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Comparative phytochemical analysis and total phenolic content in citrus fruit peel (*Citrus sinensis* and *Citrus limon*)

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Article History:	ABSTRACT (Deck for updates)	
Received on: 11.01.2018 Revised on: 08.04.2018 Accepted on: 12.04.2018	Plants are valuable source of natural products for maintaining health in hu- man beings. Phytochemicals present in plant extracts have potential antioxi- dant, antimicrobial properties which may be useful in therapeutic applica- tions. This study aims to compare and analyse the phytochemicals and esti-	
Keywords:	mate the phenolic content in two varieties citrus fruit peel namely sweet of ange (<i>Citrus sinensis</i>) and lemon (<i>Citrus limon</i>). The peels of <i>Citrus sinen</i> .	
Antibacterial, Anticancerous, Citrus fruits, Phytochemical Analysis, Phenolic Content	and <i>Citrus limon</i> were collected and the total phenolic content was calculated using Follin ciolcaletu's reagent. The phytochemicals such as phylobatannins, carbohydrates, flavonoids, alkaloids and terpenoids were present in the cit- rus fruit peels extract. The amount of terpenoids was found in greater amount in lemon peel than in orange peel. It was also found that alkaloids were present in greater amount than the other phytochemicals in orange and lemon peel. Preliminary photochemical analysis and estimation of total phe- nolic content was done in Citrus fruit peel. Thus, it is established that orange and lemon peel can be used medicinally for their antibacterial and anti can- cerous activities due to the presence of significant amount of alkaloids in them.	

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INTRODUCTION

Resistance of microbes to synthetic drugs has proportionally increased to the production of large number of new antibiotics in recent years. The microbes are having the genetic ability to transmit and gain drug resistance, which are used in therapeutic applications (Seenivasan *et al.*, 2006). Plants are valuable source of phytonutrients for maintaining the health of mankind. Medicinal plants are the most important sources for the manufacture of different drugs due to the presence of phytochemicals which are required for therapeutic applications due to their structural diversity and biological functionality (Gislene et al., 2000). Herbal extracts and phytochemicals, with antioxidant, antimicrobial activities may have therapeutic significance (Burt, 2004). Phytochemicals prevent the substances we intake, from becoming carcinogenic and stimulate the damaged cells to undergo apoptosis before they can reproduce. It also regulates hormones, prevents DNA damage and also slows growth rate of cancer. Thereby, reducing the risk of cancer and other major chronic diseases (Buckley et al., 2013). Plants possess phenolic constituents which has antioxidant, anticarcinogenic and antimutagenic properties. Phenolics are the largest group of phytochemicals responsible for antioxidant activity in plants and plant extracts (Salih and Abass, 2003).

Citrus fruits are used by humans for medicinal, herbal and agricultural reasons from time immemorial. Citrus fruits, belonging to the family of Rutaceae, are one of the main fruit tree crops grown globally (Okwu and Emenike, 2006, Pultrini *et al.*, 2006). All citrus fruits share in common their sweet and sour flavour. Flavonoids present in Citrus fruits have activities such as antibacterial, antifungal, antidiabetic, anticancer and antiviral potential (Manoharan et al., 2016). The citrus fruit peels are rich in phytonutrients, they can be used as drugs and food supplements (Ahmad et al., 2006). The Citrus fruit peel is rich in flavanones and many polymethoxylated flavones (Shahnah et al., 2007). Extract preparation of the peel, flowers and leaves of orange (Citrus aurantium L) are used to minimise disorders associated with central nervous system. The World Health Organization survey estimated that nearly 80% of the population of developing nations depend on traditional medication in primary health centres (WHO, 2002). The citrus fruit peel is a rich source of flavonoids glycosides, coumarin, beta and gamma-sitosterol, glycosides and volatile oils (Mamta Arora and Parminder Kaur, 2013).

The orange fruit (*Citrus sinensis*) is highly nutritious and rich in minerals, proteins, carbohydrates, and fat. The peel of the fruit constitutes about 10% it's total weight. Lemon (*Citrus limon*) is an important medicinal plant of the family Rutaceae. The antibacterial property in lemon extracts act against clinically significant bacterial strains (Kawaii *et al.*, 2000). The objective of this research was to evaluate the phytochemical constituents and to estimate the total phenolic content in citrus fruit peel.

MATERIALS AND METHODS

Extract Preparation

The peels of the Citrus fruits (orange and lemon) were collected and dried for a week and were crushed and powdered. An aqueous extract was prepared.

Phytochemical Analysis

1. Test for phylobatannins: To 1% hydrochloric acid 10 ml of sample was added and boiled. Formation of red coloured precipitate confirmed a positive result.

2. Test for carbohydrates:

- (a) Fehling's test: To 1ml of Fehling's A and Fehling's B reagent, 1ml of extract was added and boiled for three minutes. Red colour precipitate was formed which confirms the presence of carbohydrates in the extract.
- **(b) Benedict's test:** 1ml of the extract was boiled with 1ml of Benedict's reagent. Reddish brown precipitate was observed which confirmed carbohydrates in the extract.

3. Test for flavonoids: Sample was added to few drops in 1% liquid ammonia in a test tube. Yellow

colouration was observed which confirmed the presence of flavonoids.

4. Test for alkaloids: To 2ml of extract, add 2ml of hydro chloric acid, few drops of picric acid and 6 drops of hexane. A pale yellow precipitate indicated the presence of alkaloids.

5. Test for terpenoids: To 5ml of extract, add 2ml of chloroform and 3ml of concentrated sulphuric acid. A reddish brown ring was observed which denoted the presence of terpenoids.

Estimation of total phenolic content

Total phenolic content of both orange and lemon peel was calculated using Folin ciolcaletu's reagent against Gallic acid standard.

RESULT AND DISCUSSION

The preliminary phytochemical test indicates the presence of phylobataninns, carbohydrates, flavonoids, alkaloids and terpenoids. The study shows the presence of phytochemicals such as phylobatannins, carbohydrates, flavonoids, alkaloids and terpenes (Table 1). The phytochemicals which are frequently associated with human health are carotenoids, polyphenols and tocopherols. The comparison of the total phenolic content was also studied (Table 2) and it was found that there was a slight variation only. Bioactive constituents of plants are non-nutritive and possess specific chemical structure that help in disease prevention and antimicrobial activities and also work with dif- ferent mechanism of action (Aveen Nozad, 2005).

Table 1: Phytochemical analysis

	Orange	Lemon
	peel	peel
Test for phylobatannins	+	+
Test for carbohydrates-	++	+++
Fehling's test		
Test for carbohydrates-	++	++
Benedict's test		
Test for flavonoids	++	++
Test for alkaloids	+++	+++
Test for terpenoids	+	++

Table 2: Total phenolic contentExtractTotal phenolic contentOrange peel42 mg GAE/gmLemon peel48 mg GAE/gm

It is evident that orange and lemon peel have significant amount of alkaloids. Alkaloids and basic (alkali-like), nitrogen contains compounds mainly occurring in plants. Alkaloids and extracts of alkaloid containing plants are being used as remedies for poisons and psychoactive drugs for a long period of time (Panda and Bandyopadhyay, 2013). Alkaloids may act against the cause of an illness, or they may merely alleviate individual as caffeine acts as a stimulant. The majority of alkaloids are valuable medicines but many are powerful poisons as well. Flavanoids act as a very good antioxidant and anti-inflammatory substances whereas the alkaloids protect against chronic diseases (Rister and Bachner, 1976, Gayathri and Anuradha, 2015). Lemon is grown mainly for its alkaloids, which has anticancerous activities. The antibacterial activity in crude extracts of different parts (viz., leaves, stem, root and flower) of lemon against clinically significant bacterial strains has been studied (Kawaii *et al.*, 2000).

Thus, this study establishes that orange and lemon peel can be used medicinally for their antibacterial and anticancerous activities due to the presence of significant amount of alkaloids in them. The study also establishes that lemon peel has higher amount of terpenoids in them when compared to orange peels. Oxygenated derivatives of terpenes constitute terpenoids, which may be acyclic, cyclic, saturated or unsaturated organic hydrocarbon molecules (Balakrishnan et al., 2015). Terpenoids such as neral or geranial (E and Z isomer o citral respectively) and terpenes such as limonene may be found in large quantities in the oil sacs located in the outer, coloured portions of the rinds of many Citrus fruits (Smith et al., 2001). Terpenoids acts as anti inflammatory, expectorant, bronchodilator and local antiseptic.

Citrus fruit extracts and citrus flavonoids exhibit significant biological properties due to their phenolic content and phytonutrients which possess antioxidant properties (Menon *et al.*, 2016).

CONCLUSION

The preliminary phytochemical analysis indicates the presence of phylobatannins, carbohydrates, alkaloids, terpenes and flavonoids. Further, the presence of significant amount of alkaloids in orange and lemon peel exhibit its anticancer property. Phytochemicals prevent the substances we eat, drink and breathe, from becoming carcinogenic and stimulate the damaged cells to undergo apoptosis and also regulates hormones, prevent DNA damage and slows growth rate of cancer. Phenols give them antioxidant properties and various health benefits.

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