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PHYTOCHEMICAL SCREENING AND ANTIFUNGAL PROPERTIES OF THE PEEL EXTRACT OF *CITRUS SINENSIS* L

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Abstract

This research was carried out at the Laboratory of Food Science and Technology Department, Federal University of Technology, Owerri, Imo State, between the months of June and July, 2024. The data obtained were subjected to analysis of variance (ANOVA). The investigation revealed the presence of flavonoids, saponins, alkaloids, tannin, cardiac glycosides and steroids. Quantitative phytochemical screening of the *Citrus sinensis* L. peel extract revealed that the peel extract of *C. sinensis* L. contains alkaloids, saponins, tannins, flavonoids, and phenols. These constituents may be responsible for antifungal activity. Three different extract of citrus peel (200mg/ml, 100mg/ml and 50mg/ml) were tested for their antifungal activities against two isolates of fungi namely *R. stolonifer* and *F. oxysporum.*The percentage growth inhibition of *R. stolonifer* aqueous and ethanol *C. sinensis* L. peel extract showed that the

Introduction

Webber *et al.*, (2018) stipulated that *Citrus sinensis* L. is an orange fruit basically and its shape is round, its tree has a length of 9-10 m. Leaves of these trees are in oval shape their barks appear to be green or brown in color which is quite smooth. Leaves have a size of 4-10 cm if its length is taken in to account. The leaves of this tree are green. Leaves have smooth texture with a smell resemblance to the sweet orange. The flower of this tree consists of mainly five petals which smell same as saccharine. Valiant *et al.*, (2019) on the other hand stated that *Citrus sinensis* L. has seeds in between the

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aqueous and ethanol extract at 200mg/ml had highest % percentage inhibition of (32% and 40%) aqueous and ethanol extracts respectively. Relatively moderate inhibition were recorded at (100mg/ml) with (28 and 32%) aqueous and ethanol extracts respectively. Lowest inhibitory activities were recorded at (50mg/ml) with aqueous extract (7%) inhibition. The percentage growth inhibition of *F. oxysporum* aqueous and ethanol extracts showed that the aqueous and ethanol extracts at (200mg/ml) showed highest % inhibitory activities with (28% and 36%) respectively. Low inhibitory activities were recorded at (50mg/ml) with (10 and 20%) aqueous and ethanol extracts. The high inhibitory activities of ethanol extract could be due to solvent solubility extraction of antifungal moieties and could be linked to the fact that *C. sinensis* L. has large deposit of alkaloids, flavoniod and phenolic compounds which have antifungal properties. The *Citrus sinensis* L. peel which is normally treated as a waste product can now be used as an antifungal materials, hence reducing environmental problem particularly pollution.

Key words: Phytochemical Screening, Antifungal Properties, Peel Extract, *Citrus Sinensis*

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parts where juices are present. The seeds are green or cream in color. The fruit's flesh is mostly made of the orange sweet juicy part. The peel has orange color. The endocarp is the palatable portion, partitioned into 10-14 sections segregated by thin septa, containing up to 8 seeds/septa, but it was appeared regularly with one. Each segment consists of juice vesicles ("pulp"), with long stalks attached to the juice containing outer wall (Valiant *et al.*, 2019). *Citrus sinensis* L. (sweet orange) is the world's most widely grown and commercialized citrus specie. The fruit of *Citrus sinensis* is mostly recognized for its vitamin C content and is also an important source of other phytochemicals such as phenolics and carotenoids which are reputed to have health benefits. The sweet orange fruit is usually eaten whole or processed into juice after the peeling of the external rind (flavedo). This peeling process leads to the generation of substantial wastes. Sweet orange (*Citrus sinensis* (L.) is the world's most commonly cultivated fruit tree. It belongs to the Rutaceae family which comprises mandarins, limes, lemons, grapefruits, sour and sweet orange. Citrus fruits are of immense economic value; occupying the top position in fruit production. Orange trees are widely cultivated in tropical and subtropical climates for the sweet fruit,

which is peeled or cut (to avoid the bitter rind) and eaten whole, or processed to extract orange juice. Citrus fruits are known to contain substantial quantities of vitamin C, a potent water-soluble vitamin essential for healthy living. The term phytochemical is often used to describe a diverse range of biologically active compounds found in plants. Phytochemicals provide plants with colour, flavour and natural protection against pests. Phytochemicals are not essentially required for the sustenance of life but confer extra health benefits against pathogens. Phytochemical screening avails us with the opportunity to see at a glance the various phytochemicals present in a plant material. This may give a hint as to the possible range of bioactivities the plant product may possess. It also serves as a preliminary step in research protocol aimed at the isolation, purification and utilization of compounds inherent in the plant material for medical, pharmaceutical or agro- industrial use. The orange fruit is of unique economic importance as all portions contain potentials for diverse industrial usability. Thus far, researches into citrus wastes have been concentrated on the peels (flavedo); with little interest on the albedo and seeds.

Saadi *et al.*, (2018) noted that for a long period of time, plants has been a valuable source of natural products for maintaining human health, especially in the last decade, nowadays the use of phytochemicals for pharmaceutical purpose has gradually increased in many countries. The use of crude extracts of plants parts and phytochemicals of known antimicrobial properties can be of great significance in the therapeutic treatment of illness. In recent years, a number of studies have been conducted in various countries to prove such efficiency. Many plants have been used because of their antimicrobial traits, which are due to the secondary metabolites synthesized by the plants. These products are known by their active substances like, phenolic compounds which are part of the essential oils (Webber *et al.*, 2018). Middleton and Kandaswami, (2016) reported that citrus fruit extracts and citrus flavonoids exhibit a wide range of promising biological properties due to their phenolic profile and antioxidant properties. They have been seen to have wide range of antimicrobial activity on pathogenic microorganism. Oreopoulou and Tzia, (2020) also noted that citrus is the largest fruit crop in the world (100 million cubic tons per year) and the orange account for 60%. The remaining orange peel account for approximately 45% of the total bulk (Yeoh *et al.*, 2021). Consequently, significant amounts of orange peel are available as a by-product. The orange peel, if treated as waste materials, may create environmental problems, particularly water pollution, due to the presence

of biomaterials such as essential oil (Ferhat, 2021), pectin (Yeoh *et al.*, 2021; Berna *et al* ., 2018) and sugar. This problem could be turned into an asset, if potentially marketable active principles such as essential oil could be extracted from the peel. After extraction, the peel could be a high protein stock feed in dry form, increasing the potential return for the orange juice industry and reducing the pollution (Yeoh *et al.*, 2021). The orange peels are usually considered as waste materials, which may create environmental problems for local communities because of the presence of biomaterials in orange peel. Every ton of food waste means 4.5 ton of CO_2 emissions. (Adamu *et al.*, 2020). There is a great need for development of new and environmental friendly design processing techniques which could be turned into an asset. Also, the development of drug resistance by some microbial strain to commercial antibiotics has posed concerns to scientist which is a serious problem.

The problem of environmental pollution also can be reduced considerably, though there are several reports on antioxidant and antibacterial effect of juice and edible parts, there are meager literature on the wastes of citrus fruit besides this, the development of fungal resistance to currently available antifungal is still a threat to contend with, this necessitated the search for new antifungal agent. Therefore, this work will be done to determine the minimum inhibitory concentration and minimum fungal concentration of *Citrus sinensis* waste on fungal isolates.

MATERIALS AND METHODS

Area of Study

The research will be carried out at the Laboratory of Food Science and Technology Department, Federal University of Technology, Owerri, Imo State. Imo State University is sited at latitude 5°10' N and 6° O' N and Longitude4 6° 3.5' E and 7° O'N in Nigeria.

Source of materials

The fresh peel of *Citrus sinensis* was collected from Ekeonuwa market in Owerri. The peels will be washed thoroughly with tab water and dried under shade for 7-10 days grounded into fine powder and sieved. The peel part of *Citrus sinensis* will be collected and dried under shade. This dried material will be mechanically powdered, sheaved and stored at a dry place. This powdered material will be used for further phytochemical and antifungal analysis.

Preparation of powder

The peel part of *Citrus sinensis* will be collected and dried under shade. These dried materials will be mechanically powdered, sheaved using 80 meshes and stored in an airtight container. These powder materials will be used for further phytochemical analysis.

Extraction of the plant material

The peel of the plant will be properly washed in tap water and rinsed in distilled water. The rinsed peel part will be hot air-dried for 7-10 days at room temperature. The dried peel part of each plant will be pulverized using pestle mortar to obtain a powdered form which will be stored in airtight glass containers at 4°C until used. 50 g of powdered sample will be soaked in distilled water and methanol (200 ml and 100 ml) separately for 12 hrs at room temperature. The extracts will then be filtered with whatsman filter paper no 4 and concentrated to a final volume of 50 ml and subjected to phytochemical analysis.

Medium Preparation

A semi synthetic potato dextrose agar (PDA) medium will be used for the fungi isolate. The PDA will be prepared from commercially dehydrated medium following the manufacturer's instruction. The method to be used will be as described by Oviasogie *et al.,* (2021) with slight modification.

Twenty (20) grams of powdered (PDA) will be weighed and dissolved in 500ml of distilled water in a sterile conical flask and covered. The mixture will be autoclaved for 15 minutes as a temperature of 121^{o}C and 1.055kg/km^{2} pressure and allowed to cool. The warm medium will be dispensed into sterile petri discs aseptically and allowed to solidify at room temperature. Two drops of lactic acid will be added to exclude bacterial growth (Okigbo and Odurukwe, 2020). Awurum and Nwaneri (2022) two concentrations of both aqueous and ethanol extracts (0.5ml, and 1ml) will be prepared from the extracts for antifungal test. The effect of the plant extract on mycelium growth of the isolates will be determined using the food poisoning method technique (Sungoyomi *et al.,* 2021).

Phytochemical Test

The phytochemical screening involves the simple chemical test to detect the presence of secondary metabolites. The phytochemical test include: tests for saponins, tannins, flavonoids, alkaloids, cardiac glycosides and phenol.

Isolation and Identification of Fungi Isolates

The isolation adopted from Ugwuja and Chiejina (2015) will be used with modification. This section will be cut from periphery of disease yam and surface will be rinsed in three changes distilled water and plated in potato Dextrose Agar (PDA) plates. The plates will be incubated at room temperature (28± 2°C) for 3 -4 days subculture of colony growth will be made to obtain a clean culture. Identification will be based on observed culture wraith pattern, mycelium colour and microscopic examinations of vegetative and reproductive structures of the fungi isolates using Barnett and Hunter (2018) and Alexopulos *et al.,* (2019).

Extraction of plant samples

Fresh peel of the *Citrus sinensis* will be obtained, washed and allowed to dry in room temperature (28± 2°C) for two weeks. The dried peels will be grounded using milling machine. The grounded sample will be sieved through a 1mm musilin cloth to obtain powdered processed samples to be used for the extraction. Aqueous and ethanol extracts will be obtained using cold solvent extraction method as described by Awurum and Nwaneri, (2022). Two concentration of both aqueous and ethanol extract (0.5ml, and 1ml) will be prepared from the extract for antifungal test. The effect of the peel extract on mycelium growth of the isolates will be determined using the food poisoning method techniques (Sungoyomi *et al.*, 2021).

Statistical Analysis

Standard errors of the mean values will be calculated for the separate readings and data will be subjected to analysis of variance (ANOVA) to compare the means at 0.05 confident intervals (Obi, 2022).

RESULTS

Phytochemical Screening of *Citrus sinensis* L peel extract:

Phytochemical screening of *Citrus sinensis* L. peel extract revealed the presence of alkaloids, flavonoid, saponin, tannin, cardiac glycoside and steroids. (Table 1).

Table 1: Qualitative Phytochemical Screening of the Citrus sinensis L peel extract.

Constituents	Soxchlet	Aqueous	Ethanol
Alkaloids	-	+	+
Flavonoid	+	+	++

Saponin	+	-	-
Tannin	+	++	++
Cardiac glycosides	+	++	++
Steroids	++	-	+

+ = Present, ++ = Markedly present, - = Absent

The quantitative phytochemical screening of the *Citrus sinensis* L peel extract revealed 4.12% of alkaloids, 0.54% of saponin, 3.12% of tannin, 6.10 mgCE/g of flavonoid and 7.12 mgGAE/g of phenol in aqueous extract. The ethanol extract showed 12.34% of alkaloids, 3.17% of saponin, 4.23% of tannin, 18.75 mgCE/g of flavonoid and 17.45 mgGAE/g of phenol (Table 2).

Table 2: Quantitative Phytochemical Screening of the *Citrus sinensis* L. peel extract.

Constituents	Aqueous extract	Ethanol extract
Alkaloids %	4.12	12.34
Flavonoid (mgCE/g)	6.10	18.75
Saponin %	0.54	3.17
Tannin %	3.12	4.23
Phenol (mgGAE/g)	7.12	17.45

Antifungal Activity

For aqueous extract at 50mg/ml concentration, the percentage mycelia growth inhibition showed 7.10% for *Rhizopus stolonifer* (Penz) and 10.48% for *Fusarium oxysporum* (Mart). At 100mg/ml concentration, the percentage mycelia growth inhibition showed 28.12% and 20.01% for *Rhizopus stolonifer* (Penz) and *Fusarium oxysporum* (Mart) respectively. At 200mg/ml concentration, the percentage mycelia growth inhibition showed 32.55% and 28.10% for *R. stolonifer* and *F. oxysparium* respectively (Table 3).

For ethanol extract, at 50mg/ml concentration, the percentage mycelia growth inhibition showed 17.15% and 20.67% for *R. stolonifer* and *F. oxysporum* respectively. At 100mg/ml concentration, the percentage mycelia growth inhibition showed 32.40% and 28.15% for *R. stolonifer* and *F. oxysporum* respectively. At 200mg/ml concentration, the percentage mycelia growth showed 40.63% and 36.22% for *R. stolonifer* and *F. oxysporum* respectively (Table 3).

Table 3: Effects of Citrus sinensis L peel Aqueous and Ethanol extract on mycelia growth of fungi isolates.

Organism/ Conc. (mg/ml)	Aqueous extract			Ethanol extract		
	50 (%)	100(%)	200(%)	50 (%)	100 (%)	200 (%)
Rhizopus Stolonifer **(Penz)**	7.10^{c} ± 0.53	28.12^{b} ±0.84	32.55^{a} ±0.72	17.15^{c} ±0.72	32.40^{b}± 0.61	40.64^{a} ±0.16
Fusarium oxysporum **(Mart)**	10.48^{c}± 0.28	20.01^{b} ±0.69	28.10^{a} ±0.73	20.67^{c} ±0.39	28.15^{b} ±0.78	36.22^{a} ±0.65

Values are mean ± standard error of 3 replicates, a b c Means in a column with different superscripts are significantly different ($P<0.05$).

Discussion

Phytochemical Screening:

Akinmoladun *et al.,* (2007) stated that the medicinal value of these plants lies in bioactive phytochemical constituents that produce definite physiological action on the human body. The preliminary phytochemical investigation revealed the presence of various constituents of citrus peel as shown in Table 1. Different solvent showed different class of phytochemicals flavonoids, saponins, alkaloids, tannin, cardiac glycosides and steroids were detected. These constituents may be responsible for antifungal activity.

Quantitative Phytochemical Screening of the *Citrus sinensis* L. peel extract.

The result of this investigation revealed the phytochemical constituents of peel extract of C. sinensis L. to contain; alkaloids, saponins, tannins, flavonoids, and phenols (Table 2). These phytochemicals are secondary metabolites of natural products comprising nutritional or non-nutritional bioactive compounds found in fruits, vegetables, cereals, and other plant foods that have numerous health advantages such as reducing the risk of major (Kumar *et al.,* 2023).

The phytochemical composition of aqueous and ethanolic extracts of *C. sinensis* L. is shown in Table 2. Alkaloids were highest in the peel ethanol extract (12.34 %) while low in the aqueous extract of 4.12 %. Saponin was highest in the

ethanolic extract of the peel with 3.17 % while least in the aqueous extract of seed with 0.54 %. Tannin was of high concentrations in the peel of ethanolic extract having 4.23 % and aqueous extract at 3.12 %. Flavonoids were in high concentration in ethanolic extract of peel with 18.75 mgCE/g compositions and least in aqueous extract with 6.10 mgCE/g compositions. The maximum range of phenols was 17.45 mgGAE/g and 7.12 mgGAE/g which were in the ethanol extract of peel and aqueous extract of peel respectively.

The results are in agreement with the studies conducted by Mathew *et al.,* (2012) and Nduche *et al.,* (2019) which reported the presence of alkaloids, saponin, tannin, and flavonoids in citrus fruits. The total flavonoid and phenols of the aqueous and ethanol extracts of *C. sinensis* show the flavonoid and phenols in peel extracts were higher. This finding corroborates with the report by Li *et al.,* (2006).

Alais and Lindon (1999) and Okwu and Morah (2007) reported that, Rutaceae family generally contain host of active phytochemicals like limonin, nomilin, octanol, cineole and naringin that inhibit fungal pathogens, this compounds occur in high concentrations in grape, lemon and orange fruits and are responsible for that bitter taste of the fruits. Saponins were found in preliminary studies of the C. limon extracts, the presence of saponins may also be responsible for bitter taste and antifungal properties of this plant (Okwu *et al.,* 2007). The presence of tannins and phenolic compounds in C. limon peels indicates that this plant have antimicrobial properties since phenols and phenolic compounds are extensively used in disease prevention and remain a standard with which other bactericides or fungicides are compared (Okwu and Morah, 2007). Okwu *et al.,* (2007) reported high percentage of alkaloids and flavanoids in C. limon peels extracts which are in conformity with the preliminary screening of *C. sinensis* peels extracts in this study. Phenolics forms large group of naturally occurring, diverse and widespread compounds responsible for antifungal or bactericidal properties (Okwu *et al.,* 2007).

Antifungal Activity

Three different extract of citrus peel (200mg/ml, 100mg/ml and 50mg/ml) were tested for their antifungal activities against two isolates of fungi namely *R. stolonifer* and *F. oxysporum.*

The percentage growth inhibition of *R. stolonifer* aqueous and ethanol *C. sinensis* L. peel extract were presented in Table 3. The aqueous and ethanol extract at

200mg/ml had highest % percentage inhibition of (32% and 40%) aqueous and ethanol extracts respectively. Relatively moderate inhibition were recorded at (100mg/ml) with (28 and 32%) aqueous and ethanol extracts respectively. Lowest inhibitory activities were recorded at (50mg/ml) with aqueous extract (7%) inhibition.

The percentage growth inhibition of *F. oxysporum* aqueous and ethanol extracts was shown in Table 3. The aqueous and ethanol extracts at (200mg/ml) showed highest % inhibitory activities with (28% and 36%) respectively. Low inhibitory activities were recorded at (50mg/ml) with (10 and 20%) aqueous and ethanol extracts. The high inhibitory activities of ethanol extract could be due to solvent solubility extraction of antifungal moieties and could be linked to the fact that *C. sinensis* L. has large deposit of alkaloids, flavoniod and phenolic compounds which have antifungal properties (Patkwoski, 2006). El-Zawawy, (2015) reported that antimicrobial properties of plants extracts have been largely attributed to the presence of flavanoid and alkaloid compounds.

Okwu and Morah, (2007) stipulated that Citrus plants contain phytochemical compounds such as limolin, nomilin, octanol, cineole and terpenoids that possesses antifungal properties, this components of phytochemical which occur in high concentrations in lemon, grapes and oranges are responsible for bitter taste in this fruits and inhibits the growth of bacteria and fungi. Huang and Chung, (2003) also observed that the mechanism of inhibitory actions of phyochemicals such as alkaloids and phenolic compounds on micro organisms may be due to impairments of variety of enzymes systems in micro organisms including those involved in energy production, interference with the integrity of the cell membranes and structural components synthesisis.

According to Okwu *et al.,* (2007) 10% concentrations of peels and leaf extracts of citrus and synthetic Benomyl have inhibited the growth of *F. oxysporum* by (83.55% and 83.04%) respectively, this findings is in agreement with the present study which showed that 200mg/ml Citrus peels ethanol extracts inhibited the growth of *F. oxysporum* by 36% and *R. stolonifer* by 40%. Ortuno *et al.,* (2006) reported that Citrus flavanoid have a large spectrum of biological activities including anti bacterial, antifungal, anti diabetic, anti cancer and anti viral activities. Phenolic compounds are also considered to be fungistatic and bactereostatic. These compounds caused swelling of hyphal tips, plasma seeping around huphae, cell wall devastation, abnormal branching or fusion of hyphae and consequently wrinkling of hyphal surface (Huang and Chung, 2003). Cowan,

(2015) reported that lipophilic flavanoids may disrupt microbial membrane. Volatile oils of Citrus have anti microbial effects against bacteria and fungi (Ahonkhai *et al.,* 2009). Kumar *et al.,* (2022) noted that lemon peels have high quantity of saponin with lytic properties against many strain of bacteria and fungi.

Scientifically, it is believed that plants stores different antifungal, anti bacterial and anti viral phytochemicals on the bark, leaves and exocarp of fruits for protection especially to preserve the fruits and protect the seeds from microbial attack. This is in agreement with the findings of Okwu and Emenike (2006) who reported that phytochemicals are reserved in plants to protect the plants against invasion by pathogenic micro organisms. The extracts of peels of *Citrus sinensis* L. inhibited the growth of *R. stolonifer* and *F. oxysporum* significantly which is in agreement with the work of Okwu *et al.,* (2007). Extracts of *Citrus sinensis* L. peels contains antifungal compounds that can be used as Biofungicides. The prospect of using Citrus peels as natural fungicides is encouraging as it is biodegradable, non toxic, environmentally friendly and safe to non target micro and macro flora and fauna, less resistance development in pathogenic pest and above all cheap and readily available. Based on these findings *Citrus sinensis* L. peels that are waste by-products, its extracts can be used to control and manage fungal disease of plants as alternative to synthetic fungicides.

Conclusion

The phytochemical analysis of *Citrus sinensis* L. revealed the presence important secondary methabolites such as alkaloids, tannins, saponins, flavonoids, and phenols; thus indicating the therapeutic potentials of the plants. The higher level of flavonoids and phenols in the extract makes it a good antifungal, antioxidant and antiflammatory plant source.

Antifungal efficacy of *C. sinensis* peel extract from this study has offered a valuable source for discovering an alternative to use of synthetic chemical fungicides.

Recommendation

A further study of the extract should be carried out to isolate, characterized and elucidate the structure of the bioactive compounds present which were responsible for the potent antifungal activity *C. sinensis* peel. Also, studies should be done to know the effectiveness of the oil present in this fruit.

Contribution to Knowledge

The *Citrus sinensis* L. peel which is normally treated as a waste product can now be used as an antifungal materials, hence reducing environmental problem particularly pollution.

REFERENCES

Adamu, L. S., Seeram, N. P., Agarwal, B. B., Takada, Y., Sand, D. (2020). Pomegranate juice, total pomegranate ellagitannins, and punicalagin suppress inflammatory cell signaling in colon cancer cells. *J Agri Food Chem* 54(3): 980-985.

Ahonkhai, I., Ba, A., Edogan, O. and Mu, U. (2009). Antimicrobial activities of the volatile oils of Ocimum bacilicum and Ocimum gratissimum L. against some aerobic dental isolates. *Pakista J. Pharm. Sci.* 22:405-409.

Akinmoladun, A. C., Ibukun, E. O., Obuotor, E. M. and Farombi, E. O. (2007). Phytochemical constituent and antioxidant activity of extract from the leaves of Ocimumgratissimum. *Sci. Res.* Essay, 2, pp.163-166.

Alais, C. and Linden, G. Food Biochemistry. Aspen Publications, Maryland. (1999). 114-116.

Alexopoulos, C. O., Mims, C.W and Blackwell, M. (2019). *Introductory mycology* 4th edition. john willy and sons. inc Singapore

Awurum, A. N. and Nwaneri, J. A. (2022). Fungitaxic effect of some plant extract on wet rot of Amaranthus indicated by *Choanephoracucurbitarium*. *Nigeria journal of plant protection* 25(2) 230-236

Bernard, A., Tàrrega, A., Blasco, M. and Subirats, S. (2018). Supercritical CO_2 extraction of essential oil from orange peel; effect of the height of the bed.*Journal of Supercritical Fluids* 18: 227-237.

Cowan, M. M. (2015). Plant products as antimicrobial agents. Clin. Microbiol. Rev. 12(4), 564- 582.

El-Zawawy, N. A. (2015). Antioxidant, antitumor, antimicrobial studies and quantitative phytochemical estimation of ethanolic extracts of selected fruits peels. *Internation Journal of Current Microbial and Applied Science.* 4(5): 298-309.

Huang, J. W. and Chung, W. C. (2003). Management of vegetable crop diseases with plant extracts. pp.153-163.

Kumar KA, Narayani M, Subanthini A, Jayakumar M. (2022). Antimicrobial activity and phyochemical analysis of Citrus fruits peels-utilization of fruit waste. International Journal of Engineering Sciences and Technology. 36(6): 5414-5421.

Kumar, A. P. N., Kumar, M., Jose, A., Tomer, V., Oz, E., Proestos, C., Zeng, M., Elobeid, T. K. S. and Oz, F. (2023). Major Phytochemicals: Recent Advances in Health Benefits and Extraction Method. *Molecules.* 16; 28(2):887.

Li, B. B., Smith, B. and Hossain, M. (2006). Extraction of phenolics from citrus peels: I. Solvent extraction method, Separation and Purification Technology. 48(2):182-188.

Mathew, B. B., Jatawa, S. K. and Tiwaari, A. (2012). Phytochemical analysis of Citrus limonum pulp and peel. *International Journal of Pharmacy and Pharmaceutical Sciences.* 4:269-371.

Middleton, A. E. and Kandaswami, G. I. (2016). Phytochemical properties and antibacterial activities of the leaf and latex extracts of *Calotropisprocera*(Ait.F.)Ait.F. *Bayero J. Pure App. Sci.*, 2(1): 34-40.

Obi, I. U. (2022). Statistical Methods of Detecting Differences between Treatment Means and Research Methodology Issues in Laboratory and Field Experiments. AP Express Publishers, limited, 3 Obollo Road, Nsukka – Nigeria. pp.117.

Okigbo, R. N. and Odurukwe, C. N. (2022). Occurrence and control of fungal rot pathogen of yam (*Dioscorearotundata*Poir) with leaf extracts of *Chromolaenaodorata, Carica papaya* and *Aspilia Africana*, *Nigerian Journal of Mycology*, vol 2(1), 154–165.

Okigbo, R., Putheti, N., Ramesh, A. and Achusi, C.T. (2020). Post-harvest deterioration of cassava and its control using extracts of Azadirachtaindica and Aframomummelegucta. *E-Journal Chem* 6: 1274-1280.

Okwu, D. E. and Morah, F. N. I. (2007). Antimicrobial and phytochemical evaluation of seed of Garcinia Kola and Dennettia tripetela fruits. *Journal of Medicinal and Aromatic Plant Sciences*. 29:20-23.

Okwu, D. E., Awurum, A. N. and Okoronkwo, J. I. (2007). Phytochemical composition and in vitro antifungal activity screening of extracts from Citrus plants against Fusarium oxysporum of okra. *Pest Technology.* 1(2): 145-148.

Okwu, D. E. and Emenike, I. N. (2006). Evaluation of phytochemical constituents and vitamin contents of citrus fruits. International Journal of Molecular medicine and advanced Sciences. 2:1-6.

Oreopoulou, V. and Tzia, C. (2020). Utilization of plant by-products for the recovery of proteins, dietary fibers, antioxidants and colorants.In Utilization of by-products & treatment of waste in the food industry, Springer.

Ortuno, A. A., Baidez, P., Gomez, P. M. C., Arcas, I., Porras, A. G. and Del Rio, J. A. (2006). Citrus paradise and Citrus sinensis flavonoids: Their influence in the defence mechanism against Penicillium digitatum. *Food Chemistry.* 98(2): 351-358.

Ovrasogie, F. E. Ogofure, A. G., Beshiru, A., Ode, J. N and Omege, F. I. (2021). Assessement of fungal pathogens associated with Orange spoilage. *African Journal of microbiology research* 9(29) 1758-1763

Patkowski, E. (2006). Effectiveness of grape fruits extracts and pythium oligandrun in the control of beans and peas pathogen. *Journal of Plant Protection Research.* 46:15-28.

Saadi, A. C, Ibukun, E. O. and Obuotor, E. M,. and Farombi, E.O. (2018). Phytochemical constituent and antioxidant activity of extract from the leaves of *Ocimumgratissimum*.Sci. Res. Essay, 2, pp.163-166.

Sangoyomi, T. E., Ekpo, E. J. A. and Asiedu, R. (2021). Fungitoxic effects and attributes of *Allium sativum and Occimumgratissimum*extracts on *Rhizoctoniasolani, the causal organism of yam (Dioscorearotundata (Poir) rot disease. Nig. J. Mycol.* 2(1) : 166 -167.

Valiant, K. F., Aydin, T., Başaran, D. and Gülen, T. (2019). Antimicrobial activity of Turkish *citrus* peel oils *Pak. J.Bot.*, , 41(6), pp. 3207-3212.

Webber, J. H., Reuther, W. and Lawton, H. W. (2017). History and development of the citrus industry In: Davies, F.S., Albrigo, L.G. (Eds.). Crop production science in Horticulture: Citrus, Cab International, and Wallingford, UK, pp. 1-242.

Yeoh, S., Shi, J. and Langrish, T. A. G. (2021). Comparisons between different techniques for water-based extraction of pectin from orange peels.*Desalination* 218: 229-237.

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