THE BIOLOGICAL VITICULTURE AND GENOTYPE FUNCTIONALITY

Boris GĂINĂ[3], Eugeniu ALEXANDROV[4]

Abstract. *The expected results in modern agriculture can be obtained by applying environmental technologies and taking into account the genotype functionality factor. Knowing the functional properties of the genotype and the use of environmental technologies for growing grapes, in the future it will be possible to create balanced, sustainable and diversified agroecosystems, which will guarantee the protection of natural resources, sustainable development of society and public health. The grape sector needs to create new grape varieties, with stable productive potential, to produce high-quality derived products. As a result of the crossing of the species V. vinifera L. ssp. sativa D.C. (2n=38) with Muscadinia rotundifolia Michx. (2n=40), root-specific genotypes of grapes were obtained, to which ecological cultivation technologies can be applied, for example: Malena, Nistryana and Algumaks as table varieties and Augustine, Alexandrina and Amethyst for fresh consumption and for processing.*

Keywords: biological products, genotype, environment, functionality, ecological viticulture.

Introduction

Society is developing steadily when it consumes high-quality products of natural origin, rationally uses natural resources, and the environmental impact is minimal. Environmental protection is a global problem that should become a national priority, since it directly relates to the living conditions and health of the population, achievement of economic interests, as well as opportunities for sustainable development of society [12, 13]. Sustainable development means that way of development of human society, which is aimed at meeting the needs of the current generation, without affecting the level and quality of life of future generations. Each generation should strive to satisfy its own needs, without leaving various generations of debts for future generations, including environmental ones - depletion of natural resources or pollution of soil, water, air, etc. [13].

Organic viticulture is becoming increasingly popular around the world. This trend is based on a system of technological methods aimed at maintaining the

[3] Academician, Academy of Sciences of Moldova, e-mail: b.gaina@mail.ru
[4] Doctor habilitat in biological sciences, Institute of Genetics, Physiology and Plant Protection, e-mail: e_alexandrov@mail.ru

biodiversity of grapevine, the successful cultivation and consumption of grapes with minimal use of chemical treatments, and therefore with minimal content of their residues in the crop [2, 3, 19].

The development of human society requires special attention to problems related to environmental protection and the rational use of natural resources. There is no doubt that it is necessary to know the possibilities of the genetic potential of genotypes depending on climatic conditions, which have a significant impact on programming the quantity and quality of products [13].

Agriculture, depending on pedo-climatic and socio-economic conditions, should provide the population with high-quality food-derived natural products, supply of raw materials for industry, rational use of natural resources and conservation of biodiversity. One of the main tasks of agriculture is the identification of new genotypes that can easily adapt and develop in the context of climate change without harming the environment. To achieve this goal, you need to have genotypes with increased functionality.

Derived grape and wine products make a significant contribution to the development of the country's economy, and therefore it is necessary to pay special attention to the creation of vineyards with the introduction of environmental cultivation technologies. It is necessary to create genotypes with increased functionality and the choice of technology for growing grapes, which would allow to obtain high-quality and high productivity, with a minimum number of protection products, and maybe without them, thereby preserving the environment and biological diversity, as well as the presence of a minimum of these substances in berries and grape and wine-derived products [6, 7, 9, 14, 15].

To obtain the highest quality grape and wine derivatives, three main factors must be taken into account: genotype (variety), location of the vineyard (pedo-climatic conditions) and technology (cultivation and processing). Despite the fact that Vitis vinifera L. ssp. sativa D.C. has a high genetic potential, yet intraspecific genotypes cannot cross the genetic barrier of high sensitivity to changes in climatic conditions within the growing area, therefore it is necessary to create interspecific genotypes with high-quality products and resistant to environmental factors [6, 7, 9, 10 - 12, 14].

A research on this issue was carried out by the Research Institute of Organic Agriculture, in Switzerland, which published data on the total area of organic vineyards in the world. In 2015, this area was more than 400 thousand hectares. It has been found out that the transition of vineyards from traditional to organic technologies takes 3 years, after which the grape harvest is considered to comply with the established requirements. As for the growth rate of the area of vineyards cultivated according to the organic technologies, it was established that from 2005 to 2015, it trebled. The world leaders, in terms of area of organic vineyards and production of organic grapes, are Spain – with 85 thousand hectares, France – 75 thousand hectares, Italy – 65 thousand hectares, Mexico – 35 thousand hectares and China – 20 thousand hectares. Currently, the shares of areas of organic vineyards in European countries are as follows: Austria – 10.7 %, Italy – 10.3 %, Spain – 8.9 %, France – 8.7 %, Germany – 7.5 %, Portugal – 1.5 % [15]. More than 270 thousand hectares of organic vineyards are cultivated on the European continent, which makes up 67.5 % of the total area of organic vineyards in the world. This new trend in grape cultivation is developing rapidly in China, Turkey, Italy, Germany, Argentina, Chile, Australia and South Africa.

Materials and methods

The subject of the research was the collection of grapevine plants of the Institute of Genetics, Physiology and Plant Protection, Chisinau, Republic of Moldova. The collection includes 140 genotypes of interspecific and intraspecific, grafted and own-rooted genotypes of grapevine. The method of distant hybridization was used to create interspecific own-rooted genotypes of grapevine [4-7]. Studies were conducted in accordance with the methods of describing grapevine varieties [1, 8, 9], Methodical Recommendations for Grapevine Breeding, Study of grapes to determine how to use them. Uvology [11, 22]. The physicochemical assessment of derivative products of interspecific genotypes of grapes was performed in accordance with the methods for analysing derivative products [12, 10, 16]. To determine the resistance of the studied genotypes to phylloxera, pathogenic microorganisms etc., the methods mentioned in Normal and Pathological Anatomy of Grapevine Roots and Complex Protection of Grapevine [14-17, 21, 22] were used.

Results and discussions

Based on the functionality of grape taxa, it is possible to create genotypes that will provide an opportunity to increase the efficiency of interspecific hybridization of grapes in the process of improving valuable qualities. But, in order to achieve the set goal and obtain results, it is necessary to evaluate the genotypes included in the selection process from the point of view of functionality in relation to pedagogical conditions. To determine the functionality of genotypes, a comparative analysis of ampelographic, agrobiological, technological and physiological properties in relation to climate change is necessary. Taxa with enhanced functionality must be included in the process of creating interspecific genotypes based on pedo-climatic conditions.

In the process of selection of grapes, it is necessary to take into account the functional properties of the genotype: growing technology; fertility; productivity (in relation to the vegetative period and pedo-climatic zone); ratio of growth and fertility; histo-anatomical and biochemical properties; resistance to diseases and pests; resistance to low temperatures; the chemical composition of berry juice and derivatives (wine, distillate, etc.); bunch ripening (early - later); the appearance of a bunch; pulp properties; aroma and taste; resistance to cracking berries; the ability to store and transport grapes; use (fresh use); processing (wine, distillate, juice, etc.); recycling [1, 9, 10, 20].

Knowing the functional properties of the genotype and the use of environmental technologies for growing grapes, in the future it will be possible to ensure the creation of balanced, sustainable and diverse agroecosystems, which will guarantee the protection of natural resources, sustainable development of society and public health.

Ecotechnologies contribute to the creation of a variety of sustainable balanced agroecosystems, the rational use of natural resources, the reduction of the use of polluting technologies, the restriction of the use of chemical-synthetic substances and the reduction of potentially destructive agricultural work.

In the European market, there is a great demand for organic grapes, and it equally applies to grapes intended for consumption while fresh (table varieties) and those for the production of wine and distilled drinks. The organic cultivation of grapevine leads, on the one hand, to a reduction in the contamination of grapes with residues of the substances used to protect them from diseases and pests, and on the other hand, to a lower degree of environmental pollution (soil, water and air) [11, 12, 15].

The European Union supports organic viticulture, by subsidizing this type of activity of winegrowers (at the transition stages from traditional to organic cultivation of grapevine), covering the losses of economic agents whose vineyards have suffered from hail, epiphytotic diseases and prolonged rains that have led to crop loss. However, taking into account the fact that the demand for organic grapes (table and wine grapes) is constantly raising, not only in Europe, but also in North America and East Asia, at present, their price is 40-60 % or even 100 % higher. This, of course, attracts winegrowers, and today about 10 % of vineyards in the countries of the European Union are certified as “organic”. In France, in Alsace, some programs have been created to inform people about the peculiarities of the organic cultivation of grapevine, and their students are representatives of the grape-growing and winemaking industry of most EU countries, as well as Canada, the USA, Israel, Australia, New Zealand, China etc.

Currently, the technology of cultivation of organic grapevine must be consistent with certain standards: for example, the use of copper is limited to 3 kg per hectare per year, and the maximum sulphur application – 6 kg per hectare per year. There are countries where these standards differ in the direction of a slight decrease, but in all cases, the extract (infusion) of nettle, bark and leaves of oak, leaves of walnut, calendula etc. are widely used along with fungicides based on copper and sulphur.

However, the cultivation of grapes according to the organic technologies in the Republic of Moldova faces certain difficulties, which are the main reason behind the slow introduction of this new trend of the grape-growing and winemaking. One of them is the absence of biopreparations, produced on an industrial basis, to inhibit the growth of micromycetes (*Botrytis cinerea* etc.) and pests (leafroller moths etc.). For example, in France, a biopreparation made from *Trichoderma viride* is produced under the brand name Trichodermin B14; it has been proven to suppress the growth of mould fungi on berries and leaves in rainy weather by more than 60 % [13]. However, it has been found that this biofungicide loses a part of its action during the months with dry weather (August-September) which reduces the effectiveness of the treatment and increases the risk of micromycete infection at the beginning of the rainy period (September-October) [18]. Moreover, it has been found that the action of Trichodermin B14 is inhibited by residues of copper ions on leaves and berries, which remain after previous treatments of vineyards. In this case, the scientists from the French National Institute for Agricultural Research [3] have begun to grow the biomass of the antagonist *Trichoderma viride*, against grey mould, enriched with copper ions. Under these conditions, the biofungicide was resistant to the inhibiting effect of copper residues on leaves and berries and provided more than 75 % growth inhibition in mould fungi.

Another problem in organic viticulture is the isolation of areas with grapevine in the stages of conversion (within 3 years) from those cultivated nearby, by traditional technology. As a rule, the easiest way of solving this problem is to choose an entire vineyard (plantation), surrounded by other agricultural crops (fodder grasses, sometimes cereals etc.), or shelterbelts, often encountered in our country. The generally accepted rules for organic cultivation of grapes also stipulate the absence of any source of chemical or biological pollution (wastewater treatment plants, chemical plants, landfills etc.). In our opinion, the prices for the certification of plantations, which have been established by international organizations, licensed in this regard, are too high and unjustified, especially in cases of small plantations of grapes and other berries, fruits etc.

Other advantages of organic methods are a significant reduction in environmental pollution, lower costs for the purchase and use of expensive chemicals, as well as higher sale prices for wine and table grapes. In the EU markets, the price of certified organic grapes is 40-60 % or even 100 % higher as compared with the price of grapes cultivated by traditional methods.

Among the main problems faced by this new technology of cultivation of grapevine, there are the difficulties of using classical varieties of the genus *Vitis vinifera* L., because of the high susceptibility to the attack of micromycetes and pests, and the low frost tolerance. These are very important factors under the harsh conditions of the continental climate in our country, with high humidity and heavy rains in spring, which create favourable conditions for the development of dangerous diseases and pests, and which complicate the timely and effective use of chemical remedies in the framework of the organic technology. On the other hand, in the second half of summer and early autumn, the weather contributes to the development of other diseases, among which, powdery mildew (caused by *Oidium*) and grey mould of grapes are the most dangerous. The way out of this difficult situation may be the wide use of new grapevine varieties with higher resistance to biotic and abiotic environmental conditions [4]. The last fifty years of breeding grapevine have resulted in the creation of some promising varieties of wine grapes to be cultivated by organic methods: Viorica, Legend, Riton, Luminita, (Moldova), Bianca, Chardonel, Aletta, (Hungary), Vidal Blanc, Triumph of Alsace, Shamborsin (France), Fleurtai, Soreli, Savignon Cretos, Julius, Sagiovese etros, Merlot Chorus, Cabernet, Jerez, Julio, Caberigne Cretos, Julius, Sagiovese etret (Italy), Cabernet Jura, Pinotin, Cabernet blanc (Switzerland), Aromatny, Muscat Odessa, Zagreus, Rubin Tairovsky, Aghat Tairovsky, Golubok, Illichivsky early, Sparkling, Ovidiopol, Odessa Black, Rodnichek etc. (Ukraine). Grapevine varieties with high resistance to diseases and pests have been obtained and recommended for breeding and cultivation in Crimea – by the Institute of Viticulture and Winemaking “Magarach”, in Russia – by the All-Russian Research Institute of Viticulture and Winemaking “Ya.I. Potapenko”, in Bulgaria – by the National Institute of Viticulture and Oenology in Pleven, in Romania – by the Research and Development Institute for Viticulture and Winemaking “Valea Călugărească”. However, the above-listed varieties are susceptible to phylloxera, which makes it necessary to create vineyards from planting material grafted on phylloxera-resistant rootstocks. The viticulture sector needs new grape varieties, with stable productive potential, for the production of high-quality derivative products. The European grapevine varieties of *Vitis vinifera* L. ssp. *sativa* D.C., registered in the Republic of Moldova, as well as in other wine-producing countries, are susceptible to phylloxera (*Phylloxera vastatrix* Planch.) and that is why vineyards should be created from planting material grafted on phylloxera-resistant rootstocks. Besides, because grapevine is sensitive to low temperatures in winter, additional measures are necessary to protect plants during the period of vegetative rest [1, 7, 22].

To obtain competitive products, it is necessary to use mandatory chemical treatments to prevent or destroy pests, micromycetes and other pathogenic agents. However, these treatments affect the cost of production and pollute the environment. The creation of own-rooted grapevine plantations is a good prospect, but for this, it is necessary to enrich the grape assortment with new genotypes, resistant to diseases and pests. As a result of research, a methodology was developed for the creation of own-rooted interspecific genotypes of grapevine *Vitis vinifera* L. ssp. *sativa* D.C. x *Muscadinia rotundifolia* Michx., resistant to biotic and abiotic factors. Donors of valuable agrotechnological traits were included in the breeding process, as a result of which high-quality, stable and productive grapevine genotypes were created [2, 7].

Mastering the biological potential of interspecific genotypes will allow obtaining high-quality products from grapes, reducing costs and the use of chemicals in the process of controlling micromycetes and pests. The created genotypes have significant agrobiological and technological potential, which allows developing further research in the field of genetics and breeding of grapevine, using the method of distant hybridization. Thus, after of crossing *V. vinifera* x *M. rotundifolia,* interspecific genotypes of grapevine have been created in BC_3, with acquired agrobiological and technological properties, which allow expanding the area where grapevine can be cultivated in the northern regions and reducing the number of chemical treatments that will contribute to obtaining ecological products and protecting the environment. In the process of identifying the genetic functionality of the related taxa, *V. vinifera* and *M. rotundifolia*, characterized by low combining ability, it was found that this obstacle could be overcome by backcrossing. A wide range of recombinants, which allows improving the efficiency of distant hybridization in the process of selection of valuable characteristics, has been obtained as a result of this process [4, 5, 7].

The differences in the classification of interspecific genotypes of grapevine based on DNA profiles (SSR markers) and ampelographic criteria prove the importance of *genotype x environment* specific interactions in the development of biological and technological features of the hybrid. The multilateral research on biological and agrotechnological features, the participation in hybridization of the genotypes of different ecological and geographical origin of *V. vinifera* and *M. rotundifolia* and the elimination of aneuploid forms during subsequent crosses leads to the stabilization of the interspecific genome (2n = 38) with valuable agrobiological features and stability. The interspecific genotypes *V. vinifera* x *M. rotundifolia* can be propagated by cuttings from own-rooted, competitive planting material, to obtain early-ripening grapes [7].

When creating new grapevine varieties, by interspecific and intraspecific hybridization, it is very important to take into account the concentration, in the berries, of such chemicals as resveratrol, which ensures the resistance of the plant to adverse environmental factors. A comparative analysis of the concentration of resveratrol in the juice of wild grapes and its concentration in the berries, obtained after hybridization, has shown that, in the juice of wild grapes, the concentration of resveratrol is approximately two times higher than in subsequent generations, obtained as a result of hybridization. That is, as more generations are created, moving away from the wild representatives of the species, the concentration of resveratrol in the juice of the grapes keeps decreasing. The created interspecific genotypes of grapevine have been studied in detail according to agrobiological and technological criteria [2, 7].

The evaluation of the quality of grapes and derived products, over the years, has made it possible to select and cultivate promising own-rooted genotypes of grapevine. The interspecific genotypes of *V. vinifera* x *M. rotundifolia* are easily propagated by cuttings and can be cultivated on their own roots, thereby offering the opportunity to skip some practical steps, as well as reduce financial costs in the process of producing planting material and growing grapevine. According to the uvological and oenological criteria, the grapes of the new genotypes are not inferior to the classical varieties of *V. vinifera* in their biochemical composition and organoleptic qualities. Besides, they can be grown in the northern areas, where most plants of *Vitis vinifera* L. ssp. *sativa* D.C. do not withstand low temperatures in winter. Studying the physicochemical properties of blue-violet grapes of the interspecific genotypes (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.), it has been found that phenols, resveratrol and pectins are present in them in larger quantities than in green-yellow grapes, also exceeding the amount of these substances in the berries of the varieties of *V. vinifera* L. The quantity of resveratrol in the juice of berries of the interspecific genotypes of grapevine is 6.68 mg/l in berries with a green-yellow hue (BC_3-510 etc.), 9.3 mg/l in berries with a pink hue (BC_3-520 etc.) and 14 mg/l in berries with a blue-violet hue (BC_2-3-1, BC_3-660 etc.). From the populations of interspecific genotypes BC_3 (*V. vinifera* x *M. rotundifolia*), several promising own-rooted varieties have been selected, among them, there are table grapes, such as Malena, Nistreana and Algumax, and wine grapes: Augustina, Alexandrina and Ametist [3, 5, 7].

Using the biological potential of interspecific genotypes will make it possible to obtain high-quality derivative products under conditions of organic farming, which implies a reduction in the use of synthetic and natural chemicals in the fight against diseases and pests.

Conclusions

1. From the populations of interspecific genotypes BC_3 (*V. vinifera* x *M. rotundifolia*), several promising own-rooted varieties of table grapes have been selected, such as Malena, Nistreana and Algumax, and other selected varieties, such as Augustina, Alexandrina and Ametist, can be used as table grapes too, but also as wine grapes.

2. Growing interspecific genotypes of grapevine will decrease the negative impact on the environment by reducing the number of chemical treatments.

3. Due to the high resistance of distant hybrids to pests and diseases, the costs associated with the creation of planting material are reduced. Besides, as mentioned above, the number of chemical treatments during the cultivation process is reduced, thus minimizing environmental pollution.

4. In addition, the area of cultivation of grapevine can be expanded to the north, where the climatic conditions are unfavourable for the varieties of *V. vinifera*, which cannot tolerate the low winter temperatures, while the studied interspecific genotypes are more winter-hardy.

REFERENCES

1. Alexandrescu I., Oşlobeanu M., Jianu L., Piţuc P. Mică enciclopedie de viticultură. Iasi, 1972, 712 p.
2. Alexandrov E. The genotypes feed-back to the environmental factors. In: Scientific Papers Series Management, Economic Engineering in Agriculture and Rural Development. 2017, Vol. 17.
3. Alexandrov E. The concentration of the chemical compounds and the color of berry to the varieties of the interspecific hybrids to the vines (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.). In: Scientific Papers Series Management, Economic in Agriculture and Rural Development, 2016,Vol. 16, Issue 1. p. 53-56.
4. Alexandrov E. Interspecific hybrids of vines (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.) with increased resistance to biotic and abiotic factors. In: Scientific Papers Series Management, Economic in Agriculture and Rural Development, 2016, Vol. 16, Issue 1. p. 39-44.
5. Alexandrov E. New requirements in the creation of varieties of vine with the economic and ecological effect in the conditions of climate change. In: Scientific Papers Series Management, Economic in Agriculture and Rural Development, 2015, Vol. 15, Issue 3. p. 35-42.
6. Alexandrov E. Hibrizii distanţi ai viţei de vie (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.). Aspecte biomorfologice şi uvologice. Chişinău, 2012,140 p.
7. Alexandrov E. Crearea hibrizilor interspecifici de vita-de-vie (*Vitis vinifera L. x Muscadinia rotundifolia Michx.*). Autoreferat al tezei de doctor habilitat in stiinte biologice. Chisinau, 2017, 45 p.
8. Gaina B., Alexandrov E. Pagini din istoria si actualitatea viticulturii. Chisinău, 2015, 260 p.
9. Gaina B., Lafon-Lafourcade S., Dubos B. Incidence eonologique du traitment biologique de la vigne par *Trichoderma veride* a l`egard de la pour riture grise. In estitud d`Oenologiqic, Raport des activites de recherches, 1981-1983, p.75.
10. Gaina B., Pucheu-Plante B., Lafon-Lafourcade S. Insidene de develepment des moisissures sur le raisin a l`egard de la qalites des moutse des vins. In estitud d`Oenologiqic, Raport des activites de recherches, 1981-1983, p. 61.
11. Legea Republicii Moldova nr. 115 din 09.06.2005 cu privire la producţia agroalimentară ecologică. Monitorul Oficial Nr. 95-97 din 15.07.2005.
12. Programul naţional privind producţia agroalimentară ecologică. Hotărîrea Guvernului Republicii Moldova nr. 149 din 10.02.06. Monitorul Oficial nr. 031 din 24.02.06.
13. Strategia de mediu pentru anii 2014-2023 și Planul de acţiuni pentru implementarea acestuia. HG Rep. Moldova nr. 301 din 24.04.2014. În: Monitorul Oficial, nr. 104-109 din 06.05.2014.
14. Ampelograficeskii atlas sortov i form vinograda selekţii Naţionalinovo Naucinovo Ţentra «Instituta vinogradarstva i vinodelia im. V.E. Tairova». Kiev, 2014. 138 p.
15. Vinograd. Avt.. kol. Vlasov V.V., Muliukina N.A., Zelenianskaia N.G. i dr. Odesa, 2018, 616 p.
16. Gaina B. Anologhia i biotehnologhia produktov pererabotki vinograda. Kişinev, 1990, 167 p.
17. Nedov P. Immunitet vinograda к filoksere i vozbuditeliam gnienia kornei. Kişinev,1977, 169 p.
18. Nedov P. Filloksernaia problema i selekţia vinograda na kompleksnîi imunitet к vrediteliam i bolezniam. Ghenetika i selekţia vinograda na immunitet. Trudî Vsesoiuznovo Simpoziuma. Kiev, 1978 p. 35-45.
19. Nedov P., Guler P. Normalinaia i patologhiceskaia anatomia kornei vinograda. Kişineav, 1987, 151 p.
20. Ungureanu P. Osnovî vinodelia Moldovî. Trudî MNIIÎViV, Tom 5. Kişinev, 1960, 296 p.
21. Prinţ Ia. Vinogradnaia filloksera i merî boribî s nei. Moskva, 1965, 294 p.
22. Ănţiklopedia vinogradarstva. V 3-h tomah. Kişinev. 1986-1987.