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Modern Fruit Industry

Edited by Ibrahim Kahramanoglu,
Nesibe Ebru Kafkas, Ayzin Küden
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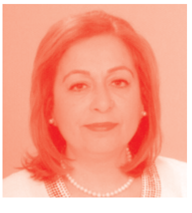
Meet the editors

Ibrahim Kahramanoglu holds a PhD in Postharvest Biology and Technology and works as a lecturer at the European University of Lefke (Northern Cyprus). He received a BSc in Horticulture and an MSc in Plant Protection. He previously worked in the alternative crops projects of USAID in Cyprus and was the managing director of Alnar Narcilik Ltd. for 8 years. He has practical experience in fruit production (especially pomegranate, olive, and citrus); control of pests, diseases and weeds; and good agricultural practices. However, his main studies and research interests are postharvest handling of fruits; use of eco-friendly bio-materials for postharvest storage; novel technologies for handling and storage; sustainable horticulture; environmental resource protection; food safety; and value adding to horticultural crops.

Ebru Yaşa Kafkas graduated from the University of Çukurova, Faculty of Agriculture, Department of Horticulture, Turkey and has worked in this department since 1996. She is now a Professor. She has experience in fruit breeding and fruit metabolomics and chromatography. Additionally, she is the author of more than 100 scientific papers (hindex: 16; total citations: 1024), and has undertaken editorial positions in scientific journals. She has participated in national and international EU projects as a Turkish coordinator. She is also a member of the International Society of Horticultural Science.



Ayzin B. Kuden, is a professor of agriculture in the Horticultural Department, University of Cukurova, Adana, Turkey. She was Vice Dean from 1997 to 1999 and Dean of the Faculty of Agriculture from 2005 to 2012. Her specializations are: physiology and breeding of stone fruits, almonds, and figs; selection, identification, and characterization of temperate and subtropical fruit germplasm. She is also interested in the use of molecular markers in breeding and dormancy breaking studies on temperate fruits. She has attended several courses in different countries for training. She has been working as the chairperson of ISHS "Temperate Fruits in the Tropics and Subtropics" working group since 1996. She has had professional memberships of ISHS and TSHS. She has contributed to the publications of four *Acta Horticulturae* editions as editor and one chapter and co-editor in two IntechOpen books and several national books. She has organized six international symposiums and two workshops, and several national meetings and a national horticultural congress. She has published around 185 peer-reviewed papers. Six PhD students of hers were appointed as academic staff in different universities in Turkey and twelve master students were appointed at good jobs. She is currently supervising six PhD and 7 master students.

Songül Çömlekçioğl is a doctor in tissue culture and growing and breeding stone fruits at Çukurova University, Turkey. She received her MSc thesis on Meristem Culture on Fig (*Ficus carica*). She has 20 years' experience in tissue culture and she completed her PhD thesis on peach and nectarine breeding. She is still working in several national and international selection breeding projects. She has published 35 scientific papers. She is currently working at Çukurova University, Agriculture Faculty, Horticulture Department as a technical staff member.

Contents

Preface

Huge efforts have been made in the new advances in the agricultural industry, i.e. chemical fertilizers and synthetic pesticides, from the 1940s to 1970s; and these resulted in a drastic increase in the productivity of global agriculture, called the "green revolution". Afterwards, along with the benefits gained, there have been several criticisms, especially about food security and environmental impacts. Furthermore, the term "sustainability" gained more importance. Sustainability is defined as "an economic activity that meets the needs of the present generation without compromising the ability of future generations to meet their needs" by World Wildlife Fund (WWF) and is based on three components: economic growth, social progress, and environmental protection. Nowadays, the demand for food is increasing due to the increase in human population but the quantity and quality of agricultural production is declining due to human-induced environmental problems, i.e. climate change and water scarcity. Moreover, our modern fruit industry needs to improve quality and quantity of fruit production using specialized production systems, mechanization, biotechnology, innovation, and science; while also protecting ecosystems by reducing energy inputs and environmental impacts. This book intends to provide the reader with a comprehensive overview of the current status of the modern fruit industry from production to consumption.

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Section 1

Advances in Postharvest Technology

Chapter 1

Alternative Eco-Friendly Methods in the Control of Post-Harvest Decay of Tropical and Subtropical Fruits

Ramsés González-Estrada, Francisco Blancas-Benítez, Rita M. Velázquez-Estrada, Beatriz Montaño-Leyva, Anelsy Ramos-Guerrero, Lizet Aguirre-Güitrón, Cristina Moreno-Hernández, Leonardo Coronado-Partida, Juan A. Herrera-González, Carlos A. Rodríguez-Guzmán, José A. Del Ángel-Cruz, Edson Rayón-Díaz, Hector J. Cortés-Rivera, Miguel A. Santoyo-González and Porfirio Gutierrez-Martinez

Abstract

The effectiveness on several fruits by the application of alternative methods against fungi is summarized in the present chapter. Several investigations have reported the efficacy of these technologies for controlling fungal infections. Currently, high post-harvest loses have been reported due to several factors such as inefficient management, lack of training for farmers, and problems with appropriate conditions for storage of fruits and vegetables. Even now, in many countries, post-harvest disease control is led by the application of chemical fungicides. However, in this time, awareness about fungi resistance, environmental, and health issues has led to the research of eco-friendly and effective alternatives for disease management. The pathogen establishment on fruits can be affected by the application of GRAS compounds like chitosan, essential oils, salts, among others; besides, their efficacy can be enhanced by their combination with other technologies like ultrasound. Thus, the applications of these alternatives are suitable approaches for post-harvest management of fruits.

Keywords: alternative systems, antifungal activity, postharvest fungi, tropical and subtropical fruits

1. Introduction

The consumption of fresh fruits and vegetables is essential for a healthy diet [1]. However, their production can be affected by microbial pathogens (mostly fungus) during the production chain [2]. In order to reduce the presence of pathogens, several post-harvest technologies have been applied [2]. One of them is the application of chemical fungicides; however, this practice is not accepted due to environmental and health issues [3]. Other alternatives are the use of eco-friendly substances such as generally recognized as safe (GRAS) compounds and emergent technologies like ultrasound and fogging. Currently, consumers demand fresh products free of chemical residues; therefore, it is necessary to develop technologies eco-friendly, effective to protect against pathogens infection, and that these technologies can maintain the fruit quality. Alternative systems such as edible coatings, essential oils, salts, natural compounds (plant extracts), among others to chemical use are suitable approaches for post-harvest disease management. These alternatives can be applied in combination with other control systems like emerging technologies (ultrasound) in order to improve their efficacy. The aim of this chapter is to make a compilation of several studies conducted on fruits for controlling important pathogens in several crops.

2. Fruit industry: importance in the world

Fruits and vegetables are essential sources for the micronutrients needed for healthier diets [4]. The potential of vegetables is to generate positive economic and nutritional impacts. The estimated farm gate value of annual global fruit and vegetable production, at nearly $1 trillion per year, exceeds the farm gate value of all food grains combined (US$ 837 billion). On the other hand, it is likely that the production of fruit and vegetable crops will not increase as rapidly as would be expected. Environmental changes can affect many different aspects of agricultural production. With greater climatic variability, temperature patterns and precipitation are some of the problems faced by fruit producers [5]. Technological advances have focused their efforts on the development of new varieties, crop management techniques, and innovations in postharvest handling and processing. Even in high-income countries such as the US, there is evidence that public funding for research in the agricultural area is less than expected, given its economic value and its contribution to human health [6].

3. Post-harvest losses: tropical and subtropical fruit

It is reported that about one-third of the production of food intended for human consumption is lost or wasted worldwide, which is roughly equivalent to 1.3 billion tons per year. This means that huge amounts of resources directed to food production are used in vain, and that greenhouse gas emissions caused by food production that is lost or wasted are also unnecessary [7]. A very significant part of the food that deteriorates or that is lost at post-harvest stage are fruits and vegetables. These losses occur throughout the management system of fruits during the harvest, transfer to the packinghouse, in the packing, during the storage, transportation and distribution to marketing centers. The causes of the losses in post-harvest are due to economic limitations, the lack of post-harvest technology as well as the lack of trained personnel about the knowledge in technology, management, physiology, and post-harvest pathology of horticultural products. This problem occurs mainly in developing countries, reaching up to 30 or 40% of post-harvest losses [8].

The fruits during their growth in the field are exposed to the attack of pathogenic fungi; they can be established as a latent infection in the fruit; and when the fruit begins its maturation process, the fungi can be activated and continue with their development, leading the deterioration of fruit. To control these pathogens, synthetic fungicides have been applied in a traditional manner; however, due to pollution and the environment issues, damage to the health of people and in general to living beings, as well as generating resistant strains of fungi, it is necessary to find alternatives to pathogen control [9, 10]. Currently, the research of alternative systems to the use of fungicides to control the losses caused by fungi, applying products of biological origin as well as organic and inorganic salts, among others, in order to control fungi infections, without contaminating the environment and without harming living beings has increased [11, 12]. Reducing the losses of post-harvest could solve the problem of hunger in many countries of the world, where not only it is producing more food but rather it is to conserve the food that is currently produced.

4. Chemical methods: applications for post-harvest disease management

Most of the post-harvest losses are attributed to the attack of a large amount of fungus in tropical and subtropical fruits. Chemical control of post-harvest diseases is widely used to maintain fruit quality [13]. There are a wide variety of fungicides for chemical control, and the vast majority is destined or directed to the pre-harvest applications, leaving aside the use in post-harvest stage [14, 15]. To make efficient the use of chemical fungicides, it is necessary to know both the pathogen and the fungicide. From the pathogen, it is necessary to know the genus and the species as well as their concentration found at pre- and post-harvest stages. On the part of the fungicide, it is necessary to know the mode and site of action, as well as the maximum residual limit (MRL) (**Table 1**) permitted on fruits and specific regulations where the fruit will be exported [16]. In **Table 1**, some fungicides used for post-harvest disease of tropical and subtropical fruit diseases are listed. The site and mode of action are summarized in the table, as well as the MRL that are allowed in the US [17] and the EU [18]. An important consequence for the inadequate use and irrational applications of chemical treatments is microbial resistance, and in this sense, it is recommended to alternate formulations to avoid this problem. Besides, post-harvest chemical control should be regionalized to the specific conditions and environment of each crop [19]. It is important to mention that agrochemical companies suggest the doses and formulations to use in a specific crop. At this point, public research centers have an important contribution, not only to verify the efficiency of fungicides but also to establish strategies for the efficient use of post-harvest chemical control [15]. Considering the new consumer tendencies, about secure products free of chemical residues, it is necessary to consider the rational use of chemical fungicides without exceeding the MRL [20–22].

Therefore, the worldwide trend for both consumers and researchers is the reduction of the use of chemical fungicides and the research for biological, organic, and environmentally friendly alternatives. All this is under certification systems to guarantee the implementation, improvement, integration, and harmonization of all mechanisms to ensure a production of healthy and good quality fruit, with high traceability (GLOBALG.AP) [23, 24].

Common name	MRL (ppm)						Chemical group	Mode action	Target site
	EU [17]			US [16]					
	Avocado	**Mango**	**Papaya**	**Avocado**	**Mango**	**Papaya**			
Cyprodinil	1.2	1.2	1.2	1	1	1	Anilino-pyrimidines	Amino acids and protein synthesis	Methionine biosynthesis (proposed)
Carbendazim	—	—	—	3	2	3	Benzimidazoles	Cytoskeleton and motor proteins	ß-tubulin assembly in mitosis
Thiabendazole	10	10	5	3	3	10			
Pyraclostrobin	0.6	0.6	0.6	—	0.05	0.2	Methoxy-carbamates	Respiration	Complex III: cytochrome bc1 (ubiquinol oxidase) at Qo site
Trifloxystrobin	—	0.7	0.7	—	0.7	—	Oximino-acetates		
Fludioxonil	5	5	5	5	5	5	Phenylpyrroles	Signal transduction	MAP/Histidine-Kinase in osmotic signal transduction
Iprodione	—	—	—	10	10	10	Dicarboximides		
Prochloraz	—	—	—	5	2	1	Imidazoles	Sterol biosynthesis in membranes	C14-demethylase in sterol biosynthesis
Difenoconazole	—	0.1	0.6	0.6	0.07	0.2	Triazoles		

Table 1.
Common name, chemical group, mode and target site as well as its MRL in some fruits for consumption [23].

5. Alternative methods in the control of post-harvest decay of tropical and subtropical fruit

5.1 Chitosan

The excessive use of agrochemicals in tropical and in subtropical fruit has leaded the search for new natural products, eco-friendly, and nontoxic to humans. In several investigations, chitosan has proved their efficacy for controlling several post-harvest diseases. Several mechanisms of action have been proposed for chitosan:

a. Pathogens: the interaction of the biopolymer with the microorganism causes changes on cell permeability affecting biochemical processes like homeostasis, fungal respiration as well as nutrient uptake and the synthesis of proteins causing severe damage on fungal cells [12].

b. Plants: induction of defense systems, by the production of important enzymes (phenylalanine ammonium lyase, polyphenol oxidase, among others) and plant immunity, favoring the adaptation of plants to biotic and abiotic stresses [25].

c. Fruits: the capability of chitosan to form mechanical barrier (coating) on fruits offers several advantages of coated fruits like a reduction on respiration rate, avoid water losses maintaining fruit firmness, maintenance of color, among others. Thus the shelf-life of fruits can be extended [36].

The induction of defense systems has been reported by the application of chitosan at post-harvest stage, preventing the development and dispersion of important pathogens such as *Colletotrichum gloeosporioides*, *Alternaria alternata*, *Rhizopus stolonifer*, and *Fusarium oxysporum* [26–28]. Enzymatic activity is also affected by the curative application of chitosan, and it increases the activity of polyphenol oxidase (PPO), peroxidase (POD), and phenylalanine amino-lyase (PAL) that induce the expression genes of β-1,3-glucanase and chitinase, involved in the defense against pathogens [29, 30]. The physiological mechanisms of the fruit are positively affected by the application of chitosan at post-harvest management under biotic and abiotic stress, that is why the post-harvest shelf-life and quality (firmness, appearance, color) of fruit can be maintained during the storage time, besides the respiration rate and ethylene production of fruits decrease [31]. Some studies reported an enhanced content of total soluble solids, ascorbic acid, the nutritional value, and acceptability [30, 32, 33]. Chitosan is compatible with other substances like organic salts, gums, or essential oils, and this alternative can improve their efficacy against pathogens due to a synergistic effect [34, 35]. Even when important information has been generated on the use of chitosan in post-harvest tropical and subtropical fruits, it is still necessary to generate information on the regulation (activation and suppression) of genes that participate in both systems of acquired resistance and those that control the processes physiological, enzymatic, and physicochemical factors of maturation in post-harvest.

5.2 Essential oils

The use of GRAS substances like essential oils (EOs) has increased in the last years, due to the research of alternatives to chemical treatments for disease control [36]. Several investigations have reported the efficacy of essential oils *in vitro* and *vivo tests* [37–40]. The application of EOs in fruits has advantages such as: high effectiveness against several pathogens and low toxicity (nontarget microorganism and humans) [36]. EOs can be applied on fruits directly (vapor phase) or incorporated as active microbial agents in different matrices (films, coatings, among others) at pre-harvest or post-harvest stages. In a recent investigation, cinnamon essential oil was added into biodegradable polyester nets for controlling *A. alternata* [41]. The results are promising due to their efficacy *in vitro* tests by stopping the development of fungus on mycelial growth (72% of inhibition) and the total control of germination. The presence of essential oil did not alter the biodegradability of nets as well as their efficiency to maintain fruit quality and the disease control on infected fruits by reducing the disease incidence. Incorporation of EOs into edible films for active packaging has demonstrated high efficacy against important post-harvest pathogens. Recently, soy protein isolate was used as a carrier for limonene with good results against *P. italicum* (isolated from infected limes) [42]. The liberation and efficacy (*in vitro* tests) of the active agents were evaluated simulating the storage conditions of limes (13 and 28°C). The mycelial growth and germination process were successfully inhibited by the incorporation of limonene into the protein matrix. The results *in vitro* were confirmed in infected limes with *P. italicum* by reducing the severity and blue mold incidence with the application of soy protein isolate with limonene added [43]. In another study, films based on chitosan, oleic acid/beeswax, and lemon essential oils were tested on tomatoes for preserving their quality. According to their results, the applications of these films improve the fruit quality by reducing water losses and the maintenance of appearance of fruit [44]. In a recent study, anthracnose and stem-end rot in the green-skinned on avocado fruits was successfully controlled by the application of thyme oil in combination with a prochloraz solution; besides, this treatment improved fruit quality (firmness) during the storage time [20]. Essential oils of copaiba and eucalyptus were tested against *Alternaria alternata* and *Colletotrichum musae* under in vitro, and the results showed good efficacy at low concentrations of the treatments (0.0–1.0%) [45].

The efficacy of black caraway (*Carum carvi*) and anise (*Pimpinella anisum*) essential oils was tested against *Penicillium digitatum in vitro* as well as *in vivo* (on oranges) evaluations. The results showed that treatments were capable to control fungi development *in vivo* and *in vitro* tests; besides, the quality of oranges was preserved by the application of the treatment of 600 μL/L [46]. In a study, lemongrass oil was tested against *Colletotrichum coccodes*, *Botrytis cinerea*, *Cladosporium herbarum*, *Rhizopus stolonifer*, and *Aspergillus niger in vitro*. According to their results, fungal development, sporulation process as well as spore germination of fungi was affected in different levels (depending on the concentration of treatment) when they were exposed to the treatments by stopping their development [47].

Thus, utilization of EOs for controlling diseases can be alternatives to chemical treatments.

5.3 Plant extracts

Currently, the study of fruits and vegetables at post-harvest stage has focused on the development of alternative control for conservation with better efficiency, sustainability, and lower cost than conventional methods. In this sense, some extracts from plants have proven to be a viable alternative for the extraction of substances with antimicrobial activity with high efficiency and low toxicity [48]. In a study, aqueous and ethanolic extracts from garlic (*Allium sativum*) were evaluated on citrus fruits (*Citrus sinensis*, *Jaffa*, and *Valencia*) with good results (**Table 2**), and it is suggested that the highest antifungal activity of the extract can be produced by the presence of allicin in the soluble fractions of extracts from garlic [49]. In the same way, extracts of garlic, ginger, and celery have been shown to have different effects on the control of the incidence of *Penicillium* sp. in fruits of the species *Citrus reticulata* Blanco [50]. In a similar study, soybean extracts were evaluated as protection method on oranges infected with *P. digitatum*, and green

Fruit	Disease	Pathogen	Extract source	Disease inhibition (%)	References
Orange	Blue mold	*P. italicum*	Garlic	92	[47]
	Green mold	*P. digitatum*	Garlic	90	[47]
			Soybean	100	[49]
	Citrus rot	*Penicillium* sp.	Garlic	80.2	[48]
			Celery	5.3	
			Ginger	16.3	
Cherry	Brown rot	*M. laxa*	*O. crenata*	76	[51]
			S. minor	89	
Nectarine	Brown rot	*M. laxa*	*O. crenata*	75	[53]
			S. minor	100	

Table 2.
Disease control of plant extracts.

mold was significantly reduced (88–100%) due to the presence of β-conglycinin in the soy protein fraction [51]. In recent years, extracts of some angiosperm species have been studied as natural fungicides, such as *Orobanche crenata* and *Sanguisorba minor*, which have shown high efficiency in the control of the diseases produced by *Monilinia laxa* in stone fruits such as apricot, cherry, and nectarine [52–54]. For the case *O. crenata* extracts, the antifungal activity is attributed for the presence of the phenolic compound verbacoside, and for *S. minor* extracts, the efficacy is related to the presence of a combination of phenolic compounds like caffeic acid, quercetin, luteolin, and kaempferol [52–54].

An important approach in fruits is quality. In this sense, the application of some plant extracts can help preserve the fruit quality and improve their shelf life. In a recent investigation, the application of guava extracts (from leaves) and lemon on banana fruits (*Musa sapientum* L.) at post-harvest stage considerably improved the shelf life of the fruits (up to 8 days) compared to untreated fruits (only 4 days), having a positive effect on the conservation of the physicochemical characteristics of fruit during the storage time [55]. Utilization of plant extracts for disease management in both pre- and post-harvest stages can be another alternative to the use of chemical treatments, due to their effectiveness and eco-friendly characteristics.

5.4 Acetic acid, peracetic acid, and hydrogen peroxide

The use of sanitizing agents such as acetic acid ($C_2H_4O_2$), peracetic acid ($C_2H_4O_3$), and hydrogen peroxide (H_2O_2) on the processing and marketing industries of fruits and vegetables have been considered a useful tool to the control of different kinds of pathogens [56]. The FDA has approved the use of these compounds because their decomposition products are water, oxygen, and acetic acid, and these are not toxic compounds and are friendly with the environment [57, 58]. For these reasons, they are classified as GRAS [56]. The acetic acid, peracetic acid, and hydrogen peroxide are recognized as antimicrobial and antifungal agents, because they have high spectrum of attack on bacteria, fungi, spores, and viruses. *In vivo* studies realized on fresh and fresh-cut horticultural products confirm their antimicrobial capacity with the reduction of human pathogens such as *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus mutant*, *Salmonella Thompson*, and *Listeria monocytogenes* [59, 60]. The application of acetic acid, peracetic acid, and hydrogen peroxide also decreases the microbial pollution of aerobic mesophiles, molds, and yeast, obtaining with these innocuous and acceptable products to the consumers [61]. In fruits like guavas, peaches, and tomatoes, the antifungal effect of these sanitizers has been confirmed with the inhibition of phytopathogens such as *Rhizopus stolonifer*, *Monilinia fructicola*, *Alternaria alternata*, *Botrytis cinerea*, *Fusarium solani*, and *Rhizoctonia solani* [62–65]. The inactivation capacity of these compounds on pathogens is based on their high oxidizing power, producing reactive oxygen species (ROS) that generate instability in biomolecules such as DNA, lipids, and proteins, which are vital for the correct cellular functioning of pathogens [56]. Usually, the application of these sanitizers at post-harvest stage in fruits and vegetables is by spraying, dipping, and fumigation. Several authors have investigated the pathogen inhibitory effect of each application system, with the finality to know which application system has better efficiency. The inhibition of *A. alternata* and *B. cinerea* in tomato fruits treated with acetic acid by immersion (50 ml/L by 3 min) and fumigation (50 µl/L by 30 min) was evaluated [64]. The results of this investigation showed the efficacy of both application systems. The growth inhibition of the pathogens tested ranged from 90 to 100% by immersion and fumigation, respectively. In strawberry fruits infected with *B. cinerea* and treated with peracetic acid, where the lower incidence of gray mold disease was obtained with the fumigation system (66%) compared to immersion method (80%) [66]. These results confirm that the capacity of inhibition of these GRAS substances against phytopathogens depends not only on the concentration used but also on the exposure time, the microorganism tested, and the application method [67]. The individual application of acetic acid, peracetic acid, and hydrogen peroxide has controlled microbial contamination in an acceptable way, but different reports have been showed that their combination with other compounds and technologies increases microbial control. Some of the technologies used include ultrasonic, organics salts, essential oils, ultraviolet light, hot water, and steam [63, 68–73]. Maintaining not only the safety but also the quality on post-harvest products is a constant challenge for the horticulture industry. The application of acetic acid, peracetic acid, and hydrogen peroxide can also improve the quality of the products, according to different in *vivo* studies carried out. In peppers, fruits treated with the solution of hydrogen peroxide and applied by dipping (15 mM by 30 min) increased their shelf life and the fruits maintained their appearance after 2 weeks stored at 20°C compared to control fruits [74]. In according to their results, fruits and vegetables such as tomatoes [75], grapes [76], nectarines [63], apples [68], and cherries [77] have been maintained the quality. Thus, the application of these sanitizers in post-harvest is a practical strategy to maintain the safe and quality of the post-harvest products.

5.5 Salts: organic and inorganic

Nowadays, there is a tendency to reduce the use of synthetic fungicides in agriculture. In this sense, organic and inorganic salts are chemical substances that are food additives generally recognized as safe (GRAS). They are widely used in the food industry due to the low toxicity and environmental impact, besides they can be combined with other control systems to control phytopathogens in post-harvest stage in different fruits and vegetables [78, 79]. It has been shown that salts such as calcium propionate completely inhibit the mycelial growth of *B. cinerea* at a concentration of 5% (w/v) [78]. This may be due to the fact that it changes the plasma membrane, thus inhibiting essential metabolic functions. Some authors have described that high concentrations of sodium benzoate inhibited the growth of *A. Alternata* [80], and this is attributed to the fact that weak acids within the cell create a dissociation, causing that protons and anions accumulate and cannot cross the plasma membrane again [81]. Bicarbonate salts, also were effective to inhibit the growth of various pathogens, the efficiency of this salt is attributed to the fact that it creates cellular ionic imbalances affecting the synthesis of polyamines and DNA during cell division [82, 83]. With the accessibility of these compounds and their effectiveness, it is possible that they can be adopted to reduce the use of traditional fungal agents. In a study, potassium sorbate was applied at low concentrations (1%) on infected tomato fruits, and significant reduction of disease incidence was obtained against *Rhizoctonia solani*, *Colletotrichum coccodes*, *Botrytis cinerea*, and *Alternaria solani* [84]. In another study, gray mold caused by *Botrytis cinerea* was totally controlled by the application of potassium sorbate at 1% [85]. The effectiveness of potassium sorbate is related to its undissociated form (sorbic acid), which has the ability to penetrate the cell membranes, causing an internal imbalance affecting enzymes related to the growth of microorganisms [86]. In a study on citrus, potassium sorbate at 3% solutions was applied in combination with heat treatment (62°C, 60 s) to evaluate their effectiveness in reducing the disease incidence caused by *Penicillium* strains. The results showed that the treatments can reduce the disease incidence on "Clemenules" (20%), "Nadorcott" mandarins (25%), "Fino" lemon (50%), tangerine "Ortanique" (80%), and "Valencia" oranges (95%) stored 20°C for 7 days. Besides, when infected and treated fruits were stored at 5°C during 60 days, on "Valencia" oranges, the green mold (*Penicillium digitatum*) was reduced up to 95% and blue mold (*Penicillium italicum*) up to 80% [87]. Green mold caused by *P. digitatum* was reduced up to 80% on infected oranges by the application of sodium benzoate (3%) in combination with hot water (53°C) during 60 s, and fruits were stored at 20°C for 7 days [88]. The effectiveness of sodium benzoate is related to its undissociated form of benzoic acid, which can enter the cell membrane, and its neutralization within the cell leads to acidification of the intracellular space, thus affecting the growth of fungus [89]. On the other hand, another alternative to avoid pathogen development is the use of silicates, and the most used in the fruit industry is: potassium, calcium, and sodium. It has been reported that the action of these organic salts causes, among other effects, inhibition of mycelial growth and alteration of the morphology of the hyphae, and in addition, the germination of conidia is inhibited and causes alterations in their external morphology. The application of sodium silicate induces alterations in the cell wall and in the morphology of the hyphae on pathogenic fungi. It is important to mention that the application of silicate as post-harvest treatment presents results similar to those reported with chitosan and tebuconazole [90]. The investigations of the use of inorganic salts in post-harvest stage are not very frequent, even there are not yet many studies that determine the effect that this may have if applied to different types of crops. These salts have been applied on orange [91], melon [92], avocado [93], and papaya [94], with good results due to the treatments, which form a barrier against pathogens on surface fruit. In addition, the application of silicates can improve some quality attributes of fruits like the maintenance of weight and reducing the respiration rate due to the capacity of the silicates to deposit between the cell wall and the cell membrane, thus decreasing the permeability, besides the stomata are covered, maintaining the humidity of the fruit and reducing its respiration [95, 96]. With the accessibility of these compounds and their effectiveness, it is possible that they can be adopted to reduce the use and applications of traditional fungal agents.

5.6 Jasmonic and salicylic acid

Resistance induced to disease in plants by biotic and abiotic elicitors is a very effective method for restricting the spread of fungal infection [97]. In general, pathogen resistance processes in plants are based on their own defense mechanisms, such as pre-existing antimicrobial compounds and inducible defense mechanisms. Resistance to diseases induced in plants and fruits by biotic or abiotic treatments is a very attractive strategy to control diseases [98]. The signal molecules salicylic acid (SA), jasmonic acid (JA), and methyl jasmonate (MeJA) are endogenous plant growth substances that play key roles in development and responses to environmental stresses. These signal molecules are involved in some signal transduction systems in plants and fruits, which induce particular enzymes catalyzing biosynthetic reactions to form defense compounds such as polyphenols, alkaloids, or pathogenesis-related (PR) proteins. This can result in induction of defense responses and provide protection for plants and fruits from pathogen attack [99]. Salicylic acid activates induction of acquired systemic resistance (SAR) response in plants, proving that in the plant-microorganism interaction, the enzyme phenylalanine ammonia lyase (PAL) is induced, which is the key in the biosynthesis of phenolic compounds [100]. Peroxide has an antibiotic activity against pathogens; it could intervene in the signaling cascade for the expression of defense genes. SA regulates activities of enzymes, peroxidase (POD), and polyphenoloxidase (PPO), that are related to induced defense of plants and fruits against biotic and abiotic stress [101]. Jasmonic acid (JA) and methyl jasmonate (MeJA) have been found to occur naturally in a wide range of higher plants. MeJA is an occurring plant growth regulator that modulates many physiological processes including responses to environmental stresses [99]. Studies indicate that acquired systemic resistance depends on signaling mediated by MeJA and is associated with some signal transduction systems, which induce particular enzymes that catalyze biosynthetic reactions to form defense compounds such as polyphenols, alkaloids, reactive oxygen species ($ROS^{\bullet}$), or PR proteins [102]. The exogenous application of MeJA induces and increases the activity of defense enzymes such as β-1,3-glucanase (β-Gluc), chitinase, polyphenoloxidase (PPO), and phenylalanine ammonia lyase (PAL), which are enzymes associated with resistance to diseases [103]. Application of MeJA effectively suppressed gray mold rot caused by Botrytis cinerea in strawberry [104] and decreased fruit decay on papaya fruit infected by *C. gloeosporioides* and *Alternaria alternata* [105]. For grape fruits inoculated with Botrytis cinerea, the application of MJ (0.01 mM) increased the enzymatic activity of PAL and PPO [106], and the same behavior was reported on Hass avocado fruits with an increase in the activity of the resistance enzymes, chitinase, β-1,3-glucanase and PAL [11]. The application of MeJA (10 mM) in cranberry fruit inoculated with *Penicillium citrinum* maintained greater POD and PAL activity [107]. There are several studies on the application of SA in fruits for the induction of defense mechanisms against pathogens. Resistance of the tomato fruit against *Botrytis cinerea* using SA as a resistance inducer, a significant increase in the expression level of the PR1 gene was observed in the fruit and a lower expression in the PR2 and PR3 genes [108]. The post-harvest application of SA (2 mg/mL) showed a decrease in the severity of anthracnose in mango cv. Kensington Pride [109]. Thus, the use of inducers offers several advantages for post-harvest disease control; besides, they can combine it with other methods to enhance their efficacy.

5.7 Coatings and edible film from natural sources

Edible coatings on fruits and films made from natural sources are a novelty method and an alternative to the use of post-harvest chemical treatments, particularly in highly perishable fruits [110]. The coatings act as a barrier during processing, handling and storage, delaying the deterioration of food, improving its quality, and extending their shelf life. The functional properties of edible coatings and films depend on their application and the characteristics of the fruit in which they are going to be used. The nature of the compound that is used to produce the coating strongly influences its efficiency, thus it must take into account both its physical and chemical properties, as well as its mechanical and permeable properties. Based on this, if the edible coatings are used properly, they may be able to delay the ripening of the fruit, slow the decomposition of the chlorophyll, reduce the weight loss, retain the ascorbic acid, improve the appearance of the fruit, and especially prolong the shelf life [111–113]. To date, the functional properties of different compounds for the production of edible coatings have been studied. Compounds from natural sources such as *Aloe vera* and waxes have shown promising properties to be applied in the preservation of tropical fruits, and therefore coatings based on these compounds can represent an innovation in the commercial application and exploitation of these resources. *Aloe vera* gel is one of the natural compounds, which has gained a great interest. Because of its nature mainly constituted by polysaccharides, it is capable to form a uniform layer on the surface of the fruit and be easily applied [114]. *Aloe vera* coating can improve the post-harvest qualitative and quantitative traits, thus it can be an alternative for chemicals preservative in the commercial storage of tropical and subtropical fruits.

Aloe vera-based coatings have been successfully tested in mango fruit ripening (*Mangifera indica L. cv. Kensington Pride*) [111]. *Aloe vera* coating reduced aroma volatile biosynthesis in the fruit pulp. Likewise, it was found that coatings delayed ripening of the fruit compared to control. They state that this effect was characterized by the suppression respiration and/or delayed climacteric peak, late fruit color development, and a greater firmness in the coated fruit compared to the uncoated ones. Similarly, the effect of edible coatings based on *Aloe vera* to extend the shelf life of the guava (*Psidium guajava*) has been demonstrated. Achipiz et al. [112] found that a coating with a concentration of 4% potato starch and 20% *A. vera* showed a favorable effect by reducing the weight loss and the respiratory rate of the fruit and increasing the firmness and retention of the vitamin C content after 10 days of storage. In another study, the effect of the *Aloe vera* gel coating on the store ability of peach fruits was evaluated [115].

The Carnauba wax stems from the leaves of the Brazilian palm *Copernicia cerifera*. It is produced as a protection method to prevent dehydration and damage. The Carnauba wax as an edible coating is being extensively studied because it has been demonstrated to reduce water loss, improve appearance, and prolong shelf life in a wide variety of fruits. An edible coating based on cassava starch and carnauba wax adding organic acids and calcium chloride was evaluated in mangoes cv Tommy Atkins minimally processed [113]. According to the results, the attributes of sensory, physical, and chemical quality were maintained, and the useful life of fruit was possible to prolong up to 24 days under refrigeration conditions (5 ± 1°C and 90 ± 2% RH).

Saucedo-Pompa et al. [116] developed an edible coating based on candelilla wax to improve avocado quality. Furthermore, they studied the effect of the ellagic acid addition in the shelf life of the fruit. The results showing the application of edible films based on candelilla wax improved the quality of the avocado fruits and extended its shelf life compared to the control fruits. Also, the addition of ellagic acid to the edible film showed an important effect, since it reduced the damage caused by the fungus *C. gloeosporioides* (the main phytopathogenic fungus for avocados) and significantly improved the quality and shelf life of avocado. Another coating based on mesquite gum-candelilla wax was evaluated in Persian limes [117]. The results showed that coatings decreased the weight loss of the fruit. In addition, by adding mineral oil (33%) to the emulsion, they observed that water vapor permeability was significantly improved, as well as its appearance.

5.8 Ultrasound

Ultrasonic is an economically and environmentally viable alternative for the processing of fruit and vegetable post-harvest [118]. Low intensity ultrasound has been used for quality control of fresh fruit and vegetables in pre- and post-harvest processes [119]. Harvesting time and storage period can be indirectly assessed by ultrasound measurements that are linked physicochemical measurements such as firmness, mealiness, dry weight percentage, oil contents, total soluble solids, and acidity [119]. On the other hand, decontamination of fresh product by ultrasound is relatively recent. The inactivation of microorganism caused by cavitation phenomenon has promoted high intensity ultrasound as method to decontaminate fruits and vegetables. The efficiency of the ultrasound process is affected by several factors such as power level, treatment time, and temperature [120]. Additionally, ultrasound can be applied directly to the medium (water) or in combination with some compounds (organic salts, organic acids, chitosan, among others) to achieve better results. Concerning individual ultrasound application, ultrasound at low frequencies (20 and 40 kHz) has demonstrated to decrease the microbial load of mesophilic aerobes in lettuce (0.9 log CFU/g) and strawberry (1.49 log CFU/g) [121, 122]. At the present time, ultrasound is being implemented in combination with various aqueous sanitizers in order to improve microbial safety and maintain food quality on organic fresh produce. *In vitro* assay, the addition of low weight chitosan (1000 ppm) enhanced the inactivation of *Saccharomyces cerevisiae* by ultrasound (20 kHz) at 45°C in Sabouraud broth (pH 5.6). After 30 min of exposure to chitosan, approximately 1-log cycle reduction of the yeast was obtained leading to a final reduction of more than three log cycles after 30 min of the ultrasonic treatment [123]. In the case of *in vivo* assays, the effectiveness of ultrasound (40 kHz, 5 min) alone and organic acids (0.3, 0.5, 0.7, 1.0, and 2.0% of malic acid, lactic acid, and citric acid) alone and their combination on reducing *Escherichia coli* O157:H7, *Salmonella Typhimurium*, and *Listeria monocytogenes* in fresh lettuce was compared. For all three pathogens, the combined treatment of ultrasound and organic acids resulted in additional 0.8–1.0 log reduction compared to individual treatments, without causing significant quality change (color and texture) on lettuce during 7 day storage. The maximum reductions of *E. coli* O157:H7, *S. Typhimurium*, and *L. monocytogenes* were 2.75, 3.18, and 2.87 log CFU/g observed after combined treatment with ultrasound and 2% organic acid for 5 min, respectively (Sagong et al., 2011). In peach fruit, the effect of ultrasound (40 kHz, 10 min) and salicylic acid (0.05 mM) either separately, or combined on blue mold caused by *Penicillium expansum* was investigated. The results showed that the application of salicylic acid alone could reduce blue mold, while the use of ultrasound had no effect. Results also revealed that salicylic acid combined with ultrasound treatment was more effective in inhibiting fungal decay during storage than the salicylic acid treatment alone. The combined treatment increased the activities of defense enzymes such as chitinase, β-1,3-glucanase, phenylalanine ammonia lyase, polyphenol oxidase, and peroxidase, which were associated with higher disease resistance induced by the combined treatment. Furthermore, the combined treatment did not impair the quality parameters of peach fruit after 6 days of storage at 20°C [124]. The incorporation of ultrasound alone or in combination with other agents in decontamination process could be a useful preservation technique for post-harvest fruits and vegetables. Combination of ultrasound and sanitizers could increase pathogen reduction without affecting the product quality, while concentration of sanitizers could be reduced as well as treatment time required, saving time and money and avoiding significant risks to consumers.

5.9 Fogging

In order to prolong the shelf life of fruits and vegetables in post-harvest periods, various technologies have been developed that maintain their integrity as well as their nutritional properties. One of the technologies little explored at present is the use of ultrasonic nebulization (Fogging) as a method of distribution of compounds that serve to prevent or control pathogenic diseases in the post-harvest period. Fogging has been used successfully for the spraying of disinfectants such as chlorine dioxide, sodium hypochlorite, hydrogen peroxide, acetic acid, and ethanol, achieving the control of epiphytic microorganisms on the surface of the strawberry (fungi and bacteria), thus reducing the decay index by up to 83.2%, demonstrating that nebulization is an effective method for the reduction of diseases in the post-harvest stage [125]. Regarding fruit quality, in a study with strawberry, peracetic acid was applied as a disinfectant by ultrasonic nebulization, and the results showed that the anthocyanin and phenolic compound contents were preserved even when the fruits were exposed to low concentrations of peracetic acid [126]. In the post-harvest and fruit storage period, it is necessary to minimize chemical products as environmental precautions and avoid adapting pathogens to various fungicides, causing high losses of between 30 and 50% in vegetables and fruits [125, 127]. The use of ultrasonic nebulization offers advantages such as the reduction of the amount of disinfectant and a better distribution of the treatment on the fruits, and in a study of figs, the inhibition of gray mold disease (B. cinerea) 80% of control was achieved with only the application of chlorine dioxide (1000 μL/L) [127]. Ultrasonic nebulization as a conservation method in the post-harvest period gives high benefits in different ways, by reducing the quantity of substances applied, the exposure time as well as a better distribution of the treatments, its application in fruits and vegetables has not been explored, thus the development of this technology can offer an alternative to the use of chemical fungicides for the control of diseases.

6. Conclusions

Considering the new tendencies in fruit industry and marketing, the use of alternative methods represents a suitable approach for several agriculture commodities not only for controlling post-harvest diseases but also for maintaining fruit quality.

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Chapter 2

Harnessing the Recent Approaches in Postharvest Quality Retention of Fruits

Nirmal Kumar Meena and Kalpana Choudhary

Abstract

Fruits are an important component of our diet. Postharvest quality retention during supply chain management is a major concern and has become a priority of today's world. Owing to this, food security is a big challenge and, to mitigate this nutritional, security is a major task. The existing technologies have brought about several desirable changes in logistics of postharvest handling of fruits. As the trend has been changing, people are moving away from synthetic treatment agents; thus, these have been replaced with eco-friendly products. Since the last few years, introduction of some environmental and consumer friendly approaches like brassinosteroids, methyl jasmonates, oxalic acid, salicylic acid, edible coatings, biocontrol agents, irradiation, and cold plasma techniques has made this line more interesting across the globe. These agents work effectively and better over traditional synthetic chemicals. Application of these formulations has been found to be better to retain the quality and fresh like appearance during storage of fruits during supply chain and storage. Thus, there is urgent need to develop some novel technologies for better establishment of fruit growing industries and their maximum retention of quality. The use of these in an integrated manner could be a better way to minimize this huge loss and maximize the quality.

Keywords: postharvest quality, respiration, fruits, novel hormones, edible coatings, enzymes

1. Introduction

Fruits are integral part of our balance diet. These provide many phytochemicals that adds variety to the diet and cure many nutrient related disorders. Food security is a major burning aspect especially in developing countries. The production has been increasing day by day but quality concept has left into backseat. Despite, increasing the food production, the postharvest shelf life and retention of quality up-to a sufficient level is still needed a lot of attention. It is well known that fruits are living commodity and more prone to postharvest decay so, they require proper management to retain quality and shelf life. The quality and shelf life of produce is affected by many preharvest factors like genotype and rootstocks [1], nutrients and foliar spray [2], quality of water [3] tree age and canopy management [4, 5] use of growth hormones and several postharvest factors like precooling, edible coatings, use of postharvest fungicides, controlled and modified storage techniques, etc. The main causes of postharvest losses are weight loss, loss of pigmentation, storage diseases and disorders and physiological changes. The rate of deterioration varies and largely depends upon intrinsic characters of produces, storage conditions and state of produces during storage [6].

The major aim of postharvest technology is to optimize and reducing the losses during unit operations by adopting the emerging technologies. There is a strong lacuna of sound postharvest management for quality retention during supply chain [7]. Existing technologies are insufficient to reduce these postharvest losses. Moreover, the awareness towards harmful chemicals, sanitizers, coatings materials and other unsafe chemicals forced the research community to invent alternatives of these technologies. To meet out the satisfactory results, several researchers applied these technologies to induce shelf life and maximize the quality of fresh fruits. These includes edible and nonedible coatings, use of salts, postharvest spray of different phytohormones, irradiation treatment, etc., but side by side, these existing technologies are being replaced with new emerging technologies. With the advancement of technology and science, many recent trends have come into existence, and they are safe to health and environment and consumer friendly. Among major recent approaches, some technologies are applied use of brassinosteroids (BRs), methyl jasmonates (MeJA), oxalic acid (OA), salicylic acid (SA), application of edible coatings and films, irradiation, use of biocontrol agents and advance storage techniques like controlled atmospheric storage and modified atmospheric packaging which really revolutionized the postharvest industry (**Table 1**). Application of these technologies proven a milestone in the supply chain management of fresh commodities and made them more market oriented by addition of some extra quality. The application methodology and concentration are specific to formulation and nature of products. Some of them are novel phytohormones which enhance the defense system of fruits and help in achieving delayed ripening and senescence. Edible coatings are also used solely or in mixture with other coatings which help in reducing the firmness and retention of quality. In addition to that, some nanomaterials, fortified materials, antimicrobials, etc. can be added to the coating mixtures which significantly reduce the microbial contamination and increase the quality. Some recent storage atmosphere techniques are available which reduce the storage disorders and also enhance the shelf life. In this chapter we jotted down the different technologies and their mechanism application, and efficacy for enhancing the quality and shelf life of the fruits.

2. Brassinosteroids

Brassinosteroids are referred as the sixth group of plants hormones [71] as well as hormone of the twenty-first century, because of its important contribution in various physiological processes [72]. At the first, this brassinolide (most active form of BRs) was extracted from the pollens of rapeseed plants (*Brassica napus*) and later on from the insect galls of chestnut and termed as "castasterone." Nowadays, many plant species are using to extract the BRs like date palm pollens, oranges pollens, loquat flower bud, seeds of persimmon and pumpkin, etc. BRs involve in many developmental process like cell division, cell elongation, vascular differentiation, photomorphogenesis, and senescence. Owing to its multiple effects, it can be used in horticulture to improve growth, yield and quality. In recent, this hormone was proved as a natural, nontoxic, and safe to environment, thus it may be used in horticultural crops to improve the quality of the produce [73]. There is several evidence that it acts as growth promoter in plants either solely or in combination with other hormones like GA3, jasmonic acid and polyamines. Several researchers carried

Name of technology	Crop with dose	Salient findings	References
BRs	Grape, mango Epi-BL (45 and 60 ng g^{-1} FW)	Enhances rate of ripening when applied exogenously	[8, 9]
	Jujube (5 μM)	Reduce senescence process in Jujube	[10]
	Strawberry (400 mm BL, 200 mm BZ)	Promote fruit ripening, fruit color development	[11]
	Grape (0.4 mg/L BRs)	Managing quality of fresh produces like more anthocyanin accumulation	[12]
		Reduced activities of PAL and PPO enzymes	[13]
	Sweet cherry (0.2 mg L^{-1})	Better shelf life	[14]
MeJA	Peaches	Delayed ripening	[15]
	Peach (0.1 mmol L^{1}), pomegranate (0.01 mM), pineapple (10^{-3}, 10^{-4} and 10^{-5}M)	Protect from chilling injury	[16–18]
	Loquats (10 μmol/L) Strawberries (1 μmol/L)	Reduced anthracnose Inhibited rot caused by *Botrytis cinerea*	[19, 20]
	Sabrosa strawberry (8 μmol L^{-1})	Enhancing total antioxidant capacity (TAC) and antioxidant enzymes activity	[21]
	Fuji apple (2240 mg L^{-1})	Higher soluble solid content, titratable acidity and delayed fruit firmness loss	[22]
	Pineapple (1 mm)	Inhibits ethylene production, weight loss, internal browning and enhances total phenol which produces during cold storage	[23]
Oxalic acid	Mango (30 mm)	Extend storage shelf-life and suppress quality deterioration	[24]
	Ber (10 mm)	Maintain enzymatic activity of phenylalanine ammonia lyase (PAL), malondialdehyde (MDA) and superoxide dismutase (SOD)	[25]
	Mature green tomatoes	Accumulation of lycopene during the postharvest ripening	[26]
	Banana (20 mm)	Reduce the deterioration and enhances their storability of fruits	[27]
SA	Guava (600 μmol)	Improve postharvest quality and delay ripening	[28]
		Retention of firmness	[28]
		Increase the shelf life during short term storage	[29]
	Apple (2 mm)	Increasing of antioxidants	[30]
	Plum (1.5 mm)	Suppresses chilling injury by altering malondialdehyde content, and enhancing polyamine accumulation	[31]
	Apple (2.0 mmol/L)	Reduce percentage of weight loss, increase total soluble solids and limiting changes in fruit color and texture	[32]
	Mango (2.0 mm)	Reduces weight loss, maintain ascorbic acid and increase shelf life at ambient temperature	[33]

Name of technology	Crop with dose	Salient findings	References
Edible coating	Papaya (papaya leaf extract + *Aloe vera* gel 1:1)	Retained firmness Delayed peel color development Reduced weight loss	[34]
	Blueberries (SemperFresh)	Slow down weight loss	[35]
	Plum (3% alginate)	Decreased weight loss Inhibits ethylene production Slowed softening process	[36]
	Fresh-cut persimmon (pectin coating + potassium sorbate, sodium benzoate)	Inhibited browning, molds, yeasts and psychrophilic aerobic bacterial growth	[37]
	Fresh-cut oranges (*Aloe vera*, gelatin, green tea)	Prevention of weight loss, retarded the microbial growth, extended the shelf life during cold storage	[38]
	Kiwi fruit slices (*Aloe vera*)	Inhibition of microbial growth	[39]
	Strawberry (CH and CMC coatings enriched with MSO) (1%)	Decrease microbial spoilage	[40]
	Pomegranate (chitosan)	Delays the ripening	[41]
	Banana (putrescine and chitosan)	Increases in phenolic compound and antioxidant activity at the end of the storage period. 1% chitosan coating enhances postharvest quality and shelf-life of banana	[42]
	Mango beeswax and chitosan	Reduces PLW, maintained firmness and reduces the activities of hydrolysis enzymes	[43]
	Pomegranate (0.5% black seed oil)	Reducing chilling injury and controlling gray mold disease	[44]
	Apple (eucalyptus, thyme 0.6% and lemon grass oil 0.8%)	Reducing the incidence of gray and blue molds	[45]
	Mandarin (coconut oil 100%)	Enhance shelf life, quality and also delayed the mold appearance	[46]
	Strawberry (chitosan and carboxymethyl cellulose (CMC))	Effective to prevents the loss of firmness and aroma volatiles, reduces the primary and secondary metabolites	[47]
	Mangoes (five nanolayers of pectin and chitosan)	Better quality in terms of weight loss, total soluble solids and titratable acidity	[48]
	Citrus (CMC/chitosan)	Maintained fruit firmness	[49]
Irradiation	Mango, pear, peach, strawberry, Nagpur mandarin and acid lime	Delaying of ripening, reduced fruit firmness, reduced rate of respiration and ethylene and lower enzymatic activities which extend the shelf life	[50–53]
	Papaya, mango (<150 Gy)	Helpful against many quarantine pests like *Bactrocera dorsalis*, fruit fly and stone weevil	[54, 55]
	Mango	Kill third instar larva of Mexican fruit fly and Mediterranean fruit fly as a quarantine treatment	[56]
	Nagpur mandarin	Delayed the Penicillium rot with higher total soluble solids	[53]

Name of technology	Crop with dose	Salient findings	References
LEL	Citrus (70 L mol m^{-2} s^{-1}), *Vitis vinifera* (80-mol m^{-2} s^{-1})	Induce disease resistance against *Penicillium digitatum* and *Botrytis cinerea*	[57, 58]
	Strawberry, citrus, peaches (40 mol m^{-2} s^{-1}), banana (464–474 nm)	Induces fruits ripening by ethylene production	[57–59]
	Banana	Positive effect on the quality (accumulations of ascorbic acid content, total sugar and phenols)	[59]
	Strawberry (blue (470 nm) light at an intensity of 40-mol m^{-2} s^{-1})	Increase total sugar content, total phenols ascorbic acid and enhances antioxidant enzyme activities (catalase, superoxide dismutase and ascorbate peroxidase)	[60]
Biocontrol agents	Citrus and stone fruits	Control green mold in citrus and brown rot (*Bacillus subtilis*)	[61, 62]
	Strawberry, grape and banana	Antagonist against gray mold and anthracnose	[63, 64]
Cold plasma	Cashew apple juice (30 mL/min)	Increase the sucrose content whereas reduces the glucose and fructose	[65, 66]
	Cut kiwifruit (dielectric barrier discharge, at 15 kV for 10–20 min)	Helpful in color retention and reduction of darkened area formed during storage	[67]
	Sour cherry, pomegranate juice (at 25 kHz, Ar, 0.75–1.25)	Increases the anthocyanin content in some fruit juices	[68, 69]
	Mandarin (2.45 GHz, 900 W, 1 L/min, 0.7 kPa, N_2, He, $N_2 + O_2$ (4:1) for 10 min)	Increases antioxidant activity, total phenolic content and also inhibits *Penicillium italicum*	[70]

Table 1.
Application of different approaches to extend the quality and shelf life of fruits.

out experiments on postharvest applications and got success to found significant achievements. A well understand correlation has been established between BRs and ripening in the fruits. Their applications alter the ripening related genes, resulting regulation in ripening and senescence. It enhances rate of ripening when applied exogenously in grape 0.4 mg L^{-1} [8], mango at the rate of 45 and 60 ng per gram of fresh weight [9], tomato in the concentration of 3 μmol L^{-1} [74] and arrest the senescence process in Jujube at a specific concentration (5 μM) [10]. However, majority of author reported that action of BRs is concentration specific and varies from nanograms to milligrams. Schlagnhaufer et al. [11] observed in his experiment that BRs application enhanced the ripening process by stimulating ethylene biosynthesis between methionine and ACC pathway. BRs (@400 mm BL, 200 mm BZ) involved in fruit color development of strawberry by expressing BR receptor, gene (*FaBRI1*) during initial red color development [12]. The applied BRs is also helpful in managing quality of fresh produces like more anthocyanin accumulation in grapes [71], reduced activities of PAL and PPO enzymes [13]. Roghabadi and Pakkish [14] found better shelf life of "Tak Danehe Mashhad" sweet cherry, when treated with BRs at the rate of 0.2 mg L^{-1}.

3. Methyl jasmonate (MeJA)

Jasmonic acid (JA) and its methyl ester (methyl jasmonate (MeJA)) are types of endogenous phytohormones that have distinct and potentially useful properties which affect plant growth and development in response to environmental stresses. Methyl jasmonate (MeJA) was discovered in *Jasminum grandiflorum* flower extracts in 1962 as a sweet-smelling compound [75]. It has inhibitory effect on ethylene biosynthesis and delayed ripening of peaches [15]. Application of jasmonate has been found effective to protect from chilling injury in many fruits crops, such as peach (@ 0.1 mmol L^{-1}) [16], pomegranate (@ 0.01 mM) [17], and "Smooth Cayenne" pineapple (@10^{-3}, 10^{-4} and 10^{-5} M) [18]. It has been discovered that exogenous treatment with MeJA enhances antioxidant capacity of different harvested fruits and horticultural crops [17, 76]. Applied MeJA also controls the postharvest diseases and decay of fruits. It was observed that application of 10 μmol/L, MeJA effectively reduced anthracnose in loquats [19] and inhibited rot caused by *Botrytis cinerea* in strawberries when treated with 1 μmol L^{-1}. [20] It has good potential to enhance the postharvest life of Sabrosa strawberry fruit (@8 μmol L^{-1}) [21]. Aglar and Ozturk [22] found that application of 2240 mg L^{-1} MeJA on fruits of Fuji apple had higher soluble solid content, titratable acidity and delayed fruit firmness loss. Boonyaritthongchai and Supapvanich [23] observed in pineapple that treatment of fruits with 1 mM MeJA inhibits ethylene production, weight loss, internal browning and enhances total phenol which produces during cold storage.

4. Oxalic acid

Oxalic acid (OA) is an organic compound which is chemically $C_2H_2O_4$. It naturally occurs in some fruits such as carambola, bilimbi, ripe papaya and kiwifruit. Oxalic acid has main role in regulating the physiology of many processes and various biochemical pathways inside the plants. It helps to increase the photosynthetic ability of plants, thereby cause increase in total soluble solids, sugars and titratable acidity. Oxalic acid reduces the production of polygalacturonase (PG) and pectin methyl esterase (PME), which are responsible for cell wall degradation, so that the treated fruit maintains the firmness in plum [77]. Zheng et al. [24] found that postharvest application of oxalic acid (@30 mM) could be a promising method to extend the storage shelf-life and suppress quality deterioration of mango fruit. It was observed in ber cv. Gola that fruit treated with 10 mM oxalic acid found to be best in maintaining enzymatic activity of phenylalanine ammonia lyase (PAL), malondialdehyde (MDA) and superoxide dismutase (SOD), their minimum values were observed with this treatment [25]. Li et al. [26], observed in mature green tomatoes that postharvest application of OA increased accumulation of lycopene during the postharvest ripening which may be due to upregulation of the expression of genes that codified for enzymes involved in carotenoid biosynthesis. Huang et al. [27] reported that dipping of banana in 20 mM oxalic acid for 10 min followed by storage at room temperature, reduced the deterioration and enhances their storability of fruits.

5. Salicylic acid

Salicylic acid (SA) or ortho-hydroxybenzoic acid is a pervasive, natural simple phenolic compound, which is frequently disseminated in plants and involved in the regulation many catabolic activities in fruits and vegetables. It is considered as a safe chemical compound for postharvest application. It has been used to improve postharvest quality such as delay ripening and retention of firmness in guava

(@600 μmol) [28, 29]) and increasing of antioxidants (@ 2 mM) in apple [30]. The application of 1.5 mM SA suppresses the chilling injury by altering malondialdehyde content and enhancing polyamine accumulation in plum [31]. Atia et al. [32], reported that the GA3 and SA treatments (2.0 mmol/L) reduced the percentage of weight loss, increase total soluble solids and efficient in limiting the changes in fruit color and texture in apple fruits. Mandal et al. [33] found that treatment of mango fruits with SA 2.0 mM reduces weight loss, maintain ascorbic acid and increase shelf life at ambient temperature. It is suggested that 600 μmol salicylic acid is beneficial to increase the shelf life of guava fruit during short term storage [29].

6. Edible coatings and films

Edible coatings are the application of commercial food grade waxes or films to product surface natural glossiness in addition to or as a replacement for natural defensive waxy coatings. These provide a barrier for moisture, oxygen and solute movement for the food and extend the shelf life by decreased respiration and ethylene, there are many commercial formulations are available in market which widely applied on the surface of fruits and vegetables. Among them Citrashine, chitosan, SemperFresh, shellac wax, carboxymethyl cellulose, guar gum, lasoda gel, *Aloe vera* gel, bee wax, etc. are common. However, plant based surface coatings and extracts are more popular than those of chemically synthesized. Use of essential oils, leaf extracts and exudations also established their strong market. These can be applied directly on the produce surface due to its edible properties, biodegradability and also healthy distinct from chemical nonedible coating which leave residual effect on the product. They have antimicrobial properties and also act as barrier. It has many positive affect on fruits such as delayed ripening in pomegranate fruits [41], inhibits ethylene production and delays softening process in plum [36], retaining firmness in papaya and [34], reduces weight loss in blueberries [35], inhibits pathogenic spoilage in kiwi fruit slices [39], in fresh cut persimmon [37], in fresh cut orange [38] and in strawberry [40]. Hosseini et al. [42] observed that coating of banana fruits with putrescine and chitosan increases in phenolic compound and antioxidant activity at the end of the storage period and it was also suggest that 1% chitosan coating is effective in enhancing postharvest quality and shelf-life of banana. It was reported that treatment of mango fruits with 2% beeswax and chitosan reduces physiological weight loss, maintained firmness by reduction in respiration rate and reduces the activities of hydrolysis enzymes [43].

Similar to that of waxes, essential oils could be effective in reducing microbial load during transportation and storage. These plant-based oils, oleoresins, leaf extracts, etc. gained popularity as surface disinfectant on fresh fruits. Many researchers have investigated the effect of leaf extracts like custard apple leaf extracts, tea extracts, neem oils, thyme oil, clove oil, ocimum oil, coconut oil, lemon grass oils, *Aloe vera* extracts, and many other herbal formulations on postharvest diseases. They may helpful in reducing many diseases like gray mold of grape, strawberry, blue mold of apple and citrus, anthracnose and stem end rot in mango and citrus, etc. and found to be effective to a certain extent. The application of these oils may increase the activities of certain defense related enzymes thus beneficial in inducing resistance mechanism in fruits. The application of 0.5% black seed oil was found effective in reducing chilling injury and also controlling gray mold disease in pomegranate (Kahramanoğlu et al. [44]). Abd-El-Latif [45] reported that application of eucalyptus, thyme and lemon grass oil (at a concentration of 0.6 and 0.8%) was found effective in reducing the incidence of gray and blue molds in apple fruits. It was observed that coating of mandarin fruits with pure coconut oil (100%) enhance their shelf life, quality and also delayed the mold appearance (Nasrin et al. [46]).

Recently, a new approach of edible coating "layer by layer coating (LBL)" is getting attention which is an electrostatic deposition technique. It is worked by combining the chitosan with other polysaccharides, like carboxymethyl cellulose (CMC). The aim of LBL was effective control the properties and functionality of material by depositing oppositely charged polyelectrolytes [47]. LBL edible coating including the five nanolayers of pectin and chitosan exhibited better quality in terms of weight loss, total soluble solids and titratable acidity in Tommy Atkins' mangoes [48]. Arnon et al. [49] reported in many citrus fruits (mandarins, "Navel" oranges, and "Star Ruby" grapefruit) that bilayer coating with CMC/chitosan slightly maintained fruit firmness. It was reported in strawberry that coating based on chitosan and carboxymethyl cellulose (CMC), (1%) found effective to prevents the loss of firmness and aroma volatiles and it also reduces the primary (involved in carbohydrate, amino acids and fatty acids metabolism) and secondary metabolites (involved in carotenoid, terpenoid, phenylpropanoid and flavonoid metabolism [47].

7. Irradiation and LED light

Fresh fruits and vegetables contain around more than 80–90% moisture. The production of fruits and vegetables has significantly reached to beyond the desired level. Despite, a significant portion of these fruits is getting spoiled due to attack of different micro-organisms during harvesting, handling and storage. Several attempts have been made to control of these microbial population. But due to risk of health hazards, nonchemical approaches emphasized over chemical methods. Food irradiation is one of the major nonthermal methods to control the disinfestations. This is a cold treatment which is highly effective against fungal, bacterial and molds. This process involves the use of ionized radiations like gamma rays, X-rays and electron beam over the food surface. Food and Drug Administration (FDA) permitted the maximum dose limit of 1 kGy for fresh fruits and vegetables [78]. Irradiation can help in delaying of ripening, reduced fruit firmness, reduced rate of respiration and ethylene and lower enzymatic activities which extend the shelf life of fruits like mango, pear, peach, strawberry, Nagpur mandarin, acid lime, etc. [50–53]. Irradiation is also helpful against many quarantine pests like *Bactrocera dorsalis* in papaya, fruit fly and stone weevil in mango [54, 55]. It was also reported that a dose of <150 Gy is sufficient to control tephritid fruit flies [55]. Similarly, Bustos et al. [56] reported that a dose of 100 and 150 Gy are enough to kill third instar larva of Mexican fruit fly and Mediterranean fruit fly in mango as a quarantine treatment. Ladaniya et al. [53] found that the irradiation dose of 1.5 kGy was delayed the Penicillium rot in Nagpur mandarin with higher total soluble solids.

Now a days, lighting based on light emitting diodes (LEDs) is one of the main emerging technologies in horticulture to enhance quality and inhibit diseases in fruits and vegetables after harvesting. LBL able to induce disease resistance in different fruit crops such as in citrus fruits against *P. digitatum*, when fruits were exposed to LBL for 3 days with 70 l mol m^{-2} s^{-1} (Ballester and Lafuente [57]), in *Vitis vinifera* against *B. cinerea* at 80-mol m^{-2} s^{-1}(Ahn et al. [79]). LED blue light induces fruits ripening by ethylene production in strawberry, in citrus fruits (Ballester and Lafuente [57]), in peaches at 40 mol m^{-2} s^{-1} (Gong et al. [58]) and in banana at 464–474 nm (Huang et al. [59]). Likewise it was found that LED light had positive effect on the quality in terms of the accumulations of ascorbic acid content, total sugar and phenols in banana (Huang et al. [59]), increase total sugar content, total phenols ascorbic acid and enhances antioxidant enzyme activities (catalase, superoxide dismutase and ascorbate peroxidase) in strawberry (blue (470 nm) light at an intensity of 40-mol m^{-2} s^{-1}) (Xu et al. [60]).

8. Use of bio control agents

All fruits and vegetables are prone to fungal and bacterial infection during storage. Due to postharvest microbial infection a significant part of fresh produce is lost during the handling, transportation and storage [80, 81]. The high moisture content and injuries make them more perishable and susceptible against microbial spoilage. Some postharvest diseases cause major breakdown in whole bulk and reduce the value of produce. The major postharvest diseases include soft rot, gray mold, anthracnose, stem end rot, blue mold, green mold, etc. that may cause huge loss. Several chemical and nonchemical approaches implemented to reduce the above said infection and to control of diseases. However, nonchemical approaches are getting more attention including use of essential oils, plant extracts and other plant based fungicides, use of bioagents. The use of bioagents is more helpful and ecofriendly approach in this line which has host specific mechanism. In this food safety line, several products were made by isolating different microorganism which as parasitic mechanism against wide range of disease causing harmful microorganisms. Some of the commercially available bioagents formulations are given in the **Table 2**.

Uses of some safe bioactive compounds have been proved beneficial in bringing down the physiological activities of fruits during transportation, storage and minimizing the overall qualitative and quantitative losses. Many antagonist species have been identified and inoculated over various fruit surface to control several disease causing microorganisms. It was reported that *Bacillus subtilis* can be used to control green mold in citrus and brown rot of stone fruits [61, 62]. Another species *Trichoderma harzianum* act as antagonist against gray mold of strawberry, grape and anthracnose of banana [63, 64]. The use of present natural mechanism of disease control could be proven a better alternatives without harm to environment and human.

Name of products	Available commercial formulation
Serenade	*Bacillus subtilis*
Messenger	*Erwinia amylovora*
Biosave	*Pseudomonas syringae*
Aspire	*Candida oleophila*

Above formulations are on the basis of availability in the market with their commercial formulations.

Table 2.
Some common commercially available bioagents formulations.

9. Cold plasma technique (CPT)

This is a nonthermal technique that has many applications in food industry. It reduces the pathological load and deactivates the enzymatic reactions thus enhances the shelf life. The most of research work has been carried out to find out the effect of CPT on microbial decontamination rather than the quality aspect [65]. Plasma generates an electromagnetic energy which ionizes the gases, however, the energy generates by the CP is different for different purposes like packaging, plastic and polymer industries. In food technology, it is widely used for surface decontamination that is achieved by placing the foods in strong electric zone resulting generates of reactive gas species that could alter the quality and sensory attributes [65].
CP technique has great impact on the quality of fruits and vegetables. It increase the sucrose content whereas reduces the glucose and fructose and this might be

attributed to high degree of polymerization in cashew apple juice (@ PE-100, 80 kHz, N_2, 10–50 mL/min, 5–15 min, 30 kPa) [65, 66]. CP has no significant effect on ascorbic acid content of the fruits and vegetables in many fruits like kiwi [67].

It was observed that exposure of CP increases the anthocyanin content in some fruit juices such as sour cherry (Garofulić et al. [68]), pomegranate juice (at 25 kHz, Ar, 0.75–1.25 dm^3/min for 3–7 min) Kovačević et al. [69]. CP treatment (dielectric barrier discharge, at 15 kV for 10–20 min) has also helpful in color retention and reduction of darkened area formed during storage of cut kiwifruit, though an immediate effect of CP treatment was slight loss of pigment which might be due to the degradation of pigments like chlorophyll and anthocyanin (Ramazzina et al. [67]). Won et al. [70] reported that CP treatment (at 2.45 GHz, 900 W, 1 L/min, 0.7 kPa, N_2, He, $N_2 + O_2$ (4:1) for 10 min) increases the antioxidant activity, total phenolic content and also inhibits *Penicillium italicum* in mandarin.

10. Conclusions

The present available technologies are getting popularity and commercial potency owing to their effectiveness, safe, cheaper, wide range and easier application methods. However, their effectiveness depends on many factors viz. formulations, nature of products and applied methodology, etc. Application of novel hormones like BRs, OA, MeJA, etc. reduces the spoilage, incidence of disorders and increase quality of produces. Use of edible coatings and some recent methodology like layer by layer coatings and their combinational effects are much more helpful in extending the shelf life of produce. Essential oils and plant extracts also found helpful in reducing the incidence of certain postharvest diseases and disinfections. Now a day, exposure of fruits to LED light could be effective in inducing phenols, antioxidants and delaying senescence process. Additionally, antimicrobial compound and some nanocomposite materials could be applied over the surface. Natural antagonistic mechanism through the application of bioagents is really helpful in maintaining ecological balance along with effective disease management. Some, newly induced technologies like cold plasma and irradiation is also helpful in minimizing disinfestation against quarantine pests and increasing the quality. So, overall effects of these technologies solely or in combination could reduce the postharvest losses and preserve the quality during supply chain management. However, there is great scope left to be carried out research and invention of new technologies with higher efficiency, ecofriendly and having broader spectrum against different postharvest losses.

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Section 2

Abiotic Stress Management

Chapter 3

Vineyard and Olive Orchard Management to Maintain Yield and Quality Under Abiotic Stress Conditions

Manuel Oliveira and Anabela Fernandes-Silva

Abstract

Yield and quality of fruits are multifaceted traits involving various plant and fruit processes that, for a given genetic makeup, depend on environmental factors and agronomic practices. Crop yield has to meet the demand of a growing population, but crop quality is a challenging issue affected by consumer's behavior and increasingly associated with food security. The projected climate scenario for South Mediterranean Europe predicts lower precipitation and higher temperatures that will negatively affect agricultural activity. A warmer and drier climate is expected to cause changes in crop yield and its quality. Higher temperatures affect photosynthesis, causing alterations in sugars, organic acids, flavonoid contents, firmness, and antioxidant activity. Reduced soil water availability will impact on the capacity of plants to accumulate biomass and when conjugated with warmer weather, it can trigger disorders like fruit sunburn further depressing crop yields. New cultivation techniques are necessary to produce sufficient food supplies to meet the basic nutrient requirements of the growing human population and support the agriculture economy. We focus on the production of olives and wine grapes, two of the most cultivated fruit crops of Southern Europe, which is certainly strongly affected by changing weather conditions. We review the recent developments on agronomic practices to counter or minimize the projected environmental changes, and we will report on our own experiences.

Keywords: abiotic stress, temperature, drought, climate change, grapevine, berry quality, olive tree, olives quality, olive oil quality

1. Introduction

Fruit production is a major agricultural activity all over the world. In 2016 the global production was above 33×10^6 metric tons and occupied about 5.6×10^6 hectares [1]. According to a survey in 2013, 1.55 million holdings in the European Union (UE) managed fruit orchards, and in 2015 3.2 million hectares were dedicated to fruit growing that represented more than 28×10^6 euros in 2018 [2]. In 2017, the world area under vines rose to 7534 kha, and grape production reached 73 million tons most of it for wine production estimated at 279 million hectoliters in 2018 [3]. Europe is a leading producer of wine grapes with 3.2 million hectares under vines worth of almost 22 billion euros of wine exports [2]. In 2018, the global production of table olives was estimated in 1.8×10^6 tons (EU 0.9×10^6) and 3.1×10^6 tons of olive oil (EU 2.2×10^6), representing a trade value of 2.8×10^6 euros for olive oil (EU 2.1 million euros) [2]. These figures render the importance of fruit production on a global scale that is likely to increase in the near future in tandem with larger demand for food, fiber, and fuel as a result of growing population, change in dietary preferences, and bioenergy policies [4]. The food demand driven by a larger and more affluent population challenges the agricultural systems to increase the output with reduced external inputs and minimal environmental impacts [5, 6] all under more difficult environmental conditions given the forecasts of an increased temperatures and more irregular rainfalls that negatively affect agriculture [7], and the Southern Europe will be strongly affected where yield losses and impaired product quality are expected [8, 9].

2. Abiotic stresses: temperature and water availability

Average temperature is expected to rise somewhere between 1.5 and 4°C from pre-industrial time until the end of the century [7]. Temperature increase affects photosynthesis, causing changes in concentration of sugars and organic acids, flavonoid contents, fruit firmness, and antioxidant activity [10].

Some unripe fruits with green skin can photosynthesize, but the fruits are primarily a sink of photosynthetic products with origin in the leaves. The temperature of the leaves follows approximately the air temperature, and net photosynthesis increases with temperature up to a certain limit that is species dependent, but in general at values greater than 35°C, there is a reduction of photosynthetic activity [11]. Temperature has strong influence on leaf water potential (ψl), stomatal conductance (gs), and intercellular CO_2 concentration [12]. Rising temperatures increase the water loss from the leaves, and their water potential becomes more negative to a point the stomata start closing reducing stomatal conductance and the flow of intercellular CO_2 that further impairs photosynthesis [13]. High temperature also diminishes starch and sucrose synthesis, by reduced activity of sucrose phosphate synthase, ADP-glucose pyrophosphorylase, and invertase [14]. Heat stress can reduce the total leaf area of the plants and trigger earlier leaf senescence that have a negative impact on the total photosynthesis performance [12]. Prolonged periods of low photosynthetic activity deplete reserves of carbohydrates, and plants might starve [15].

Soluble sugars (sucrose, glucose, and fructose) and organic acids (tartaric, malic, and citric acids) are major osmotic compounds that accumulate in fleshy fruits [15]. The biosynthesis of these compounds is related to photosynthesis that uses light (solar radiation) as source of energy, and it has been shown that the photosynthetic rate is positively correlated with light intensity till a saturation point is reached depending on the plant species and on temperature [12]. Increased light intensity also raises the temperature, and, above a critical level, protein and enzymes are broken, photosynthetic tissue is killed, cells might die, and fruits are sunburnt, all resulting in yield loss and low-quality produce [16].

At molecular level, temperature influences the protein structure, and the activity of cellular proteins becomes less stable under high and low temperatures indirectly affecting the activity of transmembrane transporters necessary to import assimilates and nutrients and to accumulate sugars and polyphenols in the fruit for the completion of the maturation process [17].

Fruits and vegetables contain different antioxidant compounds such as vitamin C, vitamin E, carotenoids, and polyphenol compounds. Among the polyphenols, flavonoids (flavanols, flavonols, and anthocyanins) are largely present in plants, and their content is partially responsible for antioxidant activity. Temperature is the most significant factor affecting antioxidant activity in vegetables and fruits, and, as temperature raises, the enzyme reactions are accelerated, antioxidant activity is augmented, and existing antioxidants decline [18].

Stressful conditions related to high temperatures are coupled with water availability that can mitigate or aggravate the effects of temperature. The increase in water demand when temperature rises is driven by plant transpiration necessary just to keep their canopies cool by water evaporation. Thus, water demand will increase at the same time as rainfall is scarcer and irregular in its frequency; therefore, an efficient use of water is particularly important for agriculture, which is the major sector for the use of freshwater resources and whose economic viability is dependent on water availability. Water uptake from the soil brings mineral nutrients that are circulated together with organic nutrients through the vascular tissues of the plant as water circulates throughout the plant. Water retention determines cell turgor, drives plant cell expansion, and permits many plant functions such as stomatal movements and transpiration [19]. Water is the abiotic factor that exerts a major effect on growth and productivity of agricultural crops, and increasingly frequent periods of drought, particularly in the Mediterranean region, is expected to be the most adverse among the abiotic factors [20].

Moderate drought reduces yields but can have a beneficial effect in fruit quality via two major mechanisms: (a) reduction in leaf stomatal conductance that results in a decrease in net photosynthesis and (b) exacerbation of oxidative stress/oxidative signaling. Net photosynthesis is responsible for primary metabolites that are the major source of precursors for the biosynthesis of phenolic compounds, carotenoids, and ascorbate. Oxidative stress may trigger the biosynthetic pathways of these compounds [21, 22]. The balance between productivity and quality benefits from a certain level of water deficit, but it depends upon the intensity, duration, and repetition of events of water deficit [23]. Water deficits during the vegetative growth stages difficult canopy development, flowering and fruit setting, and the accumulation of carbon compounds. On other hand, deficits occurring during the maturation of fruits show positive impacts on soluble sugar accumulation and enhancement of fruit aroma. The level of stress that benefits the entire cycle of the plant is likely to be depend, simultaneously, on species and environmental conditions, but when a certain threshold of water stress is surpassed, the beneficial effects are no longer observed [23, 24].

3. Wine grape quality

The fruit quality is a composition of physicochemical properties and the perception of the consumer allowing for a definition of quality that discriminates between natural characteristics of the fruit and those dictated by socioeconomic and marketing factors [25, 26]. The quality of grapes (*Vitis vinifera* L.) for winemaking is evaluated on characteristics imparted to the final product, and as such their quality is referred to the berry composition, in particular, to the content of sugars, organic acids, amino acids, phenolics, and aroma precursors that are function of genotypic, environmental, and agronomic factors [25, 27].

After fruit setting, the berry pericarp and the seeds augment in volume rapidly; organic acids, mostly malic, tartaric, and citric, accumulate in the mesocarp cell vacuoles. At the end of this period, growth slows down, and the seed maturation is completed. The maturation phase initiates at veraison, when berries of red varieties accumulate anthocyanins (red pigments) in the exocarp, glucose and fructose accumulate, malate is metabolized as a source of carbon for respiration, and volatile organic compounds are biosynthesized [28].

Abiotic stress, temperature, radiation, and water changes the pattern of growth and development of vines, and they trigger the accumulation in berry pulps, seeds, and skins of secondary metabolites (polyphenols and volatiles) as defense against cell damage [29]. The secondary metabolites present in the must contribute to wine characteristics as taste, aroma, antioxidant capacity, and stabilization during the aging process. Thus, the vineyards are managed to influence the profile and concentration of secondary metabolite to obtain the desirable wine typicity.

The Mediterranean-type climate has long warm periods well suited to growth and development of grape vines, but that period is also low in rainfall that can create serious lack of water availability. The wine regions of Northeastern Portugal are distinctively Mediterranean and have already shown the alterations in temperatures and rainfalls expected under the forecasted climatic change. Annual rainfall has been declining since the 1950s, winter temperatures are milder, and average temperatures for the grape growing season are reaching the upper limit for producing high-end wines [30, 31]. To maintain the grape quality desirable to meet the demand of a world market will be a challenging task involving the best suitable varieties and agronomic practices supported on knowledge gained by experience and research.

A clearly noticeable phenomenon resulting from higher temperatures is the advance of the phenological stages of the grapevines, with a short growing season leaving the maturation to develop under hotter and drier conditions [32, 33]. Berries maturing under high temperature have reduced content of anthocyanins [34] that are hydrophilic secondary metabolites that confer orange, red, or blue coloration to grapes and help protecting the plant tissues against abiotic stressors such as high radiation. A lower content of anthocyanins has a negative impact on the color of red wines, an important organoleptic characteristic. Sugar accumulation in the berries increases as the temperature rises, but there is a sharp fall in acidity and malate content creating an imbalance alcohol/acidity that is deleterious to high-quality wines [35, 36]. These phenomena are dependent on variety tolerance to high temperatures [35]; thus, the viticulturists must choose carefully before they start new plantations, reaching for a compromise between adaptability and quality. Usually, native varieties thrive better on a given environment than exotic ones even when conditions suffer large alterations that is the case of Touriga Nacional in Portugal, a valuable premium variety to produce high-quality wines, well adapted to regions with intense solar radiation and warm climates [37, 38], and the adaptability to different climatic conditions is a variety trait important to mitigate the effects of stressful factors on the berry quality [39, 40].

High temperatures increase evapotranspiration that coupled with declining rainfalls further deplete soil water availability, and this is actually the most pressing challenge to wine grape production in areas where aridity is advancing [41]. Nevertheless, berry quality benefits from moderate levels of water stress because skin-to-pulp ratio increases as the berries become smaller [24, 42], elevating the concentration of total soluble solids (mostly sugars) and secondary metabolites, such as phenolic compounds (especially anthocyanins) and aromatic compound (in particular norisoprenoids) whose biosynthesis is enhanced as response of the vine to water stress [43–45]. Higher content on sugar and secondary metabolites in berries will produce wines higher in alcohol, deeper in colors, and richer in aromas but also lower in titrable acidity as organic acids, mainly malic and tartaric, are depleted [24, 44].

Temperature and water stresses have different outcomes on yield and berry quality depending on their intensity and timing related to the development phase of the vine. The vineyard manager can use this knowledge to manipulate agronomic practices and obtain the desirable yield and berry quality with the most efficient use of resources, in particular, water. New vineyards must be planted with known varieties to be well to actual conditions but also with plasticity to withstand more stressful conditions; the vine rows trellised at vertical shoot position are to be oriented north to south where the terrain permits and the location carefully chosen to avoid the most severe effects expected to be brought about by coming changes. In vineyards already under commercial exploration and that are expected to have a life span of a few decades there are technical practices that can reduce the worst effects of excessive temperature and radiation, and of low amount of available soil water.

The most common result of high-intensity radiation, usually coupled with elevated temperatures, is the shriveling and sunburn of berries, reducing both yields and quality of the musts. The incidence of these phenomena is felt more acutely in vineyards where the rows are oriented west to east and one of the faces (south face in northern hemisphere) is directly illuminated all day long. One solution is to shade the lower third of the canopy with a vertically placed net close to the canopy during the period when intense radiation and its effects are expected to cause damages. Shaded vineyards maintain much higher yields than non-shaded ones but at the cost of lower concentration of anthocyanins, and their musts render wines with lighter color that can be considered detrimental to its quality [46].

Other techniques that can maintain higher photosynthetic activity and simultaneously reduce the incidence of berry sunburn are canopy coating with kaolin and intermittent nebulization with water at high pressure. They reduce plant temperature and allow for higher stomatal conductance and, consequently, increased photosynthetic rate that contributes to larger berry sugar content [33, 47]. Treated plants keep better yields, produce berries with higher concentration of sugars, and show no significant difference on other must characteristics [33].

Irrigation is already common in many wine-producing regions in Southern Europe, the Western United States, and Australia, among others, and with expected reduction in rainfall, it will be indispensable to maintain the economic activity. The challenge is to keep a balance between productivity, quality, and efficiency in the use of resources, especially water. Moderate water deficits at optimized timing can improve berry quality and support economical yields [40, 48]. Better water use efficiency was reached with no irrigation at all, but the productivity is so low that economically is not viable [49]; irrigation increases the yield but dilutes sugars and compounds responsible for color and aroma rendering a wine of lower quality. An acceptable compromise is to deficit irrigate from flowering to veraison, a phase very sensitive to severe water shortage, and no irrigation after veraison assuming that the soil still stores enough water to dispense up to the completion of the vine cycle. The effects of irrigation on berry composition are subjected to controversy with contradictory results among authors, but the source of these inconsistencies is probably related to the climatic conditions prevalent during the studies that each author carried out [50].

Under increasing competition for scarcer water resources, the imposition of moderate water stress on certain developmental periods of wine grapes is an adequate irrigation strategy to save water and maintain both yields and must quality with positive reflexes on the farm economy. Such strategy, termed by some authors as regulated deficit irrigation (RDI), has received great attention by researchers and producers [51]. Drip irrigation, from aboveground supply lines, is the most efficient form for vineyards, particularly for RDI, as it minimizes runoff and evaporation while delivering water uniformly and directly to the root zone; significant amounts of water are saved, and water contact with the plants is avoided, reducing the risks for disease development [52]. RDI with deliver lines buried underground is an expensive alternative with little benefits to offset its high costs [53].

Recent technical advances permit a more affordable and widespread use of phenomics defined by Houle et al. [54] as "a sub-discipline of biology concerned with the rapid measurement of an organism's phenotype or physical and biochemical make-up." Phenomics can help growers and managers to survey their crops to quantify spatial variability in fruit quality, yield, soil characteristics, and incidence of diseases, among other parameters, with many benefits for the more efficient use of resources and forecasts of yields [55, 56]. One example of such survey is presented by Rossi et al. [57] with the integration of soil spatial information of soil electrical resistivity, obtained automatically with a soil sensor on the go, with variation of vegetative growth and yield permitted to identify areas of a vineyard with similar traits. These areas would be subjected to differentiated agronomic practices to maximize potential benefits in yield and quality and, simultaneously, increase the efficiency of used resources.

4. Olives and olive oil quality

Climatic conditions and different agronomic practices may influence the physiological behavior of the olive tree [58] and consequently the fruit ripening process modifying both the amount and oil quality in *Olea europaea* L., although the response is cultivar dependent [59–61].

Olive oil quality may be defined from commercial, nutritional, or organoleptic perspectives. The overall quality of olive oil, from production to consumption, is strongly related to oxidative stability and its impact on the evolution of flavor, taste, color, and the content of endogenous antioxidants and other minor constituents that are beneficial to health. The International Olive Oil Council [62] and the EEC [63] have defined the quality of olive oil based on parameters that include free fatty acid (FFA) content, peroxide value (PV), ultraviolet (UV)-specific extinction coefficients (K_{232} and K_{270}), and sensory score. In particular, commercial quality is based on FFA as an important factor for classifying olive oil into commercial grades and sensory characteristics (taste and aroma). The nutritional value of olive oil arises from high levels of oleic acid and minor and health-related and antioxidative components such as phenolic compounds, tocopherols, chlorophyll, and carotenoids [64], whereas the aroma is strongly influenced by volatile compounds [65].

Fatty acid composition is one of the primary chemical parameters used to distinguish virgin olive oil from other vegetable oils [66]. Fatty acid profile may be greatly affected by environmental factors. Variability in acid composition has been correlated to the temperature sum of the period from fruit setting to fruit maturation by regulating fatty acid desaturases [67]. In fact, it has been reported that the contents of oleic acid decreased during ripening, while that of linoleic acid increased due to the transformation of oleic acid into linoleic acid by the oleate desaturase activity, which is active during triacylglycerol biosynthesis [68].

Despite the response being cultivar dependent, it has been shown that high temperatures during the maturation of olive fruits, early in the triacylglycerol biosynthesis, reduced oleic acid which is accompanied by increased palmitic and/or linoleic acids [69, 70]. In cv. Arauco, García-Inza et al. [71] observed that oleic acid concentration decreased linearly 0.7% per °C, while palmitic, linoleic, and linolenic acid percentages increased with increasing temperature. In cv. Arbequina, Rondanini et al. [72] reported a higher reduction of oleic acid with high temperatures (2% per °C).

Solar radiation and water availability are crucial not only for tree productivity, [60] but also they clearly affect olive oil quality. In cv. Frantoio, palmitoleic and linoleic acids increased in oils obtained from fruits exposed to high solar radiation intensity, whereas oleic acid and the oleic-linoleic acid ratio decreased [61].

To overcome the negative effects of combined heat and high radiation stresses, application of kaolin in olive trees growing in rainfed conditions has been used. The kaolin coat film could reduce solar radiation damage; reduce heat stress by reflecting UV light, decreasing leaf temperature, and reducing transpiration rate; increase photosynthetic efficiency in plants grown under high level of photosynthetic active radiation; and reduce heat caused by radiation [73, 74]. Khaleghi et al. [75] evaluated the effect of kaolin application in rainfed olive orchard, and they found that the highest palmitic acid was observed in olive oil obtained from untreated trees and that kaolin increased oleic acid but decreased linoleic and linolenic acid contents. Also, the percentage of monounsaturated fatty acids (MUFA) and oleic acid/linoleic acid ratio were higher in the oil obtained from trees treated with kaolin than that obtained from untreated trees. Moreover, saturated and polyunsaturated fatty acids (PUFA) were higher in untreated trees. Therefore, it can be expected that extracted olive oil from kaolin-treated trees has a higher oxidative stability and shelf life than the oil from untreated trees.

The ratio of unsaturated/saturated fatty acids influences the viscosity of oils, increasing with the amount of saturated fatty acids. This has an effect on the sensation of "fatty" on the oral cavity as a viscose oil has more time in contact with the mucous membranes of the oral cavity, giving rise to the "fatty" defect [76]. Moreover, the degree of unsaturation of fatty acids affects the oxidative stability. A high degree of unsaturation of a fatty acid increases the susceptibility to oxidation and shortens the olive oil shelf life [77]. Dag et al. [78] reported that in cv. Koroneiki, the monounsaturated/polyunsaturated fatty acid ratio and free fatty acid content generally decreased with the increased tree water deficit. Besides, olive oil oxidative stability might depend on some synergistic effects among fatty acid composition, phenolic compounds, tocopherols, carotenoids, and chlorophylls [79].

A water deficit during the initial development of the fruit can result in a decrease in the size of the cells of the mesocarp that cannot be recovered. Water deficit affects fruit maturation, which occurs earlier and more rapidly, and can result in more intense preharvest fruit fall [80]. However, a number of studies have shown that the water status of the plant has marginal, if any, effects on free acidity and peroxide value of the olive oil produced [81, 82].

Minor constituents of olive oil, such as phenolic and volatile compounds, are also influenced by the degree of maturation of the olive fruit. So, environmental factors or agronomics practices that affect the evolution of maturation of the drupe can also affect the qualitative characteristics of the resulting olive oil [83]. For example, a very high temperature sum also tends to reduce the total polyphenol content [84]. A positive correlation between the temperature sum from August to October and the total polyphenol content of olive oil was reported by Tura et al. [85].

It has been recognized that polyphenols and tocopherols are substances with natural antioxidant properties and their presence in olive oils has been associated to their general quality, improving stability, nutritional value, and sensorial properties. In cv. Cobrançosa, Fernandes-Silva et al. [82] reported a good linear relationship between total polyphenols and water stress integral. Therefore, olive trees that had been exposed to a certain level of water deficit produced oils with higher concentrations of polyphenols which were seemingly richer in the olive fruit [86, 87].

Virgin olive oil (VOO) obtained from rainfed olive orchards shows the highest resistance to oxidation in relation to irrigated olive orchards as a result of higher values of oxidative stability. The decrease in oxidative stability with water applied by irrigation is usually explained by the decrease in natural antioxidants like polyphenols and tocopherols [82, 86]. Given this assumption, several researchers have tried to find which of the mentioned substances is more correlated with oxidative stability. A number of studies have demonstrated that polyphenol contents are, among the minor compounds, the group more correlated with this parameter [88]. The antioxidant behavior of tocopherols represents a complicated phenomenon as they are efficient antioxidants at low concentrations, but they steadily lose efficiency as their oil content in the vegetables increases [76].

Olive tree water status has marked effects on concentrations of volatile compounds in the oil. Thus, olive oil from plants grown under water deficit-conditions can be bitter and pungent to the taste in opposite to those obtained in well-watering conditions [81, 89]. Williams and Harwood [90] have clearly shown that drought regimes, in Crete, reduced the relative activity of enzymes of lipoxygenase pathway and consequently the volatile compounds.

Regulated deficit irrigation (RDI) in olive orchard is an agronomic practice in which plants were irrigated avoiding water deficit during phases I and III of olive fruit growth and saving water during phase II, the noncritical phenological period of pit hardening [91]. This strategy of irrigation can affect some table olives' characteristics, for example, phenolic composition, antioxidant activity, fatty acid composition, volatile compounds, and phytoprostanes [92]. Table olives from RDI belong to a group of vegetable products named *hydroSOStainable* which are characterized by having distinctive proprieties such as high content of some nutritional and functional compounds, high intensity of sensorial attributes, and are produced with reduced use of water, which is a benefit for both farmers and for the environment [93]. Sánchez-Rodríguez et al. [93] reported that *hydroSOStainable* table olives (cv. "Manzanilla") showed the most attractive shape and color with highest fruit weight, roundest fruit, hardest texture, and a lightest and greenest color than control olives, whereas minerals, antioxidants, phenols, and organic acids and sugars of *hydroSOStainable* olives were similar to well-irrigated olives. Hence, *hydroSOStainable* table olives have advantages over those obtained in well-watered conditions reducing the use of freshwater, while they have better morphological traits that are more attractive for consumers.

5. Conclusions

Lower latitudes of temperate regions are expected to experience climate changes in coming decades that will bring about conditions less favorable to agriculture activities. Yields are likely to decrease, and the quality of produce might suffer a negative alteration. Commercial wine vines and olive trees are very sensitive to their environment, and to keep their economic value, it is necessary to adopt agronomic practices to minimize the adverse effects of climate change. The less favorable location for their growth and development might be abandoned, the choice of varieties to plant will be selected among the best adapted to future conditions, and management techniques of highly efficient irrigation, shading, spraying with reflecting materials, and tight control of canopy development, among others, will have to be commonly adopted.

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Conflict of interest

We report no conflict of interests and no other benefit from our work.

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Section 3

Berries - Recent Advances on Growing and Breeding Strategies

Chapter 4

Soil Preparation, Running Highbush Blueberry (*Vaccinium corymbosum* L.) Plantation and Biological Properties of Fruits

Zofia Zydlik, Szymon Cieśliński, Van Chung Mai, Nesibe Ebru Kafkas and Iwona Morkunas

Abstract

Due to the pro-health properties of highbush blueberry fruit, the interest in cultivation of this species has been growing significantly, which is evidenced by the current increase in world fruit production. Therefore, the aim of our review study is to present the impact of soil and climatic conditions and cultivation methods of *Vaccinium corymbosum* L. on fruit yield and quality in Central and Eastern Europe. In this region, one of the most important abiotic factors determining the yield level is the minimum temperature of the winter period and short-term increases in temperature, which are conducive to the damage to flower buds. Another factor determining the success of cultivation is soil. In addition, highbush blueberry has specific soil requirements, which result from its characteristic root structure. The adverse impact of soil factors can be mitigated to a certain extent by the use of mycorrhizal fungi. In this chapter, besides the cultivation conditions of *V. corymbosum*, the pro-health properties of fruits resulting from the presence of bioactive compounds such as polyphenolic compounds, flavonoids, especially anthocyanins, will be presented. Besides, factors, such as environmental conditions, degree of ripeness and variety, affect the content of bioactive substances.

Keywords: highbush blueberry, growing conditions, biologically active compounds

1. Introduction

Blueberries are perennial flowering plants with blue- or purple-colored berries. They are classified in the genus *Vaccinium* within the family Ericaceae (a family of flowering plants, commonly known as the heath or heather family). Blueberries are usually prostrate shrubs that can vary in size from 10 cm to 4 m in height. In commercial production of blueberries, the species with small, pea-size berries growing on low-level bushes are known as "lowbush blueberries," while the species with larger berries growing on taller cultivated bushes are known as "highbush blueberries." Due to the pro-health properties of fruit of highbush blueberry plants (*Vaccinium corymbosum L.*), the interest in cultivation of this species has been growing significantly.

Blueberries may be cultivated, or they may be picked from semiwild or wild bushes. Successful blueberry harvest requires attention to several cultivated factors.

In Central and Eastern Europe, one of the most important abiotic factors determining the yield level of highbush blueberry is the low and short-term increasing temperature in the winter, which are conducive to the damage to flower buds. Another factor determining the success of cultivation is soil moisture; water shortage results in a deterioration of the condition of shrubs as well as a decrease in quality and yield of fruit. In addition, highbush blueberry plants have specific soil requirements, which result from their characteristics of root structure. The adverse impact of soil factors can be mitigated to a certain extent by the use of mycorrhizal fungi.

Fruit of blueberries are believed to have high level of antioxidants that are thought to be responsible for much of the berries' beneficial health effects. The pro-health properties of fruits resulting from the presence of bioactive compounds such as polyphenolic compounds, flavonoids, especially anthocyanins, in the fruits of *V. corymbosum* as well as effects of factors, such as environmental conditions and degree of ripeness in the content of bioactive substances have been recorded.

This work presents the most recent state of knowledge about running a highbush blueberry plantation and properties of blueberry fruits with a wide range of biological effects that significantly improve the condition of human health.

2. Cultivation of blueberry in Central and Eastern Europe

Poland is the sixth largest producer of blueberry fruit in the world, the second largest among the European countries and the first in the group of countries of Central and Eastern Europe (FAO STAT, IBO-International Blueberry Organization). **Table 1** presents the cultivation area and the volume of blueberry fruit production in countries of Central and Eastern Europe in 2017.

The success of highbush blueberry fruit production depends on atmospheric conditions (especially on factors such as temperature and precipitation) during the growing season and during the winter resting period of the plants. Warm and dry summers are not conducive to good budding, and sudden temperature drops in winter can cause them to freeze. The climate in Central and Eastern Europe is generally considered to be favourable for the cultivation of most varieties of highbush blueberry. However, when planning the establishment of a plantation, it is necessary to analyse various climatic factors that may have a significant impact on the success of the cultivation. One of the most important factors that should be taken into account is the minimum winter temperature. Long-term observations showed that temperature dropping down to −25°C did not affect the damage to blueberry shrubs. However, it should be remembered that not only the minimum real temperatures can be harmful to plants but above all the periodical warming occurring during winter, which contributes to the hardening

Country	Cultivation area (ha)	Harvested crops (tonnes)
Bulgaria	13	99
Latvia	236	353
Lithuania	85	124
Poland	5318	16,343
Romania	130	310
Ukraine	400	1360

Table 1.
Highbush blueberry cultivation area and harvest in Central and Eastern European countries in 2017 (FAO STAT) [1].

of plants and can increase their damage caused by frost [2]. Blueberry flower buds are recognised as the most sensitive to frost damage. The studies conducted by Panicker and Malta [3] show that the resistance of highbush blueberry flowers to frost damage is significantly dependent on the concentration of abscisic acid (ABA) and the exogenous use of phytohormone may significantly reduce losses. This is confirmed in studies by Huang et al. [4] which showed a significant increase in the resistance of sugarcane plants to cold stress after application of ABA. According to observations carried out in Belarus, after the period of frost, when the temperature dropped to −27°C, and then a 10-day thaw occurred with the air temperature during the day above +5°C, those factors resulted in a significant drop in yields and even contributed to a total lack of yields. The above observations indicate a certain regularity, that is, the higher the air temperature during the winter thaw and the longer its duration and the stronger the frost after this period, the lower the blueberry crops [5]. Frosts can also be particularly harmful to blueberries in autumn and spring periods. Frosts occurring during the flowering period of shrubs are particularly dangerous. The practice shows that a drop in temperature to −7.5°C can damage up to 50% of flowers, which results in a considerable reduction of yield. In addition, blueberry growth is inhibited when the temperature drops to +3°C. On the other hand, the temperature ranging from 8 to 20°C is considered optimal for all biological processes taking place in the plant. Higher temperatures in summer foster early ripening of the fruit. On the other hand, at the temperature of +24°C, the process of flower bud formation proceeds faster [6]. In order to protect the flowers against harmful effects of spring frosts, percoronal irrigation and wind machines are used at blueberry plantations. Another threat to blueberry cultivation in Poland is hail occurrence during the vegetation period. One of the ways to reduce the risk of hail damage to plants is to use hail cannons. For example, hail cannons were installed on the farm of Jerzy Wilczewski from Białousy who cultivates high blueberry at an area of over 600 ha. The second important climatic factor influencing the success of blueberry cultivation is the length of the vegetation season. This period should last at least 160 days. In Central and Eastern Europe, it is sufficient, and it usually lasts from the last spring frosts to the first autumn frosts. The length of the growing season is the number of consecutive days on which the average air temperature stays above 5°C. The vegetation season is the time of enhanced growth and development of plants under the conditions of sufficient heat and moisture. In Poland, for example, the average length of the vegetation season for highbush blueberry ranges from 200 days in north-eastern Poland to 240 days in south-western Poland. In cooler regions of the country, it is recommended to choose varieties of blueberry that ripen earlier, because cooler July and August may delay the ripening of the fruit and may deteriorate its quality. It should be emphasised that high temperatures and the accompanying drought during the flowering of shrubs contribute to worse fruit formation, while during fruit development these factors will significantly reduce the level of yield and fruit quality [7]. High summer temperatures and low annual precipitation also contribute to the poor budding for the following year.

Precipitation is another factor that determines the success of highbush blueberry cultivation. Central and Eastern Europe have a dominating temperate climate with cooler, more humid north and warmer, drier south. It should be borne in mind that the amount of groundwater—the main source of drinking water and water used for irrigation of crops—fluctuates as a result of climate change effects and changes in groundwater levels are of seasonal nature, often leading to a water level drop of more than 0.5 m [8]. In Poland, the level of precipitation is suitable for blueberries only in the regions of Southern Poland and only if plantations are set up in areas with high groundwater levels [7]. Considerable water reserves stored in the soil in spring can satisfy the requirements of the plants during the short-term drought occurring in May and June. However, if the period of drought lasts longer, it can result in weakening or even drying out of a plant. Especially on light and poorly humus soils which do not have sufficient rainwater storage capacity, additional irrigation is necessary. It should be remembered that the depth of the main mass of blueberry roots is from 5 to 35 cm. The blueberry has the highest water requirement at the moment of intensive fruiting. Water shortage at that time usually results in a decrease in the volume and quality of the yield and a deterioration in the condition of shrubs; therefore a commodity plantation without an irrigation system poses a very high risk. One must also bear in mind that varieties derived from the *Vaccinium corymbosum* species do not tolerate cultivation on dry soils and most often require irrigation. Varieties originating from *Vaccinium angustifolium* are considered more resistant to drought [6]. Despite the common opinion that blueberries have high water requirements, one must remember that soil moisture should be stable [9, 10]. At plantations, water cannot be stored for long periods of time since plants can die if the soil is too moist (**Figure 1**).

In addition to specific water requirements, high blueberry also has specific settlement requirements. High blueberry grows best at quiet, sunny and warm positions. The neighbourhood of large water bodies which will mitigate temperatures during the year and the proximity of forests, providing protection against destructive climatic factors, has a good influence on cultivation. Blueberry plantations should never be set up in basins and recesses of the land, where cold air masses flow down creating frost stagnation. In the area where a significant lowering occurs in order to protect the blueberry shrubs from frost damage during the flowering period, it is necessary to establish a percoronal anti-frost irrigation intended to protect the flowers. Moreover, peat soils with a pH of 3.5–4.0 are best suited to the cultivation of this species in terms of soil requirements.

When choosing plantation locations, it is worth remembering that sandy soils are more suitable than heavy soils. This is confirmed in studies conducted by Cho et al. which have shown that clay soils are not useful for the cultivation of highbush blueberry in the Damyang Province, Korea [11]. It is important that the substrate has a

Figure 1.
High blueberry bush growing in a place flooded with water.

stable water content because the roots of the highbush blueberry are sensitive to both the excess and the deficiency of water [9]. It is worth remembering that like all heather plants of the Ericaceae family, the highbush blueberry has a root system consisting of very thin and numerous roots with a diameter of 17–50 μm without root hairs [2]. The lack of root hairs results in restrictions that plants have to cope with, for example, through symbiotic coexistence with mycorrhizal fungi. One of the most common types of mycorrhiza is endomycorrhiza, which is estimated to be found in almost 90% of vascular plant species. Thanks to this symbiosis, plants are better supplied with water, phosphorus, nitrogen and trace elements [12]. On the other hand, the plant provides mycorrhizal fungi with assimilates. Trophic influence of mycorrhizal fungi results from their decomposition of caries and making components in an assimilable form available to plants. A better supply of phosphorus to plants is the result of increased phosphatase secretion by fungi, which enables mineralisation of organic phosphorus compounds in the soil. This is particularly important in the case of weaker or degraded soils, where fungus cells penetrate root-inaccessible and remote areas of the soil through threaded shreds, extracting nutrients and passing them on to the plant. Proper nutrition additionally increases the plant's resistance to stress caused by drought or excessive salinity. It should be noted that mycorrhizal fungi—apart from trophic support—can protect plants against both abiotic stress (drought, inadequate acidity of the substrate, inadequate light intensity) and biotic stress (soil pathogens). The mycorrhizal mycelium surrounding the root hinders the penetration of pathogens, and the additional layer of lignin formed in cells increases their resistance to the effects of possible infection. Before deciding to use a commercially available mycorrhizal vaccine for heather plants, it is important to ensure that the soil is properly prepared. According to the research conducted by Zydlik et al. owing to the use of a mycorrhizal vaccine, it is possible, to a certain extent, to mitigate the adverse effect of pH on the biochemical properties of soil and plant yielding [13]. According to many authors, a positive effect of mycorrhiza on the growth, development and nutrition of heather plants growing on soils with inadequate pH and deficient basic nutrients was observed [14]. Mycorrhiza protects plants against abiotic stress (drought, salinity, temperature, pH) and biotic stress [15–17]. Mycorrhizal fungi facilitate transport of water and nutrients from the soil by the plant to the surface parts [18–20]. According to Smith et al. [21] and Hu et al. [22], the effect of mycorrhizal fungi decreases with a high content of nutrients, especially phosphorus in the soil. During the vegetation season, preparations improving soil properties can also be used [21, 22]. These are mostly preparations containing humus substances, which are recommended as an alternative to fertilisers in organic farms [23]. American authors pay special attention to the very high biological activity of humic acids secreted from leonardite and indicate that the content in organic materials (lignite, peat) is not as high as in the mineral [24–26].

Moreover, blueberry plantations are long lived and properly exploited and can produce satisfactory yields even for 30–40 years. Therefore, it is important to make every effort to prepare the position properly. The thorough removal of perennial weeds is particularly important. To that end, it is advisable to combine tillage (ploughing, deep loosening and harrowing of the fields) with the use of systemic herbicides. For this purpose, such agents are useful as, for example, the Roundup 360 SL herbicide in the dose of 5–8 l ha^{-1} or Glifoherb 360 SL containing glyphosate, which remains in the soil shortly, Chwastox Extra 300 SL in the dose of 3–3.5 l ha^{-1} and Starane 250 EC in the dose of 1–2 l ha^{-1}. It should be remembered that the listed herbicides are foliar systemic agents and are recommended for application to green grown weeds with a height of not less than 10–15 cm [27]. In addition, when preparing the soil for new plantings, it is worth paying attention to soil pests, which can pose a serious threat to the success of the crop. The most common are shoots, wireworms and swellings. The shoots damaging the blueberry roots are mainly larvae of the cockchafer (*Melolontha melolontha* L.). The full development of the pest from laying eggs by the females to the soil takes 3–4 years. While feeding, larvae eat small roots and bite the bark from thicker roots and from the root neck of blueberries, leading very often to weakening the growth of shrubs and in extreme cases even to their dying out. On the other hand, wireworms are larvae of the beetle family (Elateridae), the most common of which is the farmer's seeder (*Agriotes lineatus* L.). Harmfulness of wireworms in the case of highbush blueberries results from feeding on roots and drilling corridors therein, which significantly weakens plants. Adults of swellings (beetles) feed on leaves leaving characteristic crescent-shaped bites, while larvae feed on roots leading to their dying. The best period to assess the condition of soil pests is August or the first half of September, before setting up the plantation. In order to determine the risk, 32 diagonal points should be determined over the area of 1 ha. At these points, soil should be sampled from holes with the dimension of 25 cm × 25 cm and 30 cm depth (this corresponds to a field area of $2m^2$). The soil samples collected should be passed through a sieve, and the larvae of the pest species found shall be counted. The harmfulness threshold for blueberries is assumed at a level of 1 shoot and 1 wireworm and 10 swellings on the surface of 2 m^2 [9]. Before setting up the plantation, in order to limit the number of shoots, it is recommended to use a disc harrow to till the soil several times, at the turn of April and May and August, and a buckwheat tillage, which releases tannins that inhibit the development of shoots. Entomopathogenic nematodes such as *Heterorhabditis bacteriophora*, which are found in the Larvanem preparation, are used for biological control of these pests. The nematodes contained in the preparation get inside the larvae of swellings through the natural body openings (mouth, anus or lentils) and feed there. Specific bacteria are then released from the gastrointestinal tract of the nematodes, which disperse inside the insect body and multiply intensively. These bacteria break down host tissues into substances that can easily be taken up by nematodes. The larvae of a swelling die within several days. The infected larvae turn orange-red to reddish-brown. At present, there are no chemical agents for combating soil pests registered for use in blueberry cultivation. Producers in Georgia point to a new threat emerging at commodity plantations, which is the blueberry re-plantation disease. It may be caused by root nematodes (*Paratrichodorus*, *Pratylenchus*). Studies on the number of nematodes colonising the blueberry root system zone show that *Paratrichodorus renifer* and *Mesocriconema ornatum* species have the highest share in soil colonisation. The number of *Pratylenchus penetrans* species, a harmful rootstock, was definitely lower [28, 29]. One of the nematodecidic preparations approved for use is the preparation Vydate 10G containing oxamyl—it requires mixing with soil to the depth of 10–15 cm, preferably using a ripper. Nemasol 510 SL (active substance sodium methane) and Basamid 97 GR (dazomet) also belong to the group of nematocydes. These nematocydes are registered only for use on strawberry plantations. Nematodes should be combated before planting shrubs.

Another important issue in the preparation of soil for the plantation of highbush blueberry which is worth noting is the humus content of the soil. In the majority of Poland, the humus content in the soil is insufficient and usually ranges from 0.5 to 1.5%, which is definitely insufficient for high blueberry. The minimum level of humus in the soil for blueberry is 3%. Research carried out in 2015 showed that in 62.9% of soils, the humus content was 1–2%. The reason for low humus content in cultivated soils is its biological degradation. Low humus sustainability in cultivated soils is a result of intensive mineral fertilisation and mechanical treatment resulting in strong aeration of the soil. Soil caries are responsible, among others, for the tuberous structure of the soil, and the associated air-water relations increase the capacity of the soil sorption complex, thus reducing nutrient leaching to groundwater and improving soil retention properties. Humus substances are also attributed to a positive effect on the activation of microorganisms, improvement of soil physical properties, better absorption of nutrients by plants and stimulation of plant growth by supporting their life processes [30]. The most common sources of humus in the soil are plant residues, dead bodies of macrofauna and mesofauna as well as dead microorganisms and organic fertilisers in the form of manure or compost [31].

In order to increase the humus content of the soil, green fertilisers can be sown before setting up the plantation. For this purpose, it is recommended to use phytosanitary plants which show antagonistic activity towards harmful soil microorganisms and support the development of beneficial soil microflora. Phytosanitary plants include, among others, velvet (*Tagetes* L.), white mustard (*Sinapis alba*), oil radish (*Raphanus sativus* var. *oleifera*), spring rape (*Brassica napus*), oat (*Avena sativa*), rye (*Secale cereale* L.) and asparagus (*Asparagus officinalis* L.). It should be borne in mind that the action of phytosanitary plants is related to specific compounds produced by plants and released to the soil environment through roots or as a result of the biomass decomposition. For example, velvet has strong insecticidal and paralytic properties due to the rapid effect of pyrethrin on the nervous system. Velvet growing for at least 3–4 months is able to reduce the number of nematodes in the soil by up to 80%. Velvet is not only a valuable phytosanitary plant but also a rich source of organic matter and has a high fertilising value. The nematode killing effect of the velvet may also be important in reducing the population of *Pratylenchus penetrans* and *Paratrichodorus renifer* and *Mesocriconema ornatum*, which are considered as numerous parasitic species on heather plants, which may be one of the causes of blueberry replanting disease. The occurrence of these nematodes damages mainly young roots, limiting plant growth and causing a reduction in yield by 40–50%. White mustard seed (*Sinapis alba*) and oil radish (*Raphanus sativus* var. *oleifera*) also have a nematising effect [32, 33].

In addition, creating appropriate conditions for blueberry growth often requires lowering the soil pH. Soil acidification is usually carried out in two ways. The first one consists in delivering sulphur to the position and its shallow mixing with soil. For this purpose, dusty sulphur or Wigor S (granulated fertiliser containing 90% sulphur) is used. The advantage of using Wigor S is that it is not blown away from the field; it is easily soluble in water and quickly penetrates into the soil absorption complex and removed calcium from it. It should be remembered that one can carry out sulphurisation of soil at least 1 year before the planned date of plantation setup. The sown sulphur should be mixed with the soil using a disc harrow, cultivator or soil ripper [20]. Doses of sulphur depend on the type of soil and its pH (**Table 2**). The lighter the soil, the smaller the single dose of sulphur. The maximum single quantity of sulphur supplied should not exceed 300 kg. After application, sulphur should be ploughed to a depth of 10–20 cm in order to accelerate the acidifying effect of sulphur. If the current pH of the soil differs slightly from the optimum one for blueberry cultivation, acid peat, ground pine bark or sawdust from coniferous plants should be used locally in the holes under plants. In commodity plantations, furrows can be ploughed into which sawdust or pine bark will be poured, and then 150–200 kg of ammonium sulphate can be poured and mixed with soil using a ripper or cultivator. The additional use of ammonium sulphate will enable to reduce competition for nitrogen between blueberry roots and bacteria that decompose the organic material brought to the soil. The second method of acidification is very commonly used in amateur farming, where partial replacement of soil and dressing of holes before planting shrubs are easier. For hole dressing, 5–10 l of sawdust

Variety	Ripening time
Duke Spartan	Early
Bluecrop Bluegold Calypso Draper Toro Valour	Medium-early
Chandler Lateblue	Medium-late
Aurora Last call Liberty	Late

Table 2.
List of the most common varieties of commercial plantings in Poland.

or peat should be used for every plant. If the soil is very compact, hindering the development of fine roots, coarse sand can be additionally introduced at a ratio of 2:1:1. This treatment lowers the pH of the substrate while providing the plants with caries for years to come.

High blueberry shrubs, like other fruit plants, can be planted in autumn or spring. Each of these periods has its own advantages and disadvantages. The biggest disadvantage of autumn planting of shrubs is a possibility of freezing of shrubs in the winter. High blueberry shrubs are produced in containers (2–3 l pots) or root balled. Such material can be planted theoretically during the entire vegetation period, but the most often recommended time is the period of spring planting of plants. Regardless of the planting time, providing the plants with plenty of water should be always kept in mind. The material most commonly used to establish a plantation is 2 or 3 years old and has a well-developed root system. At commercial plantations, planting of blueberry shrubs is usually recommended with 3.0 × 1.0 m spacing. The belt and row planting system with 3.2 × 1.2 × 1.25 m spacing is used less frequently. Shrubs should be planted 2–3 cm deeper than they grew in the container.

In addition to the aforementioned factors related to land selection and soil preparation, the choice of variety is of great importance in the cultivation of blueberry. The value of a cultivated variety is determined by its biological properties and economic characteristics. Biological properties include the longevity of the shrubs, soil and climatic requirements, the time when the vegetation starts and ends, the time when the berries start to bear fruit, the fruitfulness and ripeness of the berries and resistance to diseases and pests. Equalisation of fruits, that is, their size and number of berries in the cluster and their taste and usable value, resistance of fruits to transport and durability in commodity trading are the economic features of significant importance. In our climatic zone, the varieties of North American origin are grown most often. Until recently, the leading variety most commonly found at plantations was the 'Bluecrop' variety, while nowadays such varieties as 'Liberty', 'Chandler', 'Duke' and 'Aurora' are most willingly planted (**Table 2**).

However, for the preparation of high blueberry plantation, the soil class is less important because the blueberry is an extremely resistant plant which has relatively low nutritional requirements. The appropriate quantity of water and regular enrichment of the settlement with humus using peat or sawdust guarantee the success of the cultivation.

3. The pro-health properties of blueberry fruits with a wide range of biological effects

Highbush blueberry (*Vaccinium corymbosum*) fruit is a particularly rich source of antioxidants [34]. Those compounds are mainly represented by flavonoids including anthocyanins, flavonols and flavanols, and polyphenolic compounds, which are represented by phenolic acids, tannins and stilbenes [35], which have high antioxidant capacity against hydrogen peroxide, superoxide radicals, peroxyl radicals and singlet oxygen [36]. Many studies have indicated that the blueberry has several beneficial health properties associated with the presence of such bioactive compounds [37]. These compounds may play a crucial role in the prevention of many chronic diseases.

3.1 Anthocyanin

Anthocyanins in blueberry fruits comprise a large group of water-soluble pigments. In fruits, they are found mainly in the external layers of the hypodermis (the skin). In cells, they are present in vacuoles in the form of various sized granules [38]. Anthocyanins are part of the very large and widespread group of plant constituents known collectively as flavonoids [39]. It has been found that blueberry anthocyanins may prevent multiple chronic diseases such as cancer [40], cardiovascular disease, diabetes [41] and age-related neurodegenerative decline [42]. Generally, anthocyanins have been reported to reduce damage caused by free-radical activity such as low-density lipoprotein oxidation, platelet aggregation and endothelium-dependent vasodilation of arteries [43]. The potential mechanisms by which anthocyanins may prevent colorectal cancer may relate to apoptosis induction and cell-cycle arrest as well as inhibition of proliferation, inflammation and angiogenesis [44].

3.2 Polyphenolic compounds

Polyphenols are a large class of natural compounds that have high antioxidant capacity and potential beneficial human health effects. These effects include antioxidant, anti-allergic, anti-inflammatory, anti-viral, anti-proliferative, antimutagenic, antimicrobial, anti-carcinogenic, protection from cardiovascular damage and allergy, microcirculation improvement, peripheral capillary fragility prevention, diabetes prevention and vision improvement [35, 45]. These classes of compounds also appear to have positive effects on the cardiovascular system, which may be due to their ability to act as free radical scavengers or by other mechanisms [46].

3.3 Antioxidant activity

Overproduction of reactive oxygen species (ROS), such as superoxide anion, hydrogen peroxide and peroxyl radicals, and reactive nitrogen species (RNS), such as nitric oxide and peroxynitrite radicals, could lead to oxidative stress and nitrosative stress, respectively. These reactive species can damage proteins, lipids and DNA, leading to lipid peroxidation, altered signal transduction pathways and the destruction of membranes and organelles [47]. The balance between ROS and antioxidants in biological systems is referred to as redox homeostasis [48]. In order to combat oxidative stress, there are several types of endogenous enzymatic antioxidants such as superoxide dismutase (SOD), catalase (CAT) and peroxidases (POD), as well as nonenzymatic glutathione, ascorbate, carotenoids and polyphenolic compounds [47].

As a rich source of antioxidants, blueberry bioactive components exert an important role against oxidative insults [49]. The antioxidative activity of compounds found in blueberry fruits relies on various mechanisms, subject to their structure. Flavonoids inhibit lipid oxidation; they chelate metals and scavenge the active forms of oxygen [50]. Anthocyanins, which are a flavonoid subgroup, inhibit the oxidation of human low-density lipoprotein and liposomes and scavenge free radicals [51]. They are also the most effective natural antioxidants and are shown to have significant anti-ageing, anticancer and immunoprotective effects [52]. Glycosylation of an anthocyanin decreases radical scavenger activity compared with the aglycone, as it reduces the ability of the anthocyanin radical to delocalise electrons [53, 54]. Anthocyanins also protect ascorbic acid against oxidation [55]. Polyphenolic compounds protect the easily oxidisable food compounds. They inhibit the oxidation of vitamin C, carotenoids and unsaturated fatty acids [56]. Blueberry fruits also contain vitamin C, provitamin A, carotenoids and E and B vitamins that can contribute to antioxidant protection [57].

In addition, the anticancer effects of berry bioactive compounds reduce and repair damage resulting from oxidative stress and inflammation [58]. Those berry phytochemicals may also potentially sensitise tumour cells to chemotherapeutic agents by inhibiting pathways that lead to treatment resistance, and consumption may provide protection from therapy-associated toxicities. These include effects on cellular differentiation and apoptosis and effects on proteins and enzymes that are involved in these processes.

3.4 Effects of blueberry fruit consumption on human health

Blueberries constitute one of the most important sources of potential health supporting phytochemicals in the human diet [58]. The dietary consumption of berries has positive effects on human health and diseases [37, 39]. Blueberry fruits are a rich source of ascorbic acid and phenolic compounds, particularly phenolic acids, anthocyanins, proanthocyanidins and other flavonoids, which prove to be beneficial to human health [58]. Their biological activities include protection against the incidence and mortality rates of cancer and protection against ischemic heart disease mortality, and they have antitumourigenic, antimicrobial, anti-inflammatory allergic and antimutagenic properties. Therefore, the use of phytochemicals as dietary supplements is growing.

However, what dose of a single antioxidant should be used as a dietary supplement? Natural phytochemicals at the low levels present in fruit and vegetables offer health benefits, but these compounds may not be effective or safe when consumed at higher doses, even in a pure dietary supplement form. Generally, taking higher doses increases the risk of toxicity. In the case of antioxidant nutrients, the proper physiologic dose should follow the recommended dietary allowance [59]. The pharmacologic dose is not equal to the physiologic dose and in some cases can be toxic. For example, in a human study, 30 healthy individuals whose diets were supplemented with 500 mg vitamin C/dose showed an increase of oxidative damage in the DNA isolated from lymphocytes [48].

4. Effect of factors on blueberry bioactive substances

Many factors that may impact antioxidant activity and composition in blueberry fruits include cultivar, environmental conditions, degree of ripeness, storage and food processing.

4.1 Cultivars

It is known that the content of phenolic compounds in berry fruits is also affected by genetic differences among cultivars of *Vaccinium corymbosum* [60–62]. Similarly, there is considerable variability in the antioxidant capacity of different cultivars [63, 64]. The genotype and maturity have effects on the activity and composition of antioxidant compounds of whole, skin and pulp fruits from highbush blueberry. Total antioxidant activity and the content of bioactive compounds in ripe fruits varied among the cultivars.

4.2 Degree of ripeness

The content of antioxidants in blueberries varied among the developmental stages of fruits. An increasing maturity at harvest of blueberry cultivars yielded fruits with higher antioxidant, anthocyanins and total phenolic contents [65, 66]. By contrast, Castrejón et al. found that polyphenolic compounds' concentration and antioxidant activity in highbush blueberry fruits decreased from unripe green to ripe blue stages of fruit maturity [67]. Total flavonoids and vitamin C levels in blueberry fruits were also decreased as the fruits grow [68]. Those antioxidants in green fruits were highest and blue fruits were lowest. Total phenolic contents and total flavonoids in fruit skin were much higher than in other tissues, which may indicate that the skin has the higher antioxidant performance.

Additionally, total antioxidant activity in unripe green and fully ripe fruits was high, whereas the lowest levels were found in intermediate ripe fruits [63]. Obvious differences in the antioxidant enzyme activities were observed among different developmental stages of fruit and tissues [68]. In the skin, pulp and blue fruit, generally SOD, POD and CAT were the highest in the skin, followed by the blue fruit, and the pulp content was the lowest, while the opposite was found in the PPO. The antioxidant enzyme activities in fruit are mainly influenced by species, environmental conditions, fruit maturation, variability over the years, harvest season and other factors.

4.3 Storage condition

Blueberry fruits are commercialised in different ways, mainly as fresh or frozen products. Freezing and drying are two possible methods to preserve blueberries, but the severity of both processes might destroy anthocyanins or their antioxidant effects [69].

The bioactive compounds in highbush blueberry fruits can be stored only for 6 weeks under controlled atmospheric conditions, in which none of the cultivars showed a significant decrease from the harvest antioxidant activity value in fruits [70].

In the condition of storage at 5°C, the level of bush ripeness had no significant effect on antioxidant activity, total phenolic content or anthocyanin content; however, fruit maturity had a significant effect on antioxidant activity, total phenolic content and anthocyanin content. The content of those bioactive substances was strongly correlated with each other. An increase in antioxidant activity, total phenolic content and anthocyanin content may occur in blueberry during cold storage and is cultivar dependent [61, 62].

The influence of storage conditions on anthocyanin stability for blueberries stored frozen was also investigated. Concentration of anthocyanins in frozen blueberries was significantly reduced; more than 50% degradation of the anthocyanins was found after 6 months of frozen storage [71]. However, when drying was preceded with osmotic dehydration, a small amount of anthocyanins in blueberry fruits were lost [69].

4.4 Food processing

Many fruit-based foods are processed into products such as beverages, baked goods or confectionaries. Processing and preservation methods, such as hot air drying, freezing/thawing, freezing/osmotic pretreatment and microwave drying, are popular techniques for blueberry fruit preservation [72]. The processing treatments that the fruit undergo may have a detrimental effect on their phytochemical antioxidant.

Regarding the influence of processing methods, the antioxidant capacity is decreased by food processing practices, such as heat or aeration [60]. There was a slight increase in anthocyanin content of the highbush blueberry fruit when processed at 20°C; however, there was no change in the oxygen radical absorbing capacity. A slight increase in total anthocyanin value after some thermal pretreatment processing was recorded [73]. Blanching of blueberries at 85°C for 3 min resulted in about 7% growth of anthocyanin. However, the anthocyanin content of thermally treated blueberries, osmodehydrated, or air dried at 70°C decreased by about 30% [74]. The amount of anthocyanins after freeze-drying is also lower probably due to their degradation.

The antioxidant capacity of the blueberries was superior. The pomace exhibited high activity, albeit lower than that of the fruit; however, after processing, the flour and the dried blueberries lost 66 and 46% of the original antioxidant activity, respectively [75]. The average anthocyanin contents of the fruits were moderate compared to other sources and species of blueberries. The pomace contains a large amount of anthocyanins, while the flour and dried blueberries exhibited a 32 and 42% loss in anthocyanin content, respectively.

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Chapter 5

The Developing Blueberry Industry in China

Jiang Jiafeng, Wei Jiguang, Yu Hong and He Shan'an

Abstract

The present situation of blueberry industry in China was summarized. The six main blueberry cultivation areas in China were reviewed and practical suggestions were made. Reference and guidance for water management of rabbiteye blueberry in Yangtze river basin was provided, and water physiological characteristics and water requirement of blueberry were also clarified so as to provide scientific management of blueberry. Effects of vinegar residue on soil physical and chemical properties, enzymatic activities, growth of blueberry, nutrient uptake, and fruit quality were studied. The effect of vinegar residue on the growth of blueberry and the mechanism revealed from the perspective of soil amelioration were also discussed from the results.

Keywords: blueberry, China, cultivation, water management, vinegar residue

1. An overview of blueberry growing areas in China

There are 91 species, 24 subspecies of *Vaccinium* L., which distributed all over the country. Among them, *V. uliginosum* L. and *V. vitis-idaea* L. are used as raw materials for making jam and juice in Northeast China for a long history. The young leaves of another species, *V. bracteatum* Thunb., distributed widely in southern China, has been used as raw material for making "black rice," a traditional health-care food, since Han dynasty about 2000 years ago. The cultivated blueberry industry in China, mainly soil cultivation, was established on the basis of introduced cultivars from abroad.

1.1 Current situation of blueberry cultivation in China

Blueberry varieties were first introduced from Northern America and Europe by Jilin Agricultural University and Institute of Botany, Jiangsu Province and Chinese Academy of Sciences in the middle of the 1980s, respectively. The expansion of commercial growing has been conducted since the beginning of the twenty-first century, and the growing areas have been developed rapidly, reaching 40,000 hm^2 so far. According to the characters of blueberry growing areas from north to south, six regions could be classified as follows (**Figure 1**):

1. Liaodong peninsula region, protected cultivation mainly

2. Shandong peninsula region, for fresh fruits mainly and processing as well

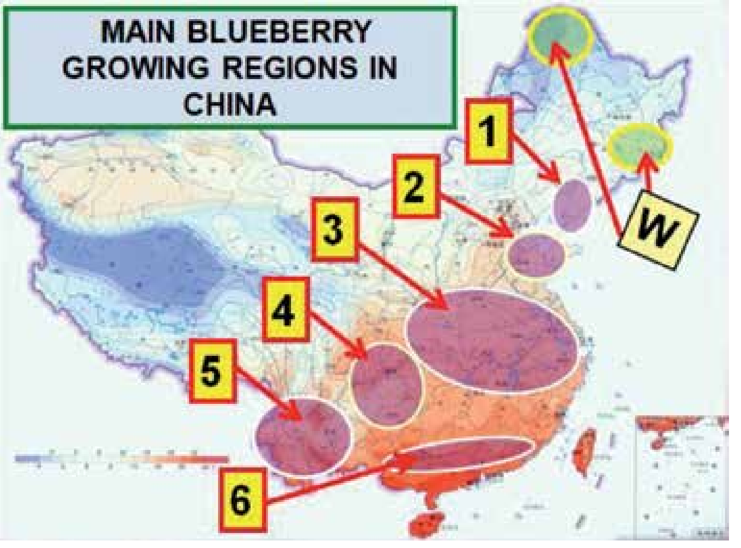

Figure 1.
Main blueberry growing areas in China. W, wild blueberries mainly; 1, Liaodong peninsula region; 2, Shandong peninsula region; 3, Changjiang River basin region; 4, Guizhou and Sichuan region; 5, Yunnan region; 6, South China region.

3. Changjiang (Yangtze) river basin region, for both processing and fresh fruits

4. Guizhou and Sichuan region, for processing mainly and fresh fruits also

5. Yunnan region, for extremely early market and dual purpose fruits

6. South China region, the newly forming region for fresh fruits

It seems that there has greater potential in the latter four southern regions, especially the Changjiang river basin region. Accordingly, because of the different ecological characteristics of every region, the appropriate varieties were different.

1.1.1 Liaodong peninsula region, Liaoning province

The climate of this region is temperate continental monsoon type, with many rainy days in summer, abundant sunshine, and frost free for 168–180 days. Slightly acidic and sandy loam is the main soil type, which is suitable for blueberry cultivation. The cultivation patterns include open field, cold shed, and greenhouse culture.

Varieties of open-field culture are “Blomidon,” “Bluecrop,” and “Duke.” Harvest time is from early July to late August, which is the latest ripening time in China. Both freezing damage and drought are the main ecological limiting factors in winter and spring. Plants must be put down and covered by soil to prevent cold and drought in winter. This measurement is barely feasible for young trees, and it is quite hard for adult plants to maintain high and stable yields. Sometimes fruit quality could be reduced by the continuous rainy days during the harvesting period.

Protected culture cannot only overcome those shortcomings of open-field culture but could also be used for forcing culture. As lower temperature usually comes early in autumn and sunshine is abundant in spring, forcing culture is a strategic choice for early harvest. But there would also be series of problems of culture techniques, facilities, and high management cost needed to be solved. Early maturing southern highbush varieties, such as “Misty,” are cultivated for pursuing high profit, which could be harvested in mid-late March of the next year as the temperature in greenhouse could warm up in mid-November of the previous year. However, due to the development of early ripening fresh fruit in Yunnan and blueberry facility cultivation in Shandong, blueberry industry in this area has great competitive pressure.

1.1.2 Shandong peninsula region, Shandong province

This region is typical of the warm monsoon climate with slightly acidic to neutral sandy loam soil. Light source is rich and there is large temperature difference between day and night. Shandong is a well-known horticultural province in China, with the unique ecological conditions which are suitable for many fruit trees and vegetables.

The annual rainfall in this area is 650–850 mm, 60% of which is concentrated in summer with rainstorm occurring sometimes, while the average annual evaporation is 1000–1200 mm, so the annual rainfall is insufficient for blueberry growing. Therefore, as for open-field blueberry cultivation, groundwater is the main water resource for irrigation. Both groundwater quality and supply are not desirable, and the air humidity is often so low; water shortage is the main limiting factor for open-field blueberry growing in this region. In addition, besides a few sites located in the coastal areas, blueberry production in this region is often threatened by hot and dry winds in late spring, as well as by freezing in winter and early spring. Therefore, culture model of greenhouse and cold shed cultivation become more popular, although open-field cultivation still exists in this area. The varieties cultivated in greenhouse have also changed from the original northern highbush varieties to much earlier maturing southern highbush varieties, which could be harvested in the mid-late March.

1.1.3 Changjiang (Yangtze) river basin region

The range of this region is from the limitation of soil pH < 6.0 from north bank of Yangtze river to roughly along about 20°C annual isothermal line on the southern bank of Changjiang river. Annual rainfall is generally >1000 mm, and summer temperature is usually high. Strong acidic red soil and yellow soil (pH 4.5–5.5) are the main acidic soil types, that is, clay, poor fertility, low organic matter, and low total nitrogen content. Therefore, corresponding measures should be taken to improve soil permeability and increase soil organic matter content. How to use local resources to increase soil organic matter in Changjiang river basin is a long-term research topic. It is suggested that the laying of drainage pipeline under the planting site could be an effective measure to prevent the soil from being too wet.

The most adverse environmental factor in this region is high temperature in summer, and the extreme maximum temperature may reach above 40°C. The number of days with the highest daily temperature greater than or equal to 35°C in this region is also increasing with the tendency of global warming and reaching 20 days in most places and even up to 30 days in some sites. This region has not only heavy rain but also drought in summer and autumn due to monsoon climate. Therefore, soil moisture regulated by drainage and irrigation is one of the key measures of blueberry culture in this region. In addition, the harvest time of some varieties in this region coincides with the plum rain season, so rain shelter should be adopted to avoid this problem. Typhoon damage should also be prevented in offshore areas.

With appropriate cultural practice, most rabbiteye varieties were adaptable to high temperature in summer, and the yield of “Brightwell” could reach more than 22.5 t per hm^2. As for southern highbush blueberries, only a few varieties with relatively strong wild traits could give high and stable yield.

1.1.4 Guizhou and Sichuan regions

The landform of Guizhou and Sichuan is complex, and blueberry is mainly planted in the low mountains and hills. The climate of Guizhou is warm and humid, belonging to the subtropical humid monsoon climate, relatively cooler in summer, and no severe cold in winter. Acid red soil is the main soil type. So, rainy weather and acid soil provide favorable environment for blueberry growing. It is practically proven that the yield of some rabbiteye varieties could reach 5–8 kg per plant. However, some weather factors, such as cloudy, foggy, and unstable spring temperature in plateau area, are the negative factors affecting the quality and yield.

Soil of the eastern Sichuan basin is mainly slightly acidic purple soil and paddy soil, which is relatively sticky and difficult to ameliorate. Other areas suitable for blueberry cultivation are mainly hilly gentle slopes, of which the soil is acid yellow brown soil and also heavy. Considering the poor local transportation, processing variety with more suitable adaptability should be adopted for the culture.

1.1.5 Yunnan region

As Yunnan is located in the low latitude plateau, the temperature varies greatly due to the influence of altitude and latitude. Except for a few places, the continuous growth season in most places lasts for more than 300 days every year. Blueberry has been introduced to Yunnan since 2004, and this region has become an early fresh blueberry producing region with a special reputation in China. Under applicable facilities, such as canopy and integration of water and fertilizer, supply of fresh fruit might last from January to August. With the cooler summer, large difference between day-night temperatures, and abundant sunshine, blueberry culture in this region could achieve high and stable yield, with qualified fruits of high sugar content; good coloring; solid, thick peel; and fragrance.

For open-field cultivation, sometimes yields would be affected by late frost in later spring and hail in spring and summer. What is more, fruit quality and harvesting of medium-maturity variety would be affected by rainy season from May to October.

1.1.6 South China region

This growing region has just been forming, including low mountains, hills, and river plains in Guangdong, Fujian and, Guangxi provinces, with the lowest latitude in all production regions in China. Annual average temperature is a little higher, and most acid soil is red soil with sticky texture and low content of soil organic matter. Considering the higher annual average temperature, some southern highbush varieties are suitable for this region; and planting areas would develop mainly around large or medium-sized cities so as to face the local consumer groups directly.

1.2 Prospects

In China, blueberry industry is a new, rising, and prospective business, but it could not be successful easily. Struggling hard and implementing hard are both needed seriously, and, among all, the key points are scientific research and practice for development.

1.2.1 Strengthening the basic construction of orchards, implementing fine cultural practice, and insuring high and stable yield with good quality

In an overall view, although the growing areas increased rapidly and qualified orchards had emerged with good yields, such as 22.5 t/hm^2, approaching the international high level, production on most plantations was low, usually only 3–4 t/ha, so the integrated level of blueberry industry was obviously lagging behind. There appeared a serious imbalance among different regions and orchards. Problems occurred during the early blind development stage, such as poor selection of orchard site, orchard infrastructure construction being not in place, and extensive management. It is helpful to be taught by all these historical experiences and to eliminate all these problems in order to improve orchard conditions to get successful and bright future.

1.2.2 Paying attention seriously to using suitable cultivars in various regions

At the beginning of commercial development stage, especially affected by the fever of making profit by selling seedlings, a number of orchards were failed by a wrong selection on cultivars, e.g., in southern China, where the soil was very heavy, people were particularly interested in southern highbush cultivars but not rabbiteye cultivars, since most southern highbush cultivars were not adapted to the ecological conditions in southern China and orchards were falling down totally in a few years. Learning from practice experience, now, most orchards have known that "cultivars with good adoption to environment is the basic principle of a good cultivar." Otherwise, no matter how nice it is, it is not a reasonable one. Even more, it has been realized that the most suitable cultivars in China should be eventually bred by Chinese breeding system in various regions.

1.2.3 Emphasizing harvesting, storing, and shipping in order to promote fruit quality

As cooling line is, so far, not perfectly used in Chinese blueberry industry, there is a big room to promote the quality by fully using these techniques. Furthermore, processing to increase economic value is a preferable way to gain profit by enterprises of blueberry industry. Accordingly, it is important to have varieties especially for processing. Recently, a favorable selection "LM-1" was released, and it possesses better adaptability to clay soil and hot summer and bears smaller fruit of approximately 1 g per berry with rich nutrition and wild fruit flavor. Its yield could reach more than 15 t/hm^2. It is one of the new representative varieties found in China.

2. Some ecophysiological response of blueberry grown under different water regimes

Blueberries (*Vaccinium* spp.) usually prefer well-drained, moist, acidic sandy loam soils with high organic content. It can tolerate moderate drought but is sensitive to soil waterlogging [1]. Inadequate or excessive soil water supply attenuates the growth and development of blueberry and makes a negative effect on both yield and fruit quality. Among the myriad of management measures for blueberry culture, scientific and reasonable water management is particularly critical [2]. Clarifying the water physiological characteristics and water requirement of blueberry is a precondition for development of water-saving and efficient irrigation schedule. Therefore, water physiology and water requirement of different types of blueberry at different growth stages under different soil and climatic conditions have been extensively studied [3–7]. Results from previous studies showed blueberry water requirements vary with plant age and cultivar, weather and soil conditions, and cultural practices [8–14]. Consequently, water demand of blueberry under different climate and soil conditions should be appropriately adjusted and revised according to local condition.

Soil types in Changjiang (Yangtze) river basin region are red soil and yellow-brown soil, which are heavy clay with poor drainage. Moreover, there are not only heavy rain days but also drought days in summer and autumn due to monsoon climate. Therefore, soil permeability improvement by soil amelioration and soil moisture regulation by precision irrigation are the key measures of blueberry cultivation in this region. Because of the different soil texture and climate conditions, it is necessary to study the water stress tolerance and water requirement of major blueberry cultivars based on local experiments in this region. It has been found that types of blueberries that best suited to grow in Yangtze river basin region are rabbiteye (*Vaccinium ashei*) and southern highbush (*V. corymbosum*). In this study, changes in growth indexes and photosynthetic characteristics of rabbiteye blueberry seedlings after drought or flooding stressed for 14 days and recovering normal water supplying were analyzed and compared. Effects of different irrigation regimes on seedling growth and water consumption pattern of "Brightwell" rabbiteye blueberry and "Misty" southern highbush blueberry were investigated. The relationship between actual water requirement of blueberry seedlings and water surface evaporation was studied, and the water requirement model based on water surface evaporation was established. Recommendations for optimum irrigation management of blueberry grown in southern China were drawn accordingly.

2.1 Studies on some ecophysiological responses of blueberry grown under different water regimes in Nanjing

2.1.1 Adaptability of blueberry to water stress

Drought and soil waterlogging were prone to occur due to the evident uneven distribution of rainfall in blueberry growing areas of the Yangtze river basin [15]. Therefore, it is urgent to carry out studies on adaptability of blueberry to water stress. Taking 2-year-old potted seedlings of cultivar "Powderblue" and "Gardenblue" of rabbiteye blueberry (*Vaccinium ashei* Reade) as research objects, changes in growth indexes of seedlings after drought or flooding stress for 14 days and recovering normal water supply for 60 days were examined and analyzed. The results showed that after drought or flooding stress for 14 days, growths of two cultivar seedlings are inhibited severely, and their root dry weight, shoot dry weight, and whole plant relative growth rate (RGR) are significantly lower than those of the control. After recovering under normal water supplying for 60 days, "Powderblue" and "Gardenblue" did not show significant differences in the root dry weight (81.4 and 83.1% of control values, respectively) between drought-stressed and control plants. Shoot dry weight of drought-stressed plants of two cultivars was still significantly lower than those of the control. RGR of drought-stressed "Powderblue" seedlings were significantly higher than that of the control, whereas "Gardenblue" did not show significant differences in RGR (75.2% of control values) between drought-stressed and control plants. Recovering under normal water supply for 60 days after flooding stress, root dry weight and shoot dry weight of the recovered "Powderblue" seedlings were significantly lower than those of the control (41.9 and 39.8% of control values, respectively), whereas recovered "Gardenblue" seedlings maintained root dry weight and shoot dry weight of 78.7 and 62.1%, respectively, of those of corresponding controls. RGR of recovered "Powderblue" seedlings were lower than that of the control, whereas RGR of "Gardenblue" was greater than that of the control seedlings (**Figure 2**). It is concluded that recovered rabbiteye blueberry seedlings preferentially partitioned assimilate to the roots during the recovery period. RGR of the recovered seedlings approached or exceeded

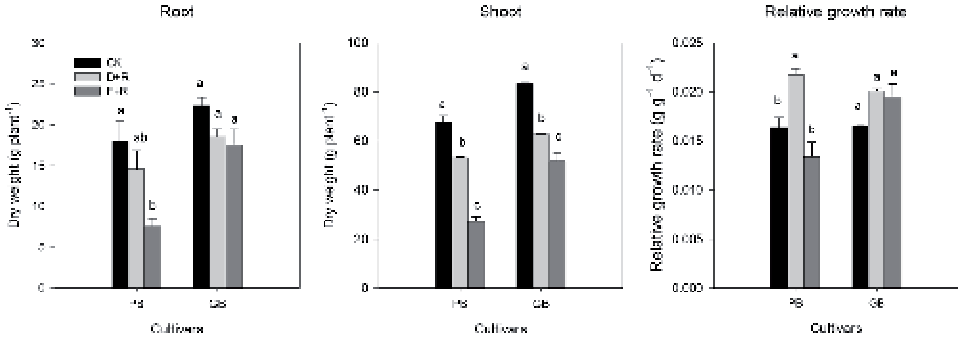

Figure 2.
Comparison on some growth indexes of "Powderblue" and "Gardenblue" of Vaccinium ashei *after water stress for 14 days and recovering under normal water supply for 60 days. Different small letters indicate the significant difference (P ≤ 0.05) among different treatments of the same cultivar.CK, the control; D + R, recovering under normal water supply for 60 days after drought stress; and F + R, recovering under normal water supply for 60 days after flooding stress.*

those of the control by the end of the experiment. "Powderblue" is more drought-resistant, whereas "Gardenblue" is more flood-resistant. Our study demonstrated that rabbiteye blueberry "Powderblue" and "Gardenblue" appear to be promising cultivars for commercial planting in Yangtze river basin region prone to drought or flooding of up to 2 weeks.

2.1.2 Study on photosynthetic physiology of rabbiteye blueberry under water stress

Study on photosynthetic response of rabbiteye blueberry to water stress was not only helpful to reveal photosynthetic adaptation to water stress in blueberry but also beneficial to selecting appropriate measures to improve the stress resistance of blueberry, creating favorable conditions for growth and yield formation of blueberry. In this study, potted, 1-year-old seedlings of rabbiteye blueberry "Brightwell" and "Gardenblue" were subjected to 14-day flooding stress, followed by a recovery for 30 days. The leaf net photosynthetic rate (P_n), stomatal conductance (g_s), and intercellular CO_2 concentration (C_i) were measured during stress, 7 days after recovery and 30 days after recovery.

Repeated measures ANOVA showed that flooding treatment and duration both had a significant effect on net photosynthesis rate (P_n), stomatal conductance (g_s), and intercellular CO_2 concentration (C_i) in both cultivars ($P \leq 0.05$). There was also a significant interaction between flooding treatment and duration ($P \leq 0.05$), indicated that the negative effects of flooding increased over time (**Figure 3A**–**F**).

The P_n in "Brightwell" decreased gradually after 3 days of flooding, and exhibited a significant drop by the 5th day. By day 11, the P_n of flooded plants in "Brightwell" dropped to the lowest values (−39% compared with that of control) and increased thereafter until the end of the experiment (**Figure 3A**). After 8 days of flooding, flooded plants in "Gardenblue" showed a significant reduction in P_n compared to control plants. The largest decline in P_n of flooded plants in "Gardenblue" was observed on days 21 (−39% compared with that of control) (**Figure 3B**). After the flooding was ended and plants were allowed to recover for 30 days, the P_n of both cultivars recovered to different degree (104 and 95% for "Brightwell" and "Gardenblue" with respect to control plants, respectively).

Flooding-induced decline in g_s in both cultivars were evident after 11 days of flooding (**Figures 3C** and **1D**; $P \leq 0.05$). The largest decline in g_s of flooded plants in both cultivars was observed on days 14 as compared to the control

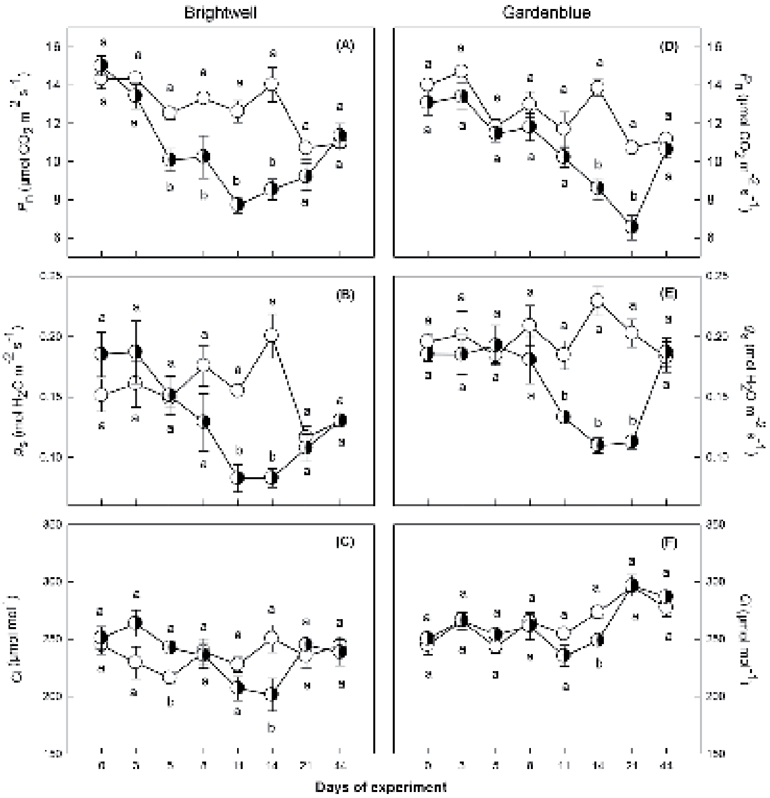

Figure 3.
Changes in the net photosynthesis rate (P_n), stomatal conductance (g_s), and intercellular CO_2 concentration (C_i) in the leaves of 1-year-old seedlings of "Brightwell" (A–C) and "Gardenblue" (D–F) under two soil conditions: Control (open circles) and flooding plus recovery (half-closed circles). Measurements were taken on days 0, 3, 5, 8, 11, and 14 (during the flooding); day 21 (7 days after flooding ended); and day 44 (30 days after flooding ended). Different lowercase letters indicate significant differences ($P \leq 0.05$) among treatments. Values are mean ± standard error.

(−59 and −52% for "Brightwell" and "Gardenblue," respectively). When the flooding treatment ended, g_s in both cultivars recovered gradually and reached a level similar to that of the control (100 and 102% for "Brightwell" and "Gardenblue" with respect to control plants, respectively) by the end of the experiment.

The C_i in both cultivars decreased gradually after 5 days of flooding. Compared with the control, the C_i of "Brightwell" and "Gardenblue" under flooding significantly decreased by 19.6 and 9.0%, respectively, by the 14th day (**Figures 3E** and **1F**; $P \leq 0.05$). After the flooding was ended and plants were allowed to recover for 7 days, the C_i of both cultivars increased rapidly and was slightly higher than that of the control. By day 44 (30 days after flooding ended), the C_i in both cultivars increased to a level similar to that in the control (97.4 and 103.4% for BW and "Gardenblue" with respect to control plants, respectively).

Decreases in carbon assimilation under the flooding condition have been demonstrated in many woody fruit species [3, 4, 16, 17]. In the present study, the leaf net photosynthetic rate (P_n) of flooded plants in both cultivars was significantly less than those of unflooded controls after 14 days of flooding. Meanwhile, the stomatal conductance (g_s) and intercellular CO_2 concentration (C_i) also decreased significantly, indicating that flooding reduced photosynthesis mainly via a decrease in stomatal conductance. Photosynthetic activity decreased during flooding in both cultivars but recovered rapidly following drainage, which indicates that decreased photosynthetic activity was temporary and that rabbiteye blueberry possessed rapid recovery ability after flood water was released. Our study demonstrated that rabbiteye blueberry could adapt to flooding stress by adjusting photosynthetic process and possess a certain extent of adaptation to flooded soil conditions.

2.1.3 Study on water requirements of blueberry

Reasonable water management is the basic guarantee for the high fruit yield and quality of blueberry [18–20]. Clarifying the water requirement of blueberry is a precondition for scientific management of blueberry orchard. In this study, weighing lysimeters were used to determine actual evapotranspiration (ET) of young seedlings of "Brightwell" rabbiteye blueberry and "Misty" southern highbush blueberry under different irrigation regimes. Effects of different irrigation regimes on seedling growth, total water consumption, and water-use efficiency (WUE) were also investigated. The results show that net increment of dry weight (IDW) per plant and total water consumption (TWC) per plant of two cultivar seedlings both elevate with increasing amount of irrigation, while water-use efficiency is lowest under 50% ET condition and the highest under 75% ET condition. As for "Brightwell," 125% ET treatments had similar values for IDW and WUE compared to 100% ET plants, whereas TWC was significantly higher than that of the control (100% ET). IDW and TWC in 50 and 75% ET treatments were significantly lower than that of the control, whereas no significant ($P > 0.05$) differences in WUE were observed. As for "Misty," IDW and TWC in 125% ET treatments were significantly higher than that of the control (100% ET), whereas WUE was not significantly different from that of the control (100% ET). IDW in 75% ET treatment was slightly lower than that of the control, and TWC was significantly lower than that of the control, whereas WUE was significantly higher than that of the control. IDW, TWC, and WUE in 50% ET treatment were all significantly lower than that of the control (**Figure 4**). It is concluded that the optimum irrigation treatment for "Brightwell" and "Misty" in southern China are 100 and 75% ET, respectively [21].

Water requirement estimation is one of the important aspects in crop water requirement research. Crop water requirement estimation by pan evaporation has been extensively used worldwide due to simplicity and low cost of the technique [22–24]. Pan evaporation method takes into account temperature, humidity, wind speed, solar radiation, and other meteorological factors, which are the same factors that affect crop

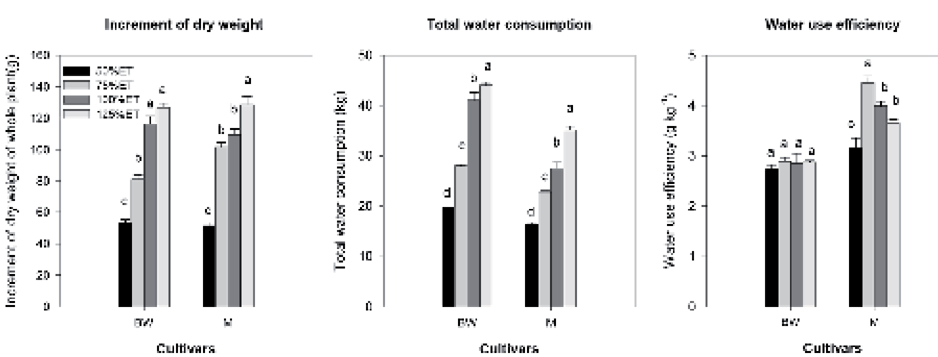

Figure 4.
Effects of different irrigation treatments on net increment of dry weight per plant, total water consumption, and water-use efficiency of seedlings of Vaccinium ashei *"Brightwell" and* V. corymbosum *"misty." different lowercases in the same column indicate the significant difference (P ≤ 0.05) among different treatments of the same cultivar.*

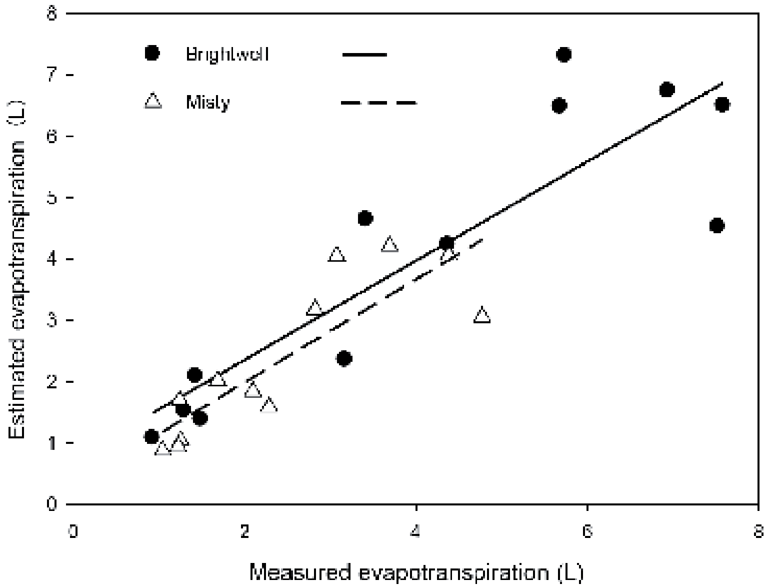

Figure 5.
The relationship between measured and estimated evapotranspiration for young seedlings of Vaccinium ashei *"Brightwell" and* V. corymbosum *"misty."*

transpiration rate. In order to clarify the feasibility of water requirement estimation by water surface evaporation, actual water requirement of young seedlings of "Brightwell" rabbiteye blueberry (*Vaccinium ashei*) and "Misty" southern highbush blueberry (*V. corymbosum*) was measured by weighing lysimeter in this study. Simultaneously, water surface evaporation above the canopy was estimated using the standard 20 cm evaporation pan. The relationship between actual water requirement and water surface evaporation was studied, and the water requirement model based on water surface evaporation was established: $Q = D^2 \times E_{p\text{-}20} \times k/1000$, in which Q is the plant water use within a certain period, D is the diameter of the plant at drip line (cm), $E_{p\text{-}20}$ is the measured evaporation from the standard 20 cm evaporation pan (cm), and k is the pan coefficient. The regression analysis results showed that estimation model for water requirement of "Brightwell" was $Q = D^2 \times E_{p\text{-}20} \times 0.282/1000$, and estimation model for water requirement of "Misty" was $Q = D^2 \times E_{p\text{-}20} \times 0.291/1000$. According to the water requirement estimation model, the fitting value of evapotranspiration was obtained, and regression analysis was conducted for the relevance between estimated and measured evapotranspiration. As illustrated in **Figure 5** there were significant positive correlations between the measured and estimated evapotranspiration ($P < 0.01$) in both cultivars, indicating that actual water requirement estimation by water surface evaporation above the canopy is feasible and effective. Our results provide reference and guidance for water management of rabbiteye blueberry under the similar climatic and geographical conditions.

2.2 Future challenges

Water management is the key factor for the success of blueberry growth. The water ecophysiology of blueberry should be further studied from the following aspects:

I. Study on blueberry water ecophysiology in the context of global climate change. Global climate change will increase the frequency and intensity of extreme weather events, which makes the blueberry growth face serious environmental challenges. Research on the ecophysiological response mechanism of water, under the global environmental change scenario, could provide the scientific evidence for developing cultural and breeding strategy to cope with global climate change.

II. Study on reliable and efficient evaluation and screening system for blueberry drought and waterlogging resistance. Plant stress resistance is a complex and comprehensive trait affected by many factors. At present, there is no widely accepted evaluation method, and the resistance evaluation screening system needs to be further improved.

III. Study on the water requirement of blueberry under open-field and protected cultivation conditions. Systematic studies should be carried out on the dynamic of water requirement of different blueberry varieties at different growth stages, so as to provide theoretical basis and technical support for resource-saving and energy-efficient water management system of blueberry orchards.

IV. Estimation of water consumption of blueberry orchard at regional scale. Along with the increase in blueberry growing areas, blueberry orchards have gradually become an important vegetation type. It is of great significance to research the water consumption of blueberry orchards at regional scale for the design of irrigation system and rational allocation of water resources.

3. Effect of vinegar residue on soil properties and blueberry growth

Blueberry grows in acidic soil with high organic content [25]. Generally it is necessary to add a large number of organic materials for soil amelioration [26] due to its low content of soil organic matter and other factors in commercial cultivation in China. Therefore, exploring suitable acidic organic materials has become one of the basic factors to ensure the sustainable development of blueberry cultivation in China.

Vinegar residue, a unique organic material, is a by-product of traditional vinegar-making industry in China. Resource of vinegar residue is abundant, as there is 2.6 million tons of vinegar residue produced annually by just one enterprise, Jiangsu Zhenjiang Hengshun Sauce and Vinegar Co., Ltd. [27]. Vinegar residue has high crude fiber content, strong acidity, and high moisture content, which makes it difficult to be disposed [28]. It is reported that vinegar residue could ameliorate soil structure and increase soil organic matter content by its large amount of grain husks [29]. Besides, it contains high organic matter, protein, as well as N, P, and K nutrients, making it to possess potential in agricultural resource business, especially in blueberry cultivation, that prefers acidic organic matter.

Therefore, the effect of vinegar residue on soil physical and chemical properties, enzymatic activities, growth of blueberry, nutrient uptake, and fruit quality should be studied. And the effect of vinegar residue on the composition and diversity of rhizosphere microbial community of blueberry should further be analyzed. By this research we would clarify the effect of vinegar residue on the growth of blueberry and the mechanism revealed from the perspective of soil amelioration, in order to provide scientific basis for the application of vinegar residue on blueberry cultivation.

3.1 Experiment setup

The pot experiment was carried out in the blueberry nursery of the Institute of Botany, Chinese Academy of Sciences, Jiangsu province, in March 2016. And three treatments were set up as CK was pure soil; VR1 was V (vinegar residue), V (soil) = 25:75; and VR2 was V (vinegar residue), V (soil) = 50:50. At harvest time in July 2017, soil and plant samples were collected, and soil physical and chemical properties (pH, conductivity, bulk density, total N content, total P content, and organic matter content), enzymatic activities (urease activity and acid phosphatase activity), growth of blueberry (plant height, basal diameter, average single fruit weight, fruit shape index, aboveground dry weight, root dry weight and root-shoot ratio), nutrient uptake (leaf N content, leaf P content and root N content, root P content) and fruit quality (total phenol and total ketone), and composition and diversity of rhizosphere microbial community of blueberry were examined.

3.2 Effect of vinegar residue on blueberry growth and fruit quality

3.2.1 Effect of vinegar residue on blueberry growth

There showed no significant differences in plant height, basal stem, single fruit weight, and fruit shape index among treatments (**Table 1**), but the aboveground dry weight of blueberry was improved significantly by vinegar residue. And the aboveground dry weight of VR1 increased 23.82% compared with CK. Root dry weight of VR1 and VR2 increased by 85.47 and 83.94%, respectively, and there was no significant difference in root dry weight between VR1 and VR2 treatments. The root-shoot ratio of plants treated with VR2 was the highest, 72.00% higher than CK, indicating that the application of vinegar residue significantly promoted the growth of blueberry roots. And 50% vinegar residue amount had the best effect on promoting blueberry growth.

3.2.2 Effect of vinegar residue on blueberry nutrient uptake

The application of vinegar residue had no significant effect on the absorption of phosphorus in blueberry leaves, roots, and fruits (**Table 2**), and there was also no significant effect on the absorption of nitrogen in blueberry leaves and fruits (**Table 2**). In terms of total nitrogen content in roots, vinegar residue significantly promoted the total nitrogen content in roots. Total nitrogen in roots of VR1 and VR2 treatment increased by 18.86 and 24.06%, respectively, compared with CK.

3.2.3 Effect of vinegar residue on blueberry fruit quality

The reducing sugar of fruits of VR1 and VR2 increased by 8.12 and 9.65% (**Table 3**) with the application of vinegar residue, and the titratable acidity content decreased by 14.28 and 7.14% compared with the control. At the same time, the soluble solid content of fruits increased significantly as soluble solid content of fruits of VR2 increased by 11.60% compared with the control. The accumulation of total phenols and ketones in blueberry fruits was significantly promoted by the application of vinegar residue (**Table 3**), and the content of total phenols and total flavonoids of VR2 increased by 71.42 and 100.00%, respectively, compared with the control. The results showed that the application of vinegar residue could significantly improve the quality of blueberry fruits.

3.3 Effect of vinegar residue on soil properties

3.3.1 Effect of vinegar residue on soil structure

Soil pH dropped by 0.27 units by the application of vinegar residue (**Table 4**), but there showed no significant difference with the control. The soil electrical conductivity increased significantly with the application of vinegar residue, and the soil electrical

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	Plant height/cm	Basal stem/cm	Single fruit weight/g	Fruit shape index	Aboveground dry weight/g	Root dry weight/g	Root-shoot ratio
CK	82.58 ± 16.15a	1.62 ± 0.14a	1.55 ± 0.29a	0.87 ± 0.06a	112.80 ± 18.87a	28.83 ± 10.48a	0.25 ± 0.06a
VR1	69.36 ± 16.86a	1.43 ± 0.09a	1.55 ± 0.32a	0.88 ± 0.06a	139.67 ± 27.02ab	53.47 ± 19.14b	0.38 ± 0.07bc
VR2	71.48 ± 7.75a	1.39 ± 0.15a	1.56 ± 0.29a	0.88 ± 0.06a	121.03 ± 21.25a	53.03 ± 11.22b	0.43 ± 0.03c

Different lowercases in the same column indicate the significant difference at 0.05 level ($P < 0.05$).

Table 1.
Effect of vinegar residue on blueberry growth.

	Leave nitrogen/mg·kg^{-1}	Root nitrogen/mg·kg^{-1}	Fruit nitrogen/mg·kg^{-1}	Leave phosphorus/mg·kg^{-1}	Root phosphorus/mg·kg^{-1}	Fruit phosphorus/mg·kg^{-1}
CK	705.03 ± 28.89a	615.88 ± 83.02a	240.40 ± 15.86a	109.10 ± 23.29a	164.60 ± 21.29a	142.89 ± 28.47a
VR1	699.35 ± 19.57a	731.83 ± 33.95b	276.03 ± 22.99a	109.10 ± 8.22a	184.10 ± 33.04a	148.49 ± 27.24a
VR2	707.68 ± 30.39a	763.03 ± 63.31b	275.40 ± 33.01a	90.15 ± 6.34a	229.85 ± 79.10a	159.67 ± 28.68a

Different lowercases in the same column indicate the significant difference at 0.05 level ($P < 0.05$).

Table 2.
Effect of vinegar residue on blueberry nutrient uptake.

	Soluble solid/%	Reducing sugar/ $mg{\cdot}g^{-1}$	Titratable acidity/%	Total phenols/ $mg{\cdot}g^{-1}$	Total flavonoids/ $mg{\cdot}g^{-1}$
CK	10.17 ± 0.70a	71.78 ± 7.12a	0.42 ± 0.07a	0.56 ± 0.13a	0.71 ± 0.11a
VR1	11.03 ± 1.81ab	77.61 ± 6.57a	0.36 ± 0.02a	0.85 ± 0.23ab	1.21 ± 0.29bc
VR2	11.35 ± 1.12b	78.71 ± 11.52a	0.39 ± 0.02a	0.96 ± 0.15b	1.42 ± 0.24c

Different lowercases in the same column indicate the significant difference at 0.05 level ($P < 0.05$).

Table 3.
Effect of vinegar residue on blueberry fruit quality.

	pH	Electrical conductivity/$\mu S{\cdot}cm^{-1}$	Bulk density/$g{\cdot}cm^{-3}$
CK	4.90 ± 0.10a	110.90 ± 9.51a	0.85 ± 0.02c
VR1	4.63 ± 0.24a	138.65 ± 12.75a	0.84 ± 0.03c
VR2	4.67 ± 0.11a	224.80 ± 49.56b	0.72 ± 0.02a

Different lowercases in the same column indicate the significant difference at 0.05 level ($P < 0.05$).

Table 4.
Effect of vinegar residue on soil structure.

	Soil nitrogen/$mg{\cdot}kg^{-1}$	Soil phosphorus/$mg{\cdot}kg^{-1}$	Soil organic matter/$g{\cdot}kg^{-1}$
CK	11.15 ± 1.84a	13.30 ± 2.78a	8.24 ± 1.11a
VR1	22.30 ± 2.44b	18.83 ± 1.11ab	29.79 ± 2.82c
VR2	37.80 ± 6.26c	25.65 ± 3.18bc	51.84 ± 1.18d

Different lowercases in the same column indicate the significant difference at 0.05 level ($P < 0.05$).

Table 5.
Effect of vinegar residue on soil nutrient.

conductivity of VR2 224.80 $\mu S{\cdot}cm^{-1}$, which was still in the range of the electrical conductivity suitable for the growth of blueberry. Soil bulk density of VR2 was 15.29% lower than the CK. The decrease of soil pH and bulk density was beneficial for the growth of blueberry after the application of vinegar residue.

3.3.2 Effect of vinegar residue on soil nutrient

The total nitrogen and total phosphorus contents of soil increased significantly with the application of vinegar residue (**Table 5**). The total nitrogen contents of VR1 and VR2 treatments were 22.30 and 37.80 mg kg^{-1}, which were 100.00 and 239.01% higher than those of the control, and the total phosphorus contents were 18.83 and 25.65 mg kg^{-1}, respectively, which were 41.58 and 92.86% higher than those of the control. The content of soil organic matter increased significantly with the increase of vinegar residue. Content of soil organic matter of VR2 was 51.84 g kg^{-1}, 5.29 times higher than that of the control.

3.3.3 Effect of vinegar residue on soil enzymes

Soil urease and acid phosphatase activities increased significantly with the application of vinegar residue (**Table 6**). Soil urease activities of VR1 and VR2 treatments were 11.67 and 26.60 $mg{\cdot}kg^{-1}\,h^{-1}$, which increased by 177.86 and 533.33%, respectively, compared with the control. And acid phosphatase activities

	Urease activity/mg·kg^{-1} h^{-1}	Acid phosphatase activity/g·kg^{-1} h^{-1}
CK	4.20 ± 1.40a	11.70 ± 1.76ab
VR1	11.67 ± 1.62b	14.54 ± 7.08b
VR2	26.60 ± 2.80d	22.87 ± 5.00c

Different lowercases in the same column indicate the significant difference at 0.05 level (P < 0.05).

Table 6.
Effect of vinegar residue on soil enzyme.

of VR2 treatments were 22.87 mg·kg^{-1} h^{-1}, which was significantly 95.47% higher than the control.

3.3.4 Effect of vinegar residue on the composition and diversity of rhizosphere microbial community

There were 12 genera of microorganisms with relative abundance of more than 1% in each treatment, and there were *Nocardioides*, *Geobacillus*, *Dyella*, *Candidatus koribacter*, *Bacillus*, *Candidatus solibacter*, *Candidatus nitrososphaera*, *Conexibacter*, *Candidatus xiphinematobacter*, *Kaistobacter*, DA101, *Mycobacterium*, *Bradyrhizobium*, *Rhodoplanes*, and *Burkholderia* (**Figure 6**). And its abundance was accounted for 20–25% of the total microbial biomass of blueberries.

The abundance of *Bacillus*, *Burkholderia*, and *Rhodoplanes* varied significantly among all treatments (**Figure** 7). The growth of *Burkholderia* and *Rhodoplanes* was inhibited by the application of vinegar residue. As *Bacillus* was an important growth-promoting bacterium in plant rhizosphere, which played an important role in plant root growth and nutrient uptake, the abundance of *Bacillus* increased to 3.5–6% with the application of vinegar residue, while it was quite low in CK.

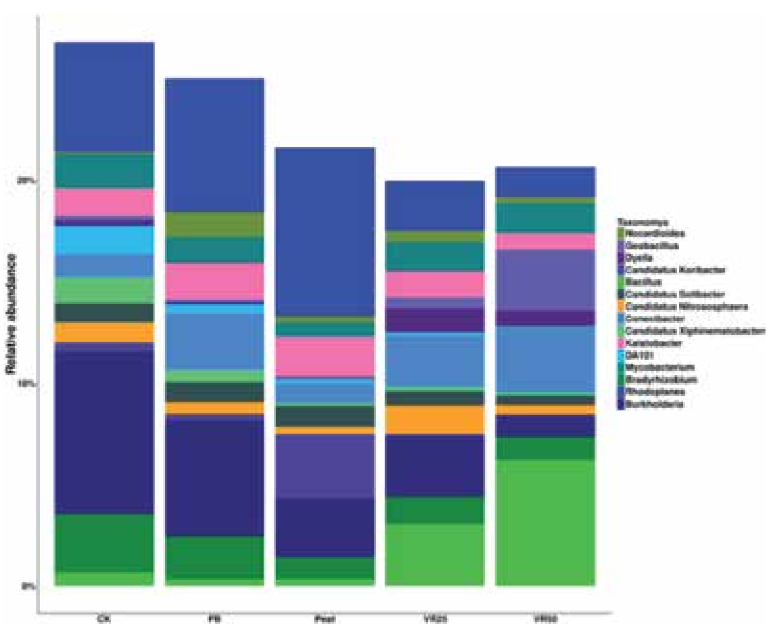

Figure 6.
Relative abundance of rhizosphere microbial community at genus level.

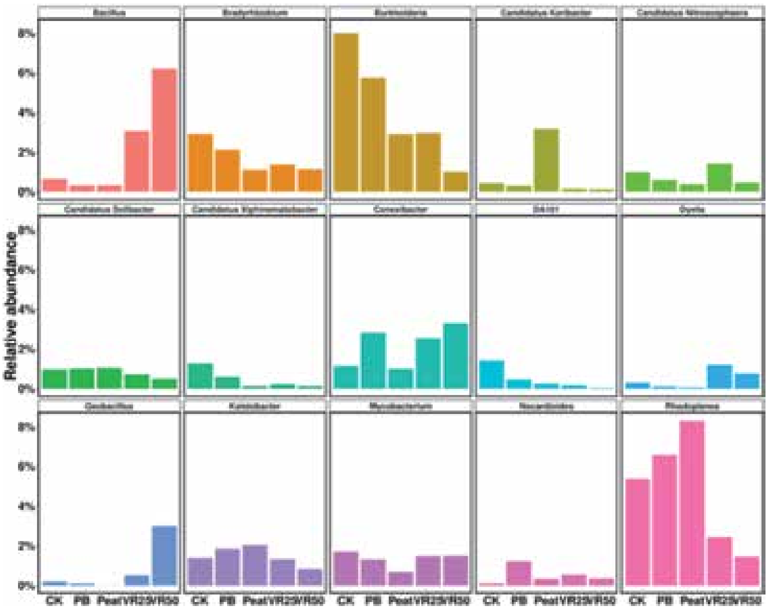

Figure 7.
Comparison of relative abundance of microorganisms at genus level.

	Chao1	Observed species	Shannon	Simpson
CK	2339 ± 159b	1407 ± 74c	7.83 ± 0.38ab	0.980 ± 0.014a
VR25	2300 ± 106b	1379 ± 61b	8.03 ± 0.14b	0.987 ± 0.003a
VR50	2108 ± 165a	1221 ± 119a	7.50 ± 0.48a	0.973 ± 0.014a

Different lowercases in the same column indicate the significant difference at 0.05 level (P < 0.05).

Table 7.
Effect of vinegar residue on the rhizosphere microbial diversity of blueberry.

From the observed species, Shannon and Simpson index decreased significantly in VR2 (**Table 7**), indicating that rhizosphere microbial diversity of blueberry decreased by the application of 50% vinegar residue.

4. Discussion and conclusion

Du et al. [30] found that vinegar residue compost amendment could promote the growth of cucumber and could be applied as a method for biological control of cucumber Fusarium wilt. Our results showed that vinegar residue could promote the growth of blueberry and increase the biomass accumulation of shoot and root, and root dry weight of blueberry under VR2 increased by 83.94% compared with CK. Previous studies have found that the application of organic materials could increase the reducing sugar content and reduce the titratable acidity content of fragrant pear fruit [31] and increase the content of Vitamin C (VC) and soluble solids in tomato fruit [32]. This research showed that the application of vinegar residue could significantly increase the soluble solid content of blueberry fruits. In addition, the total phenol and flavonoid content of the fruits of VR2 increased by 71.42 and 100.00%, respectively, compared with CK, indicating that the application of 50% vinegar residue had the best effect on promoting the quality of blueberry fruits.

Blueberry is suitable to grow in soil with abundant organic matter and pH under 5 (below 5.5) [1]. Soil pH of VR2 is 4.62, which is quite suitable for the growth of rabbiteye blueberry. Soil conductivity reflects the water soluble salt content. And soil conductivity of VR2 was 224.80 $\mu S{\cdot}cm^{-1}$, which was 102.71% higher than CK, but did not exceed the suitable range of soil conductivity for blueberry growth [33]. The exchangeability of soil salt ions and nutrient separators was higher with the increase of soil conductivity at a certain range, which is more conducive to nutrient absorption of blueberry. In addition, soil bulk density was decreased in VR2, which indicated that the application of vinegar residue could increase soil porosity. And soil pH, electrical conductivity, and bulk density were the key factors to promote the growth of blueberries by applying vinegar residue. The results showed that VR2 treatment significantly increased root-shoot rate by 72.00% and root nitrogen by 23.89% compared with the control, suggesting that vinegar residue could significantly promote root growth.

Previous studies reported that adding organic materials can significantly increase soil nutrient content and enhance soil biological activity [34]. Our results showed that the application of vinegar residue could significantly increase the content of total nitrogen, total phosphorus, and organic matter in soil. Soil nitrogen content of VR2 increased by 239.01% compared with CK, and soil organic matter content increased by 43.6 g/kg with the application of 50% vinegar residue. Improvement of soil organic matter could ameliorate soil structure and serve as resource of organic nitrogen and phosphorus. What is more, it could also enhance the activity of soil microbial. Soil urease and acid phosphatase are important enzymes for the transformation of soil nitrogen and phosphorus. Goyal et al. [35] found that the application of exogenous organic materials could improve the activity of soil urease and phosphatase and thus improve soil fertility. This study found that activities of urease and acid phosphatase were highest in VR2, with 26.60 and 22.87 $g{\cdot}kg^{-1}\ h^{-1}$, respectively, 533.33 and 95.47% more than the control. The results showed that the application of vinegar residue increased the activity of soil urease and acid phosphatase, thus increased the transformation of soil nitrogen and phosphorus, and then promoted the growth of blueberry [36, 37].

In conclusion, the application of vinegar residue could improve soil structure and increase soil nutrient and organic matter content, thus promoting the growth and fruit quality of blueberry. In the meantime, the application of 50% vinegar residue appeared to have the best effect on promoting blueberry growth.

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Section 4

Ficus

Chapter 6

Fig Production and Germplasm in Turkey

Emine Tanriver

Abstract

Turkey is one of the most important genetic origins of fig (*Ficus carica* L.) in the world, and it extended to the Mediterranean countries (Spain, Italy, France, Greece, Tunisia, Morocco, Algeria, Portugal.), USA, Syria, Iraq, Iran, Saudi Arabia, South Caucasia, and Crimea. Fig germplasm in Turkey is located mainly at the Big Meander Valley and Small Meander Valley in the Aegean Region but also widely seen in the Southeast Anatolia, the Marmara, and the Mediterranean regions. Siirt, Bottan, Diyarbakir, Elazig, Gaziantep, Besni, Kahramanmaras, Ceyhan, and Ahir Mountain are the main fig germplasm locations at the Southeast Anatolia and the Mediterranean regions. These germplasm (285 fig cultivars and genotypes) are mainly collected at the orchards of Erbeyli Fig Research Institute in Aydın Province. Fig production of Turkey is about 305.689 tons in 1.152.799 tons of world fig production. Turkey is the biggest fig-producing country and is followed by Greece, the USA, Italy, Portugal, and Spain. Dried figs (mainly Sarilop cultivar) are obtained from Aydin Province in the Aegean region, while fresh figs are obtained from the Marmara and Mediterranean regions.

Keywords: Turkey, fig, germplasm, production

1. Introduction

There are eight germplasm centers in the world which are (1) China; (2) India, Malaysia, and Thailand; (3) Middle Asia; (4) Near East; (5) Mediterranean; (6) Ethiopia; (7) South Mexico and Middle America; and (8) South America. Turkey is located both very close to the Near East and the Mediterranean basins [1, 2]. This shows the importance of Turkey as a plant germplasm center. The reason of this rich germplasm in Turkey is mainly based on the suitable ecological conditions for horticultural plants, the presence of the country on the migration routes, and Anatolia being a place where many civilizations have occurred since the first ages of the history. So, Turkey is one of the main origins of many fruit species as well as fig which was especially located at the Anatolian part of the country [3]. Some of the main fig-growing provinces are given in **Table 1**.

Ficus is a genus belonging to the Moraceae family and contains more than 800 species, among which *Ficus carica* is one of the most important edible ones [4]. Fig culture in Anatolia has begun from the very beginning of the cultural history of the human being. Fig (*Ficus carica* L.) is extended from Anatolia to the Mediterranean countries, Syria, Iraq, Saudi Arabia, South Caucasia, and Crimea. *Ficus* also originated in Egypt, India, and South China. California in the USA, South and South West Africa, and East Australia are the later cultural centers of *Ficus carica*. Fig is a special fruit called syconium, grown in temperate, subtropical, and tropical climates.

Provinces	Area (da)	Production (ton)	Yield per tree (kg)
Aydın	365.366	182.775	30
İzmir	80.778	43.741	29
Bursa	21.136	25.734	76
Mersin	3.756	7.202	62
Hatay	1.677	6.535	30
Antalya	1.377	4.319	36
Gaziantep	7.226	2.913	42

Table 1.
The production area and the yield in fig-growing provinces.

No	Name	No	Name	No	Name	No	Name
1	Göklop	30	Ada	59	Yediveren	88	Darpak
2	Bardakçı	31	Datça 5	60	Kızıl yemiş	89	Kızıl mor
3	Kuşadası Bardakçı	32	Siyah incir	61	Gök incir	90	Ekşi incir
4	Mor 1	33	Yediveren	62	Güzlük mor	91	Beyaz incir3
5	Kara Yaprak	34	Siyah	63	Haziran inciri	92	Siyah incir
6	Akça 2	35	Susak	64	Kış hayrı	93	Şeker inciri2
7	Akça 3	36	Sarı yemiş	65	Halebi	94	Melli
8	Bardak	37	Gelin yanağı	66	Azezi	95	Abbas
9	Mor 4	38	Dereköy	67	Sarı incir	96	Mut
10	Asıl bardak	39	Löp inciri	68	İstanbul inciri	97	Yeyişgüz
11	Sarılop	40	Midilli	69	Ak incir	98	Beyazorak
12	Morgüz	41	Mor incir	70	Kara sultani	99	Siyahorak
13	Karabakunya	42	Bodrum inciri	71	Baldırcın	100	Divrek kara
14	İzmir Bardacık 2	43	Armut sapı	72	Deniz inciri		
15	Bursa Siyahı	44	Mor armudi	73	Tabak inciri		
16	Sultan Selim	45	Kızıl mor	74	Turnaboyu		
17	Yediveren	46	Kırmızı incir	75	Dilaver		
18	Beyaz Seyhan	47	Kış inciri	76	Ham incir		
19	Şeker inciri	48	Yayladağ	77	Ak incir 1		
20	Morgüz	49	Aşı inciri	78	Kara incir 2		
21	Sarı çiçek	50	Osmaniyeli	79	İpek inciri		
22	Siyah kış	51	Lebi	80	Değirmen inciri		
23	Esmer bal	52	Tarak inciri	81	Ağarsak		
24	Siyah lop	53	Fetike inciri	82	Sarı incir2		
25	Datça 1	54	Lop inciri	83	Beyaz incir2		
26	Şeker	55	Gök incir	84	Kara incir3		
27	Mor bardakçı	56	Frenk inciri	85	Kabak inciri		
28	Beyaz incir	57	Kilis inciri	86	Kilis inciri1		
29	Gökçe	58	Beyaz yemiş	87	Sarı kilis		

Table 2.
Some fig cultivars at the collection plantations at Erbeyli Fig Research Institute.

Fig is a sacred fruit from the very historical times in three of the religions. Fig has high nutritional and medicinal value with the phenolic compounds, antioxidant content, vitamins, amino acids, minerals, etc.

In Turkey, fig culture is seen in almost every part of the country except the very continental climatic areas of the Middle and East Anatolia. Southeast Anatolia is very rich with fig germplasm. High extension of *Ficus carica* in Anatolia, from BC, caused the occurrence of wild genotype *F. carica* erinosyce and two other cultural forms, *F. carica* caprificus (male fig) and *F. carica* domestica (female fig). There are some other *Ficus* species and forms in Anatolia such as *F. palmata* Schweinf; F. c. var. rupestris, Hauska; *F. carica* var. kurdica, Kotschy; *F. carica*. Var. domestica, Tschirch; *F. carica* var. riparium, Hauskn; and *F. carica* var. Johannis, Boiss [5].

Almost all the coastal lines of the country such as Aegean, Mediterranean, Marmara, and Black Sea regions are the main areas for fig production. However, the best dry fig is grown at the Big Meander Valley and Small Meander Valley of the Aegean Region with a wonderful quality. In this area there is an institute (Erbeyli Fig Research Institute) of the Ministry of Agriculture and Forestry mainly carrying on *Ficus* research at the orchards with 285 fig cultivars and genotypes (**Table 2**).

According to the statistical data, among 1.152.799 tons of world fig production, Turkey takes the first place with 305.689 tons of total fig production. It was followed by Egypt with 176.000 tons, Algeria with 128.620 tons, and Morocco with 126.554 tons. The amount of dried figs are 72.000 tons among 127.500 tons of world dry fig production [1]. Dry fig exportation of Turkey is 42.227 tons, and supply is more than 60% of dry fig demand of the world. Total fig exportation of Turkey was about 63.7% of the world total fig exportation [6]. The area, production, and yield per tree considering the main fig-growing provinces are given in **Table 2** [7].

Some of the nutritional values of dry fig are 11.6 g carbohydrates, 1.2 g protein, 0.5 g fat, 1,8 g fiber, 6.4 mg vitamin A, 2.0 mg vitamin C, and 0.5 mg iron in 20 g of dry fig fruit.

2. Pomological classification and cultivars

Fig is divided into three groups for the fertilization biological characters: (a) male figs, (b) dried figs, and (c) fresh figs. The male figs take part in *F. carica* caprificus; the other two female figs take part in *F. carica* domestica.

a. Male figs contain both male (for pollen production) and short-styled female (gall) flowers, in which fig wasp (*Blastophaga psenes* L.) is grown up. Male fig fruits cannot be eaten, only used for pollen and fig wasp production.

b. Dried figs contain only long-styled female flowers in the receptacle of the syconium. This group of fig cultivars need pollination for the fruit set (Smyrna type, both breba and main crops require caprification). Sarilop fig cultivar is the main cultivar of this group.

c. Common or fresh figs contain only long-styled female flowers; however, this group of figs does not require pollination; both breba (spring crop) and main (summer crop) crops are parthenocarpic. However, some cultivars in this group need pollination for summer or autumn crop. San Pedro (while breba crop does not require caprification), main crop requires caprification [8–11]).

Caprification is to hang a profichi syconium on the branches of female trees for pollination. Caprification is very important in Smyrna-type figs. Fig wasp

(*Blastophaga psenes* L.) which is a pollination vector has a symbiotic life cycle with *F. carica*. Caprification is compulsory for cultivars like Sarılop (Smyrna type) to set economically satisfying fruit [12–14].

3. Fig research and selection studies

In the Aegean part of the country, fig is mostly grown for dry production and exportation. So, most of the research in this area was carried on the performances of dry figs, mainly on Sarilop fig cultivar. However, the increase on the exportation and transportation possibilities of fresh figs caused to work on them (such as Bursa Siyahi fig cultivar) as well. Many studies were carried out to develop fig cultivation in the country. Kaska et al. [15, 16], Kuden and Tanriver [17], and Tanriver et al. [18] worked on fertilization biology and physiology of selected fig genotypes under Cukurova conditions.

First fig selection studies have begun from the end of the 1980s with Aksoy [19] and in the 1990s with the studies of Aksoy et al. [20] at Ege University and Erbeyli Fig Research Institute and Kaska et al. [13] at Cukurova University. Kuden et al. [21], collected 38 fresh commercial fig cultivars from Southeast Anatolia region and 32 fresh commercial fig cultivars from the East Mediterranean region. They have also carried out another selection study on figs and collected and identified very promising genotypes [22]. Ilgin and Kuden [23] also identified fig germplasm in Kahramanmaras Province. All these selection studies were presented during the MESFIN network meeting [24].

Several Ms.C theses were carried out on fig selection [25–41] and also Ph.D. studies [19, 42–46]. Several selection, germplasm characterization, and cultivar identification studies were carried out by different scientists [47–50].

4. Conclusion

Fig (*Ficus carica* L.) is mainly spread in the subtropical climatic countries in the world. However, in Turkey fig is grown not only in subtropical climates, but it could be grown in less subtropical or even mild continental climatic areas. Anatolia is one of the oldest genetic origins of figs. Cultural forms of *Ficus carica* erinosyce, *Ficus carica* domestica, and *Ficus carica* caprificus are located especially in the Aegean Region having parallel life cycles with wasp (*Blastophaga psenes*) to have a special fertilization biology for figs. Three receptacle bearings occur both in male (profichi or spring crop, mammoni or summer crop, mamme or winter crop) and female (fiori or spring crop, pedagnuoli or summer crop (maincrop), cimaruoli or autumn crop). Fig wasps live inside the gall flowers in male figs and become adult and fly away from them taking pollen and enter inside the female flowers and cause the pollination.

In conclusion, Turkey has the best ecological conditions for fig production. There are 285 fig cultivars and genotypes under protection at the Ministry of Agriculture and Forestry and Erbeyli Fig Research Institute in Aydin Province. These elite plant materials were collected from the different parts of the country, since Anatolia has been the main genetic origin of fig (*Ficus carica* L.).

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Chapter 7

Chemical and Biological Characteristics of *Ficus carica* L. Fruits, Leaves, and Derivatives (Wine, Spirit, and Liqueur)

Raquel Rodríguez Solana and Anabela Romano

Abstract

Ficus carica L. is a native plant to Southwest Asia and widely spread from ancient times in the Mediterranean region. Its fruits (figs) and leaves present important nutritional components (vitamins, minerals, sugars, amino acids, etc.) and health-related effects due to their phytochemical composition. Numerous bioactive compounds, such as phenolic compounds (phenolic acids), flavonoids (flavonols, flavones, and anthocyanins), coumarins, sterols, and volatiles (monoterpenes, sesquiterpenes, norisoprenoids, ketones, alcohols, esters, etc.), among others, have been isolated from fruits and leaves of *F. carica* that are the main ingredients used in the production of different alcoholic beverages such as wine, liqueur, and spirit. This chapter aims to review the different chemical and biological characteristics found both in raw materials (fruits and leaves) and in the final product (wine, liqueur, and spirit) that have been consumed and known throughout human history.

Keywords: antioxidant capacity, biological activities, enzyme inhibitory activity, fig fruit, fig leaf, fig liqueur, fig spirit, fig wine, phenolic content, volatiles

1. *Ficus carica* L. fruits, leaves, and derivatives (wine, spirit, and liqueur)

Ficus carica L. is one of the oldest plants cultivated by humans [1]. It is native to the Southwest Asia and spread worldwide in places with typically mild winters and hot dry summers [2]. Fruits and leaves have been widely used as valuable food for people and as folk medicine due to their therapeutic effects [1, 3]. According to the FAO (2013–2017), most of the world's fig production occurs in the Mediterranean basin. The 10 main world producers include countries such as Turkey, Egypt, Morocco, Algeria, Iran, and Syrian Arab Republic. Spain is the only European country included in the list, and the American countries, United States of America and Brazil, are also included [4].

Since ancient times, *F. carica* has been ever present in different cultures. It was the first tree mentioned in the Bible and the figs the first nourishment of human beings according to the Jewish Talmud. Fig tree was linked to the paradise according to the Islamic culture, and in ancient Greece it was considered a gift from Demeter, the earth mother [5].

Figs and leaves are used in their primary and processed form to produce different traditional and industrial products (infusions, jams, wines, spirits, liqueurs, etc.). The fig is a very perishable product, and for this reason it is mainly utilized as dried fruit [6]. Either way, dry or fresh figs are well known for their nutritive value due to the high contents in minerals (mainly calcium and others like copper, manganese, magnesium, potassium, etc.), fats (source of energy), sugars, and other non-nutritive components such as water, fiber, and antioxidants like phenolic compounds [1, 6, 7]. On the other hand, infusions, decoctions, or other preparations

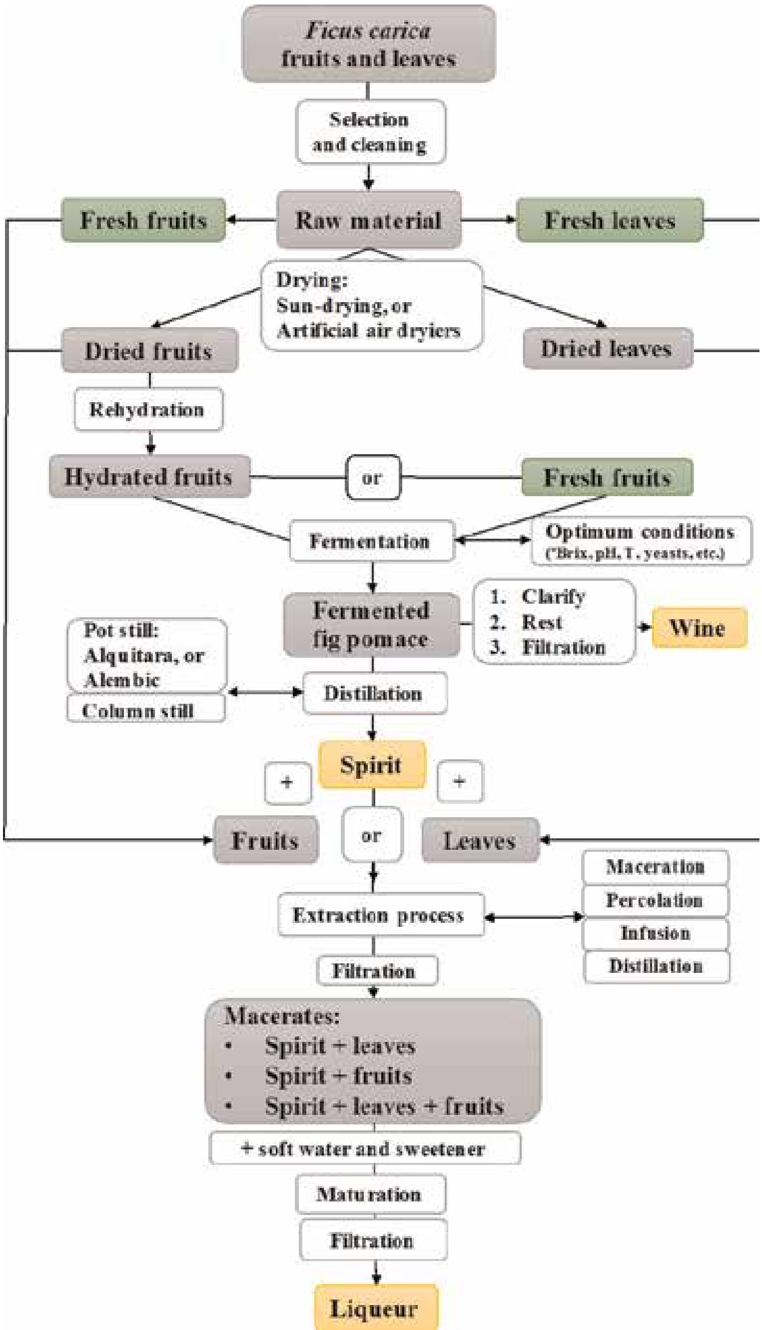

Figure 1.
Manufacturing process of Ficus carica L. wine, liqueur, and spirit from raw materials to the final beverage.

of fig leaves have been traditionally used in the treatment of different diseases due to the therapeutic effects associated with its chemical composition [8]. For all the aforementioned, *F. carica* has been included in occidental pharmacopeias (such as the Spanish and British pharmacopeias) and in therapeutic guides of herbal medicine (including the Physician's Desk Reference (PDR) for herbal medicine) [9].

The study of the extraction of phytochemicals from plants and the use of these compounds as additives has been of great importance as an efficient and safe way to add supplements in foods, to produce what is known as nutraceutical and functional food (food with a relevant effect on health or reduction in disease risk) [10]. More recently, the main objective of *F. carica* phytochemicals extraction has been based not only on finding the best extraction conditions but also the use of green extraction methods such as ionic liquids or deep eutectic solvents, environmentally friendly and sustainable for sample preparation [8]. Despite all the properties and uses found in raw materials, the scarce amount of works related to the chemical composition and biological activities of alcoholic products derived from fig fruits and leaves, specifically, wine [11–13], spirits [14–17], and liqueurs is worth mentioning [18, 19]. Furthermore, despite the mentioned characteristics of the leaves and the great tradition in using this part of the plant in the production of liqueurs [20], this is not reflected in the works cited. An outline of the process followed in the production of these beverages from the raw materials of *F. carica* to the final product can be seen in **Figure 1**. In all these products, the biotechnological step of alcoholic fermentation is required to transform the sugars present in the fruit into alcohol and produce the value-added corresponding alcoholic beverages of commercial importance. The use of yeasts (endogenous or exogenous) and controlled conditions (pH, temperature, etc.) is necessary to obtain a high-quality final product. In addition, during this fermentation process, and other processes taking place such as maceration and maturation of the beverage, new chemical compounds are produced that will contribute to the final profile of the beverages [21, 22].

In the next section, the different chemical compounds (phytochemicals) as well as the biological properties from both *F. carica* raw materials and alcoholic-derived products will be addressed in more detail.

2. Phytochemical composition of fig fruit, leaf, and the alcoholic products, liqueurs, and spirits

Phytochemicals or plant secondary metabolites are non-nutritive plant metabolites which are essential for plant survival and proper growth and reproduction [23]. Many of these components have bioactivities toward animal biochemistry and metabolism with the ability to provide health benefits. *F. carica* plant owns the highest diversity of compounds with the higher quantities of all classes of compounds (except aldehydes and monoterpenes) mainly in leaves, followed by fruits pulps and peels [2, 9, 24].

Phytochemical studies on raw materials (fruits and leaves) and derived products (wine, liqueur, and spirit) of *F. carica* revealed the presence of numerous bioactive compounds including volatiles, organic acids, phytosterols, triterpenoids, fatty acids, phenolic acids, flavonoids, coumarins, and few other classes of secondary metabolites shown in **Figure 2**.

2.1 Volatile compounds

Aroma is an important attribute of the sensory appreciation of a product and is usually used as a criterion for its quality assessment. It is a defining element of the distinct flavor of individual foods. The ripening period has an important role in the volatile composition, and many volatiles are produced during different developmental stages of plant tissues such as flowering, ripening, or maturation [1]. These volatiles are known as primary aromas, and they are responsible for varietal aromas [25]. These compounds are accumulated in plant storage sites and are released from the surface of the leaf, making this part of the *F. carica* the largest holder of compounds (except aldehydes and monoterpenes, in highest amounts in fruits [9]). On the other hand, in products such as wine, spirits, and liqueurs elaborated from fruits of *F. carica*, other types of aromas come from the different processing steps. Secondary aromas (the greatest pool of volatiles) are mainly produced by yeast as metabolism by-products, while tertiary aromas of finished alcoholic beverages are compounds that illustrate the changes made in the sample matrix during the storage and maturity stages [25].

Fruits [1, 2, 26, 27] and leaves [2, 8] of *F. carica* as well as derived products such as the alcoholic beverages, fig liqueurs [28], and spirits [14, 15, 17] consist of various volatile compounds which are identified and distributed by distinct chemical classes, such as terpenes (monoterpenes and sesquiterpenes), alcohols, aldehydes, ketones, esters, and miscellaneous compounds.

2.1.1 Terpenes

Terpenes, such as monoterpenes (C10) and sesquiterpenes (C15), are the largest class of plant secondary metabolites, as can be seen in **Figure 2**. The high vapor pressures of these compounds, at normal atmospheric conditions, allow their significant release into the air [1]. Monoterpenes such as linalool (1) and epoxylinalool (34) (more important than linalool) are related with their important role in the attraction of specific pollinators, the fig/wasp linkage [1]. Although sesquiterpenes represented just the ~3% of the total volatiles in Tunisian cultivars, it was the main class of compounds identified in leaves, and germacrene D, β-caryophyllene, and τ-elemene are the major compounds detected [9].

α-Pinene (31), one of the main monoterpenes mentioned in different works, has only been found in fruits, while the sesquiterpenes β-elemene (39), β-cubebene (60), α-ylangene (61), β-bourbonene (62), (+)-ledene (viridiflorene) (66), and α-gurjunene (67) are compounds exclusively identified in leaves [1, 2, 9]. The monoterpenes citronellol acetate (7), (E)-geranyl acetone (12), (+)-sylvestrene (16), *p*-mentha-1,3,8-triene (17), cumene (26), *o*- and *p*-cymene (27, 28), and nerol oxide (33) and the sesquiterpenes (E)-nerolidol (35), farnesyl acetate (36), α-curcumene (37), β-bisabolene (38), τ-elemene (40), (−)-δ-cadinol (45), cadina-1 (10),4-diene (48), cadalene (49), α-calacorene (50), valencene (51), acoradiene (53), δ-guaiene (α-bulnesene) (55), α-guaiene (56), isocaryophyllene (57), γ-patchoulene (65), and α-cedrene (68) are compounds only identified in Portuguese monovarietal fig spirits [14]. Linalool acetate (2), geraniol (4), (Z)-8-hydroxylinalool (5), neral (β-citral) (6), geranyl vinyl ether (8), ethyl linalool (9), nerol (10), dihydrocitronellol (11), geranial (α-citral) (13), ocimene (14), *p*-menth-3-ene (19), α-terpinolene (21), α-terpineol (23), pulegone (24), isodihydrocarveol (25), borneol (30), and (Z)- or (E)-linalool oxide (34), as monoterpenes, and cadina-1,4-diene (47) and dihydroactinidiolide (52), as sesquiterpenes, were identified in synthetic liqueurs elaborated from different Greek *F. carica* varieties [28].

On the other hand, common terpenes were found in different *F. carica* parts and/or fig spirits and synthetic liqueurs. Menthol (18), τ-muurolene (44), and τ-cadinene (46) are common compounds found in fruit and leaves [2, 9]. The

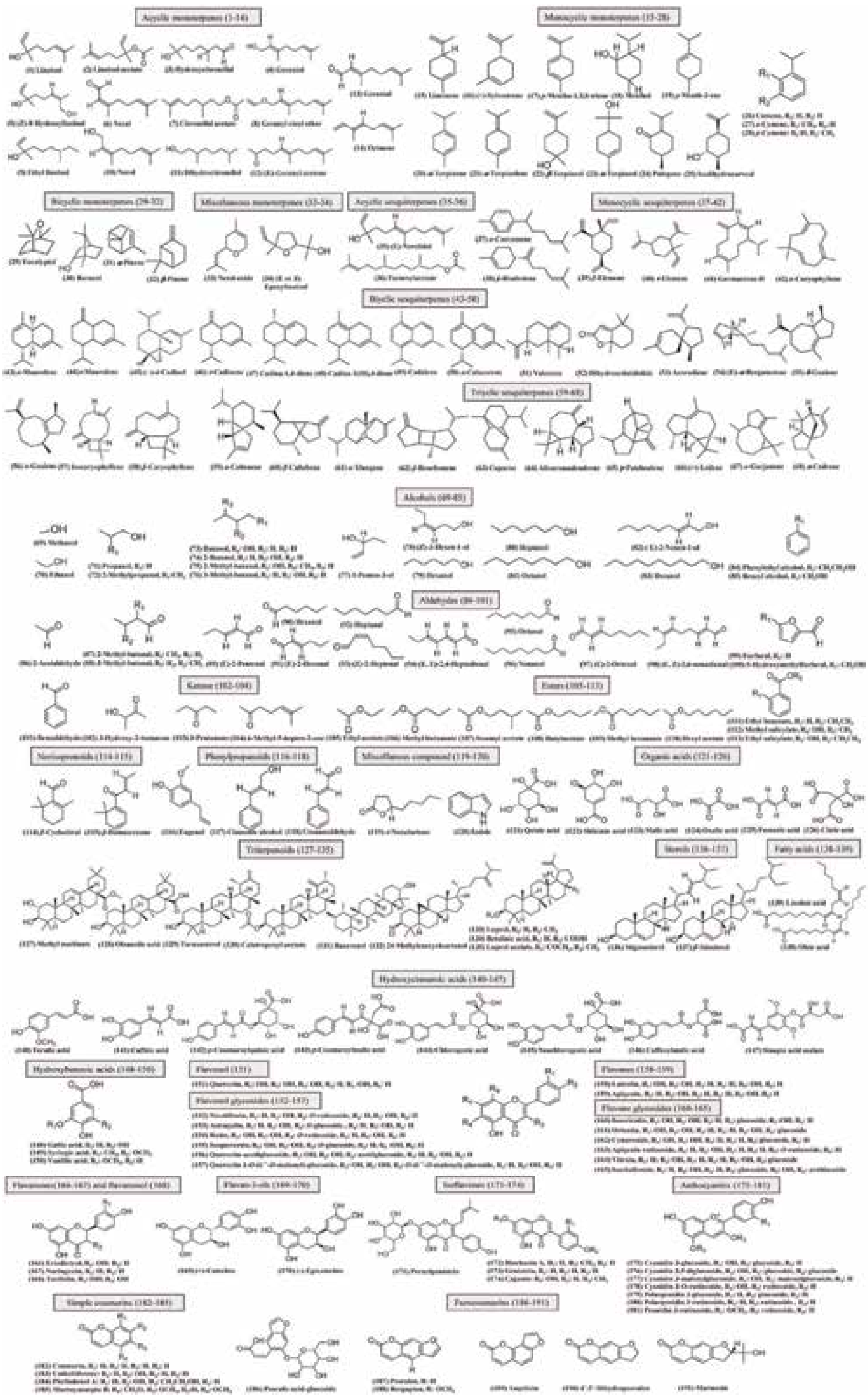

Figure 2.
Chemical structures of different phytochemicals (volatile compounds, organic acids, triterpenoids, sterols, fatty acids, phenolic acids, flavonoids, and coumarins) present in fig fruits, leaves, spirits, and liqueurs.

monoterpenes β-pinene (32) and eucalyptol (29) and the sesquiterpene (E)-α-bergamotene (54) were identified in different fig cultivars from different countries and also in fig liqueurs, while the monoterpenes linalool (1) and linalool oxide (furanoid) (epoxylinalool) (34) were isolated in fig fruits and spirits [2, 9, 28].

Other common terpenes were identified in different fig samples such as α-terpinene (20) in fig spirits and synthetic liqueurs; limonene (15), α-cubenene (59), copaene (63), and germacrene D (41) in fig fruits, leaves, and spirits; α-guaiene (57), aromadendrene (64), and α-muurolene (43) in fig leaves and spirits; and finally, β-caryophyllene (58) in fruit, leaves, spirits, and synthetic liqueurs, while α-caryophyllene (42) in fig leaves, spirits, and synthetic liqueurs [1, 2, 9, 14, 28].

2.1.2 Alcohols, aldehydes, ketones, and esters

Alcohols, ketones, and esters are the more developed compound classes in ripened fruits, representing 41% of total aroma [1].

Different alcohols were identified in fruits [(Z)-3-hexen-1-ol (78)], leaves [2-methyl-1-butanol (76) and 1-heptanol (80)], spirits [methanol (69), ethanol (70), 1-propanol (71), 1-butanol (72), 2-methyl-propanol (isobutyl alcohol) (74), 1-hexanol (79), octanol (81), and decanol (83), among others], in both fruits and leaves [1-penten-3-ol (75), benzyl alcohol (85), and (E)-2-nonen-1-ol (82)] [2, 9], leaves and spirits [1-heptanol (80)] [9, 14], and finally in raw materials and spirits [3-methylbutanol (77), phenylethyl alcohol (84)] [2, 9, 14, 15]. Methanol (69), a toxic compound formed by hydrolytic demethoxylation of esterified methoxyl groups of the pectin polymer by pectic enzymes, with a marked maximum methanol content in fruit spirits of 1500 g/hL of pure alcohol (Regulation (EC) No 110/2008), was present in fresh and dried fig spirits [14]. It should be emphasized that its concentration depends on the technological characteristics of the manufacturing process. Higher quantities were found in spirits prepared from fresh figs because this compound is naturally present in fruits and decreased in spirits prepared using fermentations with immobilized yeast cell technology [15]. In addition, greater amounts of higher alcohols [2-butanol (73) + 1-propanol (71) + 2-methyl-propanol (74) + butanol (72) + 2-methylbutanol (76) + 3-methylbutanol (77)] were also found in samples of spirits made from dried figs (approx. > 350 g/hL absolute alcohol), being indicative of worse quality of these samples [14].

The aldehydes present exclusively in fruits are heptanal (92), octanal (95), nonanal (96), 2-methyl-butanal (87), (Z)-2-heptenal (93), (E, E)-2,4-heptadienal (94), (E)-2-octenal (97), and (E, Z)-2,6-nonadienal (98) [2, 9]. Furfural (99) and 5-hydroxymethylfurfural (100), toxic compounds originated during the fired pot-still distillation process at high temperatures, and acetaldehyde (86) were identified in spirits [14, 15]. Meanwhile, common aldehydes identified in fruits and leaves were 2-methyl-butanal (87), 3-methyl-butanal (88), (E)-2-pentenal (89), hexanal (90), and (E)-2-hexenal (91); benzaldehyde (101) was present in fruits and spirits [2, 9].

The first major compound found in non-pollinated and pollinated figs was the ketone 3-hydroxy-2-butanone (acetoin) (102) [1]. Other ketones, 6-methyl-5-hepten-2-one (104) and 3-pentanone (103), were identified in, respectively, fruits (pulps and peels) and leaves of Portuguese fig varieties [2, 9].

Esters are the major contributors to fruit aroma and are the most important in ripe figs. They are produced through the esterification of alcohols and acyl-CoAs derived from both fatty acid and amino acid metabolism, in a reaction catalyzed by the enzyme alcohol-o-acyltransferase [1]. These compounds are much less developed in non-pollinated fruits, and its content decreases when using immobilized cell application during the fermentation process. In spirits, esters represent the largest class of volatiles (∼95% of total volatiles), particularly the fatty acid ethyl esters such as ethyl decanoate, ethyl octanoate, and ethyl dodecanoate. The second and third major compounds identified in fruits from Tunisian varieties were butyl acetate (108) and isoamyl acetate (107) with banana odor [1]. This last compound was also present in fig spirits [14]. Other ester identified in fruits was ethyl salicylate (113). Methyl butanoate (106), hexyl acetate (110), and ethyl benzoate (111) were found in leaves [2, 9], while methyl hexanoate (109) was a common compound in fruits and leaves and methyl salicylate (113) in fruits, leaves, and spirits [2, 9, 14]. Many methyl and ethyl esters and other esters were identified in fig spirits and are common to other alcoholic beverages. The study comparing dried and fresh fig spirits showed that dried fig spirits presented ethyl acetate (105) in higher proportion than fresh fig spirits. This compound results from the growth of acetic acid bacteria during the fermentation in aerobic conditions [14].

2.1.3 Miscellaneous compounds

The norisoprenoid β-cyclocitral (114) present in leaves and fruits [2, 9] and β-damascenone (115) characteristic of different fig spirits and synthetic fig liqueurs [14, 28] and finally the phenylpropanoids [(eugenol (116), cinnamic alcohol (117), cinnamic aldehyde (118)] and indole (120) in fruits [9] and s-nonalactone (119) [2] in leaves were other volatile compounds detected in different fig samples.

2.2 Organic acids, phytosterols, triterpenoids, and fatty acids

Some organic acids isolated from fruits and leaves of *F. carica* were the shikimic (122), malic (123), oxalic (124), fumaric (125), and citric (126) acids [2, 9, 24, 29], while the quinic acid (121) was reported only in leaves [2, 9].

Phytosterols are found in most plant foods, with the highest concentrations occurring in vegetable oils. Sterols (modified triterpenes) like β-sitosterol (137) [24] and the triterpenoids methyl maslinate (127), oleanolic acid (128), taraxasterol (129), w-taraxasterol ester, calotropenyl acetate (130), bauerenol (131), 24-methylenecycloartanol (132), lupeol (133), and lupeol acetate (135) have been reported in fig leaves [2, 9], and betulinic acid (134) in fruits [3], while stigmasterol (136) was reported in both [29, 30].

Dried and fresh fruits of *F. carica* showed polyunsaturated fatty acids with 84 and 69% of total fatty acids, respectively. Linoleic acid (139), in fresh and dried fruits, was the only polyunsaturated fatty acid identified. With respect to monounsaturated fatty acids, oleic acid (138) is the most abundant in fruits [9].

2.3 Phenolic acids, flavonoids, and coumarins

Among the different chemical structures found in *F. carica*, one of the most important for biological uses is the phenolic compounds. These play many physiological roles in plants and are also favorable to human health [2]. Fruits and leaves presented qualitative differences in phenolic acids. Leaves were richer in phenolic derivatives formed by conjugation with sugars (the hydroxybenzoic derivatives: gallic acid di-pentoside, syringic acid hexoside, vanillic acid hexoside deoxyhexoside, and the dihydroxybenzoic acids hexoside/hexoside pentoside) and organic acids including malic (the hydroxybenzoic derivative, syringic acid malate, and the hydroxycinnamic derivatives, caffeoylmalic acid, coumaroylmalic acid, sinapic acid malate, and ferulic acid malate) and quinic acid (the hydroxycinnamic derivative coumaroylquinic acid) [31, 32]. The signal of hydroxycinnamics was higher in extracts from leaves. On the other hand, in general, free forms of hydroxycinnamic acids such as caffeic acid (141), and the hydroxybenzoic acids, gallic (148) and syringic (149) acids, were only present in fruits [32]. Also the ferulic acid hexoside and the coumaroyl and ferulic hexosides were present in fruits. Moreover, the following compounds were common to both leaves and fruits: the hydroxybenzoic acids, di-/hydroxybenzoic acids and vanillic acid; the hydroxybenzoic derivatives, dihydroxybenzoic acid attached to hexoside/hexoside pentoside/pentoside/di-pentoside, vanillic acid glucoside, and gallic acid di-pentoside; and the hydroxycinnamic acids, ferulic acid (140) and the chlorogenic (3-O-caffeoylquinic acid) (144) and neochlorogenic (5-O-caffeoylquinic acid) (145) acids. The common hydroxycinnamic derivatives present in fruits and leaves were caffeoylquinic acid hexoside, dihydrocaffeic acid hexose, and the sinapic acid hexoside [2, 9, 24, 32].

Flavonols such as quercetin (151) and glycosylated flavonols such as rutin (quercetin-3-O-rutinoside) (154) (major individual phenolic identified in fruits [2]), isoquercetin (quercetin-3-O-glucoside) (155), quercetin 3-O-(6′-O-malonyl)-glucoside (157), quercetin di-deoxyhexoside hexoside, and quercetin O-di-hexoside were confirmed in fresh and dried figs and leaves [32]. Nicotiflorin (kaempferol-3-O-rutinoside) (152) and quercetin-acetilglucoside (156) were reported in fruits, while astragalin (kaempferol 3-O-glucoside) (153) only in leaves [2, 3, 9, 24, 31, 33].

Free flavones such as luteolin (158) and apigenin (159) are present in fig fruits and leaves. Also, the glycosylated flavones, isoorientin (luteolin 6-C-glucoside) (160), orientin (luteolin 8-C-glucoside) (161), cynaroside (luteolin 7-O-glucoside) (162), vitexin (apigenin 8-C-glucoside) (164), isochaftoside (apigenin 6-C-glucoside 8-C-arabinoside) (165), and apigenin 6-C-hexose-8-C-pentose [which could be identified as schaftoside (apigenin 6-C-glucoside 8-C arabinoside)], were detected in both plant parts. However, apigenin 7-rutinoside (163) and luteolin 6C-hexose-8C-pentose were present in fruits [2, 9, 33].

Another group of flavonoids identified was the flavanones, with the compounds eriodictyol (166) and eriodictyol hexoside in fruits and naringenin (167) in fruits and leaves. The flavanonol taxifolin (dihydroquercetin) (168) was identified in fruits [32]. The flavanols, (+)-catechin (169) in fruits and leaves and (−)-epicatechin (170) in leaves, were also identified [3, 33].

Genistein (173) and hydroxygenistein methyl ether malonylhexoside in leaves and prenylhydroxygenistein, prenylgenistein (171), biochanin A (genistein 4′-methyl ether) (172), and cajanin (7-methoxy 2′-hydroxy genistein) (174), in fruits and leaves, were the isoflavones identified [2, 3, 9, 24, 31, 33].

Different anthocyanin pigments, some of them containing cyanidin or pelargonidin as aglycones, as well as rutinose and glucose substituting sugars and acylation with malonic acid, were found in skin and pulp from different varieties of Iberian fresh figs with different colors (black, red, yellow, and green). These compounds include (epi)-catechin-(4-8)-cyanidin-3-glucoside, (epi)catechin-(4–8)-cyanidin-3-rutinoside,(epi)catechin-(4,8)-pelargonidin 3-rutinoside, 5-carboxypyranocyanidin-3-rutinoside, cyanidin-3-malonylglicosyl-5-glucoside, cyanidin-3-malonylglucoside, cyanidin-3-glucoside (175), cyanidin-3,5-diglucoside (176), cyanidin 3-O-rutinoside (as the main anthocyanin in different commercial fig varieties [2]) (178), pelargonidin-3-glucoside (179), pelargonidin-3-rutinoside (180) and peonidin-3-rutinoside (181). In addition, 5-carboxypyranocyanidin-3-rutinoside, a cyanidin 3-rutinose dimer, and five condensed pigments containing C–C linked anthocyanins and flavanol (catechin and epicatechin) residues were identified [9].

Coumarin (182); the hydroxycoumarins esculetin hexoside, dihydroxy-coumarin, umbelliferone (7-hydroxycoumarin) (183), and prenyl-7-hydroxycoumarin; and the furocoumarins psoralen (187) and bergapten

(5-methoxypsoralen) (188) were isolated from *F. carica* fruits and leaves [32]. Simple coumarins 6-carbaxyl-umbelliferone, phellodenol A (184), and murrayacarpin B (185) and the furocoumarins hydroxypsoralen, hydroxypsoralen hexoside, 4′,5′-dihydropsoralen (190), angelicin (isopsoralen) (189), isopentenoxypsoralen, oxypeucedanin, psoralic acid glucoside and marmesin (191) were isolated from leaves [2, 9, 31, 32].

3. Biological studies in fruit, leaf, and fig spirits and liqueurs

The leaves and fruits of *F. carica* are important in traditional medicine [24]. Many biological activities have been evaluated and confirmed on *F. carica* extracts, and the bioassay-guided fractionation in most cases allowed to assign the chemical structures responsible of such biological effects, thereby ratifying some of its folkloric uses [9]. In this section we analyzed the potential health-promoting constituents of fig fruits, leaves, and derived products, fig liqueurs, and spirits [6].

3.1 Antioxidant capacity

Among the different phytochemicals studied in *F. carica*, phenolic compounds are among the most important with antioxidant capacity (AC). Many of these compounds are able to act as antioxidants by different ways: reducing agents, hydrogen donators, free radical scavengers, singlet oxygen quenchers, and so forth [2].

3.1.1 Fig spirits and liqueurs

The antioxidant capacity (AC) by ABTS [2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonate)] and DPPH (1,1-diphenyl-2-picrylhydrazyl) assays and the total phenolic content (TPC) by Folin–Ciocalteu method were evaluated in different fig spirits and liqueurs [34]. Fig liqueurs showed high values of TPC and AC (ABTS), close to the values of other fruit spirits with highest AC such as green walnut, carob pod, and mulberry. Fig spirits presented high (third value of 15 samples) AC by ABTS assay and among the highest TPC values. However, no DPPH scavenging activity was shown for fig liqueurs and spirits.

3.1.2 Leaf extracts

The maximum total flavonoid content (25.04 mg/g) with marked scavenging activities against hydroxyl and superoxide anion free radicals in a concentration-dependent manner were found in ethanolic (40%) leaf extracts of *F. carica* (solid to liquid ratio 1:60 g/mL, temperature extraction of 60°C, and 50 min of ultrasonic treatment) [6].

3.1.3 Fruit extracts

Several works studied the AC of fruit extracts. Extracts from six commercial fig varieties were evaluated for AC by ferric reducing antioxidant method (FRAP) and also for TPC and total flavonoid content (TFC) and amount and profile of anthocyanins. The extracts exhibiting the highest AC contained the highest levels of TPC and TFC and anthocyanins (cyanidin-3-O-rutinoside as the main compound) [2, 6]. In another work, two fruit extracts [water (WE) and crude hot water-soluble polysaccharide (PS)] were evaluated for AC using the *in vitro* scavenging abilities on DPPH, superoxide, and hydroxyl radicals and reducing power assays. Both extracts have notable scavenging activities on DPPH [WE (EC50, 0.72 mg/ml) and PS (EC50, 0.61 mg/ml)], while PS showed highest scavenging activity on superoxide radical (EC50, 0.95 mg/ml) and hydroxyl anion radical (43.4% at concentration of 4 mg/ml) [6].

Ethanolic extracts from the white Beni Maouche Algerian figs were compared with carob pods and holm oak acorns [7]. Fig extracts presented lower efficacy to scavenge DPPH (20.54 ± 0.30%) than ABTS radicals (68.98 ± 0.12%) and higher reducing ability in phosphomolybdenum assay (638.23 ± 0.43 mg GAE/100 g). This extract (73.17 ± 0.16%) also inhibited the formation of the complex Fe^{2+}-ferrozine and was also able to scavenge H_2O_2 efficiently. The extracts from the three fruits evaluated (carob, acorns, and figs) showed no significant differences in nitric oxide (NO) radical scavenging activities [7].

3.2 Reactive oxygen species production, xanthine oxidase inhibition assay, and study of oxidative stress

3.2.1 Fruit extracts

The production of reactive oxygen species (ROS) in the presence of ethanolic fig extract was measured by chemiluminescence using lucigenin. This method is widely used to determine the rate of superoxide radicals in human neutrophils. Fig extract inhibited the chemiluminescence of lucigenin and ROS production and differed from each other according to the concentration of the sample and the incubation time. After 15 min of treatment, the extract tested at the highest concentration (250 μg/mL) seemed to reach its higher level of lucigenin inhibition, value 44% below that obtained with diphenylene iodonium (0.2 mM), the standard selective inhibitor of nicotinamide adenine dinucleotide phosphate (NADPH) oxidase tested [7].

Ethanolic fig extracts were able to inhibit the activity of the enzyme xanthine oxidase (XO), an enzyme that generates reactive oxygen species. Different extract concentrations were evaluated (50, 250, and 500 μg/mL), and at 250 μg/mL, ethanolic extracts presented the best inhibition, although its value is much lower (practically half) of that obtained with the positive control allopurinol, a drug clinically used for gout treatment. The extracts tested at 500 μg/mL showed a decrease in the inhibition of XO activity as the result of its prooxidant effect. The strong correlation coefficients between XO inhibition activity and phenolic compounds and flavonoids demonstrate the inhibition activity of XO [7].

3.2.2 Leaf extracts

Oxidative stress is the disturbance in the balance between the production of reactive oxygen species (free radicals) and antioxidant defenses. The role of these free radicals in the production of tissue damage in diabetes mellitus was studied in rats divided into four groups: streptozotocin-induced diabetic rats, diabetic rats that received a single dose of a basic fraction of *F. carica* leaf extract, diabetic rats that received a single dose of a chloroform fraction of the extract, and normal rats. Antioxidant status was affected in the diabetes syndrome, and *F. carica* extracts showed that they normalize it. Diabetic animals exhibited higher values for erythrocyte catalase activity, plasma levels of vitamin E, monounsaturated and polyunsaturated fatty acids, saturated fatty acids and linoleic acid than that of the control group. Both *F. carica* fractions showed that they normalize the values of the diabetic animal's fatty acids and plasma vitamin E values. They showed statistically significant differences as a function of diabetes with the vitamin E/C 18:2 ratio being normalized by the administration of the chloroform fraction (to 152.1 ± 80.3 μg/mg) and the vitamin A/C 18:2 ratio being raised relative to the untreated diabetic rats by the administration of the basic fraction (91.9 ± 14.5 μg/mg) [2].

3.3 Inhibitory activities of the enzymes α-amylase, α-glucosidase, and pancreatic lipase

3.3.1 Fruit and leaf extracts

The treatment of diabetes and obesity using the inhibition of carbohydrate (α-amylase and α-glucosidase) and lipid (pancreatic lipases)-digesting enzymes is used to reduce the digestion and absorption of carbohydrates and lipids and also to reduce significantly the blood glucose and body fat levels. Ethanolic extracts from fruits and leaves, in relation to hexane, ethyl acetate, and aqueous extracts, presented the higher α-amylase and α-glucosidase and pancreatic lipase inhibitions at the higher concentration tested (500 μg/mL). At this concentration, similar values to that of the standard acarbose were found for α-amylase and α-glucosidase inhibitions in fruit ethanolic extracts [35].

3.4 Antidiabetic, hypocholesterolaemic, and hypolipidemic activities

3.4.1 Leaf extracts

Different works on antidiabetic activity were carried out using methanolic and aqueous leaf extracts [2, 6]. The maximum glucose-lowering effect in induced diabetic rats with alloxan was observed with methanolic extracts at a concentration of 200 mg/kg and after 21 days. At these conditions, results were similar to those obtained with metformin (medication used for the treatment of type 2 diabetes). On the other hand, a clear hypoglycemic effect and reduction of total cholesterol and total cholesterol/HDL cholesterol ratio of the oral or intraperitoneal administration of aqueous leaf extracts in relation to the control group was observed in diabetic rats induced with streptozotocin.

In other work, an 8-week-old rooster's liver with high abdominal fat was used to evaluate the potential of leaf fig extract as food supplement to decrease hepatic triglyceride (TG) content and secretion of TG and cholesterol from the liver. Results showed that the leaf extract reduced the contents to the basal level in a concentration-dependent manner [2]. Another work about the intraperitoneal administration of leaf decoction extracts (50 g dry wt/kg body wt) in hypertriglyceridemia-induced rats with 20% emulsion of long chain triglycerides (LCTG) indicated a decrease in the LCTG content of 84% after 60 min and a reduction of 69% after 2 h. The results suggest the existence of compound/s in fig leaf decoction that influence the lipid catabolism [6].

3.5 Hepatoprotective activity

3.5.1 Leaf extracts

The hepatoprotective activity of methanolic leaf extract in carbon tetrachloride-induced hepatotoxicity in rats was evaluated, and the activity was comparable to that of the known hepatoprotective silymarin [6].

Petroleum ether leaf extract also showed significant reversal of biochemical, histological, and functional changes in oral rifampicin (50 mg/kg)-induced hepatotoxicity in rats [2].

3.6 Anti-herpes simples virus (HSV)

3.6.1 Leaf extracts

Water leaf extract presented low toxicity and directly killing virus effect of HSV on baby hamster kidney fibroblasts (BHK21), primary rat kidney (PRK), and human epithelial (Hep-2) cells with a maximum tolerated concentration (MTC) of 0.5 mg/mL [2]. Ethyl ethanoate and hexane fractions of methanolic extracts also showed anti-HSV-1 effect [24].

3.7 Immunomodulatory activity

3.7.1 Leaf extracts

Administration of ethanolic leaf extract presented immune modulatory activity in cellular and humoral antibody response according to various hematological and serological tests [6].

3.8 Anti-inflammatory activity

3.8.1 Leaf extracts

Different (petroleum ether, ethanolic, and chloroform) leaf extracts showed a significant reduction on carrageenan-induced paw edema in rats and a greater anti-inflammatory effect in relation to indomethacin, a standard nonsteroidal drug used for this effect [6].

3.9 Irritant potential

3.9.1 Leaf extracts

Methanolic leaf extract and isolated triterpenoids [methyl maslinate (127), calotropenyl acetate (130), and lupeol acetate (135)] exhibited irritant potential on open mice ears and were the most potent and persistent irritant effects [2].

3.10 Antimicrobial, nematicidal, and anthelmintic activities

3.10.1 Leaf extracts

Methanolic leaf extracts showed strong antibacterial activities against oral bacteria, *Streptococcus gordonii*, *S. anginosus*, *Prevotella intermedia*, *Actinobacillus actinomycetemcomitans*, and *Porphyromonas gingivalis*, with minimum inhibitory (MIC) and bactericidal (MBC) concentrations of 0.156–0.625 mg/ml and 0.313–0.625 mg/ml, respectively. These antibacterial effects may be related to some phenolic compounds isolated such as flavonoids [6]. Acetone leaf extract possessed antibacterial activity against *Staphylococcus* species and antifungal activity against *Fusarium solani*, *F. lateritium*, *F. roseum*, *Daporuthe nonurai*, and *Bipolaris leersiae* [24]. Leaf extract also showed strongest nematicidal activity against the nematodes

Bursaphelenchus xylophilus, *Panagrellus redivivus*, and *Caenorhabditis elegans* with 74.3, 96.2, and 98.4% mortality, respectively, within 72 h [2].

3.10.2 Fruit extracts

Fruit extract was found useful in protecting from bacterial pathogen attack in tomatoes [6]. Anthelmintics are drugs that either kill or expel infesting helminths living in the gastrointestinal tract or tissues. Helminths cause numerous damages to the host, for example, injury to organs, intestinal or lymphatic obstruction, causing blood loss, depriving it of food, and secreting toxins [36]. The potential of cysteine proteinases extracted from figs as a potential anthelmintic was evaluated. The experiments were carried out *in vitro* using the rodent gastrointestinal nematode *Heligmosomoides polygyrus*. A marked damage was visible within a 2-h incubation period of cysteine proteinases on the cuticle (loss of surface cuticular layers) of adult male and female *H. polygyrus* worms. The results (efficacy and mode of action) proved the potential use of cysteine proteinases as anthelmintics [6].

3.11 Antipyretic activity

3.11.1 Leaf extracts

To evaluate the antipyretic activity, different doses (100, 200, and 300 mg/kg body wt. p.o.) of ethanolic leaf extracts showed significant dose-dependent reduction in normal body temperature and yeast-induced elevated temperature (pyrexia) in albino rats. The antipyretic effect of this extract was comparable to that of the standard antipyretic agent paracetamol at 150 mg/kg body wt., p.o. The effect extended up to 5 hours after drug administration compared to that of paracetamol (150 mg/kg.b.wt., p.o.) [2, 6].

3.12 Antituberculosis activity

3.12.1 Leaf extracts

Colorimetric microplate-based assay of methanolic (80%) leaf extract exhibited effect against *Mycobacterium tuberculosis* strain H37Rv with MIC value of 1600 μg/mL [2].

3.13 Anti-calpain activity

3.13.1 Fruit extracts

Calpains are calcium-dependent enzymes that determine the fate of proteins through regulated proteolytic activity. These enzymes have been linked to the modulation of memory and are keys to the pathogenesis of Alzheimer disease [37]. Calpain activity was examined after treatment of cells with dry extracts. Fig extracts decreased the fluorescence of the fluorogenic calpain substrate tert-butoxycarbonyl-Leu-Metchloromethylaminocoumarin (t-boc-LM-CMAC) and consequently inhibited the activity of calpain. Fig extracts showed the same capacity to inhibit calpain as carob and holm oak acorn extracts. The incubation time (2, 4, and 6 h) and the concentrations tested (25, 100, and 250 μg/ml) had no effect on the inhibitory activity of calpain in the presence of fig extracts. After 2 h of treatment, the extracts already inhibited more than 50% for all the concentrations tested. This inhibitory activity of the studied extracts could be attributed to its chemical composition that contains several antioxidant groups, especially phenolic compounds such as flavonoids and flavonols [7].

3.14 Diuretic activity

3.14.1 Fruit extracts

Ethanolic fruit extracts were evaluated for the diuretic activity on individual rat through the control of the parameters, total urine volume, and urine concentration of Na^+, K^+, and Cl^-. Results showed a marked diuresis of ethanolic fruit extract treatment in rats based on the increase in urine volume and cation and anion excretions [6].

3.15 Immunity activity

3.15.1 Fruit extracts

The immunity activities of crude hot water-soluble polysaccharide (PS) were evaluated using the carbon clearance test and serum hemolysin analysis in mice. The PS (500 mg/kg) had a significant increase in the clearance rate of carbon particles and serum hemolysin level of normal mice [6].

3.16 Antispasmodic activity

3.16.1 Fruit extracts

Fig aqueous-ethanolic extract was investigated for antispasmodic effect (suppression of muscle spasms) on rabbit jejunum preparations. The extracts (0.1–3.0 mg/mL) produced relaxation of spontaneous and low K^+(25 mM)-induced contractions and with insignificant effect on high K^+ (80 mM). Similar results were observed with cromakalim, a potassium channel-opening vasodilator. This spasmolytic activity of *F. carica* fruits is probably due to the activation of $K^+{}_{ATP}$ channels [2, 6].

3.17 Antiplatelet activity

3.17.1 Fruit extracts

Proteases derived from fig aqueous-ethanolic extract were investigated on human blood coagulation using *ex vivo* model of human platelets from volunteers free of medications for 1 week. Extracts at 0.6 and 1.2 mg/mL repressed the human platelet aggregation with the agonists adrenaline and adenosine 5′-diphosphate (ADP). Ficin, a mixture of proteases derived from figs, seems to be responsible for the activation of blood coagulation factor X (vitamin K-dependent plasma glycoprotein with pivotal role in hemostasis) [2, 6, 38].

4. Conclusions

Since ancient times, the fruits and leaves of *F. carica* have been used as food and for their different therapeutic effects. In recent years several scientific works have analyzed the chemical composition of both parts of the plant to know more in depth the phytochemical compounds responsible for the biological properties demonstrated in several *in vitro* and *ex vivo* tests. In addition, the use of new environmentally friendly extraction processes, such as ionic liquids or deep eutectic solvents, and the use of fig phytochemicals as additives for new food applications (nutraceuticals and functional foods) are highly researched topics in recent times. However, research on different alcoholic beverages derived from both parts of the plant, such as wine, liqueur, and spirit, is still scarce. These beverages represent an important source of sustenance for the local economy of different countries from the Mediterranean basin, so that their study could provide an improvement in the quality of the products and publicize the chemical and biological properties derived from their consumption.

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Conflict of interest

The authors declare no conflicts of interest.

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Chapter 8

Phytochemical Composition, Antioxidant Potential, and Medicinal Significance of *Ficus*

Haq Nawaz, Rashem Waheed and Mubashir Nawaz

Abstract

Ficus, a genus of plant family *Moraceae*, includes about 850 species. Most of the species of *Ficus* are used as a source of nutrition for humans. The roots, aerial roots, stem, bark, leaves, latex, fruit, and pulp of the *Ficus* plants are medicinally important due to the presence of a variety of bioactive phytochemical compounds, such as polyphenols, phenolic acids, triterpenoids, flavonoids, flavonols, anthocyanins, carotenoids, glycosides, polysaccharides, reducing compounds, and vitamins K, E, and C. Most of these phytochemical compounds possess strong antioxidant potential in terms of metal chelating, metal reducing, lipid reducing, and free radical scavenging capacities, which may be helpful in reducing the oxidative stress in the biological systems. On account of their high phytochemical content and strong antioxidant potential, these plants show several biological activities including antimicrobial, antidiabetic, anti-obesity, hepatoprotective, cardioprotective, and renal-protective, and anticancer activities. These plants have been found to be effective in the treatment of diabetes, stomachache, piles, skin diseases, inflammation, and cancer.

Keywords: Moraceae, Ficus, Phytochemical composition, Antioxidant potential, Medicinal significance

1. Introduction

Ficus is a genus of family *Moraceae* and consists of about 850 species. About 200 different varieties of *Ficus* are present as woody trees, shrubs and vines in the forests of tropical and subtropical regions [1]. About 500 species of *Ficus* are found in the region of Asia and Australia [2]. Some species of *Ficus* are also grown as indoor as well as outdoor ornamental plants. *Ficus* species are rich in nutritional components and used as a source food in Egypt, India, south China, Turkey and Malaysia. The plants of *Ficus* species are well known in the field of traditional medicine. *Ficus* species have been found to be rich source of phenolic acid and flavonoids which make them able to protect against disorders of oxidative stress [3]. Extract of these plants have been reported to be effective in the treatment of diabetes, stomachache, piles, ulcer, dysentery, inflammation, oxidative stress and cancer [4]. Ethno-medicinal uses of *Ficus* plants have been also supported by their anti-cancer, anti-inflammatory and anti-diabetic activities [5].

Ficus plants are among the earliest cultivated fruit and ornamental tree which attract birds and mammals. *Ficus* species, such as, *Ficus carica*, *Ficus religiosa*, *Ficus benghalensis* and *Ficus racemosa* are the most important species of this genus as a spiritual, religious and historical plants to be used as folk medicine to treat various ailments, infectious diseases and cancer [6, 7]. Various parts of *Ficus religiosa*, have been reported to be used to treat high fever, chronic asthma and cancer and regulate menstrual cycle [8–11]. *Ficus carica* also known as edible fig, its fruit had been used from ancient times due to its activity against cancer, hepatomegaly, ulcer, platelets and inflammatory disorders. Leaves of *Ficus carica* used to treat dermatitis. It can activate potassium ATP channels and, hence, is used effectively in gut motility [12]. *Ficus racemosa* traditionally named as sacred fig is popular as its latex is used in treatment of ulcer, tumor, gout and aphrodisiac and fruits are used as laxative and digestive due to antitumor and antibacterial activity [13]. *Ficus benghalensis* commonly called Indian banyan has been reported to possess anti-insulinase, anthelmintic, and antitumor activity [14, 15]. Different species of *Ficus* shows different colors due to the presence of various pigments like polyphenols, flavonoids and anthocyanins. The skin of *Ficus* fruits contains comparatively higher content of phytochemicals and antioxidants than fruit pulp [16]. The wood of the *Ficus* plants contains latex like material within their vasculatures that provide protection and wound healing from physical assaults [17].

The genus *Ficus* is classified as:

Domain	Eukaryota
Kingdom	Plantae
Subkingdom	Viridaeplantae
Phylum	Tracheophyta
Subphylum	Euphyllopsidia
Infra phylum	Radiatopses
Division	Magnoliophyte
Class	Magnoliopsida
Subclass	Dilleniidae
Superorder	Urticaneae
Order	Urticales
Family	*Moraceae*
Genus	*Ficus*

2. Biochemical and nutritional composition

Since ancient times, *Ficus* species has been used as a source of food to improve the health of mankind [17]. Most of the species of *Ficus* are used in industrial products as nourishing foods. These are composed mainly of water, lipids, essential amino acids, minerals and vitamins [18]. *Ficus* genus worked as food additives that use frequently as health-promoting Mediterranean diet. It has great importance as nutraceutical and in biopharmaceutical industries [19]. They are known as rich sources of amino acids that are totally free from cholesterol and fat contents. *Ficus carica* is an excellent source of minerals containing copper, manganese, magnesium, potassium and calcium according to human needs [15, 20–22].

3. Phytochemicals of *Ficus* species

Phytochemicals are the bioactive components of plants having great importance in pharmaceutical and medicinal field. The genus *Ficus* consist of a variety

Ficus species	Plant parts	Extracting solvent	Class	Phytochemical components	References
Ficus religiosa	Barks	Water, methanol, organic solvents, helium	Polysterols	Bergapten, bergaptol, lanosterol, β-sitosterol, stigmasterol, β-sitosterol-D-glucoside (Phytosterolin)	[24, 25]
			Flavonoids	Leucocyanidin-3-O-β-glucopyranosid, leucopelargonidin-3-O-β-D-glucopyranoside, leucopelargonidin-3-O-α-L-rhamnopyranoside, lupeol, cetyl behenate, acetate and α-amyrin acetate	
			Polyphenols	Tannin, wax, saponin, leucoanthocyanidin, leucoanthocyanin	
	Fruit	Water	Flavonols	Kaempferol, quercetin, and myricetin	[26, 27]
			Miscellaneous compounds	Undecane, tridecane, tetradecane, (e)-β-ocimene β-bourbonene, β-caryophyllene, α-trans bergamotene, α-thujene, α-pinene, β-pinene, α-terpinene, limonene, dendrolasine, dendrolasine α-ylangene, α-copaene, aromadendrene, α-humulene, alloaromadendrene, germacrene, bicycle-germacrene, γ-cadinene and δ-cadinene	
	Leaves	Ethanol	Polyphenols	Eugenol, 2-phenylethyl alcohol, and benzyl alcohol, hexenol, n-hexanol, phytol, benzyl alcohol	[28]
			Miscellaneous compounds	Phenol, salicylaldehyde, phenylacetaldehyde, allyl caproate, linalool, n-nonanal, adipoin, methylcyclopentane, 2-dione, itaconic anhydride, 2-phenylethyl alcohol, benzeneacetonitrile, nonadienal, nonen-1-ol, nonadienol, linalool oxide, catechol, coumaran, cinnamyl alcohol, vinylguaiacol, hexenyl tiglate, eugenol, hexenyl hexenoate, β-ionone, dihydroactinidiolide, α-copaene, hexenyl benzoate, eudesmol, eudesmol, epi-α-cadinol, β-eudesmol, α-eudesmol, α-cadinol, pentadecanal, palmitic acid and itaconic anhydride, 3-methylcyclopentane-1, 2-dione	
Ficus auriculata	Leaves and fruits	Ether, chloroform and ethanol	Flavonols	Kaempferol, quercetin, myricetin	[29]
			Phenolic acids	Betulinic acid, lupeol	
			Sterols	Stigmasterol, bergapten, scopoletin, β-sitosterol-3-O-β-D-glucopyranoside	

Ficus species	Plant parts	Extracting solvent	Class	Phytochemical components	References
Ficus sycomorus	Whole plant	N-butanol, ethanol and methanol	Flavonoids	Quercetin, quercetin 3-O-L-rhamnopyranosyl (1-6)-β-D-glucopyranoside, quercetin 3-O-β-D-glucopyranoside (isoquercitrin), quercetin 3,7-O-α-L-dirhamnoside, quercetin, 3-O-β-D-galactopyranosyl(1-6)-glucopyranoside	[30]
			Sterol	β-Sitosterol-3-β-D-glucopyranoside	
			Phenolic acids	Gallic acid	
Ficus carica	Dried fruit	Water	Flavonoids	Alkaloids, flavonoids, coumarins, saponins, rennin, caoutchouc, resin, albumin, cerin, sugar and terpenes	[31]
	Latex	Water	Enzymes	Proteolytic enzymes, diastase, esterase, lipase, catalase, and peroxidase	[32]
			Phenolic acids	Malic acid	
	Leaves	Water	Coumarins	Psoralen and bergapten	[33]
			Flavonoids	Rutin, quercetin, and luteolin	
			Phenolic acids	Ferulic acid	
			Phytosterols	Taraxasterol, psoralen and bergapten (5-methoxypsoralen)	
	Pulp	Water	Phenolic acids	Chlorgenic acid	
	Peel	Water	Coumarins and sterol	Quercitin-3-O-rutinoside, psoralen	
Ficus benghalensis	Aerial roots	Water and methanol	Polyphenols	Saponins, tannins, glucoside and flavonoids	[14]
			Sterol	β-Sitosterol-α-D-glucose and meso-inositol	
Ficus capensis	Stem bark	Water	Polyphenols	Alkaloids, balsams, carbohydrates, flavonoids, free anthraquinones, tannins, glycosides, terpenes, resins, sterols and saponins, glycosides	[34]
	Leaves	Water	Volatile compounds	Carvacrol, α-caryophyllene, caryophyllene oxide, linalool, 3-tetradecanone, geranylacetone, 3,7,11-trimethyl-3-hydroxy-6;10-dodecadiene-1-yl acetate, hexahydrofarnesyl acetone, α-caryophyllene, 2-methyl-3-hexyne and scytalone	[35]
Ficus polita Vahl	Roots	Water	Phenolic acids	Betulinic acid and ursolic acid	[36]
			Anthocyanins	Trihydroxy-stilbene-3, 5-O-β-D-diglucopyranoside, euphol-3-ocinnamate, lupeol, taraxar-14-ene	
Ficus microcarpa	Aerial roots		Triterpenoids	Friedelin, lupeol, oleanolic acid, ursolic acids	[37]
	Leaves		Flavoinoids	Catechin, epicatechin and isovitexin	

Ficus species	Plant parts	Extracting solvent	Class	Phytochemical components	References
Ficus retusa	Leaves	Methanol	Polyphenols	1,2-Benzenedicarboxylic acid-dibutyl ester, phenol, 4-(2aminopropyl), butyrolactone	[38]
	Aerial parts	Ethanol	Flavonols	Luteolin, afzelechin, catechin, vitexin, β-sitosterol acetate, β-amyrin acetate, moretenone, β-amyrin	[39]
			Sterols	β-Sitosterol, friedelenol	
Ficus palmata	Stem bark	Water	Anthocyanins	Cetyl behenate, lupeol, α-amyrin acetate	[40]
	Leaves and bark	Water	Sterols	β-Sitosterol and a new tetracyclic tritepene-glaunol acetate	
Ficus thunbergii	Fresh leaves and stem	Methanol	Anthocyanins	Amyrin acetate, α-amyrin acetate, lupeol, β-amyrin, α-amyrin, rhoiptelenol, 3α-hydroxyisohop-22(29)-en-24-oic acid, lupenyl acetate	[41]
			Phenolic acids	Ursolic acid, betulinic acid	
Ficus cordata	Stem bark	Water	Terpenes	Pentacyclic triterpenes 8,26-cyclo-urs-21-en3β, 20β-diol and 3β-acetoxy-8, 26-cyclo-ursan-20β-ol and also 3-friedelanone	[42]
			Phenolic acids	Oleanolic acid, betulinic acid	
			Anthocyanins	Lupeol acetate, α and β amyrine, 3,5,7,4′-tetra hydroxyl flavones	
Ficus deltoidea	Leaves	Hot and cold water	Flavonols	Triterpene, conrauidienol, and dihydroflavonol, conrauiflavonol, 3,4',5-trihydroxy-6'',6''-dimethylpyrano[2,3-g]flavone	[43–45]
			Anthocyanin	β-amyrin acetate, 6β-hydroxystigmasta-4,22-dien-3-one, 8-prenylapigenin	
			Phenolic acid	Betulinic acid, ursolic acid	
			Flavonoids	Luteolin, catechin, epigallocatechin, orientin	
			Sterol	β-Sitosterol glucoside	
Ficus tsiela	Whole plant	Water	Phenolic acid	Gallic acid	[46]
			Anthocyanin	3, β-hydroksilup-20(29)-en, (lupeol)	
			Polyphenols	Carbohydrates, glycosides, saponins, resins, fat, flavonoids, tannins, and phenolic compounds. Alkaloids and steroid were absent	[47]

Table 1.
Phytochemical quality of various parts of commonly used species of Ficus.

Ficus species	Plant parts	ES	TPC	TFC	TF	AAC	TAC	TSC	TA	References
Ficus benghalensis	Roots	Ethanol	70 mg/g extract	5 mg QE/g extract	3 mg QE/g extract					[48]
Ficus deltoidea	Pulp	Water	0.49–0.88 mg GAE/g							[49]
Ficus microcarpa	Leaves	Hexane		6.6–9.5 M/TE						[50]
F virens	Dried leaves	Hexane	17.44 mg/g	3.87 mg/g						[51]
F racemosa	Dried leaves	Methanol	7.83 mg/g	1.05 mg/g						[51]
Ficus carica	Fruit	Ethanol	28.6–211.19 mg GAE/100 g FW, 11.9 mg/g of DM	2.75 μg CE/mg sample			9.6%	0.59%	0.0–298.6 μg cy-3-rutinoside/g FW	[52, 53]
Ficus deltoidea	Fruit	Hexane	259.2 mg GAE/g							[54]
		Methanol	245.2 mg GAE/g							
		Chloroform	159.2 mg GAE/g							
Ficus indica	Pulp	Methanol				28–30 mg/100 g extract				[55]

ES: extracting solvents, TPC: total phenolic content, TFC: total flavonoid content, TF: total flavonols, AAC: ascorbic acid content, TAC: total alkaloid content, TSC: total saponin content, TA: total anthocyanins, DM: dried material, QE: quercetin equivalent, TE: trolox equivalent, ep: edible pulp, GAE: gallic acid equivalent, FW: fresh weight.

Table 2.
Phytochemical content of various parts of commonly used species of Ficus.

of phytochemicals including phenolics, polyphenols, flavonoids, tannins, anthocyanins, coumarins, volatile components, glycosides, saponins, carotenoids, alkaloids, triterpenoids and vitamins. Most of these phytochemical compounds show health promoting effects in human due to their strong antioxidant potential. Higher concentrations of phytochemicals are responsible for the strong antioxidant potential of plants of genus *Ficus* and are helpful in the prevention of certain cardiovascular, neurodegenerative, and hepatic diseases caused by oxidative stress [23]. The phytochemical quality of various parts of some of the species of *Ficus* is presented in **Table 1**. It is reported that the roots, stem bark or wood, branches, fruit pulp, peel, leaves, and seeds of different species of *Ficus* plant contain the flavonoids and phenolic compounds as major phytochemical components along with polyphenol, polysterols and triterpenoids. The phytochemical content of various parts of some of the species of *Ficus* in terms of total phenolic, flavonoids, flavonols, ascorbic acid, alkaloids, saponins and anthocyanins contents in different solvents is presented in **Table 2**. The leaves and fruit pulp of various species of *Ficus* have been found to show relatively higher concentration of phenolic components due to which these parts comparatively have greater pharmacological as well as medicinal usage.

4. Antioxidant composition

Antioxidants are the substances which can scavenge free radicals and reduce the oxidative stress in the living and nonliving systems. The antioxidants possess electron donating ability and inhibit the free radical-mediated oxidative reactions by various mechanisms, such as, hydrogen donation, metal chelation, metal and lipid reduction, inhibition of lipid peroxidation and free radical inhibition [56–60]. Free radicals are the reactive oxygen and nitrogen species which are produced during various biochemical reactions particularly redox reactions. If not controlled properly, these free radicals may initiate the chain reactions in the biomolecules particularly the lipids and protein, cause the oxidative stress, and finally lead to the oxidative damage to the cell organelles, cells and tissues [24]. The oxidative damage to the cells and tissues may further lead to various health problems including cardiovascular, neurological, hepatic, and musculoskeletal abnormalities and aging. In nonliving system, the free radicals cause oxidative stress and rancidity in the food stuff for human [25]. The naturally occurring antioxidant compounds have been proved to be effective in preventing the oxidative damage to the living and nonliving systems [26]. These substances are either synthesized endogenously or taken from exogenous natural sources such as plants. The naturally occurring antioxidants include some enzymes such as glutathione peroxidase, catalase, superoxide dismutase and some non-enzymatic phytochemicals compounds including phenolic acids, polyphenols, flavonoids, anthocyanins, ascorbic acid, tocopherols, and β-carotenes [27, 28]. Some synthetic antioxidant compounds have been also reported to be effective against free radical-induced oxidative damage [29].

The antioxidant profile of various parts of *Ficus* species is presented in **Table 3**. Different parts of *Ficus* plants have been reported to showed antioxidant activity in terms of Trolox equivalent antioxidant capacity, ferric reducing antioxidant power, lipid reducing activity, inhibition of lipid peroxidation, and free radical scavenging capacity against 2,2-diphenyl picryl hydrazyl (DPPH) and 2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radicals in a dose dependent vstronger antioxidant activity due to relatively higher concentration of phenolic components [30].

Ficus species	Part	ES	TEAC*	FRAC	DPPH-RSC	ABT-RSC	ILP	LRA	References
Ficus racemosa	Stem	Methanol			16.2%	8615.3 mmol/g DM			[61]
	Bark	Ethanol			79%	10884.6 μmol/g DM			
	Roots	Water		0.5–0.26 mg/ml					
Ficus virens sublanceolata	Leaves	Water		0.13–0.66 mg/ml	IC_{50}: 0.34 mg/ml	IC_{50}: 0.23 mg/ml	83.30%		[51]
Ficus vasculosa	Leaves	Methanol		0.07–0.26 mg/ml	IC_{50}: 0.69 mg/ml	IC_{50}: 0.97 mg/ml			[51]
Ficus indica	Mouse liver	Normal saline				4.20–5.31 μmol TE/g ep	EC_{50}: 313.3 μg/ml		[55, 62]
	Chicken liver	Normal saline					EC_{50}: 333.8 μg/ml		
Ficus callosa	Fruit	Methanol		0.08–0.33 mg/ml	IC_{50}: 0.95 mg/ml	IC_{50}: 0.35 mg/ml	41–83%		[51]
Ficus palmate	Fruit	Methanol		77.6 mg AC/100 g FW	104.9 mg CE/100 g FW	577.09 mg BH/100 g FW			[63, 64]
		Ethanol		146.67 mg AC/100 g FW	146.9 mg CE/100 g FW	729.45 mg BH/100 g F W			
Ficus auriculata	Roots	Acetone		0.1–0.45 mg/ml	IC_{50}: 0.29 mg/ml	IC_{50}: 0.25 mg/ml	41–83%		[51]
Ficus virens	Bark	Water		0.06–0.32 mg/ml	IC_{50}: 1.03 mg/ml	IC_{50}: 0.48 mg/ml			[51]
	Leaves	Methanol			SC_{50} (74.00 μg/ml)				[65]
Ficus oligodon	Leaves	Acetone		0.04–0.22 mg/ml	IC_{50}: 2.54 mg/ml	IC_{50}: 0.86 mg/ml	41.40%		[51]
Ficus benghalensis	Aerial roots	Methanol			71%	6096.1 μmol/g DM			[61, 66]
		Acetone, Water		0.1–1.0 mg/ml	96.07%	6182.7 μmol/g DM			
Ficus auriculata	Stem bark	Methanol			84.088%				[67]
	Stem bark	Chloroform			83.864%				
	Stem bark	Hexane			42%				
Ficus caprefolia	Leaves	Acetone	2.32%, 4.73 mg GAE/g DW						[68]

***Ficus* species**	**Part**	**ES**	**TEAC***	**FRAC**	**DPPH-RSC**	**ABT-RSC**	**ILP**	**LRA**	**References**
Ficus carica	Leaves	Hexane, water	14.04%, 23.50 acetate/g DW	7.9–16.1 mmol/kg FW	11.42 mmol/100 g DW	6.48 mmol/100 g DW			[52, 69, 70]
Ficus carica	Fruit	Dichloromethane					IC_{50}: 0.02 mg/ml		[71]
		N hexane					IC_{50}: 1.64 mg/ml		
Ficus glomerata	Root, Bark	Water			IC_{50}: 1.62–47.50 μg/ml	IC_{50}: 0.91–6.48 μg/ml		86.13%	[72]
Ficus cordata	Leaves	Acetone	2.65%, 8.23 mg GAE/g DW						[68]
Ficus pumila L	Leaves	Ethanol			SC_{50} > 0.4 mmol/100 g DW				[73]
Ficus sur	Bark	Water	489.4 mg GAE/g DW	104.57 μmol FSE/mg DE	56.50 QE/mg DE				[74]
	Unripe fruit		62.34 GAE/g DW	19.61 μmol FSE/mg DW	7.3 QE/mg DE				
Ficus craterostoma	Leaves	Acetone	2.60%, 9.80 mg GAE/g DW						[68]
Ficus religiosa	Fruit	Methanol			55.9%			93.91%	[75]
Ficus deltoidea	Fruit	Water	5.89 mg GAE/g DW	1.82 mmol FSE/g DE	IC_{50} = 111.20 μg/ml	1.01–1.04 mmol TE/g DE			[76]
Ficus glumosa	Leaves	Acetone	2.60%, 19.24 mg GAE/g DW						[68]
Ficus microcarpa	Bark	Ethyl acetate	436 mg GAE/g DW	63.2 μg/ml	1.2 μg/ml	4.83 μg/ml			[71]
	Leaves	Ethanol						86.13%	
		Hexane						86.76%	

Ficus species	Part	ES	TEAC*	FRAC	DPPH-RSC	ABT-RSC	ILP	LRA	References
Ficus cunninghamii	Leaves	Ethanol						90.70%	[71]
		Hexane						88.97%	
Ficus mysorensis	Leaves	Ethanol						90.13%	[71]
		Hexane						94.38%	
Ficus microcarpa	Fruit	Water organic solvents	17.9 g GAE/g DW						[22]
Ficus lyrata Warb	Leaves	Ethanol			SC_{50} (8.27, 12.14 µg/ml)			80.41%	[65]
		Methanol			SC_{50} (38.37 mg/ml)				[65]
Ficus nitida L.	Dried leaves	Methanol			SC_{50} (61.67 µg/ml)				[65]
Ficus afzelii G.	Pulp	Methanol			SC_{50} (60.22 µg/ml)				[65]
Ficus decora Hort	Leaves	Methanol			SC_{50} (81.62 µg/ml)				[65]
Ficus lutea	Leaves	Acetone	3.70%, 56.85 mg GAE/g DW						[68]
Ficus natalensis	Leaves	Acetone	2.35%, 4.75 mg GAE/g DW						[68]
Ficus polita	Leaves	Acetone	3.15%, 8.04 mg GAE/g DW						[68]
Ficus religiosa	Leaves	Acetone	2.45%,5.40 mg GAE/g DW						[68]

***Ficus* species**	**Part**	**ES**	**TEAC***	**FRAC**	**DPPH-RSC**	**ABT-RSC**	**ILP**	**LRA**	**References**
Ficus sycomorus	Leaves	Acetone, hexane and methanol	2.60%, 12.33 mg GAE/g DW		SC_{50} (79.50 μg/ml)			82.35%	[65, 68]
Ficus thonningii	Leaves	Acetone	2.40%, 4.64 mg GAE/g DW						[68]
Ficus macrophylla	Leaves	Ethanol						86.40%	[71]

*****ES: extracting solvent, ABTS-RSC: azino-bis-tetrazolium sulfate radical scavenging capacity, DE: dry extract, DM: dry matter, DPPH-RSC: 2,2-diphenyl-1-picrylhydrazyl radical scavenging capacity, DW: dry weight, FRAC: ferric-reducing antioxidant capacity, FSE: ferrous sulfate equivalent, FW: fresh weight, GAE: gallic acid equivalent, IC_{50}: inhibitory concentration required for 50% inhibition, QE: quercetin equivalent, SC_{50}: scavenging concentration for required for 50% scavenging, TEAC: trolox equivalent antioxidant capacity, TE: trolox equivalent, BH: butylated hydroxyanisole, FW: fruit weight, CE: catechin equivalents, ILP: inhibition of lipid peroxidation, LRA: lipid reducing ability.*

Table 3.
Antioxidant potential of extracts from various parts of Ficus species.

5. Biological activities

On the basis of their phytochemical composition and antioxidant profile, *Ficus* species have been found to show several biological activities (**Table 4**). The studied species of *Ficus* plants were found to possess anticancer, hepatoprotective, hypoglycemic, antitumor, antioxidant, anthelmintic, analgesic, antimicrobial activity, anti-parasitic, hypolipidemic, anti-inflammatory, antibacterial, anti-ulcerogenic, mucoprotective, gastroprotective, antifungal, antiviral, antimalarial, and antiparasitic activities [43, 69]. However, the antibacterial activity has been found to be more common in different species of *Ficus*.

***Ficus* species**	**Plant part**	**Extracting solvent**	**Activity**	**References**
Ficus racemosa	Whole	Ethanol	Anticancer activity by reduction of lipid peroxidation, γ-glutamyl transpeptidase and xanthine oxidase and by generation of hydrogen peroxide	[77]
	Bark	Methanol	Hepatoprotective activity by reducing the activities of ALT, AST and ALP	[4]
	Whole	Ethanol	Hypoglycemic activity by decreasing blood glucose level	[4]
Ficus religiosa	Fruit	Water	Antitumor activity due to blockage of calcium uptake in pituitary cells	[13]
	Whole	Water	Antioxidant and antidiabetic activity with lowering the superoxide dismutase exaggerated activity	[78]
	Whole	Methanol	Anthelmintic activity with 100% effectiveness	[79]
	Whole	Water	Antimicrobial activity with inhibition zone against *B. subtilis*	[4]
	Bark	Methanol	Anti-parasitic effect with 100% lethality for *Haemonchus contortus* worms	[79]
Ficus benghalensis	Bark	Water	Antioxidant and hypolipidemic activity by reduction in lipid peroxidation, cholesterol level and triacylglycerol	[80]
	Fruit	Water	Anticancer and antibacterial activity but no antifungal activity	[81]
	Roots	Various polarity solvents	Anti-inflammatory and analgesic activity	[82]
	Whole	Methanol	Anti-inflammatory and analgesic activity due to inhibition of malanodialdehyde formation	[82]
Ficus hispida	Roots	Methanol	Antiulcerogenic activity with cytoprotective nature of constituents	[83]
Ficus arnottiana	Leafs	Methanol	Mucoprotective activity and gastric antisecretory	[23]
Ficus carica	Leaves	Methanol	Hepatoprotective activity with decrease in lipid peroxides with cytochrome p450 complex inhibition	
Ficus glomerata	Fruit	Ethanol	Gastroprotective effect	[84]
	Fruit	Phenol	Anti ulcerogenic, antimutagenic and anti cancerogenic compounds	[84]

Ficus species	Plant part	Extracting solvent	Activity	References
Ficus polita Vahl	Whole	Water	Antiviral activity due to inhibition of reverse transcriptase activity of HIV-1	[85]
	Leaves	Water	Antimalarial action against *Plasmodium falciparum*.	[86]
Ficus lyrata	Leaves	Water, ethanol	Significant antibacterial activity	[35]
	Leaves	Water	Activate against standard human pathogenic yeasts strains	[87]
Ficus Tsiela	Leaves	Diethyl ether	Anti-pneumonia activity	[88]
Ficus sycomorus L	Leaves	Water	Significant antibacterial activity but no antifungal activity	[35]
Ficus deltoidea	Leaves and fruits	Alcohol	Antifungal and antibacterial activities	[89]
Ficus platyphylla	Stem bark	Water	Antimicrobial activities against *S. aureus*	[65]
Ficus thonningii	Leaf	Water	Significant antimicrobial effect	[90]
Ficus lutea	Leaves	Acetone	Act as potent inhibitor of α-amylase	[68]

Table 4.
Biological activities of extracts from various parts of Ficus species.

6. Medicinal importance

Ficus species have been used as traditional medicines to cure diseases, such as, astringents carminatives, stomachic, vermicides, hypotensive, anthelmintic and anti-dysentery drugs [18]. *Ficus* species, such as, *Ficus racemosa*, *F. glomerata*, *F. glumosa*, *F. carica*, *F. religiosa* and *F. benghalensis* are known from ancient times as herbal medicines to treat diabetic disorders as regulating enzymatic activities, carbohydrates absorption rate, increasing insulin sensitivity, insulin secretion, hepatic glycogen synthesis, peripheral glucose uptake and antioxidant status of body [19]. The extracts of these species also reduce oxidative stress by improving weight gain in diabetic male rats [20]. Aqueous bark extract of *F. benghalensis* have been found to be active in lowering the cholesterol level in hypercholesterolemic rats [14, 15]. Methanolic extract of *F. carica* leaves prevent elevation of lipid peroxide in rats by acting as hepatoprotective agent [21]. Methanolic extracts of *F. hispida* roots exhibit anti ulcerogenic activity due to higher concentration of flavonoids in roots. Methanolic leaf extract of *F. arnottiana* exhibits both mucoprotective as well as gastric antisecretory activities due to antioxidant constituents [22, 23].

Almost all of the *Ficus* species belonging to family *Moraceae* haven traditionally used as folk medicine to cure respiratory disorders and skin diseases. The roots of *Ficus* species are important to treat gout and gums diseases that have anthelmintic activity. Fruit of *Ficus* species, such as, *F. carica*, *F. hispida*, *F. microcarpa* and *F. sycomorus* has been found to be helpful improving digestion or treating vomiting. Dried powder of bark has importance to treat burns or Asthma [4]. *F. benjamina* exhibits antitumor activity or antibacterial activity but is unable to work on fungal disorders [13, 14]. Leaves of *F. religiosa* exhibit hypotensive activity and help in treating the gastrointestinal problems [9, 56, 57]. Bark of *F. religiosa* shows hypoglycemic activity and is used against gonorrhea, bleeding, paralysis, diarrhea, bone fracture, antiseptic, astringent and antidote [58, 59]. It has been also used against liver disorders, hemorrhoid, urinary tract infections and inflammatory conditions by different mechanisms [60].

7. Conclusion

All species of *Ficus* plant possess antioxidant potential due to higher concentration of phytochemical compounds. They have a valuable role in human nutrition or have a great medicinal importance due the presence of a variety of bioactive phytochemical compounds. The principal phytochemicals present in *Ficus* species are polyphenols, phenolic acids, flavonoids, anthocyanins, glycosides, carotenoids, and some water-soluble vitamins. The presence of these phytochemicals makes *Ficus* a medicinal plant which shows various biological activities particularly the antioxidant activity. On the account of its high antioxidant potential, all parts of *Ficus* plant can be used for the management of oxidative stress and the treatment of various diseases.

Conflict of interest

The authors have no conflict of interest regarding this chapter.

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Chapter 9

A Threatened Introduced Species (*Ficus benghalensis* L.) in Ismailia, Egypt

Abdelraouf A. Moustafa

Abstract

The genus *Ficus* belongs to *Moraceae* (Mulberry family). It is comprised of around 1000 species from pantropica land subtropical origins; plants of the family are on the whole woody, ranging from trees and shrubs to climbers. *F. benghalensis* has many common names as Indian banyan tree, East Indian fig tree, and vada tree, banyan tree is considered sacred by Hindus and is largely grown near temples. It is a large and extensive growing tree of the Indian subcontinent. *Ficus* compound showed significant antioxidant effect, which might be ascribed to their polyphenolic nature. The stem bark of *Ficus benghalensis* L. and *Ficus racemosa* L. are utilized in India for the treatment of diabetes and various different illnesses. The bark of *Ficus benghalensis* decreased fasting blood sugar and glycosylated hemoglobin. The fruit of *Ficus benghalensis* is used in folk medicine for respiratory disorders and certain skin disease.

Keywords: *F. benghalensis*, banyan tree, antioxidant effect, folk medicine, age dating

1. Introduction

The genus *Ficus* belongs to *Moraceae* (Mulberry family). It is comprised of around 1000 species from pantropica land subtropical origins [1]. Plants of the family are on the whole woody, ranging from trees and shrubs to climbers [2]. *Ficus benghalensis* is an evergreen tree with a wide, spreading crown; it can grow 20–30 m or more tall. The plant usually begins life as an epiphyte, growing in the branch of another tree; as it grows older it sends down aerial roots which, when root reach the ground become much thicker and more vigorous. They supply nutrients to the fig, allowing it to grow faster than the host tree. Finally, the host dies, leaving the fig to carry on growing without competition. It can become a huge, spreading tree in time, with some specimens several hundred meters across and producing aerial roots from the spreading branches that in the long run become new trunks and enable the crown to spread considerably further.

F. benghalensis has many common names as Indian banyan tree, East Indian fig tree, and vada tree [2, 3]. Its name has been derived from the Hindu traders, called banyans, who favored the tree [2]. Banyan tree is considered sacred by Hindus and is largely grown near temples. It is a large and extensive growing tree of the Indian subcontinent. The wood is grey, moderately hard and durable under water. The wood of the "props" is stronger than that of the main trunk. The timber is used for making well-curbs, furniture, crates, door panels and cart-shafts. The props are used for tent poles and umbrella handles. The leaves are used as fodder for cattle and elephants.

The tree is harvested from the wild for its edible fruit and medicinal uses. The tree is considered sacred by Hindus and is commonly planted for religious purposes; it is also grown as an ornamental and to provide shade along roads as well as in parks and large gardens.

Ficus compound showed significant antioxidant effect, which might be ascribed to their polyphenolic nature. The stem bark of *Ficus benghalensis* L. and *Ficus racemosa* L. are utilized in India for the treatment of diabetes and various different illnesses. The bark of *Ficus benghalensis* decreased fasting blood sugar and glycosylated hemoglobin. The fruit of *Ficus benghalensis* is used in folk medicine for respiratory disorders and certain skin disease.

According to Edlin and Nimmo [4] proved that the latex (source of rubber) has been found in large quantity in the wood of *Ficus* genus, which is representing one of the largest economical uses of *Ficus* in Egypt. *Ficus benghalensis* is thought to be cultivated 150 years ago in Ismailia during the process of digging the Suez Canal back in 1859–1869.

These days, Ficus trees are subjected to many threats affecting the presence of main populations. These threats comprised by the following points: (a) human cutting for general purposes, (b) cutting by council city for shifting landscape of the city, and (c) absence of a specific pollinating wasp in order to reproduce and spread [5]. For these reasons and subsequently results, it is very hard to find a new tree individuals coming up to these areas of *Ficus*. These threats will change the landscape of the whole area in the next few years.

The present study is aimed to anticipate a strategic conservation plan for landscape construction primed by *Ficus* trees in Ismailia, Egypt and discuss the age structure of this species.

2. Distribution

Ficus benghalensis is native to South Asia particularly India, Sri Lanka and Pakistan (**Figure 1**). It is often planted around temples and a place of religious interest. It is considered as sacred tree by both Hindus and Buddhists.

Banyan tree is widely cultivated in city parks and botanical gardens throughout the New World and Old World tropics. It grows well in tropical, semi-tropical areas, monsoon and rain forests with moderate to ample rainfall. Humid air and moist soil and is hardy, drought resistance and withstands mild frost is well suited for its growth [6].

Ismailia governorate is located in the eastern part of Egypt at the middle part of Suez Canal. It is bounded from the East by Suez Canal (that penetrates Temsah Lake and Bitter Lakes), from the West by the eastern borders of Delta along Damietta Nile branch, from the South by Suez-Cairo high way, Port Said and Manzala Lake from the North (**Figure 2**). It was established as a separate governorate by the declaration law number 24 in 1960. Its area is 5067 km^2 and has seven main cities, Ismailia (the capital), Fayed, El-Tal El-Kber, El-Kantara east, El-Kantara west, El-Ksasen and Abo Souer. Human population of Ismailia governorate reaches 1.4 million individuals.

3. Description

F. benghalensis, frequently very large, up to 30 m length, have numerous aerial roots which can extend into new trunks so that the tree goes on thinning out laterally

Figure 1.
Distribution of Ficus benghalensis in South Asia according to https://media4.picsearch.com/is?JAgdqscN1wEM9xqDeneyt8fKMoNfgxAyKwy5aPsy1qo&height=301.

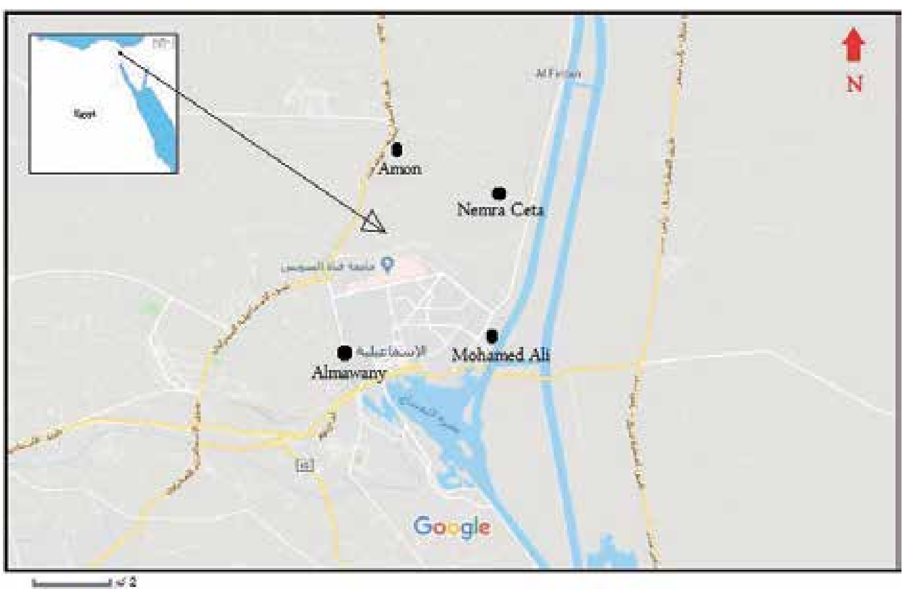

Figure 2.
Map showing the four studied localities of F. benghalensis chosen for study in Ismailia.

indefinitely; a single tree can thus cover a very wide area. The leaves are leathery, entire, ovate or elliptic, 20–40 cm long with prominent lateral veins. The figs are 1–2 cm in diameter, without stalks, in pairs in leaf axils, and when ripe are bright red.

Leaves are glossy, leathery and glabrous when mature, approximate hear the end of branches, ovate, mostly obtuse, base cordate or rounded, thickly coriaceous, basal nerves 3–7, the midrib with 4–6 pairs of secondary nerves, blade 10–20 cm, petiole 2–5 cm long, shoot pubescent, 1.5–2 cm in diameter, sessile, scarlet, red once ripe [7].

A flower has very small, separate, male and female flowers. The male flowers crowded near the mouth of the receptacle, whereas female flowers with shorter perianth, style long, male and female in the same receptacle [8].

Fruits are globular, sessile in axillary pairs, fleshy pericarp and with achenesen trenched in them, they are dark red in color, 1.5–2.0 cm diameter, red to dark purple when ripe; seeds are tiny. Fruit is not edible for humans but is eaten by birds and monkeys [8].

4. Ecology

In Egypt, various *Ficus* species are establish in streets, gardens, parks and outside the canal banks. Two of the most favorable fruits are eaten by Egyptian people (*F. carica* and *F. sycomorus*) and also they use *Ficus* in their traditional uses in folk medicine respiratory disorders and certain skin diseases [9, 10].

Mousa et al. [9, 10] detailed that there are about 20 species of *Ficus* native to Egypt; most of them are cultivated as street trees for providing shade (e.g., *F. retusa*) as in Alexandria city, while other species used as ornamental plants (e.g., *F. religiosa*). Edlin and Nimmo [4] proved that the latex (source of rubber) has been found in large quantity in the wood of *Ficus* genus, which is representing one of the largest economical uses of *Ficus* in Egypt.

5. Field visits and sampling

Four main localities characterized by large number of *F. benghalensis*, trees were selected in Ismailia city for the present study. These localities are: El-Mawany area comprises of 55 trees, Mohamed Ali area comprises of 5 trees, Amon area comprises of 16 trees, and Nemra Ceta comprise of 38 trees (**Figures 3** and **4**). In each locality, number of vegetation parameters was measured to describe the *Ficus* trees including; height, cover, circumference at base (CAB), circumference at breast height (CBH), number of aerial root and vitality. Tree vitality has also been measured using the visual assessment of crown conditions. Tree condition is often used in conjunction with other vitality assessments for verification purposes [11, 12]. However, vitality was measured according to the following scale: high, excellent healthy plant (vigor); medium, normal or some yellow leaflets and low, not healthy with yellow leaves [13].

Figure 3.
F. benghalensis trees in Ismailia roads.

Figure 4.
F. benghalensis aerial roots in one of the four studied areas in Ismailia.

6. Soil analysis

In each site, three soil samples (0–20 cm depth) were taken for soil chemical and physical analyses. Particle size analysis was done by dry sieving for the coarse sand and by pipette for fine sand, slit, and clay [14]. Soil aggregation was treated by 5% of sodium hexametaphosphate as a dispersing agent. Soil was classified based on the percentage of clay and sand using USDA limits of the basic soil textural classes [15]. Soil pH was measured electrometrically using pH meter model 1671 in soil suspension of ratio 1:2.5 soil to water. The soil-water mixture was first shacked for 2 hours, and then pH was measured [16].

The EC was measured in the soil water extract 1:1 using electrical conductivity meter model 4310 ENAWY [17]. Reported the degree of salinity for the course to loamy sand (1:1 soil water extract) as follows: 0.1–1 ds\m for non-saline, and 1.2–2.4 ds\m for slightly saline, 2.5–4.4 ds\m for modestly saline and 4.5–8.9 ds/m for strongly saline. Soil organic matter influences many soil properties including (i) the capacity of soil to supply, N, P and S and trace metals to plant's, (ii) infiltration and retention of water, (iii) degree of aggregation and overall structure that affect air and water relationships, (iv) cation exchange capacity, (v) soil color, which in turn affects temperature relationships [18]. Soil organic matter was measured using loss-on ignition (LOI) method carried out at a high temperature. This method gives quantitative oxidation of organic matter [18].

7. Age dating

Age dating of *Ficus* trees was assessed by counting the annual rings and measuring the circumference of the branches. To avoid destruction of *Ficus* trees, the counting procedure was applied to the already cut branches in each site. Annual

		Age dating scale (year)					
	Site no.	**<150**	**151–200**	**201–250**	**251–300**	**301–350**	**>350**
1	(El Mawany)	1	7	12	14	4	1
2	(Mohamed Ali)	0	9	4	3	0	0
3	(Amon)	0	0	1	1	3	0
4	(Nemra Ceta)	3	10	7	6	6	1

Table 1.
Age dating scale of F. benghalensis for the four studied localities.

rings and circumference of the available 21 cross sections of *Ficus* trees were measured. Simple regression between annual rings as dependent variable and circumference as independent variable was carried out to have the regression equation that used to figure out the age dating of the main trunk of *F. benghalensis* trees.

To describe the age dating structure of *Ficus* trees at different sites, scale as follows: <150, 151–200, 201–250, 251–300, 301–350, and >350 years (**Table 1**).

8. Data treatment

Data were statistically analyzed [19] using SPSS software (statistical package for social sciences, version 8). One-way ANOVA was carried out to test the variation of different variables between different four sites. Linear correlation coefficient, r, was estimated to find out the relationships between age dating, height and tree cover.

9. Results

9.1 Vegetation parameters

The maximum tree height was 12.5 m recorded at site four (Nemra Ceta) while the minimum tree height was 1.70 m recorded at site two (Mohamed Ali). The circumference at base (CAB) ranged between 0.56 and 1.9 m found at Nemra Ceta, while the highest circumference at breast height (CBH) was 1.8 m and lowest value of 0.65 m, both found at site one (El Mawany). The crown cover of trees ranged between 3.70 and 268.67 m^2, whereas the highest value was found at site three (Amon area) and the lowest value at site four (Nemra Ceta) (**Table 2**). The number

Site no. one (El Mawany)						
	N	**Minimum**	**Maximum**	**Mean**		**±SD**
	Statistic	**Statistic**	**Statistic**	**Mean**	**±SE**	**Statistic**
Height	55	3.52	11.52	7.0907	0.2485	1.8428
CAB	33	70	170	121.1818	4.6857	26.9171
CBH	40	65	180.03	110.8703	3.966	25.0832
Cover	55	18.31	223.14	70.1522	5.6871	42.1767
Age	40	142	394	242.425	8.6843	54.9241
Valid N (listwise)	33					
Site no. two (Mohamed Ali)						
	N	**Minimum**	**Maximum**	**Mean**		**± SD**
	Statistic	**Statistic**	**Statistic**	**Mean**	**±SE**	**Statistic**
Height	16	1.7	12	6.6875	0.5972	2.3888
CAB	7	90	125	121.1818	5.101	13.496
CBH	16	80	135.02	110.8703	4.6881	18.7523
Cover	16	13.68	104.72	70.1522	8.2362	32.9447
Age	16	175	295	242.425	10.2881	41.1525
Valid N (listwise)	7					

Site no. three (Amon area)						
	N	Minimum	Maximum	Mean		± SD
	Statistic	Statistic	Statistic	Mean	±SE	
Height	5	8.5	11	10.1	0.4301	0.9618
CAB	0					
CBH	5	109.9	157	138.998	8.2311	18.4053
Cover	5	186.17	268.67	229.786	13.2333	29.5907
Age	5	240	344	304	18.1466	40.5771
Valid N (listwise)	0					
Site no. four (Nemra Ceta)						
	N	**Minimum**	**Maximum**	**Mean**		**± SD**
	Statistic	**Statistic**	**Statistic**	**Mean**	**±SE**	
Height	38	3	12.5	6.7629	0.3142	1.9368
CAB	28	56	190	111.8929	7.3868	39.0871
CBH	33	44	163.28	105.2173	5.6693	32.5675
Cover	38	3.7	180.89	84.3755	7.9385	48.9363
Age	33	96	357	229.9697	12.4407	71.4666
Valid N (listwise)	28					

Table 2.
Vegetation parameters for the studied four sites.

	Site one	Site two	Site three	Site four
	(El Mawany)	(Mohamed Ali)	(Amon)	(Nemra Ceta)
No. of trees	55	16	5	38
Mean height	7	60.68	10.1	6.76
Mean CAB	121.8	98.82	N. R	105.21
Mean age	242.42	216.06	138.99	229.96
Mean cover	70.15	51.65	229.786	84.37

Table 3.
Mean values of height, CAB, age and cover for the four studied localities.

of aerial roots that were recognized in the studied sites fluctuated from 1 to 183 roots. Mean values of four localities are shown in (**Table 3**).

9.2 Soil properties

Soils of the examination region have two diverse surface classes, sand and loamy sand. Soil of site one (El Mawany) is loamy sand, whereas soils of the other three sites are sandy soils. Soil pH ranged from 7.43 to 9.14. Soil of site one (El Mawany) has the highest value (pH = 9.14) whereas site three (Amon) shows the lowest value (pH = 7.43) (**Table 4**).

Site no.		pH	EC (ds\m)	OM (%)	Sand (%)	Silt (%)	Clay (%)	Soil type
1	(El Mawany)	9.14	4.52	4.18	89.33	5.67	5.00	Loamy sand
2	(Mohamed Ali)	7.88	7.84	6.6	91.33	4.00	4.67	Sandy
3	(Amon)	7.43	4.56	7.3	92.67	3.67	3.67	Sandy
4	(Nemra Ceta)	7.93	5.47	7.8	91.00	3.67	5.33	Sandy

Table 4.
Soil physical and chemical characteristics for the studied localities.

As appeared (**Table 4**) summarizing the soil properties of the four sites studied, site one (El Mawany) has the lowest EC (4.52 ds/m) whereas site two (Mohamed Ali) shows the highest EC (7.84 ds\m) and both EC values of sites three (Amon) and four (Nemra Ceta) were (4.56 ds\m) and (5.47 ds\m) respectively. Site four (Nemra Ceta) shows the highest content of organic matter (7.80%) whereas site one (El Mawany) show the lowest content (4.18%), while results for sites three (Amon) and two (Mohamed Ali) were 7.30 and 6.60%, respectively. Site one (El Mawany) shows the lowest content of sand was 89.33% whereas moderate content of clay 5% and highest content of silt 5.67%. Site three (Amon) shows highest content of sand 92.67% whereas the lowest silt content and clay 3.67%. Site four (Nemra Ceta) shows the highest content of clay 5.33% whereas moderate content of fine sand fraction 91% and lowest content of silt 3.67%.

9.3 Age dating

Diameter and annual rings of each branch were measured and regression equation was calculated using the data of diameter and number of growth rings in order to figure out the age dating of the main trunk of different *Ficus* trees. The regression equation is:

No. of rings = (−0.933 + 2.195 circumference) (cm) (**Figure 5**).
$r = 0.95$ $r^2 = 0.90$ $P \leq 0.0001$

Age dating at El Mawany, ranged from 142 to 394 years. The oldest tree is 394 years, 10.51 m height and 106.73 m^2 cover while the youngest tress is 142 years, 3.52 m height and 20.58 m^2 cover. At site two (Mohamad Ali) which has 16 trees age dating of these trees ranges from 175 to 295 years, the oldest tree is 295 years, 7.50 m height and 59.42 m^2 cover, and whereas the youngest tree is 1.70 m height and 80.32 m^2 cover. Whereas site three (Amon) includes five trees, age of these trees range from 240 to 344 years, the oldest tree is 344 years, 10.50 m height and 268.67 m^2 cover; whereas the youngest tree which is 240 years old, 10 m height and 224.20 m^2 cover. Site four (Nemra Ceta) which has 38 trees, age of these trees ranges from 96 to 357 years, the oldest tree which is 357 years, 8 m height and 124.82 m^2 cover, whereas the youngest tree is 126 years, 5 m height and 13.72 m^2 cover.

Based on the regression results, the oldest *Ficus* tree is 394 years found at site one (El Mawany), followed by the second oldest tree about 357 years found at site four (Nemra Ceta).

9.4 Relationships between age and vegetation parameters

One way ANOVA (analysis of variance) of height, CAB, CBH, cover, and age have significant variation between the different sites. Multiple comparison of the

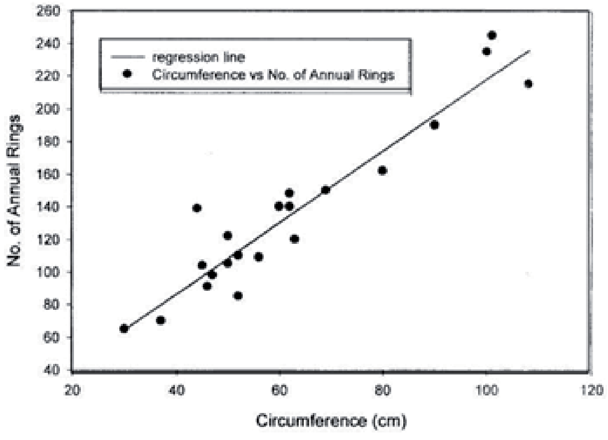

Figure 5.
Relation between number of annual rings and circumference (cm).

		Sum of squares	Df	Mean square	F	Sig.
	Between groups	51.833	3	17.278	4.619	0.004
Height	Within groups	411.47	110	3.741		
	Total	463.304	113			
	Between groups	2677.835	3	892.612	0.872	0.46
CAB	Within groups	65528.45	64	1023.882		
	Total	68206.28	67			
	Between groups	6725.245	3	2241.748	3.099	0.031
CBH	Within groups	65107.9	90	723.421		
	Total	71833.15	93			
	Between groups	131041.8	3	43680.6	23.502	0
Cover	Within groups	204448.2	110	1858.62		
	Total	335,490	113			
	Between groups	32264.92	3	10754.98	3.092	0.031
Age	Within groups	313077.7	90	3478.641		
	Total	345342.6	93			

Table 5.
ANOVA of vegetation parameters and age of F. benghalensis in four main sites.

significant variables using Duncan test showed that site three has the highest mean values of different parameters (height = 10.1, CBH = 138.9 m, cover = 229.8 m^2, and age = 304 years), followed by site number one and four. On the other hand, site number two showed the lowest mean values of these parameters (CBH = 9.8 m and cover = 51.6 m^2) (**Table 3**). Analyses of variance of vegetation parameters and age for *F. benghalensis* in the four studies area are shown in (**Table 5**).

Correlation analysis of different parameters showed highly significant direct correlation between age dating, cover, height, CAB, and CBH. Age dating data showed the highest correlation coefficient with CBH (r = 1), and CAB (r = 0.909) followed by cover (r = 0.694) and height (r = 0.651).

10. Discussion

F. benghalensis is the world's largest tree as far as its spread [20]. The tree is native to India and Pakistan (it is named for Benghal). It is a popular shade-tree, cultivated in many tropical countries [21]. The plant begins growth on other trees and eventually envelopes them completely. Aerial roots hang down from the branches and these eventually become trunks. This circle of trunks deriving from one original tree can reach an enormous size—200 m in diameter and 30 m in height. Their shade has made them important gathering places. Known in Hindu mythology as "the wish-fulfilling tree," banyans represent eternal life. The tree is sacred to Hindus and Buddhists in India and is frequently planted around temples. Being a majestic ornamental tree it is also planted in parks and along streets in the tropics. In temperate climates it is grown as an indoor plant. In general, the banyan is a source of dye and shellac—an important component in French polish—produced by lac insects which inhabit the tree as pests.

The present study showed that populations of *Ficus benghalensis* have limited distribution in Ismailia city (target study area). Four sites were only recognized in the study area. The limitation may return to the requirements of the pollinating wasp (*Eupristina masoni*). However, Ismailia is located near the midpoint of the Suez Canal, on the northwestern shore of Lake Temsaḥ. The lake, is a natural depression, was connected to the Gulf of Suez of the Red Sea in pharaonic times. The city was founded in 1863 by the French engineer Ferdinand de Lesseps, constructor of the Suez Canal, as a base camp. It was named for the ruling Egyptian khedive (viceroy) Ismail Pasha, whose elaborate palace built for the gala opening of the canal in 1869 has fallen into ruin. Laid out in the nineteenth century style, with broad avenues, tree-lined squares, parks, and gardens, it has a gridiron street plan. Ismailia was cultivated with huge number of *F. benghalensis* and now is subjected to huge human threats affecting the presence of these trees.

Nadel et al. [10] described the pollination process which is the main reason of not producing off spring of *F. benghalensis*. In general, the genus *Ficus* (*Moraceae*) is distributed in the tropics and subtropics worldwide. About one-half of the species are monoecious, the rest being gynodioecious but functionally dioecious Nadel et al. [10]. Most species grow as trees, while others are shrubs or climber. Frequently, germination in many species occurs on other trees, with the seedlings growing epiphytically while sending a set of connections of roots down to the soil, eventually "strangling" their nurse trees [10]. However, other species begin their lives on rocks or straight in soil. Pantropical in distribution, only a few fig species extend into warm temperate regions.

As expressed by Nadel et al. [10], with few exceptions, each fig species is pollinated by a different species of wasp in the family *Agaonidae* (*Hymenoptera*: *Chacidoidea*) [5, 22–24]. The pollination biology of monoecious species has been described by Galil and Eisikowitch [25, 26]. A pollen-laden female wasp enters the syconium or "fig," an urn-shaped inflorescence which, when in the receptive stage, is lined internally with dozens or hundreds of receptive female flowers and a few immature male flowers. The wasp lays her eggs through the styles into some of the ovaries, pollinating most of the flowers in the process. In dioecious fig species, pollination is more complex. The "male" tree is monoecious, having both male and female flowers in each syconium, but it has only short-styled female flowers. Nearly all of these female flowers are used by the ovipositing wasps, with the result that the tree produces no seeds, only pollen and agaonids. The female tree, on the other hand, is truly female; the syconia contain only female flowers. These flowers are all long-styled, which effectively eliminates the ability of the agaonid to oviposit in them. The absences of this process of pollination cause failure in germination of *F. benghalensis*.

Age dating results of *Ficus benghalensis* reflect the history of Ismailia area, which return to the age of Suez Canal. Digging of Suez Canal started in 1862 another canal, Ismailia canal, was constructed in the same time to sustain thousands of workers with water, food and other requirements. El-Mawany area and Nemra Ceta area showing the oldest *Ficus* trees are located near the construction of those canals. Importance of cultivating *F. benghalensis* at that time not only returns to their huge shade but also to their medicinal importance that could help in treatments of different diseases.

Based on age dating results, the oldest *F. benghalensis* is 394 years old cultivated in site number one (El Mawany), whereas the youngest tree is 96 years old cultivated in site number four (Nemra Ceta). The highest mean of age dating was recorded for the trees cultivated in site number three (304 years) located at Mohamed Ali, followed by *Ficus* trees cultivated in site number one (242 years) located at El-Mawany area. The lowest mean of age dating was recorded for *Ficus* trees cultivate in site number four and two (229, 216 years respectively). Correlation analysis indicates the direct correlation between age dating, cover, height, CAB, and CBH. CBH was the most important parameter could be used to predict the age dating of *F. benghalensis*. The results showed very important notes about growth mode of *Ficus benghalensis*: (a) average number of annual rings per centimeter ranges from 13 and 14 annual rings and (b) *Ficus* tree with age of 96 years has height 3 m and diameter at breast height equals 14 cm and the oldest tree has height and diameter equal 11 m and 57 cm, respectively.

Age dating of *Ficus benghalensis* data may be interpreted with one of the following two hypotheses. First, these trees may be introduced with age ranges from 150 to 200 years (about 20–30 cm diameter, and 5–7 m height) and connected with the Suez Canal construction for shade and medicinal importance. The second hypothesis returns the cultivation of these trees to the age before the construction of Suez Canal, and the populations of *F. benghalensis* were selected to be near to the route of Suez Canal and Ismailia canal to get benefit from their shade and medicinal importance.

10.1 Chemical constituents

Ficus benghalensis plant is an ever green plant of family *Moraceae* having many chemicals compounds present in this plant. There are so many researchers work out on this plant species. According to Patil et al. [27] described that in leaves, stem, bark; root and aerial root have different chemicals so this plant is having medicinal importance.

In *Ficus benghalensis* leaves have quercetin-3-galactoside, rutin, friedelin, taraxoseterol, lupeol, B-amyrin along with psoralen, bergapten and B-sisterol.

The bark of *Ficus benghalensis* has 5,7 dimethyl ether of lucope-largonidin 3-0-∞-L rhamnoside and 5,3, dimethyl ether of leucocynidin 3-0-∞-D galactosyl cellobioside, glycoside, 20-tetra-triaconthene-2-one, 6-heptatriacontene-10-one, pentatriacontan-5-one, beta sitosterol-alpha-D-glucose and meso-inositol Earlieds, glucoside, 20 tetrattria-conthene-2-one, 6-heptaria contene-10-one, pentatriacontan-5-one, beta sitosterol-alpha-D-glucose, and me-so inositol, leucodelphinidin derivative, bengalenoside: aglucoside, leucopelargonin derivative, leucocynidin derivative, glycoside of leucopelargonidin have been isolated from the bark of *Ficus benghalensis*.

The fruit of *Ficus benghalensis* traditional use of folk medicine for respiratory disorders and certain skin disease. According to Ayurvedic system of medicine *Ficus benghalensis* Linn (banyan tree) is well known to be useful in diabetes. This attracted the attention of many earlier workers who studied the hypoglycemic effect from the bark of *Ficus benghalensis*. *Ficus benghalensis* Linn is a large evergreen tree found throughout forest tracts of India. It is popular Indigenous system of medicine like Ayurveda, Siddha, Unani and Homeopathy. In traditional system of medicine various plant parts such as stem, bark, and root bark aerial roots, vegetative buds, leaves, fruits and latex are used in dysentery, diarrhea, diabetes leucorrhoea, menorrhagia, nervous disorders, tonic and astringent.

11. Medicinal importance of *Ficus benghalensis*

11.1 Anthelmintic activity

The extracts from *Ficus benghalensis* not only to paralyze, but also to kill the earthworms. The aqueous and methanolic extracts were found to be more effective to execute the earthworm when compared to antihelminthic drugs [28].

11.2 Anti-inflammatory activity

The ethanolic (300 mg) and petroleum ether extracts (600 mg/kg/day) of *Ficus benghalensis*, considerably abridged ($P < 0.05$) carrageenan-induced paw edema in rats. The ethanolic and petroleum ether extracts showed a greater anti-inflammatory effect compared with the standard drug Indomethacin. The results indicated the ethanolic extract of *Ficus benghalensis* exhibited more significant activity than petroleum ether in the treatment of inflammation [27].

11.3 Anti-stress and anti-allergic

Various extracts of *Ficus benghalensis* bark was screened for its anti-allergic and anti-stress potential in asthma by milk-induced leukocytosis and milk-induced eosinophilia. Aqueous, ethanol, and ethyl acetate extracts showed significant decrease in leucocytes and eosinophils in the order given while petroleum ether and chloroform extracts were inactive. This shows the application of polar constituents of *F. benghalensis* bark as anti-stress and Anti-allergic agents in asthma [29].

11.4 Antioxidant activity

The extract was examined for its antioxidant activity by DPPH radical scavenging activity, hydroxyl radical scavenging activity, reducing capacity, hydrogen peroxide activity, total phenolic content using Folin-Ciocalteu's phenolic reagent. The extract showed extreme scavenging of DPPH radical (96.07%) at 250 $\mu g\ mL^{-1}$ concentration and hydrogen peroxide (69.23%) at 1000 $\mu g\ mL^{-1}$ concentration. The extract shows good results when compared with other compounds. This shows the scavenging activity of the extract [30].

11.5 Antitumor activity

The chloroform extract of the fruit of *Ficus benghalensis* has shown toxicity in the brine shrimp (*Artemia salina*) bioassay (LC50 < 1000 μg/ml). It also possessed antitumor activity in the potato disc bioassay (% tumor inhibition >20%).

The other tested extracts showed no marked inhibition on the uptake of calcium in to rat pituitary cells GH_4C1. The results support the traditional use of these plants in Folk medicine for respiratory disorders and certain skin diseases [9, 10].

11.6 Antidiarrheal activity

The ethanol extract of the hanging roots of *Ficus benghalensis* has been evaluated for antidiarrheal activity against different investigational models of diarrhea in rats. The extract (400 mg/kg, orally) has shown significant inhibitory activity against castor oil induced diarrhea (extract fed rats had 2.21 ± 0.27 defecations per animal in 4 hours; control 4.0010.33, $P < 0.001$) and PGE2 induced enteropooling (for extract fed rats the value reported is 1.25 ± 0.15 in terms of intestinal fluid; control 0.78 ± 0.11, $P < 0.02$) in rats. The extract has also been significantly effective in reducing gastrointestinal mobility (extract fed rats: 50.2 ± 2.7%; control 79.412.76%, $P < 0.001$) in charcoal meal test in rats [31].

11.7 Antimicrobial activity

The chloroform concentrates of the product of *Ficus benghalensis* has likewise indicated inhibitory activity (0.5 mg/disc) against the bacterium *Micrococcus luteus* (18–26 mm diameter inhibition zone), which was not inhibited by kanamycin (100 μg/disc), streptomycin (100 μg/disc) or penicillin (5 μg/disc). *Streptococcus faecalis* and *Streptococcus faecium* were also inhibited by the fruit extract (17–20 mm inhibition zone). Other bacteria such as *Bacillus cereus*, *B. megaterium*, *Staphylococcus aureus*, *Streptococcus epidermis*, *Streptococcus lactis*, *Escherichia coli*, *Klebsiella pneumonia*, *Proteus vulgaris* and *Pseudomonas aeruginosa* were inhibited to a lesser extent (16–19 mm inhibition zone) [9, 10].

11.8 Antifungal activity

Mitosporic fungi and several sterile forms were isolated as endophytes from the leaf tissues and aerial roots of *Ficus benghalensis*. Although similar number of endophyte species was present in lamina and petiole, the endophytic fungi more densely colonized the petiole. The species composition and the colonization frequency of the endophytes were more for the aerial roots entering the soil when compared with those growing in the air since the roots recruited some endophytes from the soil.

The endophyte assemblages of the leaf and aerial root and of the aerial root growing in the air and soil showed little overlap suggesting that the nature of the host tissue as well as the environment determine the endophyte composition of a host [31].

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Starting in the 1940s, humans have aimed to increase agricultural productivity. However, along with the benefits gained, there have been several criticisms since the 1970s, especially about food security and environmental impacts. Nowadays, the demand for food is increasing while the quantity and quality of agricultural production is declining due to human-induced environmental problems, i.e. climate change and water scarcity. Moreover, our modern fruit industry needs to improve quality and quantity of fruit production while also protecting ecosystems by reducing environmental impacts. Hence, this book intends to provide the reader with a comprehensive overview of the new and eco-friendly technologies in the modern fruit industry.

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