CHAPTERS ON VITICULTURE

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CONTENTS

PREFACE

In Turkey, well-experienced researchers have been studying on the different subjects of the science of grapevine. So far, a book has not been written with chapters in the English language by grapevine researchers. Therefore, a book project was undertaken with fruitful collaboration. This book consists of research and reviews on grapevines based on the relevant scientific literature on grape breeding, physiological responses to drought, phenolic behavior interaction with external applications, in vitro approaches, organic and nutritional aspects, wine quality, and morpho-anatomical structure of female gametophyte turn into the berry.

We are greatly indebted to all authors for their scientific efforts and professionalism, and finally, many thanks to the publisher IKSAD to make the chapters into a book.

EDITORS

CHAPTER 1

A GENERAL OVERVIEW OF GRAPE BREEDING IN CONSIDERATION OF LITERATURE FROM PAST TO PRESENT

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INTRODUCTION

Although there are many types of *Vitis* in the world, especially *V.vinifera* has a special place. The high fruit quality and rich genetic diversity of *V.vinifera* make this species suitable (Alleweldt and Possingham, 1988; Myles et al., 2011; Zhou et al., 2017) for the wine, fruit juice, and table grape industry making it indispensable (Burger et al., 2009; Wolkovich et al., 2018). On the other hand, its sensitivity to some biotic (for example phylloxera, powdery mildew) and abiotic (for example cold damage) factors is among the disadvantages of this species (Alleweldt and Possingham, 1988).

Widely cultivated in Europe, *V.vinifera* faced the devastating destruction of diseases such as powdery mildew and downy mildew, first by phylloxera in the 1870s. In this respect, action had to be taken especially against phylloxera and powdery mildew. This necessity led to research on *Vitis* species that could be tolerant to these factors and to enter a new era in grape breeding (Galet, 1998). Since this period, breeding programs have been initiated for rootstock breeding and the development of disease-tolerant wine and table grape varieties. In these programs; *V.berlandieri*, *V.riparia*, *V.rupestris*, *V.cinerea*, *V. amurensis*, *V.labrusca*, *V.aestivalis*, *V.smalliana*, *V.caribaea*, and *V.shuttleworthii* species were used (Alleweldt and Possingham,1988; Alleweldt et al., 1991). Of these species; especially in *V.rupestris, V.riparia*, and *V.berlandieri* rootstock breeding (Alleweldt and Possingham, 1988; Alleweldt et al., 1991). *V.amurensis* and *V.riparia* have generally been used for resistance to cold and diseases, and *V.caribaea, V.smalliana* and *V. shuttleworthii* have been used in the development of cultivars that can adapt to tropical climate conditions (Camargo, 2000; Burger et al., 2009). Since the middle of the nineteenth century, it has been used to provide tolerance to various biotic and abiotic stress factors in species such as *V.pseudureticulata*, *V.balanseana*, *V.piasezkii*, (Miao et al., 1999; Zhang ve et al., 2009) *V.adstricta* (Fan et al., 2007), *V.rotundifolia*, (Wells et al., 1987; Halbrooks and Mortensen, 1989), *Vitis popenoei* Fennell (Olien, 1990; Conner, 2010) ve *Vitis arizonica* Engelm (Riaz et al., 2009). Although it is reported that the attempts for grape breeding started in the Middle Ages (Boursiquot et al. 2004), it is reported that conscious studies have been carried out since the end of the nineteenth century (Burger et al., 2009). In this study, a brief summary of the studies on vine breeding in the USA, China, France, Germany, Italy, Hungary, Spain, and the Czechsia from the end of the eighteenth century to the present is presented in the light of the literature.

United States of America

Breeding studies in the east of the USA started towards the end of the 1880s. Cultivars of *V.labrusca* and *V.vinifera* species were used as breeding materials during these periods. The first breeding efforts were aimed at improving cultivars that are cold and disease tolerant, have a high shelf life, are productive, have large berries, and have a muscat flavor. In the following years, seedless cultivar breeding was also included in these programs. (Reynolds and Reisch, 2015). Breeding studies carried out in this period began to bear their first fruits in the late 1920s. Golden Muscat and Fredonia were introduced in the 1928s. Subsequently, many varieties such as Concord seedless, Bronx seedless, Westfield, Watkins were improved. Over the next 20 years, seedless grapes such as Interlaken Seedless, Himrod, Romulus, Lakemont were introduced. Breeding studies continued with cultivar breeding programs with high fruit quality, disease, and cold tolerance by using French-American hybrids with *V.vinifera*. As a result of these studies; Cultivars such as Cayuga White, Traminette, Geneva Red, and Aromella have been introduced. In the following years, *V.rupestris* and *V.cinerea* species were included in breeding programs and focused on improving disease-tolerant varieties with high fruit quality that would minimize the use of fungicides. On the other hand, studies on seedless cultivar breeding continued and many seedless cultivars such as Marquis and Remaily Seedless were introduced to the market after the 1980s. Breeding studies have also included breeding techniques at the molecular level (Reynolds and Reisch, 2015). In studies at the molecular level, it was desired to introduce disease tolerance to *V.vinifera* by gene transfer (with the chitinase-generating gene). In such studies, some successes have been achieved with a Chancellor-based system in cultivars such as Merlot and Chardonnay (Kikkert et al., 1997; Kikkert et al., 2009). Another program investigated disease resistance in Chardonnay using brief proteins define as anti-microbial peptides (AMPs) (Vidal et al. 2006; Rosenfield et al. 2010). Such studies continued with the feature of molecular markers to determine qualified genes conferring tolerance to powdery mildew and black root (Dalbó et al., 2001; Lodhi et al., 1997). Molecular marker-based maps are also used to gear-up breeding studies in grapes (Reisch et al., 2014). Studies on molecular marker-based genome mapping are still continuing today (Reynolds and Reisch, 2015).

The breeding efforts initiated in the eastern part of the USA and the breeding activities in the Upper Midwest cover almost the same period, and it is reported that grape breeding began in the 1870s in these regions. During these periods, studies were carried out to obtain cold-tolerant cultivars, and it is reported that the pioneer of these studies was Louis Suelter (Pfaender, 1912). Louis Suelter obtained two cultivars named Suelter and Beta among the genotypes he improved from *V.riparia* x Concord crossing. Breeding studies continued at the University of Minnesota Horticulture Research Center (formerly: Fruit Breeding Farm) in 1908 (Snyder, 1982). In the following years, breeding programs were started in regions such as Nebraska, South and North Dakota, Missouri, and Illinois (Hemstad and Breeder, 2015). In the early periods, cold tolerant cultivars were tried to be improved by using combinations of Beta and *V.labruscana*. In these studies, a disease-tolerant variety called Blubell, which is used in making fruit juice and jelly, came to the fore (Wilcox, 1946). As a result of the studies of Elmer Swenson at the University of Minnesota in the 1970s, varieties named Swenson Red and Edelweiss were improved (Swenson et al., 1980). The University of Minnesota has increased grape genetic resources and studies on grape breeding since 1984 (Hemstad and Luby, 2000). Since this period, breeding programs have intensified to improve cultivars that are tolerant to cold and diseases and have high wine quality. From these studies, cultivars such as Frontenac, Frontenac Gris, La Crescent were introduced towards the end of the 1990s (Hemstad and Luby, 2003). Moreover, cultivars named Marquette, Somerset Seedless, Little friend, Petite Pearl were registered towards the end of the 2000s. Along with the applied breeding studies carried out at the University of Minnesota, marker-assisted selection methods are also applied today (Hemstad and Breeder, 2015). Vine breeding efforts in the Upper Midwest have been reported to have significant impacts on the regional grape and wine industry. Against the winter cold in this region; With the contributions of Louis Suelter and Elmer Swenson, Niels Hansen, and the University of Minnesota, many varieties that are tolerant to winter cold, have high disease resistance, and are suitable for winemaking have been improved. These improved grape cultivars are also grown by producers in different regions of the world where viticulture is made (Hemstad and Breeder, 2015).

Southern regions also play an important role in the USA's attempts to improve grapevine breeding. These regions; Although it is challenging for viticulture in terms of biotic and abiotic stress factors, *Vitis* also has gene resources that can cope with these factors. Vine breeding in the southern regions began in the early twentieth century. Early breeding efforts were particularly focused on developing grape cultivars resistant to Pierce's disease (PD) (*Xyllela fastidiosa* Wells et al.), phylloxera (*Daktulosphaira vitifoliae* Fitch), and downy mildew. *V.vinifera* and many hybrid cultivars cannot be grown in the gulf region due to Pierce's disease. In this respect, breeding PD-tolerant cultivars have been a sustained effort in the South. *V.rotundifolia*, *V.arizonica* (Riaz et al., 2009), *V.shuttleworthii* (Mortensen and Stover, 1990) species were used as a source of resistance against this disease. *V.aestivalis* Michx (Einset and Pratt, 1975; Mortensen and Stover, 1990), *V.riparia* Michx (Cousins, 2005), and *V.labrusca* species were used in cultivar breeding (Mortensen and Stover, 1990). In rootstock breeding, it is more; *V.×champinii* Planch for root-knot resistance (nematode), *V.riparia* Michx, *V.rupestris* Scheele (Cousins, 2005), *V.×doaniana* Munson ex Viala (Loomis and Lider, 1971), *V.shuttleworthii* (Mortensen et al. Stover, 1990) and *Vitis popenoei* Fennell (Olien, 1990; Conner, 2010). Apart from these species; *V.vulpina* L, *V.acerifolia* Raf., *V. mustangensis* Buckley, *V.monticola* Buckley, and *V.palmata* Vahl have been utilized in limited breeding programs (Stafne et al., 2015). Although there are many *Vitis* species in the southern regions of America, especially *V.rotundiflolia* species in the Muscadinia genus; It has a different place in terms of insect and disease tolerance, soil adaptation, berry cracking resistance, and high nutraceutical composition (Goldy and Onokpise, 2001; Andersen et al., 2013). Genotypes of Muscadine species gained importance in the early 1900s, with the initiation of studies to increase fruit quality and disease resistance in breeding programs in Carolina and North Georgia (Stafne et al., 2015). On the other hand, after the realization that crosses between local grape genotypes and cultivars of *V.vinifera* species could occur, table grape cultivation became an important undertaking in many parts of the south in the 1800s and early 1900s. During this period, new viticulture plantations were established and *V.labrusca* cultivars and some hybrid cultivars were grown in the majority of these areas (Hedrick et al., 1908). Interspecies breeding studies began in the 1800s and continued with the breeding use of local genotypes by both public and private breeders in the early 1900s (Lane, 1997). Since the 1930s, breeding programs have been initiated by some universities in North and South Carolina, Louisiana, Texas, Oklahoma, Florida, Georgia, Mississippi, Arkansas, Alabama, and Virginia (Stafne et al., 2015). In these regions; Breeding programs were focused on improving grape cultivars and seedless varieties for the wine and juice industry (Moore, 1969; Moore et al., 2011). Moreover, In these programs; Resistance to PD included a place on topics such as productivity, fruit cracking, homogeneous ripening, self-fertility, cultivar improvement suitable for mechanization (Halbrooks and Mortensen, 1989), reducing berry cracking, increasing berry size, and increasing fruit sweetness (Stafne, 2006; Conner, 2010). As a result of these studies, many varieties registered in the southern regions have been introduced to the market. Century I (Oberle, 1974), Blanc du bois (Mortensen, 1987), Foxxy Lottie (Clark, 1997; Moore et al., 2011), Southern Home, Black Beauty (Mortensen et al., 1994), Victoria Red (Moore et al., 2011) and RazzMatazz are among these cultivars. Breeding activities aimed at cultivating genotypes in the southern regions of the USA continue, especially on Muscadins. It has been reported that the development of wine and grape cultivars resistant to PD will make significant contributions to the region's viticulture, although it is difficult for consumers to adopt varieties that do not belong to the *V.vinifera* species (Stafne et al., 2015).

Another region where important studies on grape breeding are carried out in the USA is the west of the country. Breeding studies in the West date back to the 1880s. In this region, California is especially important in terms of viticulture. The University of California-Davis (UC Davis) and Fresno, (US Department of Agriculture/USDA) facility) have made significant advances in vine breeding (Lund, 2015). Apart from these institutions, some private companies have also started studies on grape breeding since the 1980s (Caputi, 2000; Lund, 2015). Studies on drought and salt stress, disease (powdery mildew), and pests (phylloxera, nematodes, PD) tolerant table and wine cultivars breeding have been focused on in the western United States (Wade, 2011; Ferris et al., 2012; Sommer, 2012; Lund, 2015). At UC Davis, Dr. Olmo, Dr. Walker, and Dr. Carole Meredith have had great success in vine breeding.

For example; Dr. He improved the O39-16 rootstock, which was successful in controlling the Olmo fanleaf virus and the dagger nematode that spreads it (Xiphinema index). As a matter of fact, it has been reported that this rootstock is the only rootstock that can be used easily in places where both fanleaf virus and X.index are present (Walker et al., 1991). Dr. Walker improved 5 rootstocks. These rootstocks are numbered from 1 to 5 with the abbreviation GRN. It is reported that these rootstocks are improved from a complex hybridization combination of many species (Ferris et al., 2012, Lund, 2015). Another researcher, Dr. Her work in molecular and genetics at Carole Meredith laid the groundwork for many of the current advances in grapevine farming. (Bowers and Meredith, 1997; Doligez et al. 2002; Riaz et al. 2004).

USDA, another institution that has achieved significant success in grape cultivation in the western regions of the USA, has been carrying out breeding studies since the 1930s. This institution has made progress in the development of table and dried cultivars. As a matter of fact, it is reported that table grape varieties developed in the USDA cover almost half of the viticulture areas in California. (California Department of Food and Agriculture, 2014b). One of the important studies conducted by this institution is the embryo rescue technique for seedless grape cultivation led by Dr. Ramming (Emershad and Ramming, 1984; Ramming, 1990). Breeding studies to obtain varieties that are especially tolerant to PD and powdery mildew are important in USDA (Walker et al., 2012). In the Western United States today, issues such as drought, pest growth, and reduction of pesticide use come to the fore. In this respect, breeding programs aimed at improving cultivars that are adaptable to drought and hot climates (Sommer, 2012) and tolerant to diseases and pests in a way that reduces pesticide use (Wade, 2011; Sommer, 2012) continue to maintain their importance (Lund, 2015).

China

It is one of the gene centers of the Chinese *Vitis* species. It is stated that there has been a viticulture culture in China for more than 2000 years (Sun, 1979). Since 1980, China has improved great momentum in establishing new plantations for viticulture. In addition to national and regional research institutes that include national grape conservation, grape genetics, and breeding programs in China, more than 20 universities and colleges conduct training and research on grapevine breeding (Lu and Liu, 2015).

It has been reported that studies on the research and collection of wild *Vitis* species in China started in the mid-1950s (Yu, 1994; Zhou and Guo, 1995; Kong, 2004). Initially, it was concentrated on the *V.amurensis* Rupr. species in Tibet and Xinjiang regions (Zhou and Fang, 1986). In these studies, *V.piasezkii* was discovered in the Xinjiang region (Lin, 1998).It is stated that there are 37 species, 1 subspecies, and 10 varieties belonging to the *Vitaceae* family in China (Li, 1998). Vine breeding began with clone selection in the 1950s. In selection studies; many outstanding clones of *V.amurensis* Rupr., *Vitis quinquangularis* Rehd, and *Vitis davidii* have been identified (Lu and Liu, 2015). To these clone; examples include *V.davidii* Tangwei (Zhang and Fan, 1985) and Xuefeng (Zhang et al., 1989) (*Vitis davidii*), Changbai No. 11 (Shuangqing) (Lin, 1982), Mushan No.1 (Shan ve et al., 2011), Shuangyou and Changbai No. 4, (*Vitis amurensis* Rupr), GSH-2, ZHJ-5, Zhonggu No.2 , Zhongjiu No.5 (Huang et al., 2003), Yeniang No. 2 (*Vitis quinquangularis* Rehd.) (Lu ve Liu, 2015). New vineyard have been established in different parts of the country with clones selected from these species (Lin, 1991; Zhu et al., 2006). Crossbreeding, especially in northern China, was initially aimed at improving cultivars tolerant of winter cold. In this respect, *V.amurensis*, which can survive down to −40°C in northern China, has been included in breeding studies as a valuable resource (Hu, 1956). In improving cold tolerant cultivars, *V.amurensis* x *V.vinifera*/*V.labrusca* combinations were also considered (He et al., 1990; (Fang et al., 1993). In the studies carried out, Heishan and Shanmeigui (Yang and Wu, 1959; Pu, 1960), Gongniang No. 1 and Gongniang No. 2 (He et al., 1990), Shuanghong (Song and Li, 1998), Zuohongyi (Song et al., 2005), and such Beichun (Fan et al., 2010) many cultivars that are resistant to winter cold have been improved. In addition to cold tolerance, studies have also been carried out on the breeding of early maturing table and wine cultivars that are tolerant to fungal diseases, colored, coarse-berry, and muscat-flavored. In these studies, apart from *V.vinifera* x *V.quinquangularis*, *V.amurensis* x *V. Vinifera* combinations, Muscat Hamburg and Kyoho, Paerl of Csaba, Queen of the Vineyard, Black Monukka and Thompson Seedless varieties were also used (Lu and Liu, 2015). In addition, hybridization between *V.vinifera* and *V.adstricta* have been made for the fruit juice industry (Fan et al., 2006). In the studies carried out; Zuijinxiang, Xiangyue, Tianfeng (He et al., 1989), Fenghou (Xu et al., 2000), Fenghuang No. 51, Jumeigui (Wang et al., 2003), Jingxiu (Yang et al., 2003), Beichun (Kong, 2004), Beizi, Beixiang and Beifeng (Fan et al., 2006), Lingyou (Huang et al., 2006), Bixiang, (Li et al., 2008), Hupei No. 1 ve Hupei No. (Jiang et al., 2008) Xiazhihong (Liu et al., 2011), Xuanlanhong (Song et al., 2012), Yueguang Wuhe, and Xiaguang (Tao et al., 2012) such cultivar were improved. In China, studies on rootstock breeding started in the mid-1980s. The vines are mostly grown on their own roots, as there is no phylloxera concern in China. However, in recent years, breeding studies have been started to cover both phylloxera and abiotic stress conditions. Huajia No.8 rootstock is the first rootstock improved in China and has been used in limited quantities in the southern regions of China (Li and Jin, 1999). Beta is used more in the northern regions. On the other hand, *Vitis davidii* Tangwei, *Vitis balanseana* (Miao et al., 1999) and *V.piasezkii* species were also used as partial rootstocks (Zhang et al., 2009). In recent years, molecular breeding strategies have been used in breeding studies to integrate *Vitis* species and high-quality cultivars of *V.vinifera* in China (Lu and Liu, 2015).

France

Beginning in the 1870s, viticulture in Europe faced three major devastating crises. First, phylloxera appeared then powdery mildew and downy mildew diseases. In the face of these factors threatening European viticulture, French breeders started breeding programs to improve rootstock and grape cultivars that are tolerant to phylloxera pests, powdery mildew, and downy mildew diseases. For this purpose, more than 10 thousand hectares of French-American crossbreeds were planted in Ontario and New York regions, and France in the late nineteenth century (Galet, 1998). In breeding studies in France; *V.vinifera*, *V.labruscana*, *V.cinerea*, *V.aestivalis*, *V.rupestris*, *V.lincecumii*, *V.riparia*, *V.amurensis* (used by Eugene Kuhlman), species were used and combined. In breeding programs in the country; famous breeders such as Georges Couderc, Albert Seibel, Francois Baco, Eugene Kuhlman, Bertille Seyve, and Ferdinand Gaillard worked (Reynolds, 2015). Many cultivars have been improved by these breeders, including Chambourcin Baco noir, Leon Millot, Marechal Foch, Aurore, Vignoles, Rosette, Rougeon, De Chaunac, Villard noir, Chelois, Chancellor Seyval blanc, Villard blanc, Vidal blanc, and Cascade are some of them (Reynolds, 2015). Breeding studies that started in France in the 1870s resulted in the improved thousands of hybrid individuals by the 1950s. In studies on rootstock breeding, by Georges Couderc; widely used rootstocks such as 3306 C, 3309 C, and 1613 C were obtained (Reynolds, 2015). Many of the cultivars improved in breeding studies have been discontinued since 1960s. Since these years, breeding programs have been started for clone selection in cultivars belonging to the *V.vinifera* species. With the breeding studies that started in France, many valuable breeding materials have been carried to the present day, causing a rich gene pool to be created. French hybrid varieties improved as a result of breeding studies were used as breeding materials in breeding studies in the second half of the twentieth century. Studies on grapevine breeding and genetic research are continuing in France (Reynolds, 2015).

Germany

The basis of breeding studies in Germany was initially based on the struggle to viral diseases (Ruehl et al., 2015). Studies started with clone selection in 1876. Previously, the mass selection method (Laufner, 1987), and then the single vine selection method was applied (Hofäcker, 2004). Until 1950 in Germany, the majority of vineyards were constituted of genotypes selected as a result of clone selection (Ruehl et al., 2015). Indexing for virus agents seen in vineyards was started in 1970. Analyzes were made with serological methods such as ELISA in the 1980s. In the early 1990s, all selected German clones were tested for the virus. Since 2013, tests on this subject are carried out according to the European Union Legislation (Schöffling and Stellmach, 1993). Clone selection; cultivars such as Silvaner, Riesling, Müller-Thurgau, Pinot noir, Pinot blanc and Pinot gris were also practised (Konradi et al., 2003; Blaich et al., 2007; Schmid et al., 2009). Clonal selections contributed to genetic diversity by enabling the selection of different clones in traditionally grown local varieties, and also had positive effects on plant health in most vineyards (Ruehl et al., 2015).

Breeding studies in Germany date back to the nineteenth century (Töpfer, 2008). Improving phylloxera-resistant cultivars were among the important goals in the initial breeding studies. After it is known that American rootstocks are resistant to this pest, more research has been started on yield and quality and disease-resistant vine breeding. In these studies, cultivars such as Siegerrebe, Scheurebe, Bacchus Morio Muskat, Kerner, Ehrenfelser and Dornfelder were improved (Ruehl et al., 2015). Since 1925, breeding studies have started to improve disease-tolerant cultivars. As a result of the studies on resistance breeding, many cultivars such as Accent, Regent, Phoenix, Cabertin, Villaris, and Cabernet Cortis, which are tolerant to powdery mildew and downy mildew, have been approved and released (Ruehl et al., 2015). These cultivars are outside of Germany; It is grown for use in winemaking in countries such as Norway, Poland, England, Ireland, Denmark, the Czechsia, Sweden, and the Netherlands. Studies on rootstock breeding have also been carried out in Germany. It has been reported that rootstock breeding has been started since the 1880s. In rootstock breeding studies, *V.riparia* seeds were used initially (Schmid et al., 2009), then crosses were made between American species and *V.vinifera* cultivars. In the following years, hybridizations were made between *V.riparia, V.berlandieri* and *V.rupestris* species (Manty, 2005). As a result of the studies, rootstocks such as 13 Gm, SO4, and Börner were improved. Considering the changing climatic conditions in Germany, studies on breeding rootstocks that are tolerant to abiotic stress factors such as drought and chlorosis are also highlighted. Molecular marker techniques are also used in breeding studies (Ruehl et al., 2015).

Italy

It is reported that grape breeding programs have been actively carried out in Italy since the late 1800s (Bavaresco et al., 2015). After the emergence of phylloxera in Italy in 1879, primarily rootstock breeding studies have started (Fregoni and Bavaresco, 1986; Di Lorenzo and Sottile, 2000). In rootstock breeding programs; The subjects of resistance to phylloxera, tolerance to lime and drought, and adaptation are emphasized. Famous breeders such as Federico Paulsen, Antonio Ruggeri, Italo Cosmo, and Alberto Pirovano have worked in rootstock breeding. In these programs; *V.berlandieri*×*V.riparia*, *V.berlandieri*× *V.rupestris, V.riparia*×*V.rupestris* species were combined. Some *Rupestris* du Lot hybrids have also been used to improve rootstocks, though in part. In these studies; rootstocks such as 779 P, 775 P and 1103 P, 240, 225, 300, 140 Ru, Cosmo 2 and Cosmo 10, Golia, Gagliardo, Star 50 and Star 74, and M series (MI, M2, M3, and M4) were improved (Bavaresco et al., 2015). In addition to rootstock breeding, studies on improving table and wine cultivars were also started in Italy in the 1875s. Cultivars belonging to the *V.vinifera* species were generally used in cultivar breeding, as well as some Hungarian (Cipriani et al., 2010) and Italian cultivars (Schneider et al., 2001; Mannini et al., 2010). Breeding programs in addition comprised clonal selection in the 1970s. Selection studies were initially based on the selection of productive clones. Afterward, qualitative characteristics began to take place among the selection criteria. Most of the cultivars improved in grape breeding programs were table grapes (Bavaresco et al., 2015). In Italy; rootstock breeding programs tolerant to drought, active limestone, salinity, and iron chlorosis continue and studies are carried out on the improvement of physiological and molecular markers in these programs. On the other hand, breeding programs are continuing to improve new grape cultivars resistant to powdery mildew and downy mildew (Bavaresco et al., 2015). In breeding studies, genetic characterization studies have been carried out with SSR and single nucleotide polymorphism markers to constitute a germplasm collection since the 1990s (Emanuelli et al., 2013). Breeding programs have been initiated to improve powdery mildew-tolerant grape cultivars by bringing resistant varieties from Hungary, Russia, Germany, the Czechsia, Serbia, and Switzerland. In breeding studies, molecular markers are used in the early selection of goal-oriented individuals (Bavaresco et al., 2015).

Hungary

Breeding studies in the country started in 1863. In Hungary, on the one hand, new table, wine, and rootstock cultivars were improved by hybridization, on the other hand, clone selections were made in old genotypes (Hajdu, 2015). After the phylloxera pest was detected in the country in 1875, especially Zsigmond Teleki took seeds from France and started to work on rootstock breeding (Bakonyi and Kocsis, 2004; Hajdu and Bakonyi, 2006; Schmid et al., 2009). In Teleki rootstock selection; he focused on phylloxera, lime, vegetative growth, and affinity (Hegedűs et al., 1966). As a result of these studies; rootstocks 5BB,125 AA SO4, 8B,5C and10 A were improved (Csepregi and Zilai, 1955). Since 1970, a new era has started in rootstock breeding with Károly Bakonyi at the Keszthely Faculty of Agriculture (Hajdu, 2015). In this period, a variety collection consisting of domestic and foreign rootstocks was established (Bakonyi et al., 1996; 1997). As a result of Bakonyi's studies on rootstock breeding; Rootstocks named Georgikon 28, Georgikon 61, Georgikon 46, Georgikon 251,Georgikon 103, and were improved (Bakonyi and Kocsis, 2006).

Grape cultivars collections have been established in the country since the late 1800s. By the end of the 19th century, individual breeders started breeding studies to develop new table grape cultivars. In 1901, in Kosice, József Mathiasz first improved a table cultivar, Darányi Ignác Muskotály. In the following years, Csaba gyöngye, an early maturing muscat flavored hybrid, was obtained by Adolf Stark (Hajdu, 2015). In 1915, Kocsis Pál started breeding studies in Kecskemét in order to obtain table grape cultivars that are drought resistant and suitable for growing in sandy soils. Many cultivars were improved in these studies. It has been reported that the most prominent variety among these hybrids is Irsai Olivér (Füri, 1977; Hajdu, 2012).

In Hungary, breeding studies for the production of table grape cultivars started in the early 1960s with the program and financial assistance of the Ministry of Agriculture. Initially, *V.vinifera* L. cultivars were used in these studies. Kozma Pálné Muskotály and Helikon Szépe are cultivars improved from these studies (Szegedi, 1968). In Hungary, the resistance breeding program was considered another high-priority program. It has been reported that French-American cultivars (Seyve Villard 12-375) and some Asian species (*V.amurensis* Rupr.) have been used for resistance breeding (Kristen, 1990; Kozma, 2002). Breeders began to improve resistant varieties in the 1950s. In this context, it was stated that the first hybrids improved and showed good development in terms of resistive to fungi diseases (powdery mildew, downy mildew, botrytis) and winter's frosts. Examples of this cultivars include Fanny, Esther, Lidi, Palatina, Pölöskei Muskotály (Kozma, 1961), Pegazus, Csépi Muskotály and Borostyán (Hajdu, 2015). Breeding programs have also been initiated to improve wine grape cultivars in Hungary. These programs first started with clone selections (Hajdu, 2002). Crossbreeding studies have also been carried out to improve wine cultivars. In these studies, it was desired to increase the quality of Eurasian genotypes, protect production safety, and obtain new varieties by crossing them with other Eurasian genotypes. Another goal is to improve resistant cultivars by using several *Vitis* species (Hajdu, 2015). A number of studies have also been carried out on Agrobacterium in resistance breeding. For example; Some breeders at the University of Horticulture, Budapest, have carried out a number of studies on the resistance of Agrobacterium, among them *V.amurensis* and various East Asian species. In these studies, they found that *V.amurensis* and some genotypes were resistant to various strains of this disease (Korbuly, 2002). On the other hand, in breeding programs initiated in the recent past, issues such as resistance to black rot, tolerance to cold, tolerance to lime and drought, and earliness are emphasized in order to improve new rootstocks and cultivars (Hajdu, 2015).

Spain

Vine clone selection studies in Spain began in the 1970s on genotypes belonging to the genus *V.vinifera* in the Catalonia and La Rioja regions. The main purpose of the initial selections was to distinguish between healthy and productive clones. In the following years, parameters such as sugar content and Ph were also taken into account. The first selection studies started in the Tempranillo cultivar (Renedo et al. 1995; Martínez et al., 2006). In Spain, after the 1980s, clone selection programs were started in different regions of the country. In these programs; It was desired to improve healthy and high quality clones with high phenolic content, loose clusters, small berry size, homogeneous ripening, tolerant to Botrytis, suitable for the Spanish climate. As a result of the studies, many valuable clones were selected (Ibáñez et al., 2015). The first *vinifera* clone (Xarello I-20) was approved in 1987 in Spain. A total of 638 clones have been reported so far, 108 of which are *V.vinifera* (OEVV-MAGRAMA, 2013). Almost all of the *vinifera* certified clones in Spain are wine cultivars. Most clones were selected from Garnacha Tinta, Tempranillo (Tempranillo CL306 is common), Palomino Fino and Cabernet Sauvignon cultivars. In selection studies, some attempts have been made to select suitable rootstocks for grape growers. There are a number of rootstock clones selected from rootstocks such as 110 R, 41B, SO4 and 161-49 C (Ibáñez et al., 2015).

It is reported that studies on vine breeding started in Spain in 1940. The first breeding program aimed to improve new wine cultivars by using some rootstocks and local genotypes as parents. It has been stated that the Redora cultivar was improved as a result of breeding studies carried out during these periods (Ibáñez et al., 2015). A breeding program was also initiated in 2003 to develop varieties that are tolerant to fungal diseases, adapt to the local ecology and have good wine quality. In this program; Merlot, Palomino Fino, Alicante Henri Bouschet, Syrah, Regent and Tempranillo cultivars were used. Some hybrids from this study (eg Palomino×Regent) were noted to be tolerant to fungal diseases. In the same year, in cooperation with IMIDA (Instituto Murciano de Investigación y Desarrollo Agrario y Alimentario) and ITUM (Investigación y Tecnología de Uva de Mesa); A breeding program was initiated to develop seedless cultivars that are productive and have high market value, and new cultivars that are tolerant to fungal diseases such as powdery mildew and downy mildew. In this program, cultivars such as Autumn Royal, Crimson Seedless, Felicia, and Gf.Ga-52-42 were used as parents (Ibáñez et al., 2015). Another breeding program using Flame Seedless, Alexandria Muscat, and Italia cultivars was started in 2007 (Carreño et al., 2009). Breeding programs in Spain continue with the cooperation of public institutions and some private companies. Among the breeding programs, efforts to improve productive and high-quality cultivars resistant to fungal diseases are among the priority targets. German wine grape cultivars (Pilzwidestandsfähige Rebsorten – PIWI) are particularly interested in resistance breeding. There is also an incentive to use hybrids from crosses between other tolerant *Vitis* species or genotypes. Molecular markers are used in selection programs to help shorten the selection procedure for seedlessness, powdery mildew, and mildew tolerance (Ibáñez et al., 2015).

Czechia

Studies on grapevine breeding in the Czechia started in the 1950s. The first breeding program; was tended at improving productive cultivars for white, colored, and aromatic wines. For this purpose, cultivars such as Gewürztraminer, Muscat Ottonel and Hárslevelű, Müller Thurgau and Prachttraube were used as parents (Pavloušek, 2015). New breeding programs were initiated in the 1960s with the main target of improving quality wine grape cultivars. By using cultivars such as Blaufränkisch and Saint Laurent as parents in these programs, was the goal to improve cultivars suitable for producing attractive and full-bodied wines with ideal tannin structure, which can adapt to the climatic factors of the Czechia (Pavloušek, 2015).

In these years, breeding programs are also started for the breeding of cultivars tolerance of fungal (powdery and downy mildew) diseases. In resistance breeding, *V.vinifera*, *V.amurensis*, and some Seibel hybrids were combined. For example, the Rondo cultivar was improved as a result of these studies and later registered in Germany. New breeding programs for resistance (powdery and downy mildew) breeding were initiated in the 1980s, using a very large gene pool of interspecies hybrids brought from different countries (Pavloušek, 2015). Seibel 13666 variety has taken an important place in studies on resistance breeding in the Czechia due to its resistance to diseases (Galet, 1988). In breeding studies; Cultivars such as Muscat Moravsky, Florianka, Alibernet, Malverina, Cerason, Kofranka, Marlen, Neronet, and Rubinet were improved (VIVC, 2022). Today, most of the Czech vineyards; It has been reported to be constituted disease-resistant cultivars such as Hibernale, Solaris Johanniter, Malverina and Regent (Pavloušek, 2015).

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CHAPTER 2

HOW GRENACHE N (*Vitis vinifera* L.) PHYSIOLOGY RESPONSES DROUGHT STRESS IN EARLY DEVELOPMENTAL STAGES?

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INTRODUCTION

Grapevine and wine production are strongly influenced by climate (Alves et al., 2013). In the context of global climate change, vineyard management must be improved to adapt to the new conditions (Grossi et al., 2016). When examining long-term climate data from Europe, results for the growing season are less clear, but drier conditions are more likely (Schultz and Lebon, 2005; Degu et al., 2019; Carvalho et al., 2022). In recent years, wine regions of southern Europe have been affected by lower rainfall, extreme temperatures, and drought (Barbagallo et al., 2021). Drought (water stress) is an abiotic stress to which plants are frequently exposed (Chacon-Vozmediano et al., 2020; Azri et al., 2021; Cataldo et al., 2022). Like all other living organisms, the vine requires water for its life cycle. It affects the composition and quality of the berries. It also affects vegetative growth and yield (Pellegrino et al., 2005; Ezzhaouani et al., 2007; Acevedo-Opazo et al., 2010). Limited water availability affects plant growth, yield components, and grape berry quality (Zufferey et al., 2020). Water availability remains the absolute most critical issue worldwide, as all vineyards require irrigation (Yzarra et al., 2015). Quality red wines have been found to require a moderate water deficit (Barbagallo et al., 2021).

Water stress has different effects on vine yield and quality components depending on the timing, intensity, and duration of water scarcity (Zufferey et al., 2020). Many researchers have found that vine water status correlates with phenology (Scholasch and Rienth, 2019). The period from bud break to blooming is a very dynamic period of shoot and root growth, and it is considerable that vines start the season with high soil water content (Rogiers et al., 2014). It is well known that flowering to maturity is often influenced by water deficit (Ollat et al., 2002; Martinez-Lüscher et al., 2015). In addition, berry volume depends on carbon uptake (source-sink ratio), vine water status, temperature, and nitrogen supply (Deloire, 2010). It is well known that limiting carbohydrate uptake minimises fruit set in grapevines (Coombe, 1962; Duchene et al., 2001). In addition, vine water status has a critical influence on berry growth (Ojeda et al., 2001), which in turn can affect yield (Dry and Loveys, 1998; Cabral et al., 2022). The degree of water scarcity and the duration of water scarcity during the grapevine growing season are of great importance (Miras-Avalos and Intrigliolo, 2017; Deloire and Pellegrino, 2021). The effect of water deficit on berry growth does not affect cell division, but is manifested by a reduction in cell volume (Ojeda et al., 2001; Chaves et al., 2010). Ollat et al. (2002) indicated that early water deficits before ripening have major effects on yield.

Grenache is generally considered a drought-tolerant cultivar (Prieto et al., 2010), and this cultivar is traditionally classified as near isohydric (Santesteban et al., 2009); this behaviour is reported as a benefit of increased drought resistance (Schultz, 2003; Scharwies and Tyerman, 2016). Dayer et al. (2020) reported that Grenache cv. exhibits hydrodynamic behaviour rather than a specific iso-anisohydric classification.

The objective of this experiment was to evaluate the effects of early water stress in grapevine cv. Grenache during the 17th to 27th stage of E-L, on plant physiology and yield. The objective of this experiment is to study the response of the vine to early water restriction.

2. MATERIAL AND METHODS

2.1. Experimental site

Grenache is the most widely grown grape variety in the world (Meneghetti et al., 2011), originating in Spain and called Garnacha cv. Garnacha tinta is called Grenache N in France, the United States and Australia (Dry, 2004; Fernandez, 2015). Vine genotypes differ in their tolerance to water stress, with cv. Grenache having low tolerance (Schultz, 1996).

The experimental vineyard was established in 2001; the Grenache cv. was grafted onto SO4 rootstocks, and trained using the Lyre system, a form of bilateral cordon with approximately six shoots of 2-3 nodes. Early water restriction was performed on *Vitis vinifera* L. cv. Grenache from mid-May to mid-June (DOY 141 to DOY 164) at the 17th to 27th E-L stages in SupAgro Montpellier-INRA vineyards in southern France (Lat. 43°37' 04", Long. -3°51' 22", Alt. 40 m) in potted (72L) vines. At the beginning of the experiment, the surface of the pots was protected from precipitation by PE. The clusters were balanced at about 19 per plant (Figure 1).

The experiment was laid out with four irrigation levels CTRL: 4L, EDI1: 3L, EDI2: 2L and EDI3: 1L of water per day. Drip irrigation was used and treatments were applied according to Carbonneau (1998). Four water deficit levels were studied: control (CTRL): (Ψ_{pd} = 0 to -0.2 MPa), EDI1 (Ψ_{pd} = -0.2 to -0.4 MPa), EDI2 (Ψ_{pd} = -0.4 to -0.6 MPa), and EDI3 (Ψ_{pd} = -0.6 to -0.8 MPa). Full irrigation, 6L per day, was initiated at the E-L growth stage 14th to17th and 27th to 38th (Eichhorn and Lorenz, 1977). All stress treatments were harvested at DOY 223. In addition, phenological stages were recorded.

Figure 1: Plants in vineyard condition, pots were covered by black PE

2.2. Grapevine water status (Ψ_{pd}) (-MPa)

The Scholander Pressure Chamber (SPC) as an instrument to measure the Ψ_w (water potential). This chamber is used to measure plant water status directly on fully developed leaves (Williams and Araujo, 2002; Cole and Pagay, 2015). Pre-dawn leaf water potential has been shown to be a powerful tool to study differences in water stress (Alves et al., 2013). In this experiment, pre-dawn leaf water potential (Ψ_{pd}) was measured at 03:00 AM at three-day intervals using a SPC.

2.3. Vegetative components

Shoot length (cm); Two plants and 3 shoots per plant were measured per meter. Shoot length was determined at 3-day intervals. Shoot growth ratio (%); Shoot length was subtracted from the length measured 3 days earlier. Shoot growth ratio was determined in cm at 3-day intervals. These components were also determined by photographs during the experimental period. They were presented according to the stages EL and DOY.

2.4. Yield components

Yield components were determined at harvest (in September 10). Harvesting was done manually, and the weight of clusters on each plant was measured with a balance. Cluster weight and yield were also expressed in g and g/per vine, respectively.

2.5. Experimental Design

A randomized complete block (RCB) trial was chosen as the experimental design. There were three replicate blocks, and each replicate plot consisted of two vines in that trial. The vines used in the experiment had the same vigor.

2.6. Statistical analysis

The experiment was performed with the statistical program MSTAT-C, and the LSD test was also used to compare the means of the treatments at the 0,01 level.

3. RESULTS AND DISCUSSION

3.1. Phenological stages

A mean temperature of 19.13°C and a mean humidity of 69.96% were recorded during the experimental period. The recorded phenological stages were not affected by EDI (Early Deficit Irrigation) in the Grenache cultivar (Table 1). Similar to our results, Ojeda et al. (2001) expressed that vine growth in the Ecotron system was coherent with that of the vineyard (Table 1). Soltekin and Altindisli (2022) found that water stress caused phenological development of plants 6-8 days earlier than control plants. Our research results contradict this result, it was suggested that this was due to the differences in terroir components.

3.2. Plant water status (-MPa) (Ψ_{pd})

The water potential of plant leaves was measured according to the threshold values of Carbonneau (1998). At the beginning of the experiment, Ψ_{pd} was about -0.40 MPa in all treatments, this value was evaluated according to Ojeda et al. (2002), Williams and Araujo (2002), Deloire et al. (2005), and a reduction in vegetative growth was observed. In DOY 148 and DOY 151,

Ψ_{pd} was highest (-0.27 to -0.37 MPa) due to rain, although the pots were insulated with PE, which could be due to humidity (average 85-93%), and vegetative growth is normal. In EDI3, the lowest Ψ_{pd} was found in DOY 157 and DOY 163 (-0.60 to -0.63 MPa).

Table 1: Grenache wine grapes phenological stages during experimental period

Month/Day-	DOY	Stages E-L	Phenology of the vine
May 14	135	14	6-7 leaves separated
May 15	136	15	7-8 leaves separated
May 17	138	16	9-10 leaves separated
May 20	**141**	**17**	**12 leaves separated single flowers separated**
May 22	143	19	Beginning of flowering
May 28	149	20	10% caps off
May 31	152	21	30% caps off
June 4	156	23	50% caps off 17-20 leaves separated
June 7	159	25	80% caps off
June 8	160	26	Cap-fall complete
June 10	162	26	Setting (2mm)
June 12	**164**	**27**	**Berries pepper corn size (4mm)**
June 16	168	29	Berries pea size (7mm)
June 20	172	31	
July 12	194	33	Berry touch
July 29	211	35	Veraison
Sept. 10	223	38	Harvest

During the experiment, CTRL was fully irrigated, and Ψ_{pd} values averaged -0.22 MPa (little or no water deficit); EDI1 and EDI2, were -0.42 MPa (moderate to severe water deficit), and EDI3 was -0.52 MPa (moderate to severe water deficit). Carbonneau (1998) and Deloire and Rogiers (2014) assessment identified CTRL (-0,22 MPa) for mild to moderate water deficit (class 1), EDI1, EDI2, and EDI3 for moderate to severe water deficit (class 2). Also according to Deloire and Heyns (2011), the thresholds for pre-dawn leaf water potential (Ψ_{plwp}) ranged from 0 to -0.3 MPa for low or no water deficit (most cultivars) and from -0.3 to -0.6 MPa for moderate-severe water deficit (cultivar dependent). On the other hand, Deloire and Rogiers (2014) indicated that thresholds of -0.3 and -0.5 MPa for pre-dawn leaf water potential have a lower effect on vegetative growth (Figure 2). In previous studies, an early water deficit (1L/per plant) was found to reduce the stress value to -0.78 MPa in cv. Merlot (Korkutal et al., 2019) and cv. Syrah (Korkutal et al., 2011) to -0.80 MPa. Since the highest stress level measured in this study was -0.63 MPa, the researchers could not obtain the same results. It was concluded that this difference was due to the variety. Thus, it can be said that the Grenache grape variety is drought-tolerant (Prieto et al., 2010). Moreover, at the end of the EDI period, it was shown that the measured water stress values could not reach the plants of CTRL, although all plants were irrigated with 6 L per day.

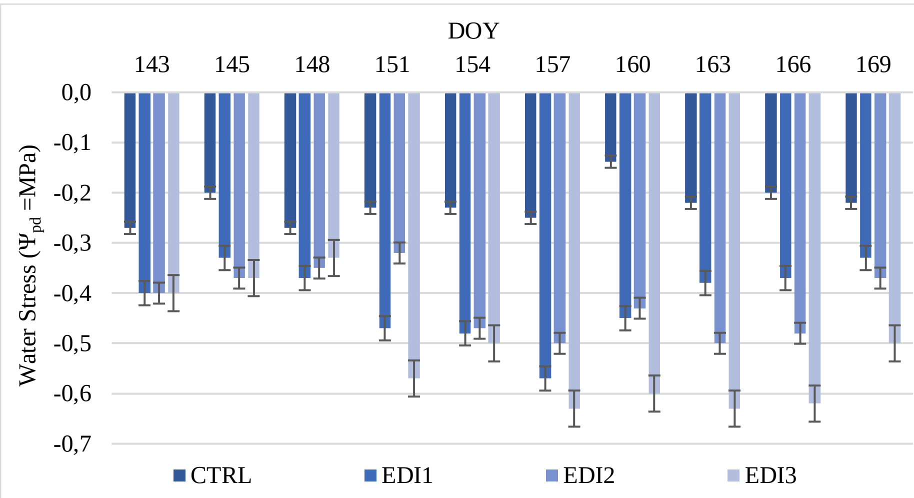

Figure 2: Changings Ψ_{pd} of Grenache N during study period

3.3. Vegetative components

3.3.1. Shoot length (cm)

At the beginning of the experiment, all shoots are about 55-70 cm long. Final shoot growth varies due to water scarcity. At CTRL, the final shoot length is 146.9 cm. The final length of the stress groups is 113.0 cm for EDI1, 102.2 cm for EDI2, and 84.7 cm for EDI3. Comparing the shoot lengths with those of plants from CTRL, EDI3 is 62.2 cm shorter than CTRL. It can be said that water deficiency has a negative effect on shoot elongation (Figure 3). A negative linear correlation (Stevens et al., 1995) was found between vine vegetative growth and EDI. These results are in the same direction as those of Korkutal et al. (2011), Korkutal et al. (2019), Deloire and Pellegrino (2021) and Candar (2022).

At the end of the first week of the experiment when the shoot elongation of EDI1 is more than 12 cm compared to CTRL, the stress value is -0.27 MPa on DOY 148. With the gradual decrease of the stress level of EDI1 on the same measurement days as CTRL, the difference in shoot length increased to 16.9 cm at a stress level of -0.57 MPa in DOY 157, this difference increased to 22.9 cm at a stress level of -0.45 MPa in DOY 160, and the difference reached 27.4 cm at a stress level of -0.38 MPa in DOY 163. At the beginning of the experiment (DOY 143), the difference in shoot length of EDI2 was 10.6 cm compared to CTRL. The difference in shoot length was 22.1 cm in DOY 157 compared to CTRL at a stress level of -0.50 MPa. The highest shoot length difference (34.4 cm) in EDI2 was observed in DOY 163 and at a stress level of -0.50 MPa under water stress conditions. The shoot length difference for EDI3 (-0.63 MPa) in DOY 157 was 34.9 cm compared to CTRL. It was found that the highest difference in shoot length at EDI3 (50.4 cm) occurred in DOY 163 and at a stress level of -0.63 MPa.

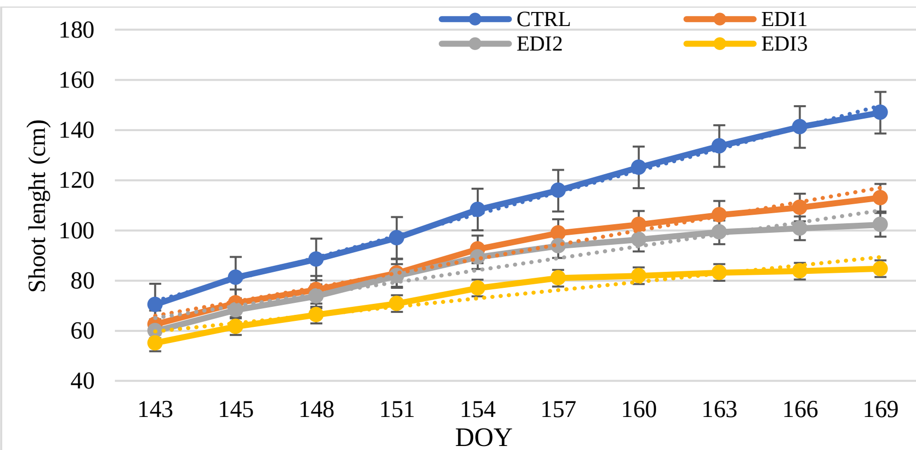

Figure 3: Shoot lengths in experimental days

3.3.2. Shoot growth ratio (%)

In Figure 4, the maximum growth is in the range of 151-154 DOY. On this day, the growth ratio ranges from 6.2 to 11.3 cm of shoot length per 3 days. The lowest ratio is found in EDI3, as expected. Buesa et al. (2017) made a similar observation that vines exposed to stress showed reduced lateral shoot growth due to higher Ψ_{pd} values. In DOY 157-160, CTRL and EDI plants showed a bright line in terms of shoot elongation ratio.

Vegetative growth and yield are close to each other (Kliewer and Dokoozlian, 2005). The main effect of water stress is the decrease in vegetative growth (Chacon-Vozmediano et al., 2020). This cycle was described using the photographic documents (Figure 5 to Figure 11). Finally, EDI affected vegetative growth, as well as in cv. Syrah (Korkutal et al., 2019), and cv. Merlot (Korkutal et al., 2011). Severe and sudden stress also severely affected growth (Bahar et al., 2011). These results are consistent with those of all researchers.

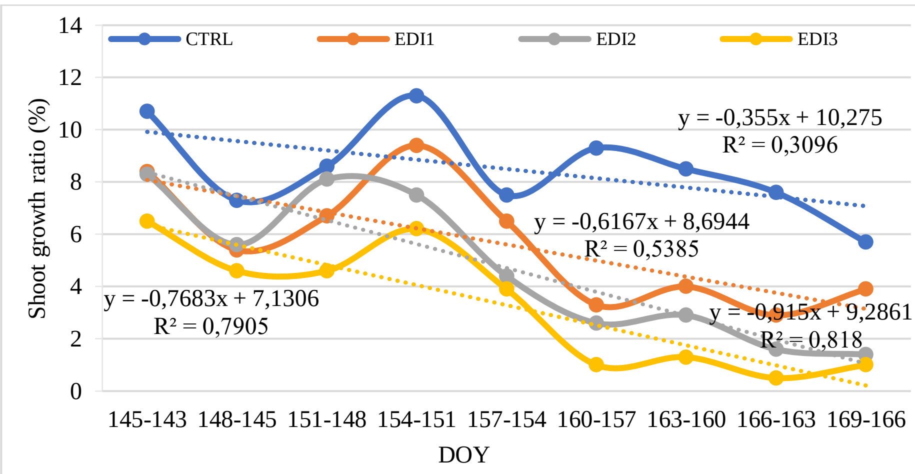

Figure 4: Shoot growth ratio of Grenache cv.

All photos in figures CTRL are the first, EDI1 is the second, EDI2 is the third and, EDI3 is the fourth. Early deficit irrigation (EDI) is a 23-day period of stabilised soil irrigation. At the beginning of the experiment, all plants were healthy (Figure 5).

DOY 145 (EL 19)			
CTRL	EDI1	EDI2	EDI3

Figure 5: In the beginning of the experiment plants of Grenache cv.

DOY 156 is at EL 23^{rd} which means that plants are flowering, 50% of the caps are removed, 17-20 leaves are falling, and berry set is normal (Figure 6). In DOY 162, Ψ_{pd} values were CTRL (-0.22 MPa), EDI1 (-0.38 MPa), EDI2 (-0.50 MPa), and EDI3 (-0.63 MPa) (Figure 7). By the EL 26^{th} the cap had completely fallen off, but the basal leaves had turned yellow. The number of yellowish leaves increased with EDI.

DOY 156 (EL 23)			
CTRL	EDI1	EDI2	EDI3

Figure 6: Flowering stage of cv. Grenache

DOY 162 (EL 26)			
CTRL	EDI1	EDI2	EDI3

Figure 7: Cap fall complete in cv. Grenache

At the EL 27^{th} stage, berries were the size of peppercorns. At stress level EDI3 (-0.63 MPa), some bunches were dry, as were some basal leaves. At EDI2 Ψ_{pd} (0.62 MPa), some leaves and grape parts were wilted. EDI1 (-0.38 MPa) had the least impact and no leaves or clusters were affected at CTRL (Figure 8).

DOY 164 (EL 27)			
CTRL	EDI1	EDI2	EDI3

Figure 8: Berries peppercorn size (4mm) in cv. Grenache

After EDI treatment, when berries reached pea size (7 mm), irrigation was maintained at 6 L per day. But EDI3 and EDI2 have a countable number of berries. Some yellowish leaves were also shed (Figure 9). This symptom is consistent with the results of Gerzon et al. (2015). Grenache plants showed reduced vegetative growth and leaf drop in response to water scarcity. In drier areas or soils, water stress generally leads to senescence and leaf drop (Munne-Bosch and Alegre, 2004) as was the case in our study.

DOY 170 (EL 29)			
CTRL	EDI1	EDI2	EDI3

Figure 9: Berries in pea size (7mm) cv. Grenache

As can be seen in Figure 10, the berries were at the berry stage and the effects of the EDI treatment were evident. The grapes subjected to water restriction had a countable number of berries.

DOY 197 (EL 33)			
CTRL	EDI1	EDI2	EDI3

Figure 10: Berry touch of Grenache cv.

A few days before harvest, the grapes were shown in Figure 11. It can be clearly seen that water deficiency has a negative effect on the quality of the grapes.

DOY 218 (about EL 38)			
CTRL	EDI1	EDI2	EDI3

Figure 11: A week earlier harvest the clusters of cv. Grenache

3.4. Yield components

Cluster weights decreased depending on the EDI value. Water stress simultaneously decreased grape bunch weight and yield. According to the OIV descriptor list, code 502, Garnacha tinta N grapes are medium sized and weigh about 500 g (OIV, 2001). On the other hand, Grenache bunch weight was defined as follows: in Parma average bunch weight 193 g and, yield 6.3 kg/vine (Shellie, 2007), in La Rioja average bunch weight 214.4 g and, yield 4.2 kg/vine (Tardaguila et al., 2008). In this study, a reduction of 23.62%, 34.59% and 62.03% was found when comparing the average bunch weights of the CTRL (EDI1, EDI2 and EDI3, respectively). A reduction of 23.08% (EDI1), 42.00% (EDI2) and 69.81% (EDI3) was also observed in yield (Figure 12).

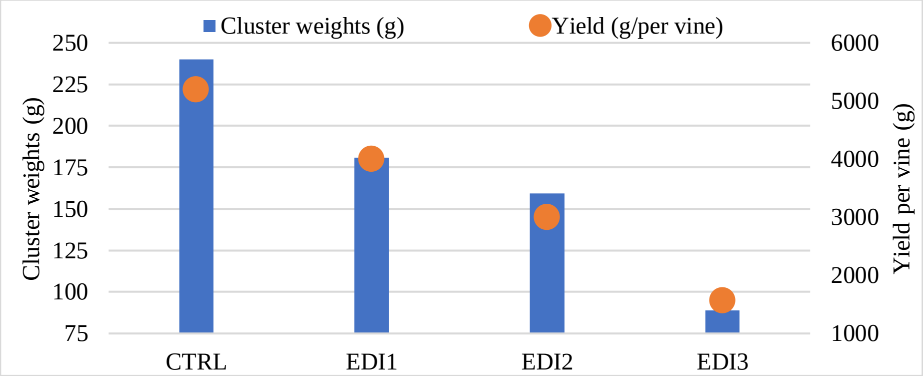

Figure 12: Cluster weight and yield under different levels of EDI

These results are in agreement with those of other researchers, such as Chacon-Vozmediano et al. (2020), who found a reduction in cluster weight and yield as a function of stress level. The lowest yield was obtained at -0.6 to -0.8 MPa. Our results show that there is a close relationship between water uptake and yield, mainly through the effects of water deficit on photosynthesis (Medrano et al., 2003; Deloire and Pellegrino, 2021). The resulting water stress reduced berry weight by about 50% (Van Leeuwen et al., 2009). Extreme water shortages also reduce yield and quality (Ojeda et al., 2002; Gambetta et al., 2020). Zufferey et al. (2020) reported that grapevine water status had no effect on cluster weight and yield at 150 to 280 DOY. Yield decreased with increasing water stress (Chacon-Vozmediano et al., 2020), and water restriction reduced yield by 10% (Buesa et al., 2017). Meanwhile, EDI resulted in an average 41%decrease in cluster weight in cv. Syrah (Korkutal et al., 2019), and 20% in cv. Merlot (Korkutal et al., 2011)

4. CONCLUSION

When water stress occurs during the most important growth phase of the vine, it has negative effects on vine growth, development, and fertility. Vegetative growth is severely affected by water stress. In addition, yield components decrease when stress levels increase. Consequently, EDI has a negative impact on the vegetative cycle and yield components.

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CHAPTER 3

PHENYLALANINE AND UREA-INDUCED CHANGES IN ANTHOCYANIN ACCUMULATION, PHENOLIC COMPOSITION AND MONOTERPENE PROFILES IN MERLOT WINE GRAPE VARIETY (*V. vinifera* L.)

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INTRODUCTION

Phenolic compounds are some of the most substantial quality components of wine owing to their effects on red wine color and oxidative processes of white wines (Cheynier et al., 2006) and as well as contribute to some major organoleptic attributes, including flavor, bitterness and astringency (Kennedy et al., 2001).

Anthocyanins are responsible for color of red wine grapes (Downey et al. 2006) and the amount and composition of anthocyanins in grape skin are important attributes in terms of grape quality (Mazza, 1995).

Monoterpenes are considered as secondary metabolites, contributing considerably to attributes of grape flavor and dividing into two different parts, including free volatile terpenes (FVTs) and potential volatile terpenes (PVTs) (Mateo and Jimenez, 2000; Sapceska et al., 2006; Kok, 2017).

All these quality attributes mentioned above are substantial measures of grape quality, so any means of raising the amount of these quality variables in grapes would be an advantage to grape grower and winemaker (Kok, 2018a; Kok and Bal, 2019).

Grape composition and associated attributes are influenced by various factors such as climate, soil type, genotype and management practices (Gladstones, 1994; Jackson and Lombard, 1993).

In wine grape growing, it may be encountered with decreases in grape quality attributes in some years and grape growers effort to explore various ways to accomplish these undesirable circumstances in grape quality. In order to overcome this unfavorable situations, grape growers have been constantly seeking for opportunities to increase grape quality. Among the grape quality improving practices in viticulture, utilization of various foliar applications on secondary metabolism in grapevines have been especially receiving much interests, since it has been displayed that these may raise accumulation of phenolic compounds and particularly anthocyanins (Kok, 2018a; b; Kok and Bal, 2019; Pastore et al., 2020; Kok, 2022).

Nitrogen fertilization application may improve yield along with grapevine overgrowth (Boonterm et al. 2013), but lead to reciprocally shaded leaves (Han et al., 2018) and alterations of grapevine canopy microclimate (Eynard et al., 2000; Martin et al., 2016; Reshef et al., 2018). However, limiting nitrogen supply in vineyards could be beneficial for high-quality grape and wine (Volpe et al., 2010).

Nitrogen is a component of proteins, nucleic acids, hormones and chlorophyll and is necessary for synthesis of both primary metabolites and production of numerous secondary metabolites in grapes (Bell and Henschke, 2005; Cheng et al., 2020).

Researchers have recently used various nitrogen sources such as arginine, phenylalanine, urea and mixture of urea and sulfur in their researches in order to increase amount of secondary metabolites, which are important elements of quality in grapes (Kok, 2018a; Cheng et al., 2020).

Phenylalanine is a vital constituent of protein in all living organisms and also serves as precursors for thousands of vital and specialized compounds in plants, including phenolic acids, flavonoids, phenolic compounds and aromatic compounds (Arora, 2010; Yoo et al., 2013).

Urea is the most broadly produced and used solid nitrogen fertilizer in the agricultural production due to its low price, low molecular weight, good solubility in water and effective absorption by plants (Lasa et al., 2012).

The application of nitrogenous compounds may bring about variations in grape composition. The goal of present study was to assess the influences induced by distinct concentrations of foliar phenylalanine and urea applications on yield and chemical attributes of Merlot wine grape variety.

1.MATERIAL AND METHODS

1.1 Research location and plant materials

The research was conducted during 2019 growing season in a commercial vineyard in Tekirdağ, Turkey (41°01´25.56´´N; 27°39´59.87´´E; 85m a.s.l.). In the current study, it was benefited from 12-year-old Merlot grapevines grafted onto Kober 5BB (*Berlandieri x Riparia Teleki 8B, Selection Kober 5BB*). The grapevines were trained to a vertical shoot-positioned trellis system on a bilateral cordon with 12 buds per grapevine, at a spacing of 2.5m between rows and 1.0m within a total of 4000 grapevines per hectare. All vineyard management practices in prevailing research area were performed in accordance with local standard practices. Meteorological data of 2019 year for Tekirdağ province were presented in Table 1.

Table 1: Meteorological data of 2019 year for Tekirdağ province

Meteorological features	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Mean temperature (°C)	5.6	5.8	9.3	11.6	17.9	24.1	23.9	25.3	21.6	17.5	15.5	9.2
Max temperature (°C)	8.1	9.1	13.1	14.9	21.3	28.2	27.7	29.6	25.8	21.1	18.9	12.5
Min temperature (°C)	3.2	3.1	5.7	8.2	14.4	20.1	19.6	21.2	17.6	14.0	12.4	6.4
Sunshine duration (h)	55.1	113.5	210.9	177.7	191.7	237.1	278.9	279.9	209.8	175.0	123.0	71.1
Precipitation (mm)	63.8	44.8	30.2	42.9	31.2	7.5	18.7	0.0	9.6	46.2	17.4	22.3
Relative humidity (%)	76.3	74.3	70.8	71.9	70.5	64.8	65.0	62.7	65.1	73.3	75.7	75.5

1.2 Foliar phenylalanine and urea applications

In existing study, uniform grapevines were chosen for foliar applications and it was made use of from distinct concentration of foliar phenylalanine and urea, including 0 mM, 2 mM phenylalanine (Phe), 4 mM phenylalanine (Phe), 8 mM phenylalanine (Phe), 20 mM urea (Ur), 40 mM urea (Ur) and 80 mM urea (Ur).

Previous to foliar spraying applications, phenylalanine (>98%, Sigma-Aldrich) and urea (>98%, Sigma-Aldrich) were employed for preparing both aqueous extracts and tween 80 as wetting agent at 0.1% was also added into all foliar solutions. A water solution of tween 80 alone was chosen for 0 mM application of phenylalanine and urea. Foliar spraying application concentrations of both phenylalanine and urea were carried out twice at 14 days before vérasion period and at vérasion period through using a back pump.

1.3 Yield and chemical attributes

In available study, berry length (mm), berry width (mm), berry weight (g), cluster length (cm), cluster width (cm) and cluster weight (g) were determined as yield attributes. Besides, total soluble solids content (%), total acidity (g/L), must pH, total soluble solids x pH^2 index (%), total phenolic compounds content (mg GAE/kg fw), total anthocyanin content (mg GAE/kg fw), free volatile terpene content (mg/L) and potentially volatile terpene content (mg/L) were analyzed as chemical attributes.

1.4 Harvest operation and preparation of berry sampling

In the course of 2019 growing season, berries on grapevines of Merlot variety were regularly monitored near the ripening period of grape variety and grapes were harvested when the berries of 0 mM phenylalanine and urea applied grapevines approximately attained total soluble solids content of 23%.

After berries had been harvested, samples of 250-berry were initially reserved for each application concentration and were ultimately employed to make analyzes for total soluble solids content, total acidity and must pH.

Additionally, it was utilized from 600-berry samples to find out total phenolic compounds content, total anthocyanin content, free volatile terpene content and potentially volatile terpene content. All berry samples were stored at -25°C until the analyzes of total phenolic compounds content, total anthocyanin content and monoterpene contents were performed. Prior to these analyzes mentioned above, berry samples were thrown out from -25°C, permitted to thaw overnight at 4°C and were homogenized in a commercial laboratory blender for 20s.

1.5 Analyzes of phenolic compounds and anthocyanins

Determination of total phenolic compounds content and total anthocyanin content were respectively conducted by using spectrophotometric methods explained by Singleton et al. (1978) and Di Stefano and Cravero (1991). Both analyzes results were clarified as milligrams of gallic acid equivalent per kilogram of fresh weight (mg GAE/kg fw).

1.6 Analysis of monoterpene compounds

Analyzes of free volatile content and potentially volatile terpene content from monoterpene compounds were also carried out by employing procedure reported by Dimitriadis and Williams (1984) as altered by Reynolds and Wardle (1989). Free volatile terpene content and potentially volatile terpene content were explained as mg/L.

1.7 Statistical analysis

The study was conducted according to a completely randomized blocks design with four replicates. All data were subjected to analysis of variance (ANOVA) through TARIST statistical software program. In order to separate differences among the means, Fisher's Least Significant Difference (LSD) multiple comparison test was employed when the ANOVA test was significant at 5% level.

2 RESULTS AND DISCUSSION

2.1 Yield attributes

In present study, significant differences were only detected in cluster length and cluster weight from yield attributes of Merlot grape variety ($p<0.05$; Table 2).

Nitrogen is one of the most important plant nutrients, encouraging plant growth due to its effects on grapevine vegetative and reproductive development (Bell and Henschke, 2005; Linsenmeir et al., 2008). Regarding berry length indicated in Table 2, distinct concentrations of foliar phenylalanine and urea applications have no significant effects on berry length ($p<0.05$) and the lowest berry length mean was 11.43 mm for 0 mM application whereas the highest berry length mean was obtained from 8 mM Phe application (11.83 mm).

Table 2: Effect of distinct concentrations of foliar phenylalanine and urea applications on yield attributes

Yield attributes	0 mM	2 mM Phe	4 mM Phe	8 mM Phe	20 mM Ur	40 mM Ur	80 mM Ur	$LSD_{5\%}$
Berry length (mm)	11.43	11.62	11.71	11.83	11.53	11.65	11.77	N.S.
Berry width (mm)	11.16	11.44	11.65	11.72	11.25	11.47	11.69	N.S.
Berry weight (g)	1.43	1.47	1.57	1.65	1.46	1.51	1.59	N.S.
Cluster length (cm)	13.60^{d}	14.21^{cd}	15.41^{ab}	16.17^{a}	13.76^{d}	15.05^{bc}	15.59^{ab}	0.85
Cluster width (cm)	10.04	10.87	11.11	11.42	10.22	10.99	11.22	N.S.
Cluster weight (g)	155.42^{c}	185.17^{b}	199.19^{ab}	212.80^{a}	165.90^{c}	191.39^{b}	201.55^{ab}	17.49

Different letters within the same parameter indicate significant differences at 5% level based on Least Significant Difference (LSD) multiple range test Phe: phenylalanine Ur: Urea
N.S.: Non-significant

It is noteworthy in Table 2 that berry width is not influenced by distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$) and higher berry width means were successively 11.72 mm (8 mM Phe), 11.69 mm (80 mM Ur), 11.65 mm (4 mM Phe), 11.47 mm mM (40 mM Ur), 11.44 mm (2 mM Phe), 11.25 mm (20 mM Ur) and 11.16 mm (0 mM).

As can be seen in Table 2, there are no significant effects on berry weight among the distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$) and berry weight means varied from 1.43 g (0 mM) to 1.65 g (8 mM Phe).

Grape cluster attributes may alter depending on genotype of grape variety and viticultural practices applied in vineyard (Çelik, 2011). Table 2 disclose that cluster length is significantly affected by distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$). The highest cluster length mean was recorded for 8 mM Phe application (16.17 cm) whereas the lowest cluster length mean was 13.60 cm for 0 mM application (Table 2).

It is obviously uncovered in Table 2 that cluster width is unaffected by distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$). While the highest cluster width mean was 11.42 cm for 8 mM Phe application, the lowest cluster width mean was recorded for 0 mM application (10.04 cm).

Based on cluster weight in presented in Table 2, distinct concentrations of foliar phenylalanine and urea applications significantly affect cluster weight ($p<0.05$). In the study, the highest cluster weight mean was obtained from 8 mM Phe application (212.80 g) when the compared to 0 mM application (155.42 g).

2. 2 Chemical attributes

In current research, all chemical attributes of Merlot grape variety were significantly influenced by distinct concentrations of foliar phenylalanine and urea applications except for total acidity and must pH ($p<0.05$, Fig. 1, 4, 5, 6, 7 and 8).

The nitrogen status of a grapevine affects the concentration of quality components of grape and thus, contributing primarily to wine grape and wine quality (Bell and Henschke, 2005; Linsenmeir et al., 2008). The qualitative compositions of grape depend on grape variety, climatic characteristics of grape growing region, viticultural practices and maturity stage of grape (Pavloušek and Kumšta, 2011; Jackson, 2020).

Sugar is usually clarified as total soluble solids concentration (pigments, acids, glycerol and sugar) and fermentable sugar concentration of grape must accounts for 90-95% of total soluble solids (Zoecklein et al., 1999). In available study, total soluble solids content is affected by distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$, Fig. 1). As represented in represented in Fig. 1, among the distinct concentrations of foliar phenylalanine and urea applications 8 mM Phe application led to the highest total soluble solids content (25.87%) when the compared with 0 mM application (23.07%).

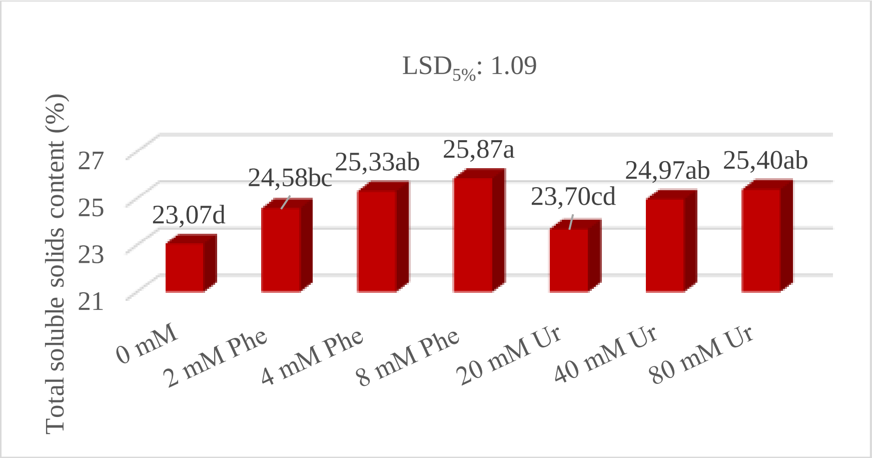

Fig. 1: Effect of distinct concentrations of foliar phenylalanine and urea applications on total soluble solids content

Titratable acidity of wine grapes ranges between 5.0 and 16 g/L as tartaric acid and the acid concentration of wine grape and resultant wine is crucial to structural and textural balance (Zoecklein et al., 1999). It is observed in Fig. 2 that distinct concentrations of foliar phenylalanine and urea applications have no significant effects on total acidity ($p<0.05$). While the lowest total acidity mean was recorded for 8 mM Phe application (8.15 g/L), higher total acidity means were in order of 10.12 g/L (0 mM), 9.47 g/L (20 mM Ur), 9.37 g/L (2 mM Phe), 9.23 g/L (40 mM Ur), 8.72 g/L (4 mM Phe) and 8.53 g/L (80 mM Ur).

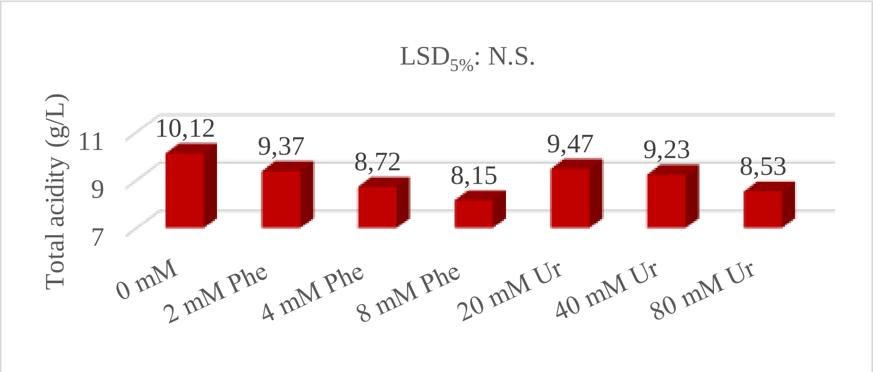

Fig. 2: Effect of distinct concentrations of foliar phenylalanine and urea applications on total acidity

Assessment of must pH is used to help determine the optimal time of grape harvest and must pH has a crucial impact on wine (Zoecklein et al., 1999). In view of must pH shown in Fig. 3, distinct concentrations of foliar phenylalanine and urea applications do not bring about significant effects on titratable acidity ($p<0.05$). In current study, must pH mean was the highest for 8 mM Phe application (3.37) compared to 0 mM application (3.30).

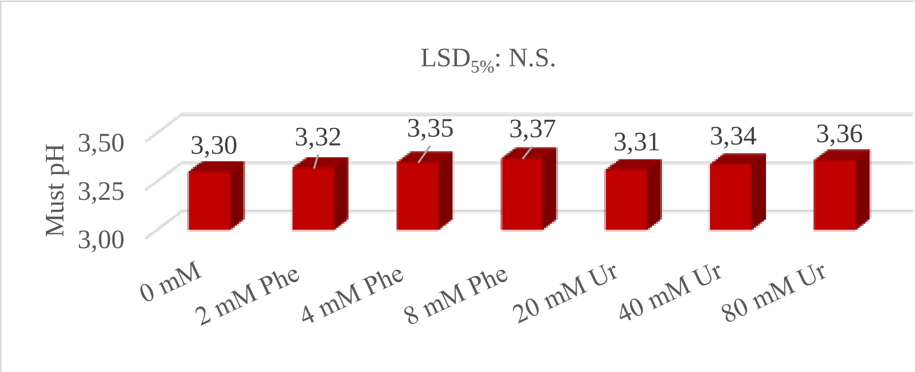

Fig. 3: Effect of distinct concentrations of foliar phenylalanine and urea applications on must pH

Total soluble solids x pH^2 index is better indicator for determining ideal ripeness status in wine grape varieties and its optimum values range from 200 to 270 (Bisson, 2001). In present study, there are significant differences among the distinct concentrations of foliar phenylalanine and urea ($p<0.05$) and index means of total soluble solids x pH^2 displayed in Fig. 4 changed from 251.70% (0 mM) to 287.45% (80 mM Ur).

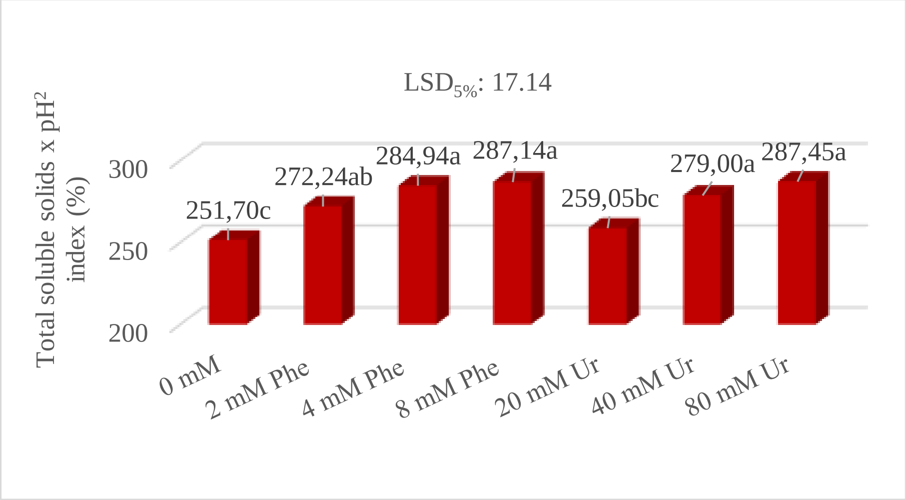

Fig. 4: Effect of distinct concentrations of foliar phenylalanine and urea applications on total soluble solids x pH^2 index

Phenylalanine is not only a nitrogen molecule but also a precursor in phenolic compound synthesis by phenylpropanoid biosynthetic pathway (Santamaria et al., 2015). Phenolic compounds have substantial role in wine grape and wine and phenolic compounds of grapes contribute to sensory properties of wine, including color, taste, mouthfeel, flavor, astringency and bitterness (Ribéreau-Gayon et al., 2006). In this study, total phenolic compounds content is significantly influenced by the distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$) and the highest total phenolic compounds content means were successively obtained from applications of 8 mM Phe (3947.75 mg GAE/kg fw), 80 mM Ur (3875.27 mg GAE/kg fw), 4 mM (3864.78 mg GAE/kg fw), 40 mM (3810.43 mg GAE/kg fw), 2 mM Phe (3751.31 mg GAE/kg fw), 20 mM Ur (3615.90 mg GAE/kg fw) when the compared with 0 mM (3211.60 mg GAE/kg fw) (Fig. 5).

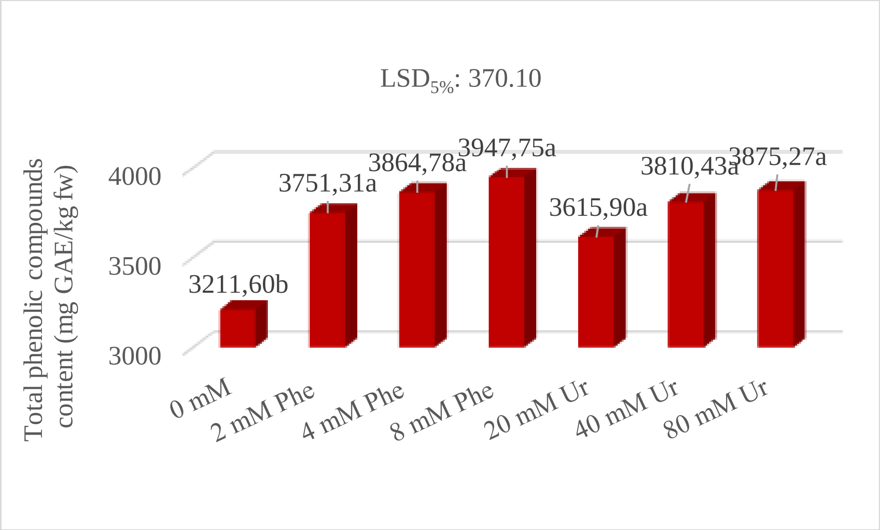

Fig. 5: Effect of distinct concentrations of foliar phenylalanine and urea applications on total phenolic compounds content

Anthocyanins located in berry skin are primary flavonoids responsible for red skin color of grape varieties (Mattivi et al., 2006). In existing study, distinct concentrations of foliar phenylalanine and urea applications have considerable effects on total anthocyanin content ($p<0.05$). The lowest total anthocyanin content was 1365.30 mg GAE/kg fw whereas the highest total anthocyanin content means were recorded for applications of 8 mM Phe (1678.25 mg GAE/kg fw), 80 mM Ur (1647.44 mg GAE/kg fw), 4 mM Phe (1642.98 mg GAE/kg fw), 40 mM Ur (1619.88 mg GAE/kg fw), 2 mM Phe (1594.74 mg GAE/kg fw), 20 mM Ur (1537.18 mg GAE/kg fw) compared to 0 mM application (1365.30 mg GAE/kg fw) (Fig. 6).

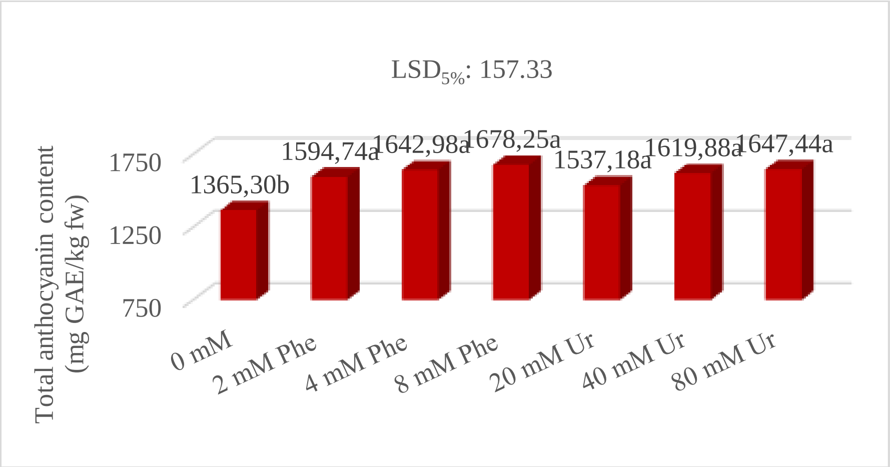

Fig. 6: Effect of distinct concentrations of foliar phenylalanine and urea applications on total anthocyanin content

Phenylalanine is precursor of 2-phenylethanol, a compound involved in wine aroma with a rose descriptor (Jackson, 2020) and of phenolic compounds (Fernández-Mar et al., 2012). Monoterpenes common ingredients of many fruits and grapes contribute strikingly to characteristic flavor of aromatic grape varieties. Merlot grape variety used in present study belongs to group of neutral grape varieties in accordance with classification of monoterpene content of grape varieties (Mateo and Jimenez 2000). Based on free volatile terpene contents of grape, significant differences were determined in distinct concentrations of foliar phenylalanine and urea applications ($p<0.05$). As shown in Fig. 7, all concentrations of foliar phenylalanine and urea applications, including 8 mM Phe (0.860 mg/L), 80 mM Ur (0.845 mg/L), 4 mM Phe (0.840 mg/L), 40 mM Ur (0.827 mg/L), 2 mM Phe (0.815 mg/L), 20 mM Ur (0.788 mg/L) led to the highest free volatile terpene content than 0 mM application (0.698 mg/L).

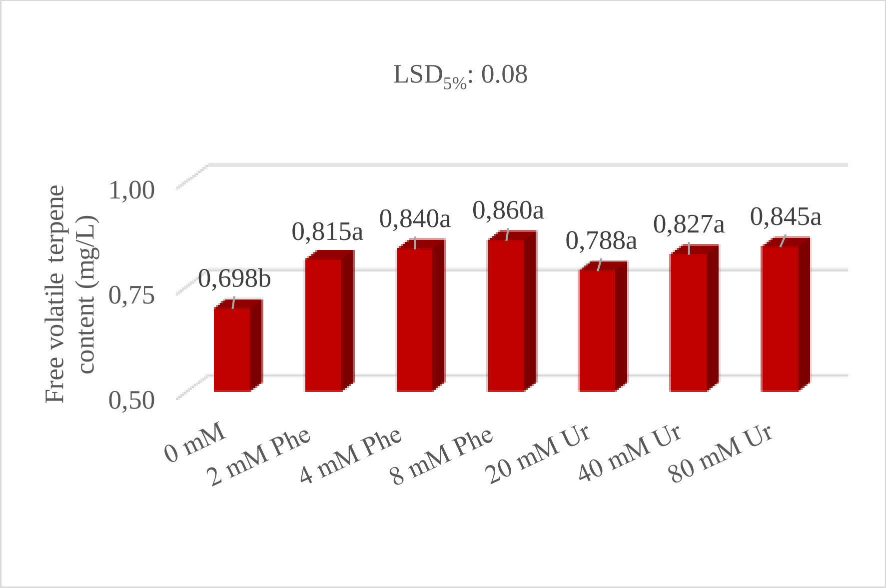

Fig. 7: Effect of distinct concentrations of foliar phenylalanine and urea applications on free volatile terpene content

The distribution of potentially volatile terpenes of grapes applied distinct concentrations of foliar phenylalanine and urea were found to be significant ($p<0.05$). The highest potentially volatile terpene contents were obtained from all concentrations of foliar phenylalanine and urea applications such as 8 mM Phe (2.130 g/L), 80 mM Ur (2.090 g/L), 4 mM Phe (2.083 g/L), 40 mM Ur (2.055 g/L), 2 mM Phe (2.023 g/L), 20 mM Ur (1.950 g/L) when the compared with 0 mM application (1.730 g/L) (Fig. 8).

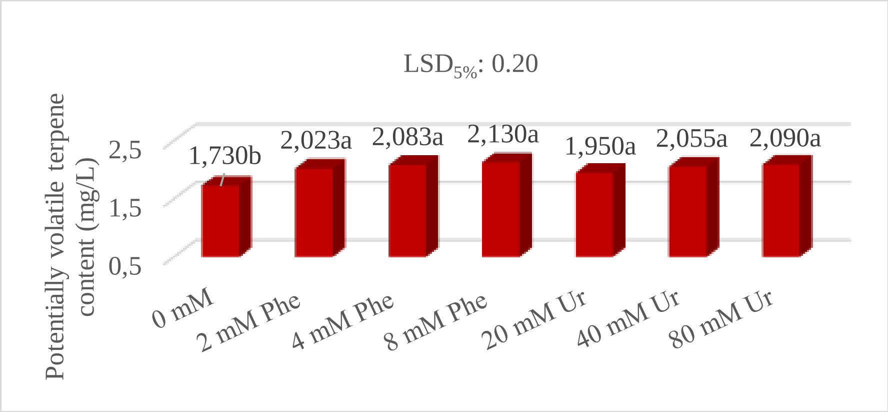

Fig. 8: Effect of distinct concentrations of foliar phenylalanine and urea applications on potentially volatile terpene content

3. CONCLUSION

In recent times, there has been a great interest in using lower concentrations of foliar nitrogen applications from various sources at certain phenological growth period of grapevine for increasing metabolic constituents of grape varieties. Nitrogen is vital for the synthesis of primary metabolites and production of several secondary metabolites, including phenolic compounds and aromatic components, which are remarkable quality attributes of grapes. In current research, study results pointed out that foliar phenylalanine application and to a lesser extent foliar urea application, to grapevine could be a useful tool for increasing wine grape quality parameters. As a result of study, 8 mM Phe application especially gave rise to the best results in chemical characteristics of Merlot grape variety and respectively followed by applications of 80 mM Ur, 4 mM Phe, 40 mM Ur, 2 mM Phe and 20 mM Ur.

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CHAPTER 4

THE USE OF SORBITOL IN THE DETERMINATION OF DROUGHT TOLERANCE OF FOX GRAPE (*Vitis labrusca* L.) *IN VITRO*

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INTRODUCTION

Drought, which is at the forefront of natural disasters, causes great losses by causing great damage to the environment and people. Forest fires due to global warming, decrease in precipitation, increase in greenhouse gases, insufficient water resources have become a major problem threatening countries (Hekimoglu and Altındeger, 2008). Drought stress has the largest share with a rate of 26% among the stress factors seen in the world's arable lands (Kalafatoglu and Ekmekci, 2005). Drought stress is more common in areas where rainfall is not sufficient, as plants cannot find the moisture they need to survive (Sircelj et al., 2007). It know that the precipitation factor, which has the highest effect on production and yield among climate factors, causes great temporal and local changes in plants (Kapluhan, 2013).
As in many parts of the world due to the climate change, it is observed that the precipitation decreases and irregular in Turkey, especially in the summer months (Kaynas and Kaynas, 2003). It is predicted that with climate change in Turkey, the temperature values will increase and the high temperature may affect the semi-arid and arid regions that do not have enough water. It is estimated that plant yield and agricultural production will be adversely affected due to these decreases for precipitation and the irregularity of the distribution of precipitation over the months (Ozturk, 2002). It has been reported that Turkey will be among the risk group countries that will likely be affected by climate change and that the Central Anatolia and Mediterranean regions will be more affected by global warming in the future (Sircelj et al., 2007). Between 2006 and 2007, Turkey experienced the fıfth driest period of the last 37 years and it was determined that Central Anatolia, Aegean and Marmara Regions were affected the most from this period (Simsek and Cakmak, 2010).

Due to the rapid increase in the world's population, the amount of water consumed per capita has also increased. While the amount of water consumed per capita was 110 m^3 in the 1700s, it is known that a large part of this amount was used for irrigation of agricultural areas, and this amount increased 40 times in the 1990s (Kılıc, 2008). More than 50% of crop loss in the world is caused by abiotic stress factors (Wang et al., 2001; Wang et al., 2003). Plants respond to abiotic stress factors such as drought, temperature and salinity with physiological and metabolic events in a way that will cause the least damage to their growth and development. Drought stress affects many physiological, biochemical and metabolic events in plants, and plants can develop some tolerance mechanisms in order to adapt to environmental conditions (Kalefetoglu and Ekmekci, 2005).

Drought stress affects plant growth and development negatively by preventing the division of cells in plants. However, with the decrease in turgor pressure and the decrease for water lost by transpiration, mineral substance uptake is prevented. The water lost negatively affects plant growth and directly reduces yield. Based on this, one of the most important limiting factors in agricultural production has been reported as drought (Capell et al., 2004, Faroog et al., 2009). With the decrease for water lost by transpiration under drought stress, morphological changes in leaves and roots absorb water from the soil with a higher strength (Cırak and Esendal, 2006; Kutlu, 2010). When plants are under drought stress, they tend to reduce water loss and transpiration by shedding their leaves and reducing leaf areas. With the decrease in leaf area, CO_2 fixation per unit area also decreases. For this reason, the plant performs less photosynthesis and slows down in development and growth as it cannot replace what it has lost. In addition, wax production on the leaf surface increases and the cuticle layer reflects the sun's rays, reducing the effect of temperature, thus reducing the transpiration rate (Costa-Franca et al., 2000; Turkan et al., 2005).

Finally, drought stress causes a decrease in plant water potential, wilting leaves, decreased cell growth and development, and closure of stomata. Severe drought can result in metabolic disturbance, reduced photosynthesis and ultimately death (Bohnert and Jensen, 1996). For this reason, water deficiency in plants is an important factor for plant cultivation, as it leads to crop losses.

Although the vine's resistance to drought stress varies according to the duration of exposure to stress, it becomes more susceptible during and immediately after flowering. The effect of water stress on crop varies according to the severity of the drought and the exposure time (Grimplet et al., 2007; Deluc et al., 2009; Chaves et al., 2010). Most of the vineyard areas are common in areas with seasonal drought, which, together with high temperatures, causes reductions in the relative humidity of the soil and air, and severely limits crop quality and yield (Chaves et al., 2007). The uniform distribution of precipitation over the months is more effective on the drought than the annual precipitation amount (Cırak and Esendal, 2006). However, based on the ecological characteristics of the vineyard areas, they are known that the amount of precipitation is generally low during the vegetation period, and it is observed that this situation has significant effects on yield and quality (Agaoğlu et al., 2003).

Some propagation methods have negative features such as allowing plant production in a limited number and in a long time, causing the transmission of diseases and pests and unwanted mutations. Due to these features, they are observed that tissue culture methods, which are faster and more effective in production of virus-free varieties, gain importance in plant reproduction today. It is known that the first micro propagation in vine was used to obtain plantlets from micro cuttings (Jean et al., 1998; Kinfe, 2010). With the propagation by tissue culture method, the production of stronger, pathogen-free and high yielding plants can be achieved with the reproduction of genetically homogeneous plant populations (Murashige and Skoog, 1974;

Blazina et al., 1991). It is possible to define tissue culture as the production or reproduction of tissue or plant anew by making use of parts of the plant such as cells, tissues or organs in an artificial nutrient medium. Plant tissue culture techniques are also widely used in plant breeding and non-breeding applications. While tissue culture is used in plant breeding, haploid plant production, in vitro selection, in vitro fertilization, conservation of genetic resources, protoplast fusion, and gene transfer, it is also used in areas such as micro propagation, synthetic seed production, and secondary metabolite production outside of breeding (Babaoğlu et al., 2001). Plant tissues grown in vitro can differentiate and form new organs such as roots, shoots and leaves directly or indirectly.

To examine the effects of abiotic stress on plants, it is necessary to control the physical environment *in vitro* (Verslues et al., 2006; Kielkowska et al., 2012). In tissue culture, stress can be induced by the addition of some specific compounds to the nutrient medium. Depending on their physicochemical properties, these compounds can be classified as stress inducing ionic and cell penetrating. They can be divided into non-ionic and penetrating (mannitol, sorbitol) and non-ionic and non-penetrating (polyethylene glycol (PEG)) (Gangopadhyay et al., 1997). In general, adding compounds such as PEG, mannitol or sorbitol to the culture medium (Ahmad et al., 2007), can create drought stress artificially. Osmotics such as mannitol or sorbitol inhibit mineral uptake from cells. For this reason, the growth and development of plants slows down and is adversely affected (Dodds and Roberts, 1985; Thomson et al., 1986). Many studies use polyethylene glycol (PEG) or sorbitol to create artificial drought stress. Sorbitol is actually a solution that is not metabolizing by plants. In addition, it reduces the osmotic potential of the nutrient medium and creates water stress that is not metabolized by plants (Rai et al., 2011; Bidabadi et al., 2012; Placide et al., 2012; Vanhove et al., 2012).

In the Black Sea Region, there is a very rich potential in terms of the types of *Vitis labrusca* known by different names such as American grape, strawberry grape, fox grape or Isabella. (Oraman, 1972; Çelik et al., 1998; Çelik, 2004). Fungal diseases do not affect grape types in this type, so they can be grown without any spraying (Cangi, 1999; Çelik, 2004).

In this study, the effectiveness of sorbitol, which provides the opportunity to create an artificial drought in vitro conditions by using single-node micro cuttings of Balıkçı Siyahı and the most effective sorbitol dose (s) for this purpose were determined.

1. MATERIAL and METHOD

In the study, 2-3 cm micro cuttings containing a single node taken from the shoots of the Balıkçı Siyahı (*V. labrusca* L.) grape type obtained from the Ordu University Research vineyard during the active growth period were used.

1.1 Sterilization of Tools and Equipment Used

The sterilization of lancets, forceps, blotting papers and test tubes (15 cm x 2.5 cm, Z681784, Merck) used in the study was carried out in an autoclave (Core, NC 90M) at 1.05 atm pressure and temperature at 121°C for 15 minutes.

1.2 Preparation of the Nutrient Medium

In the research, 1 mg/l BA (6-Benzylaminopurin, B3408, SIGMA) was added to the MS nutrient medium (Murashige Skoog 1962, SIGMA, M5519 C) as a growth regulator to form shoots from micro cuttings. 1 mg/l IBA (Indole-3-butyric acid, I5386, SIGMA) was used in rooting the shoots and creating artificial drought stress (Table 1).

After adding powdered basic MS medium (Murashige Skoog, 1962, M5519, SIGMA) and plant growth regulators (myo-inositol, BA), volume completion was performed with distilled water. The pH of the nutrient medium was fixed to 5.8 using 0.1 N HCl (Hydrochloric acid, K50244717821) and 0.1 N KOH (Potassium hydroxide, B1485433829, MERCK). 8 g/L agar was added to the medium as a solidifier and boiled. After the medium boiled, approximately 10 ml was evenly distributed in the test tubes and the caps of the tubes were closed. Afterwards, the sterilization of the prepared media was ensured by keeping them under 1.05 atm pressure and in an autoclave at 121°C for 15 minutes (Fig.1). After sterilization, the media was transferred into a sterile cabinet. In order to create artificial drought stress, 5 different doses of sorbitol (0, 0.1, 0.2, 0.3 and 0.4 M) were added to the prepared MS nutrient medium containing 1 mg/l IBA.

Fig 1: Nutrient Medium Preparation Stages (Akın, 2022)

Table 1: Content of MS Nutrient Medium (Murashige ve Skoog, 1962)

Compound	**Standard concentration (mg/l)**
Macro Elements (x10)	
$CaCl_2.2H_2O$	440
KNO_3	1900
NH_4NO_3	1650
KH_2PO_4	170
$MgSO_47H_2O$	370
Micro Elements (x 100)	
$MnSO_4.4H_2O$	22.3
H_3BO_3	6.2
$ZnSO_4.7H_2O$	8.6
$Na_2MoO_4.2H_2O$	0.25
$CuSO_4.5H_2O$	0.025
KI	0.83
$CoCl_2.6H_2O$	0.025
$Na_2EDTA.2H_2O$	37.3
$FeSO_4.7H_2O$	27.8
Vitamins (x 100)	
Glycine	2.0
Nicotinic acid	0.5
Thiamine-HCl	0.1
Pyridoxine-HCl	0.5
Growth Regulators	
IBA (Indole-3-Butyric Acid)	1 mg/l
BA (Benzyl Adenin)	1 mg/l
Organic Matters	
Myo-Inositol	100
Sucrose (g/l)	30
Agar (g/l)	8
pH	**5.8**

1.3 Preparation and Cultivation of Plant Material

In the study, 2-3 cm long micro cuttings containing a single node taken from the shoots of the Balıkçı Siyahı (*V. labrusca* L.) grape type obtained from the Ordu University Research vineyard during the active growth period were used. In order to reduce the plant-borne infections of the micro cuttings whose leaf parts were removed, a front surface sterilization trial was established by using commercial bleach at different doses and times. Microcuttings were soaked in commercial bleach at different doses and times (10 minutes in 20% commercial bleach, 15 minutes in 20% commercial bleach, 20 minutes in 20% commercial bleach, 10 minutes in 15% commercial bleach, 15 minutes in 15% commercial bleach or 20 minutes in 15% commercial bleach). As a result of the pre-sterilization trial, it was decided that 20% commercial bleach and 20 minutes surface sterilization time were appropriate and 1-2 drops of Tween 20 (P1379, Merck) were dropped into the prepared surface sterilization solution. Surface sterilization was ensured by keeping the micro cuttings in this solution for 20 minutes and then rinsed 3 times with sterile distilled water in a sterile cabinet. (Fig 2).

Fig 2: Appearance of Surface Sterilization in Micro Cuttings (Akın, 2022)

Sterilized microcuttings were cultured in MS nutrient medium with 1 mg/l BA added in a sterile cabinet, the caps of the tubes were closed, wrapped with stretch film and taken to the air-conditioning room (Fig 3).

Fig 3: Appearance of Cultivation of Explants (Akın, 2022)

After an average of 3 weeks, the shoots formed by the bursting of the node were cut and removed from the explants. In order to root and examine their response to drought, they were transferred to MS medium containing 1 mg/l IBA and different sorbitol doses (0, 0.1, 0.2, 0.3 and 0.4 M) (Fig 4).

Fig 4: Transfer of Formed Shoots to Rooting Media (Akın, 2022)

The cultured explants were kept in growth rooms with white fluorescent lamps with a temperature at 25±2 ˚C, a photoperiod of 16 hours of light and 8 hours of darkness, and an illumination of 3000-4000 lux.

Plant viability rate (%) was determined by dividing the number of survivors planted in different doses of sorbitol-added MS medium to the total number of plantlets and multiplying by 100. After the removal of the plantlets from the MS medium, the shoot lengths (cm) with a ruler; shoot fresh weights (g) weighed with precision scales sensitive to ± 0.001 g; shoot dry weights (g) were subsequently determined using a precision scale with ±0.001 g sensitivity after drying in an oven at 65°C for 72h; the number of nodes and leaf numbers were counted (pieces); the chlorophyll content (Soil Plant Analysis Development (SPAD) readings) was determined in leaf samples with a chlorophyll meter (SPAD-502, Konica Minolta Sensing, Tokyo, Japan). The degree of damage to the explants (1-4) was determined according to Sivritepe et al. (2008). The plants that were not damaged were graded as '1' degree, those with burns and dryness at the shoot tips and leaf edges of the plants were rated as '2' degree, those with necrosis in all leaves and stems of the plants as '3' degree and for dead plants as '4' degree. Shoot tolerance ratios of Balikci Siyahi under drought stress were calculated by using the following equation for each sorbitol dose in terms of shoot weight (g).

STR=Tx / To

Tx: Shoot weights of plantlets treated with sorbitol at a certain concentration (g)

To: Shoot weight of plantlets untreated with sorbitol (g)

After the removal of the plantlets from the medium, the ion flux (%) was determined using the method of Ozden et al. (2009) in the leaf samples divided into equal pieces of 0.3 g. In addition, leaf turgor weights (g) were determined by keeping the leaf samples taken from the plantlets in pure water for 6 hours. Considering the fresh weights (FW), the turgor weights (TW) determined by keeping in pure water for 6 hours and the dry weight (DW) determined after 24 hours in the oven at 80 ºC, explant water content is determined using the formula EWC (%): [(FW - DW) / (TW-DW) x 100] (Yamasaki and Dillenburg 1999).

1.4. Statistical Analysis

Experiments were conducted on randomized plot design with three replications with nine explants in each replication. Means were separated with LSD test at 5 % significance level. JMP 10.0 statistical software was used in statistical analyses. In the statistical analysis of the values expressed as percentage (%) were converted to angle transformation (arcsin√x) value and then evaluated statistically.

2. RESULTS AND DISCUSSION

2.1 Growth and Development Parameters

In the experiment, the effect of different doses of sorbitol application on the shoot growth-development parameters in cv. Balıkçı Siyahı grown under *in vitro* conditions was found statistically significant ($p<0.05$) (Table 2). The highest shoot length was determined in the application of 0% sorbitol with a value of 3 cm, and the lowest shoot length was determined in the application of 0.4% sorbitol with a value of 1 cm. The application with the highest number of nodes in the shoot was 0% sorbitol dose (2.9). 0.1% Sorbitol dose (2.7) was in the same statistical group and followed this application. The minimum number of nodes was determined in the application of 0.3% (1.6). In the experiment, the best result in terms of the effect of sorbitol applied at different doses on the number of leaves was obtained from 0 M sorbitol application (2.7). 0.1 M sorbitol (2.3) application was also included in the same statistical group and followed 0 M sorbitol application. The least number of leaves was obtained from 0.3 M (1.3) and 0.4 M (1.7) sorbitol applications in the same statistical group. In the experiment, a decrease in the amount of chlorophyll was detected with the increase of the sorbitol dose. While the highest SPAD value was 18.86 in 0 M sorbitol application, the lowest value was determined as 9.56 in 0.4 M sorbitol application. With increasing dose applications, a decrease in shoot fresh weight and shoot dry weight was observed. The highest fresh and dry weight of shoot was determined in 0% Sorbitol application (0.201 g, 0.011 g, respectively). The lowest shoot fresh weight was determined in 0.3 M and 0.4 M sorbitol applications (0.103 g, 0.090 g, respectively). The lowest value in shoot dry weight was determined in medium containing 0.4 M (0.008 g) sorbitol (Table 2).

Table 2: The Effect of Different Sorbitol Doses on Chlorophyll Content and Shoot Growth and Development Parameters of Balıkçı Siyahı Grape Type

Sorbitol Doses (M)	**Shoot Length (cm)**	**Node Number (n)**	**Leaf Number (n)**	**Chlorophyll Content (SPAD)**	**Shoot Fresh Weight (g)**	**Shoot Dry Weight (g)**
0 M	2.02 a	2.9 a	2.7 a	18.86 a	0.201 a	0.011 a
0.1 M	1.85 a	2.7 ab	2.3 ab	14.24 b	0.173 ab	0.010 ab
0.2 M	1.41 b	2.0 bc	1.7 bc	11.65 c	0.138 bc	0.009 ab
0.3 M	1.26 b	1.6 c	1.3 c	11.41 cd	0.103 c	0.009 ab
0.4 M	1.28 b	2.0 bc	1.7 bc	9.56 d	0.090 c	0.008 b
LSD 5%	0.35	0.7	0.8	2.07	0.050	0.002

**SPAD Soil Plant Analysis Development reading, LSD least significant difference, within column, means followed by a different letter differ significantly at $p< 0.05$ by LSD

Albiski et al. (2012) and Gopal ve Iwama (2007) determined a decrease in plant length due to drought stress with the increase in sorbitol concentration in potato (*Solanum tuberosum* L.). Daniel et al. (2014) determined that there was a decrease in the number of shoots and shoot length in the drought stress created by the addition of PEG 6000 (0%, 2%, 4%, 6%) in MM 106 apple rootstock. Gelmesa et al. (2017) found that the shoot length was lower at 0.1 and 0.2 M sorbitol doses in their study to define drought-tolerant potato genotypes. Gelmesa et al. (2017) found that the shoot length at 0.1 and 0.2 M sorbitol doses was lower than the control in their study to define drought-tolerant potato genotypes. The decrease in shoot length values due to increased sorbitol dose obtained from this study is also supported by the results of the researchers mentioned above. Gecene (2020) determined that the number of nodes decreased with increasing PEG doses due to drought. Low node number and leaf number results obtained from this experiment due to drought. Albiski et al. (2012), M. Harun-Or-Rashid et al. (2021) and Bilir Ekbic et. al (2022) is supported by the results of number of node and leaf. Karimi et al. (2012) found that the total chlorophyll level was lower with the increase in drought severity compared to the control group, in five almond cultivars and one hybrid rootstock (GF 677). Meşe and Tangolar (2019) created artificial drought stress on Kober 5 BB (sensitive to drought), 110 R (drought resistant) and 1103 P (moderately drought resistant) rootstocks with the addition of 0.0%, 2.5%, 5.0%, 7.5% and 10.0% PEG *in vitro* conditions. Similarly, in the mentioned study, chlorophyll content decreased due to increased drought. Gecene (2020) found a decrease in the amount of chlorophyll with increasing PEG dose with drought.

2.2 Physiological Parameters

In the experiment, the effect of different doses of sorbitol application on plant viability, degree of damage, shoot tolerance rate, explant proportional water content parameters in cv. Balıkçı Siyahı grown under *in vitro* conditions was found statistically significant ($p<0.05$). The highest viability rate was determined with a value of 90% at 0% (Control), 0.1% and 0.2% sorbitol concentrations (Table 3). The 0.4% sorbitol concentration provided the lowest plant vitality rate with a value of 65.5%. The degree of damage increased with the increase in sorbitol doses. As compared to the 0 M (1.15) application, more damage was detected in the 0.1 M and 0.2 M sorbitol applications (1.52). In 0.3 M and 0.4 M sorbitol doses, damage was observed at 1.92 and 2.22 degrees, respectively (Fig 5). There was also a decrease in shoot tolerance rate due to increasing sorbitol doses. The highest shoot tolerance rate was determined in the control group (1.000). This was followed by 0.1 M and 0.2 M sorbitol doses (0.876 and 0.800, respectively). The lowest tolerance rate was determined at 0.4 M sorbitol dose (0.641). The effect of sorbitol applications at different doses applied to Balıkçı Siyahı grape type in vitro conditions on leaf turgor weight was not found to be statistically significant. The decrease in the values due to the increase in the dose detected in other physiological parameters was not clearly determined for the turgor weight. The highest leaf turgor weight was determined in 0.1 M sorbitol (0.042 g) application and the lowest leaf turgor weight was determined in 0.3 M sorbitol (0.020 g) application. In the experiment, a decrease in explant proportional water coverage was determined with the increase of artificial drought stress created with sorbitol. The highest explant proportional water content was determined as 144.73% in 0 M (control) application. Other nutrient media containing 0.1 M, 0.2 M, 0.3 M, 0.4 M sorbitol (77.58, 65.58, 58.10, 57.41) were in the same statistical group and caused the lowest explant water content (Table 3).

Table 3: The Effect of Different Sorbitol Doses on Some Physiological Parameters of Balıkçı Siyahı Grape Type

Sorbitol Dose	Plant Viability (%)	Damage Degree (1-4)	Shoot Tolerance Rate	Leaf Turgor Weight (g)	Explant Proportional water content (%)
0	90.0 a	1.15 c	1.000 a	0.027	144.73 a
0.1 M	90.0 a	1.52 bc	0.876 ab	0.042	77.58 b
0.2 M	90.0 a	1.52 bc	0.800 b	0.038	65.58 b
0.3 M	83.2 a	1.92 ab	0.778 b	0.020	58.10 b
0.4 M	65.5 b	2.22 a	0.641 c	0.039	57.41 b
LSD 5%	10.1	0.57	0.134	N.S.	20.42

** LSD least significant difference, within column, means followed by a different letter differ significantly at $p< 0.05$ by LSD

The effect of sorbitol applications at different doses on the ion flux is shown in Figure 6. The decrease in the values due to the dose increase detected in the other investigated properties was not clearly determined for the ion flux. The highest ion flux value was determined as 119.8% at 0.2 M sorbitol dose. This application was followed by 0.4 M and 0.3 M applications with 88.4% and 88.0% values, respectively. The lowest ion flux was found to be 58% in the 0.1 M sorbitol application.

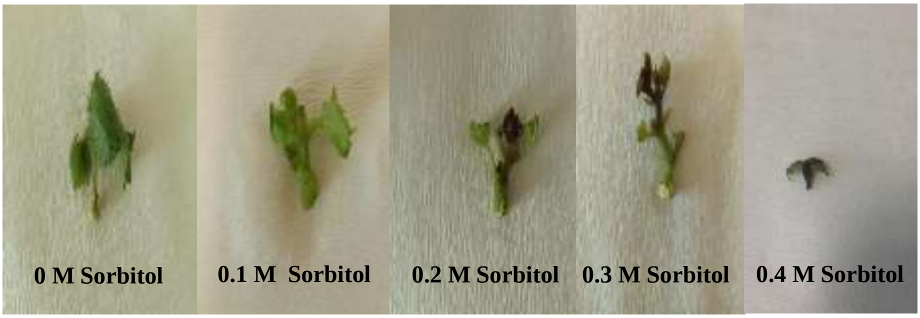

Fig 5: The Effect of Different Sorbitol Doses on the Degree of Shoot Damage of Balıkçı Siyahı Grape Type (Akın, 2022)

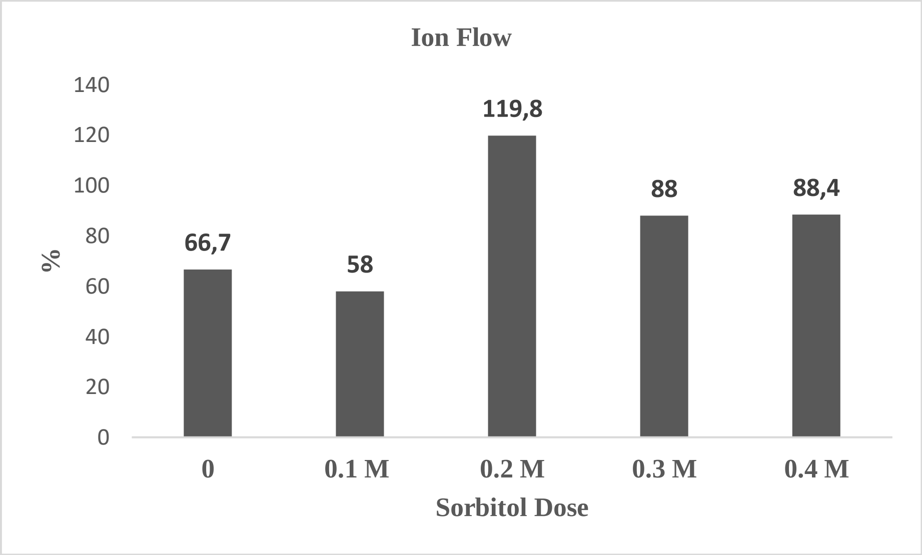

Fig. 6: Effect of different doses of sorbitol applications on ion flux in Balıkçı Siyahı grape type *in vitro* (Akın, 2022)

In the study, a decrease in plant viability was observed based on increasing sorbitol concentration. The findings of plant viability rate obtained from this study are supported by the research findings of Hassan ve Bekheet (2008), Abu-Romman (2010), Karimi ve ark. (2012) and Gecene (2020). Meşe and Tangolar (2019) determined that the degree of damage increased with drought due to increasing PEG doses in their *in vitro* study on American grapevine rootstocks. Gecene (2020) created artificial drought stress by adding five different doses of Polyethylene Glycol (PEG) (0, 1.5, 3.0, 4.5, 6.0) to the MS nutrient medium. The effect of PEG doses on the degree of damage was found statistically significant, and it was determined that the degree of damage increased with the increase of PEG doses. The same researchers determined the shoot tolerance rate as 1.00 at 0% PEG dose in the experiment. Researchers have also found a decrease in tolerance rates depending on the increase in PEG doses. The change in leaf turgor weight due to the increase in drought stress of the same researchers was similar to the results of this study. Albiski et al. (2012) induced artificial drought stress in eighteen potato cultivars (*Solanum tuberosum* L.) with the addition of 2, 4, 6, 8 and 10% sorbitol doses *in vitro*, and they found that the plant water content decreased with increasing sorbitol dose. M. Harun-Or-Rashid et al. (2021), obtained similar results of these researchers. Gecene (2020) found a decrease in the proportional water content of the explant with the increase of PEG doses in their *in vitro* research on grapevine. Karimi et al. (2012) found that the leaf water content of almond cultivar and rootstock decreased with increasing PEG concentrations. Kılıcarslan et al. (2020) also examined the effects of drought stress on beans and determined that the proportional water content of the tissue decreased in water deficiency. Gecene (2020) determined an increase in ion flux value in drought-related plants with the increase of PEG dose.

3. CONCLUSION

In the experiment, it was aimed to determine both the most effective dose of sorbitol, which provides the opportunity to create an artificial drought stress *in vitro*, and the tolerance of fox grapes to drought stress by using 2-3 cm long micro cuttings obtained from Balıkçı Siyahı grape type (*Vitis labrusca* L.). Damage and drying of leaves, necrosis of shoots and stems were observed with increasing sorbitol dose. In the study, it was determined that plant viability, plant length, number of nodes and leaves on the shoot, fresh and dry weight of shoots and damage degree decreased due to the increase in sorbitol dose compared to the control application. In terms of these properties, it was determined that 0.3 M and 0.4 M sorbitol doses were more effective than other sorbitol doses. In leaf turgor weight, the highest damage was obtained from 0.1 M sorbitol application. The shoot tolerance rate was found 0.641 in the 0.4 M sorbitol application. The proportional water content of the explant decreased with the increase of sorbitol doses and it was determined that 0.1 M, 0.2 M, 0.3 M and 0.4 M sorbitol applications were in the same statistical group. The highest values in ion flux were obtained from 0.2 M sorbitol application. With the application of 0.4 M sorbitol, the amount of chlorophyll (9.56) was found reduced by about half as compared to the control (18.86). As understood from the research findings and previous studies, drought stress is among the important factors that negatively affect many physiological and metabolic events in plants and reduce product yield and quality. It is recommended that nutrient media containing 0.3 M or more sorbitol be used to create artificial drought.

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CHAPTER 5

EVERY ASPECT OF VINE LEAVES

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INTRODUCTION

The leaf is one of the most important organs of the plant that undertakes the most basic functions. Whether it is the cotyledon leaves of a plant grown from seed or the true leaves of a plant that reproduces vegetative, the appearance of a green leaf always appears as a sign of vitality. In addition, especially for plants pollinated by insects, colorful and beautiful petals contribute to pollination and reproductıve biology, contributing to the continuity of the generation, thanks to their attractive structur.

Looking at the definition of leaf in general terms; They are known as growth-limited structures that function in photosynthesis, transpiration and respiration in plants, emerging from the nodes on the trunk side shoot. However the leaf is much more than that. Sometimes it is a marker in determining the deficiency or excess of plant nutrient elements, observing the harmful effects of diseases, sometimes a defender with the mechanisms it develops against stress factors, and sometimes it is a home for beneficial microorganisms. In this section, the importance of an organ that is so important for the plant for vine will be tried to be explained.

1. VINE BIOLOGY AND VINE PHYSIOLOGY

Vine leaves; It is a flattened organ perpendicular to the shoot axis, attached to the nodes on the summer shoot. It contains a large exterior, an abundant ventilation system and a large number of chloroplasts. Considering that the main task of the leaf is photosynthesis and transpiration, the large surface of the vine leaves is very advantageous (Ağaoğlu 1999). Vine leaves are used to distinguish and classify varieties not only by their intrinsic properties, but also by differences in color, shape, and growth habits (Chitwood and Sinha 2016).

1.1. Origin of Leaves and Their Arrangement on the Shoot

When the origin of the leaf is examined, the concept of structure formed by the differentiation of the apical meristems is encountered. Formation of leaf primordium begins with periclinal divisions under the apical meristem in the vegetation cone, and these divisions occur in layers close to the surface. At first, a flat protrusion is formed with the dividing cells, and then this protrusion shows double suffocation and forms the lower (hypophyll) and upper (epiphyll) parts of the leaf. Later, the pituitary differentiates to form the leaf sheath and stipule, and the epiphyll differentiates to form the petiole and palm. The growth of intermediate meristerms at the base of the upper part of the leaf occurs in the petiol (Yentür 1984; Ağaoğlu,1999). Different leaves of plants in morphological sense are named with the term 'phyllom'. Phylomas are classified in four parts in terms of their morphological and physiological structures and the tasks they undertake (Yentür 1984; Ağaoğlu 1999). These are briefly;

Foliar leaves; they are the main organs that perform photosynthesis and transpiration. Also called assimilated leaves.

Cataphylls; these leaves, called the bottom leaves, are located at the bottom of the shoot. Cataphylls are vestigial leaves that are either prematurely developed or inhibited (Yentür 1984). The scales on the eyes and buds are examples of these.

Hypsophiles; they are called cover leaves or upper leaves. These are the vine flower brackets the ones found in the vine are white in color and their functions are similar to those of the petal.

Cotyledons; they are described as the first leaves or the sepals. The first two cotyledons seen during seed germination represent this group. The cotyledons in the vines can be of simple structure, different forms and colors (Zankov 1981; Currle et al. 1983).

When the origin of the leaf is mentioned, it is understood that the drafts located at the nodes of the shoot model in the buds and showing a certain arrangement on it. As the shoot grows and lengthens in the vine plant, the apical bud at the tip of the shoot ensures that new leaves are constantly formed, as well as other organs (Çelik 2011).

Vine leaves show a spiral arrangement on summer shoots. In this arrangement, there is a leaf at each node and these leaves are arranged in a spiral (helical) manner around an axis. The fixed and definite intervals of the leaves arranged on the shoot axis are called divergence. In a spiral vine, only one leaf emerges from each node, and two successive leaves never overlap. Hence divergens of successive leaves are never zero. The angle between the projections of two successive leaves and the axis is called the divergence angle (Çelik 2011). Leaf divergence on the vine;

$D=P/N$ is expressed with

D= divergence

P= the number of revolutions (the number of turns until you get the overlapping leaves that are aligned)

N= number of leaves (the number of leaves counted from any leaf, following the spiral direction, spiraling on the axis until it reaches another aligned top leaf).

Leaf divergence 2/5 or 3/7 on the young shoots of the vine formed from the newly germinated core; Leaf divergence is 1/2 on 1 year old branches and mature shoots (Viala and Vermovel 1970; Ribereau-Gayon and Peynaud 1971; Gohan 1983; Yakar 1983).

2. Vine Leaf Physiology

In vine physiology, basic concepts such as seed physiology, growth and development physiology of vines, physiological functions of vines, vine-water relationship, photosynthesis, respiration, transpiration, mineral substance uptake, transport and use, carbon distribution, as well as basic physiological events such as stress physiology are examined. The vine leaf is involved in many of the physiological events mentioned above. In this section, the processes of transpiration and photosynthesis, in which the vine leaves are based, have been studied in a fundamental way.

2.1. Transpiration

As with other plants, vines lose most of the water they take from the environment in which they develop and grow throughout their lives. Water losses in vine occur in the form of transpiration, guttation and bleeding. Gutation is the loss of water in the liquid form from the holes of the leaves, called hydatids, and bleeding, especially on the wound surfaces of late pruned vines. The loss of water in grapes by evaporation is called transpiration. Transpiration means the loss of water in the form of steam through the pores (stomas), leaves, and the cuticle-covered epidermis of young organs (Kacar 1989; Çelik 2007).

In a typical grapevine leaf, only 5-10% of the total transpiration occurs in the cuticle, however the cuticle ratio can reach up to 30% in older leaves, which have a lower transpiration rate than younger leaves (Boyer et al. 1997; Keller 2010). Although the shoots and even the lignified plant parts of the vine/vineyard stem lose some water by transpiration from the bark, the main water loss occurs through the leaves. Leaf juice comes from the roots via the xylem portion of the vascular bundles and spreads widely throughout the leaf like an intricate network. In this way, the distance between the xylem main vein and the outer surface of the leaf meristem cells surrounding it is short, and there is no obstacle for water flow. Principal transpiration occurs in the outer cell walls at the intercellular boundary. The air-filled spaces around the leaf are in contact with the microscopic openings in the epidermal layer, and these are called stomata (Eşitken 2020). Although the stomata cover less than 5% of the leaf surface, the rest of the water is lost from here. For this reason, stomata have a very important role in regulating water loss by diffusion in the leaf. Acting as a pressure regulator, stomata control the flow rate, limiting pressure variations and responding very sensitively to environmental variations (Sperry et al. 2002; Keller 2010). In addition, stomata can be regulated in response to environmental events such as soil water status and plant nutrient deficiency and control water loss. Leaves consistently find a compromise between minimizing water loss by transpiration and maximizing CO_2 uptake and photosynthesis. The stoma is the strongest controller of this delicate balance. (Lombardini 2006; Eşitken 2020).

Various factors affect transpiration. These factors are examined under internal and environmental (external) factors. Internal factors; Under the title of vegetative factors, the effect of the morphological structure of the vine fleece on water loss, the effect of the anatomical structure of the vine leaf on water loss, the structure of the stomata and the water loss from them and the factors affecting the opening and closing of the stomata. Environmental external factors are light, air humidity, temperature, wind. It is examined under five headings, namely the movement of soil water and the usefulness of soil water (Çelik 2007; Kacar 2015).

2.1.1. Environmental Factors

Light; light is mainly effective for opening and closing stomata. Since the stomata are closed in the dark, the amount of water lost in the form of steam decreases significantly, while the water loss in the form of steam increases as the stomata will open in the leaves of the vines under the light.

Air humidity; the effect of air humidity on transpiration is that all or nearly the entire atmosphere inside the leaf is assumed to be saturated, and the air around the leaf is unsaturated. For this reason, a pressure gradient occurs from the inside of the leaf to the outside, and as long as the stomata are open, gases such as CO_2 and O_2 as well as water vapor pass between these two environments.

Temperature; As for the effect of temperature on transpiration, stomata close as the temperature approaches 4°C in the vine leaf, in photosynthesis they show a maximum opening of 25-30°C, which is the optimum temperature value, and gradually closes after 30°C (Kaçar 1989; Çelik 2007).

2.1.2. Internal Factors

In addition to the significant differences in transpiration in various plants grown under the same conditions, similarly, transpiration also differs between different organs of the same plant. Vine is a type of plant with culture, rootstock and wild forms. It is normal for this species, which has thousands of varieties, to show variability in terms of transpiration. The reason for these variations is the differences in the morphological and anatomical structure of the leaf, as well as the structure of the stomata and water loss from them and factors affecting the opening and closing of the stomata.

The effect of morphological structure of grape leaves on water loss; The fact that the vine leaf is covered with hairs, the intercellular spaces are different in volume and the sap is in different density, the differences in stomatic reactions cause water loss at different rates in the vine by transpiration. In general, the anatomical structure of the grape leaves is similar to each other. In addition, in two vines of the same age and the same size, the vine with a large leaf area loses more water than the vine with a narrow leaf surface. The water lost from the unit area is less in the wide leaf and more in the narrow leaf area. The different size of the leaves in the vine also affects the water uptake from the soil by the roots. For example, in the vine with the same root surface, the water uptake by the roots is very high in the small-leaved vine. The fact that the underside of the leaf is covered with different types of hairs in vines and that they cover the stomata like a felt cover also reduces the evaporation of stomata to some extent (Winkler et al. 1974; Çelik 2007).

The effect of vine leaf anatomical structure on water loss; the loss of water in the form of water vapor in the vine occurs mostly in the leaves. The vine leaf has an anatomical structure suitable for absorbing the gases it needs for its physiological activities and giving the water to the atmosphere in the form of steam. The lower epidermis of the grapevine leaf is covered with numerous small pores called stomata. The number of stomata on leaves varies according to species and cultivars. In general, the number of stomata in vine leaves per mm² varies between 50 and400. While the number of upper stomata is 250 mm²/piece in *V. vinifera* and some rootstocks, this amount is maximum 300 mm²/piece in *V. labrusca* and *V. cinereda* (Liu et al. 1978; Du ¨ring 1980; Scienza and Boselli 1981; Keller 2010).

Structure of stomata and water loss from them; Stomas are concentrated on the lower surface of the leaf and mostly around the main veins in the vine. Stomata are structures in the epidermis of plants that are small enough to be expressed in microns. When the stomata are opened, the oxygen in the intercellular spaces of the leaf is replaced by carbon dioxide and water vapor. Gas inflow and outflow are largely prevented when the stomata are closed. The opening and closing of the stomata occurs with the movement of guard cells (a special kidney-shaped structure) and auxiliary cells. Guard cells have greater cytoplasmic coverage than epidermal cells. Kidney-shaped guard cells have a thicker wall facing the stomata hole thanks to this thickening, when the cell turgor comes, the walls of the closing cells are stretched and the stomata are opened, water loss occurs with the escape of water vapor from the intercellular space (Çelik 2007).

Factors affecting the opening and closing of stomata; It is known that many factors are effective in the opening and closing of stomata. These factors are briefly mentioned.

Light; It is expressed as the most important factor in the opening and closing of stomata. The stomata are open on the leaves of vines in the light, and closed in the dark.

The mechanism of action of light on the opening and closing of pores is explained in two theories (Ribereau ve Reynold 1971; Kacar 1989; Çelik 2007). The first of these is the CO_2 and pH theory, and the second is the osmotic pressure theory. To put it simply in these two theories; starch in guard cells to water-soluble sugar; It is based on the principle that sugar turns into starch.

Water content of the leaf; leaves are the organ most affected by water deficiency. Especially in spring and summer, there is a lack of water on sunny, clear and hot days. The reason for this is that the water lost by transpiration cannot be adequately taken from the soil. This leads to a decrease in the water potential of the cells. Decreased water potential in leaf cells causes a wilting known as Incipient Wilting, although it is often invisible (Kacar 2015). With decreasing water content and lower turgor pressure, guard cells close the pores. When the water content in guard cells decreases, it leads to a decrease in the pH of the cell sap and thus to the conversion of water-soluble sugar to starch. This lowers the osmotic pressure of the guard cells and causes the pores to open (Çelik 2007).

Temperature; As mentioned in the section examining the relationship between transpiration and the environment, stomata opening increases when the temperature rises up to 25-30°C when the temperature rises and other conditions are assumed to remain the same, while stomata close in case the temperature rises above these values.

Cell Sap Density; Since it affects the turgority of guard cells, the density of the cell sap affects the opening of the stomata, albeit indirectly. In leaves with a high density of cell sap, the pores remain less open and the transpiration is less (Çelik 2007).

2.1.3. Transpiration-Reducing Agent

Substances that reduce transpiration are called antitranspirant substances. Transpiration-reducing substances can be examined in three ways; antitranspirants that close stomata, antitranspirants that form a thin film layer, and antitranspirants that are reflection type. Antitranspirants that close stomas have the feature of closing stomata directly. Fungicides such as PMA (Phenyl Mercuric Acid) and low concentration herbicides such as atrazine act as antitranspirants by promoting growth promoters stomata closure. Antitranspirants, which form a thin film layer, form a thin film coating on the leaf surface, preventing water vapor loss from the leaf. When applied to the leaves, the stomata close and therefore water from the plants reduces transpiration losses. Waterproof films on the leaf surface, these layers reduce the outflow of transpiration from the plant surface. The materials that make up this layer are silicone oil and low viscosity; waxes, silicones, octadecanol, folicote, steam gard, hexadecanol and pinolene are film coating type antitranspirants. Antitranspirants, which are reflective types, are typically white in color and have the ability to reflect light. Its mode of action is to reduce water loss from the leaf by lowering the leaf temperature. Kaolin, lime water, calcium bi-carbonate can be given as examples of these antitranspirants, which are useful in increasing the distribution of light and photosynthesis in the crown (Kacar 2015; Dündar 2019).

2.2. Photosynthesis

Photosynthesis is the conversion process of the energy necessary for the survival of all plants, including the vine. Plants make organic substances necessary for themselves by reducing the carbon dioxide they take from the air with a certain energy. In viticulture, sugar, which is the raw material of products such as fresh grapes, raisins, wine and alcohol, is the result of photosynthesis in the vine leaf, which is the total soluble dry matter. The reaction in which photosynthesis takes place is formulated as follows.

$$6CO_2 \; + \; 6 \; H_20 \xrightarrow[\text{Chlorophyll cell}]{\text{673 Kcal solar energy}} C_6H_{12}0_6 + 6O_2 + 6H_2O$$

Photosynthesis, which was formulated above in its simplest form, actually takes place with many complex events and stages. Photosynthesis is affected by vegetative (internal) and environmental (external) factors. Factors such as leaf structure and chlorophyll content, leaf age, amount of product and number of leaves, the effect of enzymes in the cell protoplasm and accumulation of photosynthesis end products are vegetative factors; light, temperature, water, oxygen density, carbon dioxide density and chemical substances constitute environmental (external) factors (İştar 1959; Kliewer 1981; Yakar 1985, Kacar 1989; Çelik 2007).

2.2.1. Internal Factors

Leaf Structure and chlorophyll content; The vine leaf has a larger leaf area than many other plants, so it benefits more from sunlight. In terms of anatomical structure, the vine leaf shows a structural feature that makes photosynthesis the best. Structural features such as stomata structure, distribution and size of intercellular spaces, structure of palisade and mesophyll parenchyma, cuticle and epidermis thickness, and development in the structuring of leaf inner vascular bundles are extremely suitable for the leaf to perform maximum assimilation as long as the environmental conditions are suitable.

As leaf cells enlarge and the rate of cell division decreases, they also form their photosynthetic machinery, namely, chlorophyll-rich chloroplasts (Keller 2010). Photosynthesis takes place in chloroplasts. After absorbing light energy and carbon dioxide, oxygen is removed and carbohydrates are formed entirely within the chloroplasts. Chloroplasts are cytoplasmic particles found especially in the mesophyll cells of the leaf. The more chloroplasts in a tissue, the higher the rate of assimilation and the higher the production of carbohydrates. Therefore, there is a linear relationship between photosynthesis and chlorophyll content. (Çelik 2010).

2.2.2. External Factors

Light; photosynthesis Greek for photos: light; synthesis: consists of the combination of synthesis words. In other words, light is a process in which sunlight is converted into chemical energy in photosynthesis and used to produce (synthesize) organic compounds within the plant from inorganic compounds obtained from the outside of the plant (Keller 2010). Different quality and intensity of light affects photosynthesis differently. The effect of light intensity on the rate of photosynthesis is significant. Producers try to make the most of the light of the vine with applications such as determining the location while establishing their vineyards, and pruning and removing leaves during cultivation. Light with a visible wavelength (400-700 nm) is used in photosynthesis. Before light energy can be used, the light must be absorbed and converted into a flow of electrons derived from the splitting of water. Chlorophyll is the main light-absorbing pigment that makes it possible to capture light energy. It is the most abundant pigment in the world. Chlorophyll is green in color because it predominantly absorbs blue (430 nm) and red (680 nm) light and reflects most green light with wavelengths between blue and red (Keller 2010). 150-200 w/m^2 of sunlight is required for the vines to assimilate at the highest level. However, for this light to have a certain intensity, it must come at a right angle to the surface of the vine leaf. There is a positive relationship between light intensity and net photosynthesis rate. It has been observed that the photosynthetic capacity of the vines grown in greenhouse conditions, the photosynthesis rate increased until the saturation point by increasing the light intensity in the greenhouse, but did not increase in any way after the saturation point (Buttrose 1969; Çelik 2007)

Temperature; The temperature is constantly changing from month to month, day to day, and even throughout the day, and plants spend most of the day at temperatures below or above the optimum temperature required for photosynthesis. An increase in temperature up to a certain level also increases photosynthesis, provided that light, carbon dioxide and other factors are not limiting. It has been reported that suitable temperatures for photosynthesis in most plants grown in temperate climate regions are between 20-30°C (Kaçar 1989). The temperature required for photosynthesis in vines can be examined in 3 parts. The first of these is the temperature that is expressed as low temperature and is below 20℃. Photosynthesis rate is lower than photochemical activity and reduction of carboxylase enzyme. Secondly, it occurs at temperatures between 25-30℃, which is expressed as Normal (optimal) temperature. It is expressed as the temperature at which maximum photosynthesis takes place. Thirdly, it is the temperatures between 30-45℃, which are specified as extreme temperatures. Between these temperatures, the amount of photosynthesis decreases rapidly (Hall et al. 1987; Çelik, 2007). It has been stated that an increase in temperature decreases the need for triose phosphate rapidly and rapidly decreases the synthesis of starch and sugar (Akman et al. 2000), while excessive increase or decrease in temperature decreases photosynthesis and stomata strength in grapevine (During 1994).

Water; Water, the basic substance of photosynthesis, is not only a factor that initiates and directs important intracellular and extracellular reactions. At the same time, it is of great importance in the opening and closing of stomata, influencing the wilting of the leaves. The water, which we express simply in the formula used in photosynthesis, combines biochemically with the carbon dioxide taken from the roots. As a result, monosaccharides are formed and 99% of the remaining water is lost by transpiration (Hall et al. 1987, Çelik 2007).

Density of oxygen; It is known that excess oxygen (O_2) in the environment retards photosynthesis in plants. This negative effect of oxygen excess on photosynthesis is called the Warburg effect. It is stated that photosynthesis is high in environments with oxygen content of less than 21%, and that photosynthesis decreases at around 21% oxygen (Kacar 2015).

Density of carbon dioxide; All of the carbon dioxide used by green plants in nature is taken in the form of soluble carbon dioxide, carbonic acid or a salt of this acid and reaches the chloroplasts. The main source of carbon dioxide for plants living on land is the atmosphere (Kacar 2015). The rate of CO_2 in the atmosphere is constant and is always 0.03%.

Chemicals; Apart from the macro and micro nutrient deficiencies, the imbalances in the amounts of the mentioned elements also affect the photosynthesis rate negatively and affect this complex event indirectly and directly. Effect of plant nutrient deficiencies on net photosynthesis; negatively affecting chlorophyll synthesis, decreasing the electron transport capacity, limiting other enzyme activities including carboxylating enzymes, negatively affecting the opening and closing of the pores and increasing respiration. At the end of the long interaction, the total photosynthesis rate decreases due to the shrinkage in leaf surface areas in plants with plant nutrient deficiencies.

3. Morphology of the Leaf

Although morphology is known as morphology, morphological features are used to define an animate and inanimate entity. If we start from this definition, similarly, the grape leaves show a structure specific to the variety and the leaf of each variety has a unique morphological structure. The morphological difference of the vine leaf is used as a measure in the identification and classification of the cultivars from the leaf in ampelography (Çelik 2011).

Leaf is a flat-shaped organ that grows perpendicular to the shoot axis, attached to the nodes on the annual shoot. Basically, the leaf consists of 3 parts. These; leaf blade (lamina), petiole (petiole) and leaf sheath (=auricle: stipula) (Figure 2.) (Ağaoğlu 1999). The form, size, color of the leaf, whether the leaf surface is smooth or carved, hairy or hairless, varies according to the variety, ecology, growing conditions and the location of the leaf on the summer shoot (Ağaoğlu 2011).

The petiole expands towards the basal and is called 'leaf sheath' in the part where it joins the branch. The veins of the same thickness, coming out of the bottom of the petiole, go to the leaf pieces (slices) as main veins and they divide into thinner veins and form a netted structure is defined as 'palmette ' veining (Çelik 2011).

The leaflets at the tip of the shoot are very hairy, yellowish, reddish or bronze in color (Oraman 1972). They are usually smaller and more sliced, with the basal leaf. The characteristic features of leaves between the 5th and 12th node are much more constant. Galet (1956), taking these features into account, grouped the leaves into three categories.

Mature leaves; 8-11 from basal on summer shoot. It is the leaves at the node and the best time to observe and measure on them is the period of I fall or the period when the shoot growth stops (Ağaoğlu 1999) (Figure 1.).

Young leaves at the tip of the shoot; until the end of the growth of the summer shoot, the first three leaves at the end are important organs in ampelography (Ağaoğlu 1999) (Figure 1.).

Young leaves at the shoot base: It is the first three leaves that are seen during the period of the primary bud (Ağaoğlu 1999)(Figure 1.).

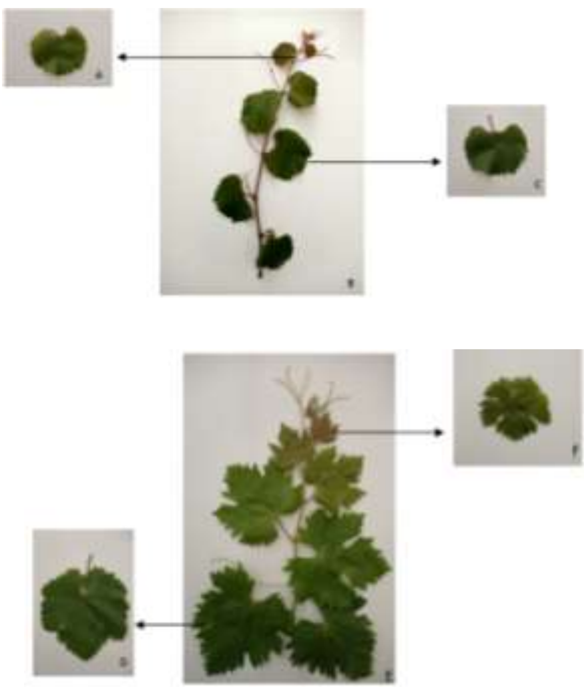

Figure 1. Young and mature leaves of 110R rootstock and Narince grape variety. A: Young leaf of 110R rootstock, B: shoots of 110R rootstock, C: mature leaf of 110R rootstock. D: Young leaf of Narince grape variety, B: shoots of Narince grape variety, C: mature leaf of Narince.

The color of the vine leaves can be light green, yellow-green, dark green or reddish. The leaves of grape varieties with red or darker fruits turn red in different tones in autumn when the growth ends; in white varieties this color turns yellow (Ağaoğlu 1999).

Leaf shape is one of the most conspicuous features of plant morphological diversity. The shape of the leaf may differ according to climate, geological time criteria, developmental differences, environmental and physiological effects (Wilf 1997; Peppe et al. 2011; Schmerleret al. 2012; Edwards et al. 2016).

In the morphological description of the vine leaf, 5 main shapes are emphasized. These are;

Heart Shaped Leaves; this leaf-shaped main vein (L_1) is longer than the other veins, the leaf length is greater than the leaf width. This leaf shape is common in grapevine and American species. For example; *V. armata, V.betulifolia, V. Rupestrist* (Rupestris du Lot) are a few (Çelik 2011).

Pentagon Shaped Leaves; the pentagonal leaf has a 5-pointed or sometimes square-like appearance. The length of the leaf is greater than its width. *V. Riparia, Riparia* x *Rupestris* hybrids, 41 B rootstock, Merlot, Hamburg Musket, Cardinal varieties are some examples (Çelik 2011).

Triangular Leaves; this leaf shape is a leaf shape formed by the change of pentagonal leaf. The angle between the veins is wide. *V. aestivalis* is given as an example (Çelik 2011).

Round Shaped Leaves; although they look round, they are leaves that are not completely round in the circular sense. It takes a round shape where the petiole joins the stem. *Vitis vinifera* leaf types fall into this group. As a few examples; Syrah, Pinot noir, Chardonnay, Gamay, Riesling, Italia, Alexandria Musk etc. can be given (Çelik 2011).

<u>*Kidney Shaped Leaves*</u>; In this leaf type, the width of the leaf is greater than its length. For example; *V. Rupestris* (Rupestris du Lot). Apart from these five shapes found on vine leaves, there are intermediate types. These intermediate leaf types are mostly seen in hybrid varieties (Çelik 2011).

<u>*Leaf size;*</u> When looking at Vitis species, very small leaves are encountered in terms of leaf size, as well as very large leaves. Although the leaf size is a feature specific to the variety, it also differs according to the fertility

of the soil, the strength of the vine, the pruning method and climatic conditions (Çelik 2011).

The size of the vine leaves is evaluated in 5 groups according to the size of the leaf blade (Fidan 1966; Oraman 1972; Galet 1979; Galet 1983).

- Very small leaf (*V.rupestris*) < 75 cm^2
- Small leaves (Grenac noir) = 76-149 cm^2
- Medium leaf (Chenin blanc) = 150-299 cm^2
- Big leaf (Carignane)= 300-600 cm^2
- Very large leaf (*V. Coignetiae, V. Riparia*) > 600 cm^2

Leaf size in the vine is explained by the leaf area. However, the area of leaves multiplied by the aspect is not the actual area. Said area is the area of a square or rectangle. In order to find the real area of the leaf, either area measuring devices should be used or an area coefficient should be used (Çelik 2011).

Leaf Slices and Sinus; In vines, the leaf can be 1-part (whole), 3-part (lobed), 5-part or rarely 7-part. In most of the cultivars, the leaf is 5-part and the edges are toothed. The palm of a mature vine leaf has a whole or different number of slices, and there are sinüs between the slices. Most of the varieties that fall under the *Vitis vinifera* species have 5 slices. The main vein coming from the petiole is divided into 5 from the place where the stem is attached, and each vein goes into a slice (Çelik 2011). There is a stalk sinus where the petiole joins the foot part. The sinus petiole can be 'V' or 'U' shaped, as well as open, closed, (O), overlapping (Ağaoğlu 1999). Many of these features are variants.

The vine leaf is in a symmetrical form. As seen in Figure 2., the slice in which the L_1 vein spreads is called the apical lobe (AL), the slices in which the $L_2L'_2$ veins spread are called the superior lobe (SL), and the slices in which the $L_3L'_3$ veins spread are called the inferior lobe (İL).

The space between the upper slices and the end slices is called the upper sinus, the upper side, and the space between the slices and the lower side slices is called the lower sinus (Çelik 2011).

Leaf Teeth; The edges of the vine leaf have a very thin or thick-sectioned appearance. Although these structures, called teeth, are between 30 and 60 in a leaf, it is possible to come across varieties with 120 teeth in some varieties. Due to their structure, they can be pointed or blunt in general (Ribereau- Gayon and

Peynaud 1971; Yakar 1983; Ağaoğlu 1999; Çelik 2011). Leaf teeth can be of the same size and shape around the leaf, or they can show an irregular structure of different sizes (Çelik 2011). According to the shape of the teeth; they are examined under 4 groups as pointed teeth, concave teeth, convex teeth and sickle-shaped teeth (Galet 1979).

Hairiness on Leaves; the hairiness of grapevine leaves is very important in ampelographic identification. As the leaves get older, hairiness generally decreases (Ağaoğlu 1999). Hairiness in vine leaves is examined in 5 groups according to their appearance and structure (Oraman 1972; Galet 1979; Anonymous 1983; Çelik 2011).

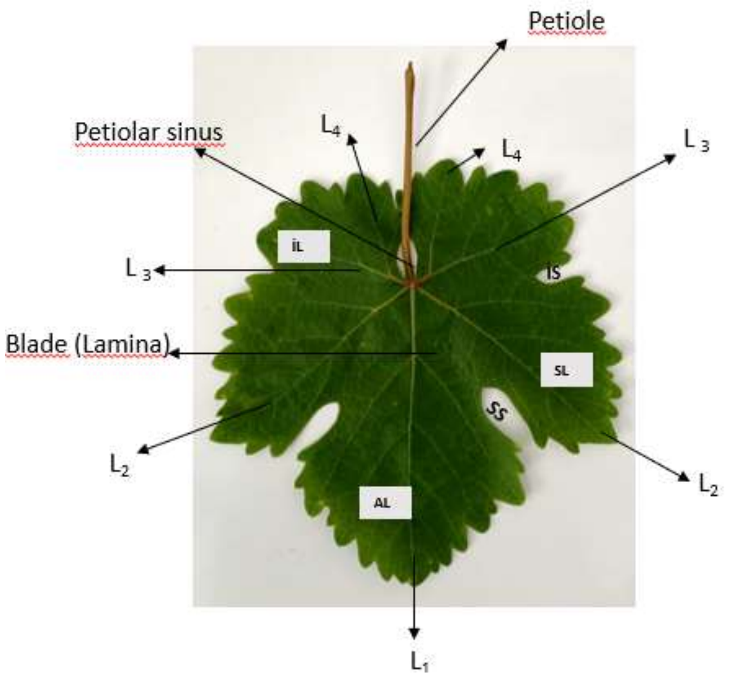

Figure 2. Parts of a mature vine leaf. AL: Apical Lobe; SL: Superior Lobe; IL: Inferior Lobe; SS: Superior Sinus; IS: Inferior Sinus; L_1, L_2, L_3, L_4 Main veins

- Woolly hairiness; It is called hairiness, which occurs when thin-long hairs spread along the main veins on the lower surface of the leaf and mingle with each other, forming frequent or sparse soft groups.

- Felt-like hairiness; it is the fact that the leaf surface is covered with very dense (felt-like) hairs. Since the hairs are very dense in this type of hairiness, the natural color on the underside of the leaf is sometimes invisible, and the color appears white or grayish due to the hairs.
- Quince-like hairiness; It is called hairiness, which is formed by very short, thin and soft yellowish-gray hairs covering the surface of the leaf very often.
- Cobweb-like hairiness; it is called hairiness, which is formed by the spread of thin long hairs on the surface of the leaf like a spider web.
- Brush-like hairiness; in this type of hairiness, the hairs have the appearance of thick and hard brush or soft velvet structure.

4. Anatomy of Leaf

There are great differences between the morphological and anatomical structures of grape leaves. The leaf blade, which is the basic part of the photosynthetic tissue in vines, has a flattened structure. The upper surface of the leaf blade is called 'ventral' and the lower surface is called 'dorsal' (Ağaoğlu 1999). Leaf is the organ where events such as photosynthesis, transpiration and gas exchange take place. When the cross-section of the leaf is taken, there is an order as follows from the upper surface to the lower surface (Çelik 2011).

- Cuticle; It is a cutin layer and covers the surface of the lower and upper epidermis. This layer is covered with waxy substances to prevent water loss from the leaf.
- Epidermis; in the vine leaf, the upper and lower epidermis is completely covered with cuticle and waxy substances, but there are stomata and leaf hairs in addition to this structure in the lower epidermis.
- Mesophyll; It is located between the lower and upper epidermis and consists of two types of parenchymatic tissues, palisade and sponge parenchyma. Palisade parenchyma; It is the part that is located just below the upper epidermis and contains plenty of chlorophyll and where the main assimilation takes place.

  Sponge parenchyma; they are located just below the palisade parenchyma and their cells are scattered. Since they have a spongy structure and the intercellular spaces are quite large, they facilitate the entry and exit of respiratory gases and water vapor.

- Stomas; in grape leaves, stomata are located on the lower surfaces. They control water loss by providing the exchange of gas and water vapor collected as a result of the physiological activity of the vine leaf.
- Intra-leaf vascular bundles; Phloem and xylem vascular bundles are lined up in the main veins spreading in the leaf blade. The vessels are composed of more than one vascular bundle. Raw and assimilation materials are transmitted to all parts of the leaf by means of veins scattered all over the leaf by branching to form a very fine network system.
- Epidermis hairs; the hairs on the vine leaf are located on the lower surface of the leaf. As mentioned before, the structure and appearance of the feathers vary according to the varieties.

5. Chemical and Phytochemical Content of Vine Leaf

There are hundreds of varieties of *V. vinifera*, which were cultivated thousands of years ago, with quality and characteristics suitable for different evaluation methods. When it comes to viticulture, the first product that comes to mind is grapes and it is evaluated as table, wine and drying. However, in the Mediterranean basin, especially in Turkey, it is seen that grapes are consumed by preserving them in brine, as well as different ways of using grapes (pestil, kome, pickles, grape juice, pickled vermicelli, grape tarhana). In recent studies, it has been seen that grape leaves are rich in bioactive substances, especially phenolic compounds, and its consumption in human nutrition has begun to increase not only in the Mediterranean basin but also in the globalizing world.

Vine leaves have a very rich and diverse chemical composition. Vine leaves contain organic acids, amino acids, sugars, some vitamins and phenolic compounds (Ribereau and Reynold 1971). Previous studies have shown the presence of low-calorie, dietary fiber, various organic acids, phenolic acids, flavones, procyanidins, vitamins, anthocyanins, minerals, oils, enzymes, terpenes, carotinoids, amino acids, reducing and non-reducing sugars in grape leaves (Ribereau et al. 1971; Hebash et al. 1991; Şendoğdu et al. 2006; Fernandes et al. 2013; Güler and Candemir 2014). Vine leaves are rich in insoluble fiber. For this reason, grape leaves are a food product that is an important source of fiber (Mürtezaoglu 2015).

The leaves' phytochemical constituents which have been recognized to date are isoprenoids, organic acids, steroids, anthocyanins, sterols, phytoalexin pterostilbene, leucoanthocyanins, rutin, quercitrin, isoquercitroside, kenferol, luteolol, esterifies and free fatty acids, tannins, vitamins, enzymes, and heterocyclic compounds (Langcake et al. 1979; Guidoni et al. 1997; Hmamouchi et al. 1997; Felicio et al. 2001; Liakopoulos et al. 2006; Batouska et al. 2008).

The nutritional value of grape leaves, which is included in our daily diet as a product of Anatolian grape culture, is reported in the literature as components in 100 g. 2.12 g total fat, 1.06 g polyunsaturated fat, 0.08 g monounsaturated fat, 93.02 calories, 5.6 g protein, 17.31 g carbohydrates, 11.00 g dietary fiber, 6.3 g sugars, 9 mg sodium, 363.08 mg calcium, 2.63 g iron, 91.02 mg phosphorus, 11.10 mg vitamin C, vitamin A is reported as 1376 ıu (Anonymous 2006b). Anonymous. 2006b. www.sigma-logic.com/nutritional_calculator.html.

In the study of Maia et al. (2021), in which they investigated the nutrient content of the leaves of 7 different grape varieties, macro and micro nutrients differ between varieties. Na, Ca and K concentrations are seen to have the highest values in all varieties (Figure 3.).

In addition to being prepared by using the leaves of the varieties grown in that region, the leaves of the varieties grown in that region, especially the leaves of Narince in Tokat, sultani seedless in the Aegean region, Yapıncak grown in Thrace and Emir in Nevşehir, are more popular in the national and international markets. Mürtezaoğlu (2015), examined the general properties of three different pickled leaves produced in Izmit, Tokat and Izmir regions and determined their nutritional values. It was determined that the protein content of 9 pickled leaves varied between 11.62 – 24.79%, fiber content between 48.70 – 58.36% and ash content between 6.32 – 11.89%. It has been reported that the water-holding and oil-holding capacity values of the cottony parts of the leaves, especially those belonging to the Tokat and Izmit regions, are quite suitable for functional use. He suggested that the glucose adsorption (7.896 - 7.537mmol/L for 100mmol/L) and α-amylase activity (7.15 - 18.27%) values of the insoluble fiber-rich parts of the leaves were close to the insoluble fiber pulp obtained from different sources.

Element (µg/g FW)	*Vitis vinifera* cv. Trincadeira	Cabernet Sauvignon	Pinot Noir	Tinta Barroca	Castelão	Bastardo	C19
Na	491.72 ± 139.27 ^{a}	661.66 ± 74.64 ^{ab}	898.26 ± 288.80 ^{ab}	746.88 ± 74.99 ^{ab}	1033.40 ± 194.20 ^{b}	371.39 ± 198.90 ^{ab}	671.68 ± 95.52 ^{ab}
Mg	21.59 ± 33.32 ^{a}	14.64 ± 32.72 ^{a}	28.61 ± 39.30 ^{a}	0.00 ^{a}	55.58 ± 57.97 ^{a}	18.51 ± 41.39 ^{a}	10.48 ± 23.44 ^{a}
Cl	21.27 ± 5.69 ^{abc}	29.01 ± 8.31 ^{bc}	9.67 ± 6.19 ^{a}	23.84 ± 6.31 ^{abc}	9.25 ± 3.28 ^{a}	25.08 ± 8.71 ^{abc}	30.02 ± 7.51 ^{c}
K	4046.22 ± 821.37 ^{a}	2817.77 ± 336.38 ^{a}	3446.68 ± 1054.71 ^{a}	3224.14 ± 252.62 ^{a}	3818.64 ± 1174.17 ^{a}	3784.25 ± 552.03 ^{a}	3467.66 ± 194.97 ^{a}
Ca	2080.56 ± 392.42 ^{abc}	1691.01 ± 106.95 ^{a}	1580.98 ± 431.74 ^{a}	2771.00 ± 327.37 ^{c}	1752.66 ± 379.56 ^{abc}	2513.26 ± 143.36 ^{abc}	2590.35 ± 149.68 ^{bc}
Sc	5.84 ± 1.41 ^{abc}	4.61 ± 0.63 ^{ab}	3.59 ± 0.82 ^{a}	6.65 ± 0.72 ^{bc}	4.86 ± 0.74 ^{abc}	6.02 ± 1.10 ^{abc}	8.65 ± 2.17 ^{c}
Ti	0.26 ± 0.25 ^{a}	0.12 ± 0.17 ^{a}	2.39 ± 3.94 ^{a}	0.27 ± 0.28 ^{a}	1.10 ± 0.92 ^{a}	0.00 ^{a}	0.30 ± 0.31 ^{a}
V	0.17 ± 0.11 ^{ab}	0.12 ± 0.12 ^{ab}	0.03 ± 0.06 ^{a}	0.26 ± 0.16 ^{ab}	0.19 ± 0.13 ^{ab}	0.150 ± 0.102 ^{ab}	0.31 ± 0.04 ^{b}
Cr	0.00 ^{a}	0.00 ^{a}	0.02 ± 0.05 ^{a}	0.03 ± 0.06 ^{a}	0.04 ± 0.09 ^{a}	0.00 ^{a}	0.00 ^{a}
Mn	9.30 ± 1.81 ^{abc}	11.77 ± 0.69 ^{abcd}	6.50 ± 0.80 ^{a}	13.01 ± 1.48 ^{cd}	9.08 ± 1.29 ^{abc}	13.24 ± 0.50 ^{d}	12.28 ± 0.42 ^{bcd}
Fe	19.79 ± 6.83 ^{a}	15.99 ± 1.62 ^{a}	14.85 ± 3.16 ^{a}	15.32 ± 1.06 ^{a}	29.26 ± 27.19 ^{a}	15.40 ± 1.01 ^{a}	20.77 ± 1.33 ^{a}
Co	0.16 ± 0.10 ^{a}	0.07 ± 0.06 ^{a}	0.10 ± 0.10 ^{a}	0.06 ± 0.08 ^{a}	0.20 ± 0.23 ^{a}	0.15 ± 0.05 ^{a}	0.10 ± 0.07 ^{a}
Ni	0.13 ± 0.04 ^{a}	0.11 ± 0.03 ^{a}	0.10 ± 0.09 ^{a}	0.11 ± 0.07 ^{a}	0.04 ± 0.05 ^{a}	0.12 ± 0.05 ^{a}	0.12 ± 0.04 ^{a}
Cu	3.26 ± 0.66 ^{ab}	3.46 ± 0.11 ^{ab}	3.69 ± 0.18 ^{ab}	2.47 ± 0.11 ^{a}	3.06 ± 0.38 ^{ab}	2.49 ± 0.18 ^{a}	4.51 ± 0.30 ^{b}
Zn	11.16 ± 2.04 ^{ab}	55.44 ± 5.41 ^{b}	30.70 ± 3.70 ^{ab}	9.41 ± 0.92 ^{a}	9.53 ± 2.43 ^{a}	9.31 ± 1.23 ^{a}	13.66 ± 0.65 ^{ab}
Se	0.00 ± 0.00 ^{a}	0.02 ± 0.02 ^{a}	0.02 ± 0.02 ^{a}	0.00 ^{a}	0.01 ± 0.03 ^{a}	0.02 ± 0.04 ^{a}	0.00 ^{a}
Br	0.45 ± 0.10 ^{abcd}	1.39 ± 0.40 ^{d}	0.14 ± 0.03 ^{a}	1.18 ± 0.071 ^{cd}	0.23 ± 0.09 ^{abc}	0.61 ± 0.38 ^{abcd}	0.97 ± 0.43 ^{bcd}
Rb	6.57 ± 1.46 ^{cd}	5.18 ± 0.29 ^{abcd}	7.16 ± 1.00 ^{d}	2.90 ± 0.15 ^{a}	5.83 ± 1.31 ^{abcd}	4.57 ± 0.98 ^{abcd}	6.04 ± 0.52 ^{bcd}
Sr	4.79 ± 1.05 ^{abc}	3.46 ± 0.35 ^{a}	4.33 ± 0.56 ^{ab}	6.39 ± 0.36 ^{c}	5.71 ± 1.02 ^{abc}	5.66 ± 0.96 ^{abc}	6.31 ± 0.35 ^{bc}
Y	2.84 ± 0.64 ^{ab}	2.88 ± 0.51 ^{ab}	4.22 ± 0.91 ^{ab}	3.89 ± 0.60 ^{ab}	4.29 ± 0.51 ^{b}	2.72 ± 0.45 ^{a}	3.37 ± 0.31 ^{ab}
I	0.34 ± 0.54 ^{a}	0.00 ^{a}	0.27 ± 0.59 ^{a}	0.56 ± 1.24 ^{a}	1.20 ± 2.21 ^{a}	0.20 ± 0.44 ^{a}	1.11 ± 0.63 ^{a}

Figure 3. Nutrient content of grape leaves of 7 different grape varieties (Maia et al. 2021)

Many sources have reported that the leaves of the plant are a source of phenolic compounds exhibiting antioxidant activity (Nishino and Yoshida 2002; Ito et al. 2002; Amakura et al. 2002; Siddhuraju and Becker 2003; Naczk et al. 2003; Amaral et al. 2005; Pari et al. 2007). This is a very important advantage, especially for a plant such as a vine, whose leaves are consumed by wrapping and stuffing. In many studies, it has been reported that the phytochemical content of the vine does not only consist of grapes and products made from grapes, but also that the leaves are rich in secondary metabolites. From the past to the present, the phytochemical compounds contained in the vine leaves; isoprenoids, organic acids, steroids, anthocyanins, sterols, leukoanthocyanins, rutin, quercitrin, isolacitroside, canpherol, luteolitins, free oils, free fatty acids, and heterocyclic compounds (Pari et al. 2007; Amarowicz et al. 2008; Jaradat et al. 2017; Babalık and Baydar 2019).

It is very important to take advantage of the beneficial phytochemicals of grape leaves and to use them in the food, pharmaceutical and cosmetic industries, as well as to benefit from the feature of an inexpensive dietary supplement. Moreover, considering that the importance of functional food components has increased in recent years, the use of vine leaves in many areas should be encouraged (Pari et al. 2007; Amarowicz et al. 2008; Babalık and Nilgün 2019).

Different researches that grapevine leaves are rich source of phenolic compounds (Bonilla et al. 2003; Doshi et al. 2006; Taware et al. 2010). Pintać et al. (2018), concluded in their study that many compounds such as phenolic acids (ellagic, chlorogenic, caffeic, p-coumaric and ferulic acids) are more in leaves than in fruit pulp. Other researchers mentioned the same trend in their studies comparing grape varieties (it is evident (Jara-Palacios Hernanz Escudero-Gilete and Heredia 2014; Luoet al. 2017).

In a study using more than one table and wine grape variety, it was determined that grape leaves also contain high phenolic substances, as in grains, and this content varies between 12.06 and 29.35 mg g^{-1}. In the research, it was determined that the leaves of wine grape varieties such as Narince, Boğazkere and Kalecik Karası have a high potential to be used in the food and pharmaceutical industry as a natural antioxidant source (Babalık and Baydar 2019).

In another study, it was reported that the total phenolic content of the extracts consisting of vine leaves was quite high (257 mg/g acetone extract and 232 mg/g methanolic extract). It has been observed that the leaf extracts of *Vitis vinifera* contain gallic, caffeic and p-coumaric acids. It was concluded that gallic acid is a dominant phenolic acid in the leaf. The analytical findings show that grapevine leaves which are waste materials from wine grape cultivation are a rich source of natural phenolic antioxidants. Therefore they might be used as functional ingredients for processing into nutraceuticals and health foods in the food (Amarowicz et al. 2008).

Doshi et al. (2006), were investigated the phenoliccomposition and antioxidant activity in grapevine parts and berries (*Vitis vinifera* L.) cv. Kishmish Chornyi (Sharad Seedless) during maturation. They have reported that the highest flavonoid values were determined in leaf samples at first harvest times (29.15 mg g^{-1} (+)-catechin equivalent).

It is important for both consumers and other usage areas to know individual bioactive phytochemicals of leaf varieties and to carry out studies to determine them. The phytochemicals contained in the vine leaf may vary according to the varieties, and it is also known that the same varieties show different bioactivities in different ecologies. In addition, studies on the bioactivity of grape leaves mostly investigate antioxidant potentials, while ascitotoxic, neuroprotective, anti-inflammatory, antimicrobial, etc. Information about the activities is still limited (Dani et al. 2010; Oliboni et al. 2011; Fernandes et al. 2013; Handoussa et al. 2013; Anđelković Radovanović Milenković Anđelković and Radovanović 2015; Abed Harb Khasib and Saad 2015; Krol,Amarowicz, and Weidner 2015; Aouey et al. 2016; Lima et al. 2016; Boraiet al. 2017; Lima Pereira Baraldi and Malheiro 2017).

6. Methods of Assessment of Vine Leaf

Vine cultivation contributes to the national economy by taking an important place in our agricultural products with different ways of evaluation, as well as being a source of livelihood for many producers engaged in agriculture (Yavaş and Fidan 1986). Grape, which is the fruit of the vine, is used as table, dried and wine grapes, as well as in different forms unique to our country (grape juice, papara, verjuice, pelverde, molasses, kome, meatballs, slice, bastik, rickshaw, raki, canned food, vinegar, pickles, tarhana, fruit pulp. etc.) are evaluated (Adır 2011). In addition to the many different ways of evaluation, Anatolian people have added a new one to their rich culinary culture by thinking of making use of the leaves of the vine with their superior intelligence and skill (Cangi and Yağcı 2017).

In the Anatolian viticulture culture, it is important to use the fresh leaves of the vine as wraps, canned or pickled. (Ağaoğlu et al. 1988). Vine leaves are used in our country to serve different purposes and flavors in the production of stuffing, stuffing, walnut batter and pita stuffing. Leaf, especially in some regions, disables the grape and emerges as the primary product in the vine (Cangi and Yağcı 2012). Vine leaf, which is an important food item on the table of Anatolian people, is low in calories and contains organic acids, sugar, phenolic substances, vitamins, dietary fiber and minerals in significant amounts (Ribereau and Reynold 1971; Cangi and Yağcı 2017).

In our country, the leaves of many grape varieties are used as canned or in pickled The leaves to be consumed for pickle are started to be taken from the ends in the spring when the shoots are still fresh. In the production of pickled vine leaves, thin-structured, hairless, fiber-free, thin-veined, less-sliced varieties that leave a sour taste on the palate are preferred. Our most important variety with these qualities is the Sultani seedless variety. In addition to grape production, this variety is a very important source of income with a high brine leaf harvest. Approximately 15000 tons of leaves are harvested annually in Manisa. Similarly, Narince, a wine and cider variety in the Tokat region, is mostly grown for its leaves (Göktürk ve ark. 1997; Anonim 2010; Cangi ve Yağcı 2012). In addition to these two varieties, the leaves of the Yapıncak grape variety are also used for pickling production in the Thrace region. Recently, pickled leaves have been used in vineyard areas in provinces such as Mersin and Denizli. The fact that some citizens in provinces such as Manisa and Tokat establish their own facilities by developing the marketing of brine leaves gives hope for the future of the sector (Göktürk et al. 1997; Çelik et al. 2010; Gülcü, 2010; Anonymous 2011c,d).

In 2016, Erbaa Narince Vineyard Leaf was registered as a geographically indicated product in the Erbaa district of Tokat. Geographical indication of a product not only provides added value to that product, but also supports local producers and rural tourism. Considering that local foods come to the fore as the main element in gastronomy tourism, which we have heard frequently recently, a vine leaf with geographical indication also contributes to the region in terms of tourism (Kaan and Gülçubuk 2008; Hazarhun and Tepeci 2018; Şimşek and Güleç 2020).



Figure 4. Photographs of Narince vine leaves harvest, sequence and cooked form

One of the most important factors to be considered in the production of pickled vine leaves is the production method that does not contain pesticides, copper and sulfur residues. In some studies by researchers, it has been reported that the active ingredients used against powdery mildew disease, which causes significant damage to the vine, are above the residual limit value (Ertürk 2009).

In the vineyard areas, powdery mildew, mildew diseases and vineyard scabies can be seen frequently. After the first (non-systemic) sprayings for downy mildew and powdery mildew, especially cultural treatments should be given priority. For this purpose, the bottom of the vineyard should be kept clean, infected leaves falling to the ground should be buried deeply by tillage, very good ventilation and sunbathing should be provided in the vineyard. As far as possible, pesticides should not be applied during the leaf harvest period in the vineyards, and when spraying is required, non-systemic drugs with a shorter duration of action should be preferred. As much as possible, foliar fertilizer should not be used during the leaf harvest period. Sulfur and copper-containing drugs with contact effect against powdery mildew, downy mildew and vineyard scabies should be used, and systemic drug use should be avoided. When collecting leaves, card leaves should not be collected from the panicle level and the lower part, and in gluttonous shoots. Herbicides should not be applied in the vineyards, especially during the leaf collection period. Because the chemicals that will come to the leaves with the wind will cause residues in the product and thus pose a risk in terms of food safety. In our country, the number of reports and studies on pesticide residues in pickled vine leaves is extremely limited (Cangi et al. 2005; Kılıç et al. 2007; Ertürk 2009; Cangi et al. 2013;Yanar et al. 2013; Gülcü and Torcuk. 2016).

In pickled leaf production, leaf harvesting should be carried out between the pre-bloom (May) and veraison period, by collecting the 4th, 5th and 6th leaves, which reach the size of 2 3-1 of the mature leaf from the tip on the shoots. Cardboard leaves that are damaged (torn, punctured, etc.) due to various reasons such as hail, wind, disease, pests should not be taken at harvest.Leaves should be collected in vineyards for a maximum of 4-6 periods, depending on ecology. If an economic return is expected from the grapes in the vineyard, it is appropriate to harvest the leaves as 2-3 cuts (Cangi and Yağcı 2012).

In case the pickled vine leaves are collected after the excess and verasion period, the grapes cannot fully mature. For this reason, leaf cutting should be done by considering the way the grapes are evaluated. Leaves should be broken early in the morning and processed into pickled in a short time. Processing the collected leaves in brine by separating the two dyes will increase the market value of the leaves. Damaged leaves, deformities and discolorations on the harvested leaves should be sorted. Again, it is beneficial to wash the leaves in order to purify the harvested leaves from foreign materials such as garbage, insects, pesticides and dust. This should be done in a way that does not scatter the decks after stacking. While producers in some regions (Manisa) sell the pickled leaves fresh, in some regions (Tokat) the producers market the harvested leaves after they are pickled (Gülcü et al. 2009; Gülcü and Torcuk 2016; Cangi and Yağcı 2017).

6.1. Vine Leaf Conservation With Different Methods

Pickled Vine Leaf; Preservation of vine leaves in pickled is a method that has existed in Anatolia for centuries. The basis of the pickled; It is based on fermentation and is based on the logic of biochemical alteration of carbohydrates, proteins and other organic substances in fresh vine leaves by microorganisms and especially lactic acid bacteria (Gülcü et al. 2009).

Canned Vine Leaf; It is the filling of fresh vine leaves in tin cans, glass jars or containers with suitable features after some pre-treatments (washing, sorting, boiling, cooling), sealing the containers in an airtight way (hermetic) and making them durable by applying heat treatment (Gülcü et al. 2009).

Unpickled Leaf; Packaging techniques are very important in recent studies to extend the shelf life of foods. There are studies on modified atmosphere packaging (MAP) techniques for the production of vine leaves without pickled (Başoğlu et al. 2004; Barazi and Erkmen 2008). This method provides some advantages such as being easy to apply, not containing salt, offering the product to the market in a short time, and not harming the environment with waste pickled (Gülcü et al. 2009).

Other Types of Conservation; Apart from the other preservation methods mentioned, in different regions under home conditions; There are different methods such as drying the leaf, dry salting, boiling in tulu water, frequently putting the leaf in jars or plastic containers and closing it, and storing it in the deep freezer (Gülcü et al. 2009).

6.2. Other Uses of Vine Leaf

Vine leaf, which has taken its place in the culinary culture of the Mediterranean basin such as Turkey and Greece, has been a tool used extensively by the people in the forms of treatment since ancient times. Since ancient times, the leaves of *V. vinifera* have been used in the treatment of jaundice, as a tonic, as a hemostatic agent, against diarrhea, hepatitis, stomachaches, antihemorrhoids, antiseptic, antianemia, diuretic, hepatoprotective, spasmolytic, hypoglycemic and vasorelaxant effects, as well as antibacterial, antifungal, anti-inflammatory, antinociceptive effects. It is used in medicine due to its various biological activities, including its antiviral and especially antioxidant properties (Kappor 1990; Hebash et al. 1991, Peterson 1991, Bombardelli and Morazzonni 1995; Baytop, 1999; Bruneton 1999; Felicio et al. 2001; Karaman and Kocabas 2001; Jaradat 2005; Gharib Naseri et al. 2006; Monagas et al 2006a; Monagas et al. 2006b, Şendoğdu et al. 2006; Orhan et al. 2007; Delíorman Orhan Et al. 2009; Koşar et al., 2007, Katalinic et al. 2009; Pari and Suresh 2009; Xia et al. 2010).The effectiveness of grape leaves used in folk medicine in human health has been supported by pharmacological studies. It has been proven that vine leaves have antidiabetic, antibacterial, antileishmanial and neuroprotective potential against peroxide damage, and that they can be used in the treatment of pain and inflammation with their anti-nociceptive and antipyretic activities (Kong et al. 2003; Şendogdu et al. 2006; Orhan et al. 2007; Dani et al. 2010; Mansour et al 2011; Aouey et al. 2016).

In a study examining the phenolic profiles and antioxidant capacities in the leaves of 20 white and colored grape varieties, it was revealed that grape leaves have rich phenolic composition and antioxidant properties. The bioactivities of the extract from the leaves indicate that they can constitute an interesting source of bioactive compounds with applicability in the food or pharmaceutical industries. It has been stated that the evaluation of this by-product in the future could have a. significant economic impact for areas devoid of agriculture (Fernandes et al. 2013).

Although Sultani Çekirdeksiz and Narince varieties come to the fore in the production of edible leaves when evaluated commercially, local varieties of each region with thin and soft structure, few slices and fine veined leaves are also widely used for this purpose (Cangi et al. 2012; Çelik 2013). Again Karaerik and Kabuğu Yufka (Sat et al. 2002), Hesapali (Ünver et al. 2007), Hamburg Misketi, Kober 5 BB and 41 B. It was determined that the leaves collected from the rootstocks (Göktürk et al. 1997) were suitable as edible. It has been stated that vine leaves with low edible quality are tested in the feeding of sheep as fresh, and these vine leaves, which are accepted as a by-product, are accepted by animals in sheep nutrition and do not have any toxic effects (Romeo et al. 2000).

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CHAPTER 6

ORGANIC GRAPE GROWING

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INTRODUCTION

Until the second world wars, farmers produced crops all naturally; they used natural products to eliminate pests, and they benefited from traditional agricultural practices that both preserved the soil and renewed it. However, in the years following the war, there was a big technological boom in the field of chemistry that also affected agriculture. A new kind of insecticide was created following Paul Miller's development of DDT in 1939 (Lockeretz, 2007). With the increasing population in the world, the demand for agricultural products is constantly increasing. Increases of up to 100% in products and food safety has been achieved, especially after the "Green Revolution", a revolution in agriculture that was initiated in the 1960s, which introduced a set of new agricultural techniques. At the time, this boom in production was considered one of the most significant technological developments of the century. However, these agricultural techniques were also starting to harm the ecosystem, causing it to deteriorate quickly. Soil, water, and the air were being polluted, and the products that were grown using various chemical pesticides and fertilizers were causing serious health problems to humans (Aksoy ve Altındişli, 1996; Ak, 2004; John and Babu, 2021). These production techniques were called "conventional"; the use of chemical fertilizers and pesticides in plant production, and the use of feed additives such as hormones and antibiotics in animal production have increased yields and reduced costs. And yet, some amounts of plant and animal-based products were left with chemical residues on them, and they harmed public health. In recent years significant increases in health problems, especially cancer, have been observed in people who consumed such products. (Aksoy ve Altındişli, 1999; Ak, 2004; John and Babu, 2021). With the increase in social awareness of the negative effects of agricultural pollution, sustainable agriculture systems such as organic agriculture that ensures the production of healthier products, are brought to the fore (Öztürk, 2004; Taşlı et al., 2003; Yürüdür et al., 2010). For a while studies on organic agriculture were carried out independently on a national basis in various countries, the 1970's they were gathered under the name of "International Federation of Organic Agriculture Movements" (IFOAM). The main purpose of this organization which is located in Germany is to set standards for organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture and to create a flow of information on organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture in the world. In EU countries, the first organic agriculture regulation, which revolved around plant production, was published on 24.06.1991. Organic agriculture regulation for livestock was published in 1999 and took effect in 2000. Today, as a result of IFOAM's hard work, organic agriculture has become a production model that is applied within the framework of laws in many countries around the World (Aksoy and Altındişli, 1996).

1. ORGANIC AGRICULTURE

Organic agriculture Organic agriculture Organic agriculture Organic agriculture Organic agriculture Organic agriculture is a production system that promotes and improves agro-ecosystem health, including biodiversity, biological cycles, and biological activity of the soil. The additives used in this system consist of biological origin materials, and mechanical and cultural methods defined in the legislation on organic agriculture. All production and sales stages of organic products are followed by an accredited and certified control and certification body. For any product to be considered an organic product, all stages from the procurement of the product to the final consumer must be inspected and documented. Inspection and certification, which is one of the main features of organic agriculture, ensure that the products grown are produced, processed, packaged, and distributed according to the organic standards specified in the legislation (İlter and Altındişli, 1996).

The purpose of organic agriculture is to eliminate environmental issues that were caused by years of unconscious soil cultivation practices and the excessive use of synthetic products and to ensure that uncontaminated areas are protected. It also aims to ensure that consumers can reach healthy products that do not endanger the environment and human health.

Organic agriculture is not “fertilizer and pesticide-free agriculture” or “natural agriculture”. Organic agriculture is one of the sustainable agriculture systems that has its own principles and practices from production to marketing. (İlter and Altındişli, 1996; Demiryürek, 2000; Dabbert et al., 2004).

1.1. Definition and Principles of Organic Agriculture

One of the most prominent definitions of organic agriculture has been created by the International Federation of Organic Agriculture Movements (IFOAM). IFOAM defines organic agriculture as an agricultural system that protects the soil, the ecosystem, and human health. This agricultural system prefers an agricultural production process that has adapted to ecology, biodiversity, and local conditions, rather than a production process that uses methods that have negative effects on the soil, ecosystem, and public health. Organic agriculture is a form of agriculture that combines tradition, innovation, and science therefore everyone can benefit from the environment that they all share. It aims to establish fair relations between all stakeholders and ensure a good quality of life for all (Luttikholt, 2007; Anonymous, 2022). According to IFOAM, organic agriculture is based on four basic principles: health, ecology, fairness, and care (Davies, 2006).

1.1.1. Health

The aim of organic agriculture is to preserve and improve the vital cycle of soil and plants and animals as an indivisible whole. According to this rule, the health activities of all communities cannot be separated from the health of the ecosystem. It offers a healthier life by growing fertile products on fertile soils.

1.1.2. Ecology

The aim of the term organic is to create a balanced ecological system and to be sustainable by working through this system. With this rule, it connects the system in organic agriculture organic agriculture to the existing ecological system. Thus, it directs production to ecological process and recycling. Nutritional balance and welfare level are provided by a certain production ecology system. This ecology system is the farm ecosystem for the soil and animals that are alive in crop production. It also provides an aquatic environment for all creatures living in the sea.

1.1.3. Fairness

The goal in organic agriculture is to provide equal relations over all common living conditions. The concept called justice means that there should be respect, equality and fairness for all living things in the world.

1.1.4. Care

In organic agriculture, it is the responsible and careful management of all living things, together with the environment, for their health and well-being. It is a dynamic and lively system that can respond to the conditions in the most correct way in the face of internal and external demands. In organic agriculture, practitioners can increase production activities and productivity in the most effective way, however this should not be in a way that endanger health and welfare. Technological innovations in organic agriculture should be reviewed and evaluated.

1.2. Organic Agriculture in the World and Türkiye

The first ideas about organic agriculture can be rooted back in the last quarter of the 19th century and the first half of the 20th century. The term “organic” was first used by W. Northbourn in 1940 in his work named W. Northbourn to describe the ecological farming system (Paul, 2014). However, the beginning of the organic agriculture movement that is market-oriented dates back to the 1970s, when the concept of "sustainability" started a debate all over the world. The International Federation of Organic Agriculture Movement-IFOAM, which was established in 1972 in Germany, was created to gather all of the organic agriculture movements in the world under one roof, to healthily direct their development, to prepare standards and regulations, and to announce developments to its members and relevant institutions. Although the trade of organic products around the world developed in the 1980s, there was an increase in demand for organic products in the 1990s, with the concerns raised by issues such as "mad cow, dioxin, GMO (transgenic products)" and the issue of sustainability all over the world (Yürüdür et al., 2010). The studies that were conducted until the 1970s started to become widespread with the establishment of IFOAM (International Federation of Organic Agriculture Movement) in

1972. Organic agricultural areas constitute 2-3% of all agriculture in European countries.

Organic agriculture in Türkiye started in 1984-1985 when European consumers demanded organic products. At that time, there was no regulation in the name of national legislation on the organic production of raisins and figs, which are traditional export products (Aksoy and Altındişli, 1999; Aksoy, 2001).

IFOAM rules were valid in organic farming. After the establishment of the Ecological Agriculture Organization Association (ETO) in 1992, the Regulation on the Production of Crop and Animal Agricultural Products with Ecological Methods, which is the first national regulation similar to the 2092/91-EU Regulation, entered into force in 1994 (Anonymous, 1994). Major additions were made to this regulation later on, and in 2002, with the "Regulation on the Principles and Implementation of Organic Agriculture" (Anonymous, 2002) numbered 24812, including organic livestock and aquaculture, and another regulation on amending the said regulation (Anonymous, 2003). "Organic Agriculture Law" (Anonymous, 2004), which is a framework law, was adopted and published on 3 December 2004. This framework law, together with the MARA, authorized the relevant institutions to regulate organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture regulations. This law also includes penalties not specified in previous regulations. The "Regulation on the Principles and Practice of Organic Agriculture" (Anonymous, 2005), which is the regulation of the law, was published in the Resmi Gazette No. 25841 on 10 June 2005 and put into practice. Some changes were made later in the said regulation (Anonymous, 2006; 2008; 2009). Finally, the "Regulation on the Principles and Practice of Organic Agriculture" (Anonymous, 2010), published in the Resmi Gazette No. 27676 on 18 August 2010, has been prepared by taking into account the latest changes in the EU legislation.

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According to Willer et al. (2022), 1.6 percent of the world's agricultural land was over 74.9 million hectares in 2020. The highest organic the total agricultural land is in Oceania (47.9%) and the lowest rate is 2.8% in Africa (Table 1).

Table 1. World: Organic agricultural land (including in-conversion areas) and regions' shares of the global organic agricultural land 2020 (Willer et al., 2022)

Region	Organic agricultural land [hectares]	Percent of the global organic agricultural land
Africa	2'086'859	2.8
Northern America	3'744'163	5.0
Asia	6'146'235	8.2
Latin America	9'949'461	13.3
Europe	17'098'134	22.8
Oceania	35'908'876	47.9
World	74'926'006	100

Australia has the biggest organic agricultural land one of country in the world. Australia is followed by Uruguay (2.74 Mha), India (2.66 Mha), France (2.55

Mha), Spain (2.44 Mha), China (2.33 Mha), USA(2.1 Mha), Italy (2.1 Mha), Germany(1.7 Mh), respectively in ten place (Figure 1).

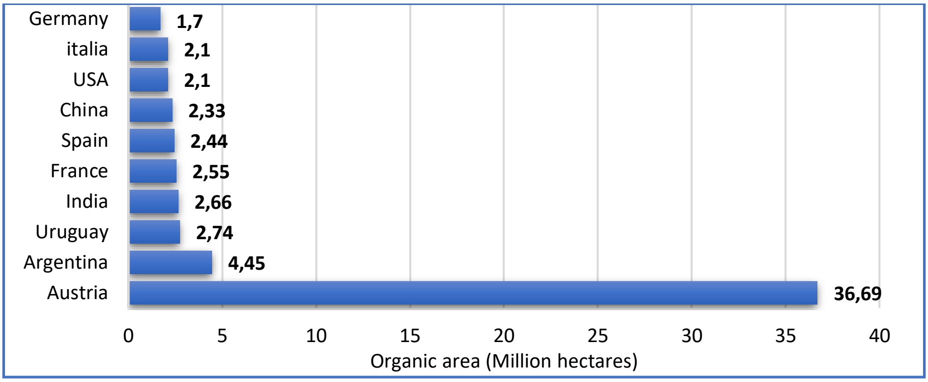

Figure 1. The ten countries with the largest areas of organic agricultural land 2020 (Willer et al., 2022)

Among organic permanent plants, the highest permanence products are olive, followed by coffee and nuts. Grape, on the other hand, ranks fourth among the permanent plants with a 10% share (Table 2).

Table 2. Distribution of organic permanent cropland and percent by crop group 2020 (Willer et al., 2022)

Organic permanent crop	Organic permanent crops land [hectares]	Percent of the global organic permanent crops land
Olives	894'989	17
Nuts	749'055	14
Coffee	744'942	14
Grapes	498'445	10
Cocoa	384'507	7
Coconut	294'234	6
Fruit, tropical and subtropical	292'535	6
Fruit of temperate climate zones	256'317	5
Tea/mate	178'928	3
Citrus fruit	140'837	3
Medicinal and aromatic plants, permanent	90'954	2
Berries	66'050	1
Fruit	62'083	1
World*	**5'238'362**	**100**

2. ORGANIC VITICULTURE

Grapevines are some of the oldest fruit plants in the world. The grape is also one of the most important horticultural crops in the world with 7'157'958 hectares and 79'125'982 tons of grape production in 2018 (Anonymous, 2022 a). Türkiye is a major producer of grapes in the world with approximately 4'670'930 hectares of vineyard area and 4.01 million tons of grape production (5^{th} in area; 6th in production). Grape production mainly consists of 52.8% table grapes, 36.4% raisins and 10.8% must-wine varieties (Anonymous, 2022 b).

The term organic viticulture is a system created for product, livestock and fish farming, which aims to use all natural agricultural techniques and to protect environmental factors. Producers working in the organic field rely on natural farming and modern scientific methods to maximize productivity in the ecosystem, increase product quality and protect the environment. Advocates on this platform argue that this method is more reliable, sustainable and less harmful (Morgera et al., 2012).

2.1. Distribution and Evolution of the World's Organic Vineyards

The European continent has an area of 431'225 ha and 87% organic vineyards. North America with 27'444 ha (5%) is second followed by Asia with 15'141 ha (3%), Latin America with 14'682 ha (3%), Oceania whit 5'783 ha (1%), and Africa with 4'291 ha (1%) (Figure 2). North America with 27'444 ha (%5) is second followed by Asia with 15'141 ha (3%), Latin America with 14'682 ha (3%), Oceania with 5'783 ha (1%), and Africa with 4'291 ha (1%) (Figure 2).

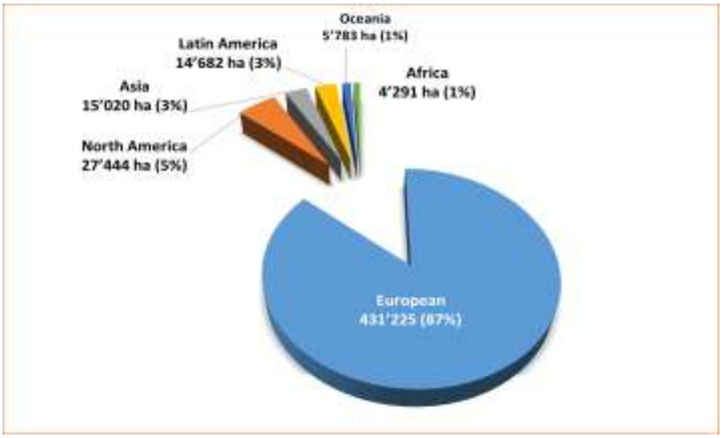

Figure 2. Organic grape area by continent in hectares (Willer et al., 2022)

Organic grape production is carried out in a limited number of countries globally. By 2020, organic viticulture was carried out in 63 countries globally, and the certified vineyard area was almost 506’400 hectares or 7.3% of the global grape area was under organic management. The largest organic vineyard areas are in France. France has 136’431 ha of organic vineyards. France is followed by Spain, Italy, the USA, china, Germany and Türkiye (FiBL, 2021) (Figure 3).

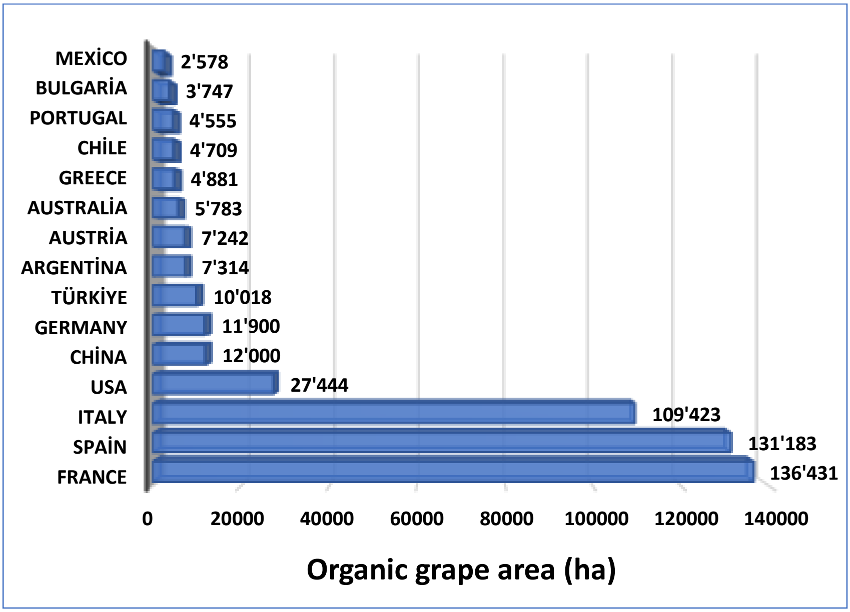

Figure 3. The countries with the largest organic grape area (ha) (Willer et al., 2022)

In terms of the distribution of organic vineyards, 15 leaders in organic production (Figure 4). The highest percent organic total agricultural land is in Poland (26.1%). Poland is followed by Belgium, France, Austria, Italy, Spain, Switzerland, Bulgaria, Germany, Lebanon, Netherlands and Mexico. Türkiye’s organic vineyard surface area has been growing at an average annual rate of 2.5% (Willer et al.2022). Since 2005, organic raisin production in Turkey is carried out intensively in the provinces of Manisa and İzmir, and it is exported especially to European countries. (Ateş, et al., 2016). According to the records of Aegean Exporters' Associations, while the income from the export of organic seedless raisins was approximately $3 million in 1997, it increased by 79% in

2004 and reached approximately $5.2 million (Kenanoglu Bektaş and Miran, 2006). It increased by 85% in 2020 and reached approximately $6.8 million (Anonymous, 2020)

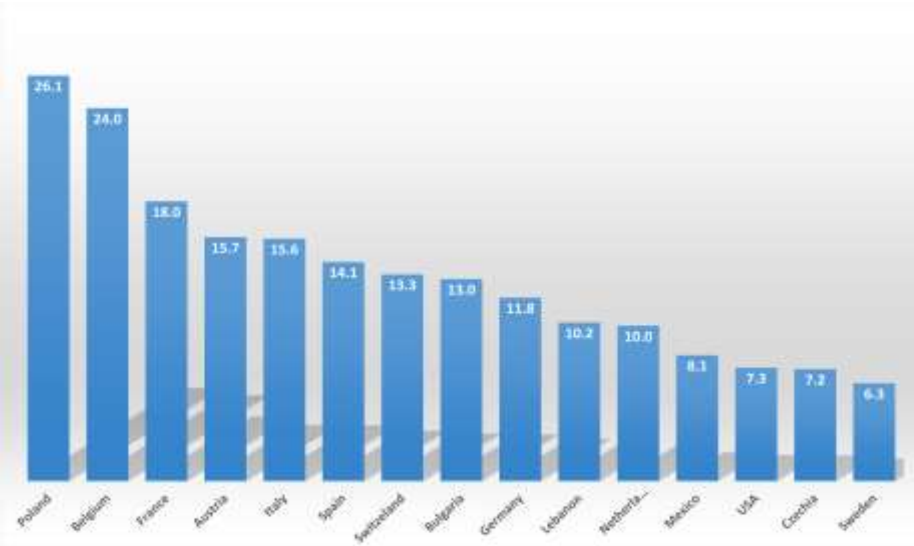

Figure 4. The countries with the highest organic grape area share (%) (Willer et al., 2022)

2.2. Technical and Cultural Practices in Organic Grape Growing

In organic viticulture, cultural management practices are the most basic method. Especially in the first establishment stage of the vineyard, varieties resistant to fungal diseases should be used. Selection of varieties resistant or susceptible to fungal diseases is of vital importance in organic grape production. Because there are a limited number of licensed fungicides in organic agriculture in the fight against diseases in organic viticulture, the repeated use of copper and sulfur to combat primary fungal diseases causes negative effects on the grape potential and causes product loss (Pedneault and Provost, 2016). It is important to effective sanitation such as removing harvest residues, pruning and similar wastes with to regulate irrigation and fertilization programs in organic vineyards

When organic viticulture is planned, organic grape production is started in three ways, according to the initial stage.

1. Continuing organic production by planting organic grapevine saplings,

2. Continuing organic production after the 3-year transition period, which started with the planting of non- organic grapevine saplings,
3. Continuing organic production in existing vineyards after a 3-year transition period

2.2.1. Planting grapevine saplings

It is recommended to plant grapevine sapling in spring or autumn. It is appropriate to plant in spring in places with a harsh climate and in autumn in places with a temperate climate. The fact that the soil is tempered during planting and the soil temperature raises above 10 ^{0}C increases the grapevine sapling retention rate.

According to the results of the soil analysis, basic fertilization should be done in the pits. This fertilization, which consists of microbial fertilizers, meets the need for these nutrients for 2-3 years. If soil analysis has not been performed, microbial fertilizer and 1 shovel of burnt farm manure should be given. Fine soil should be thrown on the fertilizers therefore it does not come into contact with the roots of the grapevine sapling.

Tubed seedlings, on the other hand, should be taken out of their containers before the mortar is dispersed, and the pit should be completely filled with soil, lightly compacted with water, and should not be trampled with feet. Cartonage seedlings are planted with their containers (Figure 5 a).

Open-rooted grapevine sapling should be placed in the pit so that the grafting site is above the soil level and the soil should be filled up to half of the pit. Soil should not be compressed too much, compression should be provided by giving life water. Too much compression prevents life water from reaching the root zone and prevents the development of young roots. After the life water is given, the rest of the soil should be filled and the soil should be compacted, and in cold regions, the top of the sapling should be covered by making a cupola (Figure 5 b).

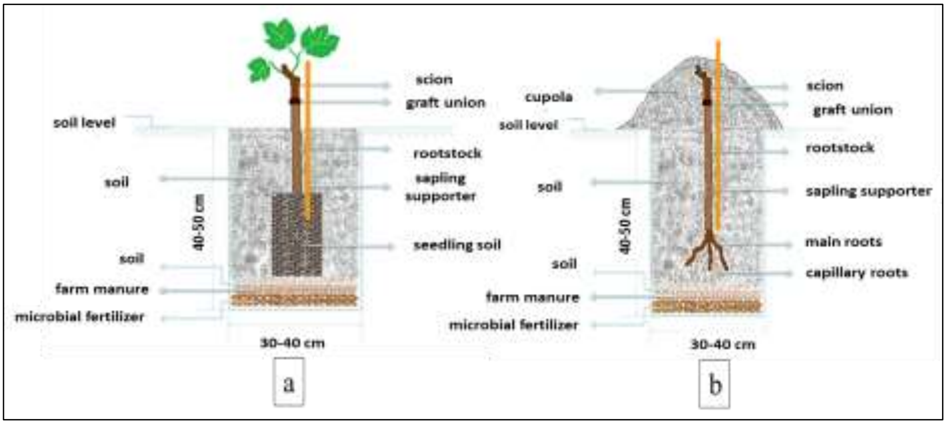

Figure 5. Planting tuberous grapevine saplings (a) and planting open-rooted grapevine saplings (b)

2.2.2. Pruning and training

There is no difference between conventional production and organic agriculture in terms of pruning and training system. Since the product is taken from one-year-old branches in the vine, it should be encouraged to form shoots. For this, regular pruning should be performed every year. Spur pruning should be performed in the bottom eye productive varieties. Cane pruning should be performed in varieties with low yield or inefficient bottom bud. In other words, the pruning required by the variety should be applied. It should be noted that overload should be avoided. Summer pruning is inevitable due to its important effect in reducing the severity of powdery mildew disease.

In terms of training systems, it is beneficial for organic viticulture in the creation of Double T, Y and V cultivation forms in terms of good ventilation of the vine and preventing congestion (Figure 6).

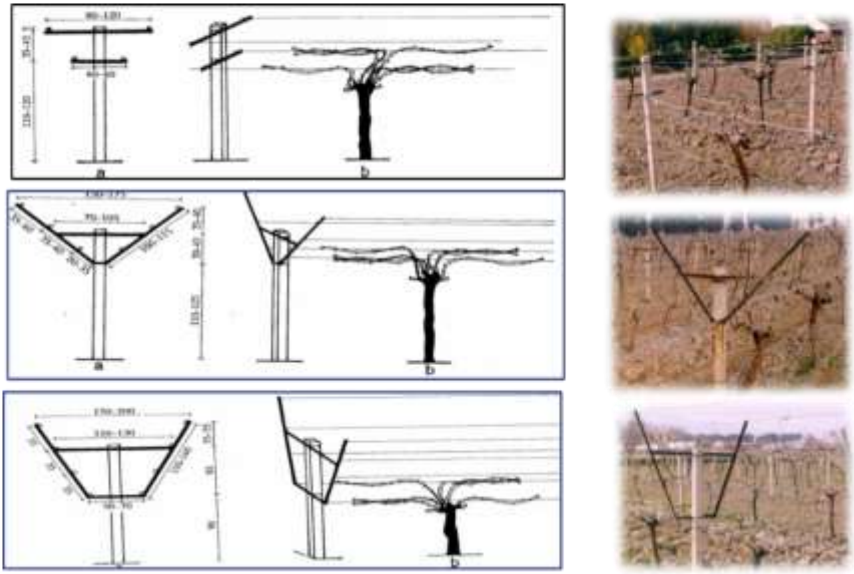

Figure 6. Training systems used in organic viticulture

2.2.3. Tillage

Expectations from tillage in organic viticulture can be listed as follows;

- Tillage should not cause water and wind erosion in the soil,
- It should prepare the best environment suitable for the living organisms in the soil,
- It should prevent the loss of macro and micro nutrients in the soil,
- It should prevent compaction in the soil, prepare the appropriate environment for the root growth of the plant,
- It should ensure that the material to be given to the soil is given under the soil and especially to the root zone.
- Tools such as plows that work by overturning the soil should be used as little as possible; instead, tools such as chisels and cultivators that work the soil without overturning should be preferred.

In minimum or reduced tillage, fewer operations are performed compared to conventional production. Although plows are used in some applications in this tillage system, soil tillage machines operating by the tractor PTO (power take-off) shaft should be preferred. Soil compaction is minimized as the field traffic is reduced as a result of the decrease in the number of operations. In addition, this method provides savings in terms of energy and human labor.

Along with applications that improve the soil structure and increase the amount of humus, tillage methods that protect the soil and do not disturb the soil dynamics should be applied. In organic viticulture, the reduced tillage method, which is expressed as more superficial tillage, is used (Sharley et al.,2008). Thus, it increases the soil organic matter and promotes the activity of microorganisms and ensures better infiltration of water into the soil.

2.2.4. Irrigation

There is a general belief in our country that vineyards are not watered. However, irrigation should be performed as needed for a good quality yield. Irrigation is a cultural practice in which yield increase is achieved by 30-40%. In our country, irrigation is carried out especially in seedless vineyards in the Lakes region and the Aegean region. Irrigation should be performed in arid and semi-arid vineyard areas. The vine needs 1 liter of water from its leaves to produce 1 g of dry matter. Under normal conditions, 20-60 ml of water per hour evaporates from the leaves from each cm^2 (20-60 ml/cm^2 /h). It also requires 450 mm/m^2 of water in vegetation. Since 250-300 mm of this evaporates during this period, approximately 700-750 mm/m^2 precipitation should be received during the vegetation period for a normal development and fruit yield in the vine. If this amount cannot be obtained from the soil, it must be met with irrigation, especially in arid and semi-arid climate conditions (İnal, 1983).

The methods recommended to be used due to the nature of organic agriculture are pressurized irrigation methods. At the beginning of these are drip and mini sprinkler irrigation methods.

The principles to be considered in irrigation practices in organic plant production are determined in Article 12 of the Organic Agriculture Regulation of the Ministry of Agriculture and Forestry. According to this article;

- Industrial and city wastewater and water obtained from the drainage system cannot be used in organic agriculture.
- Irrigation water should not cause environmental pollution. Irrigation water that complies with 7739 standards should be preferred.
- Irrigation should not cause deterioration and erosion in the soil structure.

- In addition to the ones stipulated in the regulation, there are benefits to consider the following items.
- Release irrigation techniques should not be used in agricultural areas where plant production is carried out with Organic Agriculture method.
- A water plan should be prepared according to the amount of water that the plant will consume during the vegetation period.
- Irrigation water should be given directly to the root zone in moderation.
- A drainage system should be established in soils with poor drainage, low permeability, heavy clay and loamy soils. Irrigation should not be done with the drainage water obtained from the drainage system.
- Drip irrigation method should be applied in viticulture irrigation.

2.2.5. Nutrient management

Organic fertilizers to be used in organic production vineyards should also be analyzed beforehand and the plant nutrients they contain should be determined. The purpose of organic viticulture is to maintain the vitality of the soil and to preserve its productivity. For this purpose, in addition to many applications such as rotation, cover crop, mulch application, soil cultivation suitable for organic viticulture, the use of some fertilizers and soil conditioners is allowed in cases where nutrients are not sufficient. The aim here is to maintain soil fertility and to provide adequate nutrition for plants. Organic agriculture law and relevant regulations should be taken into account in the applications of organic vineyards, it should be checked whether the fertilizers to be used have a certificate or not, and certified products should be preferred. In the areas where organic plant production will be made, if sufficient soil fertility and biological activity cannot be achieved despite the measures in paragraph 3 of the Organic Agriculture Regulation, Fertilizers, Soil Improvers and Nutrients to be Used in Organic Agriculture, which are included in the Annex-1 of the Organic Agriculture Regulation, are used (Anonymous, 2010).

Considering the Principles of Organic Agriculture, as fertilizer and soil conditioner;

2.2.5.1. Farm fertilizer

The content and amount of farm fertilizers used for organic agriculture vary according to the age and breed of animals and the litter materials used. The amount of farm manure to be given varies according to the amount of organic matter in climate and soil conditions. Farm manure should be mixed with the soil after the vineyards have shed their leaves in the autumn period. In order to prevent the negative effects of nitrogen such as pollution in water, the amount of total farm fertilizer to be used in the organic plant production phase should not exceed 170kg/N/ha/year.

2.2.5.2. Green fertilizers

Green manure is used for plants that have completed their development, and green manure plants are used for this purpose. Of the green manure plants, alfalfa, vetch and broad bean fodder crops are planted the most. These plants increase the nitrogen content of the soil by absorbing the nitrogen in the air. These plants, which are planted between the rows in winter and autumn, are mixed with the soil during the flowering period.

2.2.5.3. Composts

Composts are in the quality of humus and are obtained from the mineralization of vegetable or animal organic wastes as a result of microbial decomposition after various processes. Since they have undergone aerobic decomposition, they are organic fertilizers that are free from pathogens and look like heather soil with the smell of heather soil.

2.2.5.4. Liquid organic fertilizers

They are obtained from animal manure and plant material. Depending on the type of substances used, they are left in the water for a few days or weeks, and the organic materials are filtered and then thinned with clean water. These fertilizers are given to the plants as soil or foliar application (such as seaweed).

2.2.5.5. Natural origin mineral fertilizers

Mineral fertilizers used in organic agriculture are obtained as a result of special grinding of natural rocks. The main sources of mineral fertilizers used in organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture are rock dust, plant ashes, lime and rock phosphates, and these mineral fertilizers have rich content especially in terms of calcium and potassium. Mineral fertilizers obtained from plant ashes are rich in calcium and potassium.

2.2.5.6. Microbiological fertilizers

The commercial forms of live microorganisms, which play a role in taking some essential plant nutrients from the soil, which are necessary for plants, for use in agricultural production are called microbiological fertilizers. Non-genetically modified microorganism preparations are used in organic agriculture.

2.2.5.6.1. Plant growth promoting root bacteria (PGPR)

- Benefits of plant growth promoting root bacteria are listed below
- Conversion of some mineral substances in the soil into the form of plant nutrients that plants can benefit from,
- Easy decomposition of soil organic matter,
- Connecting the nitrogen gas in the composition of the atmosphere to the soil,
- Taking the phosphorus needed by the plants,
- They also ensure the supply of water and nutrients,

Bacteria that increase phosphorus solubility in the soil: *Rhizobium, Enterobacter, Serratia, Citrobacter, Proteus, Klebsiella, Pseudomonas* etc. Nitrogen-fixing bacteria: *Azotobacter, Rhizobium, Azospirillum, Azotobacter, Bacillus, Beijerinckia, Pseudomonas* etc.

2.2.5.6.2. Mycorrhiza

Mycorrhiza is a type of fungus that lives in symbiosis with the roots of plants. It is found in the roots of almost all plants, and its task is to improve the structure of the soil, to ensure the uptake of water and nutrients by the plants, and to supply especially useful phosphorus to the plant.

2.2.5.6.3. Trichoderma (Trichoderma)

Trichoderma is a kind of soil fungus with fungicidal and growth promoting effect.

- Stress tolerance, capillary root development,
- Dissolving and taking the nutrients bound in the soil,

- Plants have phytoalexin compounds that naturally fight in their immune systems. It usually provides disease control.
- Resistant to chemicals and heavy metals and antibiotics produced by other bacteria

2.2.5.7. Others

It is obtained by physical processes including dehydration, freezing and grinding, extraction with water or aqueous acid and/or alkaline solutions and fermentation methods.

Leonardite and elemental sulfur (S) can be used to reduce the soil reaction (pH), clinoptilolite, slag, perlite, vermiculite can be used to regulate the soil structure.

Humic acid: These are the inputs that can be used both to improve the physical properties of the soil and to facilitate the uptake of plant nutrients.

Table 3 Plant nutrition methods in organic viticulture

Table 3. Plant nutrition methods in organic viticulture (Ateş, 2007)

Deficiency in Soil	**Symptom of the plant**	**Solution**
Soil acidity	If the soil is too acid, P, Ca, Mg availability decreases, Fe, Cu, Mn toxicity can be seen.	-Aragonite -Dolomitic limestone
Boron	Slow growth, gumming, fungal in fruit	-Boron (% 10) -Boron (%14,3)
Calcium	Reddish-brown leaves, leaf margins dry	-Gypsum (for alkaline soils) -Limestone -Rock phosphate -Dolomitic limestone (for acid soils)
Copper	Leaves curl and dry like a cup	-Copper sulfate -Reducing nitrogen fertilization
Iron	intervascular chlorosis	-Iron chelate (10%) -Iron sulfate -Reducing phosphorus fertilization
Magnesium	Chlorosis of lower leaves, turning red	-Epsom salt (for alkaline soils) -Dolomitic limestone (for acid soils)
Manganese	Chlorosis that starts first in young leaves	-Manganese sulfate
Molybdenum	Yellowed and wilted leaves	-Dolomitic limestone
Nitrogen	Light green or yellowish green leaves	-Blood meal -Cotton seed flour -Corn starch flour (10-0-0)

		-Feather meal -Leather flour
Phosphorus	Leaves are dark green then red purple	-Raising the pH -Bone meal -Rock phosphate -Bat dung
Potassium	Drying on the leaf margins	-Seaweed -Granite flour -Wood ash -Potassium sulfate (0-0-52)
Zinc	small yellow leaves	-Zinc sulfate -Zinc chelate
Sulfur	yellowish leaves	-Rock phosphate -Gypsum -Elemental sulfur

2.2.6. Disease and pest management

There are differences between conventional production and organic agriculture in terms of agricultural control. It prohibits the use of synthetic chemicals that reduce biological diversity in nature and create residue problems. In the control of pests, importance is given to biological control, protection of beneficial fauna and biotechnological methods.

Knowing diseases well is the first condition of success in organic viticulture. Cultural measures should be heeded.

2.2.6.1. Disease management

2.2.6.1.1. Powdery mildew (*Erysiphe=Uncinula necator Schewein*.) management

Cultural management

The most common disease in the Aegean region is powdery mildew (Uncinula necator).

Cultural management;

- The cluster should provide ventilation by taking leaves from its surroundings.
- With a good tillage in winter, the leaves on the ground should be mixed with the soil.
- Dish pruning residues should be removed from the vineyard. Because the agent can overwinter here in another spore form.

Spraying

Wettable (WP) Sulfur:
73%……….. 500 g/ 100 l water
80%…………400 g/ 100 l water

Drinking Soda [$NaHCO_3$ (2000 gr/ 100 l water)] can be used as an alternative to sulfur (WP) in the fight against powdery mildew disease (U. necator).

Powdery mildew disease is most commonly controlled by the application of sulfur with a wettable powder, powder or soluble formulation (Crisp et al. 2006).

2.2.6.1.2. Mildew (*Plasmapora Viticola*) management

Cultural management;

- Diseased shoots should be cut from the bottom and removed from the vineyard. The bottom of the vines should be kept clean.
- Because of harboring the spores of the fungus, the leaves under the vines of the previous year should be collected and burned at the beginning of spring. Diseased leaves and weeds falling to the ground
- The prediction and early warning system is based on the fight against downy mildew.

Biological control;

- *Bacillus subtilis* and *Pseudomonas fluorescens* preparations are recommended against gray mold disease.

The medicine called burgundy slurry [Bordeaux slurry: 1% (1000 g copper sulphate + 500 g slaked lime)], which is prepared by mixing bluestone and half as much quicklime, is the most effective way to fight against mildew. It can be used in ready-made copper preparations.

The most commonly used preparation in the world today in the fight against mildew is copper. Copper, which has been used in viticulture at a rate of 80 kg/ha per year for more than 150 years, has led to copper accumulation in the upper soils of many vineyards (Rusjan et al., 2007). For this reason, restrictions have been imposed on the total use of copper preparations in the organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture regulations of the countries. Today, copper use in organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture organic agriculture is limited to 6 kg ha per year in most European countries, including Italy, France and Spain (Dagostin et al., 2011).

2.2.6.1.3. Vineyard dead arm disease (*Phomopsis viticola*) management

- It is very important to remove sick shoots and remove them from the vineyard during winter pruning.
- Winter Spraying: Claret Red Slurry: 2 – 4%
- In the spring, when the shoots are still 2-3 cm and 2 weeks after that, spraying may be required,
- For this reason, copper-based drugs can be used compulsorily.

2.2.6.1.4. Esca (*Phaeomoniella chlamydospora, Phaeoacremonium spp., Fomitiporia* spp.) management

- If the sick vines are unproductive, they should be removed together with all root parts.
- Quicklime should be poured into the pit and vines should not be planted there for several years.
- Healthy vines should not be damaged during tillage; large pruning wounds should be closed with a suitable paste. During pruning, separate scissors and saws should be used for sick vines or pruning tools should be disinfected after pruning each vine. There is no drug struggle (Çetinkaya and Ateş, 2016).

2.2.6.2. Pest management

2.2.6.2.1. Moth (*Lobesia botrana* (Lepidoptera: *Tortricidae*) management

The main pest in the Aegean region is the cluster moth (Lobesia botrana).

Cultural management;

It is recommended to suspend the vine, to make spacing and cutting in a way to keep the inner part of the vine airy, not to leave weeds, to give importance to winter cleaning, and not to water unless necessary.

Biotechnological management;

Mating inhibition (C) or staggering technique is used. There are two types of pheromone emitters in our country, similar to copper wire (Isonet-L) and capsule-shaped (Rak 2 Pro). Spreader hanging is done once during the season.

Biological control;

- -Spinosad
- -*Bacillus thuringiensis* preparations are recommended in organic viticulture to against cluster moth larvae

2.2.6.2.2. Ligament trips (*Anaphothrips vitis*; *Haplothrips globiceps; Drepanothrips reuteri*) management

Cultural management;

In or around the vineyard, shelters that may overwinter in the k phase should be destroyed,

Other host plants should not be kept in or around the vineyard.

Biological control:

- Chrysanthemum cinerariaefolium (Pyrethrin),
- Spinosad preparations are recommended in organic viticulture.

2.2.6.2.3. Vineyard leafhoppers management

- *Chrysanthemum cinerariaefolium* (Pyrethrin) [Natural Pyrethrin],
- *Azadirachta indica* (Azadirachtin),
- *Ryania speciosa* preparations are recommended in organic viticulture. However, it is not licensed in Türkiye.

2.2.6.2.4. Two-spotted red spider (*Tetranychus urticae*) management

Cultural management

- The vineyard should not be left grassy.
- If possible, measures should be taken against road dust.

Biological Control

- It can be used in biological control by protecting the beneficial ones, increasing their effectiveness, infecting the places where they are not found, and by producing and releasing them.

Spraying

- -Summer Oils

- -*Azadirachta indica* (*Azadirachtin*) preparations are recommended in organic viticulture. However, it is not licensed in Türkiye.

2.2.6.2.5. Mealy lice (*Planococcus citri*) management

Cultural management

- Bonding should not be established on the bottom land that holds a lot of water and in shady places.
- If there is a necessity, the vines should be planted sparsely and a high training system should be used.
- -The leaves of dished vines should be diluted, and the clusters should be ventilated.

Mechanical management

During winter pruning, if the bark under which the pest has overwintered is peeled and burned, the density will decrease.

Spraying

Organic preparations of summer oils are recommended in organic viticulture.

2.2.6.2.6. Brown shell lice management

Cultural management

The annual sticks that the pest has detected itself should be removed from the vineyard when pruned in the spring.

Mechanical management

Crushing on the branches where it is located also reduces the pest density. Pesticides used against mealybug and brown crustacean in Ecological Agriculture

Spraying

Organic preparations of summer oils are recommended in organic viticulture

2.2.7. Weed management

In organic viticulture, weed control, which is one of the most important parameters to be considered in order to get high quality and high yields.

2.2.7.1. Cultural management

To prevent the transmission of weeds in particular

- Weeds in organic vineyards are more farm manure
- It is transmitted by tillage tools and agricultural tools and equipment (1000 grain weight of monsoon seed = 3.82 mg)
- One of the ways weeds spread to vineyards is irrigation water. However, due to the necessity of using drip irrigation method in organic viticulture, this risk has gradually decreased.

Another cultural process that can be applied to control weeds in organic vineyard areas is the cultivation of cover crops. In our country, vetch species (*Vicia* spp.) can be used successfully as a cover plant in vineyard areas in winter and buckwheat (*Fagopyrum esculentum Moench*) in summer.

2.2.7.2. Mechanical combat

- Tillage
- Continuous mowing practice
- In brushing: It is a fairly new method for our country. It is especially effective in the control of newly emerged weeds. While the soil structure is not damaged by the brushing method, which is effective in the first few cm of the soil, the cream layer is broken.

2.2.7.3. Physical management

In the control of weeds with physical control, methods based on the principle of exposing or preventing physical parameters such as heat, light and electromagnetic waves are used. The physical methods that can be applied for weed control in vineyard areas are mainly mulching and flaming.

Mulch application: This method is to destroy the weeds between the rows by covering them with opaque materials. Covering materials such as black nylon and straw can be used for mulching.

Pine needles are an inexpensive and attractive mulch material for gardens

2.2.7.3. Biological control

As known, biological control, with its shortest definition, is the control of a living thing with another living thing. Biological weed control is all of the applications made to reduce the density of these weeds below the economic damage threshold by using living organisms that negatively affect the population of weeds (Table 4).

Table 4. Natural enemies used in the control of weeds

Weed type	**Natural enemy that can be used**
Euphorbia rigida	*Hyles euphorbia Denticera divisella*
Cynodon dactylon	*Drechslera cynodontis*
Cichorium intybus	*Amerosporium concinnum*
Centaurea solstitialis	*Ceratapion basicorne C. onopordii*
Acroptilon repens	*Auclacidea acroptilonica*
Tamarix ramosissima	*Liocleonus clatratus*
Orobanche spp.	*Phytomyza orobanchia*

2.2.8. Harvest

A few drops of grape juice are dropped into the refractometer eye and the water soluble dry matter (SSC) value can be read directly. The most important criterion showing the ripening of grapes is the accumulation of SSC in the grain. But this alone can lead to misconceptions. Therefore, the evaluation of SSC together with the amount of acid is a more realistic criterion for determining the harvest maturity. In this respect, the ratio of dry matter to acid is determined as a percentage called the maturity index. In order to harvest table grapes, the maturity index is required to be at least 20/1. Although the maturity index values vary according to the cultivars, they are generally between 25-35/1 in Türkiye.

SSC in dried grape varieties; It is a very important issue that affects many factors such as drying time, drying efficiency, storage, quality and marketing. In terms of high yield and quality, the best SSC level for seedless grapes is 22-23%. Since the SSC level is affected by many factors, especially the development of the vineyard and climatic conditions, it coincides with different times every year. Taking time into account in harvest determination can lead to quality losses. For example, while the raisin yield is around 19% in a harvest with 18% SSC, the yield reaches approximately 26% in a 23% harvest.

Grape clusters are mostly harvested by hand. Harvesting is performed using knives, scissors or power tools. After all the clusters have been cut, the workers place the clusters in the baskets. Harvesting should be performed as cool as possible, ie early in the morning. The clusters should be harvested by holding the stem without taking it into the palm. It is necessary to disinfect the tools and equipment used in Conventional Production during the harvest. Harvesting should be performed in accordance with the principles of organic agriculture.

2.2.9. Drying the Grape

Almost all of the organic certified products produced are exported to developed countries, especially EU countries, USA and Japan. Türkiye is the leading country in the dried and dried fruits market, which initially helped the development of organic agriculture.

In order to organic raisins to be preserved for a long time, the moisture content must be lowered below 15% before storage. It should be determined in a healthy way according to the TS 3411 Seedless Raisin Standard that the drying has ended. This can be easily determined either according to the maximum humidity level of 15% that can be determined in laboratory conditions, or by the fact that the grapes, which are squeezed practically by taking it in the palm, disperse easily when left. Drying of organic grapes is using the open drying method. Sulfurization method is definitely not used

2.2.10. Storage

The warehouses where the organic grapes will be stored should be clean, dry and free of fungi and insects. The materials to be used in storage should be new or the existing materials should be used in accordance with the rules, free from dirt and dishes. There should not be any substance in the warehouses that emits bad odor and is likely to contaminate the products. Organic products should not be stored together with conventional products. Any substances other than those allowed for cleaning, disinfection and organic agriculture (TS 12611) should not be used and methods should not be applied.

2.2.11. Packaging

General rules about the places where raisins will be packaged are given in TS 30576. Raisins are stored as packaged. The packages must be made of new, clean, dry, odorless material that does not impair the properties of the goods inside and that will not cause contamination. Raisin packages are stored in food-safe and UV-resistant polypropylene sacks with a net capacity of 50 kg (+/- 10%) or in food-safe plastic crates that can hold up to 400 kg of product. Maximum 8 sacks are placed on top of each other in the warehouse. The packages are kept in cool, airy places that are not wet and damp, and spaces are left between the rows in order to ventilate the stacks. The packages of organic grapes should be prepared from new, clean, dry, odorless, most suitable materials that will keep the products in good condition and not harm health during transportation, storage and marketing. According to the amount of the product in this packaging, it is preferable to have a wooden or cardboard case

2.2.12. Labeling (marking)

There should be a label on the containers and packages in which the grapes, which have been approved as organic by the Control or Certification Body, are placed in the transport, processing and storage warehouses, even temporarily, from the harvest, in order to prevent any mixing. In this label, the type and organic nature of the product should be stated, as well as the name of the manufacturer, the place of production and the name of the parcel, the lot number and the name or logo of the Control or Certification Body.

Inks and adhesives used in writings and labels should not be harmful to health. On the label to be used on the final packaging, it should be clearly stated that the product is produced organically and;

- Organic product logo (symbol) in compliance with the legislation,
- Name, code number and logo of the Control or Certification Body,
- The certificate number issued by the Control or Certification Body for this product,
- Name of the product,
- Lot number,
- Group,
- Class,
- Type,
- Length,
- Net mass (kg),
- Product year,
- Production date,
- Country (for products to be exported),

- Production place (Province, District, Village, Parcel and Lot No.),
- The mark and number of this standard,
- Recommended expiration date,
- Trade name (abbreviated name or registered trademark) and address of the marketing firm,
- Names and quantities (if used) of additives and non-agricultural ingredients.

This information can also be written in Turkish and foreign languages when requested. The information on the packaging must be legible, written or printed in such a way that it cannot be easily erased and deteriorated. Apart from these, texts and pictures can be placed as advertisements, provided that they are not contrary to the contents of the package, and are not deceptive or misleading.

Issues we are lucky in organic viticulture

- ➢ Organic Seedless Dry
- ➢ Table Grapes
- ➢ Organic Molasses
- ➢ Organic Wine

Organic viticulture development

- ➢ Development of the internal market
- ➢ Integration with different sectors (agro-ecotourism, increasing organic markets, etc.)
- ➢ Strengthening and supporting small producers in organic agriculture
- ➢ Infrastructure (input, operation, quality systems, etc.) needs to be strengthened

Organic viticulture is an "environmentally friendly" approach to viticulture and is a rapidly developing sector for grape and food production in many countries. However, it is a production system that recommends eliminating synthetic fertilizers and pesticides and controlling the organic matter cycle on the farm to increase and maintain organic soil fertility.

Organic viticulture is not a luxury. It is not an impracticable agriculture as many think, however a form of agricultural production that requires knowledge and analysis. Organic viticulture is an opportunity given to us by the low pollution of our country's geography and climate characteristics. When we evaluate these opportunities in our country, there will be an increase in organic grape production and products obtained from organic grapes.

3. CONCLUSION

In the pandemic process caused by Covid-19 at the beginning of 2020, people have tended to demand and consume products that do not disturb the natural balance in the face of the negative effects seen in humans and other living things. For this purpose, the demand for environmentally friendly farming methods is increasing day by day. Organic agriculture is one of environmentally friendly farming methods that aims to evaluate the future of human and ecosystem as an indivisible whole with healthy plant and animal production. Organic grape growing is a production system that promotes and improves agro-ecosystem health, including biodiversity, biological cycles and soil biological activity.

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CHAPTER 7

A TRAINING SYSTEM SPECIFIC TO ECOLOGY AND VARIETY: "BARAN"

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INTRODUCTION

Grapes are one of the most grown fruits among horticultural crops worldwide, and they have valuable nutritional content for human health (Keskin et al. 2009; Karadoğan and Keskin 2017; Keskin et al., 2021; Kaya et al., 2022). Grapes are produced on an area of 7.7 million ha in 93 countries around the world. The country with the largest grape production area in the world is Spain with 936.890 ha, followed by France, China and Italy, respectively. Türkiye ranks 5th with a grape production area of 405.439 ha. Grape production in the world was 77.1 million tons in 2019 and China is the country that produces the most grapes in the world with 14 million tons, followed by Italy, the United States, Spain and France, respectively. Türkiye ranks 6th in grape production with 4 million tons in the world (FAO, 2021). On the other hand, grape, which has an important production area and amount, allows the use of barren and sloping areas that are unsuitable for the growing of other agricultural products (Kaya and Kose, 2018; Kalkan and Keskin 2018; Kaya, 2020a; Kalkan et al., 2022; Keskin et al., 2022). It has, indeed, been reported that grape can be grown in many regions of Türkiye, except for some regions such as the high parts of the Central and Eastern Anatolia Region and the coastline of the Black Sea Region (Doganay, 1995; Kaya et al., 2021). Considering the grape area and production regions in Türkiye, Aegean, Mediterranean, Mid-South, Southeast, Northeast and Black Sea regions come to the fore. Apart from these regions, important grape varieties can be grown in provinces such as Erzincan and Iğdır, which have different microclimate characteristics (Kaya and Kose, 2020; Kaya, 2020b,c). Basically, the existence of natural conditions in and around the Erzincan plain has a vital role in the execution of viticulture and fruit growing activities (Kaya et al., 2021). The 'Karaerik' (*Vitis vinifera* L.) (syn) Cimin) grape variety, which is the only standard variety of the Northeast Agricultural Region, plays a key role in Erzincan viticulture. For the variety grown in 90-95% of the vineyards in the province, 6000 tons/year of product is taken from 9000 da of land (Köse et al., 2018). It has been widely grown as table grapes in Erzincan (Bayırbağ, Göller, Pişkidağ, Karakaya, Çadırtepe) for years and has a very high market value (Güneş et al., 2015). The berries of this grape variety have a special aroma (a subtle taste between slightly sweet and sour, not found in other *V. vinifera* grape varieties) (Kalkan et al., 2012). In addition, this grape variety is cultivated by applying a special training system called Baran in the region.

Baran training system, which has a history of at least 200 years in the region, is a traditional training system developed to cope with cold winter conditions (Kaya and Kose, 2020). This system is an ancient artifact that is integrated with the climatic conditions of the region and provides the opportunity to grow grapes to viticulture producers despite the cold winter conditions (Köse et al. 2018; Kaya, 2020a; 2020b,c). It also provides great advantages for bringing sloping lands to agriculture as well as preventing irrigation problems. The creation of the system requires a lot of effort and time, and it still shows as a distinctive symbol for the vineyards of the region (Kalkan et al., 2017; Kaya and Kose, 2017; Buztepe et a., 2017; Rende et al., 2018). In this study, it is aimed to introduce the Baran training system, which is an ancient system and applied in Erzincan vineyards, and to reveal its general characteristics.

Establishment of the Baran training system

The Baran system is generally composed of prostrated shaped mounds with a depth of 80-100 cm and a width of 60-80 cm, 1.5-2 m between rows and 3-3.5 m between rows (Figure 1-2). Although the length of the rows varies according to the region shape, they generally have a length of 30-50 m. After prostrate shape and planting pits are formed, 50-80 cm long grape cuttings are prepared, and before bud burst are planted in March-April at a distance of 1.5 meters to the south side of the prostrate shaped pits (Kose, 2002).

Figure 1. Preparation of the planting pit and prostrate structure for the vines (Photo: N.N. Kalkan)

After the cuttings are planted, the lower leaves and axillary shoots on the one-year-shoot in May and June are removed. Then, in July-August, the apical of the shoots are cut and the basal of the cuttings are filled with soil. As seen in Figure 2, these processes continue for 2-3 years until the shoots rise above the Baran.

Figure 2. Planting the vines on the Baran (Photo; N.N. Kalkan)

The vines, which are three or four years old, reach the level of a mound on the Baran and 4-5 shoots are left in the vines depending on the development strength. One-year-old shoots on these vines are pruned short over two, three or four buds in winter pruning (Figure 3-4)

Figure 3. One-year-old shoots on Baran (Photo: N.N. Kalkan)

When the vines are 6-7 years old, a form of training system is formed that completely covers the Barans (Figure 3).

Winter pruning in spring in the region is usually carried out in late March to early April. In this system, vines are generally pruned short depending on the development of a one-year shoot and the number of buds on the shoots varies between 2-4. Cultural practices in the vineyard are carried out in the form of aeration of the soil both between the Baran and in the prostrated shaped mounds with the form of prostration (Figure 4). In addition, green pruning is performed at regular intervals from May to mid-August.

Figure 4. Defoliation in vines trained with the Baran training system (Photo: N.N. Kalkan)

Figure 5. Cultural practices in the vineyard (Photo: N.N. Kalkan)

Advantages of Baran training system

Considering the training system of the vine and in the winter snow cover, Baran training system is possible to state that it is a natural factor in the protection of the sticks against winter cold (Kaya, 2020b,c), (Figure 6).

Figure 6. A view of the trunk and shoots from under the snow (Photo; N.N. Kalkan)

Indeed, since the trunks of the vines and the arms on these stems are buried in the soil in the Baran system, the snow falling in winter covers the vines and thus the buds are protected from the cold effect of the external environment. On the other hand, the Baran training system helps to obtain earlier and higher quality grapes by taking advantage of the soil temperature of the vines in summer (Kose, 2002). In addition, the system serves to bring such areas to agriculture by providing terracing, especially in areas with high slopes, and facilitates the need for sunbathing of the vine in the vineyards (Akpınar and Yiğit, 2006), (Figure 7).

Figure 7. A view from the baran system on a high inclined area (Photo; N.N. Kalkan

Besides, it has been reported that Barans have a feature such as eliminating the harmful effect of wind as well as water holding properties.

Disadvantages of the Baran training system

Cultural practices in this system require intense labor. Since the canopy is close to the ground, shoots and clusters cannot benefit from sunlight adequately, especially in the bottom lands, and the desired effectiveness in chemical control cannot be achieved. When the structure of the Baran system is added to negative factors such as precipitation, dew and fog, the development of fungal diseases, especially with the cracking of the berries towards the harvest time, reduces the yield and quality (Akpınar and Yiğit, 2006).

Moreover, the system is not suitable for tillage with machinery, and it is not satisfactory for adjusting the number of shoots (bud number) to be left in pruning, which is an important element in vine. However, when the modern training systems are compared with the Baran system, the proportional profitability of the stringed finishing systems was found significantly higher than the Baran system. This is due to the high production costs in the baran system, as well as the low yield per hectare (Kalkan et al., 2017;2022).

Conclusion

Factors such as vine development, climate and cost are important in the selection of cultivation systems in viticulture. The multiplicity of determining factors has led to the emergence of many training systems. There are many training systems designed in different ways for vines in different regions of our country. Baran training system, which has a history of at least 200 years in Erzincan region, which is one of these regions, is a traditional training system developed to cope with cold winter conditions. The Baran training system is an ancient legacy that is integrated according to the climatic conditions of the region and enables the producers to grow grapes despite the cold winter conditions. The Baran system also provides great advantages for the evaluation of sloping lands and to prevent irrigation problems. This system, which requires a lot of effort and time to set up, is heavily applied in the vineyards in the Erzincan region today and still stands out as a distinctive symbol of the region.

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CHAPTER 8

TURKISH AGRICULTURAL INSURANCE SYSTEM AND VITICULTURAL PRACTICES

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INTRODUCTION

The agricultural production, it is very important for the world's population, has a unique structure, which incorporates a highly sensitive field of activity and which is significantly affected from the economic, social, political, technological, and personal risks (Sümer & Polat, 2016). With playing the most important role in feeding the constantly increasing human population, the agricultural production activities are performed by taking many risks under many ecological and economic factors. Besides the human-caused failures, malpractices, and diseases and pests that can be controlled using various methods, there also are many other risks. It is impossible to estimate and control these risks. However, the measures to be taken may decrease the effects or the damages to occur may be eliminated after the incurrence of risk. The natural events such as earthquake, landslides, hail, flood, and storm have been affecting the human life since the beginning. Even though various technologies are used in estimating these natural events and various warning systems were developed for this purpose, these options mainly remain incapable. The most efficient method that can be used after taking all the required measures from the aspect of cultivation is the agriculture insurance. Since the agriculture is a risky production zone, this risk transfer instrument is irreplaceable one.

Risk can be defined as the probability of damage or the possibility of the occurrence of an undesired event. In the insurance discipline, the concept of risk, which can be defined differently in different scientific disciplines, is defined as the presence of a risk of loss, the possibility of loss, uncertainty, and the possibility of the occurrence of an outcome other than the expected one (Sarıoğuz, 2007).

From a broad perspective, risk management is to plan, organize, and control the necessary activities and resources in order to minimize the effects of uncertain events. For the stable continuity of business, the objective of risk management is to enact the necessary regulations, protect the merchandise and individuals in the organization, and keep the business' power to earn (Çağırgan, 1997).

The measures that can be taken in risk and uncertainty management would have different effects on agricultural activities. None of those measures can solely offer protection against all the risks and uncertainties. For this reason, producers should use a combination of alternative instruments. In case of insufficient technical measures and inability to measure the risk levels, agricultural products and other agricultural goods can be secured by using insurance (Sümer & Polat, 2016).

Insurance, which is a social instrument, refers to individuals and organizations supporting each other through small premiums in order to provide economic safety against large potential damages. People uniting against risks and consequently sharing the damages, which they would bear solely, constitute the basis of the insurance system. Insurance is a contract and the subject of this contract is to compensate for the relevant risk. In the insurance system, individuals and companies transfer the risk, which they bear, to the insurance company by paying premiums (Sarıoğuz, 2007).

The different practices were left via the **Law on Agriculture Insurances** No.5363 enacted in 2005 and the "State-Supported Agriculture Insurances System" gained a legal foundation (Anonymous, 2022b). In this system, in which many risks observed in herbal and animal production are covered, majority of the premiums are borne by the state.

Agricultural insurances do not only help protect the existing subjects but also further improve agriculture. Within this context, agricultural insurances play a role facilitating the producers' access to credits from the banks. Producers, who can easily access funds and gain a stable income by securing their current assets, invest in order to make use of new agricultural technologies and techniques and, thus, increase agricultural production. In conclusion, agricultural insurance is an instrument that is necessary for an advanced agricultural sector (Nnadi et al, 2013).

1. AGRICULTURE INSURANCE SYSTEM IN THE WORLD

The term "agriculture insurance" has been firstly pronounced by Benjamin Franklin. On 24 September 1788, he expressed that he had thought about if it is necessary to establish an insurance agency in order to protect the farmers against the events such as storm, disease, pests, and etc. since heavy storm damaged the products of farmers in French coasts, because a small premium that would be paid by the farmers would significantly compensate their suffer and losses (Bhavan, 2012).

The agriculture insurance implementations started in 18th century in Europe. No success could be achieved because of the reasons such as inability of preventing the catastrophic risks in agricultural activities, region's climate and soil structure, and insufficiency in recording the products being cultivated. After this unsuccessful beginning, the agriculture insurance has come to the fore since the predictability of catastrophic risks became clearer thanks to the technological advancements (especially those in meteorological estimations) in 19th century (Özdemir & Baylan, 2017). The first hail insurance program has been put into practice for the grape farmers in France and Germany in 1820, whereas the first insurance program in USA has been started for tobacco in 1883.

There is a consensus throughout the world on the fact that the agricultural producers should be protected from the covered risks since the agricultural producers face with a wide range of risks. Because, this would provide the farmers with an income security on one hand and it would also ensure the food safety of countries on the other hand. For this reason, it can be stated that the private and public sectors run a common program in agriculture insurance area (Chatterjee & Oza, 2017).

In the world, the agriculture insurance practices emerge based on various factors such as risks that are observed, risk management instruments preferred, and development level of country. For instance, in Europe where the agriculture insurance has been implemented for the first time, there is protection only against the risk of hail, whereas the agriculture insurance instruments offering protection against many risks are in use in Spain, which is also a European country. Another difference in practice is the role of state in agriculture insurances. From this aspect, the agriculture insurances are under full control of the state in countries such as Greece or completely managed by the private sector in countries such as England. On the other hand, a mixed structure is preferred in countries such as Spain.

As of the year 2007, the agriculture insurances are offered by the private insurance companies in 54% of 65 countries, whereas the percentage of countries, in which this service is provided by the public sector, is only 9%. The percentage of countries offering the agriculture insurance via public-private sector cooperation is 37%. Moreover, it can be concluded that the private insurance is more popular in developed countries when compared to the other countries (GIZ, 2016). This is of course a result the fact that the public sector bridges the gap in countries, where the private insurance companies are insufficient.

Regarding the state support to the agriculture insurances in different countries throughout the world, the highest level of state support is seen in Indonesia with 100% state support, followed by the USA with percentages between 72 and 90%, Spain with 70-90%, Japan with 50%, and Brazil with 30-60% (Reyes, 2017).

The insurance funds should cover both of the compensations and the possible disaster risks. The state-run insurance programs have advantage in accessing to the financial sources. The pool system might ensure the optimal use of funding options. On the other hand, the private insurance companies have limited funds. Thus, the compensations might not be paid rapidly and the farmers may suffer more. All of the expenses of insurance programs in Türkiye, Spain, China, Argentina, Philippines, and Ukraine are funded by the Pool system (Keskinkılıç & Alemdar, 2013).

Under the light of this information, it is impossible to discuss a uniform agriculture insurance system throughout the world and it cannot be stated that the system in any country cannot be successfully implemented in another country. However, undoubtedly, examining the failures and successes of practices in other countries would shed light the road in developing a better agriculture insurance system. Hence, the attempts to implement agriculture insurance resulted in failure in various countries (Tekin, 2015; Anonymous, 2018d).

Of course, the national conditions play important role in establishing a successful agriculture insurance system. However, the studies emphasizing the points to consider in establishing national systems should not be ignored. Making the first systematic warnings on this subject, Jain (2004) grouped the points-to-consider under 3 phases and named these steps as determining the actual status, developing the program, and ensuring the sustainability of program.

On the other hand, in a meeting organized in Vietnam in 2016, the points to consider in establishing a state-supported national agriculture insurance system were grouped under 10 subjects. However, for establishing the national agriculture insurance programs, the survey method is recommended in order to determine the producers' perspectives to and expectations from the insurance, and a survey template was presented (Asean, 2017).

Finally, since the agriculture insurance program would also cause the state to subsidize the agriculture sector in that country, it is normal that there would be differences between the countries (Branstrand & Wester, 2014). Moreover, the governments should also pay attention to the factors (population growth, size of agricultural companies, poverty in agricultural sector) influencing the agricultural policies of state and consequently the food security today and in future (Hatch et al, 2018).

2. STATE-SUPPORTED AGRICULTURE INSURANCE SYSTEM IN TURKIYE

Although the agriculture insurance is a completely commercial activity, it can be noted that the state has frequently played important roles in this field. The most important reason for this is to ensure the continuity of economic productivity and the wealth of rural population. There is no insurance product, which meets all the expectations of producers and which is universally valid (Iturrioz, 2009). From this aspect, it is normal that the countries apply acceptable and sustainable state supports through the insurance programs designed in parallel with the conditions of those countries.

In Türkiye, the agriculture insurances started with insuring the herbal products against the risk of hail. In year 1957, Şeker Sigorta (Şeker Insurance Co.) introduced the "Hail Insurance" and the "Farm Animals Insurance" was introduced by Başak Sigorta (Başak Insurance Co.) in 1959 (Perçin, 2011) (Dinler et al., 2018).

The law, which provides for the agricultural insurance system to have a legal basis in Türkiye, was published in the Official Gazette dated 21.06.2005 and entered into force (Anonymous, 2022b). Together with the Law on Agriculture Insurance No.5363, the "Risk Management in Agriculture" gained a new dimension and accelerated. The principles of compensating the losses of farmers arising from the risks specified in law were determined, as well as the implementation of premium support in agriculture insurance.

In order to establish the legal basis of TARSİM (Turkish Agriculture Insurance System), the regulations ("The Regulations of Agricultural

Insurance Pool Operating Procedures and Principles” and “The Regulation of the Application of the Agricultural Insurance”), General Terms, and Tariffs and Instructions were prepared and enacted upon the approval of Minister of Treasure and Finance. The state-supported agriculture insurance policies were issued since 1 June 2006 (Anonymous, 2018e).

Although different models are used in countries implementing the agriculture insurance in the world, it was determined at the end of analyses that the most suitable method for our country is the pool system successfully implemented in Spain. For this reason, after enacting the Law on Agriculture Insurance in 2005, a pool system was established and the TARSİM (Agriculture Insurance Pool Co.) was founded as the operator company in order to run the system (Anonymous, 2018). Issuing its first policy in 2006, TARSİM is managed by a Board consisting of 2 members from the Ministry of Forestry and Agriculture, 2 members from Insurance and Private Pension Regulation and Supervision Agency, 2 members from non-governmental organizations (Union of Turkish Agricultural Chambers and Association of Insurance, Reinsurance and Pension Companies of Türkiye) and 1 member from the operator company TARSİM. The responsibility and duties of board, pool, and operator company are clearly specified in Law on Agriculture Insurance No.5363 in Official Gazette dated 21.06.2005 (Anonymous, 2022g).

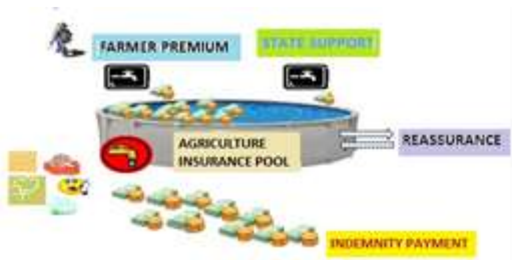

Figure 1. Agriculture Insurance Pool (TARSİM)
(Source: www.tarsim.gov.tr)

In Turkish Agriculture Insurance System (TARSİM), the premiums paid by the farmers and the premium contributions borne by the state are collected in a pool, and the damage compensations are paid to the farmers by making use of actuarial method and reassurance plans (Figure 1). Under favor of this risk transfer, by making premium payment and making use of premium contribution offered by the state, the farmers overcome the insured risk from the funds accumulated in the pool if they face with the risk that has been covered in the insurance policy. In other words, they have the chance to protect their actual assets and continue their agricultural production.

In order to purchase the protection in relation with the risks it undertakes, the Pool makes reassurance contracts with the national and international corporations. In case that the necessary protection cannot be procured from the national and international markets, the portion to be determined by the Council of Ministers shall be guaranteed by the State. In case that the protection obtained from the national and international markets (reassurance) is insufficient or the damage exceeds beyond the expectations because of extraordinary conditions, the additional allowance to be determined by The President shall be allocated to the Pool as the Allowance for Excess of Loss in order to compensate the losses (Anonymous, 2018a).

As stated by Durgut and Dumanoğlu (2016), the premium support offered by the state in agriculture insurance system encourages the producers and it also increases the premium production. This increase is a result of the increase in insured area and the number of insured animals.

The amounts of premium contribution to be offered by the State are determined by The President upon the proposal of Board of Pool and approval of Ministry in terms of the products, risks, regions, and company sizes every year. As of the year 2022, the state covers 50% of the insurance premium that should be paid by agricultural products insuring their agricultural products, greenhouses, cattle, and small cattle, poultry, aquaculture, and hives (2/3 for frost guarantee), 70% for the Village-Specific Drought Yield Insurance, and 60% for the Revenue Protection Insurance. The objective is to encourage the producers to make use of insurance (Anonymous, 2022d).

By the Article 11 of Law on Agriculture Insurance No.5363, TARSİM is subject to annual audit by the Insurance and Private Pension Regulation and Supervision Agency from the aspect of insurance practices and by the Ministry of Forestry and Agriculture from the aspects of all other administrative and financial transactions. With this structure, the system is run as an insurance system that is under supervision, audit, and guarantee of State (Anonymous, 2018a; Anonymous, 2022d).

As seen in Table 1, TARSİM constantly grew since its first day; the total sum insured increased from 211,290,594 TL in 2006 to 124 397 000 000 TL in 2021. A similar increase can also be observed in the number of policies. For instance, the number of policy that was 12 330 in 2006 increased to 2 517 704 in year 2021.

Table 1: General Appearance of Growth in Agriculture Insurance 2006 - 2021

	Total Sum Insured (TL)	**Total Premium Production (TL)**	**Number of Policies (N)**	**Loss Compensation (TL)**
2006	211 290 594	4 450 852	12 330	
2007	1 478 414 663	64 103 578	218 938	43 905 528
2008	2 224 971 605	98 443 549	260 944	44 100 874
2009	2 900 559 617	120 348 681	306 770	95 231 940
2010	3 987 866 529	185 433 744	371 116	121 399 481
2011	6 986 308 699	440 879 023	587 716	225 227 838
2012	9 497 476 828	499 348 870	744 093	280 266 706
2013	11 252 737 360	526 835 325	891 876	410 857 897
2014	13 894 743 746	683 535 994	1 086 612	532 284 864
2015	18 378 031 469	965 772 197	1 375 390	724 802 873
2016	23 080 720 277	1 299 986 302	1 444 277	840 963 512
2017	30 303 347 858	1 628 553 789	1 598 269	833 085 483
2018	42 217 541 073	2 050 635 088	1 756 428	1 065 106 035
2019	55 166 348 492	2 447 064 788	2 087 860	1 226 860 024
2020	83 146 049 745	3 198 743 163	2 235 626	1 392 944 782
2021	124 397 000 000	4 678 000 000	2 517 704	2 437 000 000

Source: www.tarsim.gov.tr (Anonymous, 2018i; 2018j; 2018k; 2018h; 2022e; 2022c; 2022g)

Successful implementation of Türkiye's State-Supported Agricultural Insurance System was analyzed and taken as a model by different countries. To establish a similar system in Azerbaijan, the "Cooperation Intention Declaration" was signed on 27th February 2020 by Tarsim and Azerbaijan Insurance Fund. Issuing its first policy, Agrar Insurance went into action on 3rd November 2020 (Anonymous, 2022f).

2.1. Insurance Experts in Agriculture Insurance System

The insurance experts play the most important role for the proper functioning of system by determining the damages, productivity, and status and reporting the results to TARSİM. The ones having "Basic Training of Insurance Expertizing" organized by the Ministry of Forestry and Agriculture and then passing the exam are entitled to work as insurance expert and they fulfill their duties independently.

With their practical knowledge and skill, the experts constituting one of the most important parts of system play important role in acceptance and popularization of Agriculture Insurance System by the farmers, as well as the other factors such as personalities and if they have moral deficiencies.

The training has a significant role in proper functioning of the system. The training and knowledge experts and agents on the subject are the most important factors influencing the success. It is clear that the training of farmers is as important as that of experts because it was determined in the previous studies (Branstrand&Wester, 2014) that the farmers generally tend towards the insurance when they face with a risk, which they cannot handle, and when they are sufficiently informed about the scope of insurance. Thus, the training becomes more important. There is no doubt that the sufficient and accurate training of farmers would have positive effect on understanding and popularizing the system and increasing the trust. Hence, Villalobos (2013) expressed that the state's role in agriculture insurance is to establish a fast, reliable, high quality, and transparent data infrastructure, to provide education, training and technical support, and to design regulatory frame.

2.2. Insurance Branches in State-Supported Agriculture Insurance System of Türkiye

In Turkish Agriculture Insurance System (TARSİM), the "**multiple risk insurance**" is implemented in 7 different branches. The "District-Based Drought Yield Insurance", which was started with wheat product in 2017, was extended in 2018 by adding the barley, rye, oat, and triticale products into the insurance coverage.

In order for the producers to be able to have insurance and benefit from the state's premium contribution, it is compulsory to be registered and updated in the relevant system in the Ministry of Agriculture and Forestry (Anonymous, 2022d).

The insurance branches and scopes of State-Supported Agriculture Insurance implemented in Türkiye have been established as follows (Anonymous, 2018h).

2.2.1. Herbal Product Insurances

- The crop losses arising from the risks of hail, storm, whirlwind, landslide, earthquake, fire, and deluge in all the herbal products; The crop losses arising from the risk of frost including the blooming period only for the fruits (in addition to the risks specified above)
- The quality loss caused from the risk of frost in vegetables, fruits, and cut flowers
- The damages in hail net and cover systems because of the risks of hail severity, storm, whirlwind, fire, landslide, earthquake, flood, and deluge in fruit orchards and vineyards
- Stem for the wheat, oat, rye, barley, and triticale products
- The crop losses in farm products, vegetables, strawberry, and sapling because of wild boars
- The crop losses in cherry in ripening period because of the risk of rainfall
- The crop losses in the leaves of grapevines grown for brined vine leave production because of the risks of hail, storm, whirlwind, earthquake, landslide, deluge, fire, and flood
- The complete damages in economically productive fruit trees and grapevines and their saplings because of hail, storm, whirlwind, fire, earthquake, landslide, flood and deluge, vehicle impact, and snow weight risks
- The loss of crop in wheat and their certified seed plants caused from the risks of drought, frost, hot wind and hot air wave, excessive moist, and the factors other than the hail package (hail, storm, whirlwind, fire, flood and deluge, earthquake)

2.2.2. Greenhouse Insurance

- The losses and damages in greenhouse construction, cover material, and technical equipment (including the products inside the greenhouse) because of hail, storm, whirlwind, fire, landslide, earthquake, vehicle impact, snow and hail weight, and flood and deluge

2.2.3. Cattle Insurance

- Any kind of animal diseases, pregnancy, labor or surgical interventions (excluding from those specified in General Terms), accidents, wild animal attacks, snake and insect bites, poisonous meadow grasses and poisoning because of fodder
- Deaths and obligatory slaughter because of natural disasters and sun prostration, fire, and explosion
- Abortion and calf deaths within one week after the birds for dairy cattle
- The deaths of foot and mouth disease of cattle in the companies, which have Certificate of Free-From-Disease, (this is an optional clause and is conditioned on paying extra premium)
- Obligatory slaughters, abortion
- Material losses of insurance holder because of calf death
- The direct losses of insured animals because of theft or attempted theft while the insured place specified in the policy or meadow(s) (which are requested asked by farmer and accepted by TARSİM) or during transportation between these addresses

2.2.4. Sheep and Goat Insurance

- Any kind of animal diseases, pregnancy, labor or surgical interventions (excluding from those specified in General Terms), accidents, wild animal attacks, snake and insect bites, poisonous meadow grasses and poisoning because of fodder, and deaths and obligatory slaughter because of natural disasters and sun prostration, fire, and explosion

- The deaths of foot and mouth disease of cattle in the companies, which have Certificate of Free-From-Disease, (this is an optional clause and is conditioned on paying extra premium),
- The direct losses of insured animals and the losses arising because of theft or attempted theft while the insured place specified in the policy or meadow(s) (which are requested asked by farmer and accepted by TARSİM) or during transportation between these addresses

2.2.5. Aquaculture Insurance

- The diseases other than the exceptions specified in General Terms,
- The deaths and losses arising from the causes such as pollutions and poisonings beyond the control of fish farmer, any sort of natural disaster, accident, predators, and algae bloom,
- Direct losses in cages and nets that the insurance holder suffer due to the causes such as natural disaster, accident, and predators

2.2.6. Poultry Insurance

- Death, killing, and obligatory slaughters arising from various poultry diseases, accidents and poisonings, natural disasters, fire, and explosion

2.2.7. Bee Hive Insurance

- Risks of storm, whirlwind, fire, landslide, earthquake, flood and deluge, vehicle impact, and wild animal attacks,
- In migratory beekeeping (in addition to the risks specified above), the risks of strike, impact, overturn, and burn during the transportation of hives

2.3. Viticultural Practices in State-Supported Agriculture Insurance System of Türkiye

The agricultural industry has played an important role in the Turkish economy and the revenues of society. Many products produced in the country such as grape, hazelnut, apricot, fig, olive, and lentil are exported and highly demanded in the global market for their quality.

With its climatic and soil characteristics, Türkiye has a large product variety and ranks high in viticultural production, especially in dried grape production. Except for the regions having a high level of precipitation in the Black Sea region, the grape can be cultivated in all the regions of Türkiye. Given the data for the year 2021, Türkiye has 3 902 211 decare area of vineyard and 3 670 000 tonnes of fresh grape yield (Anonymous, 2022h).

In Türkiye, where viticulture has such an important place, agricultural insurance is of significant importance thanks to its role in this field. As a result of global warming, natural disasters such as climate changes, uncontrolled precipitation, and frost might leave grape farmers in a difficult situation. Since it is not possible to modify or prevent the climatic factors, the best option is to protect the farmers and take various measures. Besides the measures such as conscious cultural practices, accurate variety selection, and use of technology, also the supports that would ensure sustainability in production, are needed. The presence of an instrument, which would allow the farmer to survive natural disasters by compensating for the loss of product, would ensure the continuity of production and ensure that farmer would feel secure. This is the reason for the agricultural insurance system, as well as to insure the products of farmers and keep them strong by ensuring the stability of their revenue even after a loss.

2.3.1. Guarantee Coverage in Viticulture

For a grape farmer to be able to insure the vineyard, he/she must register his/her land and products to the systems of the Ministry of Agriculture and Forestry (Farmer Registration System - ÇKS) or update the existing information/registration. Then, an insurance policy can be issued via any agent of authorized insurance companies. In the Turkish State-Supported Agricultural Insurance System (TARSİM), grape farmers are provided with coverage for grape, grapevine, sapling, hail net, cover, support systems, and grape leaf for pickling, the available coverages were determined, and insurance policies were established in this parallel (Anonymous, 2022).

Grape is under guarantee against the quality loss due to hail and the yield loss due to flood and stream, as well as vehicle hits, earthquake, landslide, fire, hail, hurricane, and storm. Moreover, in the grape product, frost risk and yield loss due to rain and hot weather can be taken into coverage optionally.

Vines and Saplings are in the scope of coverage against the hail, storm, tornado, fire, earthquake, landslide, flood, snow weight, vehicle hit, hog and frost risks.

Hail net and cover and support (wired nurture) systems are in the scope of coverage against the risks of hail, hail load, snow load, storm, hurricane, earthquake, landslide, fire, flood, stream, and vehicle hit.

Grape leaves for pickling; yield of loss in grape leaves due to hail, storm, hurricane, fire, earthquake, landslide, vehicle hit, flood, and steam risks can be taken into coverage optionally.

TARSİM has set beginning and termination times of guarantee for each product group:

- The guarantee against hail, frost, storm, hurricane, fire, earthquake, landslide, vehicle hit, flood, and stream begins with the awakening of the buds.
- Considering the guarantee against rain, grapes are under coverage between veraison and maturation/harvesting.
- Considering the hot weather, grapes are under guarantee during the blooming and fruit setting periods.

2.3.2. Government Contributions and Discounts for Premiums

For a farmer procuring agricultural insurance for his/her grape and vineyard, half of the heil insurance is paid by the state, as well as 67% of the frost insurance premium for the fruits. Moreover, while calculating the premiums, various factors can be considered in order to make discounts. These include (Anonymous, 2022ı);

- In case of renewing the policy after a no-damage policy period, the no-claim discounts applied are up to 30% for the heil package and up to 20% for frost risk.
- In case of paying the entire insurance premium in advance, a 5% cash discount is applied to the total insurance premium.

- In case that the risk prevention and minimization measures such as hail net and air stirring blades were taken, discounts up to 50% can be applied depending on the type of prevention.
- Female farmers, young farmers aged 30 years or younger, and disabled farmers can be offered different discount rates.

When the 10 products with the highest insurance rate in the Turkish State-Supported Agricultural Insurance System are examined, it is seen that grapes are in the first place in terms of premium production and second in terms of insurance price (Table 2). This shows how valuable the grape is for the farmer.

Table 2: Insurance Rates of Different Products

	Insurance Price (%)*			**Premium Production (%)***			**Number of Policies (%)***		
	2018	**2019**	**2020**	**2018**	**2019**	**2020**	**2018**	**2019**	**2020**
Wheat	24,9	20,8	21,8	12,7	11,4	12,1	40,8	35,6	33,7
Grape	**7,7**	**7,9**	**8**	**15,8**	**16,3**	**16,8**	**3,7**	**3,2**	**2,9**
Barley	6,1	6,5	7,5	3,6	4,1	5,1	12,2	13,5	16,0
Egypt	4,9	5,5	5,7	---	2,2	2,5	2,5	3,1	3,3
Sunflower	4,5	5,4	5,3	---	1,8	1,9	8,9	10,9	10,2
Hazelnut	4,1	4,2	4,6	5,8	6,0	6,1	5,6	4,8	4,8
Cotton	5,8	6,2	3,6	2,6	3,0	1,9	2,1	2,1	1,6
Olive	4,0	3,7	3,3	2,1	---	---	2,1	2,2	2,1
Rice	3,5	3,5	3,5	1,7	---	---	2,3	2,4	2,4
Apple	2,5	2,9	3,4	6,8	8,0	8,5	1,6	1,6	1,4

*: percentage of the total
Source: http://www.tarsim.gov.tr (Anonymous, 2022e; Anonymous, 2022c)

The Tarsim data are examined, it is seen in Table 3 that the risk that has occurred the most in grapes and has been paid for damage is hail. Although it varies according to the years, almost half of the total damage that occurs is hail damage. It seems that the most common form of damage after hail is frost damage (Table 3).

Table 3: The Most Common Risks in Vineyards and Their Occured Rates (%)

	2017	**2018**	**2019**	**2020**
Hail	37,4	61,7	54,7	48,8
Frost	39,8	28,4	29,3	32,3
Flood	1,8	1,8	8,4	7,4
Storm	8,0	5,5	3,9	7,2

Source: http://www.tarsim.gov.tr (Anonymous, 2022e; Anonymous, 2022c)

In their survey study on grape farmers cultivating different grape varieties in Manisa province, Yılmaz et al. determined that farmers' approach to agricultural insurance differed by educational level and age group. Reporting that the use of agricultural insurance increased with increasing educational level, researchers stated that the state support for premiums of farmers having low-income level also increased the insurance rate.

3. CONCLUSION

As known, the climate changes occurring as a result of global warming constitute an important threat for the agriculture and insurance sectors. The damages occurring at unexpected times and severity cause large-scaled agricultural losses and they also necessitate the strong agriculture insurance systems that will protect the producer against the risks. In developed countries, as in Türkiye, the support of the state strengthens the system's sustainability. This would serve for the food security of the country on one hand and the social justice and social peace on the other hand since, as a necessity of being a social state, the losses of disadvantageous (from the aspect of income level) segments of the population would be compensated.

The agriculture insurance does not only protect what currently exists but it also is an instrument contributing to the progression of agriculture sector. From this aspect, the agriculture insurance is a factor that eases the farmers' access to the bank loans. Hence, in this system, the banks act more confidently regarding the repayment of loans. The farmers, who can more easily access to the funds and achieve a stable income by protecting their current assets, invest in the new agricultural technologies and methods and, thus, they increase the agricultural production.

The agriculture insurance aiming to compensate the losses of producers, providing the long-term revenue stability, and offering social contribution are closely related with the public order from this aspect (Çiftçi, 2014).

In conclusion, any country should be able to meet its public's need for food. This, however, is closely related with the importance given to the agriculture in that country. The most important factor for the proper development of this sector, which is open to many risks, is the presence of an insurance system that protects the farmer from potential risks. Thus, it should be noted that the agriculture insurances play an important role in achieving the developed agricultural industry and ensuring the food security; the studies and subventions for improving the current system should be continued.

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CHAPTER 9

THE IMPORTANCE OF FERTILIZATION IN VINEYARD

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INTRODUCTION

Viticulture also has a very old history in Anatolia and is intertwined with Anatolian civilizations. Archaeological finds explaining the importance of viticulture have come from the Hittites, who created a great civilization of 600 years, and it has been determined that the viticulture culture in Anatolia dates back to 3500 BC (Karabat, 2000). Turkey ranks second after China in the production of table grapes worldwide. In Turkey, the Mediterranean region ranks first in terms of the total area of the vineyards where table grapes are grown, followed by the Aegean region. However, in terms of product quantity, the Aegean region is in the first place (FAO, 2019).

Sultani seedless grapes take the first place in the production of table grapes for export in Turkey. Sultani seedless grapes account for 95% of exports. Seedless grapes, which are grown in almost all provinces of Turkey and are subject to foreign trade, are mostly obtained from Manisa province (Figure 1). According to the statistics of 2021, Turkey is one of the leading countries in the world with a vineyard area of 435.227 hectares and grape production of 4 million tons. It ranks 5th in terms of area and 6th in the world in terms of production. 49.8% of the total grape production is table grapes, followed by 38.4% raisins and 11.8% wine-grape grapes (Anonymous, 2022).

Figure 1: Seedless grapes are mostly obtained from Manisa province.

Factors such as the selection and use of high-yielding varieties in the vineyards, the type of establishment of the vineyards, the maintenance, fertilization, irrigation of the soil, and the agricultural tools and machinery used to combat all kinds of pests, and their optimum use, are directly effective in increasing production and quality and play a very important role (Durgut and Arın, 2007) (Figure 2).

Figure 2: The establishment of the vineyards, the maintenance of the soil, its fertilization, irrigation

1. CLIMATE REQUESTS

Viticulture generally spreads between 200-520 latitudes in the northern hemisphere and between 200-400 latitudes in the southern hemisphere (Winkler, 1974). Although the vineyard has a warm-temperate climate; This fruit is a perennial garden plant with a high adaptability to colder and warmer climatic conditions. In terms of viticulture, climatic conditions such as temperature, sunshine duration, precipitation and wind are especially important.

With the daily average temperature above 10 °C, an increase in the vegetation period (>160 days) was detected (Ağaoğlu, 2001). A decrease in temperature below 18 °C during the development period slows down growth and development. If the air temperature rises to 35-40 °C during this period, blight (disease) occurs on shoots, leaves and grains (Çelik, 1998). Vines are sun-loving plants, as they accumulate a high amount of sugar in the grapes. During their development period, they need at least 1250-1300 hours of sun exposure (Figure 3).

Figure 3: The amount and duration of sun exposure of the vines

Viticulture can be done without the need for irrigation in places where the total annual precipitation is around 600 mm. It is not possible to make viticulture economically in areas with precipitation below 300 mm. In areas with precipitation over 900 mm, it is difficult to control fungal diseases in agricultural terms (Figure 4). At the beginning of the development period, light winds that do not exceed 3-4 m sec^{-1} are also beneficial in terms of establishing the plant-water balance in vines (Çelik, 1998). In terms of viticulture, the most unfavorable winds are winds blowing from the north and northeast in spring, reducing the temperature and causing breakage in young shoots, and strong winds blowing close to harvest, which cause injury to grains in table grape growing regions and loss of product and quality (Ağaoğlu, 2001; Kaymaz, 2010).

Figure 4: Annual precipitation of vines

2. SOIL REQUIREMENTS

The vine, which can be grown in almost any soil, is frequently preferred for reasons such as its adaptability to sloped lands, its ability to grow with less water, and the fact that it has many evaluation methods has caused it to be the most produced fruit in the world (Karabat, 2000). Adapting to different soil groups in our country as well as all over the world, the vine can grow in many different soil types, from the poorest barren soils to the most fertile alluvial soils. High and quality product is obtained from vineyards established on deep structured alluvial soils with good productivity level.

Depending on the rootstock, variety and soil structure, in some regions, the roots (pile root) of the vine can go as deep as 8 m with the effect of climatic conditions (rainfall). The capillary roots, which play a role in the nutrition of the vine, are more superficial and are more dense in the upper layers of the soil at a depth of 30-80 cm (Figure 5).

Figure 5: Annual precipitation of vines

The vines prefer well-drained soils. Heavy clay soils are not suitable for viticulture. Vines grown in humus-rich, very high N content and very moist soils are not resistant to diseases. The grains of table grapes produced in this type of soil are soft and unsuitable for transportation. Gravel and stones in the soil provide natural drainage and help warm the soil. Thus, the development of the vine increases (Çolakoğlu, 2010). Sandy soils are very suitable for viticulture due to their low water holding capacity and easy processing. In addition, since phylloxera cannot spread much in soils with a sand content of more than 60%, vines can develop more easily. Loamy soils, on the other hand, are soils containing 20-50% clay and 50-80% sand and are very suitable for quality and productive viticulture (Gücüyen, 2007). In calcareous soils containing 20% or more lime, lime-resistant rootstocks such as Fercal, 41 B can be used (Uzun, 2003) (Figure 6).

Figure 6: Lime resistant rootstocks

3. PLANT NUTRIENTS AND NUTRITIONAL DISORDERS

Plant nutrition is one of the most important elements in plant production. Fertilization accounts for 30% of the total growing cost. Due to the long-term yield of grapes, their nutritional requirements differ. Nutrients removed from the soil with the harvest must be renewed for the fertility of the soil and the sustainability of the productivity. Balanced fertilization is the only way to increase crop productivity sustainably (Lester, Jifon ve Stewart, 2007). However, the amount of fertilizer to be given to the vineyards depends on the climate, soil, rootstock, grape variety, irrigation, etc. depending on many factors such as In order to determine the fertilizer needs of the vineyards, soil and leaf analyzes must be done.

3.1. Nitrogen (N)

Nitrogen improves the green part of the plant. It is one of the most needed plant nutrients because it is the building block of proteins and it is found in chlorophyll molecules. Therefore, in N deficiency, leaves turn yellow and vegetative development stops (Atalay, 1982; Uzun, 2003). The main source of N in the soil; With the decay of organic materials (all kinds of plants, animals and microorganisms) in the soil, it creates N fertilizers that become useful to the plant and the N fertilizers we give. In addition, the N in the soil can be increased significantly with green manuring. N supplementation to the soil can be done with mineral fertilizers in different forms (ammonium, urea, nitrate, etc.). However, plants mostly take N in the form of nitrate (NO_3^-), and a small amount of ammonium (NH_4^+) and urea. For this reason, at least a few weeks must pass before the fertilizers given in the form of ammonium and urea can be converted to nitrate by the microorganisms in the soil and taken up by the vine.

Since the roots spread well around the vine in productive vineyards, N fertilizers can also be given between rows. After it is thrown into the soil, the manure should be covered with soil immediately by plowing in order to prevent N loss. N fertilizer should be applied to the vineyards in two periods, half before the buds awaken (usually in February) and the other half before flowering (in April-May). If N fertilizer is given in the late period, the lignification of the shoots is delayed and the vines may be damaged by early frosts in autumn (Atalay, 1982; Uzun, 2003).

It is possible to gain an average of 1.5-5.0 kg da^{-1} pure N per year with green fertilization made by planting legumes as intermediate agriculture in the vineyards. In order to prevent water competition with the vines, it is important to cultivate the green manure plant in winter when water is abundant. N can also be supplied to the soil with animal manure. However, in soils with salinity problems, animal manure given in excess can increase this problem and should be used carefully. It has been determined that 8.4 kg da^{-1} N is needed purely for shoot and fruit development in Sultani Seedless vineyards, and 3.0 kg of this amount is required for fruits. In another study, these amounts were determined as 6.8 and 2.8 kg da^{-1} for different varieties, respectively. In wine varieties, on the other hand, 1.43 or 1.70 kg N is needed for each ton of grapes (Uzun, 2003). The most typical symptom of nitrogen deficiency in vines, chlorosis in the leaves, appears as a homogeneous yellowing of the whole leaf. Yellowing first appears on older leaves. Shoots remain weak and short, flower and fruit set decreases, sugar rate and quality of fruit decrease. To this deficiency; Lack of organic matter in the soil, low soil temperature and extreme drought can be caused. An excess of nitrogen; It can also cause a deficiency of nutrients such as potassium, copper and boron.

Figure 7: Nitrogen deficient

3.2. Phosphorus (P)

Vines have a lower P requirement than N and K. Vines take up P in the soil in the form of H_2PO_4 and a small amount of HPO_4 ions. Although it is not possible to fully observe the symptoms of phosphorus deficiency, deficiency is more common, especially in soils with very low or very high pH. In phosphorus deficiency, a purple-red color occurs in the old leaves. The leaves are shed before flowering. Fruit is reduced on the vine and ripening is delayed. The root system weakens and winter hardiness decreases. Since the fruit set in the clusters is reduced, the yield is greatly reduced. Since the root development of the plant will slow down, there are great problems in the nutrition of the vine in general. This deficiency can also be caused by lack of organic matter in the soil, cold and humid conditions. In the use of excessive P fertilizer, Zn deficiency may occur in the vines, as the Zn uptake of the plant is adversely affected. It is sufficient to fertilize in a single band in the middle of the row in productive vineyards. However, P given in calcareous soils should be given in the form of a band, since it is easier to pass the P into the non-absorbable form. 1.0 kg da^{-1} P was detected in different organs of vines. 0.25 kg of P_2O_5 is needed for each ton of grape production (Atalay, 1982; Uzun, 2003).

Figure 8: Phosphorus deficient

3.3. Potassium (K)

Potassium increases the resistance of the vine against winter cold, it increases the color, smell, taste and juiciness of the fruits, namely the quality. In vineyards that are well fed with potassium, the water consumption of the plant is less and the water consumption of the vines decreases, since the stomata on the leaves of the plant have less time to remain open. It is also an element involved in carbohydrate metabolism, formation of sugars, protein synthesis, increased resistance of the plant to stress conditions, resistance to diseases and pests, and cell division. The period when the vines need the most is the ripening period of the grapes, that is, the summer months. Potassium deficiency firstly manifests itself in old leaves as discoloration towards the inside of the leaf margins and between the veins, then brown color and drying of these places. The green part of the leaf and the part that has changed color are separated from each other with sharp lines. Deficiency symptoms begin with fruit set and reach their most severe level at the color of the fruit. In the Sultani Seedless grape variety, the clusters begin to shrivel and dry from the tip. Since this situation is seen in the middle of summer and especially at harvest time, it can be confused with thirst symptoms. Other negative effects of potassium deficiency on fruit can be said to be the decrease in the amount of dry matter in the grain, the decrease in the must ratio and the increase in the amount of acid. When the grapes of the vines fed with sufficient K are evaluated as fresh, the shelf life is extended. In dryers, the grains are larger and homogeneous drying occurs (Atalay, 1982; Uzun, 2003).

Figure 9: Potassium deficient

3.4. Magnesium (Mg)

Magnesium is a component of chlorophyll and is needed to transport sugars from leaves to fruits. Mg, which is in the form of free ions and in the structure of chlorophyll, which gives the green color of the plant, is an important element due to its role in many enzymatic reactions within the plant. Deficiency symptoms are primarily seen on older leaves. It is in the form of yellowing between the veins and edges of the leaves. The deficiency symptoms, which start as discoloration between the veins from the petiole, continue with the formation of a reddish brown color between the veins in the later stages (Figure 10). There is a clear distinction between the green color of the leaf and the part caused by the deficiency. It is also a common symptom in sandy or acidic soils. In addition, the use of poorly burned farm manure accelerates magnesium deficiency. Deficiency symptoms occur frequently, especially because of its opposite relationship with K. In its deficiency, deterioration in fruit cluster and grain quality is observed (Atalay, 1982).

Figure 10: Magnesium deficient

3.5. Iron (Fe)

Iron is essential for the plant's photosynthesis processes. Reasons such as excess lime in the soil, bicarbonate (HCO_3) that enters the irrigation water and soil as a result of the decomposition of lime, increase in pH value, poor aeration of the soil, insufficient organic matter in the soil and high ground water can be counted as the cause of Fe deficiency. In addition, even if sufficient amount of Fe is taken by the plant, active Fe must be present in the plant. Before the establishment of viticulture, it is better to prefer varieties grafted on lime-resistant rootstocks rather than lime-sensitive rootstocks.

Symptoms of iron deficiency are seen primarily on young leaves. The most important deficiency symptom is chlorosis. All leaf veins remain green, although other parts turn from light green to yellow. It occurs primarily on young leaves. Young leaves are initially light green or yellow, veins are also green (Figure 11). However, as the deficiency increases, the veins turn yellow and the leaves appear completely yellow and even white. The vines weaken from year to year, and the yield decreases. Iron deficiency in vineyards is seen in extremely calcareous, alkaline and high ground water soils that hold iron. In order to eliminate the deficiency, Fe should be given to the plant from the soil or leaves. For this purpose, organic Fe compounds (chelates) or inorganic Fe compounds ($FeSO_4$) can be used (Uzun, 2003). Excessive Fe; It can also cause deficiencies of nutrients such as P and Mn.

Figure 11: Iron deficient

3.6. Zinc (Zn)

Zinc is an essential nutrient for the normal development of leaves, elongation of shoots and grain setting. The fact that the amount of Zn in the form that can be taken by the plant is generally insufficient in the soils of our country, the pH value is high due to excess lime in the soil, and the unnecessary use of P fertilizers in the soil causes Zn deficiency to occur in almost all plants and vineyards. The presence of excess Ca, Fe and Mn in soils and the presence of insufficient organic matter are also among the causes of Zn deficiency. Zinc element takes place in biochemical events in the plant. It plays a role in the synthesis of carbohydrates, proteins, fats and starches. Symptoms of zinc deficiency usually appear on young leaves. It causes asymmetric growth in leaves. In its deficiency, the leaves do not develop and the internodes are shortened, and an event called small-leaved (rosette) occurs. The internodes of the shoots are shortened towards the end of the shoot. Grain set decreases and pilling occurs. Excessive amount of Zn; It can also cause deficiencies of nutrients such as Mn, Fe and Cu. (Atalay, 1982; Uzun, 2003).

Figure 12: Zinc deficient

3.7. Manganese (Mn)

It is known that besides its role of accelerating the function of enzymes that play a role in growth, it also helps the formation of chlorophyll. Deficiency is mostly seen in vines grown in calcareous soils. In strongly alkaline soils with high soil pH, Mn deficiency symptoms are more common, and in very acid soils, toxicity is seen from excess Mn. Manganese deficiency symptoms usually appear on young leaves. It is in the form of yellowing between the veins and edges of the leaves. Discoloration is seen in the form of fragmented spots between the veins. Mn deficiency is more especially in the leaves in the shade (Atalay, 1982; Çolakoğlu, 2010). Although the symptoms of deficiency are similar to magnesium deficiency, the symptoms of magnesium deficiency occur primarily in the old leaves, and in manganese deficiency in the young leaves. An excessive amount of Mn; It can also cause a deficiency of nutrients such as Zn and Fe.

Figure 13: Manganese deficient

3.8. Boron (B)

Boron is necessary for seed formation and fruit set. If the amount of B in the soil is more than 1.0 mg kg^{-1}, excess B can be seen in the plant. The amount of B is high, especially in lands close to the sea, in regions with hot water sources, and in lands that are brought into agriculture after coal is extracted. The amount of B in irrigation water should not be high (more than 0.67 mg L^{-1}).

Boron deficiency is more common in soils with low pH and in regions with abundant rainfall. Boron deficiency causes yellowing of young leaves, asymmetric leaf growth in grapes. Young leaves shrivel and curl, often thickening and taking on a dark blue-green color. It has a positive effect on cell division (proliferation) at growth points, on the development of meristem tissues in the crown buds and on auxin production in the cells in this part. In its deficiency, as in all fruit trees, there is a decrease in fruit set because it affects the formation of pollen. In boron deficiency, the internodes in the shoot narrow, the leaves at the ends of the shoots start from the leaf margin and become discolored and dry towards the inner parts, and the leaves become smaller. In excessive B deficiency, the dried leaves are shed. In the shoots, short but more numerous armchair shoots occur. Defoliated shoots take the form of a bush. Shrub and defoliation starts from the tip of the shoot and progresses downwards. (Atalay, 1982; Çolakoğlu, 2010).

Figure 14: Boron deficient

4. FERTILIZATION

Plants need absolute nutrients in order to develop and produce products. In nutrient deficiencies or excesses, many visible symptoms occur in organs such as roots, shoots, leaves and fruits. It is known that vineyards with large leaves with prominent veins reflect nutritional problems well. Early and correct diagnosis will increase the success rate of fertilization applications to be made in the treatment of diseases.

The amount of fertilizer to be given to the vineyards depends on the climate, soil, rootstock, grape variety, irrigation, etc. depending on many factors such as In order to determine the fertilizer needs of the vines, soil and leaf analyzes must be done.

4.1. Fertilization of Vineyards According to Their Development Periods;

4.1.1. Plant Fertilization

Before establishing the vineyard, soil samples should be taken from 0-30 cm and 30-60 cm and analyzed, taking into account the root depth, in order to determine the rootstock to be used and to determine the fertilization program. The point to be considered in plant fertilization is that it may be necessary to use P fertilizers according to the lime and pH value of the soil, and K and Mg fertilizers according to the % clay and available K and Mg amount of the soil. If the pH value of the soil is above the desired level, it is important to reduce it with S, and if the pH value is too low, it is important to increase it with liming (dolomite) (Atalay, 1982; Çolakoğlu, 2010). In the fertilization of young vineyards, it is necessary to give weight to fertilizers with P and K, which have the effect of improving the root system of the vine. Fertilization should be done in three different development periods of the youth age (first application 2-3 weeks before the start of bud growth, the second after flowering and when the grains are in the size of a scatter), as in the full yield period in the vineyard.

Figure 15: Terrain view of young vineyards

In suspended viticulture, fertilizers are applied 60-70 cm away from the main body by tape method and mixed into the soil 20-25 cm deep. With base fertilization, all of the P and K needed by the plant and a part of N are given. In top fertilization, K fertilizer should be applied with the remaining part of N. From the planting year, the vines should be fertilized for three years until they reach the full yield age (Kovancı et al., 1984).

4.1.2. Yield Age Fertilization

In the fertilizer recommendation to be made in the vineyards at full yield age, the purpose of growing the vine and irrigation type should be taken into consideration. Drip-appropriate fertilizer advice should be given monthly. Monthly recommended fertilizer amounts should be applied by dividing the number of irrigation to be performed during that month. As fertilization time, as in young vineyards, base fertilization should be done before bud growth, and top fertilization should be done when the grains are scattering or in the size of a grove. The nutrient requirement of the vine plant varies depending on the development periods. The plants water consumption and nutrient intake are in parallel. Plants need the most nutrients between bud activity (eye swelling) and flowering, flowering and moles because they take nutrients in dissolved form in water. These periods should be considered in fertilization (Atalay, 1982).

Figure 16: Yield age fertilization

5. CONCLUSION

Increasing the agricultural production in parallel with the increase in the world population causes the use of more inputs. Considering the population growth and the need for agricultural production, the most intense input consumption will be in developing countries. In this sense, countries with the potential to develop agriculture, such as Turkey, are candidate countries to meet their future food needs. Some of the negativities caused by the intensive use of chemical fertilizers, which are at the beginning of the inputs used in agriculture, are worrying. Damages resulting from excessive and unbalanced fertilization highlight the increase in controlled fertilizer consumption. It is possible to increase productivity without using unconscious and intense inputs and without destroying nature and the environment. Nutrient deficiencies in the soil prevent healthy and quality production increase. More intensive research should be done on this subject, fertilizer consumers should be made aware and encouraged, and the use of micro element fertilizers should be expanded as necessary.

In order to get the most effective benefit from fertilizers, it is necessary to use the right fertilizer in the right plant, in the right place, at the right time, taking into account plant demands, climate, soil structure, irrigation status and vegetation period. In order to get more efficiency with a balanced fertilization and to obtain quality and healthy products; It is necessary to determine the fertilizer need correctly and to keep records of information such as the type and amount of fertilizer, application method and frequency. The purchasing power of the producer is also very important in fertilizer consumption. Fertilizer-product-price relationship is an important factor to be considered in fertilizer consumption. Improvement and improvement of new plant varieties with high fertilizer use efficiency in order to benefit from fertilizers more effectively will significantly reduce excessive fertilizer use and input costs.

Due to the low organic matter content and high pH of the soils of Turkey, the use of soil conditioners and organic fertilizers should be expanded in order to increase the availability of plant nutrients in the soil that cannot be taken up by the plant. Considering the positive effects of organic materials on soil fertility, studies on organo-mineral fertilizers should be given due importance. Nutrient deficiencies in the soil prevent healthy and quality production increase. More intensive research should be done on this subject, fertilizer consumers should be made aware and encouraged, and the use of micro element fertilizers should be expanded as necessary. In our country, where a wide variety of cultivated plants are produced and has different soil properties, paying attention to the production and use of plant-specific fertilizers will contribute significantly to the use of effective fertilizers. Increasing the efficiency of fertilizer use will also play an important role in meeting the plant nutrient requirement by reducing the risks. Measures to increase fertilizer efficiency are important in terms of product, environmental and economic aspects.

By applying the required type and amount of fertilizer; As a result of excessive use of fertilizers, problems such as quality deterioration in yield, loss of productivity of agricultural lands, negative effects on the environment, waste of resources, etc. will be prevented, as well as the decrease in yield and quality encountered as a result of using less than necessary. The most effective way of using the right fertilizer in accordance with the principles of sustainable agriculture is applications based on soil and plant analysis. One of the most important problems is that the farmers do not have the right information on the use and consumption of fertilizers. Particular attention should be paid to ensuring the flow of information between researchers and farmers conducting scientific studies, and their regional experiences should be taken into account by ensuring effective participation of farmers. Training activities should be given to research, extension and fertilizer dealers and farmers in order to ensure the use of the right type and amount of fertilizer. Conscious producers who do not harm the environment and human health, protect natural resources, ensure food safety, and make agricultural production that can be traced at all stages should be the most important target.

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CHAPTER 10

WINE QUALITY OF THE UNIQUE HIGH ALTITUDE GRAPE CULTIVAR "ERCİŞ" FROM VAN-ERCİŞ VINEYARDS

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INTRODUCTION

Van province (Figure 1) is known as one of the ancient grape growing area, in the World (Keskin, 2011; Keskin et al. 2018). Fossills that are obtained around Hoşap Castle (Figure 2) show us, viticulture back to the iron age.

Figure 1. Van province

Figure 2. Hoşap castle (Photo: Murat Cacim)

As in ancient time, today "Erciş" grape cultivar (Figure 3) is a unique variety in Van province. Archeological resources explained that this unique variety had a great importance in the life of Urartians especially in 600-900 BC.

Berry Characteristics
Color: Black with grey bloom
Form: Round
Size: Medium 2.0-2.5 g
Seed: 2-3
Flavor: Distinct varietal flavor
Cluster Characteristics
Forum: Winged conical
Size: Medium, 250 g
Compactness: Compact
Cultural Characteristics
Ripenning: Midseason
Pruning: Spur

Figure 3. "Erciş" grape cultivar (Photo: Nurhan Keskin)

Wine quality is a term that is so hard to explain because of human based subjetcive decisions and that is why science try to explain “quality” objectively all the time. Product based researches are mostly made by chemical and sensory analyses and the researches that aim to determine the fingerprint of the final products are getting more important especially for wines nowadays.

Altough Van region has very compelling ecological factors such as temperature, altitude, vegetation period for viticulture, "Erciş" cv. has been adapted these situations for centuries at its original terroir. Since most of the vineyards (Figure 4) are neglected by social and economic problems of the region, "Erciş" cv. has been in extinction danger. Therefore, studies on "Erciş" grape cultivar is very important to save and improve this unique high altitude variety in Van province.

Figure 4. "Erciş" cv. Vineyards (Photo: Nurhan Keskin)

Molecular characterisation (Gazioğlu-Şensoy, 2008) and ampelographic description (Kelen, 1991) and clone selection studies (Uyak, 2002) have been brought to "Erciş" cv. in a a recognisible level. Also chemical quality content (Keskin and Kunter, 2007; Gazioğlu-Şensoy, 2010; Gazioğlu-Şensoy, 2012; Keskin et al., 2012; Gazioğlu-Şensoy, 2015; Keskin, 2017; Baytin and Keskin 2018; Keskin, 2018) response to abiotic stress conditions (Keskin and Kunter 2007; Ersayar, 2017; Gökçen, 2022) and suitability for storage are also among the topics studied (Keskin et al., 2014). The resveratrol concent of the cultivar is close to popular grape varieties such as "Cabernet Sauvignon", "Öküzgözü" and "Kalecik Karası" (Keskin, 2007). It also has a high potential in terms of callus formation for resveratrol producing rate *in vitro* studies (Keskin and Kunter 2010).

This study aims to provide new insights into understanding some phytochemical characteristics of the wines produced from "Erciş" grape cv. This research is a first attempt for evaluation of the cultivar as a wine grape with phenolic characteristics.

2. MATERIAL AND METHODS

Preparation of Wine Samples

In this study, 19.420 tones of "Erciş" cv. grapes are harvested at original terroir-Van-Erciş province in 2021 vintage and varietal young red wines are produced at Akberg Winery.

Brix Determination of Must

Brix was determined by a digital refractometer (Atago Rx-7000 α, Tokyo, Japan).

Density Analyses

Density was determined by a mustimeter (Dujardin Salleron, France). Sugar content of the must was also calculted by the mustimetre (hydrometer).

Determination of Titratable Acidity

Titratable acidity was determined by titration of 5 mL wine samples with 0.067 N NaOH, till the pH 8.1 (IFU, 1968). The spent volume of 0.067 N NaOH while titration, gave directly total acidity as tartaric acid (g/L).

pH Analyses

pH was determined by a pH-meter at 20ºC (WTW Inolab Level 1, Weilheim, Germany).

Alcohol Analyses

Alcohol content (v/v) was determined by an electronic ebulliometer (Dujardin Salleron, France).

Chemical Composition Analyses

Chemical composition of the wine (sugar content, volatile acidity, organic acids, YAN, Oe, glycerol) was determined by Anton-Paar brand laboratory instrument.

3. RESULTS AND DISCUSSION

Evaluation of wine quality have been investigated both in must and wine (Table 1).

Table. 1. Basic analytical wine analysis in "Erciş" wine

	Must	Wine
Ethanol (v/v)	--	10.16
Glucose (g/L)	--	1.1
Fructose (g/L)	--	1.9
°Bx	19	--
pH	3.39	3.37
Titratable acidty (pH=7.0)	--	14.32
Titratable acidty (pH=8.2)	--	14. 21
Volatile acidity (g/L)	--	0.02
Malic acid (g/L)	--	1.25
Lactic acid (g/L)	--	1.07
Tartaric acid (g/L)	--	2.89
Density (g/L)	1077	997
Extract (g/L)	--	32.6
Must weight (Oe)	77.12	-1.06
Glycerol (g/L)	--	6.0
YAN (mg/L) (N)	--	344
Reducing sugars (g/L)	--	0.0

Results showed that inadequate maturity level has engendered low quality. Erciş grapes had to be harvested at 19 °Bx level and this is a disadvantage to keep grapes on vine up to high brix level because of cold climate conditions. In the previous studies maturity level of "Erciş" cv. was determined as 13.67 and 12.33 °Bx. (Yaşar 2005), 12.90 and 16.20 °Bx (Çelik, 2016), 16.50 and 17 °Bx (Keskin, 2017). Basic analytic assay of samples showed us "Erciş" cv. wine has satisfactory data for vinification.

Keskin et al. (2012), were carried out must and wine analyses in 2011 vintage of "Erciş" cv. In this study °Bx, density, titrable acidity (as tartaric acid), pH, alcohol and sugar content were determined as 19.0 °Bx, 1077 g/L, 9.32 g/L, 3.28, 10.4° (v/v) and 16.10 % (w/w). Our results are in agreement with the this preliminary results.

4. CONCLUSION

These results provided that "Erciş" cv. can be considered as a valuable cultivar for quality red wine making. It is worth to study beginning from vineyard in order to obtain high quality yield to the end of wine processing. In the light of the data obtained from this study, it is expected that supporting viticulture based on Erciş cv. will provide positive economic and social results for the Van region.

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CHAPTER 11

FLORAL TRAITS, OVULE, AND EMBRYO SAC DEVELOPMENT BY VISUAL REPRESENTATION IN *VITIS VINIFERA* L. GRAPEVINES

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INTRODUCTION

In grapevines, there is the possibility of utilizing different fruit set mechanisms for breeding new grape variants. The development of an ovary into a grape berry depends on the anatomical structure of ovules and pollination characteristics. With respect to this matter, the anatomical characterization of ovule development in the grapevine varieties is relevant for both basic and applied research, and it is important for breeding approaches.

The grapevines have a mixed bud type that contains the structures that will yield shoots, leaves, and flowers in the developmental cycle of a year. In other words, shoots carry leaves, flowers, and fruits together in every vegetation year. Flowering and fruit set is required for two growing seasons (Hellman 2003, Monteiro et al 2021, Keller 2020).

In the first year, buds containing inflorescence primordia keep developing in the leaf axil at the area of the connection point of the leaf on the shoot. Buds will contain flower cluster primordia before the end of the growing season, and then buds will go into dormancy until bud-burst in the next growing season In a compound bud structure, fruitfulness is mainly regarded as the number of flower clusters in the primary bud. The primordia of all vegetative and reproductive organs occur inside the bud. Flowering and fruit set occurs in the following season of the same buds. After bud-burst, depending on the genotype, inflorescence primordia become visible in soon. (Figure 1).

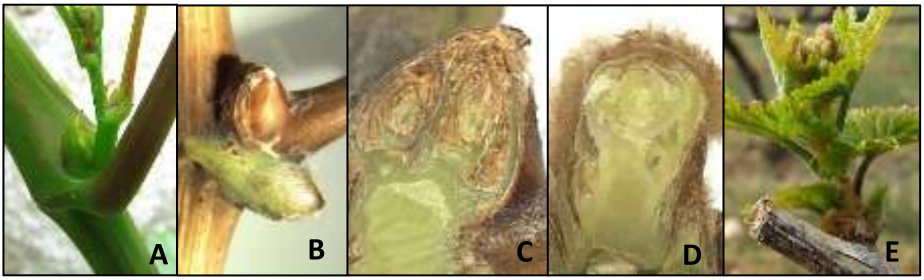

Figure 1: The first development year of compound bud in the base of the petiole; (A); Appearance of bud in the dormant season (B); Primary, secondary, and tertiary buds inside of the dormant bud (C); The vegetative and reproductive primordia in the primary bud (D); The emergence of inflorescence at the bud-burst (E)

While the shoot development proceeds, inflorescences continue differentiation to form flower clusters and the flowers are formed. It is well known that the flowering process is strongly dependent on temperature (May 2004, Keller 2020). About 20-35 days after bud burst flowers are ready for anthesis (Figure 2).

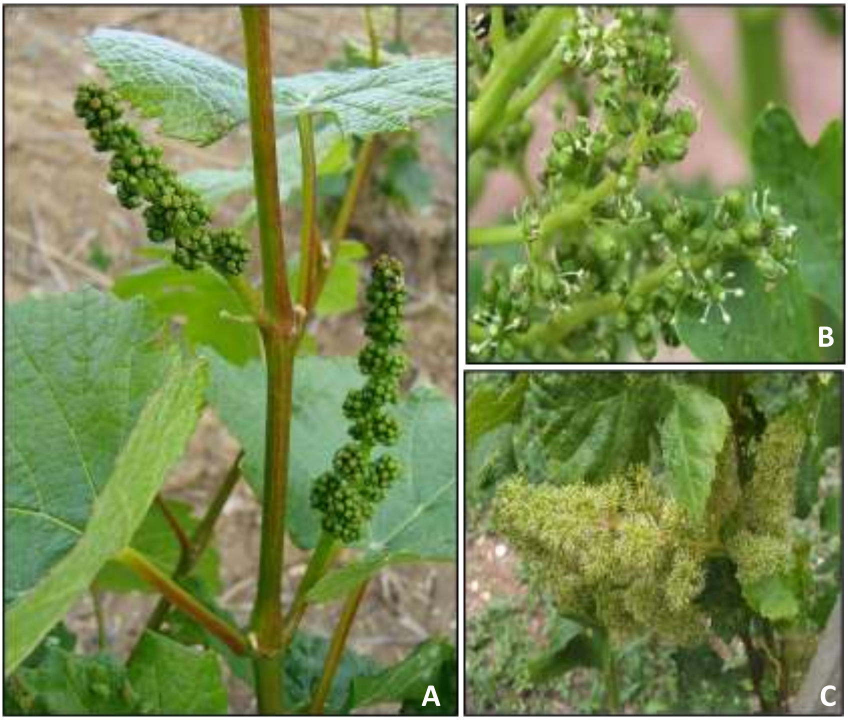

Figure 2: Inflorescence turn into flower cluster. Inflorescence branching (A); Early flowering of flower cluster (B); Anthesis, full flowering (C)

The morphological and anatomical structures of pistil and pollen have determinative effects on pollination, embryogenesis, and fruit set in grape cultivars. Therefore, studies dealing with the hybridization of grapevines and, understanding the fruit set in individuals are interested in the structure of ovule and embryo development.

The presented chapter comprises detailed microscopic descriptions of ovule development concerning fruit-set characters in *Vitis vinifera* L. based on the data of our previous studies and the relevant literature. In this context, ovule and embryo development of *Vitis vinifera* L. was exhibited by using original macro-microphotographs, that are obtained by Marasali (1992, Marasalı 2002) and unpublished studies of B. Kunter (Marasalı).

1. Flower types in *Vitis vinifera* L. Grapevines

Vitis vinifera L. species is characterized by having two types of flowers. Fertilization takes place either self or cross-pollination considering the flower type.

Varieties of this species mostly have perfect hermaphrodite flowers with normal stamens and much viable pollen. The perfect hermaphroditic flowers have mainly five, sometimes six, or rarely seven functional stamens. The flower carries one fertile pistil developing into a berry. It is the flower type with the highest incidence rate in *Vitis vinifera* L. grapevines.

The other flower type is termed functionally pistillate or functionally female flower. The number of reproductive organs of the flowers is the same as perfect hermaphrodite flowers. However, the stamens in this type of flower are morphologically short and recurved that indicating non-viable pollen. In contrast to abnormalities of stamens, the pistil is productive, generally. For this reason, cross-pollination is essential to obtain the fruit set. In *Vitis vinifera* L. germplasm, a limited number of them have functionally female flowers. Although this occurrence is said to be rare in grapevines, there are some well-known grape cultivars such as Çavuş, Tahannebi, Karagevrek, and Hönüsü for fresh consumption. The structurally pistillate type genotypes can use as mother plants in hybridization studies without the emasculation process. The advantage of the genotypes with pistillate flower structures is that they can be used as the mother plants in hybridization without the need for emasculation.

In both flower types, the main parts are the calyx, corolla, stamen, and pistil. On a grapevine flower, calyx consisting a ring of five fused sepals. The sepals are in vestigial form. Reproductive organs are covered by corolla (or calyptra, flower cap). Corolla consists of five combined petals. Petals are green and corolla likes a cap. Therefore many times it is called flower cap. The general feature of *Vitis* grapevines is that the petals of the corolla open upward from the base of the flower where they are connected. During the bloom, corolla falls, stamen and pistil appear. In this stage, the morphological differences between the two flower types become visible. Regarding the morphological characteristics of stamens and pistils, a wide range of differences has been seen among the individuals (May 2004, unpublished results B. Kunter (Marasalı)).

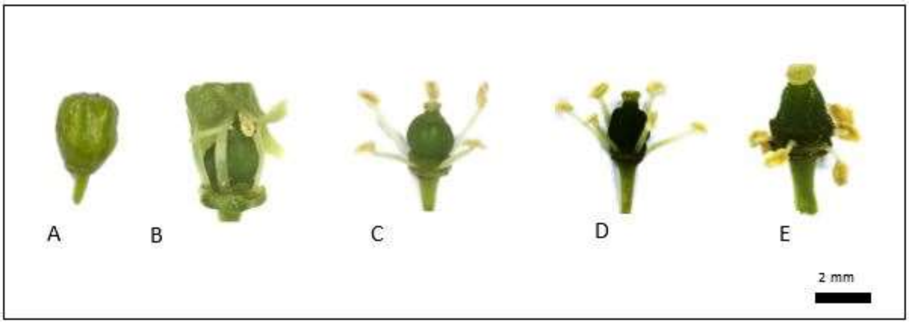

Figure 3: *Vitis vinifera* L. grapevine flowers. Corolla covers floral organs (A); Corolla detaching from flower base (B); Perfect hermaphrodite flower with five stamens (C); Perfect hermaphrodite flower which has six anthers (D); Functionally pistillate flower which has recurved stamens (E)

2. Ovule structure and its development after fertilization

In *vinifera* grape varieties, pollination and fertilization are essential for transforming the ovary into a berry. The ovule and embryo sac development influence the change of the ovary becoming a berry and its subsequent development In grapevine, three types of ovule-embryo sac structure concerning seed development and fruit set may be described refer to relevant literature: (1). Perfect ovules with functional embryo sac- Seeded grapes; (2). Imperfect ovules with aborted embryos- Stenospermic seedless grapes (Stenospermocarpy); (3). Imperfect ovules with aborted embryo sac- Parthenocarpic seedless grapes

In addition to the given abortions, in the class of seeded grapes, some cultivars show non-viable seed formation termed “empty-seededness”.As a result of this development, seed germination percentage is very low. Such seeds are morphologically normal but non-viable due to embryo and endosperm abortion and limit the success of hybridization programs. It can be characterized as the problem of seed development rather than fruit set, since the qualityof berries is not changed at all. Here, the anatomical structure of empty seediness, which has been explained in the Çavuş variety, has not been discussed since have focused on the abnormalities in relation to fruit set.

2.1. From perfect ovules to fertile seed and normal seeded fruit set

The development of the fertilized ovule turns into a berry with fully developed seeds is described as the normal seeded fruit set. The ovary contains two ovules in each carpel in grapevine flowers, either perfect hermaphrodite or functionally pistillate flowers. It means that a grape berry contains four seeds if all ovules are well-fertilized (Figure 4).

Figure 4: Longitudinal section of grape flower before flower cap fall. Ovary two ovules in one carpel

In the early stage of the development of the ovule, ovular primordia become anatropous generally 14 days before anthesis. The initial differentiation of the embryo sac begins the distinction of the *megaspore mother cell*. Megaspor mother cell is generally determined 8-11 days before anthesis. The cell undergoes meiosis resulting in the production of four megaspores. Out of four haploid cells, only one survives (Marasalı 2002). It is the chalazal megaspore that forms eight nucleates by repeated mitotic nuclear divisions. Meanwhile, the nucellar tissue develops into several layers and forms a narrow zone that is micropyle. Depending on the division of the megaspore and the number of nuclei in the embryo sac, female gametophyte formation in *Vitis vinifera* L. grapevines is classified as *monosporic 8-nucleate type* (Haig 2020).

The three nuclei placed at the chalazal end are antipodal cells. In grapevines, it is thought that the antipodal cells do not play a well-known role in berry and seed development. In the blooming period, two nuclei from each pole migrate to the center of the embryo sac. These are called polar nuclei. Polar nuclei are generally observed just before anthesis. When anthesis begins, the two nuclei fuse to form the *secondary nucleus*. After fusion, the secondary nucleus in the embryo sac migrates toward the micropylar end. An egg cell and two synergids lie at the micropylar end of the embryo sac. The egg cell is not to be said neither a consistently larger size nor a more conspicuous nucleus than the synergids. It is sometimes slightly larger than the synergids. After the pollen tubes reach the micropyle, they pass through into the embryo sac. The fusion of the egg apparatus and sperm and the fusion of the second sperm and secondary nuclei realize. This is double fertilization. In our former studies, it was observed that double fertilization took place 8-11 days after the beginning of flowering.

After double fertilization takes place, for at least 2-4 weeks the zygote stays in a rest period. But the endosperm nuclei continue dividing. The first mitosis of the zygote is the first step in embryonic development. Pro-embryo consists of a terminal cell and basal cell. In our previous studies, two-celled embryos were observed 9-21 days after fertilization in Karasakız and Çavuş cvs. (Marasalı 1992). Within one month after anthesis, the embryo becomes heard-shape and, at the berry ripening period reaches its full size. In the development of normal seed, there is a rapid development of the outer integument of the sclerenchyma. Ovules that turn into normal viable seeds contain a well-developed embryo that size about 2.0-3.0 mm. Microphotographs have presented the above-explained female gametophyte development in Figure 5.

In grapes, being able to develop after fertilization into one or more viable hard seeds is the rule for the seeded fruit set. Many of the cultivated *Vitis vinifera* L. grape varieties bear seeded fruit and the seeds are noticeable when the berries are eaten. But in many seeded grape varieties, seed formation is relatively less than the ovule number. Studies show that when the flowers are about to open four ovules are presented in the ovules. After fertilization, some ovules can not develop. It is considered that ovule abortions in seeded genotypes are hereditary. That is why seeded varieties show differences in seed numbers and sizes.

2.2. Imperfect ovules

Imperfect ovules result in the most valuable berry developments in *Vitis vinifera* L. grapevines. But such developments are together with seedlessness or immature and low germinative capacity seeds. The seedless fruits of grapes are two main types: stenospermocarpy and parthenocarpy. They arise as the result of different abnormalities concerning immature ovule developments.

In stenospermocarpic grapes such as Sultani, flowers are the perfect hermaphrodite type and pollen is highly viable. Female gametophyte development is identical to a fertile embryo sac up to the beginning of embryo development. The egg, two synergids, and fusion of polar nucleus are differentiated in the embryo sac at the anthesis. On the other hand, abnormalities exist in integument tissue at the time of anthesis. At this early stage of the ovule development, the tips of inner integuments elongated. In some ovules, tips are curved together in the same direction. Integument tissue can not become hardened in progressing developments. For the typical stenospermic seed degenerations appear soon after fertilization in the feeble formation of sclerenchyma, and seed development is incompetent. Depending on the genotype, a limited number of cell divisions takes place in the zygote and endosperm nuclei (Figure 6). Concerning the degeneration stage of embryonic cells, seed trace is different in size (Değirmenci ve Marasalı 2002).

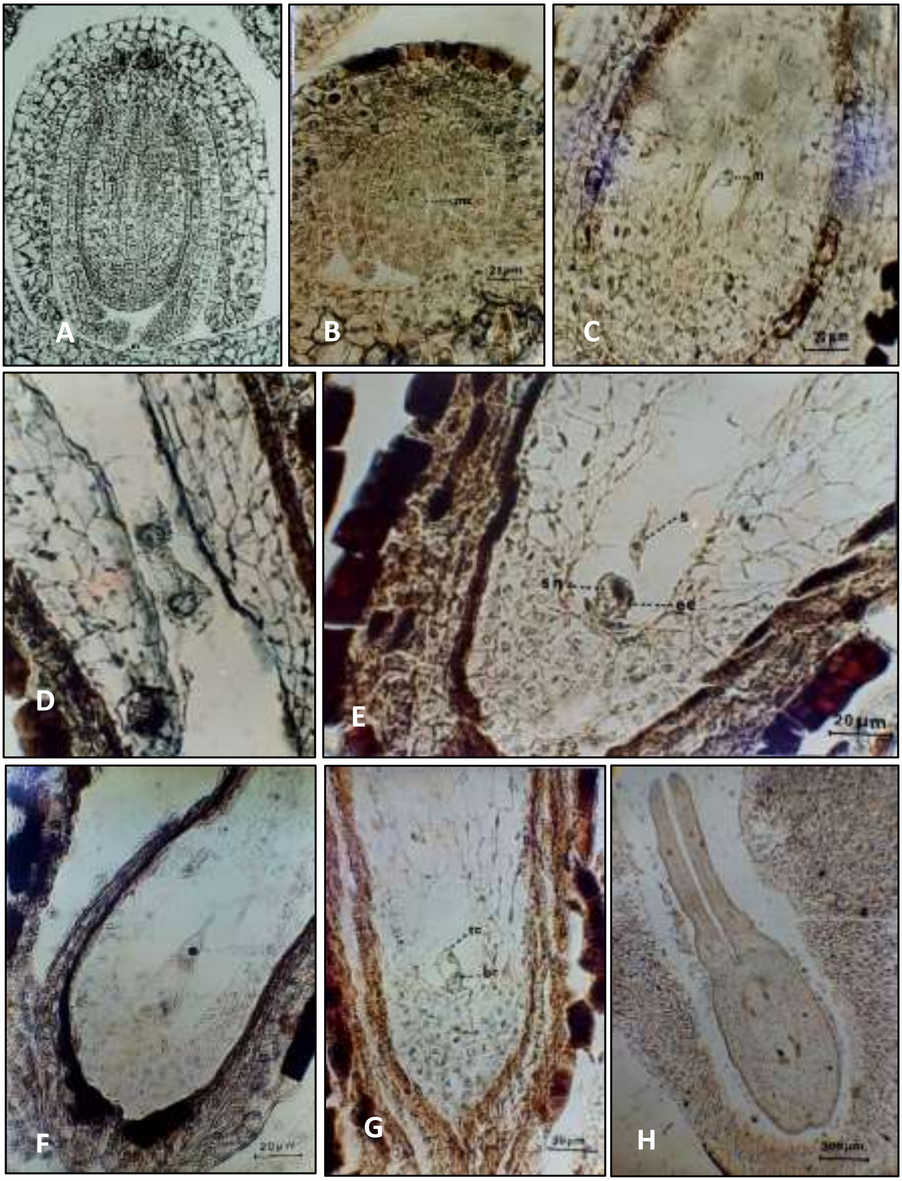

Figure 5: Embryo sac development and double fertilization in the ovule of grapevine. Early stages of ovule development. Anatropous ovule having nucellus tissue (A); Differentiated megaspore mother cell (mc) in nucellus tissue (B); Megaspore nuclei (C); Two polar nucleus (D); Secondary nuclei (s), egg cell (ec) and synergid (sn) in the sac (E); Double fertilization (F); First dividing of zygote for embryo development, terminal and basal cell (tc,bc) (G); Embryo (H)

In the previous studies, it is concluded that seed trace size in stenospermocarpic cultivars is related to the relative time of embryo/endosperm breakdown. Cultivars known for very small seed traces typically have embryo/endosperm abortion at a very early developmental stage (Ledbetter and Ramming 1989).

Finally, integument and embryo sac abortions are effective on the shape, size, and character of the seed formation. Stenospermocarpic fruit set is of great importance in viticulture since including the anatomical structure for breeding new seedless cultivars. Seedless grapes such as Sultani Çekirdeksiz, Yuvarlak Çekirdeksiz, Ergin Çekirdeksizi, Perlette, Beauty Seedless, Flame Seedless, Crimson Seedless, Superior Seedless, etc. are economically valuable via stenospermocarpic fruit set. Therefore the inheritance of stenospermic ovules is paramount in advanced selective breeding.

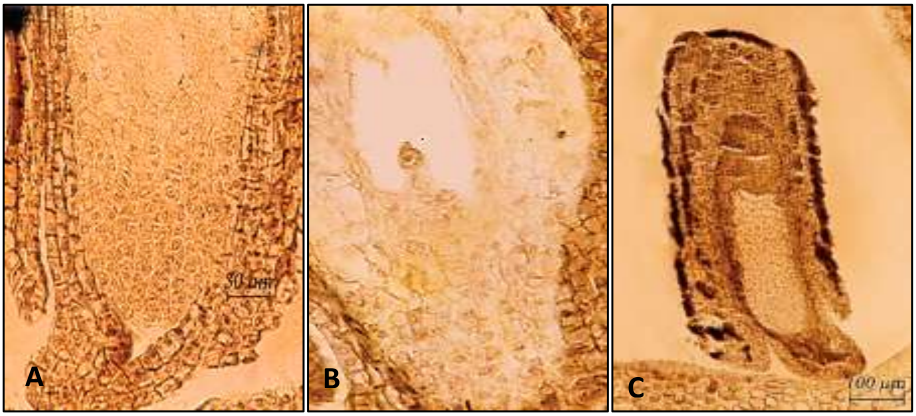

Figure 6: Longitudinal sections of stenospermocarpic ovules. Elongated inner integument (A); The zygote after fertilization (B); Stenospermic seed trace (C)

Imperfect ovule development also results in parthenocarpic fruit set in grapevines. Parthenocarpic grapes have the extreme type of defective ovules. Ovules have only the outer integument. There is no embryo sac. found. Nucellus grows extremely and exceeds beyond the integument., defective ovules are fully misshaped and can not differentiate to form an embryo sac (Figure 7). As a result, berries are entirely seedless and small-sized. In rare cases, reporting a limited number of seeded berries in certain clusters has been attributed to mutation from the seedless to the seeded type (Stout 1936).

Parthenocarpy is said to be cultivar specific. Corinth grapes are the well-known varieties that have imperfect ovules without embryos and small-sized seedless fruits of parthenocarpy.

Figure 7: Entirely misshaped ovule. One integument; nucellus tissue bends outward between the integument tissue

CONCLUSION

Hundreds of flowers form a grape cluster, and these flowers may be in different stages of reproductive development. That is why pollination and following developments of berry formation could be considered complicated in grapes. The fruit set is not only the result of the fertilization process but also the development of the ovule and embryo. Studies on the ovule and embryo development are important to understand the type of fruit set in a variety and, the degree of seed structures in hybrids for breeding new grape varieties. Considering the dynamic improvement of table grape breeding programs, cytological studies on ovule development will always be in focus. In this chapter, it has been presented developments of ovules in *Vitis vinifera* L. in series stages by microphotographs.

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