World Inventia Publishers

Journal of Pharma Research

http://www.jprinfo.com/

Vol. 7, Issue 10, 2018 ISSN: 2319-5622 USA CODEN: JPROK3

Research Article

SCREENING OF IN VITRO ANTIOXIDANT ACTIVITY OF SELECTED FRUIT PEELS: A COMPARATIVE STUDY

Gana Manjusha K. [1] *, Dr. Ganga Rao B. [2]

*[1] Vignan Institute of Pharmaceutical Technology, Andhra Pradesh, INDIA.

[2] University College of Pharmaceutical Sciences, Andhra University, Andhra Pradesh, INDIA.

Received on: 10-10-2018; Revised and Accepted on: 25-10-2018

ABSTRACT

The present was planned to evaluate the in vitro antioxidant activity of selected peel powers such as Annona sqaumosa, Actinidia deliciosa, Cucumis melo and Mallus pumila using different in vitro assays including the scavenging activities of hydroxyl radical, hydrogen peroxide, Nitric oxide and DPPH radical scavenging activities. The antioxidant activity of peels was compared with standard antioxidant rutin. The comparative study resulted that the Mallus pumila showed better antioxidant activity against the hydroxyl radical, hydrogen peroxide, Nitric oxide and DPPH radicals. The antioxidant activity of selected peels is due to its rich source of anthocyanins, carotenoids, tannins, flavanoids and phenols of fruit peels.

KEYWORDS: *Fruit peels, Antioxidants and Rutin.*

INTRODUCTION

Herbs are the source of treatment options of various ailments uncured by the existed therapies for the sake of mankind. Thousands of plant species are available throughout the world with phytoconstituents, which are the reasons of medicinal property [1]. Synthetic and natural antioxidants are available. Now a day interest is going in the identification of new herbal antioxidants as substitutes to the synthetic antioxidants, in order to avoid the un towards effects. Among the natural ones, the fruits and vegetables occupied because of high levels of antioxidant or secondary metabolites. Epidemiological studies have indicated that frequent consumption of natural antioxidants is associated with a lower risk of oxidative stress, which is the major cause of many dysfunctions. In general terms, oxidative stress is caused by the excessive generation of free radicals.

Free radicals are capable to reacts more fast might be due its nature of containing one or more unpaired electrons acts by donating or extracting the other molecules of carbohydrates, proteins, lipids and nucleic acids etc [2] Hence, free radicals are termed as reactive oxygen species (ROS). ROS are various forms of activated oxygen, which include free radicals such as superoxide anion radicals (.O2-) and hydroxyl radicals (OH·), as well as non free radicals (H_2O_2) and singlet oxygen [3]. The oxidation process that occur under the influence of atmospheric oxygen or reactive oxygen species can be delayed or inhibited by compounds called "Antioxidants". Nutritonal supplements and pharmaceutical products containing antioxidant active principles can satisfy the need of exogenous antioxidants. Amongst the most important exogenous antioxidants, vitamin E, vitamin C, β-carotene, vitamin E, flavonoids, mineral Se are well Exogenous antioxidants can derive from natural sources (vitamins, flavonoids, anthocyanins, some mineral compounds), but can also be synthetic compounds, like butylhydroxy anisole, butylhydroxytoluene, gallates, etc [4].

***Corresponding author:**

Gana Manjusha K.

Vignan Institute of Pharmaceutical Technology,

Andhra Pradesh, INDIA.

** E-Mail: manjusha.kondepudi.g@gmail.com*

DOI: https://doi.org/10.5281/zenodo.1471138

Among the selected plants parts, the fruit peels were also showed presence of phytoconstituents which might show the beneficial effects such as suger, protein, lipids, tannins, glycosides, vitamins and some trace elements. These peels are also contains notable amount of chlorophyll, carotene and anthocyanins. Hence, fruit peel has great potential for development due to its significant action over many health problems. Therefore it is to required extraction of its functional and medicinal components, in order to increase its potential values [5]. Research and development of fruit peel is still in the initial stage and need to develop new component from fruit as nutrition and in treatment major ailments. Hence the present study was planned to work on the peels of *Annona sqaumosa*, *Actinidia deliciosa*, *Cucumis melo* and *Mallus pumila*.

MATERIALS AND METHODS

Plant materials and preparation:

The ripened custard apple, kiwi, musk melon and apple were obtained from local market. The peels were manually separated and shade dried. The peels were grounded and passed through 40 mesh size. The moisture content of peel powder was found to be 12%. The powder was suspended in 2% gum acacia and used in the *in vitro* studies.

Hydroxyl radical scavenging activity:

Scavenging activity of hydroxyl radical was measured by the method of Halliwell et al., 1989.[6] Hydroxyl radicals were generated by a Fenton reaction (Fe^{3+}-ascorbate-EDTA-H_2O_2 system), and the scavenging capacity of the extract and standard towards the hydroxyl radicals was measured by using deoxyribose degradation method. The reaction mixture contained 2-deoxy-2-ribose (2.8 mM), phosphate buffer (0.1 mM, pH 7.4), ferric chloride (20 µM), EDTA (ethylene diamine tetra acetic acid) (100 µM), hydrogen peroxide (500 µM), ascorbic acid (100 µM) and different concentrations of the test sample and the final volume is 1 ml. The reaction mixture was then incubated for 1 hour at 37 °C. After the incubation an aliquot of the reaction mixture (0.8 ml) was added to 2.8% TCA (trichloro acetic acid) solution (1.5 ml), followed by TBA (thiobarbituric acid) solution (1 ml of 1% in 50 mM sodium hydroxide) and sodium dodecyl sulphate (0.2ml). Then the mixture was heated for colour development. The mixture is then cooled and the absorbance was measured at 532nm against an appropriate blank solution. All experiments were performed in triplicates. The percentage of inhibition was expressing as

$$\% \text{ Inhibition} = A_0 - A_1 / A_0 X\ 100$$

Where A_0 was the absorbance of the control without a sample, A_1 is the absorbance in the presence of the sample.

Hydrogen peroxide radical scavenging activity:

The hydrogen peroxide scavenging assay was carried out by the method described by Ruch et al., 1989 [7]. The principle of this method is that there is a decrease in absorbance of H_2O_2(hydrogen peroxide) upon oxidation of H_2O_2. A solution of 43 mM H_2O_2 was prepared in 0.1M phosphate buffer (Ph 7.4). The test sample at various concentrations in 3.4mL phosphate buffer was added to 0.6mL of H_2O_2 solution (43mM) and absorbance of the reaction mixture was recorded at 230 nm. Sodium phosphate buffer without H_2O_2 the blank solution.

DPPH radical scavenging activity:

The potential AA of extracts, fractions and pure compounds was determined on the basis of the scavenging activity of the stable 1, 1 -diphenyl-2-picrylhydrazyl (DPPH) free radical. Aliquots of 1ml of a methanolic solution containing each pure compound were added to 3ml of 0.004% MeOH solution of DPPH. Absorbance at 517 nm, against a blank of methanol without DPPH, was determined after 30 min (UV, Perkin-Elmer-Lambda 11 spectrophotometer) and the percent inhibition activity was calculated [8]. IC50 values denote the concentration of sample required to scavenge 50% DPPH free radicals. All tests were run in triplicate and averaged.

Nitric oxide radical scavenging activity:

Nitric oxide generated from sodium nitroprusside in aqueous solution at physiological pH interacts with oxygen to produce nitrite ions, which were measured by the Griess reaction [9]. The reaction mixture (3 ml) containing sodium nitroprusside (10 mM) in phosphate buffered saline (PBS) and IBPG and the reference compound in different concentrations (20, 40, 60, 80 and 100 µg) were incubated for 150 min at 25°C. After incubation 1.5ml of the Griess reagent (1% sulphanilamide, 0.1% naphthyl ethylene diamine dihydrochloride in 2% H3PO4) was added. The absorbance of the formed chromophore was measured at 546nm. The percentage inhibition was measured by comparing absorbance values of control and test samples.

Table No. 1: Inhibitory concentration (IC50) values of selected fruit peels in *in vitro* antioxidant models

Method	IC50 (µg)				
	APAS	APAD	APCM	APMP	Rutin
Hydroxyl radical scavenging activity	50.24±0.76	46.00±0.24	74.35±0.22	26.69±0.19	35.55±0.84
Hydrogen peroxide radical scavenging activity	4.99±0.49	3.52±0.71	3.53±0.58	3.24±0.59	3.77±0.61
Nitric oxide radical scavenging activity	45.75±0.47	37.13±0.61	47.09±0.39	23.83±0.49	11.38±0.72
DPPH radical scavenging activity	127.92±0.56	149.59±0.81	181.84±0.51	114.02±0.64	88.79±0.70

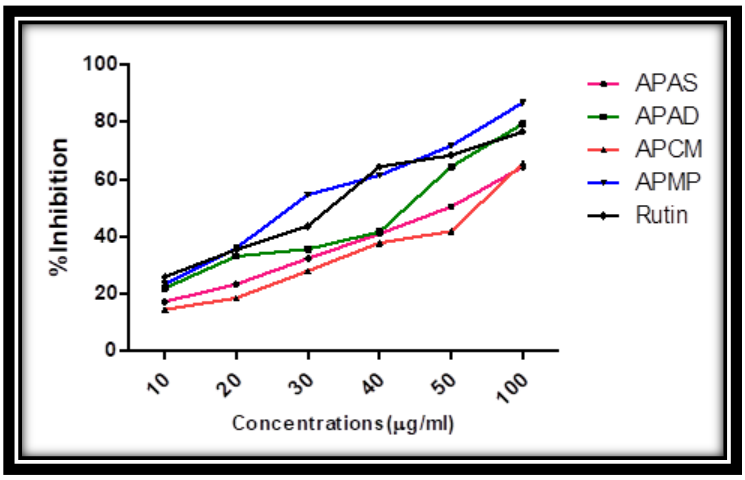

Fig. 1: Effect of fruit peels and Rutin on hydroxyl radical scavenging activity

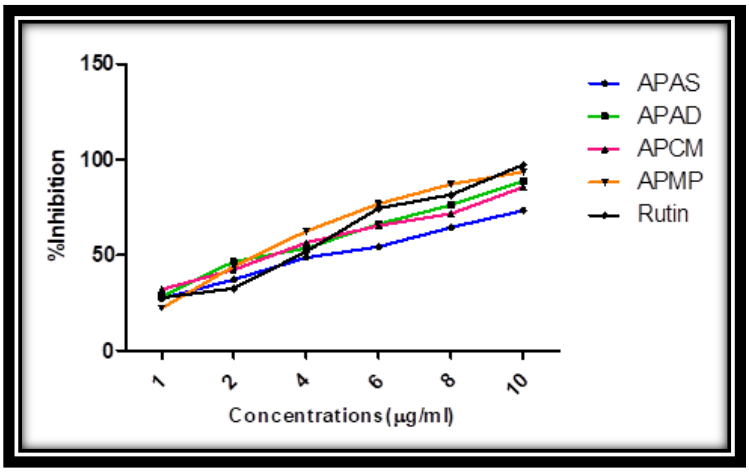

Fig. 2: Effect of fruit peels and Rutin on hydrogen peroxide scavenging activity

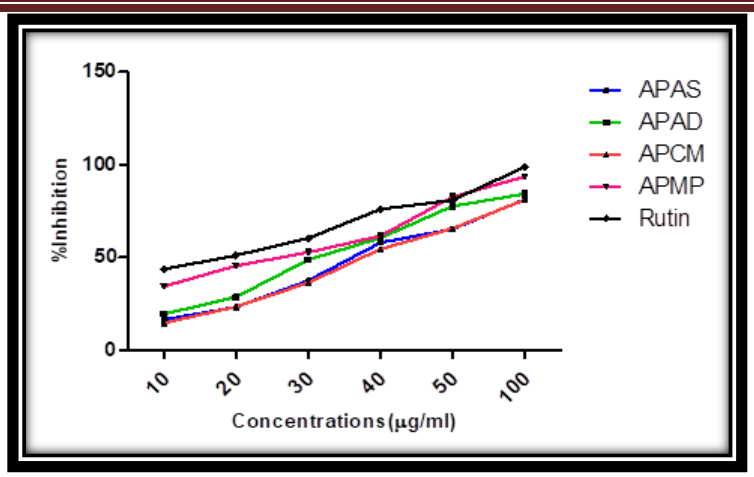

Fig. 3: Effect of fruit peels and Rutin on Nitric oxide radical scavenging activity

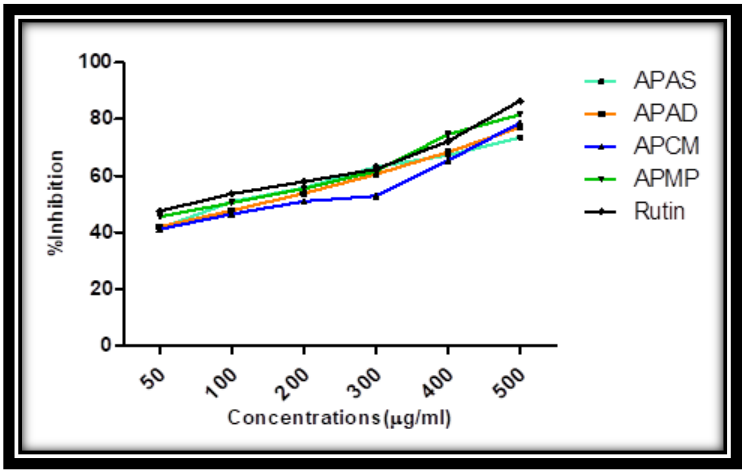

Fig. 4: Effect of fruit peels and Rutin on DPPH radical scavenging activity

RESULTS AND DISCUSSION

The loss of balance between the generation of free radicals and antioxidant enzymes in human body is a consequence is called as oxidative stress [10]. Several research supporting that the plant based drugs were reported to cure chronic metabolic diseases acts by protecting the body defense system and by acts against the free radicals [11]. To act against the both enzymatic and nonenzymatic source of free radicals, antioxidants need to protect organisms from inflammation caused by excessive generation of ROS like superoxide, hydroxyl, hydrogen peroxide and nitric oxide.

Oxidative stress was defined as the lack of balance between the occurrence of reactive oxygen/nitrogen species and the organism's capacity to counteract their action by the antioxidative protection systems.

The human body possesses defense mechanisms against free radical-induced oxidative stress, which involve preventative mechanisms, repair mechanisms, physical defense and antioxidant defenses. Super oxide dismutase (SOD), Glutathione peroxidase (GPx), Catalse (CAT) are some of the enzymatic antioxidant defenses. Ascorbic acid (Vitamin C), α-tocopherol (Vitamin E), glutathione (GSH), Carotenoids, Flavonoids etc are non enzymatic antioxidants.

Recently, peels are gaining more popularity due to its phytoconstituents and wide range of biological activities comparing with other parts of peels. The peels occupy almost 30% total weight of the fruits and utilized as a by-products of fruits [12]. The activity might be due to the presence of phenolics compounds, carotenoids, vitamins, and terpenoids. These compounds have potency to scavenging the free radical in order to reduce the development of oxidative stress in many chronic diseases [13]. Hence the present investigation was designed to evaluate the antioxidant activity of selected fruit peels such as *Annona squamosa* (APAS), *Actinedia deliciosa* (APAD), *Cucumis melo* (APCM) and *Mallus pumila* (APMP).

The Hydroxyl radicals are interacts with the tissue lipids and leads to peroxide reactions by abstracting the hydrogen atoms from the membranes and involved in metabolic functions and lipid peroxidation [14] . The hydrogen donating capability of the plant extracts are able to inhibit the hydroxyl radical formation by terminating the peroxide reactions. These radicals interacts with macromolecules and damage the normal functioning of carbohydrates, proteins, lipids and nucleic acid by mutations and by the formation of lipid peroxidation [15] Hydroxyl radical-scavenging activity of the selected fruit peel extracts was investigated using the Fenton reaction mechanism. The activity of APMP was found to be more among the selected fruit peel extracts in scavenging hydroxyl radicals in a concentration dependent manner (graph 1). The order of potency in scavenging the hydroxyl radical was APMP>Rutin>APAD>APAS>APCM. The APMP showed significant percent of inhibition when compared with standard rutin and other peels.

The hydrogen peroxide radical are generally originated from the hydrogen peroxide. The H_2O_2 radical is not much toxic but the product hydroxyl radical is toxic. The H_2O_2 scavenging of the reported plant extracts might be attributed to their phenolics and reported to have electron donating capability there by it neutralizes the water molecule. The activity of APMP was found to be more among the selected fruit peel extracts and standard rutin in scavenging hydrogen peroxide radical in a concentration-dependent manner (Fig. 2). The order of potency in scavenging the hydrogen peroxide was APMP > APAD > APCM > Rutin > APAS. The APMP showed significant percent of inhibition when compared with standard rutin and other selected fruit peel extracts.

The product of nitrogen stress is nitric oxide radical and it is unstable and it interacts with the oxygen and generates the both nitrate and nitrite by the formation of NO_2, N_2O_4. It is easily react with superoxide anion to form peroxynitrile, a potent oxidizing molecule [16]. It makes the production of tissue damage by the process id lipid peroxidation by cellular damage and inflammation. The activity of APMP was found to be more among the selected fruit peel extracts and standard rutin in scavenging nitric oxide radical in a concentration-dependent manner (Fig. 3). The order of potency in scavenging the nitric oxide radical was Rutin > APMP > APAD > APAS > APCM. The APMP showed significant percent of inhibition when compared with standard rutin (table 1).

The DPPH is a exogenous free radical and the DPPH radical scavenging activity works on the principle that conversion of blue colored diphenylpicrylhydrazyl to yellow-colored diphenylpicryl hydrazine. The phytochemicals with the hydrogen donating capability showed significant DPPH radical [17]. The activity of APMP was found to be more among the selected fruit peel extracts and standard rutin in scavenging DPPH radical. The extracts were capable of scavenging DPPH radical in a concentration-dependent manner (Fig. 4). The order of potency in scavenging the DPPH radical was Rutin > APMP > APAS > APAD > APCM. The APMP showed significant percent of inhibition when compared with standard rutin (table 1).

CONCLUSION

However the potential application of fruit peels in food supplementation depends strongly on their chemical composition and its antioxidant power. In the present study resulted that the selected peels showed significant antioxidant activity against both exogenous and endogenous free radicals in a concentration dependent manner. Among all the selected peels the *Mallus pumila* showed better antioxidant activity among selected peel powders. The study suggested that the peels are not waste products to discard, it also one of the part that might contains high levels of phytochemicals reported to possess many pharmacological activities.

REFERENCES:

1. Aqil F, Ahmed I, Mehmood Z. Antioxidant and free radical scavenging properties of twelve traditionally used Indian medicinal plants. *Turk J Biol* **2006;**30:177-183.
2. Jayaprakasha GK, Jaganmohan Rao L, Sakariah KK. Antioxidant activities of flavidin in different *in vitro* model systems. *Bio org Med Chem* **2004**;12:5141-5146.
3. Halliwell B. How to characterize an antioxidant: an update. *Biochem Soc Symp* **1995**;61:73-101.
4. Liyana-Pathirana CM, Shahidi F and Alasalvar C. Antioxidant activity of cherry laurel fruit (Laurocerasus officinalis Roem.) and its concentrated juice. *Food Chem* **2006**;99:121-128.
5. Chaojin W and Qinghua L. Advances in comprehensive utilization of fruit peel in China, *Acad J Agri Res* **2016**;4(9):589-592.
6. Halliwell B, Gutteridge JMC. & Aruoma OI. The deoxyribose method: A simple test tube assay for determination of rate constants for hydroxyl radicals. *Anal Biochem* **1989**;165:215–219.
7. Ruch RJ, Cheng SJ, Klaunig JE, Prevention of cytotoxicity and inhibition of intracellular communication by antioxidant catechins isolated from Chinese green tea, *Carcinogenesis* **1989**; 10:1003–1008.
8. Braca A, Tommasi N, Bari LD, Pizza C, Politi M, Morelli I. Antioxidant principles from Bauhinia terapotensis. *J Nat Prod* **2001**;64:892–895.
9. Ebrahimzadeh MA, Nabavi SF. In vitro antioxidant and free radical scavenging activity of Leonurus cardiaca subsp. Persicus, *Grammosciadium platycarpum* and Onosma demawendicum. *Afr J Biotech* **2010**;9:8865-8871.
10. Abuashwashi MA, Palomino OM, Gómez-Serranillos MP. Geographic origin influences the phenolic composition and antioxidant potential of wild Crataegus monogyna from Spain. *Pharm Biol* **2016**;54(11):2708–2713.
11. Zeidan S, Hijazi A, Rammal H, Al Bazzal A, Annan H and Al-Rekaby AAN. Determination of bioactive compound content and antioxidant activity of the Labanese Eryngium creticum. *Eu Chem Bul* **2015**;4(11):498-502.
12. Moon JK and Shibamoto T. Antioxidant assays for plant and food components. *J Agri and Food Chem* **2009;**57:1655-1666.
13. Khansari N, Shakiba Y, Mahmoudi M. Chronic inflammation and oxidative stress as a major cause of age-related diseases and cancer. *Recent Pat Inflamm Allergy Drug Discov* **2009**;3(1):73-80.
14. Massey KA, Nicolaou A. Lipidomics of polyunsaturated-fatty-acid-derived oxygenated metabolites. *Biochem Soci Transact* **2011**;39(5):1240–1246.
15. Fukuda T, Ito H. & Yoshida T. Effect of the walnut polyphenol fraction on oxidative stress in type 2 diabetes mice. *BioFactors* **2004**;21:251–253.
16. Carolina P, María NÁ and Rafael R. Peroxynitrite, a potent macrophage-derived oxidizing cytotoxin to combat invading pathogens. *Biofactors* **2014**;40(2):215–225.
17. Barreira JCM, Ferreira ICFR, Oliveira MBPP, Pereira JA. Antioxidant activities of the extracts from chestnut flower, leaf, skins and fruit. *Food Chem* **2008**;107:1106-1113.

How to cite this article:

Gana Manjusha K, Dr. Ganga Rao B. SCREENING OF IN VITRO ANTIOXIDANT ACTIVITY OF SELECTED FRUIT PEELS: A COMPARATIVE STUDY. J Pharm Res 2018;7(10):227-230. **DOI: https://doi.org/10.5281/zenodo.1471138**

Conflict of interest: The authors have declared that no conflict of interest exists.

Source of support: Nil