International Archive of Applied Sciences and Technology
Int. Arch. App. Sci. Technol; Vol 10 [1] March 2019 : 119-122
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[ISO9001: 2008 Certified Organization]
www.soeagra.com/iaast.html

IAAST
ONLINE ISSN 2277- 1565
PRINT ISSN 0976 - 4828

CODEN: IAASCA RESEARCH ARTICLE
DOI: .10.15515/iaast.0976-4828.10.1.119122

Antioxidant analysis of different parts of raw *Carica papaya*

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ABSTRACT
Over centuries, papaya (Carica papaya Linn.) belonging to Caricaceae family is a renowned nutritious and medicinal plant. Each and every part of the papaya has its own nutraceutical properties. This study sought to examine the total phenolic content (TPC) and total flavonoid content (TFC) from different parts of raw papaya such as peel, seed, pulp and leaves.1,1- Diphenyl-2-Picryl-Hydrazyl (DPPH) free radical scavenging assay was performed to evaluate the antioxidant potential of the samples and was correlated with total phenolic and flavonoid contents of samples. The total phenolics were expressed as mg GAE/100 gm i.e. mg/100g Gallic acid equivalent and the total flavonoids were expressed as mg QE/100 gm i.e. mg/100g Quercetin equivalent. The total phenol and flavonoid content of the leaves were found to be maximum with positive correlation with DPPH radical scavenging assay. Therefore, the leaf extracts of Carica papaya demonstrated potent antioxidant activity and could be of immense value in the pharmaceutical and food industry.
Keywords: *Carica papaya, antioxidants, phenolics, flavonoids, DPPH, pharmaceutical.*

Received 23.12.2018 Revised 28.01.2019 Accepted 21.02.2019

CITATION OF THIS ARTICLE
P Rajput, K Desai, N Khan, P Rajput, S Dasgupta.Antioxidant analysis of different parts of raw *Carica papaya*.Int. Arch. App. Sci. Technol; Vol 10 [1] March 2019 : 119-122

INTRODUCTION

Carica papaya Linn. belonging to *Caricaceae* family, is a widely grown perennial tropical tree, grows up to about 10 m in height with an erect trunk. Its leaves are huge, measuring about 50-70 cm in diameter, deeply palmately lobed with seven lobes [1]. Numerous species of *Caricaceae* have been used as medication against a variety of diseases [2]. Antioxidants act as cooperative networks employing a series of different redox reactions [3]. Antioxidant such as phenolics are the most pronounced secondary metabolites found in plants, and their distribution is shown throughout the entire metabolic process. These phenolic substances contain numerous varieties of compounds: simple flavonoids, phenolic acids, complex flavonoids and coloured anthocyanins [4]. These phenolic and flavonoid compounds are usually related to defence responses in the plant. However, these metabolites play an important part in other processes, for instance incorporating attractive substances to assist pollination, colouring for camouflage and defence against herbivores, as well as antibacterial and antifungal activities [5-7]. Thus, the aim of the study is to determine the total antioxidant activity (TAA) by determining total phenolic content (TPC), total flavonoid content (TFC) and DPPH free radical scavenging activity from the different parts of papaya tree including their unripe fruits peel, pulp, seeds and the young leaves.

MATERIALS AND METHODS

Collection of plant material

Natural leaves, and raw fruit of *Carica papaya* were collected very carefully from local residence of Surat, Gujarat. The peel, pulp and seed of raw papaya fruit was carefully separated out and cleaned properly.

Preparation of Extracts

Fresh leaves and other parts of raw papaya fruit were rinsed in water, dried on paper towel and were chopped into small pieces. 1gm of the chopped plant material was homogenised with 10 ml of 80% ethanol, then centrifuged at 10,000 rpm for 10 minutes. The pellet was discarded and the supernatant was used for further analysis.

Estimation of Total Phenol

The total phenolics of each fruit extract were determined by the Folin-Ciocalteu method [8]. The diluted aqueous solution of each extract (0.5 ml) was mixed with 2.5 ml of 0.2N Folin Ciocalteu reagent. This mixture was allowed to stand at room temperature for 5 minutes and then 2 ml of 7.5% sodium carbonate solution was added. After 2 hours of incubation, the absorbance was measured at 760 nm against water as blank. A standard calibration curve was plotted with Gallic acid. The results were expressed as mg/g Gallic acid equivalents (GAE)/ g of fresh plant material.

Estimation of Total Flavonoid

The total flavonoids were estimated by aluminium trichloride colorimetric method [9]. 2 ml of 2% aluminium trichloride ($AlCl_3$) in methanol was mixed with the same volume of the extract solution. Following 10 minutes of incubation, the absorbance was taken against a blank that consist of the same solution but without the $AlCl_3$ at 415 nm using UV-spectrophotometer. Quercetin was used as reference compound to produce the standard curve, and the results were expressed as mg of Quercetin equivalents (QE)/g of fresh plant material.

DPPH free radical scavenging assay

Effect of the sample extracts on DPPH radical was measured by using a slightly modified method [10]. Briefly, 1mM DPPH in 99.5% Ethanol. To 0.5 ml of DPPH radical solution, add 2 ml of the prepared extracts of plant material and the reaction mixture is vortexed for 10s and allow to stand at room temperature for 30 minutes. The absorbance is recorded at 517 nm by using UV Spectrophotometer and 75% ethanol act as control. Ascorbic acid is used as reference antioxidant compound.

The percentage of DPPH radical scavenging activity is determined as:

DPPH radical scavenging effect (%) = [1- (Test sample absorbance/blank sample absorbance)] ×100 (%).

RESULTS AND DISCUSSION

This study sought to examine the total phenolic content (TPC) and total flavonoid content (TFC) from different parts of raw papaya such as peel, seed, pulp and leaves.

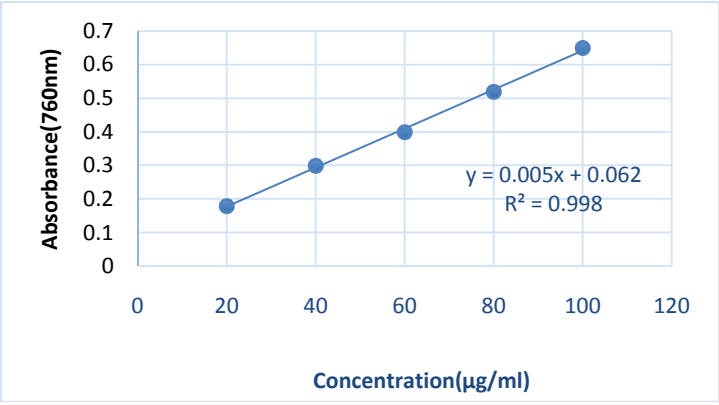

Fig 1: Standard curve of Gallic Acid

Phenolic compounds are widely distributed in plants [11], which have gained greatly attention, due to their antioxidant activities and free radical-scavenging abilities, which

potentially have beneficial implications for human health [12]. The TPC was determined in comparison with standard Gallic acid and the results were expressed in terms of mg Gallic aid equivalent (GAE)/ gm) using the standard curve equation: y=0.0058x + 0.062, R^2=0.9982, Where y is absorbance at 760 nm and x is total phenolic content. (Fig. 1)This study showed that the selected parts of the papaya plant varied significantly. It ranged from 4 mg GAE/gm in peel, 4.8 mg GAE/gm in seed to 5.6 mg GAE/gm in pulp and 9.2 mg GAE/gm in leaf. The result also directs that the young leaves contained high phenolic content that may provide good sources of dietary antioxidant.

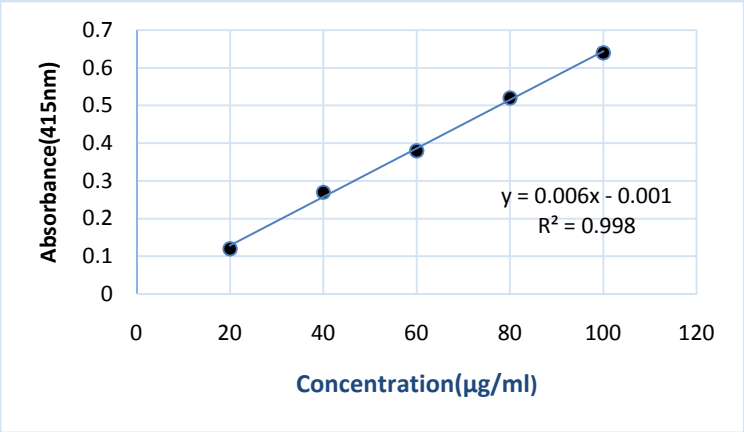

Fig 2: Standard curve of Quercetin

TFC of the extracts in terms of mg/gm quercetin equivalent antioxidant capacity (mg QEAC/gm) using the standard curve equation: y=0.0065x-0.001, R^2=0.9981, Where y is absorbance at 415 nm and x is total Flavonoid content. (Fig. 2) Flavonoids have antioxidant activity and could therefore lower cellular oxidative stress, which has been implicated in the pathogenesis of various neurodegenerative diseases, including Alzheimer's disease, Parkinson's disease, and amyotrophic lateral sclerosis [13].The total flavonoid, testified as Quercetin equivalent antioxidant capacity (QEAC). In present study TFC was obtained 8 mg QEAC/gm of seed, 9.2 mg QEAC/gm of peel, 9.6mg QEAC/gm of pulp and 25.2 mg QEAC/gm of leaf. In recent years, studies have shown that papaya fruit contains not only vitamins and other nutrients but also contains biologically flavonoids [14].

The free radical scavenging activity of different extracts was studied by their ability to reduce the DPPH. DPPH is a stable free radical and accepts an electron or hydrogen radical to turn into a stable diamagnetic molecule [15]. A newly prepared DPPH solution is of deep purple colour with absorption maximum at 517 nm and in the presence of antioxidant this colour disappears due to quenching of DPPH free radicals and transform them into a colourless product i.e. 2,2-diphenyl-1-hydrazine. Antioxidant mechanism performed by providing hydrogen atoms or electron [16]. The results revealed that DPPH radical scavenging ability ranged from 72% in seed, 80% in peel and 82% in pulp to 90% in leaf. This increase in radical scavenging ability could be attributed to the increase in the total phenol and flavonoid content [13].

CONCLUSION

Carica papaya leave, seed, pulp, and peel aqueous extracts exhibits noticeable antioxidant activity. Hence, the use of unripe papaya peel, seed along with pulp and leaf could be helpful in the treatment of some oxidative stress induced human diseases.

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