Recent Studies on Strawberry Breeding in Türkiye

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Abstract

Scientific research highlights the benefits of berries for human health, and increases the demand of consumption and production all over the world including Turkey. For this reason, berry breeders have started to work on the development of new varieties that are especially important for human health and nutrition. For this purpose, within the scope of the European Union project called "Breeding Value", which includes 8 different countries and 20 participants under the coordination of Professor Bruno Mezzetti, Turkey was involved in this project and the nutritional contents of local berries were examined, as well as their resistance to some biotic and abiotic stress factors. On the other hand, our country has participated in breeding studies to develop varieties resistant to gray mold disease, which causes significant commercial losses in strawberries, and to determine QTLs related to the disease, within the scope of the "MedBerry" project, which is also supported by the European Union, under the coordination of Elena Baraldi, in which it is a part of 5 different European projects. In addition, development of new strawberry varieties tolerant to water stress by crossbreeding method and identification of candidate genes related to water stress", supported by national foundation TUBITAK (The Scientific and Technological Research Council of Türkiye) is continuing. Finally, in our project on the development of new varieties resistant to powdery mildew and gray mold in strawberries with different breeding strategies, were supported by the national TUBITAK with our international stakeholders Mezzetti and Baraldi, we will study classical hybridization and protoplast fusion techniques with the varieties determined within the scope of our MedBerry project has already finalized. In this article, the breeding studies we have done on strawberries in recent years are presented.

Keywords: Strawberry, breeding, botrytis, drought stress

INTRODUCTION

Strawberries (*Fragaria×ananassa* Duchesne ex Rozier) are enjoyable fruits and consumed by people of all ages and cultivated almost in all over the world. Recently its consumption and production trend increase year by year. The most important feature of strawberry is

that it is suitable for different growing systems due to its high adaptability to different climate and soil conditions. This allows us to consume it fresh or processed on our tables throughout the year. Strawberries can be produced throughout the year with applying different growing systems such as outdoor and indoor open-field, under protected, in soil and soilless conditions.
The main reasons for this are the scientific understanding of the importance of strawberries in terms of human health and nutrition, the fact that their fruits are loved and consumed by everyone due to their unique pleasant aroma, and the fact that they can be consumed in a wide variety of ways, such as jam, marmalade, ice cream, yoghurt and cake, in addition to fresh consumption. It has a market advantage, adapts to different climate and soil conditions, and brings high income per unit area. With attention focused on the effectiveness of naturally produced antioxidants, in recent years there has been intense interest in functional foods and especially nutraceuticals, that is, due to their basic nutritional properties as well as their health benefits. Strawberries are rich in polyphenolic compounds with antioxidant properties (Kafkas, 2017; Ayvaz Sönmez, 2022; Ürün, 2023).

Strawberry production in Türkiye expanded every year with the latest data indicating the last 10 years the production of 372.498 tons in 2013 and increased to 728.112 tons in 2022. Although the majority of strawberry production is done in the open, in recent years undercover and high polyethylene tunnels with Spanish type-high tunnels have become widespread. Recently, there has been an increasing trend towards soilless culture, as crop rotation, chemical fumigants are not effective against soil-borne diseases, and the ease of cultural processing, especially the convenience it provides in harvesting.

In Türkiye, the scientist Prof. Nurettin Kaşka at the University of Cukurova played an important role in the advancement and improving cultivation technologies of strawberry culture in Türkiye. As a result of a few cross-breeding programs a few Turkish cultivars were obtained and the first breeding program was started by Ataturk Horticultural Central Research Institute and 'Yalova-15', 'Yalova-104', 'Eren 77', 'Ata 77', 'Erenoğlu 77', 'Hilal 77', 'Dorukhan 77', 'Doruk 77', 'Bolverim 77' cultivars developed from this program by Dr. Onur Konarlı and Dr. Burhan Erenoğlu (Yılmaz, 2009). In the second cross-breeding programme, the scope of her doctoral thesis, and research assist. Ebru Kafkas made many hybridizations to transfer the aroma of the Ottoman strawberry variety to large, hard-fruited, high-yielding strawberry varieties and registered 3 strawberry varieties named Kaşka', 'Sevgi' and 'Ebru' by the Ministry of Agriculture. Then two more varieties were also registered as 'Seyhun' and 'Ceyhun' from Yaltır company and cooperation with Ebru Kafkas (University of Çukurova). "Ottoman" varieties were used in all breeding programs. However, since the "Ottoman" strawberry variety is morphologically male sterile, it was only used as the maternal parent in hybridizations. According to the obtained results, it was concluded that the fruit flesh of most of the hybrid individuals, especially the flavored ones, was soft and the firmness of the fruit flesh was mostly inherited from the main parent. For this reason, another breeding program was started with support from national and international institutions and organizations by the leadership of Ebru Kafkas.

Due to the climate changes experienced in recent years, effective and environmentally friendly management with biotic and abiotic stress factors is of great importance in strawberry cultivation, as in other fruit species. Developing varieties with high tolerance

to diseases and pests is one of the most important breeding purposes especially since strawberry fruits have soft flesh and are not suitable for washing with much water, and the pesticide residue applications affect human health negatively. When the pesticide is applied, the chemicals penetrate the fruit easily because of its soft and perishable fruit characteristics and has no fruit shell. Due to high sensitivity to fungal pathogens up to 11-12, fungicide applications can be necessary every year to secure production yield. This increases public concern for environmental food safety and makes urgent the need to develop sustainable Integrated Pest Management (IPM) alternatives. Strawberry major diseases are known as grey mold, anthracnose and powdery mildew caused by the fungal pathogens *Botrytis cinerea*, *Colletotrichum* spp. and *Podosphaera aphanis,* respectively. All over the world every year these pathogens affect serious product losses and applying agrochemicals cannot be phased out entirely soon. For this reason, to develop tolerant varieties applying new breeding strategies with new control methods has a vitally importance. For this reason, Prima MedBerry Project was carried out by the collaboration of Italy, Spain, France, Morocco and France. The project was to set up new genetic strategies to improve strawberry resistance to diseases and climate adaptation, maintain production of fresh high-quality fruit, and assess new molecular technologies to develop small molecules to directly control pathogens on plants with the limited need of agrochemical application and the discovery of cross-kingdom communication mechanisms between plant and pathogens, inspires the use of new molecular technologies (RNAi) for the development of small molecules alternative to agrochemical and active in pathogen control and adaptability of GMO-free biotech solutions. As a Turkish partner, our objective was to screen our germplasm strawberry genetic resources and to create a population crossing between tolerant and susceptible varieties to detect Quantitative Trait Loci (QTL) regions related to Botrytis cinerea resistance in leaf and fruit in strawberry. In total, 945 and 984 SNPs were mapped in the 'Fortuna' maternal and the 'Rubygem' paternal maps, respectively. Five significant QTLs are associated with resistance to *Botrytis cinerea* in the leaf in both parents. The QTLs linked to resistance to strawberry fruits were identified within a total of three LGs in different positions. The compressive genetic linkage maps constructed in 'Fortuna x Rubygem' population can be used in genetic and QTL studies for important agronomical traits. The recognized QTLs linked by Botrytis can be preferred for future strawberry breeding programs to carry out marker-assisted selection.

We were involved as a partner Breeding Value Project entitled "Pre-breeding strategies for obtaining new resilient and added value berries" supported by EU Horizon 2020 in 2019 under the coordination of Prof. Bruno Mezzetti. Totally 20 partners from 8 different countries interacted for the betterment of the future strawberry industry and the effective use and evaluation of genetic resources, to identify the most important resilience traits and fruit quality traits to develop studies on EU consumers' preferences; to apply Life Cycle Analyses to evaluate the ecological benefits, to generate a novel efficient data analyses strategy; to develop prototype visualization tools; and to disseminate and communicate the results to all beneficiaries interested in strawberries (Senger el al., 2022). As a Turkish partner, we are involved in almost all working packages providing our genetic material for allelic diversity and QTL studies, metabolomic analysis, panel and consumer tests, LCA analysis, evaluation of biotic and abiotic stress tolerance of our genetic resources based on their water deficient stress and botrytis tolerance degreed and postharvest and laboratory ring test studies on strawberries.

In Türkiye, recently, farmers started to complain about lower yields and reduced fruit

quality caused by water scarcity. Water stress is one of the main constrains of strawberry cultivation caused by the water scarcity in both soil and soilless culture. It is estimated that by the end of this century, the temperature in Mediterranean countries will increase about 4 - 5 °C and this will increase water scarcity and have a negative effect on berry yield, quality and protection management. Some agricultural management techniques were developed for solving this problem however all of these techniques are expensive and cannot bring a permanent solution for the problem. Thus, improving water stress-tolerant varieties is the best solution. Although there are limited studies about water stress in the world however there are no such studies in Turkey. Tolerant varieties can be achieved by traditional breeding methods like crossing and selection but it takes a long time. Nowadays classical breeding is improved by the use of marker-assisted breeding and genetic engineering tools which are more efficient and effective. With this project cultivar breeding program will be started related to deficit water stress tolerance on strawberry. Although there are many QTL studies on strawberry related to fruit quality and disease resistance. However, no QTL studies conducted related to drought stress or any abiotic stress studies on strawberries. In our ongoing European Union Horizon 2020 project, the susceptibility level of strawberry germplasm resources was identified. The highly tolerant and sensitive strawberry varieties were used in this project to develop highly tolerant varieties with high yields and desirable fruit quality by crossings. The obtained individuals will be screened based on the morphological and some physiological parameters under deficit water stress conditions. In the second year, according to the evaluation results of in vitro and in vivo water stress experiments, approximately 20-30 F1 lines will be selected and then vegetatively propagated and exposed to water deficit stress and evaluated based on some morphological and physiological parameters. Among them, 5-10 lines will be selected and an adaptation experiment will be done in two different locations. In the last step, only the DEGs (differentially expressed genes) in the QTL region will be selected and validated by combining QTL-seq and RNA-seq results.

The scope of this planned project, it is aimed to develop midway materials and strawberry varieties that are tolerant to gray mold (Botrytis cinerea) and powdery mildew and have high nutritional content, by following a multidisciplinary path in strawberry, which mainly includes morphological, metabolomic and genomic approaches. Within the scope of our PRIMA project (MedBerry), our strawberry variety candidates selected as having high tolerance to gray mold disease constitute the main materials of the ongoing TUBİTAK 1004 project. With this project, it was aimed to cross promising candidates of highly tolerant to botrytis and powdery mildew diseases and then select the tolerance status of the selected individuals using the methods we developed within the scope of our previous PRIMA (MedBerry) project. In addition, it was aimed to obtain new individuals that are tolerant to gray mold by combining protoplast cell sources of strawberry varieties and variety candidates with the electrofusion technique in in-vitro. In this context, the ploidy levels of the somatic hybrids that will be formed by plant regeneration from the calli obtained from the protoplasts derived from anthers to be cultured will be determined by flow cytometry. The variety or variety candidates to be developed by both classical hybridization breeding and protoplast fusion methodologies. Then putative lines will be propagated vegetatively and evaluated in terms of fruit yield, quality and disease tolerance in different ecologies (Adana, Erzurum and Tekirdağ).

With this proposed project, the first step will be taken in a strawberry breeding program, which has been carried out in limited numbers in the world, especially on protoplast

fusion, and will be implemented by us for the first time in our country. In addition, master's and doctoral students and postdoctoral researchers who will take part as scholars in our project will be provided with the opportunity to develop themselves, and by collaborating with foreign researchers who are experts in their fields from abroad. Strawberry breeders, producers and seedling producers, as well as scientists working on this subject, will be able to benefit from these results.

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