiley & Sons, New York, pp. 289-477.

Yuldasheva NK, Ul'chenko and Glushenkova A I (2010). Wheat germ oil. J.Chemistry of Nautral Compounds, 46:97-98.

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Variety of Bread Wheat "Aldane"

"Aldane" is a winter bread wheat (*Triticum aestivum* L.) variety developed by Trakya Agricultural Research Institute (TARI) and registered in 2009. Aldane cross is Bul2477-2/3/093-44/Au//Bez with TE5255-2T-1T-1T-0T pedigree. Crossing was made in 1996 and yield test began in 2001-2002 growing year.

The spike of the Aldane cultivar is moderately long, white, smooth, awnless and compact. It resembles cultivar Pehlivan. The flag leaf is twisted, light-green, and with low glaucousity. Grain is long-oval, hard and red colour and with 37.6 g Thousand Kernel Weight. Aldane is a medium-tall cultivar, similar to Pehlivan. Plant height is between 95 and 105 cm depending on the growing conditions. It is medium early and as it has good adaptation ability, it has been grown throughout Trakya-Marmara region and some other parts of Turkey. It gives high yield both on fertile and less fertile soils. It has resistance to winterkilling and is tolerant to medium drought conditions. Aldane is tolerant to powdery mildew (*Erysiphe graminis* f. sp. *tritici*) and stripe rust (*Puccinia striiformis* f. sp. *tritici*). It is resistant to leaf rust (*Puccinia recondita*). It is susceptible to root rot diseases; if it is grown on flat area lodging could be problem.

Yield test of the Aldane was done for 12 years in Trakya region in Turkey. Yield potential is medium high however, high yield can be obtained if environmental conditions are favorable and applied good agronomic practices. The highest grain yield obtained was 9220 kg ha-1 and mean yield of last 12 years (2003 and 2014) was 6652 kg ha-1 in Trakya growing conditions. The highest yield (9220 kg ha^{-1}) was obtained in 2004-2005 growing season in Tekirdag location. Suggested planting rate is between 450-500 seeds/m^2.

Grain quality is excellent. The mean values of some bread making qualities of the last 5 years (2009 and 2013) are; test weight 81.6 kg, protein content 13.5 %, gluten value 38.2%, gluten index 89.8 % and sedimentation 59.5 ml. The highest quality values in last 5 growing seasons were; test weight 84.1 kg, protein content 15.6 %, gluten value 46.0 %, gluten index 96.5 % and sedimentation 69.8 ml. Some other bread wheat quality characters are; absorption (%) 60-65, and energy value (W) 250-280.

Pre-Basic and Basic seeds of the Aldane cultivar have been produced by Trakya Agricultural Research Institute (TARI). Certified seed of the Aldane are produced by both private companies and state farms.

Picture 1. Spike and grain of the Aldane cultivar

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References

Anonymous (2010). Cool season cereals variety registration reports. Variety Registration and Seed Certification Center, Ankara, Turkey.

Öztürk I and Korkut KZ (2011). Characterization of Drought Resistance and its Relations with Quality in Bread Wheat (Triticum Aestivum L.) Genotypes. Namık Kemal Uni. 2011. (Ph. D. Thesis)

Öztürk I, Babaoğlu M, Avcı R, and Tuna B (2012). Trakya Agricultural Research Institute Annually Reports, Edirne, 2012. Trakya Agr. Res. Inst., Edirne, Turkey.

Süzer S ve Öztürk, I (2011). Trakya Bölgesi'ne uygun buğday tarımı ve yeni çeşitlerinin özellikleri. Hasad. Aylık Tarım Dergisi. Sayı:310, S: 96-100. İstanbul.

Variety of Bread Wheat "Selimiye"

"Selimiye" is a winter bread wheat (*Triticum aestivum* L.) variety developed by Trakya Agricultural Research Institute (TARI) and registered in 2009. Selimiye cross is Lau/Agd/3/Odes95//Olv/B16 and selection history is TE5402-4T-1T-2T-0T. Cross was made in 1997-1998 and yield testing began in 2003-2004 growing year.

The spike of the Selimiye cultivar is moderately long, red colour, smooth, awnless and compact. Spikelet and glume is very though. Appearance of the spike looks like Pehlivan and Aldane but colour is red. The flag leaf is twisted, dark-green, and with gloucoisity similar to Pehlivan. The grain is oval, hard, red colour and with 37.1g Thousand Kernel Weight. Selimiye is medium-tall cultivar with plant height 90 to 105 cm depending on the growing conditions. It has resistance to winterkilling, tolerant to medium drought condition and medium early. Selimiye has been grown throughout Trakya-Marmara region. It has high productive tillering capacity. It is suitable for growing on fertile and less fertile soils. Selimiye is susceptible to powdery mildew (*Erysiphe graminis* f. sp. *tritici*). It has

tolerance to yellow rust (*Puccinia striiformis* f. sp. *tritici*) and leaf rust (*Puccinia recondita*) and some root rot diseases. If the variety is grown on fertile and flat area in combination with the use of high seed rate and fertilizers, lodging could be problem.

Selimiye was tested for 11 years in Trakya region in Turkey. It has high yield potential. Average yield of the last 11 years between 2004 and 2014 growing year in Edirne condition was 7553 kg ha^{-1}. The highest yield with 9180 kg ha-1 was obtained in 2013-2014 growing season in Edirne location. Suggested planting rate is 450-500 seeds/m^2.

Selimiye has good bread making quality characteristics. Some of the quality mean value of the last 5 years (2009 and 2013) are; test weight 83.2 kg, grain protein content 12.4%, gluten value 37.2 %, gluten index 84.7% and sedimentation 51.6 ml. In the same period (2009-2013) the highest values were; test weight 85.9 kg, grain protein content 14.7%, gluten value 44.1%, gluten index 92.9% and sedimentation 68.8 ml.

Pre-Basic and Basic seeds of the Selimiye cultivar have been produced by Trakya Agricultural Research Institute (TARI). Certified seeds of Selimiye are produced by both private companies and farmers and state farms.

Picture 1. Spike and grain of the Selimiye cultivar

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References

Anonymous (2010). Cool season cereals variety registration reports. Variety Registration and Seed Certification Center, Ankara, Turkey.

Öztürk I and Korkut KZ (2011). Characterization of Drought Resistance and its Relations with Quality in Bread Wheat (Triticum Aestivum L.) Genotypes. Namık Kemal Uni. 2011. (Ph. D. Thesis)

Öztürk I, Babaoğlu M, Avcı R, and Tuna B (2012). Trakya Agricultural Research Institute Annually Reports, Edirne, 2012. Trakya Agr. Res. Inst., Edirne, Turkey.

Süzer S ve Öztürk, I (2011). Trakya Bölgesi'ne uygun buğday tarımı ve yeni çeşitlerinin özellikleri. Hasad. Aylık Tarım Dergisi. Sayı:310, S: 96-100. İstanbul.

Variety of Triticale "Ümranhanım"

"Ümranhanım" is a hard red winter triticale (x*Triticosecale* Witt.) cultivar developed by East Anatolia Agricultural Research Institute and registered in 2010 for its high level of winter hardiness. Under the controlled environment experiment for cold tolerance Ümranhanım was resistant to -17^{0}C. It has the highest survival rate among the tested genotypes.

Ümranhanım was selected from the advanced material provided by Bahri Dagdas International Agricultural Research Institute (BDIARI) in 2002. Ümranhanım was released primarily for its superior adaptation to dryland agriculture in East Anatolia. It is tall (120-130 cm) with good straw strength, high yielding, early and resistant to foliar and spike diseases. When it was tested for 4 years at 4 locations (Ilıca, Pasinler, Mus, Erzincan) in the eastern part of Turkey, yield of Ümranhanım cv (4862 kg/ha) was higher than check varieties Tatlıcak 97 (4672 kg/ha), MIKHAM (3825 kg/ha), Melez (3931 kg/ha) and Karma (4144 kg/ha). In addition, Ümranhanım has the best yield stability in the region compared to other varieties.

It has slightly colored spikes with awns. Spikes are long, and dense with brown glumes. Kernels are semi-hard and red in color. The seed is elongated and oval-shaped.

It has a winter growth habit with high-tillering capacity. It is mid-early maturing, drought and lodging resistant and its response to fertilizer is high. Threshing ability is good.

1000-kernel weight 22,4-45,0 g; the long-term range is 9,9-16,2% grain protein content; test weight is 71,3-76,6 kg/hl; digestible protein is 9,5-10,8; crude oil is 2,3-2,5%; crude cellulose is 1,8-2,0%; grain hardness is 57,3%. Ümranhanım is resistant to yellow rust (*Puccinia striiformis*), leaf rust (*Puccinia recondita*), stem rust (*Puccinia graminis*), powdery mildew (*Erysiphe graminis*), common bunt (*Tilletia foetida*) and smut (*Ustilago tritici*) in field conditions.

It has higher grain yield than barley and oats under dryland conditions in the region. It is recommended for dryland areas in Eastern Anatolia Region and the other cold areas. Its grain can be used both for human consumption as well as animal feed.

Certified seeds of Ümranhanım are produced by both private companies and state farms.

Figure 1. Spike and grain of the Ümranhanım cultivar

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References

Anonymous (2013). Registration Committee Report. Variety Registration and Seed Certification Center, Ankara, Turkey.

Anonymous (2013). Property Documents of Ümranhanım Variety. Variety Registration and Seed Certification Center, Ankara, Turkey.

Variety Registration and Seed Certification Center.

Genç I 1972. Research on yield and yield-related characteristics of domestic and foreign bread and durum wheat. Dissertation. Ankara University. Faculty of Agriculture.

Köycü C 1979. Research on yield, yield-related traits and some morphological characters of domestic and foreign winter bread wheat (Triticum aestivum L.). Atatürk University Faculty of Agriculture. Department of Field Crops. Erzurum.

Tıschner T, Köszegi B, Veizs O 1997. Climatic Programmes Used In The Mortanvasar Phytotron Most Frequently in Recent Years. Acta Agronomica Hungaria, 45(1):85-104.

Variety of Barley "Altıkat"

"Altıkat" is six rowed spring barley (Hordeum vulgare L.) cultivar developed by GAP International Agricultural Research and Training Center (GAPIARTC) and it was registered in 2011.

Altıkat was selected from the cross Arta/4/Arta/3/Hml-02//Esp/1808-4L, which was made by International Center for Agricultural Research in the Dry Areas (ICARDA). The selection history is ICB96-0601-0AP-10AP-0AP. It was distributed by ICARDA in IBYT-LRA-M trial and selected by GAP IARTC in Diyarbakır in 2003-2004 growing season.

This cultivar was first tested in yield trial for two years (2003/04-2004/05) and selected for regional trials. It was tested for three years and in seven locations in Southeastern Anatolia Region of Turkey. It was proposed as candidate cultivar to registration and certification center in 2007, because of its high stability and superiority. It was registered in 2011.

The morphological characters: Altıkat has vertical development, white leaf color, long stems, early maturity. The plant height of Altıkat (100-130 cm) is shorter than Şahin 91 and Sur 93, also the maturity of Altikat is earlier than Şahin 91 and Sur 93 cultivars.

Test weight of Altıkat is similar to Akhisar 98 and Vamıkhoca 98, but less than Sur 93 and Şahin 91. The grain aleurone layer color is weak in seed with thin chaff. The technological characteristics of Altıkat were determined through 3 years testing by Ankara Central Quality Laboratory for field crops with Şahin 91, Sur 93, Atılır and Fırat check cultivar. Altıkat's average thousand grain weight, hectoliter weight and protein content were less (5%-15%) than two rowed barley cultivars which were used as check in study.

Altıkat was tested by registration and certification center in 7 environments in 2008-2009 and 2009-2010 seasons. The yield average of Altıkat was 4745 kg ha-1, while the yield average of checks was 4475 kg ha-1. The results of yield trials showed that Altıkat has 6% more than checks averages. The stability results indicated that Altıkat had b value (0.9777) to near 1 and positive a value (35.8). Moreover, Altıkat was the best yielding genotype in bad conditions, while second after Atılır cultivar in good conditions. The results showed that the yield of Altıkat could be higher after Atılır in good conditions, but it is yielding 6% more than checks in bad conditions. Altıkat is recommended for farms because of high yielding and stability in Southeastern Anatolia Region of Turkey.

Figure 1. Spike and grain of the Altıkat cultivar

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References

Anonymous (2011). The results of registration and certification center trials. Annual reports. Variety Registration and Seed Certification Center, Ankara, Turkey.

Kendal E (2011). The barley cultivation in the GAP Region, Journal of Food, Agricultural and Livestock Directorate of Mardin, Page: 44-45.

Kendal, E, Tekdal, S, Aktas, H, Karaman, M, Berekatoglu, K, Dogan H (2014). Determination of yield and yield components of spring barley genotypes using biplot analysis. Balkan Agricultural Congress, Abstract book, Page: 510, 8-11 September 2014.

Kendal, E. and Dogan H (2014). Impact of two-six row in spike on yield, some quality and morphological parameters in barley. Turkish Journal of Agriculture and Natural Sciences, 1(2), 131-142, 2014.

Kendal, E (2014). Comparison of traditional and bed planting system on some agronomical characteristics in barley. Dicle University, Institute Journal of Science and Technology, 3(1), 43-52,2014.

Kilic H (2014). Additive main effect and multiplicative interactions (AMMI) Analysis of grain yield in barley genotypes across environments. Journal of Agricultural Science.20, 337-344.

http://arastirma.tarim.gov.tr/gaputaem. The results of barley breeding projects. Annual reports (2003-2014).