

Investigation on phytochemical profile of *Citrus limonum* peel extract

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Abstract

In the present day scenario perishable fruit peels are considered as a new era of pharmaceutical products as they are rich in phytochemicals and act as antioxidant agents. Lemon peel which is a good source of several bioactive compounds is usually discarded or thrown away after extracting the juice from it. The present study was carried out to determine the presence of various phytochemicals such as terpenoids, alkaloids, glycosides, phytosterols, diterpenes, phenols, tannins, flavonoids and saponins in lemon peel. Secondary derived plant metabolites such as phenols, flavonoids, diterpenes, phytosterols and cardiac glycosides were present in both methanol and acetone extract while saponins was present only in methanol extract. The presence of various phytoconstituents was further confirmed using thin layer chromatography technique. Hence the result of the present study indicates the use of lemon peel in various value added products thereby benefitting several industries.

Keywords: lemon peel, bioactive compounds, pharmaceutical agent

1. Introduction

According to the World Health Organization (WHO) medicinal plants are considered to be the best source to obtain a variety of drugs. India is the largest producer of medicinal herbs and is appropriately called the botanical garden of world [1]. Citrus fruits have long been utilized as traditional Asian medicine for centuries as they are considered to be a rich source of secondary metabolites which possess a broad spectrum of biological activities [2]. Phytochemicals accumulate in different parts of the plants, such as in the roots, stems, leaves, flowers, fruits or seeds particularly most of them are often concentrated in the outer layers of the various plant tissues. The levels may vary depending upon the variety, processing, cooking and growing conditions [3].

Citrus fruits such as orange, lemon and lime have been cultured widely and processed into juices. During the manufacture of citrus juice, citrus peels are discarded as waste products. Citrus flavonoids have a broad spectrum of biological activity such as antimicrobial, antioxidant, anti-diabetic and anticancer properties [4]. Lemon which belongs to Rutaceae family is a rich source of vitamin C, alkaloids, flavonoids and essential oils which have antimicrobial and anticancer property. Studies showed that essential oils, protopine, corydaline, alkaloids, limonoids, lactons, polyacetylene, acyclic sesquiterpenes, hypericin and pseudohypericin compounds present in *Citrus limonum* is responsible for its therapeutic properties [5].

In the present day scenario attention has been focused on industrial wastes, especially those containing residual phenols from the used plant raw material. Keeping in view the medicinal and therapeutic values of *Citrus limonum*, the present study was carried out in an effort to make beneficial use of peel which is usually thrown out as waste.

2. Materials and Methods

2.1 Plant material and preparation of extract

Fresh lemons were purchased from a local market in Chennai and washed thoroughly under tap water. The peels were

removed using a sterile knife and shade dried for 4-5 days at room temperature. The dried peels were pulverized using an electric blender and stored in airtight containers for further use. Two different solvents namely methanol and acetone were used for extraction. 5grams of lemon peel powder was soaked in 100 mL of the respective solvents for 72 hours by maceration technique. The supernatant was filtered using Whatmann filter paper 1 and Buchner funnel and concentrated using rotary evaporator and dry residue was preserved at 5°C until further use. The percentage yield obtained was calculated using the following formula:-

$$\% \text{ yield of extract} = \frac{\text{Extract weigh}}{\text{Dry weight powder}} \times 100$$

2.2 Qualitative phytochemical screening

Phytochemical screening was carried out to analyze the presence of various phytoconstituents such as alkaloids, glycosides, saponins, phenols, flavonoids, terpenoids, steroids, carbohydrates and protein according to standard method described by (Mace [6]; Wagner [7]; Wagner *et al.* [8]; Evans [9]; Kokate [10]). The presence of carbohydrates and proteins was also tested according to standard method given by Ramakrishnan *et al.* [11]). General reactions in this analysis revealed the presence or absence of these compounds.

2.3 Thin Layer Chromatography

The thin layer chromatography method of the sample spotting was used to determine the R_f value of the separated compounds. TLC plate was prepared using silica gel G and was left overnight for drying. Aliquots of the extracts were applied on the TLC plates which was 0.2 mm above from the bottom with the help of a capillary tube. Then the plate was placed in a glass beaker containing solvents such as toluene: ethyl acetate: acetic acid as mobile phase for acetone extract. The solvents used as mobile phase for methanol extract was ethyl acetate: toluene. The zone of spot in the chromatogram corresponding to the band was noted. The R_f value was calculated using the following formula:-

$$R_f = \frac{\text{Distance travelled by the solute}}{\text{Distance travelled by the solvent}}$$

3. Results and Discussion

The percentage yield of extracts presented in Table 1 reveals that

the acetone extract had the lowest percentage yield (5.8%) while methanol extract had the highest yield (16.8%). The difference in percentage yield could be due to several reasons such as the solvent used for extraction, mode of extraction and time period required for extraction.

Table 1: Yield (%) of extracts of *Citrus limonum* peel

Extract	Yield (%)
Methanol extract	5.8
Acetone extract	16.8

The results of phytochemical analysis in Table 2 indicates that both the extracts showed the presence of various secondary plant metabolites such as phenols, flavonoids, diterpenes, phytosterols and cardiac glycosides while saponins was present only in methanol extract. On the other hand, alkaloids, tannins and glycosides were absent in both the extracts. Matthew *et al.* [12] reported that ethanolic and aqueous extracts of lemon peel and lemon pulp also showed the presence of various phytochemicals such as alkaloids, phenols, flavonoids, diterpenes, cardiac glycosides, tannins and saponins. Kumar *et al.* [13] also reported the presence of various phytochemicals in ethanolic extract of

lemon peel. As citrus peels are rich in phytonutrients they are used in drug production or as food supplements [14].

Phytochemicals (derived from the Greek word *phyto*, meaning plant) are non-nutritive biologically active, naturally occurring chemical compounds found in plants, which imparts health benefits such as antioxidant activity, antimicrobial effect, modulation of detoxification enzymes, stimulation of the immune system, cholesterol lowering ability, decrease of platelet aggregation, modulation of hormone metabolism and anticancer property [15, 12].

Table 2: Phytochemical screening of *Citrus limonum* peel

S. No.	Phytochemicals	Tests performed	Methanol extract	Acetone extract
1	Alkaloids	Mayer's test	-	-
		Wagner's test	-	-
		Hager's test	-	-
2	Saponins	Froath test	+	-
		Foam test	+	-
3	Phenols	Ferric chloride test	+	+
4	Tannins	Gelatin test	-	-
5	Flavonoids	Alkanine reagent test	+	+
		Lead acetate test	+	+
6	Diterpenes	Copper acetate test	+	+
7	Glycosides	Modified bortrager's test	-	-
8	Phytosterols	Salkowshi's test	+	+
9	Cardiac glycosides	Legal's test	+	+
10	Carbohydrates	Fehling's test	-	-
11	Proteins	Xanthoproteic test	-	-

The presence of various phytoconstituents was further confirmed using thin layer chromatography technique. Thin layer chromatography is commonly used in the detection of compounds through a separation process. The number of spots observed and their corresponding R_f values is shown in Figure 1 and 2 and presented in Table 3. Five spots with R_f values 0.97,

0.91, 0.85, 0.73 and 0.67 were observed for acetone extract of lemon peel while five spots with R_f values such as 1, 0.87, 0.33, 0.22 and 0.11 were observed for methanol extract respectively. Kumar *et al.* [13] reported the presence of β carotene in lemon peel with a R_f value of 0.91. The obtained result of β carotene was also matched with standard.

Table 3: Thin Layer Chromatography

Compounds observed under UV @ 235nm	Acetone extract R_f value	Methanol extract R_f value
1	0.97	1
2	0.91	0.87
3	0.85	0.33
4	0.73	0.22
5	0.67	0.11

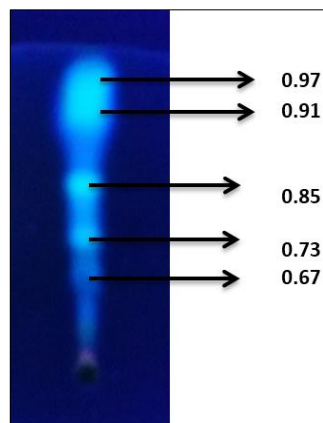


Fig 1: TLC of isolated compounds in UV of acetone extract of lemon peel

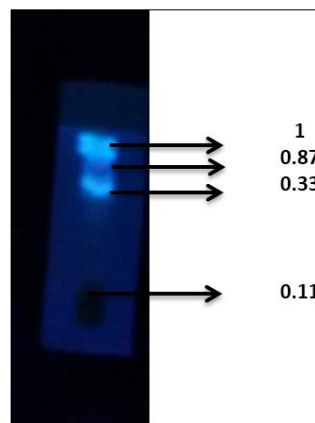


Fig 2: TLC of isolated compounds in UV of methanol extract of lemon peel

4. Conclusion

The study reveals that lemon peel is a rich source of active compounds with various medicinal and pharmacological properties making them to be utilized as an attractive, alternate and cheap source of functional ingredients for the formulation of functional foods and nutraceuticals. Further studies are also required to unravel and characterize active components present in lemon peel.

5. References

- Shariff N, Sudarshana MS, Umesha S, Hariprasad P. Antimicrobial activity of Rauvolfia tetraphylla and Physalis minima leaf and callus extracts. African Journal of Biotechnology. 2006; 5(10):946-50.
- Kalpa S, Mahinda S, Won-WL, Young-TK, Jae IK, Myung CO. Antibacterial effect of citrus press-cakes dried by high speed and far-infrared radiation drying methods. Nutrition Research and Practice. 2012; 6(3):187-194.
- King A, Young G. Characteristics and Occurrence of Phenolic Phytochemicals. Journal of the American Dietetic Association. 1999; 24:213-218.
- Olaniyan AM. Development of a small scale orange juice extractor. Journal of Food Science and Technology. 2010; 47:105-108.
- Maruti JD, Chidamber BJ, Jai SG, Kailash DS. Study Antimicrobial Activity of Lemon (*Citrus lemon* L.) Peel Extract. British Journal of Pharmacology and Toxicology. 2011; 2(3):119-122.
- Mace ME. Histochemical Localization of phenols in healthy and diseased tomato roots. Phytopathology. 1963; 16:915-925.
- Wagner H. Pharmazeutische Biologie 5th edition AUFI. 15 BN 3-437-20 498-X.Gustav fisher Vwelog. Stuttgart. Germany. 1993.
- Wagner HXS, Bladt Z, Gain EM. Plant drug analysis. Springer Veralag. Berlin. Germany. 1996.
- Evans WC. Trease and Evans Pharmacology 14th edition. Harcourt Brace and company. Asia. Pvt. Ltd. Singapore. 1997.
- Kokate CK. Practical pharmacognosy.4th edition. Vallaph prakashan publication, New Delhi, India. 1999.
- Ramakrishnan SK, Prasannan G, Rajan R. Textbook of medical biochemistry. Orient Longman, New Delhi. India. 1994, 582.
- Mathew B, Jatawa SK, Tiwaari A. Phytochemical analysis of *Citrus limonum* pulp and peel. International Journal of Pharmacy and Pharmaceutical Sciences. 2012; 4(2):269-371.
- Kumar SP, Pushpak A, Shilpi M, Anup K, Shraddha V. Phytochemical analysis of *Citrus limonum* and *Citrus sinensis* peels and identification of β carotene pigment using ethanolic extract. International Research Journal of Pharmacy. 2014; 5(10):789-791.
- Chede PS. Phytochemical analysis of *Citrus sinensis* peel. International Journal of Pharma and Bio Sciences. 2013; 4(1):339-343.
- Savithamma N, Linga RM, Suhrulatha D. Screening of Medicinal Plants for Secondary Metabolites. Middle East Journal of Scientific Research. 2011; 8(3):579-584.