Chia (Salvia hispanica L.): Chemical composition, phenolic compounds, antioxidant activity, and antitumor activity

https://doi.org/10.56238/sevened2024.018-039

Daisy Naomi Tan[1], Marcelo Alexandre Prado[2] and Sheila Oliveira-Alves[3]

ABSTRACT

The intake of bioactive compounds, such as phenolic compounds and omega 3 fatty acids, from foods of animal and plant origin, is important for protection against chronic non-communicable diseases, such as cardiovascular disease, diabetes, cancer, among others. The increase in cancer incidence is related to changes in eating habits, exposure to pollutants, and factors such as consumption of foods with simple sugars and saturated fats. Studies show that the consumption of natural antioxidants present in fruits, vegetables and vegetable oils can reduce the risk of cancer and other chronic diseases. Chia seed, rich in phenolic compounds and α-linolenic fatty acid, has been linked to benefits such as reducing cardiovascular disease, controlling diabetes, and protecting against oxidative stress. Research on chia seeds seeks to understand its health benefits and its potential in preventing diseases related to metabolic imbalance and chronic inflammation, including cancer.

Keywords: Chia (Salvia hispanica L.), Antioxidante, Antitumoral.

[1] Graduated in Food Engineering
Institution: State University of Campinas (UNICAMP)
E-mail: dnaomi20@gmail.com
[2] Doctor in Food Science
Institution: State University of Campinas (UNICAMP)
E-mail: mprado@unicamp.br
[3] Doctor in Food Science
Institution: National Institute of Agrarian and Veterinary Research (INIAV)
E-mail: sheilacris.oliveira.alves@gmail.com

INTRODUCTION

CHIA

Salvia hispanica L. is a biannually cultivated herbaceous plant, belonging to the family Lamiaceae, superdivision of Spermatophyta, and Kingdom Plantae (Mohd Ali et al., 2012). Salvia hispanica L., popularly known as chia, is native to southern Mexico and northern Guatemala, being an important staple food cultivated in the Pre-Columbian period, as the ancient Aztecs and other cultural groups in Mesoamerica cultivated and harvested chia seeds extensively, and its use in cooking occurred in the form of whole grains and crushed (Cahill, 2003).

The chia plant can grow up to 1 meter tall, has opposite arranged leaves, with its flowers being purple or white, with a size between 3 to 4 mm, and the fused parts of the flower contribute to a high rate of self-pollination (Mohd Ali et al., 2012). The color of the seed varies between black, gray and black mottled with white, and its shape is oval, with length ranging from 1.90 to 2.37 mm and diameter from 1.21 to 1.43 mm (Ixtaina et al., 2008), as shown in Figure 1.

Figure 1. Planting chia (a) and chia seed (b). (Source: Tosco 2004; Coelho & Salas-Mellado, 2014).



Currently, chia is cultivated in Argentina, Colombia, Ecuador, Peru, Bolivia, Paraguay and Australia, for commercial purposes of its seeds (Coates & Ayerza, 1996; Busilacchi et al., 2013). Seeds soaked in water, fruit juice, and soups are the most consumed form in the regions of Mexico (Cahill, 2003; Reyes-Caudillo, 2008). In Brazil, chia has been consumed along with breakfast cereals, fruits, yogurts and refreshing drinks such as fruit juices and smoothies.

Chia seeds are a pseudocereal, gluten-free, and therefore have been incorporated into various types of foods, beverages and mixed flour formulations for the bakery industry. In addition, the seeds have been used as nutritional supplements as well as in the manufacture of breakfast cereal bars and biscuits in the United States, Latin America, and Australia (Coelho & Salas-Mellado, 2014).

Chia seeds have about 25.0 to 35.0 g of oil/100 g, and the fatty acids present in its oil are highly unsaturated. Essential fatty acids are the main components in chia oil, linoleic acid (LA, C18:2ω-6, 17.0-20.3 g/100 g oil) and α-linolenic acid (ALA, C18:3ω-3; 60.2-67.8 g/100 g oil),

according to Coates & Ayerza (1996), Álvarez-Chávez et al. (2008), Peiretti & Gai (2009), Ixtaina et al. (2011), Marineli et al. (2014), and Boidora et al. (2017).

The nutritional quality of chia seeds is directly related to its maturation stage, because with increasing maturity, there are reductions in the levels of fatty acids, especially α-linolenic acid, and in the crude protein content (Peiretti & Gai, 2009). Maturity, in fact, is the most important factor affecting seed quality, due to the difference in proportion between plant tissue components, increased lignification during seed development, and increased fiber fractions in plant tissues (Morrison, 1980; Wilson et al., 1991; Peiretti & Gai, 2009).

The crude protein content in the seed varies from 15.95 to 26.03 g/100 g (Ayerza & Coates, 2011; Ayerza, 2013; Marineli et al., 2014), and its content is higher than in other traditional cereal crops, such as wheat (11.00 g/100 g), maize (8.80 g/100 g), rice (13.30 g/100 g), oats (14.92 g/100 g), barley (19.04 g/100 g) and sorghum (12.10 g/100 g) (Ragaee et al., 2006; Rupollo et al. 2006). Although chia is not commercially cultivated as a source of protein, its amino acid profile is balanced, and no limiting factors are observed for an adult's diet, as the seed has considerable levels of threonine (5.91 mg/g), lysine (7.65 mg/g) and leucine (9.75 mg/g), and high levels of arginine (16.34 mg/g). asparagine (13.28 mg/g), and glutamine (25.95 mg/g), as reported by Ayerza (2013).

In addition to proteins, chia seeds have 90 to 94 g/100 g of dry matter, which is composed of carbohydrates (26-41 g/100 g), dietary fiber (18-38 g/100 g), ashes (4-5 g/100 g), minerals and vitamins, according to Mohd Ali et al. (2012) and Marineli et al. (2014). In addition to the nutrients present, when the chia seed is humidified, a mucilaginous transparent gel is formed that remains firmly attached to it, as in its epicarp there are cells that produce mucilage when moistened (Ixtaina et al., 2008). The mucilage formed is a natural exudate, composed mainly of xylose, glucose, and glucuronic acid, forming a branched polysaccharide of high molecular weight (Lin et al., 1994, Muñoz et al., 2012).

Reyes-Caudillo et al. (2008) reported that chia seeds contain about 5 to 6 % mucilage, while Muñoz et al. (2012) cited 7 % mucilage. Coorey et al. (2014) analyzed the mucilage formed in the chia seed, and reported that it has 58% crude fiber and 34% carbohydrates. The mucilage extracted from chia seeds has great potential in food formulations as a thickening agent, emulsifying agent and as a stabilizer.

The dietary fiber content in the seed varies from 18 to 38 g/100 g, with most of this content represented by insoluble dietary fiber (14-35 g/100 g), and the rest by soluble dietary fiber (2.4-3.6 g/100 g). Dietary fiber includes cellulose, hemicellulose, lignin, pectins, gums, mucilage, and other associated polysaccharides and oligosaccharides, and are resistant to digestion and absorption in the human small intestine and complete or partial fermentation in the large intestine (Esposito et al., 2005; Capitani et al., 2012). Soluble fibers are fermented in the colon, while insoluble fibers have an

action in the formation of fecal bolus and have limited fermentation in the colon (Anderson et al., 2009).

In addition, dietary fibers play an essential role in the gut health, nutritional and physiological effects of consumers, as they are significantly associated with a lower risk of developing coronary heart disease, hypertension, diabetes, and obesity (Willem van der Kamp et al., 2010).

CHIA PRODUCTS

Chia oil can be obtained by different methods, such as solvent extraction, pressing and supercritical extraction (Martínez et al., 2012). In the industry, the oil is obtained by cold pressing of the chia seed by expeller process, to obtain an oil with quality and nutritional parameters, according to the values determined by the Codex Alimentarius legislation, however cold pressing results in a partial extraction of the oil (Ixtaina et al., 2010).

The acidity index and the peroxide index are described as reference parameters to determine the quality of the conservation of the oils. According to the quality standard for vegetable oils, the legislation determines a maximum value of 3.30 g oleic acid/100 g oil and 20 mEqO2/kg oil, respectively (Codex Alimentarius, 2003). Ixtaina et al. (2012) reported the value of 1.30 oleic acid/100 g oil for the acid value and 1.00 mEqO2/kg oil for the peroxide index in commercial chia oil obtained by the cold pressing system, a method currently used for the extraction of commercialized chia oil.

The commercialization of chia oil has been growing annually due to its high content of polyunsaturated fatty acids (PUFA). PUFA play an important role in the prevention of chronic diseases, such as hypertension, coronary artery disease, diabetes, and cancer (Poudyal et al., 2012).

PUFAs vary in chia oil in the range of 80.60 to 84.09 g/100 g, and the main fatty acids identified are linoleic and α-linolenic (Álvarez-Chávez et al., 2008; Ixtaina et al., 2011; Marineli et al., 2014; Bodoira et al., 2017). Chia oil can present high variation in the content of polyunsaturated fatty acids, and these differences are attributed to different environmental conditions, such as temperature, light, soil type and available nutrients. Ayerza (1995) reported variations in the concentrations of oleic, linoleic and linolenic acid in the oil due to the location of the seed cultivation, while Ayerza & Coates (2004) reported that chia seeds grown in different ecosystems of South America showed significant differences in oil contents and fatty acid composition. Generally, the content of α-linolenic acid varies in the range of 60.20 to 67.80 g/100 g, as described by Coates & Ayerza (1996); Álvarez-Chávez et al. (2008); Peiretti & Gai (2009); Ixtaina et al., (2011); Marineli et al. (2014) and Boidora et al. (2017).

In addition, edible oils have phytosterols, which have been widely studied for their hypocholesterolemic and anticarcinogenic effects (Pelletier et al., 1995; Phillips et al., 2002;

Kozłowska et al, 2016). Generally, sterols are mainly in free and esterified form, and free sterols can have different physiological effects (Miettinen & Gylling, 1999; Phillips et al., 2002). In chia oil, the total sterol content is between 8.15 and 12.60 g/kg oil, and these levels are similar to evening primrose oil (Oenothera biennis), in which the total sterol content is approximately 10 g/kg oil, but in other unrefined oils, such as olive oil, peanut, rapeseed, safflower, sesame and sunflower, the total sterol content ranges from 1.50 to 8.00 g/kg oil (Álvarez-Chávez et al., 2008). In chia oil, β-sitosterol (7.96 g/kg) is the main component in relation to stigmastanol (1.78 g/kg) and stigmasterol (2.17 g/kg), in general, β-sitosterol represents about 60% of the total sterols in crude vegetable oils (Álvarez-Chávez et al., 2008).

Vegetable oils are major sources of tocopherols, which are natural antioxidants and play important roles in the reproduction and antioxidant mechanisms of animal and plant tissues (Guinazi et al., 2009). Chia oil has a total tocopherol content between 443 and 480 mg/kg, with γ-tocopherol being the most abundant, in the range of 415 to 463 mg/kg (Ciftci et al., 2012; Ixtaina et al., 2012; 2015).

After the extraction of the chia oil by cold welding, the fraction of the residual seed is dried, crushed at 435 μm, thus obtaining the fibrous chia flour. Therefore, flour is the residue of the process of extracting oil from the chia seed. Fibrous flour has a high protein content (32.0-35.0 g/100 g) and crude fibre (21.0-29.0 g/100 g), and considerable lipid content (8.7-14.0 g/100 g), even after processing (Capitani et al., 2012; Segura-Campos et al., 2013; Coorey et al., 2014).

According to Capitani et al. (2012), the levels of dietary fiber in fibrous flour range from 44 to 46 g/100 g, with most of this content represented by insoluble dietary fiber (40-41 g/100 g) and the rest by soluble dietary fiber (4-5 g/100 g). Insoluble fibers contribute to increased fecal bolus volume, reduced intestinal transit time, delayed glucose absorption, and starch hydrolysis. High consumption of soluble dietary fiber reduces postprandial glucose responses after carbohydrate-rich meals, as well as lowering LDL (Low Density Lipoprotein) and total cholesterol levels (Weickert & Pfeiffer, 2008).

In addition, Capitani et al. (2012) report that fibrous chia flour has a high capacity to retain and absorb water, and can be used as an emulsifying agent and stabilizer of emulsions.

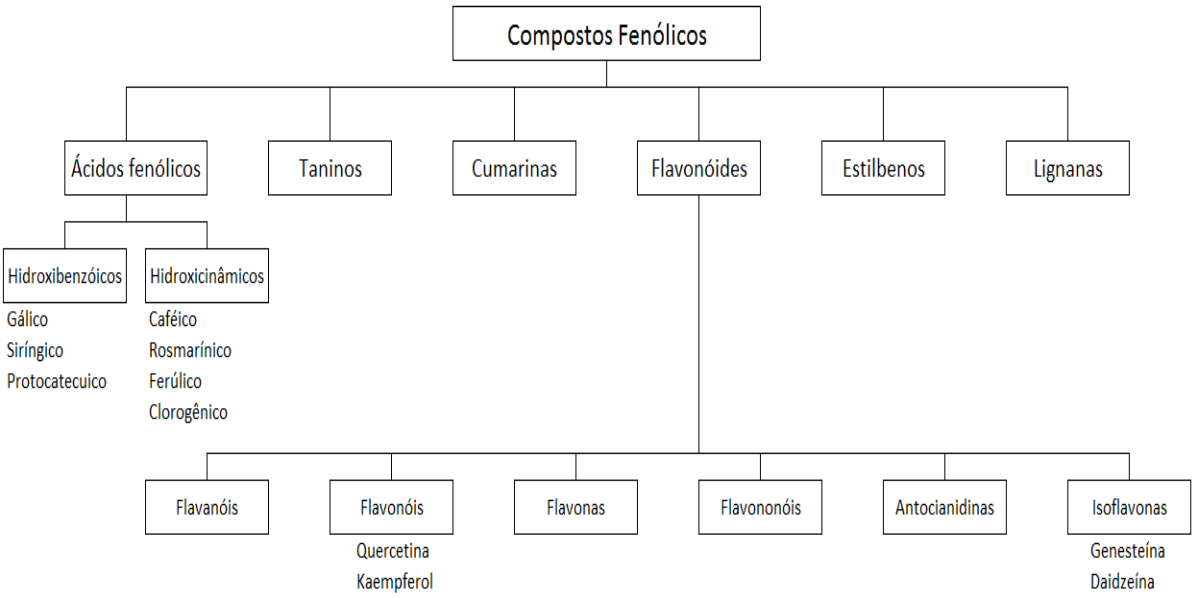
Fibrous flour is a source of minerals, with high levels of calcium (5615.0 mg/kg), magnesium (4624.0 mg/kg), iron (117.7 mg/kg) and phosphorus (9988.5 mg/kg), but low levels of copper (18.7 mg/kg) and zinc (96.0 mg/kg), as described by Capitani et al. (2012). Fibrous chia flour has been used as a raw material in cereal bar production due to its high protein and mineral contents.

PHENOLIC COMPOUNDS

Phenolic compounds are secondary metabolites present in plants and range from simple molecules to others with a high degree of polymerization, and may be present in free form, linked to sugars (glycoside) and proteins in various parts of plants (Acosta-Estrada et al., 2014; Heleno et al., 2015). Phenolics are characterized by the presence of one or more aromatic rings attached to at least one hydroxyl radical and/or other substitutes, and can be divided according to the number of phenolic rings and the structures to which they are attached (D'Archivio et al., 2007; Oliveira & Bastos, 2011).

In addition, phenolic compounds play an important role in plants in growth and reproduction, act as antipathogenic agents and also contribute to plant pigmentation. In foods, phenolic compounds can contribute to bitterness, astringency, color, flavor, aroma, and oxidative stability (Naczk & Shahidi, 2006). Among the more than five thousand phenolics described, flavonoids, coumarins, tannins, lignans, stilbenes, and phenolic acids stand out, as they have antioxidant activity (Shahidi & Ambigaipalan, 2015), as shown in Figure 2.

Figure 2. Classification of phenolic compounds. (Adapted from Shahidi & Ambigaipalan, 2015).



The most abundant groups of phenolic compounds in food are flavonoids, phenolic acids, and lignans. (D'Archivio et al., 2007; Oliveira & Bastos, 2011). Phenolic acids and flavonoids usually occur in conjugated soluble (glycoside) and insoluble forms (Nardini & Ghiselli, 2004; Acosta-Estrada et al., 2014). In nature, phenolic acids occur mainly in insoluble or bound forms, while flavonoids are presented as glycosides, with a single or multiple sugar moieties linked through an OH group (O-glycoside) or through carbon-carbon bonds (C-glycoside).

Phenolic acids are present in almost all plant-derived foods, representing a significant portion of the human diet. The average intake of phenolic acids in humans has been reported to be on the order of 200 mg/day, depending on dietary habits and preferences (Clifford & Scalbert, 2000).

The bioavailability of phenolic acids is crucial for their biological properties (Heleno et al., 2015). When ingested in free form, phenolic acids are rapidly absorbed by the small intestine and subsequently conjugated, however, the chemical structures of the compounds can also influence conjugation reactions, as well as the amount of metabolites formed by the gut microbiota in the colon (Scalbert & Williamson, 2000; Heleno et al., 2015).

After ingestion and absorption, phenolic acids are conjugated by methylation, sulfation, and glucuronidation reactions, which are controlled by specific enzymes that catalyze these steps, and conjugation reactions vary according to the nature of the phenolic acid and the amount ingested (Heleno et al., 2015).

Several factors alter the bioavailability of phenolic acids present in foods, such as the complexity of the food matrix, the chemical form of the compound, intestinal transit time, gastric emptying, compound metabolism and degree of conjugation, possible interactions with proteins in the bloodstream and tissues, composition of the gut microbiota, and the genetic profile of the individual (Crozier et al., 2009; Oliveira & Bastos, 2011)

The analysis methodology used to determine the free and bound forms of phenolic acids generally consists of extraction with aqueous organic solvents to obtain soluble phenolics, followed by a hydrolysis treatment to obtain bound phenolic compounds. Generally, ultrasound-assisted hydrolysis associated with acid hydrolysis has been used to leachate and hydrolyze phenolic compounds more rapidly than traditional methods, since the surface contact area between the solid and liquid phases is increased by particle rupture (Herrera & Luque de Castro, 2004).

In the separation, identification and quantification of phenolic compounds, advanced techniques are used, such as High Performance Liquid Chromatography (HPLC). This technique is the most commonly used, coupled to detectors such as diode array (DAD), fluorescence or mass spectrometer (MS). The use of HPLC is a tool that helps the most varied studies, as it separates compounds present in a matrix, which can be compared to standards for their identification and quantification.

The main class of phenolic compounds identified in chia seeds, using high-performance liquid chromatography analysis, were phenolic acids, which can be divided into two main groups, hydroxybenzoic and hydroxycinnamic acids, which are synthesized from the shikimic acid pathway (D'Archivio et al., 2007; Oliveira & Bastos, 2011).

Martínez-Cruz et al. (2014) analyzed chia seed by ultra-high performance liquid chromatography coupled to diode array detector (UHPLC-DAD), and the main compounds

quantified were rosmarinic acid (0.92 mg/g seed), protocatechuic ethyl ester acid (0.74 mg/g seed), caffeic acid (0.02 mg/g seed), gallic acid (0.01 mg/g seed) and daidzein (0.006 mg/g seed).

Reyes-Caudillo et al. (2008) identified chlorogenic acid (0.10 mg/g seed) and caffeic acid (0.06 mg/g seed) in chia seeds, while quercetin (0.26 mg/g seed) and kaempferol (0.50 mg/g seed) were identified in the hydrolyzed extracts by high-performance liquid chromatography coupled to the diode array detector (HPLC-DAD).

Chlorogenic acid, caffeic acid, quercetin, myricetin and kaempferol have also been identified in fibrous flour and chia oil (Capitani et al., 2012; Ixtaina et al., 2012). Phenolic compounds, especially phenolic acids and flavonoids, have been widely studied due to their various health benefits as antioxidants, in the prevention of chronic inflammation, cardiovascular disease, cancer, and diabetes.

The high diversity of phenolic acids isolated from Salvia plants has shown excellent antimicrobial activity, as well as antioxidant activity, some of which have been used against numerous pathological disorders, such as atherosclerosis and brain dysfunction (Cvetkovikj et al., 2013; Martínez-Cruz & Paredes-López, 2014).

ANTIOXIDANT ACTIVITY

Oilseeds are sources of phenolic compounds with high antioxidant activity, which can sequester free radicals. Among the phenolic compounds are phenolic acids, flavonoids, quinones, coumarins, lignans, stilbenes, tannins, vitamins, terpenoids, and some other endogenous metabolites (Cai et al., 2004; Naczk & Shahidi, 2006).

Phenolic antioxidants interfere with the oxidative process as free radical deactivators and also as metal chelators. The antioxidant potential of phenolic compounds depends on the number and arrangement of hydroxyl groups in the molecule (Cao et al., 1997). Therefore, phenolics have been widely studied, as they have diverse bioactivities that are beneficial to human health, reducing the risk of cancer, heart disease, and diabetes; as well as exerting anti-bacterial, antiviral, anti-inflammatory and anti-allergic activities (Yao et al., 2004; Oak et al., 2005; Shahidi & Ambigaipalan, 2015).

According to Bianchi and Antunes (1999), antioxidants are free radical acceptors, intercepting these compounds generated by cellular metabolism or exogenous sources, avoiding the formation of lesions, the loss of cellular integrity; and reconstituting already damaged cell membranes.

The antioxidant activity in seeds can be determined by a variety of assays with different mechanisms, including hydrogen atom transfer (HAT), electron transfer (ET), metal reduction power, and metal chelation. The FRAP (Antioxidant Power of Iron Reduction) assay is a method that

measures the power of reducing metals, i.e., it measures the ability of antioxidants to reduce the ferric ion complex (Fe3+) to the ferrous complex (Fe2+) of intense blue color in an acidic medium (Shahidi & Zhong, 2015).

The ORAC (Oxygen Radical Reduction Capacity) assay is a method based on the transfer of hydrogen atoms, and measures the ability of an antioxidant against the peroxyl radical, where the antioxidant and a fluorescent marker (fluorescein) compete kinetically for peroxyl radicals, generated through the decomposition of nitrogenous compounds, such as AAPH (2,2'-azobis-(2-methylpropionamidine)-dihydrochlorinated) at 37 °C (Ou et al., 2013). ORAC is an important in vitro assay to evaluate antioxidant activity, as it uses a biologically relevant free radical (peroxyl radical), which is prevalent in human biology. This method considers both the time of inhibition and the degree of inhibition of the release of free radical action caused by antioxidants (Ou et al., 2013, Prior, 2015).

Jiménez et al. (2010) evaluated the antioxidant activity of the oil and the whole chia seed, and the oil showed low values of antioxidant activity compared to that of the whole seed. The antioxidant activity of chia seed was obtained in the range of 45.5 to 98.73 µmol TE/g while that of the oil was from 1.32 to 4.58 µmol TE/g, indicating that the antioxidant content in chia seed is hydrophilic in nature.

Marineli et al. (2014) determined the antioxidant activity of chia seeds, obtaining a value of 514.30 µmol TE/g for the ORAC assay, and a value of 405.71 µmol TE/g for the FRAP assay. Likewise, the antioxidant activity of fibrous flour was 577.2 µmol TE/g, which is higher than that found in wheat bran, sorghum and barley (48.5, 51.7 and 14.9 µmol TE/g), according to Ragaee et al. (2006).

Phenolic acids, the majority class of phenolic compounds in chia seeds, behave as antioxidants, due to the reactivity of their phenolic fraction, i.e. the presence of hydroxyl in the aromatic ring. Although there are several mechanisms, the predominant mode of phenolic acids' antioxidant activity is thought to be the deactivation of radicals via hydrogen atom donation (Shahidi & Ambigaipalan, 2015). Therefore, chia seeds are considered as functional ingredients with high antioxidant potential in food products with commercial applications.

ANTITUMOR ACTIVITY

Currently, cancer is among the leading causes of death in the world, and is defined as the set of diseases in which abnormal cells can divide and grow disorderly, being able to invade other tissues, and can spread throughout the body through the circulatory system establishing itself in other organs and tissues (NCI, 2017).

Studies report that the increase in the incidence of cancer may be related to changes in eating habits, increased life expectancy, increased pollutants and contact with carcinogenic compounds, whether due to these factors or others, this pathology is today one of the biggest public health problems (Monteiro et al., 2014; Siegel et al. 2016; Wang et al., 2016).

Investigators report that oxidative stress is one of the key components in linking environmental toxicity to the carcinogenic process, as reactive oxygen species (ROS) are generated in response to both endogenous and exogenous stimuli (Ziech et al., 2010; Fuchs-Tarlovsky et al., 2013). Evidence, both in vivo and in vitro studies, points to environmental agents, such as radiation, xenobiotics and chlorinated compounds, as significant inducers of cellular damage via ROS-mediated toxicity (Klaunig & Kamendilus, 2004; Garis et al., 2008; Fuchs-Tarlovsky et al., 2013). Studies have described the relationship between increased oxygen reactive cell radicals and the pathogenesis of several chronic diseases, including cancer (Shobha & Andallu, 2013; Prasad et al., 2016).

ROS are constantly generated within the body and are necessary to drive regulatory pathways, but they are also one of the causes of several pathological conditions, including cancer. Numerous lines of evidence suggest that ROS can promote as well as suppress cancer cell survival. ROS are known to regulate every step of tumor development, including transformation, survival, proliferation, invasion, metastasis, and angiogenesis (Prasad et al., 2016).

Studies report that ROS can mediate an indirect attack on DNA, mainly through reaction with other cellular components, such as phospholipids, resulting in the generation of reactive secondary intermediates and irreversible coupling to the DNA base, forming DNA adducts (Marnette, 2000; Fuchs-Tarlovsky et al., 2013). The formation of DNA adducts is central to the carcinogenesis process, because if such adducts escape cellular repair mechanisms and persist, they can induce errors, and ultimately, mutations (Wogan et al., 2004). Therefore, oxidative lesions have been implicated in the etiology of cancer, and lesions serve as a critical biomarker of oxidative DNA damage (Valko et al., 2004; Fuchs-Tarlovsky et al., 2013).

Evidence from experimental studies has shown that natural compounds, including phenolic compounds, act as positive anticancer regulators by adjusting the oxidative stress response, inhibiting cancer cell proliferation, and modulating autophagic signaling (Hasima & Ozpolat, 2014; Lang et al., 2015; Irimie et al., 2015; Guaman-Ortiz et al., 2017).

Studies of preclinical and clinical models have indicated that natural antioxidants promote reduced risk of cancer, and epidemiological data suggest that people who consume diets rich in natural antioxidants, which come from fruits and vegetables, have a lower risk of developing chronic disease and mortality than those who eat low amounts of fruits and vegetables (Shanmugam et al., 2016; Prasad et al., 2016; Prasad et al., 2017).

An in vivo study with a diet containing cereals and vegetables, such as "Wushen" (a mixture of foods containing 55 natural ingredients, including plants, meats, cereals and legumes), demonstrated high antitumor activity, reducing tumor growth by 48.52% in an experimental model with mice implanted subcutaneously with murine sarcoma S180 cells, and this antitumor potential of the "Wushen" diet is directly associated with its antioxidant properties (Wang et al., 2014).

Eating diets containing herbs, fruits, and vegetables significantly increases the antioxidant capacity of plasma, as the bioactive compounds present in vegetables are antioxidants, and prevent the formation of radicals, removing radicals before damage can occur, repairing oxidative damage, eliminating damaged molecules, and preventing mutations (Shobha & Andallu, 2013).

Phenolic compounds can limit the formation of initiated tumor cells by stimulating DNA repair (Yang et al., 2001). Quercetin, catechins, isoflavones, lignans, flavanones, ellagic acid and resveratrol, induce a reduction in tumor growth (Shobha & Andallu, 2013). Phenolic acids, such as gallic acid, chlorogenic acid, caffeic acid, have inhibitory properties against the invasive (adhesion, migration and angiogenesis) and metastatic behaviors of a variety of cancer cells in vitro and in vivo (Weng & Yen, 2012; Roleira et al., 2015).

Studies with polyunsaturated fatty acids (PUFA) present in vegetable oils and oilseeds have been investigated in tumor assays due to their anti-inflammatory and anticarcinogenic action. The high consumption of PUFA, specifically ω3-PUFA, can contribute to the reduction of chronic inflammatory processes involved in the development of many tumors (Vendramini-Costa & Carvalho, 2012). In addition, PUFA present in vegetable oils exert antitumor effects, perhaps affecting gene expression or activating signal transduction molecules involved in the control of cell growth, differentiation, apoptosis, angiogenesis, and metastasis (Espada et al., 2007).

Miranda-Vilela et al. (2014) evaluated the effects of supplementation with pequi oil (Caryocar brasiliense) on oxidative damage induced by doxorubicin in mice with solid Ehrlich tumor, with a protective effect on oxidative damage induced by doxorubicin and containment of tumor growth due to the high concentration of PUFA in pequi oil.

Among the models of in vivo studies to evaluate the anticancer activity, Ehrlich's tumor has been used because it is a practical and transposable murine tumor model for the analysis of the antiproliferative effects of several compounds, and this tumor can develop in the solid form, when inoculated subcutaneously, and in the ascitic form, when the cells are inoculated into the animal's peritoneum (Nascimento et al, 2006; Vendramini-Costa, 2010).

Espada et al. (2007) investigated the antitumor effects of diets with 6% chia oil and 6% safflower oil, consumed over a period of 45 days, in a model of murine mammary adenocarcionoma transplanted subcutaneously in Balb/C mice, obtaining a significant reduction of 29.68% in tumor mass and 88.89% in the number of metastases in the groups that consumed the diet with chia oil,

however, the diet with safflower oil did not significantly reduce tumor mass. The results indicate that chia oil, with a high content of ω3-PUFA, is a potential antitumor agent, and may have preventive action. Therefore, more studies should be carried out in order to prove its preventive potential and its mechanism of action.

REFERENCES

1. Acosta-Estrada, B. A., Gutiérrez-Uribe, J. A., & Serna-Saldívar, S. O. (2014). Bound phenolics in foods, a review. *Food Chemistry, 152*, 46–55.

2. Álvarez-Chávez, L. M., Valdivia-López, M. L. A., Aburto-Juárez, M. L., & Tecante, A. (2008). Chemical characterization of the lipid fraction of Mexican chia seed (*Salvia hispanica* L.). *International Journal of Food Properties, 11*(3), 687-697.

3. Anderson, J. W., Baird, P., Davis, R. H., Ferreri, S., Knudtson, M., & Koraym, A. (2009). Health benefits of dietary fiber. *Nutrition Reviews, 67*(4), 188-205.

4. Ayerza, R. (1995). Oil content and fatty acid composition of chia (*Salvia hispanica* L.) from five northwestern locations in Argentina. *Journal of the American Oil Chemists' Society, 72*, 1079–1081.

5. Ayerza, R. (2013). Seed composition of two chia (*Salvia hispanica* L.) genotypes which differ in seed color. *Emirates Journal of Food and Agriculture, 25*(7), 495-500.

6. Ayerza, R., & Coates, W. (2004). Protein and oil content, peroxide index and fatty acid composition of chia (*Salvia hispanica* L.) grown in six tropical and sub-tropical ecosystems of South America. *Tropical Science, 44*(3), 131–135.

7. Ayerza, R., & Coates, W. (2011). Protein content, oil content and fatty acid profiles as potential criteria to determine the origin of commercially grown chia (*Salvia hispanica* L.). *Industrial Crops and Products, 34*, 1366–1371.

8. Bianchi, M. L. P., & Antunes, L. M. G. (1999). Radicais livres e os principais antioxidantes da dieta. *Revista de Nutrição, 12*(2), 123-130.

9. Bodoira, R. M., Penci, M. C., Ribotta, P. D., & Martínez, M. L. (2017). Chia (*Salvia hispanica* L.) oil stability: Study of the effect of natural antioxidants. *Food Science and Technology, 75*, 107-113.

10. Busilacchi, H., Quiroga, M., Bueno, M., Di Sapio, O., Flores, V., & Severin, C. (2013). Evaluation of *Salvia hispanica* L. cultivated in the south of Santa Fe (Argentina). *Cultivos Tropicales, 34*(4), 55-59.

11. Cahill, J. P. (2003). Ethnobotany of chia, *Salvia hispanica* L. (Lamiaceae). *Economic Botany, 57*, 604–618.

12. Cai, Y., Luo, Q., Sun, M., & Corke, H. (2004). Antioxidant activity and phenolic compounds of 112 traditional Chinese medicinal plants associated with anticancer. *Life Sciences, 74*, 2157–2184.

13. Cao, G., Sofic, E., & Prior, R. L. (1997). Antioxidant and prooxidant behavior of flavonoids: Structure-activity relationships. *Free Radical Biology and Medicine, 22*, 749–760.

14. Capitani, M. I., Spotorno, V., Nolasco, S. M., & Tomás, M. C. (2012). Physicochemical and functional characterization of by-products from chia (*Salvia hispanica* L.) seeds of Argentina. *Food Science and Technology, 45*, 94-102.

15. Ciftci, O. N., Przybylski, R., & Rudzinska, M. (2012). Lipid components of flax, perilla, and chia seeds. *European Journal of Lipid Science and Technology, 114*, 794–800.

16. Clifford, M. N., & Scalbert, A. (2000). Ellagitannins-occurrence in food, bioavailability and cancer prevention. *Journal of the Science of Food and Agriculture, 80*, 1118–1125.

17. Coates, W., & Ayerza, R. (1996). Production potential of chia in northwestern Argentina. *Industrial Crops and Products, 5*, 229-233.

18. Codex Alimentarius. (2003). World Health Organization Codex Alimentarius Commission Joint WHO/FAO, Codex Stan 33 – Codex standard for olive oils and olive pomace oils. Joint FAO/WHO Food Standards Programme. Geneva: WHO/FAO (1981 (Rev. 2-2003), 1-9.

19. Coelho, M. S., & Salas-Mellado, M. M. (2014). Revisão: Composição química, propriedades funcionais e aplicações tecnológicas da semente de chia (*Salvia hispanica* L.) em alimentos. *Brazilian Journal of Food Technology, 17*(4), 259-268.

20. Coorey, R., Tjoe, A., & Jayasena, V. (2014). Gelling properties of chia seed and flour. *Journal of Food Science, 79*(5), 859-866.

21. Crozier, A., Jaganath, I. B., & Clifford, M. N. (2009). Dietary phenolics: Chemistry, bioavailability and effects on health. *Natural Product Reports, 26*, 1001.

22. Cvetkovikj, I., Stefkov, G., Acevska, J., Petreska Stanoeva, J., Karapandzova, M., Stefova, M., Dimitrovska, A., & Kulevanova, S. (2013). Polyphenolic characterization and chromatographic methods for fast assessment of culinary *Salvia* species from South East Europe. *Journal of Chromatography A, 1282*, 38–45.

23. D'Archivio, M., Filesi, C., Benedetto, R., Gargiulo, R., Giovannini, C., & Masella, R. (2007). Polyphenols, dietary sources and bioavailability. *Annali dell'Istituto Superiore di Sanità, 43*(4), 348-361.

24. Espada, C. E., Berra, M. A., Martinez, M. J., Eynard, A. R., & Pasqualini, M. E. (2007). Effect of chia oil (*Salvia hispanica*) rich in ω-3 fatty acids on the eicosanoid release, apoptosis and T-lymphocyte tumor infiltration in a murine mammary gland adenocarcinoma. *Prostaglandins, Leukotrienes and Essential Fatty Acids, 77*(1), 21-28.

25. Esposito, F., Arlotti, G., Bonifati, A. M., Napolitano, A., Vitale, D., & Fogliano, V. (2005). Antioxidant activity and dietary fibre in durum wheat bran by-products. *Food Research International, 38*, 1167-1173.

26. Fuchs-Tarlovsky, V. (2013). Role of antioxidants in cancer therapy. *Nutrition, 29*, 15–21.

27. Garis, D., Skiada, V., & A., B. (2008). Redox signaling and cancer: The role of "labile" iron. *Cancer Letters, 266*, 21–29.

28. Guamán-Ortiz, L. M., Orellana, M. I. R., & Ratovitski, E. A. (2017). Natural compounds as modulators of non-apoptotic cell death in cancer cells. *Current Genomics, 18*, 132-155.

29. Guinazi, M., Milagres, R. C. R. M., Pinheiro-Sant'Ana, H. M., & Chaves, J. B. P. (2009). Tocoferóis e tocotrienóis em óleos vegetais e ovos. *Química Nova, 32*(8), 2098-2103.

30. Hasima, N., & Ozpolat, B. (2014). Regulation of autophagy by polyphenolic compounds as a potential therapeutic strategy for cancer. *Cell Death & Disease, 5*, e1509.

31. Heleno, S. A., Martins, A., Queiroz, M. J. R. P., & Ferreira, I. C. F. R. (2015). Bioactivity of phenolic acids: Metabolites versus parent compounds, a review. *Food Chemistry, 173*, 501-513.

32. Herrera, M. C., & Luque de Castro, M. D. (2004). Ultrasound-assisted extraction for the analysis of phenolic compounds in strawberries. *Analytical and Bioanalytical Chemistry, 379*(7–8), 1106–1112.

33. International Agency for Research on Cancer (IARC). (2011). Disponível em: http://www.globocan.iarc.fr. [Acesso em: 05 de maio de 2017].

34. Irimie, A. I., Braicu, C., Zanoaga, O., Pileczki, V., Gherman, C., Berindan-Neagoe, I., & Campian, R. S. (2015). Epigallocatechin-3-gallate suppresses cell proliferation and promotes apoptosis and autophagy in oral cancer SSC-4 cells. *OncoTargets and Therapy, 8*, 461-470.

35. Ixtaina, V. Y., Julio, L. M., Wagner, J. R., Nolasco, S. M., & Tomás, M. C. (2015). Physicochemical characterization and stability of chia oil microencapsulated with sodium caseinate and lactose by spray-drying. *Powder Technology, 271*, 26–34.

36. Ixtaina, V. Y., Martínez, M. L., Spotorno, V., Mateo, C. M., Maestri, D. M., Diehl, B. W. K., Nolasco, S. M., & Tomás, M. C. (2011). Characterization of chia seed oils obtained by pressing and solvent extraction. *Journal of Food Composition and Analysis, 24*, 166–174.

37. Ixtaina, V. Y., Nolasco, S. M., & Tomás, M. C. (2008). Physical properties of chia (*Salvia hispanica* L.) seeds. *Industrial Crops and Products, 28*, 286–293.

38. Ixtaina, V. Y., Nolasco, S. M., & Tomás, M. C. (2012). Oxidative stability of chia (*Salvia hispanica* L.) seed oil: Effect of antioxidants and storage conditions. *Journal of the American Oil Chemists' Society, 89*(6), 1077–1090.

39. Ixtaina, V. Y., Vega, A., Nolasco, S. M., Tomás, M. C., Gimeno, M., Bárzana, E., & Tecante, A. (2010). Supercritical carbon dioxide extraction of oil from Mexican chia seed (*Salvia hispanica* L.): Characterization and process optimization. *Journal of Supercritical Fluids, 55*, 192–199.

40. Jiménez, F. E. G., Beltrán-Orozco, M. C., & Martínez, M. G. V. (2010). The antioxidant capacity and phenolic content of chia (*Salvia hispanica* L.) integral seed and oil. Special Abstracts / *Journal of Biotechnology, 150S*, S315.

41. Klaunig, J., & Kamendilus, L. M. (2004). The role of stress in carcinogenesis. *Annual Review of Pharmacology and Toxicology, 44*, 239–267.

42. Kozłowska, M., Gruczynska, E., Scibisz, I., & Rudzinska, M. (2016). Fatty acids and sterols composition, and antioxidant activity of oils extracted from plant seeds. *Food Chemistry, 213*, 450–456.

43. Lang, F., Qin, Z., Li, F., Zhang, H., Fang, Z., & Hao, E. (2015). Apoptotic cell death induced by resveratrol is partially mediated by the autophagy pathway in human ovarian cancer cells. *PLoS One, 10*(6), e0129196.

44. Lin, K.-Y., Daniel, J. R., & Whistler, R. L. (1994). Structure of chia seed polysaccharide exudate. *Carbohydrate Polymers, 23*(1), 13–18.

45. Marineli, R. S., Moraes, E. A., Lenquiste, S. A., Godoy, A. T., Eberlin, M. N., & Maróstica Jr., M. R. (2014). Chemical characterization and antioxidant potential of Chilean chia seeds and oil (*Salvia hispanica* L.). *Food Science and Technology, 59*, 1304-1310.

46. Marnette, L. J. (2000). Oxyradicals and DNA damage. *Carcinogenesis, 21*, 361–370.

47. Martínez-Cruz, O., & Paredes-López, O. (2014). Phytochemical profile and nutraceutical potential of chia seeds (*Salvia hispanica* L.) by ultra high performance liquid chromatography. *Journal of Chromatography A, 1346*, 43–48.

48. Martínez, M. L., Marín, M. A., Faller, C. M. S., Revol, J., Penci, M. C., & Ribotta, P. D. (2012). Chia (*Salvia hispanica* L.) oil extraction: Study of processing parameters. *Food Science and Technology, 47*, 78-82.

49. Miettinen, T. A., & Gylling, H. (1999). Regulation of cholesterol metabolism by dietary plant sterols. *Current Opinion in Lipidology, 10*, 9–14.

50. Miranda-Vilela, A. L., Grisolia, C. K., Longo, J. P. F., Peixoto, R. C. A., Almeida, M. C., Barbosa, L. C. P., Roll, M. M., Portilho, F. A., Estevanato, L. L. C., Bocca, A. L., Báo, S. N., & Lacava, Z. G. M. (2014). Oil rich in carotenoids instead of vitamins C and E as a better option to reduce doxorubicin-induced damage to normal cells of Ehrlich tumor-bearing mice: Hematological, toxicological and histopathological evaluations. *Journal of Nutritional Biochemistry, 25*, 1161–1176.

51. Mohd Ali, N., Yeap, S. K., Ho, W. Y., Beh, B. K., Tan, S. W., & Tan, S. G. (2012). The promising future of chia, *Salvia hispanica* L. *Journal of Biomedicine and Biotechnology, 2012*, 1-9.

52. Monteiro, L. S., Bastos, K. X., Barbosa-Filho, J. M., Athayde-Filho, P. F., Diniz, M. F. F. M., & Sobral, M. V. (2014). Medicinal plants and other living organisms with antitumor potential against lung cancer. *Evidence-Based Complementary and Alternative Medicine, ID 604152*, 15.

53. Morrison, I. M. (1980). Changes in the lignin and hemicellulose concentrations of ten varieties of temperate grasses with increasing maturity. *Grass and Forage Science, 35*, 287–293.

54. Muñoz, L. A., Cobos, A., Diaz, O., & Aguilera, J. M. (2012). Chia seeds: Microstructure, mucilage extraction and hydration. *Journal of Food Engineering, 108*(1), 216–224.

55. Naczk, M., & Shahidi, F. (2006). Phenolics in cereals, fruits and vegetables: Occurrence, extraction and analysis. *Journal of Pharmaceutical and Biomedical Analysis, 41*, 1523–1542.

56. Nardini, M., & Ghiselli, A. (2004). Determination of free and bound phenolic acids in beer. *Food Chemistry, 84*(1), 137–143.

57. Nascimento, F. R., Cruz, G. V., Pereira, P. V., Maciel, M. C., Silva, L. A., Azevedo, A. P. S., Barroqueiro, E. S. B., & Guerra, R. N. M. (2006). Ascitic and solid Ehrlich tumor inhibition by *Chenopodium ambrosioides* L. treatment. *Life Sciences, 78*(22), 2650-2653.

58. National Cancer Institute (NCI). (2017). Disponível em: https://www.cancer.gov/. [Acesso em: 05 de maio de 2017].

59. Oak, M. H., El Bedoui, J., & Schini-Kerth, V. B. (2005). Antiangiogenic properties of natural polyphenols from red wine and green tea. *Journal of Nutritional Biochemistry, 16*, 1–8.

60. Oliveira, D. M., & Bastos, D. H. M. (2011). Biodisponibilidade de ácidos fenólicos. *Química Nova, 34*(6), 1051-1056.

61. Ou, B., Chang, T., Huang, D., & Prior, R. L. (2013). Determination of total antioxidant capacity by oxygen radical absorbance capacity (ORAC) using fluorescein as the fluorescence probe: First action 2012.23. *Journal of AOAC International, 96*(6), 1372–1376.

62. Peiretti, P. G., & Gai, F. (2009). Fatty acid and nutritive quality of chia (*Salvia hispanica* L.) seeds and plant during growth. *Animal Feed Science and Technology, 148*, 267–275.

63. Pelletier, X., Belbraouet, S., Mirabel, D., Mordret, F., Perrin, J. L., Pages, X., & Debry, A. (1995). A diet moderately enriched in phytosterols lowers plasma cholesterol concentrations in normocholesterolemic humans. *Annals of Nutrition and Metabolism, 39*, 291–295.

64. Phillips, K. M., Ruggio, D. M., Toivo, J. I., Swank, M. A., & Simpkins, A. H. (2002). Free and esterified sterol composition of edible oils and fats. *Journal of Food Composition and Analysis, 15*, 123–142.

65. Poudyal, H., Panchal, S. K., Waanders, J., Ward, L., & Brown, L. (2012). Lipid redistribution by alpha-linolenic acid-rich chia seed inhibits stearoyl-CoA desaturase-1 and induces cardiac and hepatic protection in diet-induced obese rats. *Journal of Nutritional Biochemistry, 23*, 153–162.

66. Prasad, S., Gupta, S. C., Pandey, M. K., Tyagi, A. K., & Deb, L. (2016). Oxidative stress and cancer: Advances and challenges. *Oxidative Medicine and Cellular Longevity, 2016*, ID 5010423.

67. Prasad, S., Gupta, S. C., & Tyagi, A. K. (2017). Reactive oxygen species (ROS) and cancer: Role of antioxidative nutraceuticals. *Cancer Letters, 387*, 95–105.

68. Prior, R. L. (2015). Oxygen radical absorbance capacity (ORAC): New horizons in relating dietary antioxidants/bioactives and health benefits. *Journal of Functional Foods, 18*, 797–810.

69. Ragaee, S., Abdel-Aal, M. E.-S., & Noaman, M. (2006). Antioxidant activity and nutrient composition of selected cereals for food use. *Food Chemistry, 98*, 32–38.

70. Reyes-Caudillo, E., Tecante, A., & Valdivia-López, M. A. (2008). Dietary fibre content and antioxidant activity of phenolic compounds present in Mexican chia (*Salvia hispanica* L.) seeds. *Food Chemistry, 107*, 656–663.

71. Roleira, F. M. F., Tavares-da-Silva, E. J., Varela, C. L., Costa, S. C., Silva, T., Garrido, J., & Borges, F. (2015). Plant derived and dietary phenolic antioxidants: Anticancer properties. *Food Chemistry, 183*, 235–258.

72. Rupollo, G., Gutkoski, L. C., Martins, I. R., & Elias, M. C. (2006). Efeito da umidade e do período de armazenamento hermético na contaminação natural por fungos e a produção de micotoxinas em grãos de aveia. *Ciência e Agrotecnologia, 30*(1), 118-125.

73. Scalbert, A., & Williamson, G. (2000). Dietary intake and bioavailability of polyphenols. *Journal of Nutrition, 130*, 2073S–2085S.

74. Segura-Campos, M. R., Salazar-Vega, M. I., Chel-Guerrero, L. A., & Betancur-Ancona, D. A. (2013). Biological potential of chia (*Salvia hispanica* L.) protein hydrolysates and their incorporation into functional foods. *Food Science and Technology, 50*(2), 723-731.

75. Shahidi, F., & Ambigaipalan, P. (2015). Phenolics and polyphenolics in foods, beverages and spices: Antioxidant activity and health effects – A review. *Journal of Functional Foods, 18*(Part B), 820–897.

76. Shahidi, F., & Zhong, Y. (2015). Measurement of antioxidant activity. *Journal of Functional Foods, 18*, 757–781.

77. Shanmugam, M. K., Lee, J. H., Chai, E. Z. P., Kanchi, M. M., Kar, S., Arfuso, F., Dharmarajan, A., Kumar, A. P., Ramar, P. S., Looi, C. Y., Mustafa, M. R., Tergaonkar, V., Bishayee, A., Ahn, K. S., & Sethi, G. (2016). Cancer prevention and therapy through the modulation of transcription factors by bioactive natural compounds. *Seminars in Cancer Biology, 40-41*, 35–47.

78. Shobha, R. I., & Andallu, B. (2013). Oxidative stress and cancer: Role of anti-carcinogenic herbs and spices. *American Journal of Phytomedicine and Clinical Therapeutics, 3*, 351-369.

79. Siegel, R. L., Miller, K. D., & Jemal, A. (2016). Cancer statistics. *CA: A Cancer Journal for Clinicians, 66*(1), 7-30.

80. Tosco, G. (2004). Os benefícios da "Chia" em humanos e animais. *Atualidades Ornitológicas, 119*, 7.

81. Valko, M., Izakovic, M., Mazur, M., Christopher, J., Rhodes, C., & Telser, J. (2004). Role of oxygen radicals in DNA damage and cancer incidence. *Molecular and Cellular Biochemistry, 266*, 37–56.

82. Vendramini-Costa, D. B., & Carvalho, J. E. (2012). Molecular link mechanisms between inflammation and cancer. *Current Pharmacological Design, 18*, 3831-3852.

83. Vendramini-Costa, D. B., Castro, I. B. D., Ruiz, A. L. T. G., Marquissolo, C., Pilli, R. A., & Carvalho, J. E. (2010). Effect of goniothalamin on the development of Ehrlich solid tumor in mice. *Bioorganic & Medicinal Chemistry, 18*, 6742–6747.

84. Wang, C., Peng, R., Wang, L., Chen, P., Wang, S., Xu, X., Zhang, Q., Lin, S., & Hu, X. (2014). Wushen, a food mixture containing 55 different natural ingredients, inhibits S180 tumor growth in vivo. *Food Function, 5*, 1475–1480.

85. Wang, H., Yin, Y., Wang, P., Xiong, C., Huang, L., Li, S., Li, X., & Liu, F. (2016). Current situation and future usage of anticancer drug databases. *Apoptosis, 21*(7), 778-794.

86. Weickert, M. O., & Pfeiffer, F. H. A. (2008). Metabolic effects of dietary fiber consumption and prevention of diabetes. *Journal of Nutrition, 138*(3), 439–442.

87. Weng, C.-J., & Yen, G.-C. (2012). Chemopreventive effects of dietary phytochemicals against cancer invasion and metastasis: Phenolic acids, monophenol, polyphenol, and their derivatives. *Cancer Treatment Reviews, 38*, 76–87.

88. Willem van der Kamp, J., Jones, J., McCleary, B., & Topping, D. (2010). Dietary fibre: New frontiers for food and health. *Wageningen Academic Publishers*.

89. Wilson, J. R., Deinum, B., & Engels, F. M. (1991). Temperature effects on anatomy and digestibility of leaf and stem of tropical and temperate forage species. *Netherlands Journal of Agricultural Science, 39*, 31–48.

90. Wogan, G. N., Hecht, S. S., Felton, J. S., Conney, A. H., & Lu, Y. (2004). Environmental and chemical carcinogenesis. *Seminars in Cancer Biology, 14*, 437–486.

91. Yang, C. S., Landau, J. M., Huang, M. T., & Newmark, H. L. (2001). Inhibition of carcinogenesis by dietary polyphenolic compounds. *Annual Review of Nutrition, 21*, 381–406.

92. Yao, L. H., Jiang, Y. M., Shi, J., Tomas-Barberan, F. A., Datta, N., Singanusong, R., & Chen, S. S. (2004). Flavonoids in food and their health benefits. *Plant Foods for Human Nutrition, 59*, 113–122.

93. Ziech, D., Franco, R., Georgakilas, A. G., Georgakila, S., Malamou-Mitsi, V., Schoneveld, O., Pappa, A., & Panayiotidis, M. I. (2010). The role of reactive oxygen species and oxidative stress in environmental carcinogenesis and biomarker development. *Chemical Biology & Interactions, 188*, 334–339.