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POLYPHENOLS AND ANTIOXIDANT ACTIVITY OF FOUR FRUITS NATIVE TO THE COAST OF CEARA UNDER DIFFERENT MATURATION STAGES[1]

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ABSTRACT – This study aimed to quantify polyphenolic compounds and antioxidant activity of four fruits native to the coast of Ceara under different maturation stages aiming their use for a healthier diet. Myrtle (*Eugenia punicifolia* (Kunth) DC.) fruits were collected at the Botanical State Park of Ceara, in Caucaia-CE and guajiru (*Chrisobalanus icaco* L.), manipuça (*Mouriri cearensis* Huber) and murici-pitanga (*Byrsonima gardneriana* A. Juss.) fruits were collected at the Botanical Garden of São Gonçalo, São Gonçalo do Amarante-CE. Fruits were collected and transported to the Laboratory of Plant Ecophysiology, being characterized, processed and frozen for chemical assessments at the Laboratory of Physiology and Postharvest Technology - *Embrapa Agroindústria Tropical*: polyphenolic compounds and total antioxidant activity. The ESTAT software was used for statistical analyses. The polyphenolic content had the highest and the lowest value for guajiru fruits with 480.73 and 10.90 mg/100 g, respectively and total antioxidant activity, the highest value was found for manipuça fruits, with 42.99 μM of Trolox/g of pulp and the lowest value for guajiru fruits, with 1.45 μM of Trolox/g of pulp. There was a significant correlation between polyphenolic content and total antioxidant activity obtained for guajiru fruits, which showed high correlation coefficient, $R = -0.95$ ($P < 0.05$).
Index terms: *Eugenia punicifolia*, *Chrisobalanus icaco*, *Mouriri cearensis*, *Byrsonima gardneriana*.

POLIFENÓLICOS E ATIVIDADE ANTIOXIDANTE DE QUATRO FRUTAS NATIVAS DO LITORAL CEARENSE EM DIFERENTES ESTÁDIOS DE MATURAÇÃO

RESUMO - A presente pesquisa objetivou quantificar os compostos polifenólicos e a atividade antioxidante de quatro frutas nativas do litoral cearense, em diferentes estádios de maturação, visando a sua utilização para uma alimentação mais saudável. Os frutos da murta (Eugenia punicifolia (Kunth) DC.) foram colhidos no Parque Estadual Botânico do Ceará, em Caucaia-CE, e os do guajiru (*Chrisobalanus icaco* L.), do manipuçá (*Mouriri cearensis* Huber) e do murici-pitanga (*Byrsonima gardneriana* A. Juss.), no Jardim Botânico de São Gonçalo, em São Gonçalo do Amarante–CE. Os frutos foram coletados e transportados ao Laboratório de Ecofisiologia Vegetal, sendo caracterizados, depois processados e congelados para as avaliações químicas, no Laboratório de Fisiologia e Tecnologia Pós-Colheita, da Embrapa Agroindústria Tropical: compostos polifenólicos e atividade antioxidante total. Utilizou-se o programa ESTAT nas análises estatísticas. O teor de polifenólicos teve o maior e o menor valor para o fruto de guajiru, com 480,73 e 10,90 mg/100 g, respectivamente, e à atividade antioxidante total, o maior valor foi no fruto do manipuçá, com 42,99 μM de Trolox/g de polpa, e o menor valor, no fruto do guajiru, com 1,45 μM de Trolox/g de polpa. Observou-se correlação significativa e negativa entre os polifenólicos e a atividade antioxidante total obtidos nos frutos do guajiru, que apresentou alto coeficiente de correlação, R= -0,95 (P < 0,05).
Termos para indexação: *Eugenia punicifolia*, *Chrisobalanus icaco*, *Mouriri cearensis, Byrsonima gardneriana.*

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INTRODUCTION

Fruit consumption has been associated with a decrease in cardiovascular and cancerous diseases, and this effect is promising due to the presence of antioxidants, which neutralize reactive species or free radicals, thus reducing oxidative damage in cell metabolism (SOUZA et al. 2014).

In order to understand the bioavailability of antioxidants and the functions of phenols of plant origin in the human body, it is important to determine their quantity and chemical structures (KOOLEN et al., 2013, BATAGLION et al., 2014, CARVALHO-SILVA et al., 2014 BATAGLION et al., 2015, BERTO et al., 2015).

The frequency and development of chronic diseases are partly due to the imbalance of functional components in the biological parameters, among which phenolic compounds, carotenoids, flavonoids, anthocyanins, terpenes, vitamin C and other phytochemicals present in almost all fruits found in Brazilian fruit species. In addition, substances contained in fruits can be responsible for the protection of cells, prevention of infections and diseases, providing sustainability, maintenance of recovery and compensatory metabolisms, such as antioxidant function (ROCHA et al., 2013).

Recently, natural antioxidants from medicinal plants have been intensively investigated in order to discover compounds capable of protecting from diseases related to oxidative stress and damage induced by free radicals. In this sense, polyphenolics have gained importance due to their potential as prophylactic and therapeutic agents in many diseases; therefore, many studies have been conducted by the scientific community reporting their antioxidant effects (KINDL et al., 2015).

The aim of this study was to quantify polyphenolic compounds and the antioxidant activity of four native fruits of the coast of Ceará at different maturation stages, myrtle (*Eugenia punicifolia* (Kunth) DC), guajiru (*Chrisobalanus icaco* L.), manipuçá (*Mouriri cearensis* Huber) and murici-pitanga (*Byrsonima gardneriana* A. Juss.), aiming at their use for a healthier diet.

MATERIAL AND METHODS

The work was carried out at the Laboratory of Plant Ecophysiology, State University of Ceará - UECE, and at the Laboratory of Post-Harvest Physiology and Technology, Embrapa Agroindústria Tropical, both in Fortaleza-CE, from February 2013 to November 2014.

Vouches specimens of guajiru - *Chrysobalanus icaco* L. (57408), manipuçá - *Mouriri cearensis* Huber (57407), murici-pitanga - *Byrsonima gardneriana* A. Juss. (57409) and myrtle - *Eugenia punicifolia* (Kunth) DC. (57410) were found deposited in the Prisco Bezerra Herbarium, Federal University of Ceará - UFC, Fortaleza-CE.

Myrtle fruits were collected in the State Botanical Park of Ceará, Caucaia-CE, whose coordinates are 3º44’10”S and 38º39’11”W, consisting of sandbanks, Caatinga and mangrove forests, with minimum precipitation from August to October (0.0 mm) and maximum in May (157.4 mm) (FUNCEME / IPECE, 2015), and equatorial climate of Savannah with dry summer (As), according to the Köppen-Geiger climatic classification (KOTTEK et al., 2006).

Guajiru, manipuçá and murici-pitanga fruits were collected from the Botanical Garden of São Gonçalo, São Gonçalo do Amarante-CE, located in coordinates 3º 36 ‘26”S and 38º 58’ 06”W, consisting of sandbanks, with minimum precipitation in August (1.5 mm) and maximum in April (281.8 mm) (FUNCEME / IPECE, 2015), and equatorial climate of Savannah with dry summer (As), according to the Köppen-Geiger climatic classification of (KOTTEK et al., 2006).

Fruits of the four species native to the coast of Ceará were manually and randomly collected from 10 previously selected plants of each plant species, approximately 2.5 kg of fruits per maturation stage. Guajiru, manipuçá, murici-pitanga and myrtle were characterized in the following stages (E), respectively: 5 (E_1-very immature, E_2-immature, E_3-mature, E4-ripe and E5-senescent), 5 (E_1-very immature, E_2-immature, E_3-mature, E_4-ripe, and E_5-senescent) and 6 (E_1-very immature, E_2-moderately immature, E_3-immature, E_4-mature, E_5-ripe and E_6-senescent).

Initially, seeds were removed from fruits, and the pulp was processed and frozen in freezer at -20 ° C ± 1, to finally analyze the total extractable polyphenols (POL) and total antioxidant activity (AAT).

POLs were determined using the Folin-Ciocalteau reagent and a standard curve of gallic acid as reference, according to methodology described by Larrauri; Rupérez; Saura-Calixto (1997).

AAT was determined by assay with free radical ABTS described by Rufino et al. (2007).

The experimental design was a completely randomized design, with five or six treatments, depending on the maturation stage of fruits, and four replicates consisting of 200 fruits each. Results were submitted to analysis of variance, observing the significance by the F test and, when significant, the Tukey test was carried out, at the 5% probability level, using the <V.1.0> - ESTAT Statistical Analysis System. To verify the linear correlations among variables, significance was verified by the t test, using the Assistat 7.7 beta statistical software.

RESULTS AND DISCUSSION

The results obtained for total extractable polyphenols (POL) at the different maturation stages of fruits (Figure 1) presented the following values: in guajiru fruits, minimum of 10.90 mg / 100 g in E_4 and maximum of 480.73 mg / 100 g in E_3; in manipuçá fruits, minimum of 18.28 mg / 100 g in E_1 and maximum of 94.91 mg / 100 g in E_4; in murici-pitanga fruits, minimum of 153.51 mg / 100 g in E_3 and maximum of 373.98 mg / 100 g in E_2; and in myrtle fruits, minimum of 75.51 mg / 100 g in E_6 and maximum of 180.68 mg / 100 g in E_2.

Analyzing POLs obtained from viuvinha fruits (*Myrcia splendens* (Sw.) DC.), Façanha (2012) obtained maximum of 415 mg/100 g no E_2 and in the case of mapirunga fruits, maximum of 414.61 mg / 100 was obtained in E_1; therefore, these values were higher than the maximum values obtained in myrtle fruits in this study (E_2-180,68 mg / 100 g).

Silveira (2008) analyzed POLs among 15 genotypes of mature puçá fruits (*Mouriri elliptica* Mart.), and genotype 14 presented the lowest value, 71.35 mg / 100 g, and genotypes 17 and E2 obtained the highest values, 180.45 mg / 100 g and 179.95 mg / 100 g, respectively; the lowest result reported by the author was lower than the mature manipuçá fruit analyzed in this study (E_4-94.91 mg / 100 g), while the highest values reported were higher.

For the total antioxidant activity (AAT), the results obtained in the different maturation stages of the four native fruits analyzed are described according to Figure 2. In guajiru fruits, minimum of 1.45 µM Trolox / g in E_5 and maximum of 20.20 µM Trolox / g in E_4; in manipuçá fruits, minimum of 9.28 µM Trolox / g in E_2 and maximum of 42.99 µM Trolox / g in E_1; in murici-pitanga fruits, minimum of 4.42 µM of Trolox / g in E_3 and maximum of 7.40 µM of Trolox / g in E_2; and in myrtle fruits, minimum of 6.69 µM of Trolox / g in E_5 and maximum of 18.46 µM of Trolox / g in E_2.

Studying the AAT of 15 genotypes of mature puçá fruits, Silveira (2008) found that genotype E1 presented lower value, 9.10 µM Trolox / g, and genotype 2 presented higher value, 26.49 µM Trolox / g; therefore, the lower result reported by the author was lower than mature manipuçá fruits in this study (E_4-14.08 µM Trolox / g), while the highest reported value was higher.

Studying the AAT of the pulp of mature Amazon fruits, including murici (*Byrsonima crassifolia*), Canuto et al. (2010) obtained value of 1.5 µM Trolox / g, which is lower than the value found for mature Murici-pitanga fruits in this study (E_4-6.16 µM Trolox / g).

Evaluating the AAT of mature murici fruits (*Byrsonima verbascifolia*), Morais et al. (2013) obtained value of 15.63 µM Trolox / g, which is higher than that obtained in mature murici-pitanga fruits in this work (E_5-7.06 µM Trolox / g).

Analyzing the AAT of mature uvaia fruits (*Eugenia pyriformis* Cambess), Pereira et al. (2012) obtained value of 336.29 µM Trolox / g, therefore it presents higher antioxidant potential compared to mature myrtle fruits in this work (E_5-6.69 µM Of Trolox / g).

In this study, a correlation was observed between the same species only for guajiru fruits (Table 1). A negative and significant correlation was obtained at 5% probability level between POLs and AAT, with R = -0.95. On the other hand, negative and significant correlations were observed among different species: GPOL and MPPOL (R = -0.99, P <0.01); GPOL and MUPOL (R = -0.99, P <0.01); MPPOL and MUPOL (R = -0.96, P <0.05); GAAT and MUAAT (R = -0.95, P <0.05); MPAAT and MUAAT (R = -0.99, P <0.01); as well as positive and significant correlations: MPPOL and GAAT (R = 0.98, P <0.05); GAAT and MPAAT (R = 0.98, P <0.05).

Analyzing 15 genotypes of puçá fruits, Silveira (2008) verified a positive and significant correlation at 1% probability level between POLs and AAT (R = 0.66, P <0.01); however, this behavior was not observed for manipuçá fruits in this study, since it did not present any significant correlation.

Kuskoski et al. (2006) verified a positive and significant correlation in 15 tropical fruits at 1% probability level between POLs and AAT, with R = 0.99, which differs from that obtained among these variables in guajiru fruits studied in this study (R = -0.95, P <0.05).

Similarly, working with pure cupuaçu pulps, Santos (2007) found a positive and significant correlation between POLs and AAT (R = 0.74, P <0.05), which also differs from results of this study for guajiru fruits.

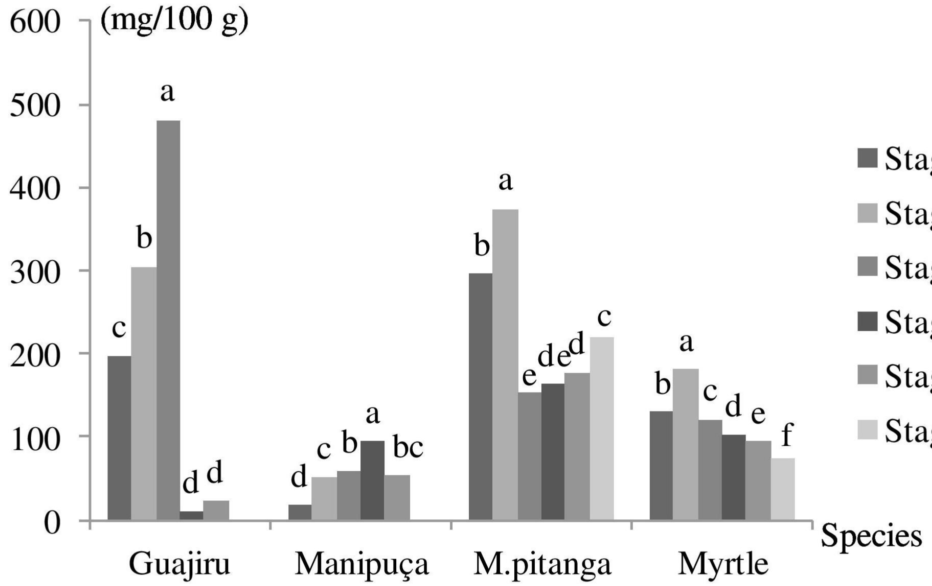

FIGURE 1 - Total extractable polyphenols (mg / 100 g) of guajiru (*Chrysobalanus icaco* L.), manipuçá (*Mouriri cearensis* Huber), murici-pitanga (*Byrsonima gardneriana* A. Juss.) and myrtle fruits (*Eugenia punicifolia* (Kunth) DC.) at different maturation stages.

Means followed by the same letter in the same species do not differ from each other by the Tukey test at 5% probability level.

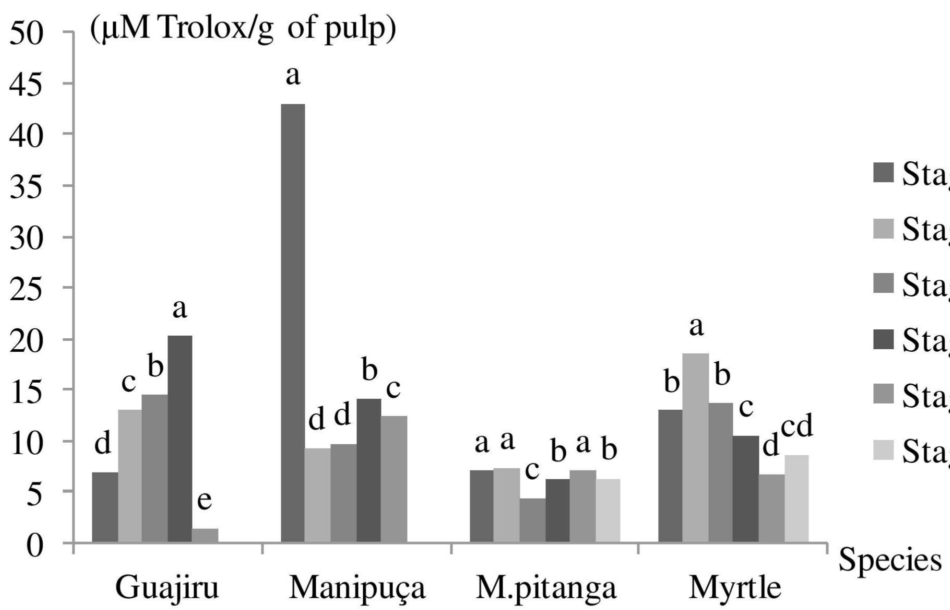

FIGURE 2 - Total antioxidant activity (µM Trolox / g) of guajiru (*Chrysobalanus icaco* L.), manipuçá (*Mouriri cearensis* Huber), murici-pitanga (*Byrsonima gardneriana* A. Juss.) and myrtle fruits (*Eugenia punicifolia* (Kunth) DC.) at different maturation stages.

Means followed by the same letter, in the same species, do not differ from each other by the Tukey test at 5% probability level.

TABLE 1 - Correlation matrix between total extractable polyphenol (POL) and total antioxidant activity (AAT) of guajiru (G), manipuçá (M), murici-pitanga (MP) and myrtle (MU) fruits at different maturation stages.

Variables	MPOL	MPPOL	MUPOL	GAAT	MAAT	MPAAT	MUAAT
GPOL	0.39^{ns}	-0.99**	-0.99**	-0.95*	-0.00^{ns}	-0.89^{ns}	0.83^{ns}
MPOL	-	-0.51^{ns}	-0.27^{ns}	0.64^{ns}	0.91^{ns}	-0.76^{ns}	0.83^{ns}
MPPOL	-	-	-0.96*	0.98*	-0.13^{ns}	0.94^{ns}	-0.89^{ns}
MUPOL	-	-	-	0.91^{ns}	0.12^{ns}	0.83^{ns}	-0.75^{ns}
GAAT	-	-	-	-	-0.29^{ns}	0.98*	-0.95*
MAAT	-	-	-	-	-	-0.44^{ns}	0.55^{ns}
MPAAT	-	-	-	-	-	-	-0.99**

*, ** Significant, respectively, at 5 and 1% of probability level, by the t test.
^{ns} Not significant.

CONCLUSIONS

Among the maturation stages of the four fruits studied, stage 3 for guajiru fruits, stage 4 for manipuçá fruits, stage 2 for murici-pitanga fruits and stage 2 for myrtle fruits showed the highest polyphenolic values when compared to the other stages, although guajiru fruits stood out, which presented the highest value.

For the antioxidant activity, maturation stage 1 of manipuçá fruits stood out, as it presented the highest antioxidant content among the fruits studied, although this stage had the lowest polyphenolic content, which suggests the action of other antioxidants, such as vitamins C and E, chlorophyll and carotenoids. However, relevant values were also obtained at maturation stage 4 for guajiru fruits, maturation stage 2 for myrtle fruits and maturation stage 2 for murici-pitanga fruits.

For the antioxidant activity, maturation stage 1 for manipuçá fruits stood out, as it presented the highest antioxidant content among the fruits studied; however, relevant values were also obtained at maturation stage 4 for guajiru fruits, maturation stage 2 for myrtle fruits and maturation stage 2 for murici-pitanga fruits.

Guajiru and manipuçá fruits can be used for a healthier diet, since guajiru has the highest polyphenolic values, and manipuçá, the highest antioxidant activity; therefore, both can be recommended to be commercially exploited by the food industry.

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