European Journal of Applied Sciences – Vol. 10, No. 3
Publication Date: June 25, 2022
DOI:10.14738/aivp.103.12456.
Sobowale, A. A., Agbane, J., Bamkefa, B. A., & Aduramigba-Modupe, A, O. (2022). Fungitoxic Potentials of *Ocimum Gratissimum* L. *and Zingiber Officinale* (L.) Roscoe Extracts Against Fungi Associated with Postharvest Rot of *Carica Papaya* L. European Journal of Applied Sciences, 10(3). 659-670.

Fungitoxic Potentials of *Ocimum Gratissimum* L. *and Zingiber Officinale* (L.) Roscoe Extracts Against Fungi Associated with Postharvest Rot of *Carica Papaya* L.

Sobowale Ayodele Adegboyega
Department of Botany, University of Ibadan, Ibadan, Nigeria

Agbane John
Department of Botany, University of Ibadan, Ibadan, Nigeria

Bamkefa Bukola Ayodeji
Department of Biological Sciences, Lead City University, Ibadan, Nigeria

Aduramigba-Modupe Adefoyeke Olufunmilayo
Department of Crop Protection and Environmental Biology
University of Ibadan, Ibadan, Nigeria

ABSTRACT

The fungitoxic potentials of Ocimum gratissimum and Zingiber officinale extracts on fungi associated with rotting Carica papaya were investigated. Rotting and healthy pawpaw species of Carica papaya were obtained from Bodija market, Ibadan, Nigeria. Isolation of fungi from the rotting fruit was done using standard procedures. Pure cultures of the isolated fungi were obtained on Acidified Potato Dextrose Agar (APDA). Leaves of O. gratissimum and rhizome of Z. officinale were obtained from Botanical Garden, University of Ibadan, Nigeria. Crude extracts (aqueous and methanol) of the plants were obtained using standard procedures. The antimicrobial potentials of the extracts on the isolated fungi in vitro at concentrations of 25%, 50%, 75% and 100% were evaluated. Their interactive effects on the pathogens were also evaluated. Cultures with 0% methanol and water served as controls. All experiments were done in triplicates. Incubation of all Petri plates were done at room temperature for 7 days. Radial and diametric growth of the fungi was measured every 24 hours using a meter rule. Data obtained were subjected to analysis using SAS (version 9.2). Mean separation was done using Duncan Multiple Range Test (DMRT) at p≤0.05). The isolated fungi were Colletotrichum gloeosporioides, Lasiodiplodia theobromae, and Aspergillus flavus. Inhibitory effects of extracts of both plants and of both solvents on the isolated pathogens were significantly better than in control. Inhibitory effect of the methanol extracts was better than that of aqueous extracts. Inhibitory effect of O. gratissimum extract was also significantly better than that of Z. officinale (p≤0.05). Inhibition at all extract concentrations was significantly better than in control. However, inhibition at 100% and 75% was higher than that at other concentrations. Inhibition obtained with the different treatment combinations was better than that with individual concentrations. Generally, inhibition of Lasiodiplodia theobromae was significantly better than that of Aspergillus flavus, which in turn was significantly better than that of Colletotrichum gloeosporioides (p≤0.05). Ocimum

gratissimum and Zingiber officinale could be said to possess promising fungitoxic potentials. More work is required to ascertain their potentials in vivo.

Keywords: Carica papaya, postharvest rot, fungitoxicity, Ocimum gratissimum, Zingiber officinale

INTRODUCTION

Pawpaw (*Carica papaya*) is an indigenous, multi-purpose, perennial herbaceous plant that is cultivated mainly for its fruit. It is cultivated and consumed throughout the tropical and warmer subtropical areas of the world. Major producers of the fruit include Mexico, Brazil Germany including Nigeria [1]. However, the ripe fruits are known to be susceptible to attack by microorganisms such as fungi, bacteria, nematodes and viruses including rot pathogens. Spoilt pawpaw fruits are characterized by excess softening, mycelia growth, loss of moisture, unpleasant odour, shrinkage and total drying up of water in the fruits [2]. Infection has been reported to occur during growth season, harvesting, handling, transport, postharvest storage and/or after purchasing by the consumer. [3], [4], [5], [6].

Generally, biological control of postharvest diseases including the use of botanicals has continued to gain increasing attention [7]. Basil (*Ocimum gratissimum*), is famous for its local cultivation in Africa. In Nigeria, it is commonly used as spice and its medicinal properties are well known among the locals [8], [9]. It has been reported to exert inhibitory effect against different fungal pathogens. Ginger (*Zingiber officinale*), a creeping perennial rhizome, is also famous for its several medicinal properties. It is as well widely used around the world in foods as a spice [10], [11], [12], [13], [14], [15]. According to FAO [16], Nigeria is reputed to be among the largest consumers of the rhizome in the world. Extracts from the rhizome is known to show varying antimicrobial activities. Fresh juice of the rhizome has also been reported to exert inhibitory action against several fungal pathogens [17]. Very many plants, including basil and ginger, as well as their parts have been reported to show antimicrobial activities because they have some naturally occurring substances which play an effective role in plant disease resistance [18]. Antimicrobial potentials of plant extracts against plant diseases have been severally documented. However little has been done on the interactive effects of these extracts on plant diseases. The experiment examined the interactive effects of crude extracts of *Zingiber officinale* and *Ocimum gratissimum* on fungi associated with rots of *C. papaya.*

METHODOLOGY

Collection of samples

Diseased pawpaw was identified by physical examination using the method of Balali *et al*. [19]. Rotting pawpaw fruits were purchased from Bodija market, kept in sterile polythene bags and brought to the Plant Pathology Laboratory, Department of Botany, University of Ibadan. *Zingiber officinale* (rhizome) and *Ocimum gratissimum* (leaves) were obtained from the same market and later verified and authenticated in the Herbarium, Department of Botany, University of Ibadan, Nigeria.

Isolation and identification of fungi

The pawpaw samples were washed with sterile distilled water and surface-sterilized with 70% methanol. Rotting sections from the samples were cut with sterilized scalpel and surface-sterilized with 1% sodium hypochlorite for 30 seconds and rinsed in five changes of sterile

distilled water. They were dried with sterile paper towel and later inoculated on Petri plates of Potato Dextrose Agar (PDA). All plates were incubated at 28°C for 7 days and observed daily for fungal growth. The fungi were sub-cultured to obtain pure cultures which were later put on PDA slants. Morphological characterization and identification of all isolated fungi was done following standard procedures [20], [21], [22], [23]. Pathogenicity tests was conducted for the isolated fungi following standard procedures.

Preparation of plant extracts

Fresh bulbs of ginger (*Zingiber officinale*) and fresh fully expanded leaves of African basil (*Ocimum gratissimum*) were washed thoroughly under running tap water and later soaked in 1% Sodium hypochloride for 30 seconds before rinsing in 5 exchanges of sterile distilled water. They were air dried at room temperature for two weeks and then grinded to get fine powder. The powdered samples were added to a different conical flasks containing the extraction solvent (sterile distil water and methanol). The mixtures were allowed to stand for 48 hours with periodic shaking using rotary shaker in order to homogenize. Filtration was done through a double layered muslin cloth and No. 1 Whatman filter paper prior to evaporation. The filtrate was dried and concentrated using rotary evaporator. The dried extract was stored in sterile bottle at -20°C for further use.

Evaluation of effect of the plant extracts on growth of the fungal isolates

The extracts were prepared at various concentrations of 25%, 50%, 75%, and 100% represented as 0.025 g/ml, 0.05 g/ml, 0.075 g/ml and 0.01 g/ml. For each extract, 2ml of each concentration was transferred into empty sterile Petri plate under sterile conditions, after which 15ml of molten PDA was dispensed into each Petri plate under sterile conditions. The plates were gently rotated to ensure even dispersion of the extracts and then allowed to gel. Each fungus was then inoculated at the center of each Petri plate. This was done by inoculating a 4 mm diameter mycelia disc of 7 days old culture of each of the three test fungi. Petri plates with 0% extract served as control. All experiments were done in triplicates and incubation was done at 28°C for 7 days. Diametric and radial growth of the growing fungi was measured at 24 hours interval for 7 days using meter rule along two perpendicular lines drawn on the reverse of the plates. Effects of the extracts on growth of the fungi was examined using the method of Sangoyomi [24]. The effects aqueous and methanol extracts of ginger *Zingiber officinale* (Zo) and basil *Ocimum gratissimum* (Og), as well as their interactive effects on growth of the isolated fungi was examined at different concentrations, viz., *Zo*25%+*Og*25%, *Zo*25%+ *Og*50%, *Zo*25% + *Og*75%, *Zo*25%+ *Og*100%, and *Zo*50% + *Og*25%, *Zo*50%+ *Og*50%, *Zo*50%+ *Og*75%, *Zo*50%+ *Og*100%, and *Zo*75%+ *Og*25%, *ZO*75%+ *Og*50%, *Zo*75%+ *Og*75%, *Zo*75%+ *Og*100%, and *Zo*100%+ *Og*25%, *Zo*100%+ *Og*50%, *Zo*100%+ *Og*75%, and *Zo*100%+ *Og*100%.

Data Collection and analysis

Radial and diametric growth of the fungi were measured daily using a meter rule while growth inhibition was measured using the formula below:

Growth inhibition (%) = [(DC- DT)/DC] **X** 100

Where DC = average diameter of control

DT = average diameter of fungi colony with treatment

All data were subjected to analysis of variance analysis (ANOVA) using General Linear Model (GLM) procedure of SAS (version 9.2). Means were separated using Duncan's Multiple Range Tests (DMRT) at p≤0.05.

RESULTS

Isolated fungi and effects of the plant extracts

The fungi isolated from the diseased pawpaw fruit include *Colletotrichum gloeosporiodes* (Plate 1), *Botrydiplodia theobromae* (Plate 2), and *Aspergillus flavus* (Plate 3). Their pathogenicity was confirmed in Plate 4. Table 1 shows the overall effects of *Z. officinale* and *O. gratissmum* extracts of different extraction solvents, on mycelia growth of *Collectotrichum gloeosporioides, Lasiodiploda theobromae* and *Aspergillus flavus*, after 7 days of incubation. Generally, inhibitory effect of the methanol extracts was significantly higher than that of aqueous extracts. Inhibitory effect of *O. gratissimum* extract was significantly better than that of *Z. officinale* (p≤0.05). However inhibitory effects of extracts of both solvents were significantly better than in control (Figure 1). Table 2 shows the overall impact of concentration on inhibition potential of *Ocimum gratissimum and Zingiber officinale* extracts against the isolated fungi at different incubation days. Generally, growth inhibition at all the concentrations was significantly better than in control. Inhibition at 100% and 50% was also higher than that at other concentrations (p≤0.05). Table 3 gives the overall growth inhibition of the isolated fungi by different concentrations of the extracts. Generally, growth inhibition at 100% concentration was significantly higher than that at other concentrations. Inhibition at 50% was also significantly higher than that at 75% concentration, which in turn gave significantly higher inhibition than that at 25% concentration (p≤0.05).

Table 4 shows the overall growth inhibition of each isolated fungus by all the extracts. Generally, inhibition of *Lasiodiplodia theobromae* was significantly better than that of *Aspergillus flavus,* which in turn was significantly better than that of *Colletotrichum gloeosporioides* (p≤0.05).

Table 5 gives the interactive effects of 25% *Z. officinale* (Zo) and all concentration levels of *O. gratissimum* (Og) on growth (cm) of the isolated fungi. Generally, 25%Zo+50%Og gave the best inhibition followed by 25%Zo+25%Og compared to other treatment combinations. Table 6 shows the interactive effects of 50% *Z. officinale* (Zo) and all concentration levels of *O. gratissimum* (Og) on growth of the isolated fungi. Generally, 50%Zo+50%Og gave the best inhibition compared to other treatment combinations. Table 7 gives the interactive effect of 75% *Z. officinale* (Zo) and all the concentration levels of *O. gratissimum* (Og) on growth of the isolated fungi. Generally, 75%Zo+50%Og gave the best inhibition followed by 75%Zo+25%Og compared to other treatment combinations. Table 8 gives the interactive effects of 100% *Z. officinale* (Zo) and all concentration levels of *O. gratissimum* (Og) on growth of the isolated fungi. Generally, 100%Zo+100%Og gave complete growth inhibition of the pathogens followed by 100%Zo+50%Og compared to other treatment combinations. Table 9 gives ANOVA table for fungitoxic activities of *O. gratissimum* and *Z. officinale* extracts on the isolated fungi. The F values for concentration, day, organism, and treatment (P>0.0001) were all highly significant. The F values (P>0.0001) for interaction between organism and concentration, treatment and concentration, organism and day, treatment and day, organism and treatment and also concentration and day (P>0.0037), were all highly significant. The F values (P>0.0001) for interaction among organism, concentration and day, treatment, concentration and day,

organism, treatment and concentration as well as organism, treatment and day were all also highly significant.

DISCUSSION

The fungitoxicity of *Ocimum gratissimum* and *Zingiber officinale* extracts against *Colletotrichum gloeosporiodes Lasiodiplodia theobromae*, *Aspergillus flavus* isolated from the rotting pawpaw fruit (C*. papaya*) underscored their growth inhibitory potentials against the fungal pathogens. *Colletotrichum gloeosporiodes Lasiodiplodia theobromae* and *Aspergillus flavus* amongst others have been reported to be the causal agents of postharvest rot of pawpaw fruit in storage [25]. The extracts of *Ocimum gratissimum* and *Zingiber officinale* have been reported to have anti-microbial and anti-fungal properties of which their derivatives are of great importance in public health, cosmetics, medicine and agriculture [26]. The higher significant growth inhibition by *Z. officinale* and *O. gratissmum* extracts of both extraction solvents compared to control shows the appropriateness of the two solvents (methanol and water) as extraction solvents for the extracts of *Z. officinale* and *O. gratissmum.* Both extraction solvents could thus be said to be promising extraction solvents especially for leaves of *Ocimum gratissimum* and of *Zingiber officinale* rhizomes. However, the different results obtained with different concentrations of extracts obtained with different extraction solvents agree with the work of Azwanida [27] who reported that different plants require certain extraction method in order to ensure the antifungal properties of the plant employed.

The significantly higher growth inhibition by all the extracts at all concentrations compared to control underscores the promising fungitoxic potentials of extracts of the two plants against the pathogens, even at low concentrations. Results obtained with extracts of the different concentrations might be indicative of 100% and 50% as preference extract concentration over the others. However, overall growth inhibition of the three pathogens (*Lasiodiplodia theobromae, Aspergillus flavus, Colletotrichum gloeosporioides),* shows the higher sensitivity of *Lasiodiplodia theobromae,* and *Aspergillus flavus* to the extracts compared to *Colletotrichum gloeosporioides.* This points to high chances of obtaining better growth inhibition of *Lasiodiplodia theobromae,* and *Aspergillus flavus* results with these extracts. The improved growth inhibition obtained with various treatment combinations underscores the preference of treatment combinations over individual treatments for more effective inhibition of the pathogens. This shows that synergy between the different treatment combinations will most likely give better growth inhibition of the pathogens than the individual treatments.

The highly significant F value (P>0.0001) for concentration showed the critical impact of extract concentration on effectiveness of the extracts in inhibiting the isolated pathogens. It explained the significant growth inhibition of some of the pathogens at 100% and 50% extract concentration compared to other concentrations. Onuh *et al*. [28], in their experiment reported increase in fungi toxicity of plants extracts at higher concentrations. The highly significant F value (P>0.0001) for organism showed that toxicity of the isolated pathogens to the extracts differed significantly from one pathogen to the other. This means the fungal growth did not respond to the extracts the same way. The highly significant F values (P>0.0001) for day and treatment shows that inhibition of the pathogens by the extracts also differed significantly among days of incubation and treatments. Significant F value (P>0.0001) for interaction between organism and concentration means that growth inhibition of each of the pathogen differed significantly from one extract concentration to the other. Significant F value

(P>0.0001) for interaction between treatment and concentration means that effectiveness of any particular treatment depended significantly on the concentration of the extract. Significant F value (P>0.0037) for interaction between concentration and day means that effectiveness of most of the extract concentrations in growth inhibition of the pathogens depended significantly on the day of incubation. Duration of contact between each pathogen and extract could therefore be said to play a critical role in effective growth inhibition of the pathogen. Significant F values (P>0.0001) for interaction between organism and day, as well as treatment and day, also indicated the significant role of duration of contact between pathogen and extract in effective growth inhibition of the pathogen by each of the treatments. However, significant F value (P>0.0001) for interaction between organism and treatment showed that growth inhibition of each of the pathogens differed significantly from one treatment to the other. Significant F value (P>0.0001) for interaction among treatment, concentration and day showed that effective growth inhibition of any particular pathogen (*Colletotrichum gleosporiodes or Lasiodiplodia theobromae* or *Aspergillus flavus*) by any of the treatments depended significantly on the extract concentration and day of incubation. This is also shown by the significant F value (P>0.0001) for interaction among organism, treatment and concentration.

CONCLUSION

The leaves of *Ocimum gratissimum* and of *Zingiber officinale* rhizomes could be said to possess promising antimicrobial potentials against fungi associated with rotting *C. papaya* such as *Colletotrichum gleosporiodes* and *Lasiodiplodia theobromae*. Leaves *Ocimum gratissimum* might possess better inhibitory potential than *Zingiber officinale*. Extract concentration of 100% and 75% give better growth inhibition than 50% and 25%. However, a lot of work must still be done in the field to ascertain the fungal growth inhibitory potentials of extracts from these plants *in vivo*.

References

[1] FAO, 2004. FAO Statistics. Food and Agricultural organization, Rome, Italy. The International Tropical Fruits Network (TFNet),

[2] Frazier, W.C. and Westhoff, D.C. 1995. Food Microbiology 4th Edition, McGraw Hill, New Delhi, pp.83-98.

[3] Snowdon, A.L., 1990. A Color Atlas of Postharvest Diseases and Disorder of Fruits and Vegetables, Vol. I: General Introduction and Fruits. Wolfe Scientific, London.

[4] Singh, D. and Sharma R. R. 2007. Postharvest disease of fruit and vegetables and their management. In: Prasad, D edition sustainable pest management. Daya Publishing House, New Delhi, India.

[5] Barth, M; Hankinson, T.R; Zhuang H. and Breidt F. 2009. Microbiological spoilage of fruits and vegetables. W.H. Sperber, M.P. Doyle (editions), Compendium of the Microbiological Spoilage of Foods and Beverages, Food Microbiology and safety. C Springer Science + Business Media, LLC, pp13-183.

[6] Chukwuka, K.S., Okonko, I.O. and Adekunle A.A. 2010. Microbial Ecology of Organisms Causing Pawpaw (*Carica Papaya* L.) Fruit Decay in Oyo State, Nigeria. American- Eurasian Journal of Toxicological Sciences, 2(1): 43-50.

[7] Wilson, C.L. and Wisniewski, M.S., 1989. Biological control of postharvest diseases of fruits and vegetables: an emerging technology. Annual Review of Phytopath. 27: 425-441.

[8] Okigbo RN, Okorie, R, Puthefi R. 2009. *In-vitro* effects of Garlic (Allium sativum L.) and African Basil (*Ocimum gratissimum*) on pathogens isolated from rotted cassava root. 34:742-747.

[9] Euloge. A, Edwige. A, Soumanon. M. 2012. Journal of microbiology biotechnology and food extracts. Food and nutrition sciences, 6, 1381-1394.

[10] Awang, D.V.C. 1992. Ginger. Can. Pharm. J. 125, 309–311.

[11] Wang, P.H., Chung, C.Y., Lin, Y.S. and Yeh, Y. 2005. Use of polymerase chain reaction to detect the soft rot pathogen, *Pythium myriotylum,* in infected ginger rhizomes. Letters Appl. Microbiol. **36:** 116–120.

[12] Khair-ul-Bariyah, S., Ahmed, D., Ikram, Aujla, M. 2012. Comparative Analysis of *Ocimum basilicum* and *Ocimum sanctum* : Extraction Techniques and Urease and alpha – Amylase inhibition. Pak. J. Chem., 2(3):1-8.

[13] Zaker, M. 2014. Antifungal Evaluation of Some Plant Extracts in Controlling *Fusarium solani*, the Causal Agent of Potato Dry Rot In vitro and In vivo. Inter. J. Agri. Biosci., 3(4): 190-195.

[14] Sharma S, Vijayvergia R, Singh T. 2010. Evaluation of antimicrobial efficacy of some medicinal plants. J Chem.Pharm. Res. 2 (1):121-124.

[15] Ogbuehi, H. C., Emeribe, E. O., & Asagwara, J. O. 2016. ER (*Zingiber officinale*) extracts on shelf life of eggplant (*Solanum melogena* L.), 171–178.

[16] FAO. 2012. Feedipedia. Animal feed resources information system. http://www.fao.org

[17] Kapoor A. 1999. Antifungal activities of fresh juice and aqueous extracts of turmeric and ginger (*Zingiber officinale*). J Phytological Res. 10:59.

[18] Srivastava D, Kumari A, Misra N. 2009. Fungal spoilage of some fruits and screening of some leaves for fungitoxicity. Proc Nat Acad Sci Sect B, 79: 421-430.

[19] Balali, G. N., S. M. Scott, E.S. Whisson, D.L. and Wicks, T.J. 1995. Anastomosis group and pathogenicity of isolate of *Rhizoctonia solani* potato crop in South Australia. 3rd ed.

[20] Sutton B.C. 1973. Coelomycetes. In Ainsworth G.C., Sparrow F.K., Sussman A.S. (Eds.) The Fungi: An Advanced Treatise. Academic Press. New York. USA. pp 513-574.

[21] Nelson P.E., Toussoun T.A., Marasas W.F.O. 1983. *Fusarium Species*: An Illustrated Manual for Identification. Pennsylvania State University. Univer- sity Park, PA, USA. 193 pp.

[22] Oyeleke, A. and Manga, S.B. 2008. Essential of Laboratory Practice, 3rd edition, Tobest Publishers, Minna, Niger State, Nigeria, Pp.12- 29.

[23] Watanabe, T. 2002. Morphologies of cultured fungi and key to species. In: pictorial Atlas of soil and seed fungi (Ed.). haddad, S., dery, E., Norewitz, B.E. and Lewis, R (Ed.) (CRC Press LLC, N.W. Corporate Blvd., Boca Raton, florida 33431. 1- 48.

[24] Sangoyoni T.E. 2004. Post- Harvest Fungal Deterioration of Yam (*Dioscorea rotundata* Poir) and its control. Thesis. Inter- national Institute of Tropical Agriculture. Ibadan, Nigeria. 179 pp.

[25] Okigbo R.N. 2005. Biological control of postharvest fungal rot with *Bacillus subitlis*. Mycopathologia 159 (2):307-314

[26] Coyne, R. S., Nicholas, A. Stover and wei, Miao. 2012. Whole Genome studies of Tetrahymena. 109:53-81.

[27] Azwanida, N. N. 2015. A Review on the Extraction Methods use in Medicinal plants, principle, strength and limitation. Medicinal of Aromatic Plants.4: 196, doi: 10.4172/2167-0412.1000196. Faculty of Agro- based industry, University of Malaysia Kelantan, Jeli, Kelantan, Malaysia.

[28] Onuh, J., Shrikiti, D., Ubwa, S. and Shambe, T. 2015. Isolation of six microorganisms from rotten. *Dioscorea alata* (water yam), and antimicrobial sensitivity test with nine plant extracts. Food and nutrition sciences, 6, 1381-1394.

Table 1: Overall effects of the plant extracts on growth (cm) of all the isolated fungi

Solvents	Treatment	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Aqueous	*Z. officinale* extract	1.01^{b}	0.97^{b}	1.20^{b}	1.49^{b}	1.77^{c}	1.89^{c}	1.86^{c}
	O. gratissimum extract	0.65^{c}	0.87^{b}	1.04^{bc}	1.14^{bc}	1.84^{c}	1.44^{cd}	1.53^{cd}
	Z. officinale + *O. gratissimum*	1.31^{a}	1.81^{a}	2.34^{a}	2.97^{a}	3.71^{a}	4.18^{a}	4.70^{a}
Methanol	*Z. officinale* extract	0.42^{de}	0.49^{cd}	0.62^{de}	0.77^{c}	0.78^{de}	0.85^{ef}	0.92^{de}
	O. gratissimum extract	0.22^{ef}	0.28^{de}	0.28^{ef}	0.28^{d}	0.33^{de}	0.33^{fg}	0.33^{ef}
	Z. officinale + *O. gratissimum*	0.12^{f}	0.14^{e}	0.14^{f}	0.16^{d}	0.16^{e}	0.17^{g}	0.18^{f}

Means with different letters in a column are significantly different (p≤0.05)

Table 2: Impact of concentration on inhibition potential of *Ocimum gratissimum and Zingiber officinale* extracts against the isolated fungi at different incubation days

Parameters	Variable	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day7
Concentration	25%	0.76^{ab}	0.95^{abcde}	1.23^{bcde}	1.24^{edfg}	1.67^{bcd}	1.82^{cde}	1.87^{def}
	50%	0.66^{ab}	0.73^{de}	0.87^{cde}	1.15^{efg}	1.30^{cd}	1.32^{de}	1.41^{ef}
	75%	0.77^{ab}	0.91^{bcde}	1.05^{bcde}	1.16^{efg}	1.60^{cd}	1.41^{de}	1.41^{ef}
	100%	0.52^{b}	0.66^{de}	0.81^{de}	0.97^{fg}	1.00^{d}	1.06^{cd}	1.10^{f}
	Control	0.99^{edefg}	1.07^{ghi}	1.33^{fghi}	1.85^{hig}	2.11^{hi}	2.15^{ijk}	2.32^{f}

Means with different letters in a column are significantly different (p≤0.05)

Table 3: Overall growth inhibition of the isolated fungi by different concentrations of the extracts

Percentage concentration of all extracts	Average mean (cm)
25%	1.39^{a}
50%	1.07^{c}
75%	1.20^{b}
100%	0.88^{d}

Means with different letters in a column are significantly different (p≤0.05)

Table 4: Overall growth inhibition of each isolated fungus by all the extracts

ISOLATED FUNGI	AVERAGE MEAN (cm)
Colletotrichum gleosporiodes	2.04[a]
Lasiodiplodia theobromae	0.59[c]
Aspergillus flavus	0.77[b]

Means with different letters in a column are significantly different (p≤0.05)

Table 5: Interactive effects of 25% *Zingiber officinale (*Zo) and all concentration levels of *Ocimum gratissimum* (Og) on growth (cm) of the isolated fungi

Conc.	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
25%Zo+ 25%Og	0.97[a]	1.33[ab]	1.60[ab]	1.92[abcd]	2.27[abc]	2.62[abc]	2.85[abcd]
25%Zo+ 50%Og	0.65[ab]	0.83[cde]	1.07[bce]	1.35[cdefg]	1.55[cd]	1.73[cde]	2.00[cdef]
25%Zo+ 75%Og	0.93[a]	1.32[ab]	1.98[a]	2.48[a]	3.10[a]	3.27[a]	3.52[a]
25%Zo+ 100%Og	0.73[ab]	1.07[abcd]	1.43[abc]	1.65[bcdef]	2.22[abc]	2.55[abc]	2.95[abc]

Means with different letters in a column are significantly different (p≤0.05)

Table 6: Interactive effects of 50% *Zingiber officinale (*Zo) and all concentration levels of *Ocimum gratissimum* (Og) on growth (cm) of the isolated fungi

Conc.	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
50%Zo+25%Og	0.82[ab]	1.07[abcd]	1.32[bcd]	1.52[bcdef]	1.80[bcd]	2.07[bcd]	2.27[bcde]
50%Zo+50%Og	0.47[b]	0.55[e]	0.65[e]	0.75[g]	0.87[de]	0.95[e]	1.03[f]
50%Zo+75%Og	0.83[ab]	1.67[abcd]	1.58[ab]	2.07[abc]	2.70[ab]	3.20[a]	3.45[a]
50%Zo+25%Og	0.82[ab]	1.07[abcd]	1.32[bcd]	1.52[bcdef]	1.80[bcd]	2.07[bcd]	2.27[bcde]

Means with different letters in a column are significantly different (p≤0.05)

Table 7: Interactive effects of 75% *Zingiber officinale* (Zo) and all concentration levels of *Ocimum gratissimum* (Og) on growth (cm) of the isolated fungi

Conc.	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
75%Zo+ 25%Og	0.50^{b}	0.77^{de}	1.00^{bcde}	1.30^{defg}	1.68^{bcd}	2.00^{cd}	2.37^{bcde}
75%Zo+ 50%Og	0.83^{ab}	0.97^{abcde}	1.17^{bcde}	1.32^{defg}	1.60^{cd}	1.83^{cde}	2.00^{cdef}
75%Zo+ 75%Og	0.92^{a}	1.30^{abc}	1.55^{ab}	2.18^{ab}	2.68^{ab}	2.98^{ab}	3.27^{ab}
75%Zo 100%Og	0.78^{ab}	0.95^{abcde}	1.22^{bcde}	1.67^{bcdef}	2.17^{abc}	2.50^{abc}	2.82^{abcd}

Means with different letters in a column are significantly different (p≤0.05)

Table 8: Interactive effects of 100% *Zingiber officinale* (Zo) and all concentration levels of *Ocimum gratissimum* (Og) on growth (cm) of the isolated fungi

Concentration	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
100%Zo+ 25% Og	0.92^{a}	1.40^{a}	1.53^{ab}	1.82^{abcde}	2.22^{abc}	2.17^{bcd}	2.77^{abcd}
100%Zo+ 50% Og	0.48^{b}	0.78^{de}	1.00^{bcde}	1.23^{defg}	1.50cd	1.70^{cde}	1.93^{cdef}
100%Zo+ 75%Og	0.95^{a}	1.10^{abcd}	1.37^{bcd}	1.92^{abcd}	2.33^{abc}	2.67^{abc}	2.93^{abc}
100%Zo+100%Og	0.00^{c}	0.00^{f}	0.00^{f}	0.00^{h}	0.00^{e}	0.00^{f}	0.00^{g}

Means with different letters in a column are significantly different at (p≤0.05) level

Table 9: ANOVA table for fungitoxic activities of *Z. officinale* and *O. gratissium* on the fungi isolated from rotting *Carica papaya*

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Concentration	3	51.7990966	17.26636554	66.22	0.0001**
Day	6	124.9294492	20.82157494	79.86	0.0001**
Organism	2	629.8967128	314.9483564	1207.91	0.0001**
Replicate	2	0.2490315	0.1245158	0.48	0.6204
Treatment	5	660.7050170	132.1410034	506.79	0.0001**
Concentration*Day	18	10.0605846	0.5589214	2.14	0.0037**
Organism*Concentration	6	46.3161195	7.71935325	29.61	0.0001**
Concentration*Replicate	6	1.4362427	0.2393738	0.92	0.4810
Treatment*Concentration	15	142.9743066	9.53162044	36.56	0.0001**
Organism*Day	12	90.6891363	7.55742803	28.98	0.0001**
Day*Replicate	12	1.6513061	0.1376088	0.53	0.8977
Treatment*Day	30	58.0419370	1.93473120	7.42	0.0001**
Organism*Replicate	4	0.4892658	0.1223164	0.47	0.7584
Organism*Treatment	10	555.4402044	55.54402044	213.03	0.0001**
Treatment*Replicate	10	2.9810969	0.2981097	1.14	0.3262
Organism*Concentration*Day	36	23.0323019	0.63978629	2.45	0.0001**
Concentration*Day*Replicate	36	6.5586669	0.1821852	0.70	0.9095
Treatment*Concentration*Day	90	53.4121383	0.59346823	2.28	0.0001**
Organism*Concentration*Replicate	12	5.9339125	0.4944927	1.90	0.0312
Organism*Treatment*Concentration	30	179.0006271	5.96668764	22.88	0.0001**
Treatment*Concentration*Replicate	30	6.6980815	0.2232694	0.86	0.6894
Organism*Day*Replicate	24	3.5954160	0.1498090	0.57	0.9500
Organism*Treatment*Day	60	65.1469969	1.08578328	4.16	0.0001**
Treatment*Day*Replicate	60	9.5386222	0.1589770	0.61	0.9916
Organism*Treatment*Replicate	20	4.6533867	0.2326693	0.89	0.5974
Error	474	939.173	1.981		
Corrected total	503	2974.730			
R square	0.6843				

*** = Significant, ** = Highly Significant**

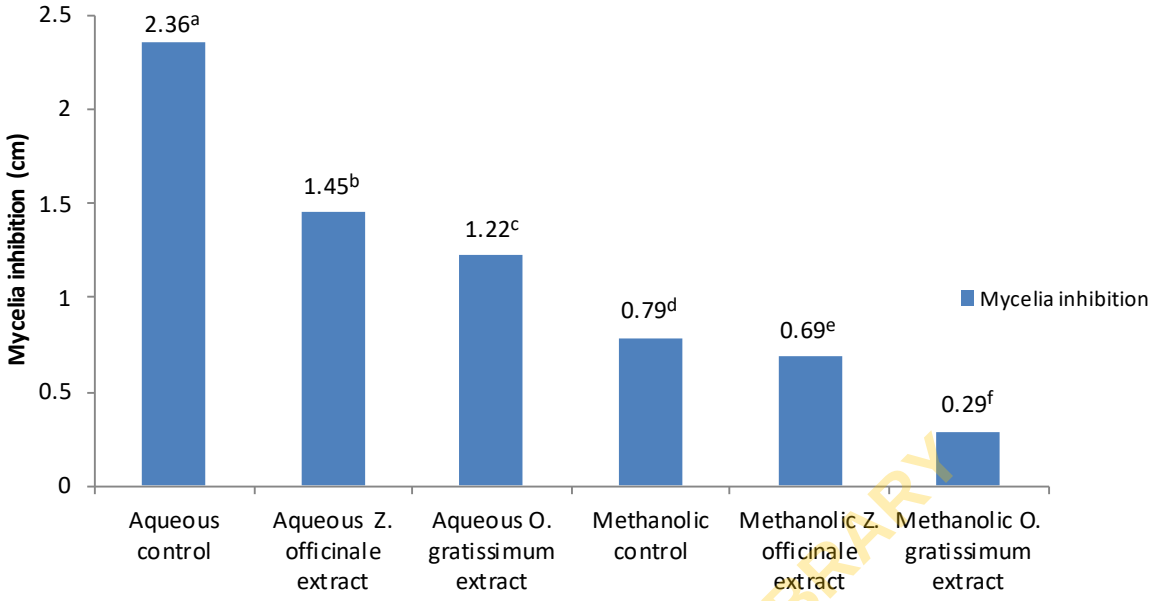

Figure 1: Overall growth inhibition of all isolated fungi by the extracts.