PHYTOCHEMICAL AND ANTIMICROBIAL ACTIVITY OF DIFFERENT CITRUS FRUIT PEELS

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ABSTRACT
Amongst the largely produced fruits citrus fruits, rank first in fruit industry. They are a very good source of essential oils, phytochemicals and nutrients. Due to the presence of different phytochemicals the citrus fruits show anti-tumor, anti-inflammatory and anti-microbial properties. In the present study dried peels of different citrus fruits (Citrus sinensis, Citrus limon and Mangifera indica) were used. The phytochemicals were extracted using ethanol, acetone, water, petroleum ether and ethyl acetate. The ethanolic extract was further used for determining anti-microbial activity as maximum number of phytochemicals were extracted using ethanol. The anti-microbial activity was checked against E. coli and B. subtilis. It was found that all the plants show anti-microbial activity against the used cultures with M. indica giving the highest zone of inhibition (6-7 mm).

KEYWORDS: Citrus Fruits, Phytochemicals, Anti-Microbial Activity.

INTRODUCTION
Citrus fruits are the most abundant fruit produced worldwide contributing 100 million tons annually to the fruit industry (Jwanny et al., 2012). Citrus fruits peels are the most familiar and rich source of nutrients, essential oils and phytochemicals (Tao et al., 2008) due to which they exhibit anti-microbial, anti-tumor, anti-oxidative and anti-inflammatory properties (Aruoma et al., 2012; Karimi et al., 2012). Therefore, these can be efficiently used as drugs or as food supplements too (Nychas et al., 1995; Sokovic et al., 2007). The peel of citrus fruit is rich in flavonones, glycosides and many polymethoxylated flavones which are very rare in other plants (Ahmad et al., 2006). Citrus peels have been used due to their anti-microbial properties produced by compounds synthesized in the secondary metabolism of the plant such as phenolic compounds which are a part of the essential oils, as well as tannin (Tyagi and Malik, 2010; Oikeh et al., 2016). Another reason behind using plant extracts as anti-microbial agents is that they do not show side effects like synthetic anti-microbials and micro-organisms so not develop resistance to them. Hence, the phytochemical extracts of plants have evoked interest as alternative remedies for the treatment of many infectious diseases (Parekh et al., 2005). Citrus peels have been known to relieve various diseases. The use of plant extracts and phytochemicals, both with known anti-microbial properties, can be of great significance in therapeutical treatments (Seenivasan et al., 2006). There are numerous advantages of secondary metabolites of plants. In order to evaluate the presence of bioactive components from peel extracts, the present study has been planned. The present investigation has been undertaken to screen the presence of phytochemicals extracted from peels of various citrus fruits (Citrus sinensis, Citrus limon and Mangifera indica).

MATERIALS AND METHODS
Preparation of Solvent Peel Extract
The peels of the citrus fruits (C. sinensis, C. limon and M. indica) were collected, shade dried and powdered. Solvents like acetone, ethanol, petroleum ether, ethyl acetate, methanol, aqueous, hexane and water were used for the preparation for extracts.

Phytochemical Screening
The extracts were subjected to preliminary phytochemical screening following the methodology of Kokate (2001) with slight modifications. The extracts were subjected to various phytochemical tests to determine the active constituents (alkaloids, carbohydrates, proteins, tannins, steroids, diterpenes, phytosterols, saponins, flavanoids, anthraquinones, phlobatannins, terpenoids, chalcones and phenols) present in the crude extracts obtained with different solvents.

Antibacterial Activity
The sterile agar plates were inoculated with Escherichia coli and Bacillus subtilis. Anti-microbial activity of various fruit peel extracts were tested by using agar well diffusion method. The wells were then filled with about...
100µl of plant extract. The plates were then incubated at 37°C for 24 hrs. After incubation, zone of inhibition was measured.

RESULTS AND DISCUSSIONS
The phytochemical analysis of citrus fruit peels in different solvents is listed in table 1. Aqueous extract of C. limon (lemon) peels showed the presence of alkaloids, reducing sugars, tannins, steroids, phytosterol, saponins, flavonoids, cardiac glycosides and terpenoids and absence of diterpenes, anthraquinones, phlobatannins, chalcones and phenols. The results were in confirmation with the findings of Dhanavade et al. (2011) who also found presence of alkaloids, carbohydrates, reducing sugars, tannins, steroids, diterpenes, saponins, flavonoids, cardiac glycosides and terpenoids and absence of diterpenes, anthraquinones, phlobatannins, terpenoids, chalcones and phenols were absent in water. On the other hand phlobatanins and terpenoids were present in acetone extract. The results were similar to those found by Mathur et al. (2011).

Aqueous extract of C. sinensis (orange) peels showed the presence of tannins, saponins and flavonoids whereas alkaloids, steroids, diterpenes, phytosterol, anthraquinones, cardiac glycosides, phlobatannins, terpenoids, chalcones and phenols were absent in water. The phytochemical analysis of acetone extract of lemon peels showed the presence of alkaloids, tannins, steroids, flavonoids and phlobatannins (Kumar et al., 2011).

Table 1: Phytochemical analysis of Citric Fruits.

<table>
<thead>
<tr>
<th>Solvents</th>
<th>Plants</th>
<th>Citrus limon(Lemon)</th>
<th>Citrus sinensis (Orange)</th>
<th>Mangifera indica (Mango)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aqueous</td>
<td>Alkaloids, Tannins, Steroids,</td>
<td>Tannins, Saponins, Flavonoids</td>
<td>Tannins, Steroids, Diterpenes,</td>
<td>Tannins, Steroids, Diterpenes,</td>
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<tr>
<td></td>
<td>Phytosterols, Saponins, Flavonoids, Terpenoids</td>
<td></td>
<td>Saponins, Flavonoids, Phlobatannins, Terpenoids</td>
<td>Saponins, Flavonoids, Phenols</td>
</tr>
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<td>Acetone</td>
<td>Alkaloids, Tannins, Steroids,</td>
<td>Tannins, Saponins, Flavonoids,</td>
<td>Alkaloids, Tannins, Steroids,</td>
<td>Alkaloids, Tannins, Steroids,</td>
</tr>
<tr>
<td></td>
<td>Flavonoids, Phlobatannins</td>
<td>Flavonoids, Phlobatannins, Terpenoids</td>
<td>Flavonoids, Phenols</td>
<td>Saponins, Flavonoids, Phenols</td>
</tr>
<tr>
<td>Ethanol</td>
<td>Alkaloids, Tannins, Steroids,</td>
<td>Alkaloids, Tannins, Steroids,</td>
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<tr>
<td></td>
<td>Diterpenes, Phytosterols,</td>
<td>Saponins, Flavonoids, Phlobatannins,</td>
<td>Saponins, Flavonoids, Terpenoids</td>
<td>Saponins, Flavonoids, Phenols</td>
</tr>
<tr>
<td></td>
<td>Terpenoids, Chalcones, Phenols</td>
<td>Terpenoids</td>
<td></td>
<td></td>
</tr>
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<td>Petroleum ether</td>
<td>Alkaloids, Tannins, Steroids,</td>
<td>Alkaloids, Steroids, Diterpenes,</td>
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<tr>
<td></td>
<td>Diterpenes, Saponins, Flavonoids, Terpenoids</td>
<td>Flavonoids, Phlobatannins, Terpenoids</td>
<td>Saponins, Flavonoids</td>
<td>Saponins, Flavonoids</td>
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<tr>
<td>Ethyl acetate</td>
<td>Flavonoids</td>
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<td>Alkaloids, Tannins, Steroids,</td>
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<td></td>
<td></td>
<td>Phenobatannins, Terpenoids</td>
<td>Flavonoids, Phlobatannins</td>
<td>Saponins, Flavonoids, Phenols</td>
</tr>
</tbody>
</table>

Tannins, steroids, diterpenes, saponins, flavonoids, terpenoids and phenols were found in aqueous extract of M. indica (Mango) while alkaloids, phytosterol, anthraquinones, cardiac glycosides, phlobatannins and chalcones gave the negative results. Ethanol was found not to be so active in extracting more amount of phytochemicals and showed the presence of only few phytochemicals including alkaloids, tannins, steroids, diterpenes, flavonoids, terpenoids and phenols. Some findings are in accordance with the Reda et al., 2010 who reported the presence of only alkaloids, tannins, steroids, flavonoids and phenols in ethanolic extract of mango peels.

Anti-microbial Activity
Since maximum compounds were extracted in ethanolic extract therefore, it was used to determine anti-microbial activity. Anti-microbial activity using ethanolic extract of C. sinensis, C. limon and M. indica was determined. All the plants showed activity against B. subtilis (Figure 1) and E. coli (Figure 1). The zone of inhibition for C. sinensis 2-5 mm whereas for C. limon it was 5-6 mm. M. indica showed the highest zone of inhibition i.e. from 6-7 mm. Mathur et al. (2011) checked the anti-microbial activity of ethanol extract of orange, kinnow and shaddock and found that ethanolic extract of orange possessed maximum anti-microbial activity in comparison to other solvent extracts.
CONCLUSION
The extracts from peels of different fruits (C. limon, C. sinensis and M. indica) were recovered and screened for their phytochemical property. Five solvents (acetone, ethanol, ethyl acetate, petroleum ether and water) were used for the extraction of various phytochemical compounds. The phytochemical compounds vary with the species of plants, plant material used and solvent used. The ethanolic extract was used to determine the anti-microbial activity and it was found that all the extracts possess the anti-microbial activity.

REFERENCES