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Citrus sinensis: A potential plant source for antimicrobial activity and value added products

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Abstract
Plants and their by-products have been used for therapeutic purposes from time immemorial. The *Citrus sinensis* (sweet orange) is the most widely farmed and commercialised citrus species. *C. sinensis* fruit is most known for its vitamin C content, but it also contains other phytochemicals such as phenolics and carotenoids that are considered to have health advantages. After peeling the external skin (flavedo), the *C. sinensis* fruit is normally eaten whole or processed into juice. This peeling procedure generates a significant amount of debris. The majority of research has been on citrus fruits, with little attention paid to citrus peels, despite the fact that they have been widely utilised as food ingredients and as a significant herb since ancient times. Citrus peels contain a range of bioactive compounds, especially flavonoids, and they are a significant source of essential oil (EO) and pectin as well as other nutrients. Citrus peel's health-promoting biological effects include antimicrobial, antioxidant, anti-inflammatory, analgesic, and anticancer properties, allowing it to be exploited as a source of functional components and preservatives in the production of nutritionally safe novel foods. *C. sinensis* have demonstrated antimicrobial activity against bacteria, fungus, and multidrug-resistant (MDR) organisms. The researchers developed modern, cost-effective, and time-saving methods for extracting antimicrobial compounds and producing antimicrobial films from the peel of *C. sinensis*. Many novel value-added products are developed from citrus peels, in addition to its medical value. Citrus peel valorisation is beneficial to the environment as well as the bio-economy. Citrus waste management is a rural economy resource that is underutilized. This review summarises the major antimicrobial activities of *C. sinensis*, and possibilities for value addition via waste management measures.

Keywords: *C. sinensis*, bioactive compounds, antimicrobial activity, value-added products, food supply chain

Introduction
The genus Citrus (Rutaceae) is an ancient, widely traded, and popular crop. Its cultivation dates back to 2100 BC [1]. Citrus trees and shrubs are small to medium-sized shrubs or trees that are grown in the tropics and subtropics [2]. Citrus fruits account for 14 % of India's fruit harvest, placing the country sixth in the world among the major fruits grown there (bananas account for 33 %, mangoes for 21 %, papayas for 6 %, guava for 4 %, grape for 3 %, and apple for 3 %) [3, 4]. Orange is a flowering tree that is evergreen. The height of an orange tree is usually between 9 - 10 metres (although very old specimens have reached 15 m). The orange fruit is a hesperidium. Berry varieties vary widely in terms of their size, colour, shape, and the quality of their juice. The fruit shape range in form from globose to ovoid. In addition to having smooth skin, wild orange fruit has whole petiole wings. When compared to the sweet orange, the petioles of sour orange leaves are substantially larger. "Orange" comes from the Sanskrit word "narang." In a typical fruit, there are 11 separate components. The orange fruits have fine texture skin and orange in colour similar to their pulpy flesh. The diameter of an orange is normally between 2 to 3 inches [2]. The orange is a cross between pomelos (*Citrus maxima*) and mandarins (*Citrus reticulata*). Oranges come in two varieties: sweet oranges (*Citrus sinensis*) and sour or bitter oranges (*Citrus aurantium*). In most species, the flowering period is between March and May, the fruiting period is between October and December, and the growth season of late maturing species is between February and April of the following year [2]. Citrus fruits can also be utilised as additives, spices, cosmetic compounds, and chemoprophylactic medications in the food, beverage, cosmetic, and pharmaceutical industries [5, 6]. Several active secondary metabolites from the fruit, peel, roots, leaves, and juice of *C. sinensis* contribute to the plant's medicinal activity namely flavonoids, hydroxyamides, alkanes, steroids, and fatty acids, carbohydrates, coumarins, peptides, alkylamines, and carbamates, volatile compounds, carotenoids, and minerals such as magnesium, potassium, sodium, and calcium [7]. Representative images of *C. sinensis* plant, fruit, and dried peel is shown in Figure 1.

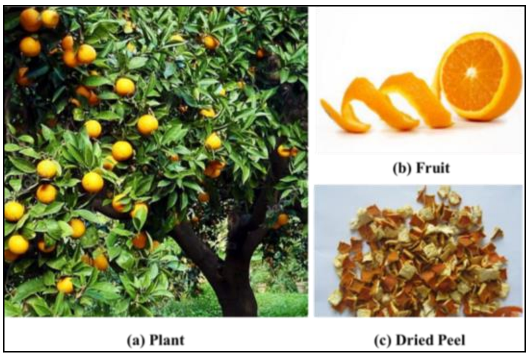

Fig 1: Representative images for *C. sinensis* (a) Plant, (b) Fruit and (c) Dried Peel

The antioxidant, anticancer, anti-ulcer, anti-anxiety, antibacterial, larvicidal, anti-diabetic, and anti-inflammatory activities of *C. sinensis* are among its most important bioactive properties [2].

Citrus by-products are the most important solid derivatives of the citrus processing sector, accounting for approximately 50% of the total weight of fresh fruit produced [8]. They are composed primarily of the peel (60–65 %), internal tissues (30–35 %), and seeds (0–10 %) [9]. Citrus residues have no commercial value, despite the fact that they contain a high concentration of EO, soluble sugars, pectin, cellulose, and hemicellulose, all of which might be used as building blocks in a variety of industrial processes [9]. In view of the massive amount of "wastage" that is generated throughout the food supply chain, orange peels have enormous potential to be utilised as a value-added product, including the recovery of natural antioxidants, enzymes, pectin, or organic acids, EO, the production of ethanol, and prebiotics single cell protein [9]. Strikingly, the most important by-product of citrus processing is EO. Because citrus EOs have been designated as generally regarded as safe, they are widely employed as natural food additives in a variety of food and beverage items [10, 11]. Citrus EOs are also exploited as natural preservatives because of their wide range of biological activities, which include antibacterial and antioxidant properties [12].

Antimicrobial activity of *C. sinensis*

Disc diffusion method was used to demonstrate the antimicrobial activity of EO extract from peels of *C. sinensis*. The results displayed significant antimicrobial activity against *Bacillus subtilis, Escherichia coli, Salmonella typhi, Staphylococcus aureus, Aspergillus flavus*, and *Candida albicans*. Dipentene was identified as the biologically most active antibacterial component using thin layer chromatography-bioautography [13]. Investigators have reported that the orange juice processing waste as a powder is a feasible antimicrobial agent for main food spoilage microorganisms such as *Aspergillus niger*, *Botrytis cinerea, and Penicillium sp.* Without the need of extraction, the orange juice processed waste can be utilized as antimicrobial agent, as this is a cost-effective and less time-consuming approach to obtain antimicrobial activity from the waste. The first development of antimicrobial films using orange peel as a powder (OPP) instead of the extracted EO was reported. The OPP inhibition effects on conidia germination and mycelial growth reduction of the fungal organisms were determined by quantifying the amount needed to develop antimicrobial films [14]. The methanol and ethyl acetate solvent extracts of *C. sinensis* (juice, fruit peel, stem, leaves, and seeds) were subjected to check its anti-MDR activity against bacterial strains, *E. coli*, *Pseudomonas aeruginosa, Acinetobacter baumannii, S. aureus,* Methicillin-resistant *S. aureus* (MRSA), vancomycin-resistant *S. aureus* (VRSA), VRSA and coagulase negative, and methicillin-resistant and vancomycin sensitive *S. aureus*. Additionally, methanolic and ethyl acetate extracts were then labelled with chitosan modified compound (CMC) and the results were compared. Among the two solvents, ethyl acetate extracts displayed a greater activity compared to methanolic extracts and a significant activity was observed by the combinatorial effect of CMC and *C. sinensis*. The study reported that the *C. sinensis* may be a possible therapeutic action against superbugs [15].

The antimicrobial activity of three commonly available fruits peel waste [Orange (*C. sinensis* L.), yellow lemon (*Citrus limonia* Osbeck), and banana (*Musa acuminata*)] was evaluated on wide range of microorganisms namely *P. aeruginosa, E. coli, Klebsiella pneumoniae, S. typhi, Serratia marcescens, Proteus vulgaris, S. aureus, Streptococcus pyogenes, Aeromonas hydrophila*, *Enterococcus faecalis*, *Lactobacillus casei*, and *Listeria monocytogenes, A. niger, Penicillium citrinum*, *Saccharomyces cerevisiae and C. albicans*. The solvents namely methanol, ethanol, ethyl acetate, and distilled water were employed for extraction process. The prepared extracts were evaluated for antimicrobial activity against the selected test organisms by the well-bore method. The individual extract's minimum inhibitory concentration was also determined. The descending order of the yield of extracts from fruit peels using solvents was reported as distilled water, methanol, ethanol, and ethyl acetate, while the descending order of antimicrobial activity efficiency of fruit peel extracts was yellow lemon, orange, and banana peel. The study reveals that respective fruit peel waste can be employed for therapeutic purposes in the combat against MDR bacterial infection. The study confirms that the potential of respective fruit peel waste to be used for therapeutic purpose to combat the MDR microorganism infection as well as proposes that the utilization of waste fruit peels will also result in reduction of waste material and repurposing them for beneficial purposes in a cost-effective and ecologically responsible manner [16].

The pathogens of dental caries, namely *Lactobacillus acidophilus,* and *Streptococcus mutans* were inhibited maximum by hot ethanolic extract of *C. sinensis* peel followed by cold ethanolic extract while at a very high concentrations aqueous extracts were effective. Against both the dental caries pathogens, the minimum inhibitory concentration of hot and cold ethanolic extracts of *C. sinensis* peel ranged from 12-15 mg/ml. The study concludes that *C. sinensis* peel extract contains compounds with therapeutic promise and proposes further toxicity studies and clinical trials on the effects of these peel extracts are needed before large-scale therapy is recommended. [17].

The phenolic content and antimicrobial activities were investigated from the fresh and dry *C. sinensis* peel extracts. The results demonstrated that the fresh *C. sinensis* peel extract possess more phenolic and has better antimicrobial activities against the studied microbial strains *S. aureus and E. faecalis, P. aeruginosa, E. coli, Salmonella typhimurium, C. albicans, A. niger* and *Penicillium notatum* compared to the dry peel extract. The study's findings show that drying plant materials before extraction may not always be the best choice, as some active pharmacological chemicals may be lost in the process. Drying plant parts before extracting phytonutrients has been reported to result in the loss of active components, according to investigators. [18]. The anti-fungal activity of *C. sinensis* [orange peel polyphenolic extract (OPE)] was investigated against *B. cinerea, Monilinia fructicola, and Alternaria alternata* the three related fungal pathogens of post-harvest. The three fungal mycelial growth and conidial germination was inhibited at a concentration of 1.5g/L OPE extract. The results clearly displays that orange peel waste is an excellent anti-fungal compound source and the study proposes that using ferulic acid or ferulic acid-rich extracts, either individually or in formulation with other post-harvest treatment, that may be suitable as a natural alternative to decrease post-harvest damages and, also, improve the shelf-life of fruit [19]. The antifungal effects of *C. sinensis* (L.) Osbeck epicarp EO on *A. niger* growth and morphogenesis were investigated. The loss of cytoplasm in fungal hyphae and hyphal tip budding were the predominant alterations seen under light and scanning electron microscopy following oil treatment. The diameter of the hyphal became significantly thinner, distorted, and cell wall was disrupted. The EO of *C. sinensis* has fungitoxic properties that limit the growth of *A. niger*, causing irreversible morphological changes, and the study suggests that it be used in the biomanagement of *A. niger* [20]. On food contact surfaces, the EO derived from *C. sinensis* var. Liucheng was found to be an efficient agent in inactivating *Vibrio parahaemolyticus*, *S. typhimurium*, *E. coli*, but not *S. aureus* [21].

Generation of Value Added Products through Food Supply Chain Management

The growing awareness of more sustainable uses of natural resources [22] and the transition to a resource-efficient economy [23] are spurring legislative initiatives and institutional procedures toward the creation of bio-economy solutions. The bio-economy has been identified as a strategic lever for employment development in rural and urban regions, as well as for reducing reliance on fossil fuel energy, so promoting both environmental and economic sustainability [24]. The bio-economy concept is linked to the circular economy by completing the material loop through recycling and reusing products, hence lowering raw material consumption. Agricultural wastes and by-products are gaining prominence because they may be used to support an effective bio-economy strategy by converting waste materials into value-added products [25]. Waste materials can be used to create lucrative innovative goods, unlocking new sources of economic value for supply chain actors [26]. The efficient utilisation of agricultural wastes and by-products, which effectively convert waste materials into value-added goods, is seen as critical for an effective bio-economy plan for rural development. Citrus waste valorization is a good example of generating additional market and non-market values [27]. Citrus waste management is a crucial challenge for citrus processors in this context. It does, however, represent a potentially underutilised resource for rural sustainable development.

The global volume of citrus treated each year is approximately 31.2 million tonnes [28], with 50-60% being trash [29, 30]. Citrus waste is mostly made up of water (75–85%), mono- and disaccharides (6–8%), and traces of oil [31]. However, if EO are not properly disposed of, they might constitute a harm to the environment [29, 32]. Various technical breakthroughs for valorizing citrus waste have been created, with the primary goal of converting potential environmental dangers and economic issues [30] into a lucrative resource. Pectin extraction [33], dietary fibre extraction [34], biogas generation [35], ruminant feeding [36], and EO (especially d-limonene) extraction are some of these ways. Adoption of disruptive technology frequently necessitates structural supply chain rearrangement. Several research have been conducted over the last decade to investigate the elements that influence green and sustainable supply chain implementation [37]. One of the most significant impediments to technology adoption is a lack of cooperation and coordination among players in the new supply chain [38].

Conclusion

MDR microbial strains have emerged as a serious threat to worldwide public health and MDR is a topic of considerable interest around the world. Naturally derived compounds have been and will continue to be a significant source of novel medicinal drugs. Research on natural product has recently seen a resurgence of attention, and they are now being looked into more as potential new leads for producing better antimicrobial drugs. Bioactive compounds found in *C. sinensis* have been identified as prospective therapeutic candidates for antimicrobial activity and also MDR prevention. *C. sinensis* EO, with the exception of mild phototoxicity, are generally safe to use and have little human toxicity. EO will definitely continue to play a major part in the food and beverage industry, as well as in pharmaceutical, cosmetic, and "green" pest control applications. It is important to note that, despite the fact that studies on the health-promoting properties of citrus peel extracts have yielded encouraging results *in vitro* and *in vivo*, these breakthroughs are still at an early stage and require further examination before moving forward with clinical trials. Despite the fact that *C. sinensis* peels are agricultural wastes generated in the food supply chain, their vast availability could be used as value-added products using green technology. The study concludes by suggesting that public awareness is needed in order to create value-added products from waste generated from the *C. sinensis* food supply chain.

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