Antioxidant activity, phenolic content and fatty acid profile for seeds *Physalis angulate* L

Atividade antioxidante, teor de fenólicos e perfil de ácidos graxos de sementes da *Physalis angulate* L

DOI:10.34117/bjdv6n12-570

Recebimento dos originais: 10/11/2020
Aceitação para publicação: 22/12/2020

Tânia Granzotti da Silva
Mestre em Ciência e Tecnologia Ambiental pela Universidade Federal da Grande Dourados, CEP 79.825-070 – Dourados-MS, Brasil
E-mail: taniagranzotti@hotmail.com

Cintia Granzotti da Silva Scudeler
Doutora em Biotecnologia e Biodiversidade, Universidade Federal da Grande Dourados, CEP 79.825-070 – Dourados-MS, Brasil
E-mail: cintiagranzotti@hotmail.com

Euclésio Simionatto
Programa de Pós-graduação em Recursos Naturais, Universidade Estadual de Mato Grosso do Sul, 79950-000 Navirai-MS, Brasil
E-mail: euclesio@uems.br

Rogério Cesar de Lara da Silva
Programa de Pós-graduação em Recursos Naturais, Universidade Estadual de Mato Grosso do Sul, 79950-000 Navirai-MS, Brasil
E-mail: rcsilva@uems.br

ABSTRACT
Brazil has a large diversity of plants that have not been discovered, or have been little studied before their potential in treating various diseases. The *Physalis angulata* L., family Solanaceae is popularly known as "Winter Cherry", "camapum", "bag guy", among others, and is used in folk medicine for various treatments. This study were evaluated the antioxidant potential and phenols for leaves, fruit peels, and the chromatographic profile of fatty acids of the ground cherry seeds. For the determination of antioxidant potential was used the method free radical DPPH (0.004%) and rutin as positive standard. The phenolic content was determined by the Folin- Ciacalteu method. The profile of fatty acids of the seeds was analyzed by gas chromatography with flame ionization detector (GC-FID). Through this evaluation gave significant results to test the antioxidant in leaves showing inhibition values of the 98.15%. However, the fruit pulp showed a percentage of 90.87% of antioxidants against the DPPH. For extracts of bark and fruits also yielded satisfactory results as the antioxidant potential. The fruit pulp was also rich in phenolic compounds having a contente of 113.18 mg GAE / mg extract. The chemical composition for the fatty acids present in seeds have a high content of linoleic acid (C18:2n6c) with 69.9%. Further studies should be conducted in relation to compounds present in *Physalis angulata*, so you can develop new products to benefit human health.

Keywords: Medicinal plant, DPPH, Gas chromatography, fatty acids, phenols.
RESUMO
O Brasil possui uma grande diversidade de plantas que não foram descobertas ou foram pouco estudadas antes de seu potencial no tratamento de várias doenças. A *Physalis angulata* L., família Solanaceae é popularmente conhecida como “Cereja de Inverno”, “camapum”, “Bag guy”, entre outras, e é utilizada na medicina popular para diversos tratamentos. Neste estudo foram avaliados o potencial antioxidante e fenólicos para folhas, cascas de frutas e o perfil cromatográfico de ácidos graxos de sementes de cereja moídas. Para a determinação do potencial antioxidante foi utilizado o método radical livre DPPH (0,004%) e a rutina como padrão positivo. O conteúdo fenólico foi determinado pelo método de Folin-Ciocalteu. O perfil de ácidos graxos das sementes foi analisado por cromatografia gasosa com detector de ionização de chama (GC-FID). Através desta avaliação deu-se resultados significativos ao testar o antioxidante em folhas apresentando valores de inibição da ordem de 98,15%. Porém, a polpa da fruta apresentou um percentual de 90,87% de antioxidantes contra o DPPH. Já os extratos de casca e frutas também apresentaram resultados satisfatórios quanto ao potencial antioxidante. A polpa da fruta também foi rica em compostos fenólicos, contendo 113,18 mg de GAE / mg de extrato. A composição química dos ácidos graxos presentes nas sementes apresentam alto teor de ácido linoléico (C18:2n6c) com 69,9%. Mais estudos devem ser realizados em relação aos compostos presentes na *Physalis angulata* L., para que seja possível desenvolver novos produtos que beneficiem a saúde humana.

Palavras-chave: plantas medicinais, DPPH, cromatografia gasosa, perfil de ácidos graxos.

1 INTRODUCTION

The knowledge of medicinal plants becomes as old as the human species. Its use in symbolic treatments often the only therapeutic option in different communities. Through the use and established efficacy of many medicinal plants of various species contributed to the advancement and development of herbal medicine (Marciel and others 2002).

The use of plants for treating diseases initiated empirical, is normally used for preparations leaves, fruits, roots and other plant parts. This knowledge is passed from generation to generation as oral or books left by ancient civilizations, being initiated by the use of herbal medicine (Cunha and others 2003).

Today, herbal medicine, yet it is based on traditional use, going to check irrelevant aspects of the previous period, as well as being increasingly supported by the characteristics of quality, efficacy and safety (Cunha and others 2003).

In recent years there is growing concern about "live longer and live with quality", so science is constantly changing, conducting several surveys in order to improve the quality of life through organic and inorganic substances used as drugs. In Brazil, research on organic framework has been highlighted due to diversity of riches found in fauna and flora, thus arousing the interest of researchers in relation to their active ingredients and medicinal properties (Farias 2003; Strathern 2002).

Brazil has the greatest biological diversity in the world, because of their wealth, just the cerrado biome contains more than 6,000 plants vinculares (Mendonça and others 1998), many with food and medical value (Almeida and others 1998), country is rich in biodiversity. The use of certain plants as a
healing medium is a highly widespread and popular activity, often used wrongly, many plants have toxic principles and their indiscriminate use can cause serious problems, thus encouraging research in the area should be motivated to guarantee the required safety possible. Often researchers modify the basic structure of plant constituents in order to increase their activity or make it specific for a particular purpose. Thus, medicinal plants have many compounds that can influence their action. These compounds have the function of protecting the active components of certain changes, including oxidation, hydrolysis among others, may also enhance absorption by the body, to facilitate passage through membranes or by inhibiting enzymatic systems. The action of the plant or extract, often has greater activity than the same amount of active constituent isolated, so there is a great interest in herbal medicines (Cunha and others 2003). In organic chemistry, in about fifty years (the early and mid-nineteenth century) was characterized by high numbers of isolated organic compounds. In this period also gave a theoretical evolution of organic chemistry, this system of identification of the composition percentage of organic compounds to organic chemical analysis gave a significant contribution (Neves and others 2008).

Through matching the chemical composition of Physalis angulata L. studies, we have obtained the isolation of flavonoids, alkaloids, steroids and ceramides (Ismail and others 2001). However, a majority group of secondary metabolites characterized as vitaesteroides has been found in roots and leaves of P. angulata, being called physalins. The plant in question have already been isolated and identified several physalins as B, E, F and G (Tomassini and others 2000). The same has been used in traditional medicine as an analgesic, anti-rheumatism, which aids in the treatment of sore throat and abdomen (Bastos and others 2008). Is also cited as a healing, purifying the blood, kidneys shortener albumin, fortifying the optic nerves. It is indicated as an adjunct in the treatment of prostate carcinoma and high cholesterol, fight diabetes, hepatitis, chronic rheumatism, skin diseases, bladder, kidneys and liver. Some biological activities of the extracts of P. angulata, such as antibacterial, anticancer and anti-inflammatory (Ismail and others 2001) are reported.

Under this hypothesis, the objectives of this study were to evaluate the antioxidant activity and phenolic compounds in ethanol extracts and chemical composition for fixed oils derived from Physalis angulata L. obtained from cerrado in Brazil.

2 MATERIALS AND METHODS
2.1 OBTAINING THE SAMPLE

The fruits of Physalis angulata L. were obtained from the cerrado near the city of Naviraí - Mato Grosso do Sul state, Brazil, and were collected from September to January. After collection, the samples were washed in tap water and sanitized in a solution of sodium dichloroisocyanurate dihydrate.
0.66% (active chlorine content 3%). Were separated into parts (leaf, bark and seed) holding a piece of whole fruit, before performing the analysis. The parts of the fruit were dried in air circulating oven for 24 hours at 35 °C. But the fruit that remained whole not undergone previous drying. After this process they were subjected to methods of extraction and chemical tests.

2.2 OBTAINING THE ETHANOL EXTRACTS

To obtain the extract, collected 185.52 g of leaves and the same were stored in glass jar with lid and ethanol at a ratio of 2 : 1 being left for 15 days at rest to obtain a coloring intense extracts. After this period, a filtration was performed using filter paper, using the assistance of a Buchner funnel and 250 ml erlemmeyer. The extract was placed in a 250 mL volumetric flask and evaporated in a rotary evaporator will vacuum to obtain the crude ethanol extract of the leaves. To prepare the bark extract were necessary 145.00 g and of the fruit extract 54.30 g. For the determination of phenolic compounds were used the same above-mentioned methodology.

2.3 Fixed oil extraction and characterization.

The fruits were analyzed for fixed oils content that makes up the lipid fraction of plant metabolism. Samples of the collected seeds were subjected to oven drying and air circulation in order to remove moisture. The extraction the lipids of oil were performed through the using the Soxhlet extractor with adding hexane solvent by 4 hours. It was used about 130 g of the material (seed) ground in a Wiley mill turning the seeds into small particles. Then the solvent was evaporated on a rotary evaporator in order to remove excess solvent contained in the oil and recovering the same.

2.4 QUALITATIVE ANTIOXIDANT ASSAY

Prepared was a solution of 2,2-diphenyl-1-picryl-hidrazila (DPPH) 0.04 g in 100 ml of methanol and kept refrigerated and sealed until use. The sample (0:05 g) was dissolved in methanol and applied to capillary TLC plate for revelation with DPPH. The yellow spots show the presence of an antioxidant.

2.5 QUANTITATIVE ANTIOXIDANT ASSAY

To test the DPPH antioxidant was used 10 mg of extract for leaf, bark and fruit of Physalis angulata, dissolving in 5 mL methanol. Dilutions were conducted at concentrations of 10.0; 5.0; 2.5; 1.25; 0.62 mg mL\(^{-1}\) for the extracts and supplemented with 1 mL of DPPH and methanol. After a 30 min. incubation at room temperature, protected from light, measured by reduction of DPPH free radical by reading the absorbance in a UV-Visible spectrophotometer from Varian - Cary 50 CON (USA) at 517nm. In this test, a natural antioxidant rutin was used as a positive standard. The percentage of
inhibition of DPPH in the samples was calculated using the following equation: % inhibition of DPPH = [(AB - AA) / AB] x 100 where AB = absorbance of the blank (t = 0 min) AA = absorbance of the sample (t = 30 min).

2.6 DETERMINATION OF TOTAL PHENOLIC COMPOUNDS

For the determination of total phenolic content of the Folin-Ciocalteu method descripts by Singleto and Rossi (1965) was utilizable. To analyses of the extracts 5 mg were dilutec in 5 mL of methanol to be utilized. An aliquot of 100 µL of this solution was transferred to flasks of 5 mL. To this solution was added 1 mL of distilled water and then 0.2 ml of Folin-Ciocalteu reagent. Finally, we added 0.6 ml of a 20% Na₂CO₃ solution and completed to volume with distilled water. After 90 min, the absorbance of the samples was measured at 750 nm using a quartz vial (1 cm), with the "white" the methanol and all reagents except the extract. The total phenols (TP) was determined by interpolating the absorbance of the samples against a calibration curve constructed with standard gallic acid (50 and 500 mg mg⁻¹) and expressed as mg of GAE (gallic acid equivalents) per mg of extract.

2.7 FIXED INDEX SAPONIFICATION OF OIL

To carry out the saponification was weighed 1.0 g of the oil, and added 50 mL of 0.5 M KOH and transferred to a volumetric flask under reflux for 30 min at a temperature between 50 °C to 60 °C. After cooling the solution, the solution was transferred to beaker and added 5 drops of phenolphthalein indicator. The solution was titrated with H₂SO₄ 0.5 M. For achieving free saponification of the oil, was added in a 50 mL erlenmeyer 0.5 M KOH and phenolphthalein (5 drops). The solution was titrated with H₂SO₄ 0.5M. The calculation was done as follows: (Vb x Va) x N x MM (KOH) / m (g) sample, and Vb = free KOH (volume); Va = KOH oil (volume); N = Normality or MM = Mass Concentration and Molar KOH. The tests were performed in triplicate.

2.8 ACID INDEX OF FIXED OIL

To carry out the acid number was weighed 1 g of oil being added to a solution of 15 ml ether / ethanol and 2: 1, and added 3 drops of phenolphthalein indicator solution and titrated with 0.1 M KOH . For free solution used the same procedure in the absence of oil. For the calculation should follow: IA = V x C x MM (KOH) / m (g) sample, and IA = index of acidity; V = Volume (mL); C = concentration (M); MM = Molar mass (g) and m = mass. The tests were performed in triplicate.
2.9 CHARACTERIZATION OF FATTY ACIDS

The characterization of the fatty acids present in the oil was performed according to the method described in ISO 5509, 1978. The treated samples were analyzed by gas chromatograph HP 5890A Series II (USA), equipped with polyethylene glycol chromatographic column (60m x 0.25mm x 0.25 μm) with flame ionization detector (GC-FID). The chromatographic conditions were: initial temperature 160 °C / 5 min → 5 °C / min to 220 °C / 10 min → 3 °C / min to 235 °C / 60 min. An aliquot 50 μL of fatty material was dissolved in heptane (1 mL) followed by addition of 1 ml (2 mol L⁻¹ KOH) plus addition of 1 mL of heptane. The organic phase of heptane containing methyl esters of fatty acids was collected and 2 μL injected in split mode (1:80) GC-FID. The identification of fatty acids was carried out by comparing the retention time of a mixture of fatty standard the 32 compounds (2000 mg L⁻¹ - Sigma-Aldrich) fatty acids. The quantification was carried out by area normalization of the chromatographic peaks.

2.10 STATISTICAL ANALYSIS

Analyses were performed in triplicate and the results shown by the mean (n = 3) and standard deviation. Comparisons between groups were subjected to analysis of variance (ANOVA), with significant differences determined by Tukey test p ≤ 0.05.

3 RESULTS AND DISCUSSION

The amount of plant material collected and the yield obtained for each are shown in Table 1. The DPPH test, in addition to quantifying the antioxidant activity can also be used for qualitative test where a permanent yellow color on a purple background indicates a positive test for antioxidant compounds. Best results were obtained for the extract of the leaves and bark than to the fruit flesh. The oil extracted from the seeds also presented a slow activity antioxidant.

| Samples  | Average (g) | Average of extracts (g) |
|----------|-------------|-------------------------|
| Husk     | 145.00±0.03 | 10.26±0.85              |
| Leaf     | 185.52±0.10 | 25.62±0.50              |
| Fruit polp | 54.30±0.07 | 6.03±0.34               |
| Seed     | 130.00±0.74 | 12.00±0.12              |

Avarage of samples n=3.

The quantitative antioxidant tests were performed with the ethanol extract obtained crude of the leaf, bark and fruit of *Physalis*. Was used for this test solution of DPPH (0.004%). This method of assessment of the percentage of antioxidant activity occurs change in coloration of the sample, and,
when added to the sample aliquot in the DPPH solution that has a purple color after, if the sample is active gets a yellow tint.

These color changes are related to the antioxidant activity present in swatches. The extracts of *P. angulata* L. were analyzed and showed a significant amount of antioxidant activity; it was shown inhibitions between 39-98% for concentrations of 0.62, 1.25, 2.5, 5 and 10 mg mL\(^{-1}\). Although higher antioxidant activity has been to the leaves, the extract obtained for the fruit pulp presented a percentage of 90.87% inhibition. The results are showing in Table 2.

Table 2. Values in percentage inhibition of prepared with different concentrations of the extracts of leaf, bark and whole fruit of *Physalis angulata*.

| Concentration (mg mL\(^{-1}\)) | % Inhibition | % Inhibition | % inhibition | % inhibition |
|---------------------------------|--------------|--------------|--------------|-------------|
|                                 | Leaf         | Husk         | Fruit        | Rutin       |
| 0.62                            | 39.76±0.01\(^{a\;}\) | 30.74±0.02\(^{b\;}\) | 25.92±0.05\(^{c\;}\) | 92.05±0.01\(^{d\;}\) |
| 1.25                            | 57.68±0.60\(^{a\;}\) | 45.78±0.40\(^{b\;}\) | 39.48±0.20\(^{c\;}\) | 93.79±0.50\(^{d\;}\) |
| 2.5                             | 78.34±0.08\(^{a\;}\) | 76.45±0.10\(^{b\;}\) | 71.51±0.13\(^{c\;}\) | 95.94±0.09\(^{d\;}\) |
| 5                               | 84.64±0.10\(^{a\;}\) | 84.45±0.50\(^{a\;}\) | 84.13±0.40\(^{a\;}\) | 97.47±0.10\(^{b\;}\) |
| 10                              | 98.15±0.17\(^{a\;}\) | 91.50±0.11\(^{b\;}\) | 90.87±1.00\(^{b\;}\) | 98.34±0.14\(^{a\;}\) |

Results expressed by the average of three replicates and standard deviation. *Different letters (in line) significantly different (p ≤ 0.05) by Tukey test.

The equation of the calibration curve obtained for galic acid was \( y = 0.0031x + 0.0386 \) with a correlation coefficient \( r = 0.9897 \). Total phenols were determined by the colorimetric method using Folin Ciocalteu. The tests were performed in triplicate and the results are expressed in mg equivalent to gallic acid (mg GAE.mg\(^{-1}\) of extract) in Table 3. The results obtained in the determination of the phenols by Folin-Ciocalteu method, showed high contents of bioactive phenolics compounds for the three constituent parts of the plant, as well as the results presented to the antioxidant activity.

Table 3. Average values for concentrations of phenols (mg GAE / mg of extract), the Folin-Ciocalteu method.

| Extract | Total phenols |
|---------|---------------|
| Leaf    | 130.05±0.12\(^{a\;}\) |
| Husk    | 116.40±0.17\(^{b\;}\) |
| Fruit   | 113.18±0.10\(^{c\;}\) |

Results are expressed by the mean value of three replicates and standard deviation. *Different letters (in the same column) show a significant difference (P ≤ 0.05) by Tukey test.

The saponification index (SI) is a measure of free and combined fatty acids that exist in the oil and is directly proportional to the average molar (MM) mass. The smaller the MM, the greater the SI (Carvalho, 2011), compared with values from literature data are shown in Table 4. Analyzing the comparative data can be observed that the saponification index for crude oil gave a result *Physalis*
50.47±0.12 mg g⁻¹, well below the other oils that showing IS greater than 180 mg KOH g⁻¹ as described by Carvalho (2011).

Table 4. Determination of saponification index (mg KOH g⁻¹) and acidity index (mg KOH g⁻¹) Gross Oil: Canola, Physalis angulata and Olive.

| Oil                  | Saponification index | Acidity index   |
|----------------------|----------------------|-----------------|
| Canola oil           | 182-193              | 0.061 ± 0.001   |
| Physalis angulata    | 50.47±0.12           | 0.029 ± 0.001   |
| Olive oil            | 182-193              | 0.079 ± 0.002   |

About the acid index, these data show that the oil used has a low acidity for crude oil Physalis. Due to the low acidity of the vegetable oil analyzed, it is estimated that the fatty acids are linked to glycerol molecules, being the predominant form triacylglycerol’s, as described in the literature. As shown in Table 5 the data were compared with literature data described by Milinsk (2007) for canola oil and olive oil, and showed a higher acid value than oil in the ground cherry.

Table 5. Percentage obtained for each fatty acid Physalis angulata L.

| Fatty acids | Percentage |
|-------------|------------|
| Palmitic (C16:0) | 11.3±0.12 |
| Estearic (C18:0) | 3.2±0.1   |
| Oleic (C18:1n9c) | 10.7±0.09 |
| Linoleic (C18:2n6c) | 69.9±0.21 |
| SFA         | 14.5       |
| UFA         | 10.7       |
| PUFA        | 69.9       |
| SFA / PUFA  | 1/5.55     |
| Sum         | 95.1       |

SFA: saturated fatty acids; PUFA: polyunsaturated fatty acid; UFA: unsaturated fatty acid.

Through analysis of oil from the seeds of Physalis by GC-FID was possible to identify the main esters being palmitic (C16: 0); stearic (C18: 0); oleic (C18: 1n9c) and linoleic (C18: 2n6c) acid. As compared to the literature data reported by Milinsk (2007), it can be seen that the olive oil also presents oleic esters (C18: 1n9c); palmitic (C16: 0) and linoleic (C18: 2n6c), not only presenting stearic (C18: 0). Therefore canola oil esters presents α - linoleic acid (C18: 3n3); oleic (C18: 1n9c) and linoleic (C18: 2n6c), differing only palmitic and stearic. Table 5 shows the percentage of fatty acids determined for the seeds of Physalis. Comparing the relationship between saturated and unsaturated fatty acids analyzed in this work with those cited by Borges and others (2007), for common oils such as peanut, corn, soybean, it was found that the oil from the seeds of Physalis (1 / 5.5), below are the reasons for peanuts (1 / 2.8), soy (1 / 5.7) and maize (1 / 6.7). As for the oil from the seeds umbu studied by Borges and others (2007), found the ratio between saturated / unsaturated was lower (≈ 1 / 1.2) showing has a
low content of unsaturated fatty acids. This indicates that the Physalis fruit seeds contain higher concentrations of unsaturated mainly from linoleic acid (C18:2n6c) with 69.9%.

4 CONCLUSION

Ethanol extracts of Physalis angulata L. were obtained from leaf, bark and fruit, according to the methodology described in the literature were determined antioxidant activity (against free radical DPPH) using rutin as standard positive and total phenols using the method applies Folin- Ciocalteu reagent. Analysis showed high antioxidant activity for the three parties investigated, and the percentage inhibition to the DPPH radical was large, tests have shown that the amount of extract used were sufficient to cause inhibition of free radicals. The content of phenols (bioactive compounds) was also with high values, which explains the high percentage inhibition against DPPH. To oil the tests showed satisfactory results, offers great possibilities for food use, as it presented a high content of linoleic fatty acids (69.9%). However should be done further study in relation to the compounds present in Physalis angulata L., so you can develop new products to benefit human health.

ACKNOWLEDGMENTS

The authors thank CAPES and CNPq
REFERENCES

Almeida, S.P.; Proença, C.E.B.; Sano, S.M.; Ribeiro, J.F. 1998. Cerrado: Espécies vegetais úteis. DF: EMBRAPA – CPAC. Planaltina. 464p.

Bastos, G.N.T.; Silveira, A.J.A.; Salgado, C.G.; Picanço-Diniz, D.L.W.; Do Nascimento, J.L.M. 2008. Physalis angulata extract exerts anti-inflammatory effects in rats by inhibiting different pathways. Journal of Ethnopharmacology, 118, 617 – 626.

Borges, S.V.; Maia, M.C.A.; Gomes, R.C.M.; Cavalcanti, N.B. 2007. Chemical Composition of Umbu (Spondias tuberosa Arr. Cam) SEEDS. Química Noval. 30 (1), 49-52.

Carvalho, C.O. 2011. Comparação entre métodos de extração do óleo de Mauritia flexuosa L.f. (Arecaeeae - buriti) para o uso sustentável na reserva de desenvolvimento tupé: rendimento e atividade antimicrobiana. Dissertação de Mestrado, Biotecnologia e Recursos Naturais, Universidade do Estado do Amazonas, Manaus, Amazonas, 110p.

Cunha, A. P.; Silva, A. P.; Roque, O. R. 2003. Plantas e produtos vegetais em fitoterapia. Fundação Calouste Gulbenkian. Lisboa, 701p.

Farias, R. F. 2003. Para gostar de ler: História da Química II. Editora: Átomo, Campinas, SP, Brasil.

Ismail, N.; Alam, M. 2001. A novel cytotoxic flavonoid glycoside from Physalis angulata. Fitoterapia, 72: 676-679.

Maciel, M.A.M.; Pinto, A.C.; Junior, V.F.V.; Grynberg, N.F.; Echevarria, A.; Grynberg, N.F.; Echevarria, A. 2002. Plantas medicinais: a necessidade de outros multidisciplinares. Química Nova, 25, 429-438.

Mendonça, R.; Felfili, J.M.; Walter, B.M.T.; Silva, J.M.C.; Rezende, A.V.; Filgueiras, T.S.; Nogueira, P.E.N. 1998. Flora vascular do Cerrado. EMBRAPA-CPAC.

Milinsk, M.C. 2007. Análise comparativa entre oito métodos de esterificação na determinação quantitativa de ácidos graxos em óleo vegetal. Tese de Doutorado em Química, Universidade Estadual de Maringá. 118p.

Peres, M. T. L. P.; Simionatto, E.; Hess, S. C.; Bonani, V.F.L.; Candido, A.C.S.; Casteli, C. Poppi, N. R.; Honda, N. K.; Cardoso, C. A. L.; Faccenda, O. 2009. Estudos químicos e biológicos de Microgramma vacciniifolia (Langsd. & Fisch) Copel (Polypodiaceae). Química Nova. 32 (4): 897-1.

Singleton, V, L.; Orthofer, R.; Lamuela-Raventós, R.M. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Methods in Enzymology. 299: 152-78.

Souza, G. H. B.; Mello , J. C. P.; Lopes, N. P. 2012. Farmacognosia: Coletânea científica. 1 ed. Ouro Preto: Editora UFOP.

Souza, C. M. M.; Silva, H. R.; Vieiras, J. G. M.; Ayres, M. C. C.; Costa, C. L. S.; Araújo, D. S.; Cavalcante, L. 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. Química Nova. 30 (2): 351-5.
Shahidi, F. 2008. Antioxidants: extraction, identification, application and efficacy measurement. Electronic Journal of Environmental, Agricultural and Food Chemistry. 7 (8): 3325-30.

Singleton, V. L.; Rossi, J. A. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Vitic., 16: 144-158.

Smith, P. M. 1976. The chemotaxonomy of plants. Edward Arnold, Bristol.

Strathern, P. 2002. O sonho de Mendeleiev, a verdadeira história da química. Jorge Zahar, Rio de Janeiro. 264p.

Simon, D. 2001. O guia Decep Chora de ervas: 40 receitas naturais para uma saúde perfeita. Rio de Janeiro: Campus.

Su, B., Misico, R., Park, E. J., Santarsiero, B. D. 2002. Isolation and characterization of bioactive of the leaves and stems of Physalis philadelphica. Tetrahedron. 58 (17): 3453-3456.

Tomassini, T. C. B., Barbi, N. S., Ribeiro, I. M., Xavier, D. C. D. 2000. Gênero Physalis: uma revisão sobre vitéasteróides. Química Nova, São Paulo. 23(1): 47-57.

Vale, N.B. A farmacobotânica, ainda tem lugar na moderna anestesiologia? Ver. Brás. Anestesia, 2002.

Veras, M. L., Bezerra, M. Z. B., Lemos, T., Lin, G., Uchoa, D. E. A., Braz, F. R., Chai, H. B., Cordel, G. A., Pessoa, O. D. 2004. Cytotoxic Withaphysalins from the Leaves of Acnistus arborescens. J. Nat. Prod. 67(4):710-713.

Wu, S. J., NG, L. T., Chen, C. H., Lin, D. L., Wang, S. S., Lin, C. C. (2004). Antihepatoma activity of Physallis angulata and P. peruviana extracts and their effects on apoptosis in human Hep G2 Cells. Life Science. 74(16): 2061-73.