

Review

Phytochemical importance and utilization potential of grape residue from wine production

Marina Gonçalves Cirqueira¹, Samantha Serra Costa^{1*}, Josiane Dantas Viana¹, Carlos Antônio Borges Cohim Silva², Marcelo Andres Umsza-Guez³ and Bruna Aparecida Souza Machado¹

¹Faculty of Technology, SENAI/CIMATEC, National Service of Industrial Learning - SENAI, Salvador, Bahia, Brazil.

²Consulado do Vale, Petrolina/PE – Brazil.

³Federal University of Bahia, Salvador, Bahia, Brazil.

Received 31 August, 2015; Accepted 16 January, 2016

Grapes are placed at a distinct position, both in the economy and at nutritional and phytochemical levels. The number of studies performed aimed at identifying the various bioactive compounds of grapes, how their cellular structures are linked, efficient methods of extraction and its effect on human health are rather relevant. The use of grapes for wine production is a worldwide activity, therefore a great quantity of residues are generated. These, in turn, are commonly used in soil fertilization and animal feed. Research in the food, pharmaceutical, medical and agricultural industries have revealed a great potential of using grape residue to recuperate phenolic compounds, flavonoids and especially the resveratrol. These have been found by these researches to offer benefits associated to its consumption, such as improvements in glucose tolerance in diabetic patients, decrease in the occurrence of cardiovascular diseases, diminishing the symptoms of menopause, protection against osteoporosis, cancer and Alzheimer's disease. The present work aimed at gathering information about the characterization of the phytochemical content of grapes and its residues, as well as gathering data about the extraction process of such compounds and their application in the food industry.

Key words: *Vitis vinifera*, phenolic components, antioxidant capacity, extraction methods, vinification.

INTRODUCTION

Grape production is one of the most important activities in agriculture. Over ten thousand varieties are known around the world. It is estimated that grape production around the world has increased from 59.74 millions of tons in 1990 to 68.31 million in 2010. In the last twenty years, there was an annual average increase of around

0.5% a year (FAO, 2013).

Grapes (*Vitis* sp.) are one of the most important fruit crops worldwide. There are about 60 grape species in the genus of *Vitis*, and the species *Vitis vinifera*, or European grapes, is most widely cultivated (Liang et al., 2014; Urcan et al., 2017). The three main species of grapes are

*Corresponding author. E-mail: manthasc@yahoo.com.br.

the European (*V. vinifera*), the American (*Vitis labrusca* and *Vitis rotundifolia*) and the French hybrid grapes. The phenolic compounds are the most important phytochemicals in the grapes, as they possess various biological activities and benefits to health (Wada et al., 2007). They are consumed as fresh fruits as well as wine, juice and other processed products (Liang et al., 2011; Liang et al., 2014; Xu et al., 2017). About 27% of the grapes are consumed as fresh fruit (table grapes) and 2% as dried fruit, whereas 71% of the crop is processed, especially for winemaking (Wang et al., 2013).

When grapes are processed to produce its derivatives, great quantities of residues are also generated each year, stimulating the development of other economic viable forms of using the waste. The use of grape residue and its components has an important environmental impact in the reduction of waste and the possibility of creating products with high added value. Various studies have been performed to explore the content of bioactive compounds derived from these residues, besides seeking the best method for extraction and its possible incorporation in food products (Ping et al., 2011a, b; Burin et al., 2014; Machado et al., 2014; Yazykova and Andreevna, 2015; Medina-Meza et al., 2015; Mildner-Szkudlarz et al., 2015; Santos et al., 2016). Polyphenols have been associated with the bioactive potential of grapes due to their antioxidant, anti-inflammatory, anticarcinogenic and antibacterial activities (Burin et al., 2014; Sahpazidou et al., 2014; Li et al., 2015; Casazza et al., 2016). To obtain an assessment of the full range of phytochemicals in grapes, Liang et al. (2011) we recently analyzed 36 phenolic compounds in the berry samples of 344 representative *V. vinifera* cultivars while Liang et al. (2014) investigated and analyzed phenolic profiles, antioxidant and antiproliferative activities of twenty-four selected *Vitis vinifera* grape cultivars. It was evidenced the phytochemical potential of the samples investigated.

The present work aims at gathering information about the characterization of grapes, its residues and derived products regarding their phenolic compounds content, as well as their extraction mechanisms and application in the food industry.

GRAPES

The three main grapes varieties are the European type (*Vitis vinifera* L.), the American type (*Vitis labrusca* and *Vitis rotundifolia*), and the hybrid French variety. Grapes are classified according to the purpose of their use, as table grapes, wine grapes and dried (raisins). Besides, they can be classified as seeded or seedless. As a tradition, the quality of the products derived from grapes is directly related to aspects of its manufacture, but strongly dependent on the physical-chemical characteristics of the raw material that originated them, that is, the properties originally contained in the grape

(Prozil et al., 2012; Ma et al., 2015; Nogales-Bueno et al., 2017).

The European grapes (*V. vinifera* L.) produce the fine wines, and are a variety of great importance in the global context, as well as widely known in wine making. Examples are the white grapes Chardonnay, Sauvignon Blanc and Gewürztraminer and the red grapes Cabernet Sauvignon, Cabernet Franc, Merlot and Pinot Noir. The American grapes (*V. labrusca*), also known as table grapes, are those used for consumption as a fruit and used for juicing, table wines and cooking in general (Ping et al., 2011a; Liang et al., 2014; Cerqueira and Machado, 2016).

Regarding the nutritional profile, the fruit has various important elements, such as vitamins, minerals, carbon hydrates, fibers and phytochemicals. The chemical composition of grapes varies especially according to climate, soil, variety and cultivar. The physical-chemical properties of the fruit can determine the characteristics of its derivatives. The acidity in the juice, for example, is an indicative of the presence of the organic acids tartaric, malic and citric, which give the product a low pH, contributing to a balance between the sweet and acid tastes. The total soluble solids content and the rate soluble solids/acidity are indicative of the grape ripeness and are the most employed criteria for choosing the best time to commercialize or process the fruit (Gil and Pszczolkowski, 2007; Santiago et al., 2014).

The nutritional importance of the grapes and the products derived from it, such as juices, wines, jams and raisins has been constantly reported (Djilas et al., 2009). The direct correlation between the consumption of grapes and wines as beneficial to the health is due to the high quantity of phenolic compounds present in the fruit, which have a significant antioxidant power (Djilas et al., 2009; Syed et al., 2017; Dumitru and Antoce, 2016).

Grapes are an important source of different phenolic compounds in high concentrations. Therefore, the subproducts of its processing retain significant quantities of these substances, such as flavonoids (anthocyanins, catechins and flavonols), stilbenes (resveratrol), phenolic acids (especially benzoic and hydroxycinnamic acids) and a large variety of tannins and proanthocyanidins. These compounds are extensively recognized and reported to have beneficial effects on the human health, including antioxidant, anti-inflammatory, anticancer, vasodilator and antimicrobial activities (Kchaou et al., 2013; Ahmed et al., 2015).

SOLID RESIDUES FROM GRAPES RESULTING FROM WINE PRODUCTION

Wine making involves the procedures and processes applied to transform ripe grapes in wine. The main phases in the production of white, red and rose wine are harvesting, storing and analysis of grapes; stemming and

crushing; addition of sulphite to wort; addition of pectinolytic enzymes; removal of particulate matter; addition of yeast; adjustment of sugar levels or chaptalization; alcoholic fermentation; maceration; separation of solid and liquid phases; pressing of bagasse; malolactic fermentation; racking; attesting; stabilization; filtrations; stabilization in wooden barrels; cuts; bottling and corking; aging in bottles (Prozil et al., 2012; Cirqueira et al., 2014).

Due to the large number of phases, the processing of grapes for wine making generates a great quantity of residues, derived from different phases of the process. It is estimated that 80% of the global production of grapes is aimed at wine manufacturing. Also, for each 100 L of wine produced, 25 to 31 kg of residues are generated, of which 13 to 17 kg are made of bagasse, which explains why it is considered the main product of grape residues (Spigno et al., 2008; Amarin et al., 2015). With such large quantities of subproducts and residues generated, there is an increasing interest in using these products as raw material for other industries, both for economic reasons and environmental concerns (Prozil et al., 2012; Spigno et al., 2008; Oliveira et al., 2012).

The bagasse is made of skin, stems and seeds. These residues have some important characteristics, such as low pH, a high phytotoxic content and phenolic substances, with high antibacterial properties, which lead to resistance to biological degradation. Normally, these residues are used as fertilizers; however, new alternatives are necessary for its utilization, once the high levels of phenolic compounds in the soil are known to cause germination problems (Negro et al., 2003; Soquetta et al., 2016; Casazza et al., 2016).

Another end given to the grape skin and/or bagasse is its use in making animal feed. This purpose is also limited due to its composition, rich in phenolic compounds, which represents an antinutritional factor. Such fact makes the grape bagasse a residue of great interest for the recovery of these potentially bioactive compounds, recognized by their high antioxidant activity (Ping et al., 2011a, b; Prozil et al., 2012; Lima et al., 2014).

The grape's skin is an expressive source of anthocyanidins and anthocyanins, which are natural dyes and have antioxidant properties, such as the inhibition of lipoperoxidation and antimutagenic activity. The rachis has a high quantity of polyphenol, especially tannic compounds, which have a high nutraceutical and pharmacological potential. Its presence in excess in wine can cause a high astringence to the product, therefore its removal during processing is essential (Souquet et al., 2000; Lima et al., 2014).

The lack of applicability of these residues in new areas is mainly associated to the lack of knowledge about the chemical composition and the structure of the main components of the grape's skin, which corresponds to approximately 90% of the total residue. The majority of the studies are more related to different classes of

extracts, such as anthocyanins, hydroxycinnamic acids, flavonoids, flavonols and glycosides (Kammerer et al., 2004; Zhang et al., 2016a) and less concerned with the evaluation of the basic macromolecular components of the grape's skin (Arnous and Meyer, 2008; Zhang et al., 2016b).

The grape's skin that has been through processing for wine production can be a cheap and valuable raw material for the recovery of biologically interesting polyphenolic compounds and products based on these compounds. The flavonoids present in the grape's skin, especially the anthocyanins, flavonols and flavonoids have been widely studied (Yang et al., 2009). However, there needs to be a more accurate investigation concerning the stilbenes, especially resveratrol, extracted from the skin of different grape varieties and found in considerable quantities (Kammerer et al., 2005; Cirqueira et al., 2013; Rocha et al., 2016).

BIOACTIVE COMPOUNDS OF GRAPE'S SKIN

The red colour and the sensory attributes of a wine's quality, especially red wines, are largely due to the phenolic substances, taste and precursors existent on the cellular wall of the skin. The grapes' skin represents 5 to 10% of the total dry weight of the fruit and acts as a hydrophobic barrier that protects the grapes from physical and other climatic traumas, dehydration, infection by fungi and UV light. During the wine making process, the transference of phenolic compounds of the red grapes for the wine occur mainly from the grape's skin during the maceration phase, which, in the case of red wine making, occurs directly on the crushed fruits (Spigno et al., 2008; Mildner-Szkudlarz et al., 2015).

The phenolic content on the grapes' skin varies from 285 to 550 mg/kg of grapes' skin, depending on the variety of the grapes and on the type of pre-treatment (Pinelo et al., 2005a, 2005b; Rombaut et al., 2014). The potent antioxidant activity of wine and grape extracts on controlling the oxidation of *in vitro* low density lipoproteins were significantly correlated with the action of phenolic compounds present in the samples, especially on the grapes' skin (Tomera, 1999).

Various studies were developed seeking ways of recovering the phenolic compounds from subproducts of wine making and its application as potentially antioxidant food products (Spigno et al., 2008; Pinelo et al., 2005a; Yilmaz et al., 2015).

When the red grapes are processed for wine making, the skin and seeds usually remain in contact with the wine in fermentation for various days. This material is then submitted to a careful extraction, and has a high content of phenolic compounds. This residue still has high levels of phenolic compounds retained in the skin's matrix, which stimulates its use as a source for recovery of phenolic compounds (Kammerer et al., 2005). Various

Table 1. Bioactive compounds and function performed in the food matrix.

Bioactive compound	Function in food matrix	References
Condensed tannin	Anti-microbial and anti-parasite	Naczka and Shahidi (2006)
Cyanidin	Pigmentation	Monagas et al. (2006); Gallego et al. (2013)
Polyphenols	Nutraceutical and antimicrobial	Xu et al. (2017)
Delfinidin	Pigmentation	Revilla et al. (1998)
Flavonoids	Antioxidant	Spinelli et al. (2016)
Flavonoids	Antifungal, cellular antioxidant	Cook and Samman (1996)
Phenolics	Antioxidant	Yilmaz et al. (2015)
Kaempferol Malvidin quercetin	Pigmentation	Nile et al. (2013); Monagas et al. (2006); Montana et al. (2007)
Resveratrol	Antifungal, cellular antioxidant	Ahmed et al. (2015)

factors such as cultivar, growing conditions of vines, period of contact between skin and wine in fermentation, temperature of the process and the presence of seeds and rachis, all affect the transference of phenolics to the wort wine, which in turn determines the available quantity of phenolic components in the bagasse (Pinelo et al., 2005a, b; Mohamed et al., 2016).

In various experiments performed with the objective of recovering bioactive compounds, it was a consensus that it was necessary to determine the best conditions for extraction which benefit the release of phenolic compounds from grapes' subproducts in different solvents (Pinelo et al., 2004; Meyer et al., 1998). Besides the complex composition and the connections between phenolic compounds and the different components of the grape's skin, these aspects are important to increase the efficiency of the extraction process (Vidal et al., 2001).

During the maturation of grapes, environmental factors and endogenous enzymes promote changes that affect the composition and structure of the sugars and phenolic compounds contained in the grapes. Research demonstrates that wines made from more mature grapes, in general have a higher content of anthocyanins, a lower rate anthocyanins -flavonol and a higher quantity of some simple phenolic compounds, such as gallic acid and siringic acid (Vidal et al., 2001).

Generally, despite the fact that the phenolic composition varies greatly depending on the variety and growing conditions of the grapes, the skin has the higher content of tannins of the fruit. These tannins differ depending on the fractions of the grape, and could present a higher degree of polymerization and a lower quantity of gallates (Souquet et al., 2000). Catechin, epicatechin and epicatechin gallate are the main unities that constitute the tannins of the skin (Sun et al., 1996). However galocatechin and epigallocatechin are also present in lower quantities (Souquet et al., 2000; Chira et al., 2015). Other compounds, such as quercetin 3-glycuronide were also detected in considerable quantities in the seeds, followed by catechin, caftaric acid and astilbin (Souquet et al., 1996; Lima et al., 2014). Table 1 presents other studies that report different functions

performed by the bioactive compounds of grapes in food.

The grape's skin can be divided in three layers. The outer layer, or cuticle, is composed of hydroxylated fatty acids called cutina, and it is covered by hydrophobic wax (Lecas and Brillouet, 1994). The intermediate layer, the epidermis, consists of one or two layers, which appear as a group of regular cells. The internal layer, hypodermis, which is the closest to the pulp, is composed by various layers of cells containing the majority of the phenolic compounds of the grape's skin (Lecas and Brillouet, 1994; Zhang et al., 2015).

The cellular wall of the grapes forms a barrier against the diffusion of various components, including smells and phenolic compounds, being composed of 30% neutral polysaccharides (cellulose, xyloglucan, arabinan, galactan, xylan and mannan), 20% of pectinic acid substances (of which 62% are methyl sterified), approximately 15% of insoluble proanthocyanidins, and < 5% structural proteins (Lecas and Brillouet, 1994). Bioactive compounds that occur in plants can be linked to the polysaccharides of the cellular wall, confined in vacuoles, associated to the nucleus through different chemical chains, or even joined by physical chains, depending on the composition and disposition of both the phenolic compound and the polysaccharide (Le Bourvellec et al., 2005).

The retention of phenolic compounds on the cellular wall depends on compositional and structural parameters such as stereochemistry, conformational weight or molecular flexibility. Besides that, physical characteristics of the cellular wall, such as surface topography, porosity and chemical composition can also influence the eventual aggregation between conformational polysaccharides from the cellular wall and phenolic substances (Le Bourvellec et al., 2004).

The majority of data about complexation of phenolic compounds with polysaccharides of the cellular wall of plants were obtained using cyclodextrines, polysaccharides in solid state or compounds from the cellular wall of apples prepared by certain chemical treatments. Two mechanisms of association have been proposed to explain the complex links between the

phenolic compounds and the polyssacharides. The first corresponds to hydrogen chains between the hydroxyl groups of the phenolic compounds and the oxygen atoms of ether crossed chains. This way, dextrane gels would be able to encapsulate phenolic compounds inside its pores (Le Bourvellec et al., 2005; Freitas et al., 2003). The second mechanism corresponds to hydrophobic interactions that occur as a result of the ability of certain polyssacharides to develop secondary structures, that is, the nanotubes, or gels, which result in hydrophobic regions. The pockets or hydrophobe cavities can be capable of encapsulating the phenolic compounds, as it has been shown between β -cyclodextrin and different phenolic compounds, such as caffeoylquinic and flavonoids (Le Bourvellec et al., 2005).

Studies have reported an association between flavonoids as the cellular nucleous of various plants. The link of quercetin-3-sulfate to the protein of cellular nucleous of *Flaveria* has already been reported (Grandmaison and Ibrahim, 1996). The association of flavonoids inside the cell nucleous was confirmed by other studies where microscopic techniques made possible the detection of this phenomenon in other vegetable species. Therefore, the substantial quantity of catechin, epicatechin and proanthocyanidins were detected in the cellular nucleous of five different tree species, as well as in flower teas (Feucht et al., 2004a).

However, despite the fact that various studies reveal the existence of an association between flavonoids and the nucleous of vegetal cells, little is known about the specific link between flavonoids and the components of the cell nucleous. In any case, the occurrence of flavonoids related to the nucleous gives rise to new relative questions, such as the possible ability of the phenolic compounds to protect DNA against the oxidative stress through removing free radicals (Albersheim, 2006).

PHENOLIC COMPOUNDS

The phenolic compounds are largely found distributed in plants, and constitute a very diverse group of phytochemicals derived from phenylalanine and tyrosine, including simple molecules and molecules with a high degree of polymerization. In food, the phenolic compounds are responsible for the colour, astringence, scent and oxidative stability (Alasalvar et al., 2001; Antonioli et al., 2015).

The phenolic compounds are included in the category of neutralizers of free radicals, being very efficient in the prevention of auto-oxidation. Phenolics from grapes and red wines were associated to the inhibition of human LDL oxidation (*low-density lipoprotein*) *in vitro*, to the prevention of atherosclerosis and to anti-mutagenic and anti-viral effects (Kaur and Kapoor, 2001; Jara-Palacios et al., 2014). Chemically, the phenolic compounds are defined as substances with an aromatic ring with one or

more hydroxilic substitutes, including its functional groups. The antioxidant activity depends on its structure, particularly on the number and position of the hydroxyl groups, as well as on the nature of the substitutions on the aromatic rings. There are around 8.000 different phenolic compounds which, according to their chemical structure, are divided in classes: Phenolic acids, flavonoids, stilbenes and tanins (Balasundram et al., 2006).

The phenolic compounds present in the grape's skin are linked and/or attached to lignin and polyssacharides present in the matrix of the cellular wall. These compounds are linked by hydrophobe interactions and hydrogen bridges, and can also be in other areas of the vegetal cell, including vacuoles and cell nucleous. The release of the bioactive compounds on the grape's skin starts with the degradation of polyssacharides on the cellular wall, either in wine-making process to enrich the wine, or for obtaining compounds from the bagasse for various purposes (Fang et al., 2008).

The phenolic compounds contained in the vacuoles of the cellular wall are weakly linked to the structure, and are presumably, the most susceptible to be affected by variables such as temperature, solvent-solute ratio and type of solvent used, all of which can modify the balance and conditions of the solid-liquid extraction. The temperature is one of the most important variables to affect the release of the phenolic compounds of the grape's skin; increases in the temperature of extraction contribute to improve both the solubility of the solute and the diffusion coefficient. Consequently, in high temperatures, there is an increase in the content of extracted phenolic compounds (Pinelo et al., 2005a; Xu et al., 2014a).

Despite the positive effects of using higher temperatures for the benefit of extractions, the temperature cannot be increased indefinitely, as in temperatures above 50°C there can be instability of the phenolic compounds and the denaturation of the membranes. The increase of the solvent-solute ratio has also been suggested to increase the yielding of phenolic compounds. However there needs to be a balance between the use of high and low ratio of solvent-solids, in order for an equilibrium between the high costs and the solvents residues to occur, aiming at avoiding the effects of saturation (Pinelo et al., 2005b; Pinelo et al., 2004).

The type of solvent used is also one of the variables that mostly influences the extraction process. Methanol, ethanol and water are the most used solvents for the extraction of phenolic compounds from the grape's skin and bagasse derived from wine production. Among these solvents, the methanol presents a higher capacity to extract phenolic compounds, followed by ethanol and lastly water (Pinelo et al., 2005a). When alcohols are used as solvent in extraction, a progressive release of phenolic compounds from the grape's skin is observed, relative to time of extraction. When water is used as

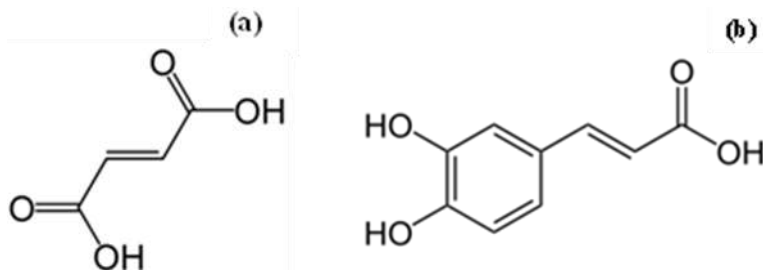


Figure 1. Molecular structure of fumaric acid (a) and caffeic acid (b).

solvent, the time of contact is not so important. Other variables, such as a smaller particle size and higher quantity of sample also benefit the release of phenolic compounds, especially in continuous extraction (Hayouni et al., 2007).

The conditions of extraction can also promote the formation of phenolic compounds that do not occur naturally in the grape's skin. In a study performed (Pinelo et al., 2005b), the formation of polymers of flavan-3-ol was observed in the grape's skin mass, when submitted to a continuous extraction with an ethanolic extractor. Despite observing some of these structural alterations, other factors can occur simultaneously, such as variations in their functional properties, when opting for the limitation in the presence of oxygen during the extraction process of phenolic compounds (Pinelo et al., 2004).

PHENOLIC ACIDS

Phenolic acids are found in higher quantities in the tissues of grape pulp, around 80 to 85%. However, its concentration decreases with the ripening of the fruit and varies according to the cultivar. They are simple compounds formed by an aromatic ring with substitutes capable of sequestering reactive species linked to its structure, such as the radical hydroxyl and the singlet oxygen. The phenolic acids are divided in two groups, the benzoic acids, which have six carbon atoms (C6-C1), such as the gallic acid, *p*-hydroxybenzoic, protocatechuic, vanillic and syringic, and the cinnamic acids (Balasundram et al., 2006). The other group has nine carbon atoms (C6-C3), such as the caffeic, ferulic, *p*-coumaric and sinapic acids (Balasundram et al., 2006). Figure 1 presents the molecular structure of the fumaric acid and caffeic acid.

The relationship between the structures of the phenolic acids and its antioxidant activity has already been established. The antioxidant capacity of the phenolic acids and its esters depends on the number of hydroxyl groups present in the molecule and its position in relation to the carboxyl functional group. Derivates of cinnamic acid are more active as antioxidants than the derivates of

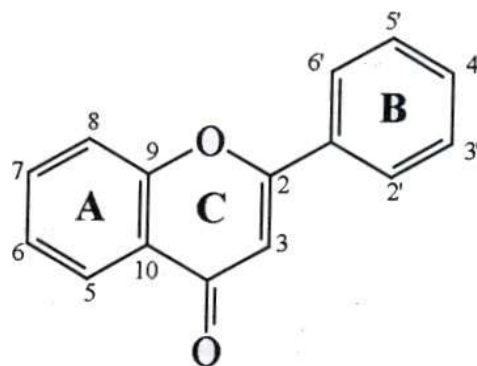


Figure 2. Basic structure of flavonoids.

benzoic acid. This is due to the fact that the first compound presents a higher number of hydroxyl groups in relation to the second, which guarantees a higher ability to donate H⁺ ions and stabilize radicals. The introduction of a second hydroxyl group in the *ortho* or *para* position also increases the antioxidant activity of these compounds (Pinelo et al., 2004).

FLAVONOIDS

Flavonoids constitute the larger group of plant phenolic compounds, being responsible for the colouring of flowers and fruit. They are substances of low molecular weight, composed by 15 carbon atoms. Its general structure is essentially formed by two aromatic rings at the extremities, linked by a bridge of three carbons, usually in the form of a heterocyclical ring (Figure 2). Variations in the substitution configuration of the ring placed between two rings result in the majority of subclasses of flavonoids, which are flavones, flavanones, isoflavones, catechins and anthocyanins (Pinelo et al., 2004).

According to the literature, these compounds have demonstrated activity against allergies, high blood pressure, viral infections, inflammations, arthritis, mutations and carcinogeneses, cancer and AIDS. Its antioxidant potential depends on the number and position

of the hydrogen groups and its conjugations, and also due to the presence of electrons on the benzenic rings. In general, the presence of hydroxyl groups in positions 3, 4 and 5 of the ring at the right extremity has been described as being responsible for increasing the antioxidant activity; however, depending on the flavonoids subclass, the effect could be the opposite (Pinelo et al., 2004).

ANTHOCYANINS

Anthocyanins are flavonoids which are largely distributed in nature, and are responsible for the majority of blue and violet colours, and almost all tones of reds that appear in flowers, fruit, some leaves, stems and roots of plants. In grape vines, they are responsible for the colour of the red grapes, and they transfer, in part, to the wine during vinification, being also found in the pulp of some grape varieties (Versari, 2008).

The use of anthocyanins as colouring arises great interest, but, at the moment, this happens more due to its antioxidant capacity, which is even greater than vitamin E, butyllated hydroxyanisole (BHA) and butyllated hydroxytoluene (BHT). Highly polar, they can substitute the lypophilic antioxidants, such as vitamin E. It is possible to delay the oxidation of frozen fish by adding grape procyanidins (Kang et al., 2003; Kruger et al., 2014). Besides that, both *in vitro* and *in vivo* studies demonstrate the capacity of anthocyanins in reducing the proliferation of cancerous cells and inhibit the formation of tumours (Kang et al., 2003; Lila, 2004).

STILBENES- RESVERATROL

The family of the stilbenes is vast. The resveratrol is the main representative of this group, and it occurs naturally in various plant species, such as mulberry, peanuts and grapes, especially red grapes. The interest of the scientific community for resveratrol was originally due to epidemiological studies which indicated an inverse relationship between the moderate consumption of wine and the risk of coronary disease. Besides, the prevention properties of resveratrol were observed both *in vitro* and *in vivo* (Jang et al., 1997).

The resveratrol (3,5,4-trihydroxi-*trans*-stilbene) is a biologically active substance, belonging to the phytoalexins group (Figure 3). It can be found in nature in the aglycosidic or glycosidic forms, the latter having different denominations, depending on the glycon involved and its geometric form, *trans* and *cis*. The isomer *trans*, which is predominant, is generally more biologically active, however it is also thermo- and photo-sensible, being transformed into *cis* in the presence of visible light, which makes its identification difficult depending on the method used (Vitac et al., 2005).

The majority of its cardio-protective properties are

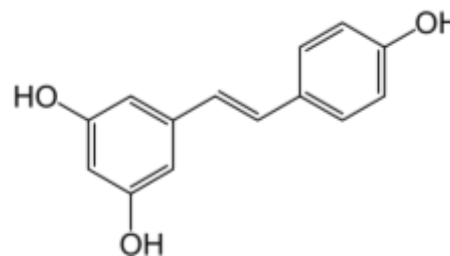


Figure 3. Molecular structure of resveratrol.

associated to its capacity to exert vaso-relaxation and anti-inflammatory response. Among other benefits for the health, the resveratrol has anti-tumoral, anti-diabetic and anti-obesity properties (Li et al., 2012; Jeong et al., 2012).

Generally, the stilbenes are recognized as biological active compounds with anti-fungal action against many pathogens (Jeandet et al., 2002). The most reported anti-microbial effect of resveratrol and other stilbenes is against a common grape vine pathogen, *Botrytis cinerea*, which causes significant losses in grape vines production around the world (Filip et al., 2003). The anti-microbial activity of resveratrol against micro-organisms that cause skin diseases, such as the bacterias *Staphylococcus aureus*, *Enterococcus faecalis* and *Pseudomonas aeruginosa* and the fungi *Trichophyton mentagrophytes*, *Trichophyton tonsurans*, *Trichophyton rubrum*, *Epidermophyton floccosum* and *Microsporum gypseum*, was evidenced (Jeandet et al., 2002). Recently, other important biological actions of resveratrol were reported, such as the capacity to improve tolerance to glucose in diabetic patients, alleviate the symptoms of menopause and protect against osteoporosis, cancer and Alzheimer's disease (Li et al., 2012).

A growing number of studies have examined the pharmacological properties of resveratrol related to many human diseases, including cardiovascular diseases, diabetes mellitus, neurodegenerative diseases and cancer (Saleem et al., 2005). The antioxidant and anti-inflammatory effects of resveratrol perform a crucial role in the therapeutic treatment, although the action mechanisms need to be evaluated more thoroughly (Jeandet et al., 2002; Ksiezak-Reding et al., 2010; Mossalayi et al., 2014).

The content of resveratrol in grapes decreases drastically during its ripening, being practically undetectable in the ripe fruit. Besides, during the process of wine fermentation, the maceration of the bagass into alcohol transfers a great deal of resveratrol from the grape into the wine, reducing, that way, its availability in the bagass (Jeandet et al., 2002; Oliveira et al., 2015).

In grapes, the resveratrol is synthesized almost exclusively in the peel, and peaks of synthesis occur shortly before the grapes reach maturity. The terminal enzyme involved in the biosynthesis of resveratrol is

stilbene synthase. Its activation happens in response to exogenous stress factors, such as lesions, ultraviolet radiation and chemical signs from fungal pathogenic agents. The peak level of resveratrol occurs in a period of approximately 24 h after exposure to stress and decreases after 42 to 72 h, as a result of the activation of stilbene oxidase. The degree of increase in the levels of resveratrol in grapes depends on the variety and exposure to stress (Adrian et al., 2000). Due to resveratrol being produced in response to external stimuli, it is expected that the grapes and the wine have variations in their levels, according to regions and crops. Besides that, many factors such as the increase in temperature, higher levels of SO₂ and decrease of pH, result in higher levels of resveratrol during the process of vinification (Gambutu et al., 2004).

PHENOLIC EXTRACTION DURING VINIFICATION

The general variations in the proportion solids-liquids which affect the yield of phenolic compounds extraction, as well as various technical variables, were specifically referred to as having an influence in the phenolic concentration in wines. Some of these refer to the bursting of the grape cell, breaking the links and rigid structure of the cellular wall, allowing for an increase in the release of phenolic compounds. In general, high fermentation temperatures increase the efficiency of phenolic extraction. Previous works have shown that there is an increase in the total yielding of phenol, but there is only a small difference in the content of anthocyanins in wine whose fermentation had temperatures varying from 15 to 30°C. The levels of sulphur dioxide (SO₂) and the temperatures commonly used for the fermentation of red wine do not affect considerably the extraction of phenolic compounds (Girard et al., 2001; Antonioli et al., 2015).

The use of low temperatures, normally on the range of 10 to 15°C, for many days prior to fermentation does not produce a negative effect on the phenolic composition of the resulting wines. Freezing the wine before fermentation has a potentially bigger effect. Due to freezing, the grapes undergo structural damage and the cellular membranes are broken, thereby releasing the anthocyanins on the (mosto) (Sacchi et al., 2005).

The high temperatures are used in thermovinification, where there the peels are heated up at 60 or 70°C during a short period of time, extracting the phenolic compounds together with the juice, pressing before fermentation. The treatment using heat damages the hypodermic cell membranes, releasing anthocyanins and causing the denaturation of the polyphenoloxidase enzyme, which stops darkening. The pectinolytic enzymes are used to break the (lamella media) between the cells of the pulp and the cellular walls of the peel, releasing the pigments. An improvement in the production of juice and its

colouring has been reported using these enzymes in the extraction process (Ducruet et al., 1997; Rolle et al., 2015).

Regarding maceration, a prolonged period of maceration, of around 4 to 10 days, increases the concentration of anthocyanins and tanins after around a year on bottled wine. It was reported that the reduction in the size of the particles of grape bagass had a positive effect on the recuperation of phenolic compounds of wine bagass. That means that there was an increase in the polysaccharide hydrolysis of the cellular wall, catalysed by several pectinolytic enzymes and mixed preparations of degrading enzymes (Meyer et al., 1998).

METHODS OF EXTRACTION OF BIOACTIVE COMPOUNDS

Use of organic solvents

The use of organic solvents on the extraction of phenolic compounds is one of the most traditional methods of extraction, being greatly used for the obtainment of extracts from various vegetal matrixes (Pinelo et al., 2004).

The conventional techniques of extraction using organic solvents, such as maceration and Soxhlet, are commonly applied in the chemical, pharmaceutical and food industries. This is used for the obtainment of varied extracts and can use a variety of solvents such as methanol, hexane, chloroform, ethyl acetate, acetone, ether, etc. However, these techniques require a high energetic cost and can degrade thermally sensitive substances, as they use high temperatures for extraction or separation of solute-solvent mixture (Castro and Capote, 2010; Kemperman et al., 2013). Assisted maceration by ultrasound is a relatively new technique, based on the use of energy from sound waves, mechanical vibrations transmitted in a frequency superior to the auditive human capacity. In the last decade, its analytical application had a significant increase, particularly in the preparation of samples, rupture of cellular structure and for favouring and accelerate the release of compounds, chemical reactions and physical transformations (Orozco-Solano et al., 2010). There are, basically, two types of device which generate ultrasound waves: water bath and probe. In both, the ultrasound energy is produced by a piezoelectrical ceramic placed between two metallic plates – piezoelectric transducer (Luque-Garcia and Castro, 2003).

The efficiency of this extraction technique is said to be equivalent or higher than that obtained with the Soxhlet extractor. Its advantages are translated into time reduction, temperature of extraction and quantity of reagents, use of different solvents and mixtures, favouring of reactions that would not occur in normal conditions and a consequent increase in yielding (Castro

and Capote, 2010).

Use of enzymes

One of the tools used in the process of decomposing the structure of the cellular wall are the enzymes such as the cellulases, hemicellulases and pectinases, which are capable of catalysing the hydrolysis of the polysaccharides links of the cellular wall (Kashyap et al., 2001; Xu et al., 2014b). Factors such as the relationship between time and temperature of the enzymatic treatment, relationship enzyme and substrate or the type of solvent used in extraction influence the liberation of phenolic compounds of the grape peel. In using acetone at 70% as a solvent and size of the particles between 125 and 250 μm , extracts were obtained from the grape bagass at a concentration of 6055 mg GAE/L of phenolic compounds after 8 h extraction, using a pectinase Grindamyl. When no enzyme was used, however, the concentration of phenolic compounds on the extract decreased to 4615 mg GAE/L, confirming that the assisted enzymatic extraction is one of the most efficient techniques available to increase recuperation of phenolic compounds (Meyer et al., 1998).

Although recuperations significantly better of phenolic compounds from grape peel bagass have been obtained with previous enzymatic treatment and posterior methanolic or aqueous extraction, the data available indicate that only 5 to 10% of the dry matter weight is degraded, which suggests that the degree of degradation of the polysaccharides from the cellular was low (Landbo and Meyer, 2004).

Pectinases are the enzymes mostly used during vinification. Some studies have demonstrated the positive effect of using pectinases during maceration on the increase of phenolic compounds of wine during the processing and conservation. An increase of approximately 40% in the quantity of anthocyanins, corresponding to 220 for 305 mg/L was observed during vinification and conservation of wines treated with pectinases (Palma and Taylor, 1999).

In another study (Bautista et al., 2005), significant differences were reported in the quantity of total phenolic compounds extracted, when the same wine was submitted to the action of pectinase. It is known that the presence of lignin can delay degradation of polysaccharides, performed by enzymes through the unproductive adsorption of proteic enzyme by lignin. Besides that, the enzymes can also be inhibited by the presence of tanins. The improved knowledge about a particular polysaccharide on the cellular wall and a better understanding of how the lignin links to the phenolic compounds in the matrix of the cellular wall of grapes would bring new perspectives for the use of specific enzymes, and therefore, an improvement in the recuperation of these from grapes' peel (Schofield et al.,

1997).

In recent years, the trend towards improving the efficiency of phenolic extraction have concentrated mainly on using enzymes, independently or combined with other enzymes. As highlighted previously, the knowledge about the specific link and distribution of phenolic compounds can allow the utilization of more specific enzymes. The purification of enzymatic mixtures, as well as the synthesis of new enzymes, can promote an improvement of the phenolic yields during extraction (Kammerer et al., 2004; Mohamed et al., 2016).

Besides that, new extraction principles and optimization of the extraction conditions, such as the relationship temperature, solvent-solid, the use of supercritical fluids and projects of extraction of new cells have shown to be promising to optimize the release of phenolic compounds of the grapes' peel and valorization of bagass derived from wine production (Hayouni et al., 2007).

SUPERCritical EXTRACTION

An alternative method to the conventional techniques is the supercritical extraction, which uses supercritical fluids as solvents (Pereira et al., 2004; Shao et al., 2014; Machado et al., 2013). The extraction with supercritical fluid consists in the transference of mass based on the use of fluids to temperatures and pressure above the critical values. The critical temperature is the highest one, at which the gas can be converted in liquid through the increase in pressure. The critical pressure is the highest one, at which the liquid can be converted in gas through the increase in the liquid's temperature (Gomez and Ossa, 2002; Machado et al., 2015; Machado et al., 2016a).

The extraction with supercritical fluid stands out for representing a technology that allows the obtaining of high quality extracts and which minimizes damage to the environment, due to the absence of solvent in the final product. That way, the use of supercritical fluids has been considered a great option for the extraction and fractioning of natural products, particularly for the pharmaceutical and food industries (Pereira et al., 2004; Machado et al., 2013).

Thermal degradation of sensitive compounds is avoided due to being operated in lower temperatures, whilst the absence of light and oxygen prevents oxidative reactions. This constitutes a great advantage for the extraction of antioxidants, ensuring the conservation of its biological properties. The materials processed by supercritical fluids do not require a separate phase for sterilization, once the pressure gradient at the exit of the extractor can generate extracts free from live organisms and spores (Adil et al., 2007; Duba et al., 2015b; Machado et al., 2016b).

In practice, over 90% of extractions performed with supercritical fluids are realized with carbon gas for

Table 2. Extraction methods used to recuperate bioactive compounds in residues derived from grape processing.

Food matrix	Bioactive compound	Functional activity	Extraction method	Extractive conditions	References
Grape peel	Overall Phenolics	Antioxidant	Extraction with supercritical fluid	CO ₂ + ethanol, 170 kgcm ⁻² , 43°C	Pinelo et al. (2005)a
Grape peel	Anthocyanins overall	Antioxidant and anti-inflammatory	Extraction with supercritical fluid	CO ₂ + ethanol, 170 kgcm ⁻² , 46°C	Pinelo et al. (2005b)
Grape bagass	Overall Phenolics	Antioxidant	Ultrasound	Ethanol	Silva et al. (2008)
Grape peel	<i>Trans</i> -resveratrol	Vasodilator and anti-inflammatory	Extraction with supercritical fluid	CO ₂ + ethanol, 40 MPa, 35°C	Casas et al. (2010)
Grape peel	<i>Trans</i> -resveratrol	Vasodilator and anti-inflammatory	Ultrasound	Ethanol	Silva et al. (2008)
Grape peel	Polyphenols	Antioxidant	Counter-current chromatography	methanol-water (80/20; v/v) followed by 3.0 L of acetone-water (75/25; v/v)	Luo et al. (2016)
Grape bagass	<i>Trans</i> -resveratrol	Vasodilator and anti-inflammatory	Ultrasound	Ethanol	Silva et al. (2008)
Seed oil	Tocopherols, tocotrienols, chlorophylls, carotenoids and total phenol contents	Antioxidant	Extraction with supercritical fluid	CO ₂ + ethanol 50 MPa, 50°C	Mohamed et al. (2016)
Grape bagass	Flavonoids	Antioxidant and anti-inflammatory	Extraction with supercritical fluid	CO ₂ + ethanol, 40 MPa, 32°C	Casas et al. (2010)
Grape	Anthocyanin and phenolics	Antioxidant	Radiation	15°C, up to 2 KGy	Gupta et al. (2015)
Grape peel	Flavonoids	Antioxidant and anti-inflammatory	-	Ethanol, 60°C	Katalinic et al. (2010)

*Grape bagass, Peel, seeds and stems. * ERPE, Electronic resonance paramagnetic espectroscopy.

a series of reasons. Besides showing relatively low pressure and temperature (73,8 bar and 31,1°C), CO₂ is inert, non-toxic, non-inflammable, of relatively low cost, easily available in high purity, odourless, and can be readily removed from the final product, leaving no residues (Gomez and Ossa, 2002; Diaz-Reinoso et al., 2006; Machado et al., 2013).

The addition of organic co-solvents such as ethanol, methanol, acetone, among other polar solvents, increases the solvation power of CO₂ and the yield of extraction of polyphenols (Adil et al., 2007; Ni et al., 2015). Despite the supercritical water being frequently used for the destruction of risk-prone organics, the high temperature, above 374°C, the pressure above 220 bar, combined with the corrosive effect of water in these

conditions, have a limited practice in extraction of oil from plants. Water in subcritical conditions has shown to be an effective fluid for the extraction of various classes of oils. This practice is called subcritical extraction or extraction with pressurized liquid (Maier et al., 2009).

In a previous work (Louli et al., 2004) the antioxidant activity of extracts from the bagass of grapes obtained with different liquid solvents was evaluated. It was also submitted to different pre-treatments, posterior to the supercritical extraction, to increment the content of phenolic compounds, the antioxidant activity and the organoleptic properties of these extracts.

In another study (Maier et al., 2009), the residue from grapes seed was used, already used for obtainment of oil through pressing, to recuperate

phenolic compounds. The supercritical extraction has also been previously used as an enzymatic pre-treatment of grape seed to increase the rate of burst/intact cells, favouring the extraction and incrementing the yield of oil (Pazos et al., 2006), as well as for the obtainment of grape seed oil, using ethanol as modifying solvent (Silva et al., 2008). Table 2 shows the extraction methods used in different works in obtaining bioactive compounds.

APLICABILITY OF RESIDUES DERIVED FROM GRAPE PROCESSING

Many studies search the applicability of residues from grape processing, especially aiming at

increasing the input of bioactive components in food products. Products based on grape components are already commercialized in the form of supplements, in powder or capsules (Monagas et al., 2006; Duba et al., 2015a; Güzel and Saygılı, 2016; Machado et al., 2014).

A study was developed (Perin and Schott, 2011) with the aim to elaborate a type of flour from the bagass generated from the processing of grape juice. This was intended to be used in the manufacture of a cookie, added with 5, 10 and 15% of this flour, and the bioactive compounds present in the residue, in the flour and the manufactured cookie would be evaluated. The content of total polyphenols detected had a descending value, from the grape bagass, flour and cookie, respectively, with 56, 18 to 12.07 mg of galic acid/100 mL extract. Both the flour and the cookie suffered some processes which used relatively high temperatures for a significant amount of time, which could have led to alterations/degradations of these components.

In the work developed (Özvural and Vural, 2011), the effects of incorporating grape seed flour obtained from subproducts of wine processing in sausages was investigated. The variations in the physical, nutritional and sensorial parameters were observed in different concentrations of flour. The colour values (L^* , a^* and b^*) of the sausages decreased, in general, with the increasing quantity of grape seed flour. The use of this flour has also led to a decrease in the level of oxidation of the products, probably due to its antioxidants content. The increment of grape seed flour in sausages reinforced the contents of protein, total dietary fibre and capacity of water retention in the treatments. Although the level of grape seed flour above 0.5% reduced the general acceptability, the sausages with levels of up to 2% had marks above average. The evaluation of the incorporation of residue from vinification in the production of healthier and more functional sausages was reached by the study; however the importance of further research aiming to improve the palatability of the products was mentioned.

The input of bioactive compounds from food matrixes has been profusely reported in the scientific area, due to its beneficial importance to health. In the last few years, scientific studies have focused on the use of residues derived from food processing. These residues have great quantities of significant bioactive compounds, concomitantly pushing forward advances regarding extraction techniques and improvements in yielding.

The content of the bioactive compounds obtained from food residues shows great potential for incorporation in food products. It can offer a functional character to products originally poor in such compounds, an important aspect in the attempt to stimulate a healthy life style.

In summary, information reported in this work shows a growth in the number of studies performed in the last few years, as well as the use and possible combination of new techniques.

Conflicts of Interests

The authors have not declared any conflict of interests.

REFERENCES

- Adil IH, Çetin HI, Yener ME, Bayindirli A (2007). Subcritical (carbon dioxide + ethanol) extraction of polyphenols from apple and peach pomaces, and determination of the antioxidant activities of the extracts. *J. Supercrit. Fluids* 43:55-63.
- Adrian M, Jeandet AC, Breuil D, Levite S, Debord S, Bessis R (2000). Assay of Resveratrol and derivate stilbenes in wines by direct injection high performance liquid chromatography. *Am. Enol. Vit.* 51(1):37-41.
- Ahmed HH, Abdel-fatah HM, Hamza AH, Mahmoud RH (2015) Grape seed extract restrains hepatocellular carcinoma: pre-clinical study. *Int. J. Pharm. Biosci.* 6:514-525.
- Alasalvar CJM, Grigor D, Zhang PC (2001). Comparison of volatiles, phenolics, sugars, antioxidant vitamins and sensory quality of different colored carrot varieties. *J. Agric. Food Chem.* 49(3):410-1416.
- Albersheim P (2006). Radical scavenging activity of various extracts and fractions of sweet orange-peel (*Citrus sinensis*). *Food Chem.* 94:19-25.
- Amorin FL, Rocha IS, Ferreira ES, Machado BAS, Umsza-Guez MA (2015). Technological prospecting related to related patent deposits Bioactive compounds present on grapes. *Cadernos de Prospecção.* 8(4):801-807.
- Antoniolli A, Fontana AR, Piccoli P, Bottini R (2015) Characterization of polyphenols and evaluation of antioxidant capacity in grape pomace of the cv. Malbec. *Food Chem.* 178:172-178.
- Arnous A, Meyer AS (2008). Comparison of methods for compositional characterization of grape (*Vitis vinifera* L.) and apple (*Malus domestica*) skins. *Food Bioprod. Process* 86:79-86.
- Balasundram N, Sundram K, Samman S (2006). Phenolic compounds in plant and agri-industrial byproducts: antioxidant activity, occurrence, and potential uses. *Food Chem.* 99:191-203.
- Bautista AB, Martinez A, Ros JM, Lopez JM, Gomez E (2005). Improving colour extraction and stability in red wines: The use of maceration enzymes and enological tannins. *Int. J. Food Sci. Technol.* 40:867-878.
- Burin VM, Ferreira-Lima NE, Panceri CP, Bordignon-Luiz MT (2014). Bioactive compounds and antioxidant activity of *Vitis vinifera* and *Vitis labrusca* grapes: Evaluation of different extraction methods. *Microchem. J.* 114:155-163.
- Casas L, Mantell C, Rodríguez M, Ossa ELMI, Roldán A, Ory ID, Caro I, Blandino A (2010). Extraction of resveratrol from the pomace of Palomino fino grapes by supercritical carbon dioxide. *J. Food Eng.* 96:304-308.
- Casazza AA, Aliakbarian B, Lagazzo A, Garbarino G, Carnasciali MM, Perego P, Guido Busca (2016). Pyrolysis of grape marc before and after the recovery of polyphenol fraction. *Fuel Process. Technol.* 153:121-128.
- Castro MDL, Capote FP (2010). Soxhlet extraction: Past and present panacea. *J. Chromatogr. A* 1217:2383-2389.
- Cerqueira JC, Machado BAS (2016) Obtaining and characterization of grape-peel extract rich in bioactive compounds. *Res. Technol. Innov.* 1:34-44.
- Chira K, Zeng L, Floch AL, Péchamat L, Jourdes M, Teissedre PL (2015). Compositional and sensory characterization of grape proanthocyanidins and oak wood ellagitannin. *Tetrahedron* 71(2):2999-3006.
- Cirqueira MG, Costa SS, Bramont WB, Costa AS, Alves ARC, Dantas EA, Machado BAS (2013). Physical-chemical, microbiological and phytochemical characterization of grapes and residues from juice production. XVIII ENAAL e IV Congresso Latino Americano de Analistas de Alimentos 1(1):1-5.
- Cirqueira MG, Oliveira RS, Umsza-Guez MA, Amorin F, Silva CC, Machado BAS (2014). Desenvolvimento y cuantificación de compuestos bioactivos de producto a base de cascaras de uva

- originária de la producción de jugo. In: V Congreso Internacional de Ciencia y Tecnología de Alimentos.
- Cook NC, Samman S (1996) Flavonoids- chemistry, metabolism, cardioprotective effects, and dietary sources. *Nutr. Biochem.* 7:66-76.
- Díaz-Reinoso B, Moure A, Domingues H, Parajó JC (2006). Supercritical CO₂ extraction and purification of compounds with antioxidant activity. *J. Agric. Food Chem.* 54:2441-2469.
- Djilas S, Canadianovic-Brunet J, Cetkovic G (2009). By-products of fruits processing as a source of phytochemicals. *Chem. Ind. Chem. Eng. Q.* 15:191-202.
- Duba KS, Fiori L (2015a). Extraction of bioactives from food processing residues using techniques performed at high pressures. *Curr. Opin. Food Sci.* 5:15-22.
- Duba KS, Fiori L (2015b) Supercritical CO₂ extraction of grape seed oil: Effect of process parameters on the extraction kinetics. *J. Supercrit. Fluids* 98:33-43.
- Ducruet JA, Dong RM, Glories Y (1997). Influence des enzymes pectolytiques selectionees pour loenologie sur la qualite et la composition des vins rouges. *Rev. Fran. Oenol.* 155:16-19.
- Dumitru E, Antoche AO (2016). A New Early Ripening Table Grape Cultivar Obtained in Pietroasa Research Station: *Vitis vinifera L. cv. Mihaela*. *Agric. Agric. Sci. Procedia* 10:215-221.
- Fang XK, Gao J, Zhu DN (2008). Kaempferol and quercetin isolated from *Euonymus alatus* improve glucose uptake of 3T3-L1 cells without adipogenesis activity. *Life Sci.* 82(11):615-622.
- FAO. FAOSTAT. Database (2012). Available at: <http://www.fao.org/faostat/en/#data>.
- Feucht W, Dithmar H, Polster J (2004). Nuclei of tea flowers as targets for flavonoids. *Plant Biol.* 6:696-701.
- Filip V, Plocková M, Smidrkal J (2003). Resveratrol and its antioxidant and antimicrobial effectiveness. *Food Chem.* 83:585-593.
- Freitas V, Carvalho E, Mateus N (2003). Study of carbohydrate influence on protein-tannin aggregation by nephelometry. *Food Chem.* 81:503-509.
- Gallego MAG, García-Carpintero EG, Sánchez-Palomo E, Viñas MAG, Hermosín-Gutiérrez I (2013) Evolution of the phenolic content, chromatic characteristics and sensory properties during bottle storage of red single-cultivar wines from Castilla La Mancha region. *Food Res. Int.* 51(2):554-563.
- Gambutti A, Strolo D, Ugliano M, Lecce L, Moio L (2004). Transresveratrol, quercetin, (+)-catechin, and (-)-epicatechin content in South Italian monovarietal wines: Relationship with maceration time and marc pressing during winemaking. *J. Agric. Food Chem.* 52:5747-5751.
- Gil GF, Pszczolkowski P (2007). Viticultura: fundamentos para optimizar producción y calidad. Ediciones Universidad Católica de Chile. 535.
- Girard B, Yuskel D, Cliff MA, Delaquis P, Reynolds AG (2001). Vinification effects on the sensory, colour, and GC profiles of Pinot noir wines from British Columbia. *Food Res. Int.* 34:483-499.
- Gomez AM, Ossa EML (2002). Quality of borage seed oil extracted by liquid and supercritical carbon dioxide. *Chem. Eng. J.* 88(1-3):103-109.
- Grandmaison J, Ibrahim RK (1996). Evidence for nuclear binding of flavonol sulphate esters in *Flaveria choraefolia*. *J. Plant Physiol.* 147:653-660.
- Gupta S, Padole R, Variyar PS, Sharma A (2015). Influence of radiation processing of grapes on wine quality. *Radiat. Phys. Chem.* 11:46-56.
- Güzel F, Saygılı H (2016). Adsorptive efficacy analysis of novel carbonaceous sorbent derived from grape industrial processing wastes towards tetracycline in aqueous solution. *J. Taiwan Inst. Chem. Eng.* 60:236-240.
- Hayouni EA, Abedrabba M, Bouix M, Hamdi M (2007). The effects of solvents and extraction method on the phenolic contents and biological activities in vitro of Tunisian *Quercus coccifera L.* and *Juniperus phoenicea L.* fruit extracts. *Food Chem.* 105:1126-1134.
- Jang M, Cai L, Udeani GO, Slowing KV, Thomas CF, Beecher CW (1997). Cancer chemopreventive activity of resveratrol, a natural product derived from grapes. *Science* 275:218-220.
- Jara-Palacios MJ, Hernanz D, Escudero-Gilete ML, Heredia JF (2014). Antioxidant potential of white grape pomaces: Phenolic composition and antioxidant capacity measured by spectrophotometric and cyclic voltammetry methods. *Food Res. Int.* 66:150-157.
- Jeandet P, Douillet-Breuil AC, Bessis R, Deborad S, Sbaghi M, Adrian M (2002). Phytoalexins from the Vitaceae: biosynthesis, phytoalexin gene expression in transgenic plants, antifungal activity and metabolism. *J. Agric. Food Chem.* 50:2731-2741.
- Jeong JK, Moon MH, Bae BC (2012). Autophagy induced by resveratrol prevents human prion protein-mediated neurotoxicity. *Neurosci. Res.* 73(2):99-105.
- Kammerer D, Claus A, Carle R, Schieber A (2004). Polyphenol screening of pomace from red and white grape varieties (*Vitis vinifera L.*) by HPLC-DAD-MS/MS. *J. Agric. Food Chem.* 52:4360-4367.
- Kammerer D, Claus A, Schieber A, Carle R (2005). A Novel Process for the Recovery of Polyphenols from Grape (*Vitis vinifera L.*) Pomace. *J. Food Chem. Toxicol.* 70(2):157-163.
- Kang Y, Siegel PM, Shu W, Drobnjak M, Kakonen SM, Cordon-Cardo C, Guise TA, Massague J (2003). A multigenic program mediating breast cancer metastasis to bone. *Cancer Cell* 3:537-549.
- Kashyap DR, Vohra PK, Chopra S, Tewari R (2001). Applications of pectinases in the commercial sector: a review. *Bioresour. Technol.* 77(2):215-227.
- Katalinic V, Možina S, Skroza D, Generalić I, Abramović H, Miloš M, Boban M (2010). Polyphenolic profile, antioxidant properties and antimicrobial activity of grape skin extracts of 14 *Vitis vinifera* varieties grown in Dalmatia (Croatia). *Food Chem.* 119:715-723.
- Kaur C, Kapoor HC (2001). Antioxidants in fruits and vegetables – the millennium's health. *Int. J. Food Sci. Technol.* 36:703-725.
- Kchaou W, Abbes F, Blecker C, Attia H, Besbes S (2013). Effects of extraction solvents on phenolic contents and antioxidant activities of Tunisian date varieties (*Phoenix dactylifera L.*). *Ind. Crops Prod.* 45:262-269.
- Kemperman RA, Gross G, Mondot S, Possemiers S, Marzorati M, Wiele TV, Doré J (2013). Impact of polyphenols from black tea and red wine/grape juice on a gut model microbiome. *Food Res. Int.* 53:659-669.
- Kruger MJ, Davies N, Myburgh KH, Lecour S (2014). Proanthocyanidins, anthocyanins and cardiovascular diseases. *Food Res. Int.* 59:41-52.
- Ksiezak-Reding HHOL, Santa-Maria I, Diaz-Ruiz C, Wang J, Pasinetti GM (2010). Ultrastructural alterations of Alzheimer's disease paired helical filaments by grape seed-derived polyphenols. *Neurobiol. Aging* 33(7):1427-1439.
- Landbo AK, Meyer AS (2004). Effects of different enzymatic maceration treatments on enhancement of anthocyanins and other phenolics in black currant juice. *Innov. Food Sci. Emerg. Technol.* 5:503-513.
- Le Bourvellec C, Bouchet B, Renard CMGC (2005). Non-covalent interaction between procyanidins and apple cell wall material. Part III: Study on model polysaccharides. *Biochim. Biophys. Acta* 1725:10-18.
- Le Bourvellec C, Guyot SE, Renard C (2004). Noncovalent interaction between procyanidins and apple cell wall material Part I Effect of some environmental parameters. *Biochim. Biophys. Acta* 1672:192-202.
- Lecas M, Brillouet JM (1994). Cell wall composition of grape berry skins. *Phytochemistry* 35:1241-1243.
- Li SH, Zhao P, Tian HB, Chen LH, Cui LQ (2015) Effect of Grape Polyphenols on Blood Pressure: A Meta-Analysis of Randomized Controlled Trials. *PLoS One* 10(9):e0137665.
- Li Z, Pang L, Fang F (2012). Resveratrol attenuates brain damage in a rat model of focal cerebral ischemia via up-regulation of hippocampal Bcl-2. *Brain Res.* 1450:116-124.
- Liang ZC, Cheng L, Zhong GY, Liu RH (2014). Antioxidant and Antiproliferative Activities of Twenty-Four *Vitis vinifera* Grapes. *PLoS One* 9(8):e105146.
- Liang ZC, Christopher OL, Zhong GY, Cheng LL (2011) Polyphenolic profiles detected in the ripe berries of *Vitis vinifera* germplasm. *Food Chem.* 129:940-950.
- Lila MA (2004). Anthocyanins and human health: an *in vitro* investigative approach. *J. Biomed. Biotechnol.* 5(5):306-313.
- Lima MS, Silani ISV, Toaldo IM, Corrêa LC, Biasoto ACT, Pereira GE, Bordignon-Luiz MT, Ninow JL (2014). Phenolic compounds, organic acids and antioxidant activity of grape juices produced from new Brazilian varieties planted in the Northeast Region of Brazil. *Food Chem.* 61:94-103.

- Louli V, Ragoussis N, Magoulas K (2004). Recovery of phenolic antioxidants from wine industry by-products. *Bioresour. Technol.* 92(2):201-208.
- Luo L, Cui Y, Zhang S, Li L, Li Y, Zhou P, Sun B (2016) Preparative separation of grape skin polyphenols by high-speed counter-current chromatography. *Food Chem.* 212:712-721.
- Luque-Garcia JL, Castro MDL (2003). Ultrasound: a powerful tool for leaching. *Trends Anal. Chem.* 22(1):41-47.
- Ma M, Yang Y, Zhang H, Yuan M, Zhu W, Ning J, Yang W, Yang M (2015). Grape pre-evaluation by berry-leaf biochemistry quantitative correlation analysis. *Afr. J. Agric. Res.* 10(50):4543-4553.
- Machado BAS, Silva RPD, Barreto GA, Costa SS, Silva DF, Brandão HN, Rocha JLC, Dellagostin AO, Henriques JAP, Umsza-Guez MA, Padilha FF (2016a). Chemical Composition and Biological Activity of Extracts Obtained by Supercritical Extraction and Ethanolic Extraction of Brown, Green and Red Propolis Derived from Different Geographic Regions in Brazil. *Plos One* 11:e0145954.
- Machado BA, de Abreu Barreto G, Costa AS, Costa SS, Silva RP, da Silva DF, Brandão HN, da Rocha JL, Nunes SB, Umsza-Guez MA, Padilha FF (2015). Determination of Parameters for the Supercritical Extraction of Antioxidant Compounds from Green Propolis Using Carbon Dioxide and Ethanol as Co-Solvent. *PLoS One* 10(8):e0134489.
- Machado BAS, Costa SS, Silva RPD, Alves ARC, Guarieiro LLN, Padilha FF (2016b). Use of Patent Indicators as a Methodology for Evaluating the Development of Supercritical Extraction Technology. *Revista Virtual de Química* 8(4):1079-1093.
- Machado BAS, Pereira CG, Nunes SB, Padilha FF, Umsza-Guez MA (2013). Supercritical Fluid Extraction Using CO₂: Main Applications and Future Perspectives. *Sep. Sci. Technol.* 48:2741-2760.
- Machado BAS, Silva CC, Guedes CMC, Umsza-Guez MA, Cirqueira MG, Oliveira RS (2014). Process for the preparation of concentrate rich in bioactive compounds and product obtained. Invention Patent National Institute of Industrial Property. BR1020140302425,
- Maier T, Schieber A, Kammerer DR, Carle R (2009). Residues of grape (*Vitis vinifera* L.) seed oil production as a valuable source of phenolic antioxidants. *Food Chem.* 112:551-559.
- Medina-Meza IG, Barbosa-Cánovas GV (2015). Assisted extraction of bioactive compounds from plum and grape peels by ultrasonics and pulsed electric fields. *J. Food Eng.* 66:268-275.
- Meyer AS, Jepsen SM, Sorensen NS (1998). Enzymatic release of antioxidants from grape pomace. *J. Agric. Food Chem.* 46(7):2439-2446.
- Mildner-Szkudlarz S, Siger A, Szwengiel A, Bajerska J (2015). Natural compounds from grape by-products enhance nutritive value and reduce formation of CML in model muffins. *Food Chem.* 172:78-85.
- Mohamed HB, Duba KS, Fiori L, Abdelgawad H, Tili I, Tounekti T, Zrig A (2016). Bioactive compounds and antioxidant activities of different grape (*Vitis vinifera* L.) seed oils extracted by supercritical CO₂ and organic solvent. *Food Sci. Technol.* 74:557-562.
- Monagas M, Gomez-Cordoves C, Bartolome B (2006). Evaluation of the phenolic content of red wines from *Vitis vinifera* L. during ageing in bottle. *Food Chem.* 95:405-412.
- Montana MP, Pappano N, Giordano SO, Molina P, Debattista NB, Garcia NA (2007). On the antioxidant properties of three synthetic flavonols. *Die Pharmazie* 62:72-76.
- Mossalayi MD, Rambert J, Renouf E, Micouleau M, Méron JM (2014). Grape polyphenols and propolis mixture inhibits inflammatory mediator release from human leukocytes and reduces clinical scores in experimental arthritis. *Phytomedicine* 21:290-297.
- Naczki M, Shahidi F (2006). Phenolics in cereals, fruits and vegetables: Occurrence, extraction and analysis. *J. Pharm. Biomed. Anal.* 41:1523-1542.
- Negro C, Tommasi L, Miceli A (2003). Phenolic compounds and antioxidant activity from red grape marc extracts. *Bioresour. Technol.* 87(1):41-44.
- Ni Q, Gao Q, Yu W, Liu X, Xu G, Zhang Y (2015). Supercritical carbon dioxide extraction of oils from two *Torreya grandis* varieties seeds and their physicochemical and antioxidant properties. *Food Sci. Technol.* 60:1226-1234.
- Nile SH, Kim SH, Ko EY, Park SW (2013). Polyphenolic Contents and Antioxidant Properties of Different Grape (*V. vinifera*, *V. labrusca*, and *V. hybrid*) Cultivars. *BioMed Res. Int.* 2013:1-5.
- Nogales-Bueno J, Baca-Bocanegra B, Jara-Palacios MJ, Hernández-Hierro JM, Heredia FJ (2017). Evaluation of the influence of white grape seed extracts as copigment sources on the anthocyanin extraction from grape skins previously classified by near infrared hyperspectral tools. *Food Chem.* 221:1685-1690.
- Oliveira LC, Barcellos AD, Machado BAS, Druzian JI (2012). Antioxidant activity of phenolic compounds in red wine: search in scientific and technological foundations. *Cadernos de Prospecção* 5(4):221-228.
- Oliveira RS, Barreto GA, Silva CC, Umsza-Guez MA, Machado BAS (2015). Evaluation of solvent systems in the extraction of bioactive compounds of red grape waste. *Res. Technol. Innov.* 1(1):12-22.
- Orozco-Solano M, Ruiz-Jimenez J, Castro MDL (2010). Ultrasound-assisted extraction and derivatization of sterols and fatty alcohols from olive leaves and drupes prior to determination by gas chromatography-tandem mass spectrometry. *J. Chromatogr. A* 1217:1227-1235.
- Özvural EB, Vural H (2011). Grape seed flour is a viable ingredient to improve the nutritional profile and reduce lipid oxidation of frankfurters. *Meat Sci.* 88:179-183.
- Palma M, Taylor LT (1999). Extraction of polyphenolic compounds from grape seeds with near critical carbon dioxide. *J. Chromatogr. A* 949:117-124.
- Pazos M, Alonso A, Fernandez-Bolanos JL, Medina I (2006). Physicochemical properties of natural phenolics from grapes and olive oil byproducts and their antioxidant activity in frozen horse mackerel fillets. *J. Agric. Food Chem.* 54(2):366-373.
- Pereira CG, Marques MOM, Barreto AS, Siani AC, Fernandes EC, Meireles MAA (2004). Extraction of indole alkaloids from *Tabernaemontana catharinensis* using supercritical CO₂ + ethanol: an evaluation of the process variables and the raw material origin. *J. Supercrit. Fluids* 30(1):51-61.
- Perin EC, Schott IB (2011). Use flour extracted from grape residue in the preparation of biscuit type cookie. Universidade Tecnológica Federal do Paraná. 62. http://repositorio.roca.utfpr.edu.br:8080/jspui/bitstream/1/296/1/FB_C_OALM_2011_2_06.pdf
- Pinelo M, Del Fabbro P, Manzocco L, Nunez MJ, Nicoli MC (2005a). Optimization of continuous phenol extraction from *Vitis vinifera* byproducts. *Food Chem.* 92:109-117.
- Pinelo M, Rubilar M, Jerez M, Sinero J, Nunez MJ (2005b). Effect of solvent, temperature, and solvent-to-solid ratio on the total phenolic content and antiradical activity of extracts from different components of grape pomace. *J. Agric. Food Chem.* 53:2111-2117.
- Pinelo M, Rubilar M, Sineiro J, Nunez MJ (2004). Extraction of antioxidant phenolics from almond hulls (*Prunus amygdalus*) and pine sawdust (*Pinus pinaster*). *Food Chem.* 85:267-273.
- Ping L, Brosse N, Chrusciel L, Navarrete P, Pizzi A (2011a). Extraction of condensed tannins from grape pomace for use as wood adhesives. *Crops Prod.* 33:253-257.
- Ping L, Pizzi A, Guo ZD, Brosse N (2011b). Condensed tannins extraction from grape pomace: characterization and utilization as wood adhesives for wood particleboard. *Crops Prod.* 34:907-914.
- Prozil SO, Evtuguin DV, Lopes LPC (2012). Chemical composition of grape stalks of *Vitis vinifera* L. from red grape pomaces. *Crops Prod.* 35:178-184.
- Revilla E, Ryan JM, Martin-Ortega G (1998). Comparison of several procedures used for the extraction of anthocyanins from red grapes. *J. Agric. Food Chem.* 46(11):4592-4597.
- Rocha GS, Ferreira RS, Pimenta NMA, Fonseca LM, Mafra D, Blondet V, Fonseca AS, Barroso SG (2016). Effects of resveratrol, grape juice or red wine consumption Irisin levels and fibronectin type III domain containing protein 5 and uncoupling protein gene expression modulation in rats. *Clin. Nutr. Exp.* 5:1-5.
- Rolle L, Torchio F, Giacosa S, Río Segade S (2015). Berry density and size as factors related to the physicochemical characteristics of Muscat Hamburg table grapes (*Vitis vinifera* L.). *Food Chem.* 173:105-113.
- Rombaut N, Savoie R, Thomasset B, Bélliard T, Castello J, Van Hecke E, Lanoisellé JL (2014). Grape seed oil extraction: Interest of supercritical fluid extraction and gas-assisted mechanical extraction

- for enhancing polyphenol co-extraction in oil. C. R. Chim. 17:284e292.
- Sacchi KL, Bisson LF, Adams DO (2005). A review of the effect of winemaking techniques on phenolic extraction in red wines. Am. J. Enol. Vitic. 56:197-206.
- Sahpazidou D, Geromichalos GD, Stagos D, Apostolou A, Haroutounian SA, Tsatsakis AM, Tzanakakis GN, Hayes AW, Kouretas D (2014). Anticarcinogenic activity of polyphenolic extracts from grape stems against breast, colon, renal and thyroid cancer cells. Toxicol. Lett. 230(2):218-224.
- Saleem M, Kim HJ, Ali MS, Lee YS (2005). An update on bioactive plant lignans. Nat. Prod. Rep. 22:696-716.
- Santiago WE, Teruel BJ, Tinini RCR, Figueredo DG (2014). Postharvest dehydration of Syrah grapes (*Vitis vinifera* L.) under controlled temperature conditions with real-time monitoring of mass loss. Afr. J. Agric. Res. 10(4):229-234.
- Santos APC, Vanderlinde R, Machado BAS, Mamede MEO (2016). Improving production of aromatic compounds by indigenous yeasts in Chenin Blanc grape must. Afr. J. Agric. Res. 11:2433-2442.
- Schofield P, Pell AN, Krause DO (1997). Molecular beacons: Development of a fluorescence-based hybridization technique for ecological studies with ruminal bacteria. Environ. Microbiol. 63:1143-1147.
- Shao Q, Deng Y, Liu H, Zhang A, Huang Y, Xu G, Li M (2014). Essential oils extraction from *Anoectochilus roxburghii* using supercritical carbon dioxide and their antioxidant activity. Ind. Crops Prod. 60:104e112.
- Silva MV, Loureiro A, Falcão A. (2008). Effect of food on the pharmacokinetic profile of *trans*-resveratrol. Int. J. Clin. Pharmacol. Ther. 46(11):564-570.
- Soquetta MB, Stefanello FS, Huerta KM, Monteiro SS, Rosa CS, Terra NN (2016). Characterization of physicochemical and microbiological properties, and bioactive compounds, of flour made from the skin and bagasse of kiwi fruit (*Actinidia deliciosa*). Food Chem. 199:471-478.
- Souquet JM, Cheynier V, Brossaud F, Moutounet M (1996). Polymeric Proanthocyanidins from grape skins. Phytochemistry 43(2):509-512.
- Souquet JM, Labarbe B, Le Guerneve C, Cheynier V, Moutounet M (2000). Phenolic Composition of Grape Stems. J. Agric. Food Chem. 48:1076-1080.
- Spigno G, Pizzorno T, De Faveri DM (2008). Cellulose and hemicelluloses recovery from grape stalks. Bioresour. Technol. 99:4329-4337.
- Spinelli FR, Dutra SV, Carnieli G, Leonardelli S, Drehmer AP, Vanderlinde R (2016). Detection of addition of apple juice in purple grape juice. Food Control 69:1-4.
- Sun BS, Spranger MI, Silva JMR (1996). Extraction of grape seed procyranidins using different organic solvents. In: Proceedings of the 18th International Conference on Polyphenols. Groupe Polyphénols, Bordeaux (France).
- Syed UT, Brazinha C, Crespo JG, Da-Silva JMR (2017). Valorisation of grape pomace: Fractionation of bioactive flavan-3-ols by membrane processing. Sep. Purif. Technol. 172:404-414.
- Tomera JF (1999). Current knowledge of the health benefits and disadvantages of wine consumption. Trend Food Sci. Technol. 10:129-138.
- Urcan DE, Giacosa S, Torchio F, Segade SR, Raimondi S, Bertolino M, Gerbi V, Pop N, Roll L (2017). 'Fortified' wines volatile composition: Effect of different postharvest dehydration conditions of wine grapes cv. Malvasia moscata (*Vitis vinifera* L.). Food Chem. 219:346-356.
- Versari A (2008). A comparison of analytical methods for measuring the color components of red wines. Food Chem. 106(1):397-402.
- Vidal S, Williams P, O'Neill MA, Pellerin P (2001). Polysaccharides from grape berry cells walls. Part I: tissue distribution and structural characterization of the pectic polysaccharides. Carbohydr. Polym. 45:315-323.
- Vitac X, Bonert A, Vanderlinde R, Valls J, Richard T, Delaunay JC, Merillon JM, Teissedre PL (2005). Determination of stilbenes (δ -viniferin, *trans*-astringin, *trans*-piceid, *cis*- and *trans*-resveratrol, *eviniferin*) in Brazilian wines. J. Agric. Food Chem. 53:5664-5669.
- Wada M, Kido H, Ohyama K, Ichibangas T, Kishikaw N, Ohba Y, Nakashima MN, Kurod N, Nakashima K (2007). Chemiluminescent screening of quenching effects of natural colorants against reactive oxygen species: evaluation of grape seed, monascus, gardenia and red radish extracts as multi-functional food additives. Food Chem. 101:980-986.
- Wang L, Xu M, Liu C, Wang J, Xi H, Wu B, Loescher W, Duan W, Fan P, Li S (2013). Resveratrols in Grape Berry Skins and Leaves in *Vitis Germplasm*. PLoS One 8(4):e61642.
- Xu C, Yagiz Y, Borejsza-Wysocki W, Lu J, Gu L, Ramírez-Rodriguez MM, Marshall MR (2014b). Enzyme release of phenolics from muscadine grape (*Vitis rotundifolia Michx.*) skins and seeds. Food Chem. 157:20-29.
- Xu C, Yagiz Y, Hsu WY, Simonne A, Lu J, Marshall MR (2014a). Antioxidant, antibacterial and antibiofilm properties of polyphenols from muscadine grape (*Vitis rotundifolia Michx.*) pomace against selected foodborne pathogens. J. Agric. Food Chem. 62(28):6640-6649.
- Xu C, Yagiz Y, Zhao L, Simonne A, Lu J, Marshall MR (2017). Fruit quality, nutraceutical and antimicrobial properties of 58 muscadine grape varieties (*Vitis rotundifolia Michx.*) grown in United States. Food Chem. 215:149-156.
- Yang J, Liu RH, Martinson T (2009). Phytochemical profiles and antioxidant activities of wine grapes. Food Chem. 116(1):332-339.
- Yazykova MY, Andreevna KK (2015). The biological properties of extract from seeds, peel and combs of red grapes Marselan Intra-ENTAV 980. J. Biotechnol. 208:S97.
- Yilmaz Y, Göksel Z, Erdogan SS, Öztürk A, Atak A, Özer C (2015). Antioxidant activity and phenolic content of seed, skin and pulp parts of 22 grape (*Vitis Vinifera* L.) cultivars (4 common and 18 registered or candidate for registration). J. Food Process. Preserv. 39:1682-1691.
- Zhang P, Fuentes S, Siebert T, Krstic M, Herderich M, Barlow EWR, Howell K (2016a). Comparison data of common and abundant terpenes at different grape development stages in Shiraz wine grapes. Data Brief 8:1127-1136.
- Zhang P, Fuentes S, Siebert T, Krstic M, Herderich M, Barlow EWR, Howell K (2016b). Terpene evolution during the development of *Vitis vinifera* L. cv. Shiraz grapes. Food Chem. 204:463-474.
- Zhang S, Cui Y, Li L, Li Y, Zhou P, Luo L, Sun B (2015). Preparative HSCCC isolation of phloroglucinolysis products from grape seed polymeric proanthocyanidins as new powerful antioxidants. Food Chem. 188:422-429.