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Original scientific article

MOLECULAR CHARACTERIZATION OF WILD GRAPES FROM NORTHEASTERN PART OF TURKEY

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Progress in grape breeding requires the exploitation of genetic variation among market classes, races and gene pools. Wild grapevines (*Vitis vinifera* ssp. *sylvestris*) are being endangered in their natural habitats and high priorities should be given to the wild germplasm.

Turkey is one of the richest sources of wild grapevine and they mostly grown on forest trees on river basin. The present study was carried out to determine the amount of genetic variation and the degree of relatedness among 23 wild grape genotypes using 17 simple-sequence-repeat markers (SSR). Two international grape cultivars, Cabernet Sauvignon and Merlot are also included study. Number of alleles per locus of the 17 Simple Sequence Repeat (SSR) markers ranged from 3.0 to 14.0 and a total of 162 alleles with an average of 9.53 alleles per locus. The average expected and observed heterozygosity values were 0.773 and 0.781, respectively, which exhibited high level of genetic diversity in the wild grape germplasm. The unweighted pair group method with arithmetic mean analysis revealed three main genetic clusters that partially separated wild grape genotypes each other and. The international cultivars formed a out group. The high genetic diversity among native wild grapes from Coruh valley is suggesting that this area could be one of the centre of diversity of the specie. The results indicate a substantial genetic diversity in *V.*

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vinifera ssp. *sylvestris* and the need of exploring a wider area to increase the chance of finding a particular genotype.

Keywords: Wild grape, SSR, molecular characterization, diversity

INTRODUCTION

The plant genetic resources found in all continents of the world as an important part of agriculture ecosystem and they included wild relatives of cultivated species, traditional varieties, modern cultivars, crop wild relatives and other wild plant species. They also accepted as valuable breeding material due to richness of different gene combinations. Plant genetic resources important natural biological resources as well and they have been using centuries to obtain varieties and hybrids that increased economic benefits and sustainability (ERCISLI, 2004). They are also the sources of rare genetic traits needed for crop improvement to cope with environmental stresses, plant disease and pests etc.

Genetic erosion, loss of genetic diversity, was occurred in many cultivated plant species. The main reason of loss of wild relatives is related mainly to the reduction or loss of habitat due to land use for agriculture, urbanization and industrialization (RIAZ *et al*., 2018). The loss of wild relatives at an alarming rate and unless they are conserved now before it is too late, the genes contained in them will not be available when required in future. Thus collecting, preserving and characterizing of grape wild relatives are an important task to obtain new cultivars with higher yield and better quality (EMANUELLI *et al*., 2013).

Turkey is one of significant country in the world from the standpoint of plant genetic resources and plant diversity. Two of the Vavilov's Centre of Origin (i.e., Near Eastern and Mediterranean Centres) extends into Turkey. This, of course, indicates that Turkey is one of the center of origin and/or centre of diversity of several crop plants and many plant species (KARAGOZ, 2003; EYDURAN *et al*., 2015).

Turkey has important wild grapevines populations and grape cultivars, which offers to grapes breeders a valuable gene pool from where to extract genes of interest. Turkey has only *Vitis vinifera* ssp. *sylvestris* as a wild grape. Wild grapes have valuable features regarding the biotic and abiotic stress factors such as resistance to lime, drought, pest and disease. That is why they are of great interest for researchers and many studies have been made to determine their distribution in Turkey. Wild grapes are distributed all over the country, mainly in river basins and forests. Wild grapes have distinct characters when compared to seedlings of cultivated grapes (UZUN and BAYIR, 2010).

The traditional methods for the identification and differentiation of grape cultivars are based on ampelographic characteristics, which based on morphological differences between the cultivars (BENJAK *et al*., 2005; KHADIVI-KHUB *et al*., 2014; SOYLEMEZOGLU *et al*., 2014). However, the phenotype of plants is heavily influenced environmental conditions as well as nutritional state and health. Plant developmental stages are also affecting morphology of plants and to make correct ampelographic characterization fully developed grape leaves are needed. Application of ampelographic methods requires skilled individuals as well. For these reasons, alternative and reliable methods for cultivar identification are required.

The conservation and sustainable use of plant genetic resources require accurate identification of their accession. The dramatic advances in molecular genetics over the last few

years have provided workers involved in the conservation of plant genetic resources with a range of new techniques for easy and reliable identification of plant species. Many of these techniques have been successfully used to study the extent and distribution of variation in species gene-pools and to answer typical evolutionary and taxonomic questions (KARP *et al*., 1997). Properties desirable for ideal DNA markers include highly polymorphic nature, co-dominant inheritance (determination of homozygous and heterozygous states of diploid organisms), frequent occurrence in the genome, selective neutral behavior (the DNA sequences of any organism are neutral to environmental conditions or management practices), easy access (availability), easy and fast assay, high reproducibility, and easy exchange of data between laboratories (ERCISLI *et al*., 2008; ERCISLI *et al*., 2011; EYDURAN *et al*., 2016).

SSR also known as microsatellite repeats, consist of short nucleotide sequences that are repeated many times in tandem. The number of SSR tandem repeats can vary in a sequence, and many such variants (alleles) can exist in a population (POWELL *et al*., 1996). SSR markers tend to be amongst the most polymorphic genetic marker types and have been introduced into the process of cultivar identification as well as in pedigree reconstruction and genetic mapping (CASTRO *et al*., 2011).

The main objective of this study was to analyze the pattern of genetic diversity within wild grapes from the Northeastern Turkey near Caucasia considered to be the center of grape domestication.

MATERIAL AND METHODS

Plant material

In the study, we used 23 wild grape (*Vitis vinifera* ssp. *sylvestris*) naturally grown Yusufeli district in northeastern part of Turkey near Caucasia. The wild grape (*Vitis vinifera* ssp *sylvestris*) samples were collected from plants located in their natural habitats mostly along the Coruh and Barhal rivers in 2017. Care was taken to select plants that were dioecious and notes were made for the flower phenotype and leaf morphology. We added 2 well-known foreign grape cultivars namely Cabernet Souvignon and Merlon as reference in SSR analysis.

DNA extraction

Genomic DNA was extracted from young leaf tissue using the Wizard® Genomic DNA Purification Kit (Promega, Madison, WI) according to the instructions provided by the manufacturer. Subsequently, an RNAse treatment was performed the eluted DNA samples. Purity and concentration of the DNA were checked both on 1% (w/v) agarose gels and by NanoDrop® ND-1000 Spectrophotometer.

SSR analysis

A total 17 SSR primers that widely used in *Vitis* for molecular characterization have been used (Table 1). *Polymerase Chain Reaction* (PCR) was conducted in a volume of 10 µL and contained 15 ng genomic DNA, 5 pmol of each primer, 0.5 mM dNTP, 0.5 unit GoTaq DNA polymerase (Promega), 1.5 mM $MgCl_2$ and 2 µL 5X buffer. The forward primers were "labelled" with WellRED fluorescent dyes D2 (black), D3 (green) and D4 (blue) (Proligo, Paris, France). Reactions without DNA were included as negative controls. PCR amplification was performed using the Biometra® PCR System. The amplification conditions consisted of an initial

denaturation step of 3 min at 94°C, followed by 35 cycles of 1 min at 94°C, 1 min at 52-56°C and 2 mins at 72°C with a final extension at 72°C for 10 mins. Determination of polymorphisms, the PCR products were run on CEQ™ 8800 XL Capillary Genetic Analysis System (Beckman Coulter, Fullerton, CA). The analyses were repeated at least twice to ensure reproducibility of the results.

Data analysis and genetic relationships

The genetic analysis program "IDENTITY" 1.0 (WAGNER and SEFC, 1999) was used according to PAETKAU *et al*. (1995) for the calculation of number of alleles, expected and observed heterozygosity. Genetic similarity was determined by the program "MICROSAT" (version 1.5) (MINCH *et al*., 1995) using proportion of shared alleles, which was calculated by using "ps (option 1 - (ps))", as described by BOWCOCK *et al*. (1994). The results were then converted to a similarity matrix, and a dendrogram was constructed with the UPGMA method (SNEATH and SOKAL, 1973) using the software NTSYS-pc (Numerical Taxonomy and Multiware Analysis System, version 2.0) (ROHLF, 1988).

RESULTS AND DISCUSSION

In SSR analysis, 17 highly polymorphic simple sequences repeat (SSR) primer pairs screened for amplification of 23 wild grape genotypes and 2 reference cultivars. In experiment, all SSR primers gave reproducible and scorable amplification product from 23 wild grape genotypes and 2 standard grape cultivars. In general, we obtained higher levels of heterozygosity in sylvestris because of its obligate out-crossing nature.

Table 1. Simple sequence repeats (SSRs), number of detected alleles, observed heterozygosity (Ho) and expected heterozygosity (He) of 17 SSR markers on 23 wild genotypes and 2 grape cultivars investigated.

SSR Primers	Number of alleles	Expected heterozygosity (*He*)	Observed heterozygosity (*Ho*)
VMC1B11	10	0.727	0.690
VMC4F3.1	14	0.824	0.911
VVIB01	7	0.657	0.693
VVIH54	11	0.822	0.887
VVIN16	3	0.701	0.645
VVIN73	9	0.580	0.597
VVIP31	13	0.867	0.858
VVIP60	10	0.739	0.698
VVMD5	9	0.783	0.807
VVMD7	12	0.768	0.790
VVMD21	6	0.657	0.711
VVMD24	5	0.840	0.805
VVMD25	9	0.745	0.758
VVMD27	8	0.830	0.812
VVMD28	12	0.907	0.886
VVMD32	11	0.849	0.816
VVS2	13	0.833	0.904
Total	162	13.129	13.268
Average	9.53	0.773	0.781

Table 1 shows codes of SSR primers, the number of alleles for each primer, expected heterozygosity and observed heterozygosity. The results showed that all SSR primers used gave polymorphic alleles. A total of 162 polymorphic alleles were obtained across 25 grape genotypes and cultivars. The number of amplified fragments (polymorphic alleles) ranged from 3 (VVIN16) to 14 (VMC4F3.1), with an average of 9.53 fragments per primer (Table 2). Among SSR markers, VMC4F3.1 gave the highest number of allele (14 allele), followed by VVIP31 and VVS2 (13 allele) and VVMD7 and VVMD28, (12 allele). The lowest number of alleles was observed in VVIN16 SSR marker as 3 (Table 1).

Expected heterozygosities (*He*) were variable across loci reflecting the different number and frequencies of the alleles found. For 17 loci, VVIN73 had the lowest expected heterozygosity (*He*) as 0.580 while VVMD28 loci gave the highest expected heterozygosity value as 0.907. It was especially visible for VMC4F3.1 where a higher (*Ho*) was observed as 0.911. Observed heterozygosity was the lowest as 0.597 in VVIN73 loci. In general, considering average value, expected heterozygosity (*He*) was slightly lower (0.773) than the observed values (*Ho*) (0.781) (Table 1).

This study has demonstrated the utility of universal 17 SSR markers among wild grape germplasm in Turkey. KARATAS *et al*. (2014) used 21 wild grape samples from southern Anatolia and compared with Turkish and international grape cultivars by using 22 SSR markers. They detected a total of 222 alleles at the 22 SSRs loci analyzed and the number of alleles per SSR locus ranged from 4 (VVIN16) to 20 (VVIV67) and the mean allele number per locus was 10.09 indicating similarities with our results. They found VVIn16 as the lowest polymorphic loci and VMC4f3 as the most polymorphic loci as we determined. They also reported expected and observed heterozygosity ranged from 0.586 (VVIb01) to 0.898 (VVIV67) and 0.545 (VVMD21) to 0.906 (VVMD28). ERGUL *et al*. (2011) used 15 SSR primer combinations efficiently for wild grape accessions. They obtained the number of scoreable fragments amplified by each SSR primer pair from 6 (VVIN16 and VVIB01) to 21 (VMC4F3.1) with an average of 12.26 per primer combination. They obtained 184 scoreable alleles (12.27 allele per loci) were detected among the 84 wild grape genotypes. They also reported average observed and expected heterozygote values as 0.748 and 0.811 indicating higher expected value than observed value. We obtained higher allele number with VMC4f3, VVIP31, VVS2, VVIP60, VVMD5 and the lowest values were obtained from VVIN16, VVIB01, VVIN73 and VVMD25 (Table 1). RIAZ *et al*. (2018) used a wide number of wild grape genotypes originating from Asia to Europe and characterized by 20 SSR loci. They found that VMC4f3 was the most effective loci with an average of 20.95 alleles per locus. They obtained that observed and expected heterozygosity varied greatly among loci. They reported *He* values ranged from 0.477 (VVIn73 locus) to 0.803 (VVS2), with a mean value equal to 0.678. While the *Ho* values varied from 0.535 (VVIn73) to 0.845 (VVIp31) and the mean overall value was 0.742 indicating similarities with our study. LIU *et al*. (2012) studied genetic diversity among wild grapes in China and found that VVS2 and VVMD7 were the effective primers to discriminate genotypes. They reported the number of alleles from 6-12 with an average of 9 alleles per locus, which is in agreement with our result. In Iran, to reveal genetic diversity and provide information to conserve valuable grapevine germplasm, 63 wild accessions collected from five forest locations of the Zagros Mountains were genotyped by using 23 simple sequence repeats (SSR) loci. All used primer pairs produced

polymorphic banding patterns. In total, 182 alleles ranging from 3 (VVMD21) to 13 (VVMD5 and VVMD8) and with an average of 7.9 alleles/locus were amplified (BANEH *et al.*, 2015). They reported average expected and observed heterozygosity as 0.74 and 0.69, respectively. The number of average polymorphic alleles per primers was higher than obtained by BANEH *et al*. (2015). In our study VVMD5 primer gave 11 allele per loci indicating close value with above study. ARROYA-GARCIA *et al.* (2016) characterized wild grapevine in Spain by using SSR primers and they found the mean number of alleles between the different populations ranged from 6.3 to 9.1. They found that *Ho* ranged between 0.608 and 0.687, whereas the values of *He* were slightly higher, ranging between 0.636 and 0.766 which in accordance with our results.

The dendrogram resulting from UPGMA cluster analysis showed that the studied genotypes and cultivars could be divided three main clusters. The first cluster contained 11 *Vitis vinifera* ssp *sylvestris* originated from Coruh valley in Turkey and cluster I further divided 2 subgroups. In cluster I, the closest Turkish wild grape genotypes were genotype 2 and genotype 19 with 73% similarity ratio. Cluster II included 12 wild grape genotypes. The standard international cultivars Cabernet Souvignon and Merlon clustered together as out-group from wild genotypes. Based on SSR profiles of 23 wild grape genotypes and 2 foreign cultivars, we could not observed that 23 wild grape genotypes and cultivars genetically identical.

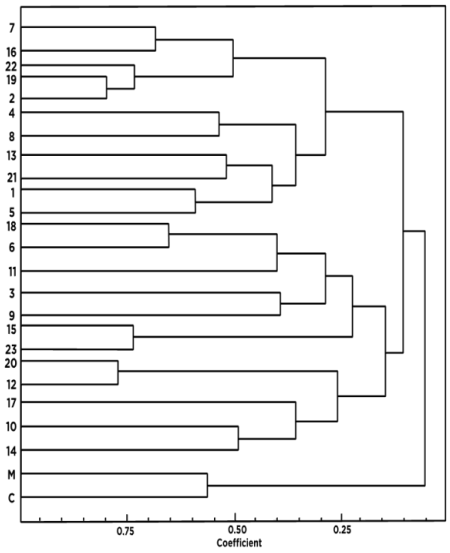

Figure 1. The UPGMA dendrogram based on simple matching similarity matrix obtained using 17 SSR markers, illustrating the relative similarity among 23 wild grape genotypes and 2 international grape cultivars. C: Cabernet Sauvignon, M: Merlot

Previous studies indicated great diversity among wild grape genotypes from different parts of the world and no identical genotypes has been reported in these studies (ERGUL *et al*., 2011; LIU *et al*., 2012; KARATAS *et al*., 2014; BANEH *et al*., 2015; ARROYA-GARCIA *et al*., 2016;

RIAZ *et al*., 2018) supporting our results. The study showed that molecular marker technologies are the most advanced and, possibly, the most effective means for understanding the basis of genetic diversity in wild grape. They are efficient and accurate tools with which genetic variation can ultimately be identified and assessed in a rapid and thorough manner. In fact, numerous previous studies showed that horticultural plants are genetically very diverse due to open pollination nature (ERCISLI *et al*., 2003; ZIA-UL-HAQ *et al*., 2013; DOGAN *et al*., 2014; EYDURAN *et al*., 2014; GUNDOGDU *et al*., 2014; GECER *et al*., 2020; KASKONIENE *et al*., 2020; ENGIN and MERT, 2020)

By applying molecular technologies to approach the biological questions underlying the understanding of genetic diversity, we can make significant progress in the speed and depth at which we attain adequate and appropriate conservation and, thus, genetic resources made available for its use in crop improvement.

Associated with the high reproducibility of the SSR markers, the results obtained in this study support the use of these markers as an important tool in the molecular characterization of wild grape genotypes in germplasm banks, in the identification of duplicates, in the correct identification of cultivars and of genetically divergent potential parents to be used in breeding programs.

CONCLUSION

In conclusion, the gene pool of the wild Vitis surveyed in Northeast Anatolia has significant amounts of genetic variation. In regard to germplasm management, our results show that the germplasm collection is highly variable and most variation is common to all genetic groups identified. To our knowledge, this is the first comparison of the discriminating capacity, efficiency and ability of marker system in Turkish wild grape in Northeastern Anatolia. The results of the present work will be the basis for future studies on the phylogenetic relationships and germplasm organization in the genus Vitis.

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REFERENCES

ARROYO-GARCIA, R., M., CANTOS., M., LARA., M.A., LÓPEZ., A., GALLARDO., C.A., OCETE., A., PÉREZ., H., BÁNÁTI., J.L., GARCÍA., R., OCETE (2016): Characterization of the largest relic Eurasian wild grapevine reservoir in Southern Iberian Peninsula. Span. J. Agric. Res., *14* (3): e0708.

BANEH, H.D., S.A., MOHAMMADI., M., LABRA., F., SHAFAIE (2015): Genetic diversity of wild grape (*Vitis vinifera* ssp. *sylvestris*) populations in Zagros forests as revealed by SSR markers. Acta Hortic., *1082*:189-194.

BENJAK, A., S., ERCISLI, A., VOKURKA, E., MALETIC, I., PEJIC (2005): Genetic relationships among grapevine cultivars native to Croatia, Greece and Turkey. Vitis, *44*(2):73–77.

BOWCOCK, A.M., A., RUIZ-LINARES., J., TOMFOHRDE, E., MINCH, J.R., KIDD, L.L., CAVALLI-SFORZA (1994): High resolution of human evolutionary trees with polymorphic microsatellites. Nature, *368*: 455-457.

CASTRO, I., J.P., MARTIN, J.M., ORITZ, O., PINTO-CARNIDE (2011): Varietal discrimination and genetic relationships of *Vitis vinifera* L. cultivars from two major Controlled Appellation (DOC) regions in Portugal. Sci. Hortic., *127*: 507-514.

DOGAN, H., ERCISLI, S., TEMIM, E., HADZIABULIC, A., TOSUN, M., YILMAZ, S.O., ZIA-UL-HAQ, M (2014): Diversity of

chemical content and biological activity in flower buds of a wide number of wild grown caper (*Capparis ovate* Desf.) genotypes from Turkey. C.R. Acad. Bulg. Sci. *67*: 1593-1600.

EMANUELLI, F., J., BATTILANA, L., COSTANTINI, L., LE CUNFF, P., THIS, M.S., GRANDO (2010): A candidate gene association study for Muscat flavor in grapevine *Vitis vinifera* L. BMC Plant Biol., *10*:241.

ENGIN, S.P., MERT, C (2020): The effects of harvesting time on the physicochemical components of aronia berry. Turk. J. Agric. For. *44*:361-370.

ERCISLI, S., ESITKEN, A., CANGI, R., SAHIN, F (2003). Adventitious root formation of kiwifruit in relation to sampling date, IBA and *Agrobacterium rubi* inoculation. Plant Growth Regul. *41*: 133-137.

ERCISLI, S. (2004): A short review of the fruit germplasm resources of Turkey. Genet. Res. Crop Evol., *51*:419-435.

ERCISLI, S., E., ORHAN, A., ESITKEN, N., YILDIRIM, G., AGAR (2008): Relationships among some cornelian cherry genotypes (*Cornus mas* L.) based on RAPD analysis. Genet. Res. Crop Evol., *55* (4), 613-618.

ERCISLI, S., A., IPEK, E., BARUT (2011): SSR marker-based DNA fingerprinting and cultivar identification of olives (*Olea europaea*). Biochem Genet., *49*(9-10): 555-561.

ERGUL, A., G., PEREZ-RIVERA, G., SOYLEMEZOGLU, K., KAZAN, R., ARROYO-GARCIA (2011): Genetic diversity in Anatolian wild grapes (*Vitis vinifera subsp. sylvestris*) estimated by SSR markers. Plant Genet. Res., *9*:375-383.

EYDURAN, S.P., ERCISLI, S., AKIN, M., BEYHAN, O., GECER, M.K (2014): Organic Acids, sugars, vitamin c, antioxidant capacity, and phenolic compounds in fruits of white (*Morus alba* L.) and black (*Morus nigra* L.) mulberry genotypes. J. Appl. Bot. Food Qual., *88* :134-138.

EYDURAN, S.P., M., AKIN, S., ERCISLI, E., EYDURAN, D., MAGHRADZE (2015): Sugars, organic acids, and phenolic compounds of ancient grape cultivars (*Vitis vinifera* L.) from lgdir province of Eastern Turkey. Biol. Res., *48*:2.

EYDURAN, S.P., S., ERCISLI, M., AKIN, E., EYDURAN (2016): Genetic characterization of autochthonous grapevine cultivars from Eastern Turkey by simple sequence repeats (SSRs), Biotechnol. Biotechnol. Equip., *30*:26-31.

GECER, M.K., KAN, T., GUNDOGDU, M., ERCISLI, S., ILHAN, G., SAGBAS, H.I (2020): Physicochemical characteristics of wild and cultivated apricots (*Prunus armeniaca* L.) from Aras valley in Turkey. Genet. Resour. Crop Evol. *67*:935-945.

GUNDOGDU, M., OZRENK, K., ERCISLI, S., KAN, T., KODAD, O., HEGEDUS, A (2014): Organic acids, sugars, vitamin C content and some pomological characteristics of eleven hawthorn species (Crataegus spp.) from Turkey. Biol. Res. *47*: 21.

KARAGOZ, A. (2003): Plant genetic resources conservation in Turkey. Acta Hortic., *598*:17-25.

KARATAS, D.D., H., KARATAS, V., LAUCOU, G., SARIKAMIS, L., RIAHI, R., BACILIERI, P., THIS (2014): Genetic diversity of wild and cultivated grapevine accessions from southeast Turkey. Hereditas, *151*:73-80.

KARP, A., S. KRESOVICH, K.V., BHAT, W.G., AYAD, T., HODGKIN (1997): Molecular Tools in Plant Genetic Resources Conservation: A Guide to the Technologies. IPGRI Technical Bulletin No. 2; International Plant Genetic Resources Institute; Rome, Italy: 1997.

KASKONIENE, V., BIMBIRAITE-SURVILIENE, K., KASKONAS, P., TISO, N., CESONIENE, L., DAUBARAS, R., MARUSKA, A.S (2020): Changes in the biochemical compounds of *Vaccinium myrtillus*, *Vaccinium vitis-idaea*, and forest litter collected from various forest types. Turk. J. Agric. Fo. *44*:557-566.

KHADIVI-KHUB, A., A., SALIMPOUR, M., RASOULI (2014): Analysis of grape germplasm from Iran based on fruit characteristics. Braz. J. Bot., *37*,105-113.

LIU, C.H., X., FAN, J., JIANG, D., GUO, H., SUN, Y., ZHANG, J., FENG (2012): Genetic diversity of Chinese wild grape species by SSR and SRAP markers. Biotechnol. Biotechnol. Equip., *26*(2):2899-2903.

MINCH, E., A., RUIZ-LINARES, D.B., GOLDSTEIN, M., FELDMAN, L.L., CAVALLI-SFORZA (1995): Microsat (Version 1.4d): a

computer program for calculating various statistics on microsatellite allele data. Stanford University Medical Center, Stanford.

PAETKAU, D., W., CALVERT, I., STIRLING, C., STROBECK (1995): Microsatellite analysis of population structure in Canadian polar bears. Mol. Ecol., *4*: 347-354.

POWELL, W., M., MORGANTE, C., ANDRE, M., HANAFEY, M., VOGEL, S., TINGEY, A., RAFALSKI (1996): The comparison of RFLP, RAPD, AFLP and SSR (microsatellite) markers for germplasm analysis. Mol. Breeding., *2*: 225–238.

RIAZ, S., G.D., LORENZIS, D., VELASCO, A., KOEHMSTEDT, D., MAGHRADZE, Z., BOBOKASHVILI, M., MUSAYEV, G., ZDUNIC, V., LAUCOU, M.A., WALKER, O., FAILLA., J.E., PREECE, M., ARADHYA, R., ARROYO-GARCIA (2018): Genetic diversity analysis of cultivated and wild grapevine (*Vitis vinifera* L.) accessions around the Mediterranean basin and Central Asia. BMC Plant Biol., *18*:137.

ROHLF, F.J. (1988): NTSYS-PC Numerical Taxonomy and Multivariate Analysis System. Exeter Publishing, New York.

SNEATH, P.H., R.R., SOKAL (1973): Numerical Taxonomy. Freeman, San Francisco.

SOYLEMEZOGLU, G., Y.S., AGAOGLU, I., UZUN (2014): Ampelographic characteristics and isozymic analysis of *Vitis vinifera* spp. sylvestris (Gmel.) in Southwestern Turkey. Biotechnol. Biotechnol. Equip., *15*(2):106-113.

UZUN, H.I., A., BAYIR (2010): Distribution of wild and cultivated grapes in Turkey. Not. Sci,. Biol., *2*: 83–87.

WAGNER, H.W., K.M., SEFC (1999): Identity 1.0. Centre for Applied Genetics. University of Agricultural Science, Vienna.

ZDUNIĆ, J., E., MAUL, K., HANČEVIĆ, M., LEKO, L., BUTORAC, A., MUCALO, T., RADIĆ, S., ŠIMON, I., BUDIĆ-LETO, M-Z., MIHALJEVIĆ, E., MALETIĆ (2017): Genetic diversity of wild grapevine (*Vitis vinifera* subsp. *sylvestris* Gmel Hegi) in the eastern Adriatic region. Am. J. Enol. Vitic., 2017, *68*:252-257.

ZIA-UL-HAQ, M., AHMAD, S., QAYUM, M., ERCISLI, S (2013): Compositional studies and antioxidant potential of *Albizia lebbeck* (L.) Benth. Pods and seeds. Turk. J. Biol. *37* (1): 25-32.

MOLEKULARNA KARAKTERIZACIJA DIVLJE VINOVE LOZE IZ SEVEROISTOČNOG DELA TURSKE

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Izvod

Napredak u oplemenjivanju grožđa zahteva korišćenje genetskih varijacija među klasama, rasama i genskim *pool*-ovima. Divlja vinova loza (*Vitis vinifera* ssp. *sylvestris*) je ugrožena u svojim prirodnim staništima, a divljoj germplazmi treba dati prioritet. Turska je jedan od najbogatijih izvora divlje vinove loze i uglavnom se uzgaja na šumskom drveću u slivu reka. Ova studija je sprovedena da bi se utvrdila količina genetskih varijacija i stepen srodnosti među 23 genotipa divlje vinove loze koristeći 17 SSR markera. Uključene su i dve međunarodne sorte grožđa, kaberne sovinjon i merlot. Broj alela po lokusu 17 SSR markera kretao se od 3,0 do 14,0 i od ukupno 162 alela, prosečno je bilo 9,53 alela po lokusu. Prosečne očekivane i zabeležene vrednosti heterozigotnosti bile su 0,773, odnosno 0,781, što je pokazalo visok nivo genetske raznolikosti divlje vinove loze. Neponderisana metoda parnih grupa sa analizom aritmetičke sredine otkrila je tri glavna genetska klastera koja su delimično razdvajala genotipove divljeg grožđa. Međunarodne sorte formirale su spoljnu grupu. Visoka genetska raznolikost samoniklog divljeg grožđa iz doline reke Joruh sugeriše da bi ovo područje moglo biti jedno od centara divergentnosti ove vrste. Rezultati ukazuju na značajnu genetsku raznolikost *V. vinifera ssp. sylvestris* i na potrebu za istraživanjem šireg područja kako bi se povećala šansa za pronalaženje određenog genotipa.

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