# Phenolic content and antioxidant activity of medicinal plants

Conteúdo fenólico e atividade antioxidante de plantas medicinais

Contenido fenólico y actividad antioxidante de las plantas medicinales

Received: 02/07/2023 | Revised: 02/27/2023 | Accepted: 02/28/2023 | Published: 03/05/2023

Vanessa Cristina Fernandes ORCID: https://orcid.org/0000-0003-0877-8741 Instituto Federal de Educação Ciência e Tecnologia do Triângulo Mineiro, Brasil E-mail: vanessacrisfernandes124@gmail.com **Carla Regina Amorim dos Anjos Oueiroz** ORCID: https://orcid.org/0000-0002-1031-0403 Instituto Federal de Educação Ciência e Tecnologia do Triângulo Mineiro, Brasil E-mail: carlaregina@iftm.edu.br **Eduardo Santos Almeida** ORCID: https://orcid.org/0000-0002-1778-4521 Instituto Federal de Educação Ciência e Tecnologia do Triângulo Mineiro, Brasil E-mail: eduardoalmeida@iftm.edu.br Cláudia MariaTomás Melo ORCID: https://orcid.org/0000-0002-3086-0613 Instituto Federal de Educação Ciência e Tecnologia do Triângulo Mineiro, Brasil E-mail: claudiamelo@iftm.edu.br

#### Abstract

Medicinal plants have played an essential role in the development of human culture. Today, the contribution of plants in industries such as fine chemicals, cosmetics, pharmaceuticals, drugs, and industrial raw materials is remarkable. Recent studies have shown that plants have constituents with functional properties that effectively treat and prevent chronic degenerative diseases. Based on the importance of medicinal plants, this study aimed to quantify the total phenolic content and antioxidant activity of 10 medicinal plants. The phenolic content was quantified using the Swain and Hillis method with a Folin-Ciocalteau reagent, while the antioxidant activity was determined using a 2,2-diphenyl-1-picrylhydrazyl (DPPH) reagent. The samples were dried in a circulating air incubator at 60 °C for 48 h before phenolic extraction. The compounds were extracted with 50% v/v ethanol, 50% v/v acetone, and distilled water. The results of this study showed that 80% of the plants analyzed had similar phenolic contents that were not statistically different, with an average content of 4.864 mg GAE g-1. The antioxidant activity ranged from 30% to 75%. The ability to scavenge free radicals was expressed as the percentage of inhibition of the oxidation of the radical. In addition, no significant differences were observed in the yield obtained from the extraction solvents, 50% v/v ethanol and 50% v/v acetone, used in this study.

Keywords: Chemical analysis; Solvent extraction; Total phenolic.

#### Resumo

As plantas medicinais têm desempenhado um papel essencial no desenvolvimento da cultura humana. Hoje, a contribuição das plantas é notável na indústria da química fina, cosméticos, produtos farmacêuticos, medicamentos e matérias-primas industriais. Estudos recentes mostraram que as plantas possuem constituintes com propriedades funcionais que são eficazes no tratamento e prevenção de doenças crônico-degenerativas. Com base na importância das plantas medicinais, este estudo teve como objetivo quantificar o teor de fenólicos totais e a atividade antioxidante de 10 plantas medicinais. O conteúdo fenólico foi quantificado pelo método de Swain e Hillis com reagente Folin-Ciocalteau, enquanto a atividade antioxidante foi determinada pelo reagente 2,2-difenil-1-picrilhidrazil (DPPH). As amostras foram secas em estufa com circulação de ar a 60 °C por 48 h, antes da extração dos fenólicos. Os compostos foram extraídos com etanol 50% v/v, acetona 50% v/v e água destilada. Os resultados deste estudo revelaram que 80% das plantas analisadas apresentaram teores semelhantes que fenólicos que não diferiram estatisticamente, com média de 4,864 mg EAG g<sup>-1</sup>. A atividade antioxidante variou entre 30 e 75%. A capacidade de sequestrar radicais livres foi expressa como a porcentagem de inibição da oxidação do radical. Além disso, não foram observadas diferenças significativas no rendimento obtido a partir dos solventes de extração, etanol 50% v/v e acetona 50% v/v, utilizadas neste estudo.

Palavras-chave: Análise química; Extração por solvente; Fenólicos totais.

#### Resumen

Las plantas medicinales han jugado un papel esencial en el desarrollo de la cultura humana. Hoy, el aporte de las platas es notable en la química fina, cosmética, farmacéutica, medicamentos y materias primas industriales. Estudios

recientes han demostrado que las plantas tienen constituyentes con propriedades funcionales que son eficaces en el tratamiento y prevención de enfermedades crónico-degenerativas. Basado en la importancia de las plantas medicinales, este estudio tuvo como objetivo cuantificar el contenido fenólico total y la actividad antioxidante de 10 plantas medicinales. El contenido fenólico se cuantificó por el método de Swain y Hillis con el reactivo de Folin-Ciocalteau, mientras que la actividad antioxidante se determinó con el reactivo 2,2-difenil-1-picrilhidrazilo (DPPH). Las muestras se secaron en una estufa con circulación de aire a 60°C durante 48 horas, antes de extraer los fenoles. Los compuestos se extrajeron con etanol al 50% v/v, acetona al 50% v/v y agua destilada. Los resultados de este estudio revelaron que el 80% de las plantas analizadas presentaron contenidos similares de fenoles que no difirieron estadísticamente, con un promedio de 4.864 mg EAG g<sup>-1</sup>, la actividad antioxidante varió entre 30 y 75%. La capacidad de capturar radicales libres se expresó como el porcentaje de inhibición de la oxidación de radicales. Además, no se observaron diferencias significativas en el rendimiento obtenido con los solventes de extracción, 50% v/v etanol y 50% v/v acetona, utilizados en este estudio.

Palabras clave: Análisis químico; Fenólicos totales; Extracción por solvente.

# **1. Introduction**

### 1.1 Healthy eating

Currently, the well-informed public has shown a tendency to consume products with functional properties. This trend may be attributable to studies showing the beneficial effects of these products on human health; these health-promoting effects of the products are because of their curative and preventive potential, which are due to the presence of antioxidant compounds. There is evidence that these compounds reduce the risk of developing some chronic degenerative diseases (Wicklund et al., 2005).

Foods and medicinal plants containing compounds with antioxidant activity are of interest since they can neutralize free radicals produced by the human body (Asif, 2015).

Some researchers have quantified the phenolic content and antioxidant activity of plant foods such as vegetables and fruits (Velioglu et al., 1998; Oliveira et al., 2009), spices (Morais et al., 2009a) and medicinal plants (Sousa et al., 2007; Dallaqua & Damasceno, 2011).

Clinical and epidemiological studies have shown that phenolic antioxidants in cereals, fruits, and vegetables are the main factors that have contributed to the low and significant decrease in the incidence of chronic degenerative diseases in populations that consume diets rich in these foods. Therefore, it is of great importance to study antioxidants derived from renewable and natural sources, mainly from well-known plants or plants used daily by the population (Shahidi, 1996; Jayaprakaskha & Jaganmohan Rao, 2000; Roesler et al., 2007).

In this context, the aim of this study was to quantify the phenolic content and determine the antioxidant activity of ten medicinal plants, using three different solvent extracts for each.

#### 1.2 Phenolic content and antioxidant activity

Free radicals are oxidative agents characterized as atomic or molecular species with one or more unpaired electrons in their outer orbital, making them highly reactive (Gillhan et al., 1997).

According to Melo et al. (2011) and Moraes et al. (2012), free radicals and other oxidants are responsible for aging and degenerative diseases such as cancer, cardiovascular diseases, cataracts, and impaired immune and cerebral function. Studies have shown that free radicals are controlled in living organisms by various antioxidant compounds, which may be endogenous to the body or obtained from dietary sources. Antioxidants can react with free radicals through various mechanisms, such as inhibiting the peroxides-forming chain reaction (Vázquez et al., 2007).

A diet rich in fruits, wine, chocolate, and vegetables may reduce oxidative stress in the body (Cerqueira et al., 2007; Pieniz et al., 2009). The intake of flavonoid-rich foods such as fruits, vegetables, tea, wine, and chocolate has been associated with a reduction in the risk of developing several chronic diseases, because the protective effect of these foods is due in part to their antioxidant properties, which reduce oxidative stress (O'Byrne, 2002; Halliwell et al., 2005; Cerqueira et al., 2007; Pieniz et al., 2009).

Therefore, the use of substances with antioxidant properties may be of great importance in treating and preventing diseases associated with an increase in oxidative stress.

According to Krinsky (1994), an antioxidant is a compound that protects the biological system from the harmful effects of processes and reactions that may cause excessive oxidation. Furthermore, according to Pietta (2000), antioxidants are substances that reduce the rate of oxidation through one or more mechanisms such as free radical inhibition and metal complexation.

In small amounts, oxidants are important for cell membrane renewal, inflammatory responses, and fighting microorganisms. However, excessive levels of oxidants may attack the cellular DNA, leading to mutations. They can also attack the lipid molecules that compose the cell membrane, thereby destroying its structure (Carper, 1995).

One of the main theories proposed to explain the curative and preventive potential of foods is based on the presence of antioxidant compounds. Recent studies correlate the consumption of fruits and vegetables with antioxidant properties with the evidence of reduced risk of developing some chronic degenerative diseases (Wicklund et al., 2005). Therefore, it is of great importance to quantify antioxidant compounds in different food sources that may be consumed in the daily diet.

Natural antioxidants are widely distributed in foods and medicinal plants. These natural antioxidants, especially polyphenols, and carotenoids, exhibit a wide range of biological effects, including anti-inflammatory, anti-aging, anti-atherosclerosis, and anticancer. Effective extraction and proper evaluation of antioxidants from food and medicinal plants are crucial to explore the potential antioxidant sources and promoting their application in functional foods, pharmaceuticals, and food additives (Xu et al., 2017).

Free radicals are oxidative agents characterized as atomic or molecular species with one or more unpaired electrons in their outer orbital, making them highly reactive (Gillhan et al., 1997). They and other oxidants are responsible for aging and degenerative diseases such as cancer, cardiovascular disease, cataracts, and impaired immunological and cerebral function (Melo et al., 2011; Moraes et al., 2012). In small amounts, oxidants are important for cell membrane renewal, inflammatory responses, and fighting microorganisms. However, excessive levels of oxidants can attack cellular DNA, leading to mutations. They can also attack the lipid molecules that make up the cell membrane, thus destroying its structure (Carper, 1995).

Antioxidants are compounds that protect the biological system from the harmful effects of processes and reactions that can cause excessive oxidation (Krinsky, 1994). They can react with free radicals by reducing the rate of oxidation through one or more mechanisms, such as inhibition of the chain reaction that forms peroxides, inhibition of free radicals, and complexation of metals (Pietta, 2000; Vázquez et al., 2007).

Studies have shown that free radicals in living organisms are controlled by various antioxidant compounds, which may be endogenous to the body or obtained from food sources (Vázquez et al., 2007). The presence of antioxidant compounds in flavonoid-rich foods, such as fruits, vegetables, tea, wine, and chocolate is one of the main theories explaining the curative and preventive potential of foods in reducing the development of various chronic degenerative diseases. The protective effect of these foods is partly due to their antioxidant properties, which reduce oxidative stress (O'Byrne, 2002; Halliwell et al., 2005; Wicklund et al., 2005; Cerqueira et al., 2007; Pieniz et al. 2009).

Therefore, the use of substances with antioxidant properties may be of great importance for the treatment and prevention of diseases associated with increased oxidative stress. Thus, it is of great interest to quantify antioxidant compounds in different food sources that can be consumed in the daily diet.

#### **1.3 Medicinal plants**

The search for and the use of plants with therapeutic properties has been handed down from generation to generation preserving millennia-old traditions, as evidenced by several phytotherapy treatments (Correa Jr., 1991)

At present, from the poorest regions of the country to the largest Brazilian cities, medicinal plants are widely used to treat and cure diseases, sold on the streets and in popular markets, and found in residential backyards. Moreover, in rural areas, they are often the only therapeutic resource for many people (Maciel et al., 2002).

The main factors influencing the use of medicinal plants are the low standard of living of the population and the high cost of medicines. Therefore, plant users around the world continue to consume phytotherapeutics, validating some of the therapeutic information accumulated over the centuries (Newall et al., 2002).

In 2005, on the recommendation of the SUS (*Sistema Único de Saúde* – Brazilian National Health System), Brazil proposed the inclusion of medicinal plants and phytotherapy as treatment options in the National Health System, provided that these products based on medicinal plants comply with the standards of the current legislation (Brasil, 2006).

Hoeffel et al. (2011) conducted interviews with people knowledgeable about medicinal plants and were able to list 186 species that were mentioned by the interviewees including saffron (*Curcuma longa*), blackjack (*Bidens pilosa* L.), ginger (*Zingiber officinale*), fennel (*Foeniculum vulgare Mill*), elderberry (*Sambucus nigra*), wormseed (*Dysphania ambrosioides*), and numerous others.

The use of medicinal plants in the treatment and prevention of diseases is called phytotherapy, which is characterized by treatment with crude plants as opposed to isolated active ingredients. These raw plant materials help the body normalize disturbed physiological functions, restore immunity, promote detoxification, and gradually rejuvenate (Schenkel et al., 2000; Franca et al., 2008; Firmo et al., 2011). Gurib-Fakim (2006) reported that 25% of medical prescriptions consist of formulations that are based on substances derived from plants or synthetic analogs.

In the scientific community, there is little information on the reliability and safety of most medicinal plants. However, there has been an increase in ethnopharmacological research and the use of modern pharmacological, biochemical, toxicological, and molecular biological techniques to evaluate and validate medicinal plants and to advocate their use, which has effectively shortned the time needed to develop new drugs (Firmo et al., 2011).

Sousa et al. (2007) studied the antioxidant activity and phenolic compounds in five medicinal plants and found that all the extracts analyzed had a high phenolic content (11.55 to 97.6 mg of gallic acid equivalents [GAE]  $g^{-1}$  of dry material). Velioglu et al. (1998) analyzed sunflower seeds, flaxseed, wheat germ, several fruits, vegetables, and medicinal plants and obtained a total phenolic content ranging from 169 to 10548 mg 100  $g^{-1}$  (equivalent to 1.69 to 105.48 mg  $g^{-1}$ ).

Calixto (2000) reported that domestication, biotechnological study, and genetic improvement of medicinal plants can be advantageous to obtain plants with better uniformity and quality.

# 2. Methodology

### 2.1 Sample collection and raw material preparation

Twenty grams of each one of the 10 medicinal plants were collected from street markets and backyards in the city of Araguari, MG, in three repeated collections and were stored at the Laboratory for Physicochemical Analysis of the Federal Institute of the Triângulo Mineiro. For the extraction of phenolic compounds, three different solvents were used namely distilled water, 50% v/v acetone, and 50% v/v ethanol. After the extraction, both phenolic content and antioxidant activity were quantified in the samples as shown in the flow chart in Figure 1.



Figure 1 - Flowchart of extraction and determination of total phenolics and antioxidant activity in medicinal plants.



The samples were dried in a circulating air incubator at 60 °C for 48 h prior to phenolic extraction as shown in Figure

#### 2.2 Obtaining the extract

1.

The plant material was extracted with 50% v/v acetone-distilled water and 50% v/v ethanol-distilled water. Beakers containing 0.5 g of the previously dried and macerated plant material were filled with 30 mL of the solvent. During the extraction process, the samples were stirred regularly and extracted for 24 h according to the method of Bertoldi (2006) with modifications.

#### 2.3 Spectrophotometric determination of total phenolic compounds

For the determination of the total phenolics, the procedure proposed by Swain and Hillis (1956) was adopted with modifications.

After filtering the extracts obtained in section 4.2, 0.5 mL of the supernatant from each was pipetted into Falcon tubes, and then 2.5 mL of 10% v/v Folin-Ciocalteau and 2.0 mL of freshly prepared 7.5% w/v sodium carbonate were added to each tube. The sample mixtures were held in a water bath at a temperature of 50 °C for 5 min and then allowed to stand until they reached a temperature of approximately 25 °C.

The absorbance was then measured at 760 nm using a spectrophotometer against a blank containing the solvent and the same proportions of the reagents (Folin-Ciocalteau and sodium carbonate). The phenolic content of each extract was quantified using a prepared standard curve. It was used 0.5 mL of a stock solution of gallic acid at concentrations of 0, 10, 20, 30, 40, and 50 mg L<sup>-1</sup> and expressed as gallic acid equivalents (GAE). For analysis, 2.5 and 2.0 mL of Folin-Ciocauteau and sodium carbonate, respectively, were added to the sample to give a total volume of 5.0 mL, corresponding to each test sample volume. Along with the samples, a blank was also measured by replacing the sample with 0.5 mL of the appropriate solvent (distilled water, ethanol, or acetone at 50% v/v) and adding the same volume and concentration of the reagents used to construct the standard curve.

# 2.4 Antioxidant activity -Free radical scavenging capacity

Antioxidant activity was determined by a spectrophotometric method using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) reagent. This method is based on the percentage of the decrease in absorbance induced by the extract, and the samples are read with a spectrophotometer at 517 nm (Brand-Williams et al., 1995; Meda et al., 2005; Wu et al., 2005). In this reaction, the DPPH species are reduced by the antioxidant components present in the organic compounds being analyzed (Morais et al.,

2009b). The ability to scavenge free radicals was expressed as the percentage of inhibition of the oxidation of the radical, as expressed in Equation 1.

#### % Inhibition = 100 x (Aa-[Ab-Ac]) / Aa Equation (1)

Where, **Aa** is the absorbance of the DPPH incubation solution without sample addition (2 mL of DPPH 0.1 mM + 3 mL of methanol), **Ab** is the absorbance of the incubation mixture consisting of DPPH 0.1 mM, and the sample (0.5 mL of the sample plus 2 mL of DPPH 0.1 mM plus 2.5 mL of methanol), and **Ac** is the absorbance of the blank solution without DPPH 0.1 mM (0.5 mL of the sample plus 4.5 mL of methanol).

Aliquots of the extract were used to assess the scavenging ability of the free radical DPPH. Briefly, 2 mL of the free radical DPPH 0.1 mM methanolic solution and 2.5 mL of methanol were added to each 0.5 mL sample. After incubation for 30 min at room temperature in the dark, the reduction of DPPH 0,1 mM free radical was measured by reading the absorbance at 517 nm.

# 3. Results and Discussion

Table 1 shows the mean total phenolic content and antioxidant activity of the 10 studied medicinal plants expressed as mg GAE g<sup>-1</sup>. The results are presented as mean  $\pm$  standard deviation (SD), and the data were subjected to analysis of variance and comparison of means using Tukey's test. The level of statistical significance was set at a 5% probability level.

Table 1 - Content of phenolic compounds as milligrams of gallic acid equivalents per gram (mg GAE  $g^{-1}$ ) of medicinal plant samples.

Plants		Phenolic Content	Antioxidants
Scientific Name	Brazilian Popular Name	(mg EAG g <sup>-1</sup> )	(%)
Mentha spicata	Mint	$6.05 \pm 1.30$ a	68.37 ± 13.36 abc
Lupinus luteus L	Alfavaca	$5.28 \pm 1.16 \text{ ab}$	$50.40 \pm 17.69$ abcd
Rosmarinus officinalis	Rosemary	4.86.±.1.24 ab	$75.89 \pm 22.43a$
Arnica chamissonis	Arnica	$4.76 \pm 0.80 \text{ ab}$	$52.06 \pm 16,38$ abcd
Matricaria chamomilla L	Chamomile	$4.62 \pm 1.36 \text{ ab}$	67.27 ± 13.31abc
Rubus rosifolius,	Blackberry leaf	4.53 ± 1.75 ab	71.56 ± 18.58 ab
Melissa officinalis	Lemon balm feaf	$4.40 \pm 1.33$ ab	$40.06\pm13.36~\text{cd}$
Vernonia condensata Backer	Boldo	$4.40 \pm 1.07 \text{ ab}$	$46.53 \pm 27.65 abcd$
Zingiber officinale	Ginger	$3.71\pm1.16~\text{b}$	$42.70 \pm 25.58$ bcd
Alternanthera brasiliana	Terramycin	$3.67\pm0.60~b$	$30.88 \pm 18.77 d$

\*\*Significant at 1% probability level (p<0.01). Means followed by the same letter are not different by Tukey test, p<0.05. Data are presented as mean of 3 replicates. Source: Authors.

From the experimental results (Table 1), it can be observed that the phenolic content of *Mentha spicata* plant was significantly different from that of the terramycin (*Alternanthera brasiliana*) and ginger (*Zingiber officinale*), but similar to that of the other plants analyzed. Pastor-Cavada et al. (2009) found concentrations of total phenolics ranging from 8.7 to 11 mg  $g^{-1}$  in *Lupinus luteus* seeds collected in southern Spain, a concentration higher than that obtained in the leaf of this plant (Table 1).

Ribeiro et al. (2019) analyzed the total phenolic compounds in two varieties of peppers, obtained with seven different extraction solvents, and found that, for dried peppers, the result ranged from 1.92 to 5.44 mg EAG g<sup>-1</sup>, from 1.04 to 1.76 mg

EAG  $g^{-1}$  for fresh peppers. To four dried peppers the result of total phenolics is reasonably close to the results obtained in this research for medicinal plants.

Port (2011) obtained average results of total phenolic compounds much higher than the one obtained in this research for the plants he evaluated, and the values obtained ranged from  $27.04 \pm 3.03$  to  $721.08 \pm 108.51$  (mg EAG g<sup>-1</sup>) for *C. citratus* and *C. railway*, respectively. The highest phenolic values obtained can be related to the type of plant, the degree of grinding, the solvent, and the form of extraction among other peculiarities of the chemical analysis.

Regarding the solvents, no statistical difference in extraction efficiency was observed between the solvents used for the extraction of phenolic compounds (Table 2). This shows that it is possible to extract phenolics from dried plants using water with the same efficiency as other more expensive or toxic solvents.

 Table 2 - Total phenolic content as milligrams of gallic acid equivalents per gram (mg GAE/g) obtained from three different solvent extracts.

Solvents	Phenolic Content <sup>ns</sup> (mEAG g <sup>-1</sup> )	Antioxidants (%)*
Water	4.63 ± 1.73 a	$46.53 \pm 21.74 \text{ b}$
Alcohol 50% v/v	$4.40 \pm 1.16 \text{ a}$	$55.21\pm24.08~ab$
Acetone 50% v/v	$4.86 \pm 1.15$ a	$61.97 \pm 22.45$ a
Average	4,63 ± 1,35	

<sup>ns</sup> not significant (p>0.05). \* Significant at 5% probability (p <0.05). Source: Authors.

Bertoldi (2006) observed that pure solvents such as water or alcohol have lower extraction efficiency than mixed solvents do, which is not in agreement with the results obtained in this study. In addition, the same author previously reported that 70% and 90% v/v acetone have the highest mean extraction rates for phenolic compounds compared to pure water and acetone.

Babbar et al. (2014) confirmed through experimental results that methanol and ethyl acetate were better than the other two solvents (chloroform and n-hexane extract) for the extraction of phenolic compounds. This could be due to their higher polarity and better solubility for phenolic components present in plant materials (tomato peel, pea pod, cauliflower waste, and potato peel).

According to Beal (2006), the total phenolic content analyzed showed significant differences, ranging from 525.42 - 476.72 mg 100 g<sup>-1</sup> GAE in the ethereal extracts of ginger. These extracts also had a good amount of phenolics (365.07 - 279.95 mg 100 g<sup>-1</sup> GAE in water), which is particularly important since ginger is traditionally prepared for consumption in teas or even soups with the inclusion of water in the process. However, the extraction efficiency of acetone was not as evident as in the other extracts, presenting an amount of phenolics that varied between 176.33 - 130.99 mg 100 g<sup>-1</sup> GAE. The results obtained by the referred author are close to those obtained in this research, for the same plant, showing that one can have a more efficient extraction depending on the solvent used.

The antioxidant activity of rosemary (*Rosmarinus officinalis*) was 75.89%, the highest among all the plants analyzed in this study (Table 03), followed by *Rubus rosifolius, Mentha spicata, Matricaria chamomilla L, Arnica chamissonis, Lupinus luteus L* and *Vernonia condensata Backer*, all with statistically similar antioxidant activities, ranging from 71.56% to 46.53%. *Zingiber officinale* presented 42.70% of antioxidant activity, close to that of *Melissa officinalis* with 40.06%. *Alternanthera brasiliana* was the plant that presented the lowest antioxidant activity with 30.88%, although this plant is rich in natural

antioxidants, which justifies its use in popular medicine, especially in the treatment of free radical-mediated disorders (Enechi et al., 2013).

According to Beal (2006), when studying the antioxidant activity and identification of phenolics in *Zingiber officinale*, the ginger samples whose state of maturity was more advanced showed a higher content of total phenolics and also greater antioxidant activity, which can happen with other medicinal plants.

A linear correlation between total phenolic content and antioxidant activity was not observed in the present study as shown in Figure 2.



Figure 2 - Correlation between total phenolic content and total antioxidant capacity of medicinal plants.

#### Source: Authors.

Spiridon et al., (2011) found a different result from this research, as there was a linear correlation between total phenolics and antioxidants in the study of herbal medicinal plants.

Port (2011) agrees with Sharma and Bhat (2009) when she states that it is difficult to compare the antioxidant potential of different extracts due to the different ways in which the results are presented in the literature. The DPPH• index (I%) only shows the capacity of the extract at a fixed concentration used to reduce or not reduce the DPPH• radicals, where in many cases, increasing the concentration of the extract, the percentage of the DPPH index is increased. As for the inhibitory concentration test (IC50), it shows the extract concentration necessary to reduce the initial concentration of DPPH• by 50%, however, using different concentrations of DPPH•, as is the case in some researchers found in the literature, the results are different for the same sample. Thus, the antioxidant activity index is related to the concentration of DPPH• used in IC50 assay of the sample, resulting in constant data for each plant compound or extract, since extraction procedures or different sampling locations may give different results (Scherer; Godoy, 2009). The IC50 and the percentage of inhibition of radical oxidation (Eq 01) are two ways of expressing how free radicals are inhibited by the presence of a phenolic and, consequently, how to know which plants or foods provide substances to inhibit free radicals present in the body in greater or lesser quantities.

Souza et al. (2007) show that the results of the quantitative evaluation of the antioxidant activity of the ethanolic extract of *T. brasiliensis* (leaf and bark), *T. fagifolia* (leaf), *C. macrophyllum* (leaf), *Q. grandiflora* (leaf) and *C. prunifera* (root) and positive controls, at concentrations of 100, 50 and 25  $\mu$ g mL<sup>-1</sup>, determined by the DPPH assay, that all species have DPPH radical scavenging activity, however the extract of *C. prunifera* was the less active in the three concentrations. The other samples of extracts, at a concentration of 100  $\mu$ g mL<sup>-1</sup>, showed antioxidant activity above 60%, reaching a maximum of 91.36 ± 0.55% for the

extract of *T. brasiliensis* bark against 94.84  $\pm$  0, 44% of gallic acid and 89.25  $\pm$  0.25% of rutin. These results show that the concentration of DPPH can affect the sequestration activity, as well as the reference phenolic compounds such as gallic acid, rutin, among others, making it very difficult to compare information regarding the quantification of phenolics and antioxidant activity.

Ribeiro et al. (2019) obtained a higher average antioxidant activity (75.66%) in two pepper varieties when the extract was obtained with distilled water and 50% v/v methanol, both of which had the same average amount of total phenolics before to analysis. In this research the antioxidant activity of the extract with 50% v/v acetone was higher than that of water, both being statistically similar to 50% v/v ethanol (Table 2).

The extract of distilled water had the lowest extraction for antioxidant activity, with a result significantly different from that of acetone and statistically similar to the alcohol extract at 50% v/v. Thus, when phenolic content and antioxidant activity are determined simultaneously, the best solvent to use is acetone 50% v/v.

Fatima et al. (2017) studied the antioxidant activity in the leaves and fruits of five medicinal plants, and found that the percentage of free radical inhibition ranged from 37.66% to 77.33%. The results showed that all the medicinal plant species studied exhibited remarkable antioxidant activity in different concentrations when their different parts were tested in ethanolic extracts, so all these species could be used as a valuable drug against various antioxidants produced in the body. The antioxidant activity of the fruit of *Solanum nigrum* (37.66%) presented the lowest percentage of inhibition and the highest inhibition was presented by the fruit of *Datura inoxia* (77.33%), using the same method used in this research.

According to Beal (2006), the ethereal ginger extracts showed the highest antioxidant capacities (317.30 - 251.29  $\mu$ M g<sup>-1</sup> TEAC) when compared to the others, followed by the aqueous extracts (161.44 - 115.30  $\mu$ M g<sup>-1</sup> TEAC). The alcoholic and acetonic extracts showed much lower antioxidant capacity than the others (78.94 - 34.87  $\mu$ M / g TEAC and 60.77 - 32.34  $\mu$ M g<sup>-1</sup> TEAC, respectively).

With the experimental results, it can be said that the analyzed medicinal plants are sources of phenolic compounds that have antioxidant activity and that the greater or lesser amount of these components in the plant may be related to the degree of maturation, extraction method, degree of sample crushing, analysis method, among other experimental variables, as verified by Beal (2006).

# 4. Conclusion

Eighty percent or more of the evaluated medicinal plants showed the same content of phenolic compounds, which were not statistically different and ranged from 4.40 to 6.05 mg EAG  $g^{-1}$ .

The extraction of total phenolics with distilled water, acetone 50% v/v, and alcohol 50% v/v did not differ statistically, showing that it is possible to use the inexpensive solvent, distilled water, to extract these compounds from dried plants.

The antioxidant activity ranged from 30.88% to 75.89% in the plants studied, suggesting that this property is not only dependent on the total phenolic content of the plants, but also on the solvent used in the extraction, among other variables.

In future research, it is expected the addition of antioxidant compounds as food preservatives and/or as a source of substances with functional properties.

### Acknowledgments

The authors thank IFTM-Campus Uberlândia for providing the laboratory and the chemical reagents for the development of this research.

# References

Asif, M. (2015) Chemistry and antioxidant activity of plants containing some phenolic compounds. Chemistry International, 1(1): 35-52.

Babbar, N., Oberoi, H. S., Sandhu, S. K. & Bhargav, V. K. (2014). Influence of different solvents in extraction of phenolic compounds from vegetable residues and their evaluation as natural sources of antioxidants. *J Food Sci Technol.* 51(10):2568–2575. 10.1007/s13197-012-0754-4

Beal, B. H. (2006). Atividade antioxidante e identificação dos ácidos fenólicos do gengibre (Zingiber officinale Roscoe). Dissertação (Mestrado em Ciência dos Alimentos) – Centro de Ciências Agrárias, Universidade Federal de Santa Catarina, 87p.

Bertoldi, M. C. (2006). Atividade antioxidante in vitro da fração fenólica, das oleorresinas e do óleo essencial de pimenta rosa (Schinus terebinthifolius Raddi). Dissertação (Mestrado em Ciência e Tecnologia de Alimentos) - Universidade Federal de Viçosa - UFV, Viçosa, MG, 96 p.

Brand-Williams, W., Cuvelier, M. E. & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. Lebensmittel Wissenscharft and Technologies. 28: 25-30.

Brasil. (2006). *Ministério da Saúde. Política Nacional de Plantas Medicinais e Fitoterápicos.* Brasília. DF. http://bvsms.saude.gov.br/bvs/publicacoes/politica\_nacional\_fitoterapicos.pdf

Calixto, J. B. (2000). Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (phytotherapeutic agents). Braz. J. Med. Biol. Res. 33: 179-189.

Carper, J. (1995). Alimentos: o melhor remédio para a boa saúde. Ed. Campus. 632 p.

Cerqueira, F. M., Medeiros, M. H. G. & Augusto, O. (2007). Antioxidantes dietéticos: controvérsias e perspectivas. Quim. Nova. 30: 441-449.

Correa Júnior, C., Ming, L. C. & Scheffer, M.C. (1991). Cultivo de plantas medicinais, condimentares e aromáticas. EMATER-PR, 151 p.

Dallaqua, B. & Damasceno, D. C. (2011). Comprovação do efeito antioxidante de plantas medicinais utilizadas no tratamento do Diabetes mellitus em animais: artigo de atualização. *Rev. Bras. Pl. Med.* 13: 367-373.

Enechi, O. C., Odo, C. E. & Wuave, C. P. (2013). Evaluation of the in vitro anti-oxidant activity of alternanthera brasiliana leaves. Journal of Pharmacy Research. 6(9): 919-924.

Fatima, I., Hussain, T., Rafay, M., Akram, M., Bano, S., & Shabbir, S. (2017). Evaluation of antioxidant activity of leaves and fruits extracts of five medicinal plants. *Pak J Pharm Sci.* 30:1625–1628.

Firmo, W. C. A., Menezes, V. J. M., Passos, C. E. C., Dias, C. N., Alves, L. P. L., Dias, I. C. L., Neto, M. S. & Olea, R. S. G. (2011). Contexto histórico, uso popular e concepção científica sobre plantas medicinais. *Cad. Pesq.*, 18.

França, I. S. X., Souza, J.A., Baptista, R. S. & Britto, V. R. S. (2008). Medicina popular: benefícios e malefícios das plantas medicinais. *Rev. Bras. de Enferm.* 61: 201-208.

Gillhan, B., Papachristodoulou, D. K. & Thomas, J. H. (1997). Will's biochemical basis of medicine. (3a ed.), Butterworth Neinemann. London, 1997.

Gurib-Fakim, A. (2006). Medicinal plants: traditions of yesterday. Molecular Aspect of Medicine. 27: 1-936.

Hoeffel, J. L. M., Gonçalves, N. M., Fadini, A. A. B. & Seixas, S. R. C. (2011). Conhecimento tradicional e uso de plantas medicinais nas apas's cantareira/Sp e Fernão DiaS/MG. *Revista Vitas*. 11.

Halliwell, B., Rafter, J. & Jenner, A. (2005). Health promotion by flavonoids, tocopherols, tocotrienols, and other phenols: direct or indirect effects? Antioxidant or not? Am J Clin Nutr. 81:268S-276S.

Jayaprakasha, F. K. & Rao, L. J. (2000). Phenolic constituents from lichen *Parmotrema stuppeum* (Nyl.). Hale and their antioxidant activity. Z. *Naturforsch*. 55:1018-1022.

Krinsky, N. I. (1994). The biological properties of carotenoids. Pure Appl. Chem. 66:1003-1010.

Maciel, M. A. M., Pinto, A. C., Veiga, J. R. V. F., Grynberg, N. F. & Echevarria, A. (2002). Plantas medicinais: a necessidade de estudos multidisciplinares. *Quim. Nova.* 25: 429-438.

Meda, A., Lamien, C. E., Romito, M., Millogo, J. & Nacoulma, O. G. (2002). Determination of the total phenolic, flavonoid and proline contents in Burkina Fasan honey, as well as their radical scavenging activity. *Food Chemistry*. 91:571-577.

Melo, C. M. T., Costa, L. A., Bonnas, D. S., Chang, R. (2011). Compostos fenólicos e capacidade antioxidante de pimentas Capsicum chinense (bode), Capsicum baccatum variedade praetermissum (cumari) e Capsicum frutescens (malagueta). *Enciclopédia Biosfera*. 7:1-6.

Morais, S. M de., Cavalcanti, E. S. B., Costa, S. M. O. & Aguiar, L.A. (2009a). Ação antioxidante de chás e condimentos de grande consumo no Brasil. *Brazilian Journal of Pharmacognosy.* 19: 315-320.

Morais, S. A. L., Aquino, F. J. T., Nascimento, P. M., Nascimento, E. A. & Chang, R. (2009b). Compostos bioativos e atividade antioxidante do Café Conilon submetido a diferentes graus de torra. *Quim. Nova.* 32:327-331.

Moraes, L. P. de., Paz, M. F da., Argandoña, E. J. S., Silva, L. R. da. & Zago, T de. O. (2012). Phenolic Compounds and Antioxidant Activity of Fermented "Dedo-de moça" Pepper Sauce. Compostos Fenólicos e Atividade Antioxidante de Molho de Pimenta "Dedo-de-Moça" Fermentado. *Biochemistry and Biotechnology Reports.* 1:33-38, 2012. Newall, C. A., Anderson, L. A. & Phillipson, J. D. (2002). Plantas Medicinas: Guia para profissional de saúde. Ed. Premier, 300.

O'Byrne, D. J., Devaraj, S., Grundy, S. M. & Jialal, I. (2002). Comparison of the antioxidant effects of Concord grape juice flavonoids  $\alpha$ -tocopherol on markers of oxidative stress in healthy adults. *American Journal of Clinical Nutrition*. 76:1367-1374.

Oliveira, A. C. de., Valentim, I. B., Goulart, M. O. F., Silva, C. A., Bechara, E. J. H. & Trevisan, M. T. S. (2009). Fontes Vegetais Naturais de Antioxidantes. *Química Nova*. 32:689-702.

Pastor-Cavada, E., Juan, R., Pastor, J. E., Alaiz, M., Giróncalle, J. & Vioque, J. (2011). Antioxidative activity in the seeds of 28 Vicia species from southern Spain. J. Food Biochem. 35:1373-1380.

Pieniz, S., Colpo, E., Oliveira, V. R. de., Estefanel, V. & Andreazza, R. (2009). Avaliação in vitro do potencial antioxidante de frutas e hortaliças. R. Ciênc. Agrotec. 33:552-559.

Pietta, P. (2000). Flavonoids as antioxidants. J Nat. Prod. 63:1035-1042.

Roesler, R., Malta, L. G., Carrasco, L. C., Holanda, R. B., Souza, C. A. S. & Pastore, G. M. (2007). Atividade Antioxidante de frutas do cerrado. Ciênc. Tecnol. Aliment. 27:53-60, 2007.

Port's, P. S. da. (2011). Compostos fenólicos e potencial antioxidante de ervas consumidas na região amazônica brasileira. Dissertação (mestrado) – Universidade Estadual de Campinas. Faculdade de Engenharia de Alimentos. Campinas, SP.

Ribeiro, M. S. da., Melo, C. M. T., Almeida, E. S. & Queiroz, C. R. A. A dos. (2019). Extraction of total phenolic content and antioxidant activity of *Capsicum chinese* and *Capsicum spp* in different solvents and extraction time. *Biosci. J.*, Uberlândia, 35(4):1110-1119, 2019.

Sousa, C. M. M., Rocha e Silva, H., Vieira Jr, R. G. M., Ayres, M. C. C., Costa, C. L. S., Araújo, D. S., Cavalcante, C. D., Barros, E. D. S., Araújo, P. B. M., Brandão, M. S. & Chaves, M. H. (2007). Fenóis totais e atividade antioxidante de cinco plantas medicinais. *Quim. Nova.* 30:351-355.

Schenkel, E. P., Zannin, J. C. P., Mentz, L. A., Bordignon, S. A. L., Irgang, B. Plantas tóxicas. In: Simões, C. M. O., SchenkeL, E. P., Gosmann, G., Mello, J. C. P., Mentz, L. A. & Petrovick, P. R. (2000). (Org) *Farmacognosia: da planta ao medicamento*. Porto Alegre, Editora da Universidade/UFRGS, Florianópolis, Editora da UFSC, p. 755-788, 2000.

Shahidi, F. (1996). Natural Antioxidants: An Overview "in" Natural Antioxidants Chemistry, Health Effects, and Applications. AOCS Press: Champaign, Illinois, p. 1-11.

Spiridon, C. S., Anghel, N., Teaca, C. A., Bodirlau, R. & Armtu, A. (2011). Antioxidant capacity and total phenolic contents of orégano (*Origanum vulgare*), lavender (*Lavandula angustifólia*) and lemon balm (*Melissa officinalis*) from Romania. *Nat Prod Res.* 17:1657-61, 2011. doi: 10.1080/14786419.2010.521502.

Swain, T. & Hillis, W. E. (1959). The phenolic constituents of Prunus domestica. The quantitative analysis of phenolic constituents. *Journal. Science. Food Agricuture*. 10:63-68, 1959.

Vázquez, A. M., Totosaus, A., González, L. R. G., Salazar, K. A. F. & Martínez, I.G. (2007). Antioxidantes I. Chile ancho (*Capsicum annum* L. grossum sendt.) y romero (*Rosmarinus officinalis* L.) como fuentes naturales de antioxidantes. *Ciencia y Tecnologia*. 6:112-116.

Xu, D. P., Li, Y., Meng, X., Zhou, T., Zhou, Y., Zheng, J. J. & Li, H. B. (2017). Natural Antioxidants in Foods and Medicinal Plants: Extraction, Assessment and Resources, *International Journal of Molecular Sciences*. 18 (96). https://doi.org/10.3390/ijms18010096.

Velioglu, Y. S., Mazza, G., Gao, L. & Oomah, B. D. (1998). Antioxidantactivity and total phenolics in selected fruits, vegetables, and grain products. J. Agric. Food Chem. 46:4113–4117.

Wicklund, T., Rosenfeld, H. J., Martinsen, B. K., & Sundfor, M. W. (2005). Antioxidant capacity and colour of strawberry jam as influenced by cultivar and storage conditions. LWT - Lebensmittel-Wissenschaft und Technologie, 38:387-391.

Wu, J. H., Tung, Y. T., Wang, S. Y., Shyur, L. F., Kuo, Y. H., & Chang, S. T. (2005). Phenolic antioxidants from the heartwood of Acacia confusa. J. Agric. and Food Chem. 53:5917-5921.