

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



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 high-bush 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 $+5^{\circ}\text{C}$, 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 $+3^{\circ}\text{C}$. 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 $+24^{\circ}\text{C}$, 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 2 m²). 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 m² [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 nematocides. These nematocides 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, peroxy 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 peroxy 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.

IntechOpen

Author details

Zofia Zydlik^{1*}, Szymon Cieśliński¹, Van Chung Mai², Nesibe Ebru Kafkas³
and Iwona Morkunas^{4*}

1 Department of Dendrology, Pomology and Nursery Production, Faculty of Horticulture and Landscape Architecture, Poznań University of Life Sciences, Poznań, Poland

2 Department of Plant Physiology, Faculty of Biological Sciences, University of Vinh, Vinh, Vietnam

3 Department of Horticulture, Faculty of Agriculture, University of Cukurova, Adana, Turkey

4 Department of Plant Physiology, Faculty of Horticulture and Landscape Architecture, Poznań University of Life Sciences, Poznań, Poland

*Address all correspondence to: zofia.zydlik@up.poznan.pl;
iwona.morkunas@gmail.com and iwona.morkunas@mail.up.poznan.pl

IntechOpen

© 2019 The Author(s). Licensee IntechOpen. This chapter is distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/3.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. 

References

- [1] FAO. FAOSTAT. Rome: Food and Agriculture Organisation of the United Nations; 2017 Available from: <http://faostat.fao.org>
- [2] Rejman A, Pliszka K. Highbush Blueberry. Warsaw: PWRiL; 1991. p. 112. ISBN: 83-09015-14-3 (in Polish)
- [3] Panicker GK, Malta FB. Effect of abscisic acid and paclobutrazol on cold hardiness of rabbiteye blueberry (*Vaccinium ashei* Reade). *Acta Horticulturae*. 2016;(1117):315-320
- [4] Huang X, Chen MH, Yang LT, Li YR, Wu JM. Effects of exogenous abscisic acid on cell membrane and endogenous hormone contents in leaves of sugarcane seedlings under cold stress. *Sugar Tech*. 2015;17(1):59-64
- [5] Ruban NN, Pawłowski NB. Influence of climatic conditions on the fruiting of *Vaccinium corymbosum* L. in Belarus. In: International Conference "Growing Blueberries and Cranberries with Elements of Ecology"; 29-22 June 2006; Skierniewice. pp. 187, 2006-193. ISBN 83-88707-92-2 (in Polish)
- [6] Pliszka K. Blueberry. Agricultural and Forestry Publishing House, Warsaw; 2002. ISBN 83-915617-8-X (in Polish)
- [7] Stępień W. Impact of precipitation and temperature on the yield of two blueberry varieties in many years fertilizing experiments in Skierniewice. In: International Conference "Growing Blueberries and Cranberries with Elements of Ecology"; 29-22 June 2006; Skierniewice. pp. 53, 2006-58. ISBN 83-88707-92-2 (in Polish)
- [8] Garamhegyi T, Kovcas J, Pongracz R, Tanos P, Hantrani IG. Investigation of the climate-driven periodicity of shallow ground water level fluctuations in a central-eastern European agricultural region. *Hydrogeology Journal*. 2018;26(3):677-688
- [9] Moore JN. Adapting low organic upland mineral soils for culture of highbush blueberries. *Acta Horticulturae*. 1993;346:221-229
- [10] Davies FS, Johnson CR. Water stress growth and critical water potentials of rabbiteye blueberry. *Journal American Society for Horticultural*. 1983;107:6-8
- [11] Cho HS, Cho YS, Park MY, Kim ES, Ji SM, Jung BJ. Comparative growth of blueberry (*Vaccinium corymbosum*) cultivated in two different planting ridge heights against water-logging damage. *Acta Horticulturae*. 2016;1117:331-334
- [12] Sumorok B, Michalska-Hejduk D, Zdanowicz A, Drobnińska A, Sas Paszt L. The occurrence of mycorrhizal fungi in selected plant communities in the Kurówka River valley. *Water-Environment-Rural Areas*. 2009;3(27):195-204 (in Polish)
- [13] Zydlik Z, Pacholak E, Rutkowski K, Styła K, Zydlik P. The influence of a mycorrhizal vaccine on the biochemical properties of soil in the plantation of blueberry. *Zemdirbyste-Agriculture*. 2016;103(1):61-66
- [14] Borkowska B, Krzewińska D. Inoculation of young cranberry and blueberry plants with fungi isolated from polish ecosystems. *Acta Horticulturae*. 2009;(810):689-696
- [15] Cartmill AD, Valdez-Aquilar LA, Bryan DL, Alarcon A. Arbuscular mycorrhizal fungi enhance tolerance of vinca to high alkalinity in irrigation water. *Scientia Horticulturae*. 2007;15:275-284
- [16] Ortas I. Field trials in mycorrhizal inoculation in the eastern

Mediterranean horticultural region.
In: Feldman F, Kapulnik Y, Baar J,
editors. Mycorrhiza Works. Germany:
Hannover; 2008

[17] Porras-Soriano A, Soriano-
Martín ML, Porras-Piedra A, Azcón R.
Arbuscular mycorrhizal fungi increased
growth, nutrient uptake and tolerance
to salinity in olive trees under nursery
conditions. *Journal of Plant Physiology*.
2009;**166**:1350-1359

[18] Chatzistathis T, Orfanoudakis M,
Aligras D, Therios I. Colonization
of Greek cultivars' root system by
arbuscular mycorrhiza fungus: Root
morphology, growth, and mineral
nutrition of olive plants. *Scientia
Agricola*. 2013;**70**(3):185-194

[19] Gamal MA, Abdul-Wasea AA,
Salem MA, Eslam MA. Influence of
arbuscular mycorrhiza and phosphorus
fertilization on the gas exchange
status, growth and nutrient contents
of soybean (*Glycine max.* L) plants
grown in a sandy loam soil. *Journal of
Fruit Agriculture and Environment*.
2014;**12**(1):150-156

[20] Asrar AA, Abdel-Fattach M,
Elhindi KM. Improving growth, flower
yield and water relations of snapdragon
(*Antirrhinum majus* L.) plants grown
under well-watered and water-stress
conditions using arbuscular mycorrhizal
fungi. *Photosynthetica*. 2012;**50**:305-316

[21] Smith SE, Jakobsen I,
Grønlund M, Smith FA. Roles of
arbuscular mycorrhizas in plant
phosphorus nutrition: Interactions
between pathways of phosphorus
uptake in arbuscular mycorrhizal
roots have important implications
for understanding and manipulating
plant phosphorus acquisition. *Plant
Physiology*. 2011;**156**:1050-1057

[22] Hu J, Lin X, Wang J, Dai J, Cui X,
Chen R, et al. Arbuscular mycorrhizal
fungus enhances crop yield and

P-uptake of maize (*Zea mays* L.): A
field case study on a sandy loam soil
as affected by long-term P-deficiency
fertilization. *Soil Biology and
Biochemistry*. 2009;**41**(12):2460-2465

[23] Mayhew L. Humic substances
in biological agriculture. In:
Productkey PL. [online]. Last
actualisation: 2015-11-03. 2004
[Pozyskano: 2018-01-13; 16:09]. 2004.
Dostępny w: [http://productkeys.us/s/
Humic-Substances-Dr-Mayhew.pdf](http://productkeys.us/s/Humic-Substances-Dr-Mayhew.pdf)

[24] Aitken JB, Acock B, Senn TL. The
characteristics and effect of humic acids
derived from leonardite. South Carolina
Agricultural Experiment Station
Technical Bulletin. 1964;**1015**:1-28

[25] Dudley JB, Pertuit AJ, Toler JE.
Leonardite influences zinnia
and marigold. *HortScience*.
2004;**39**(2):251-255

[26] Ece A, Saltali K, Eryigid N,
Uysal F. The effects of leonardite
applications on climbing bean
(*Phaseolus vulgaris* L) yield and some
soil properties. *Journal of Agronomy*.
2007;**6**(3):480-483

[27] Lisek J. Ways to reduce weeds
on highbush blueberry plantations.
In: National Conference "Growing
highbush blueberries"; 21 April 2010;
Skierniewice. pp. 64-70. ISBN 978-83-
60573-41-9 (in Polish)

[28] Zasada T, Pinkerton IJ, Koch C. Host
status and damage potential of
Paratrichodorus renifer Forge and
Pratylenchus penetrans (Nematoda)
to blueberry (*Vaccinium* spp.).
Canadian Journal of Plant Pathology.
2010;**34**(2):277-282

[29] Jagdale GB, Holladay T,
Brannen PM, Cline WO, Aquedelo P,
Nyczepir AP, et al. Incidence and
pathogenicity of plant-parasitic
nematodes associated with blueberry
(*Vaccinium* spp.) replant disease in

Georgia and North Carolina. Journal of Nematology. 2013;**45**(2):92-98

[30] Gawlik A, Gołębiowska D. The influence of humic acid on growth of "Ramrod" pea (*Pisum sativum* L.) plants. In: Book of Abstracts: Biostimulators in Modern Agriculture; 7-8 February; Warsaw. 2008. p. 108

[31] Paul EL, Clark F. Microbiology and soil biochemistry. UMCS, Lublin. 2000;**400** (in Polish)

[32] Zydlik P. Phytosanitary plants for growing strawberries, In: Strawberry, raspberry, blueberries, Plantpress Sp. zoo., Kraków, 2016;**11-12**:12-13 (in Polish)

[33] Oleszczak M. Fertilizers that enrich the soil with an organic substance. Monthly Practical Orchard SAD. 2018;**2**:59-62 (in Polish)

[34] Szajdek A, Borowska EJ. Bioactive compounds and health-promoting properties of berry fruits: A review. Plant Foods for Human Nutrition. 2008;**63**:147-156

[35] Moyer RA, Hummer KE, Finn CE, Frei B, Wrolstad RE. Anthocyanins, phenolics, and antioxidant capacity in diverse small fruits: *Vaccinium*, *Rubus*, and *Ribes*. Journal of Agricultural and Food Chemistry. 2002;**50**(3):519-525

[36] Wang SY, Jiao H. Scavenging capacity of berry crops on superoxide radicals, hydrogen peroxide, hydroxyl radicals, and singlet oxygen. Journal of Agricultural and Food Chemistry. 2000;**48**:5677-5684

[37] Seeram NP. Berry fruits: Compositional elements, biochemical activities, and the impact of their intake on human health, performance, and disease. Journal of Agricultural and Food Chemistry. 2008;**56**(3):627-629

[38] Ehlenfeldt MK, Prior RL. Oxygen radical absorbance capacity (ORAC)

and phenolic and anthocyanin concentrations in fruit and leaf tissues of highbush blueberry. Journal of Agricultural and Food Chemistry. 2001;**49**:2222-2227

[39] Battino M, Beekwilder J, Denoyes-Rothan B, Laimer M, McDougall GJ, Mezzetti B. Bioactive compounds in berries relevant to human health. Nutrition Reviews. 2009;**67**(Suppl 1):145-150

[40] Wang LS, Stoner GD. Anthocyanins and their role in cancer prevention. Cancer Letters. 2008;**269**:281-290

[41] Joseph JA, Denisova NA, Arendash G, Gordon M, et al. Blueberry supplementation enhances signaling and prevents behavioral deficits in an Alzheimer disease model. Nutritional Neuroscience. 2003;**6**:153-162

[42] Harris GK, Gupta A, Nines RG, Kresty LA, et al. Effects of lyophilized black raspberries on azoxymethane-induced colon cancer and 8-hydroxy-2'-deoxyguanosine levels in the Fischer 344 rat. Nutritional Cancer. 2001;**40**:125-133

[43] Heinonen IM, Meyer AS, Frankel EN. Antioxidant activity of berry phenolics on human low-density lipoprotein and liposome oxidation. Journal of Agricultural and Food Chemistry. 1998;**46**:4107-4112

[44] Yun JM, Afaq F, Khan N, Mukhtar H. Delphinidin, an anthocyanidin in pigmented fruits and vegetables, induces apoptosis and cell cycle arrest in human colon cancer HCT116 cells. Molecular Carcinogenesis. 2009;**48**:260-270

[45] Rahman M, Riaz M, Desai UR. Synthesis of biologically relevant bioflavonoids-a review. Chemistry and Biodiversity. 2007;**4**:2495-2527

[46] Andriantsitohaina R, Auger C, Chataigneau T, Étienne-Selloum N,

Li H, Martínez MC. Molecular mechanisms of the cardiovascular protective effects of polyphenols. *British Journal of Nutrition*. 2012;**108**:1532-1549

[47] Slemmer JE, Shacka JJ, Sweeney MI, Weber JT. Antioxidants and free radical scavengers for the treatment of stroke, traumatic brain injury and aging. *Current Medicinal Chemistry*. 2008;**15**:404-414

[48] Droge W. Free radicals in the physiological control of cell function. *Physiological Reviews*. 2002;**82**:47-95

[49] Lowe F. Biomarkers of oxidative stress. In: Laher I, editor. *Systems Biology of Free Radicals and Antioxidants*. 1st ed. Vol. 2. Berlin, Heidelberg: Springer; 2014. pp. 65-87

[50] Heim KE, Tagliaferro AR, Bobilya DJ. Flavonoid antioxidants: Chemistry, metabolism and structure–activity relationships. *Journal of Nutritional Biochemistry*. 2002;**13**:572-584

[51] Kähkönen MP, Heinonen M. Antioxidant activity of anthocyanins and their aglycons. *Journal of Agricultural and Food Chemistry*. 2003;**51**:628-633

[52] De Souza VR, Pereira PAP, da Silva TLT, de Oliveira Lima LC, Pio R, Queiroz F. Determination of the bioactive compounds, antioxidant activity and chemical composition of Brazilian blackberry, red raspberry, strawberry, blueberry and sweet cherry fruits. *Food Chemistry*. 2014;**156**:362-368

[53] Wang H, Cao G, Prior RL. Oxygen radical absorbing capacity of anthocyanins. *Journal of Agricultural and Food Chemistry*. 1997;**45**:304-309

[54] Fukumoto L, Mazza G. Assessing antioxidant and prooxidant activity of phenolic compounds. *Journal of*

Agricultural and Food Chemistry. 2000;**48**:3597-3604

[55] Sarma AD, Sreelakshmi Y, Sharma R. Antioxidant ability of anthocyanins against ascorbic acid oxidation. *Phytochemistry*. 1997;**45**:671-674

[56] Shahidi F, Naczk M. Phenolic compounds in fruits and vegetables. In: *Phenolics in Food and Nutraceutical*. Vol. 2004. Boca Raton, USA: CRC Press, Taylor and Francis Group, LLC; pp. 131-156. ISBN 978-0-203-50873-2

[57] Nile SH, Park SW. Edible berries: Bioactive components and their effect on human health. *Nutrition*. 2014;**30**:134-144

[58] de Castro DDSB, Teodoro AJ. Anticancer properties of bioactive compounds of berry fruits: Review. *British Journal of Medicine and Medical Research*. 2015;**6**(8):771-794

[59] Halliwell B. Oxidative stress and neurodegeneration: Where are we now? *Journal of Neurochemistry*. 2006;**97**(6):1634-1658

[60] Kalt W, Howell A, Duy JC, Forney CF, McDonald JE. Horticultural factors affecting antioxidant capacity of blueberries and other small fruit. *HortTechnology*. 2001;**11**(4):523-528

[61] Connor AM, Luby JJ, Hancock JF, Berkheimer S, Hanson EJ. Changes in fruit antioxidant activity among blueberry cultivars during cold-temperature storage. *Journal of Agricultural and Food Chemistry*. 2002;**50**(4):893-898

[62] Connor AM, Luby JJ, Tong BS, Finn CE, Hancock JF. Genotypic and environmental variation in antioxidant activity, total phenolic content, and anthocyanin content among blueberry cultivars. *Journal of the American Society for Horticultural Science*. 2002;**127**:89-97

- [63] Ribera AE, Reyes-Díaz M, Alberdi M, Zuñiga GE, Mora ML. Antioxidant compounds in skin and pulp of fruits change among genotypes and maturity stages in highbush blueberry (*Vaccinium corymbosum* L.) grown in southern Chile. *Journal of Soil Science and Plant Nutrition*. 2010;**10**(4):509-536
- [64] Pertuzatti PB, Barcia MT, Jacques AC, Vizzotto M, Godoy HT, Zambiasi RC. Quantification of several bioactive compounds and antioxidant activities of six cultivars of Brazilian blueberry. *The Natural Products Journal*. 2012;**2**(3):188-195
- [65] Prior RL, Cao G, Martin A, Sofic E, McEwen J, O'Brien C, et al. Antioxidant capacity as influenced by total phenolic and anthocyanin content, maturity, and variety of *Vaccinium* species. *Journal of Agricultural and Food Chemistry*. 1998;**6**:2686-2693
- [66] Zadernowski R, Naczek M, Nesterowicz J. Phenolic acid profiles in some small berries. *Journal of Agricultural and Food Chemistry*. 2005;**53**(6):2118-2124
- [67] Castrejón ADR, Eichholz I, Rohn S, Kroh LW, Huyskens-Keil S. Phenolic profile and antioxidant activity of highbush blueberry (*Vaccinium corymbosum* L.) during fruit maturation and ripening. *Food Chemistry*. 2008;**109**:564-572
- [68] Sun Y, Li M, Mitra S, Muhammad RH, Debnath B, Lu X, et al. Comparative phytochemical profiles and antioxidant enzyme activity analyses of the southern highbush blueberry. *Molecules*. 2018;**23**:2209. DOI: 10.3390/molecules23092209
- [69] Lohachoompol V, Srzednicki G, Craske J. The change of total anthocyanins in blueberries and their antioxidant effect after drying and freezing. *Journal of Biomedicine and Biotechnology*. 2002;**2004**(5):248-252
- [70] Skrovankova S, Sumczynski D, Mlcek J, Jurikova T, Sochor J. Bioactive compounds and antioxidant activity in different types of berries. *International Journal of Molecular Sciences*. 2015;**16**:24673-24706. DOI: 10.3390/ijms161024673
- [71] Reque PM, Steffens RS, Jablonski A, Flores SH, de O Rios A, de Jong EV. Cold storage of blueberry (*Vaccinium* spp.) fruits and juice: Anthocyanin stability and antioxidant activity. *Journal of Food Composition and Analysis*. 2014;**33**:111-116
- [72] Zielinska M, Sadowski P, Błaszczak W. Freezing/thawing and microwave-assisted drying of blueberries (*Vaccinium corymbosum* L.). *LWT Food Science and Technology*. 2015;**62**:555-563
- [73] Giovanelli G, Brambilla A, Rizzolo A, Sinelli N. Effects of blanching pre-treatment and sugar composition of the osmotic solution on physico-chemical, morphological and antioxidant characteristics of osmodehydrated blueberries (*Vaccinium corymbosum* L.). *Food Research International*. 2012;**49**:263-271
- [74] Giovanelli G, Brambilla A, Sinelli N. Effects of osmo-air dehydration treatments on chemical, antioxidant and morphological characteristics of blueberries. *LWT Food Science and Technology*. 2013;**54**:577-584
- [75] Reque PM, Steffens RS, Da Silva AM, Jablonski A, Flores SH, de Oliveira Rios A, et al. Characterization of blueberry fruits (*Vaccinium* spp.) and derived products. *Food Science and Technology (Campinas)*. 2014;**34**(4):773-779