

# Phytochemical and Antimicrobial Activities of *Acalypha wilkesiana*

Erhumuoghene Mary Enakireru, Olasupo John Ilori\* and Williams Omotola Tanimowo.

Department of Biological Sciences, Anchor University, Lagos, Nigeria,

\*Corresponding author

## Abstract:-

**Background:** There is a considerable concern among consumers regarding the health risk of using synthetic additives for food preservation. Therefore, there is need for the use of plant products for food preservation.

**Objectives:** This study was designed to identify the phytochemicals in aqueous and methanol extracts of *Acalypha wilkesiana* and examine the effects of extracts of *Acalypha wilkesiana* on the growth of fungi and bacteria isolated from spoilt fruits and vegetables.

**Material and methods:** The extracts were prepared according to standard methods. The extract solution (100%) was further diluted to various concentrations (75%, 50%, 25%). Spoilage organisms were cultured on potato dextrose agar, Macconkey agar and Nutrient agar. Pure cultures were identified using a variety of biochemical tests, microscopy and their cultural characteristics. These microbes were subjected to the different extract and the data obtained were analysed by Analysis of Variance (ANOVA) to determine significant ( $P < 0.05$ ) effects. Significant differences between means were determined using Duncan's Multiple Range Test DMRT.

**Results:** The phytochemical screening indicated the presence of alkaloids, phenols, glycosides, flavonoids, terpenoids, saponins, quinones, anthraquinones, steroids and phlobatanins. The extract had inhibition properties against the spoilage microorganisms. The extract showed antibacterial effects against *Pseudomonas aureginosa*, *Salmonella enterica*, *Citrobacter freundii*, *Proteus vulgaris*, *Salmonella typhi*, *Enterobacter aerogenes*. Also, extract showed antifungal effects against *Candida tropicalis*, *Rhizopus microsporus*, *Aspergillus niger*, *Trichophyton equinum*, *Zygosaccharomyces* sp.

**Conclusion.** The findings of this research showed that *Acalypha wilkesiana* is an excellent options for management of spoilage caused by the test isolates and are relevant for consideration in development of new natural preservations.

**Keywords:-** Phytochemical, Antimicrobial, Extracts, *Acalypha wilkesiana*.

## I. INTRODUCTION

Food spoilage and food losses are important issues for human beings with regards to food safety and food security, since people started producing and storing food products. These losses result to one or more problems in agricultural production (Rezaei and Liu, 2017). The main agents responsible for microbial spoilage include parasites, bacteria, and/or fungi. Consumers in recent years have high preference for natural preservatives to chemical-based preservatives due to their adverse health effects. This natural preservative are alternatives to chemical preservatives so as to prolong the shelf life and safety of food (Arora *et al.*, 2013). Many researchers have investigated the antimicrobial effects of plant extracts on food spoilage microbes (Díaz Dellavalle *et al.*, 2011; Mohanka and PriyankaInt, 2014; Clarke *et al.*, 2017; Oso *et al.*, 2018; Kursa *et al.*, 2022; Atwaa *et al.*, 2022; Akinduti *et al.*, 2022; Alhadad *et al.* 2022).

*Acalypha wilkesiana* belongs to the family *Euphorbiaceae*. The preservation of food crops is necessary to prevent and reduce spoilage caused by microorganisms. It also helps to increase the shelf life of food crops and meet the needs of the demands of consumers for safe and natural products without chemical preservatives. Therefore, the objective of this study was to determine the phytochemicals in the extracts of *A. wilkesiana* and examine the effects of the extracts on the growth of fungi and bacteria in spoilt food.

## II. MATERIALS AND METHODS

### ➤ Plants Extraction.

Extraction procedures was carried out according to standard methods. The leaves of *Acalypha wilkesiana* were thoroughly washed and 20 g of the leaves were extracted in water and methanol. Various concentrations (100%, 75%, 50% and 25%) of water and methanolic extract were prepared by the way of dilution.

### ➤ Phytochemical Analysis

Phytochemical screening for alkaloids, phenols, flavonoids, saponins, terpenoids and glycosides, steroids, quinones and anthraquinones, sulphuric acid was carried according to Sofowora (1982).

➤ *Isolation and Identification of Test Organisms*

The microbes were isolated from spoiled food samples by using the pour plate method. Biochemical and morphological examination tests were employed to identify the bacterial and fungi.

➤ *Screening For Antimicrobial Activity Of The Extracts*

The aqueous extracts were evaluated for antibacterial activity and antifungal activity according to agar well diffusion method of Daoud *et al.* (2015). Muller Hinton agar medium was prepared and poured into sterile Petri dishes by pour plate technique. Using a sterile cork borer (9 mm) wells were made in the agar medium. The extracts were introduced into the well and all the plates were incubated at 37°C for 24 hours. The diameters of zone of inhibition produced by the extracts were measured in mm for antibacterial activity. Similarly the extracts were screened for antifungal activity

using agar well diffusion method. Potato Dextrose Agar (PDA) medium was used for inoculation of fungal isolates.

➤ *Statistical Analysis*

The data obtained were analysed by Analysis of Variance (ANOVA) to determine significant (P< 0.05) effects. The significant differences between means were determined using Duncan’s Multiple Range Test DMRT. The result of the study is presented as Mean ± standard error of the trials.

**III. RESULTS AND DISCUSSION**

➤ *Phytochemical Analysis of Alcalypha wilkesiana*

The phytochemical analysis carried out on different extracts of *Acalypha wilkesiana* showed the presence of alkaloid, glycosides, flavonoid, saponins, phenols, terpenoids, steroid, quinone and phlobatanins. Anthraquinones was absent in all the extracts. (table 1).

Table1: Phytochemical analysis results of *Alcalypha wilkesiana*

S/N	PHYTOCHEMICALS	Hot water extract	Distilled water extract	Methanol extract
1.	Alkaloids	+	-	-
2.	Phenols	+	+	+
3.	Glycosides	+	+	+
4.	Flavonoids	-	-	+
5.	Terpenoids	+	+	+
6.	Saponins	+	+	+
7.	Quinones	+	+	+
8.	Anthraquinones	-	-	-
9.	Steroids	+	+	+
10.	Phlobatanins	+	+	+

Key (+) positive (-) negative

Most of the extracts of *Acalypha wilkesiana* showed antifungal activities while more antibacterial activities were observed in methanol extracts. The extract prepared from methanol had more effect on *Pseudomonas aeruginosa* and *Salmonella enterica* compared to that of distilled water extracts had more effects on *Citrobacter freundii*, *Proteus vulgaris*, *Salmonella typhi*, *Enterobacter aerogenes* (Table 2). The methanol extracts had more effect on *Aspergillus flavus* while distilled water extracts had more effects on *Aspergillus niger* and *Trichophyton equinum*. The hot water extracts showed more antifungal activities against *Rhizopus microspores* and *Zygosaccharomyces* (Table 3).

Table 2: Antibacterial effect of different extracts of *Acalypha wilkesiana* on *Pseudomonas aureginosa*, *Salmonella enterica*, *Citrobacter freundii*, *Proteus vulgaris*, *Salmonella typhi*, *Enterobacter aerogenes*.

Extract	Conc.	<i>P.aeruginosa</i>	<i>S.enterica</i>	<i>C. freundii</i>	<i>P. vulgaris</i>	<i>S. typhi</i>	<i>E. aerogenes</i>
Hot water	100%	12.0 ± 0.00	-	9.50± .50	11.0±0.00	11.0±0.00	-
	75%	12.0 ±0.00	-	9.50±0.50	10.0±1.00	9.50±0.50	-
	50%	11.0 ± 0.00	-	9.50± .50	9.50±0.50	9.50±0.50	-
	25%	-	-	9.50±0.50	9.00±0.00	9.50± .50	-
Distilled water	100%	11.0 ± 0.00	-	13.0±2.00	12.5±0.50	10.0± .00	10.0 ± 1.00
	75%	10.50± .50	-	10.0±1.00	12.0± .00	10.0±0.00	9.50 ± 0.50
	50%	11.0 ± 1.00	-	9.50±0.50	9.50±0.50	-	-
	25%	-	-	-	9.50±0.50	-	-
Methanol extract	100%	16.0 ± 3.50	10.0± .00	-	9.50± .50	10.5±1.50	9.50 ± 0.50
	75%	12.50 ± 1.50	10.0±1.00	9.50±0.50	10.5 ± 1.5	9.50± .50	-
	50%	10.50 ± 1.50	10.0± .00	-	-	-	-
	25%	10.50 ± 1.50	-	-	-	-	-

Table 6: Antifungal effect of different extracts of *Acalypha wilkesiana* against *Candida tropicalis*, *Rhizopus microsporus*, *Aspergillus niger*, *Trichophyton equinum*, *Zygosaccharomyces* sp

Extract	Concentration	<i>Aspergillus niger</i>	<i>Aspergillus flavus</i>	<i>Rhizopus microsporus</i>	<i>Trichophyton equinum</i>	<i>Zygosaccharomyces Sp</i>
Hot water	100%	10.5 ± 0.50	28.0 ± 16.0	19.0 ± 3.00	11.5 ± 0.50	19.0 ± 3.00
	75%	9.5 ± 0.50	12.0 ± 1.00	17.5 ± 5.50	13.0 ± 2.00	17.5 ± 5.50
	50%	8.0 ± 1.00	11.0 ± 2.00	15.0 ± 6.00	-	15.0 ± 6.00
	25%	7.50 ± 0.50	10.5 ± 1.50	13.0 ± 5.65	-	13.0 ± 5.65
Distilled water	100%	16.5 ± 2.50	25.0 ± 5.00	16.5 ± 4.50	21.5 ± 0.50	16.5 ± 4.50
	75%	12.5 ± 1.50	18.0 ± 2.00	13.0 ± 3.00	11.0 ± 0.00	13.0 ± 3.00
	50%	8.00 ± 0.00	14.0 ± 0.00	11.0 ± 0.00	-	11.0 ± 0.00
	25%	8.00 ± 0.00	13.0 ± 1.41	12.5 ± 1.50	-	12.5 ± 1.50
Methanol extract	100%	14.5 ± 5.50	27.5 ± 2.50	11.0 ± 0.00	17.0 ± 6.00	11.0 ± 0.00
	75%	8.50 ± 0.50	22.5 ± 2.50	-	-	-
	50%	8.50 ± 0.50	15.0 ± 4.00	15.5 ± 4.50	-	15.5 ± 4.50
	25%	8.50 ± 0.50	12.5 ± 1.50	11.5 ± 2.50	-	11.5 ± 2.50

Despite the proven efficiency of chemical preservative in prevention and outbreak control of food poisoning diseases, their repeated applications has resulted in the accumulation of chemical residues in food chain, microbial resistance and unpleasant side effects of these chemicals on human health (Bialonska *et al.*, 2010). Because of such concerns, efforts have been focused on developing potentially effective, healthy safer and natural food preservatives. The phytochemical screening indicated the presence of alkaloids, phenols, glycosides, flavonoids, terpenoids, saponins, quinones, anthraquinones, steroids and phlobatanins. The results demonstrates that both aqueous and methanolic extracts of *Acalypha wilkesiana* had significant invitro antimicrobial activity, however the degree of inhibition varied among the extracts. This is possibly due to the differences in solubility of the bioactive components in the extracts. This is in agreement with earlier report by Ibekwe *et al.*(2001). From the results, it is evident that the extract had a high inhibition properties against the spoilage microorganisms. Fungal species isolated from this study have been implicated in food infection (Tafinta *et al*, 2013). Fungal isolates were mostly inhibited by extracts from *Acalypha wilkesiana*. Antimicrobial activities of crude leaf extracts of *Acalypha wilkesiana* is well documented (Adesina *et al*, 2000).

#### IV. CONCLUSION

The present study showed that all the fungi and bacteria were inhibited by the extract of red *Acalypha* leaf. Therefore, the findings of this research showed that *Acalypha wilkesiana* is an excellent options for management of spoilage caused by

the test isolates and are relevant for consideration in development of new natural preservatives.

#### REFERENCES

- [1]. Adesina, S.K., Idowu, I.O., Ogundaini, A.O., Oladimeji, H., Olugbade, T.A., Onawunmi, G.O. and Pais, M. (2000). Antimicrobial Constituents of the Leaves of *Acalypha wilkesiana* and *Acalypha hispida* *Phytother. Res.* 14, 371–374
- [2]. Akinduti, P.A., Emoh-Robinson, V., Obamoh-Triumphant, H.F. Obafemi, Y. D. and Banjo, T. T. (2022) Antibacterial activities of plant leaf extracts against multi- antibiotic resistant *Staphylococcus aureus* associated with skin and soft tissue infections. *BMC Complement Med Ther* 22, 47.
- [3]. Alhadad, A. , Salem, G. , Elmhdwi, M. , Hussein, S. and Elshareef, S. (2022) Assessments of Antibacterial and Antioxidant Properties in the Methanolic and Aqueous Leaf Extracts of *Pistacia lentiscus* against Different Antibiotic Resistance Pathogenic Bacteria. *Advances in Bioscience and Biotechnology*, 13, 113-133.
- [4]. Arora, D. S., Onsare, J. G., and Kaur, H. (2013):Bioprospecting of Moringa (Moringaceae): microbiological perspective. *Journal of pharmacognosy and phytochemistry*, 6, 193-215.
- [5]. Atwaa, E.S.H., Shahein, M.R., Radwan, H.A., Mohammed, N.S., Aloraini, M.A., Albezrah, N.K.A., Alharbi, M.A., Sayed, H.H., Daoud, M.A. and Elmahallawy, E.K. (2022) Antimicrobial Activity of Some Plant Extracts and Their Applications in

- Homemade Tomato Paste and Pasteurized Cow Milk as Natural Preservatives. *Fermentation*, 8, 428.
- [6]. Bialonska, D., Ramnani, P., Kasimsetty, S. G., Muntha, K. R., Gibson, G. R., and Ferreira, D. (2010). The influence of pomegranate by-product and punicalagins on selected groups of human intestinal microbiota. *International journal of food microbiology*, 140(2-3), 175-182.
- [7]. Clarke, D., Tyuftin, A. A., Cruz-Romero, M. C., Bolton, D., Fanning, S., Pankaj, S. K., and Kerry, J. P. (2017). Surface attachment of active antimicrobial coatings onto conventional plastic-based laminates and performance assessment of these materials on the storage life of vacuum packaged beef sub-primals. *Food microbiology*, 62, 196-201.
- [8]. Daoud, A., Malika, D., Bakari, S., Hfaiedh, N., Mnafigui, K., Kadri, A. and Gharsallah, N. (2019). Assessment of polyphenol composition, antioxidant and antimicrobial properties of various extracts of Date Palm Pollen (DPP) from two Tunisian cultivars. *Arabian Journal of Chemistry*, 12(8):3075-3086.
- [9]. Díaz Dellavalle, P. Cabrera, A., Alem, D., Larrañaga, P. Ferreira, F. and Rizza, M. (2011). Antifungal activity of medicinal plant extracts against phytopathogenic fungus *Alternaria* spp. *Chilean Journal of Agricultural Research*. 71. 231-239.
- [10]. Ibekwe V.I., Nnanyere N.F. and Akujobi C.O. (2001). Studies on Antibacterial activity and phytochemical qualities of extracts of orange peels. *Int. J. Environ., Health and Human Development*. 2(1):41-46.
- [11]. Kursa, W., Jamiołkowska, A., Wyrostek, J. and Kowalski, R. (2022) Antifungal Effect of Plant Extracts on the Growth of the Cereal Pathogen *Fusarium* spp.— An In Vitro Study. *Agronomy*, 12: 3204.
- [12]. Mohanka, R. and PriyankaInt (2014). Plant extract as natural food preservative against spoilage fungi from processed food *J.Curr.Microbiol.App.Sci* 3(8) 91-98.
- [13]. Oso, B., Ogunnusi, T. and Fagbemi, M.. (2018). Antimicrobial Effect of Some Plant Extracts on Plant Pathogens that Cause Food Spoilage. *Microbiology Research Journal International*. 24: 1-15.
- [14]. Rezaei, M., and Liu, B. (2017): Food loss and waste in the food supply chain. *International Nut and Dried Fruit Council: Reus, Spain*, Pp26-27.
- [15]. Sofowora, A. (1982). *Medicinal Plants and Traditional Medicine in Africa*. John Wiley and Sons Ltd Nigeria pp. 33-34.
- [16]. Tafinta, I.Y., Shehu, K., Abdulganiyyu, H., Rabe, A.M. and Usman, A. (2013) *Nig. J. Basic Appl. Sci.*. 21(3): 193-196