

**MITIGATING FRUIT ROT PATHOGENS OF PINEAPPLE WITH AQUEOUS EXTRACTS OF DATURA STRAMONIUM, ZINGIBER OFFICINALE, KHAYA GRANDIFOLIOLA, HYPIS SUAVEOLENS AND CALOPHYLLUM INOPHYLLUM**

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**ABSTRACT**

Antifungal effects of aqueous extracts of *Hyptis suaveolens* (Pignut), *Khaya grandifoliola* (large leave khaya), *Zingiber officinale* (Ginger), *Calophyllum inophyllum* (Mast wood) and *Datura stramonium* (Jimson weed), on the radial growth of fungal pathogens of pineapple fruit was investigated, varied concentrations of extracts from the different plant parts were tested against the rot organisms, the highest aqueous extract inhibitory effect of *Zingiber officinale* was on *Aspergillus nidulans* (26.00%) at 1.0g/mL while the least inhibitory effect was on *Colletotrichum fruticola* (22.00% ) at 1.0g/mL, the highest aqueous extract inhibition of *Datura stramonium* was on *Aspergillus niger* (14.00%) at 1.0g/mL while the least inhibitory effect was (0.00%) on *Aspergillus flavus* and *Colletotrichum fruticola* at all concentrations, the highest aqueous extract inhibition of *Calophyllum inophyllum* was (15.00%) on *Aspergillus fumigatus* and *Aspergillus nidulans* at 1.0g/mL while the least inhibitory effect was 12.00% on *Aspergillus tubingensis* at 1.0g/mL, the highest aqueous extract inhibition of *Hyptis suaveolens* was 18.00% on *Trichoderma harzianum* and *Aspergillus fumigatus* at 1.0g/mL while the least inhibitory effect was 10.00% on *Aspergillus niger* at 1.0g/mL, the highest aqueous extract inhibition of *Khaya grandifoliola* was 18.00% on *Aspergillus flavus* at 1.00g/mL while the least inhibitory effect was 10.00% on *Aspergillus niger* at 1.0g/mL, the fungitoxic potential of these plant extracts on rot inducing fungi of pineapple fruit is an indication of their use by farmers as alternative to commercial or synthetic fungicide that pose danger to our ecosystem and health. The effects of rot organisms on the proximate composition of the fruit were not significant on the nutrient level of the fruit.

**Keywords:** Pineapple, Fruit Rot Pathogens, Proximate Composition, Plant Extracts.

**1. INTRODUCTION**

Pineapple (*Ananas comosus* L. Merr.) belongs to the family Bromeliaceae. Pineapple is a non-climacteric, parthenocarpic, multiple fruit often called as syncarp or sorosis, and composed of some 100–200 berry-like fruitlets, sometimes called eyes, attached to a central core (Assumi, *et al.*, 2021). Pineapples are commercially grown over a wide range of latitude from 30° N in the northern hemisphere to 33° 58S in the South (Manik *et al.*, 2019). The fruits can be harvested from May to June or November to January. The fruits with crown can be kept for 10-15 days after harvesting (Jonathan, 2025).

Pineapple is the third most important tropical fruit in the global market after banana and citrus. Pineapple is produced for both fresh consumption and processing (Manasij Das and Swarnali Bhattacharya (2024) and fruits are often called nature's wonderful medicines, rich in vitamins, minerals, anti-oxidants and phyto-nutrients without which human body cannot maintain proper health and develop resistance to disease (Lobo and Siddiq, 2017). Pineapple fruit is widely distributed in tropical regions. In recent years, it has become one of the most demanded exotic fruits (FAOSTAT, 2017). It is also used in food processing for meat tenderization and as a dietary supplement (Mohanty *et al.*, 2016).

In Nigeria, pineapple is produced in many states of South West Nigeria. North Eastern states produce major share of pineapples in Nigeria (Oniah and Edem, 2022). Even though, Nigeria is one of the major producers of fresh pineapples, its position in world export is significantly low, less than 2% of total production is used for processing (Oniah and Edem, 2022). The stem end rot (SER) infection slows down in lower temperature but continues to grow internally. SER disease of pineapple forms the major limiting factor during storage (Manik et al, 2019)

Post-harvest losses are costly in terms of money and manpower and these can be catastrophic for developing countries (Jainu, *et al.*, 2016). At present, black rot of pineapple is controlled by the application of fungicides. Despite high cost, the problem of pesticide residue is becoming acute, as they were reported to have carcinogenic, teratogenic, oncogenic, neurotoxic and genotoxic health hazards (Rohrbach *et al.*, 2003). In case of *C. paradoxa* the problem to device control strategies is multifarious due to its facultative pathogenic nature of infection (Reyes *et al.*, 2004). Spoilage is caused by bacteria, yeasts, fungi and moulds. Susceptibility of pineapples to microbial colonization is due to its differential chemical composition such as high level of sugar, low pH and its high water activity which favours the growth of microorganisms in pineapple is recognized as source of potential health hazard to man and animals, this is due to their production of toxins which are capable of causing disease like respiratory tract infections, meningitis, gastroenteritis, diarrhea in man following ingestion (Lobo and Siddiq, 2017).

## **2. MATERIALS AND METHODS**

### **Samples collection**

Pineapple fruit samples were purchased from a neighbourhood market in Ado-Ekiti, Nigeria. These were transported in sterile polythene bags to the laboratory of Science Laboratory Technology Department, Federal Polytechnic, Ado-Ekiti for fungal isolation and further analyses.

### **Isolation of pathogens**

The surface sterilized fruits with symptoms of diseases were then cut into 2 mm pieces and plated on to sterile potato dextrose agar (PDA) in Petri dishes supplemented with 250mg chloramphenicol to guide against bacterial invasion (Serag *et al.*, 2022). The plates were incubated at  $28 \pm 2^{\circ}\text{C}$  for 2-3 days and observed for fungal mycelial growth and emerging mycelia were later sub cultured into fresh PDA medium. The developed colonies were counted and repeatedly sub-cultured on PDA plates to get pure cultures. Pure isolates were later stored on PDA slants for identification, characterization and subjection to test plant for antifungal bioassay. Obtained pure isolates were identified on the bases of macro and micro morphological characteristics. Morphological characteristics of the fungi (mycelium coloration or pigmentation, presence or absence of septate, spore morphology) were recorded. In some cases, the infected

tissues were stained by cotton blue and lactophenol (McClenny, 2005) and observed under microscope. Morphological identification of fungi was based on the morphology of the fungal colony or hyphae, the characteristics of the spores and reproductive structures (Agrios, 2005).

### **Pathogenicity test**

Pathogenicity test was carried out using the techniques described by Akintobi *et al.* (2011). Healthy pineapple fruit samples were obtained from the market with zip lock bag and transported to Microbiology Laboratory of Ekiti State University, Ado Ekiti. The pineapple fruits were washed under running tap to eliminate contaminants from their surfaces. These were then surface sterilized in 1% NaOCl for three minutes. Thereafter, they were rinsed in three changes of sterile distilled water and wiped dry using a sterile blotting paper. A sterile inoculating needle containing fungal spore was used to punch the pineapple fruits. Each fungal isolates was inoculated into healthy pineapple fruits (Wani, 2006). Control was also set in the same manner without fungal isolate. Disease development was checked after 24 hours. The diameters of rot portion of the fruits were measured and each fungus was later re-isolated from the inoculated samples and the isolates were compared with the initial isolates.

### **Preparation of plant crude extracts**

Crude plant extracts were obtained from *Datura stramonium*, *Calophyllum inophyllum*, ginger rhizome (*Zingiber officinale*), leaf of *Hyptis suaveolens* and *Khaya grandifoliola*. The extraction process followed the procedure described by Handa *et al.* (2008). The leaves were washed under tap water, rinsed in three changes of sterile distilled water and dried using sterile blotting paper. The leaves were then placed and dried at a temperature of 40°C for three weeks. The rhizomes were also washed under tap water and rinsed in three changes of sterile distilled water. These were blotted dry using sterile blotting papers, peeled, cut into smaller pieces and placed in the oven at the same temperature for three days. All the plant materials were pulverized using sterile mortar and pestle in order to rupture the leaf tissues to release the active cellular contents. The extracts were placed in sterile specimen bottles. This was done to maximize the surface area which in turn enables the mass transfer of active ingredients from the plant material to the solvent.

Fifty (50gm) each of the test plant powder was placed in separate sterile conical flasks and 200mL of solvent (ethanol and aqueous) was added to each of the plant powder, ensuring that the powder was completely immersed into the solvent, this was then shaken vigorously at different intervals for two days, this was allowed to stand on the bench at room temperature. A sterile funnel was placed into a 500mL conical flask and then a Whatman's (No.1) filter paper was folded and placed into the funnel. The extract was poured gradually into the filter paper and allowed to trickle into the conical flask. The filtrate was then poured into sterile universal bottles. The crude extracts in the universal bottles were placed in a rotary evaporator for 60 minutes at 50°C to concentrate the extracts by evaporating the solvent. The concentrated crude extracts were dried in the oven at 40°C for two days until a powder like substance remained at the bottom of the universal bottles. The labeled universal bottles containing the powder were stored in the refrigerator at 4°C.

### **Preparation of inoculum**

The fungal inoculum was prepared from 5-day old culture grown on PDA medium. The Petri dishes were flooded with 8 to 10 ml of distilled water and the conidia were scraped using sterile spatula. The spore density of each fungus was adjusted with spectrophotometer ( $A_{595}$  nm) to obtain a final concentration of approximately  $10^5$  spores/mL according to Cheesbrough, (2002).

### **Effects of crude plant extracts on growth of fungal mycelia**

The fungicidal effects of the crude extracts in controlling pineapple fruit rots were evaluated on eight pathogenic fungal isolates namely: *Aspergillus flavus*, *Rhizopus sp*, *Trichoderma harzianum*, *Colletotrichum fruticola*, *Aspergillus niger*, *Aspergillus fumigatus*, *Aspergillus tubingensis* and *Aspergillus nidulans*.

### **Determination of the effects of the crude extracts on the fungal isolates**

The method of Amadioha and Obi (1999) was used to determine the effects of the crude extracts on the fungi. Different concentrations of the crude extracts were prepared by weighing separately 0.2, 0.4, 0.6, 0.8 and 1.0mg of *Datura stramonium*, *Calophyllum inophyllum*, ginger rhizome (*Zingiber officinale*), leaf of *Hyptis suaveolens* and *Khaya grandifoliola* powder respectively. Each powder was dissolved in 1ml sterile distilled water to form solutions of different concentrations. The Muller Hinton agar medium was prepared according to the manufacturer's instruction and this was autoclaved at the  $121^\circ\text{C}$  for 15 min. The media was poured into each Petri dish and set aside to solidify under the laminar hood. After solidifying the media, a sterile glass spreader was used to uniformly spread the inoculum throughout the medium. Then, 100  $\mu\text{L}$  of each extract was adjusted to the same concentration (50 mg/mL) and perforated filter papers (disc) were soaked for two hours before placing them on the agar plate. The agar plate was allowed to rest for 1 hour in the incubator at  $37^\circ\text{C}$  for a daytime.

The sensitivity of the test microorganisms was determined by assessing the diameter of the zone of inhibition in diameter. Inhibition percentage was taken thus:

$$\text{Growth inhibition (\%)} = \frac{(\text{DC} - \text{DT}) \times 100}{\text{DC}}$$

Where DC = Average diameter of colony with control

DT = Average diameter of colony with treatment.

### **Proximate Analysis**

The proximate composition of each pineapple fruit infected with each of the isolated fungus was determined according to AOAC, (2000)

#### **i. Moisture content determination**

Two grams (2.0g) of each sample was placed in an oven, maintained at  $100 - 103^\circ\text{C}$  for 16 hours with the weight of the wet sample and the weight after drying were noted. The drying was repeated until a constant weight was obtained. The moisture content was expressed in terms of loss in weight of the wet sample.

$$\% \text{ moisture content} = \frac{\text{weight of moisture}}{\text{weight of sample}} \times 100$$

**ii. Ash content determination**

Two grams (2.0g) of each of the oven-dried samples in powder form were accurately weighed and placed in crucible of known weight. These were ignited in a muffle furnace and ashed for 8 hours at 550°C. The crucible containing the ash was then removed, cooled in desiccators and weighed and the ash content expressed in term of the oven-dried weight of the sample.

$$\% \text{ Ash content} = \frac{\text{weight of ash} \times 100}{\text{weight of sample}}$$

**iii. Protein content determination**

The protein nitrogen in 1g of the dried samples was converted to ammonium sulphate by digesting with concentrated H<sub>2</sub>SO<sub>4</sub> and in the presence of CuSO<sub>4</sub> and Na<sub>2</sub>SO<sub>4</sub>. These were heated and the ammonia evolved was steam distilled into boric acid solution. The nitrogen from ammonia was deduced from the titration of the trapped ammonia with 0.1M HCl with Tashirus indicator (double indicator) until a purplish pink color was obtained. Crude protein was calculated by multiplying the value of the deduced nitrogen by the factor 6.25mg.

**iv. Crude fibre content determination**

Two grams of each sample was weighed into separate beakers, the samples were then extracted with petroleum ether by stirring, settling and decanting 3 times. The samples were then air dried and transferred into a dried 100mL conical flask, 200cm<sup>3</sup> of 0.127M sulphuric acid solution was added at room temperature to the samples. The first 40cm<sup>3</sup> of the acid was used to disperse the sample. This was heated gently to boiling point and boiled for 30 minutes. The contents were filtered to remove insoluble materials, which was then washed with distilled water, then with 1% HCl, next with twice ethanol and finally with diethyl ether. Finally, the oven-dried residue was ignited in a furnace at 550°C. The fibre contents were measured by the weight left after ignition and were expressed in term of the weight of the sample before ignition.

**v. Fat content determination**

The lipid content was determined by extracting the fat from 10g of the samples using petroleum ether in a Soxhlet apparatus. The weight of the lipid obtained after evaporating off the petroleum ether from the extract gave the weight of the crude fat in the sample.

**vi. Carbohydrate content determination**

The carbohydrate content of the samples was determined as the difference obtained after subtracting the values of protein, lipid, ash and fibre from the total dry matter (AOAC, 2009).

**3. RESULTS**

Table 1. The highest inhibitory effect of aqueous extract of *Datura stramonium* was recorded on *A. niger*, *A. fumigatus* and *A. tubingensis* which had the highest zone of inhibition of 14.00, 12.00 and 12.00% respectively at (1.0g/mL) and was also active at all concentrations.

**Table 1: Antifungal activities of *Datura stramonium* aqueous extract on fruit rot fungal pathogens**

| Isolates                        | Diameter of zones of inhibition (mm) |        |        |       |        |
|---------------------------------|--------------------------------------|--------|--------|-------|--------|
|                                 | Concentrations (g/mL)                |        |        |       |        |
|                                 | 1.0                                  | 0.8    | 0.6    | 0.4   | 0.2    |
| <i>Aspergillus flavus</i>       | 0.00d                                | 0.00d  | 0.00c  | 0.00c | 0.00c  |
| <i>Rhizopus sp</i>              | 12.00b                               | 10.00b | 8.00ab | 8.00a | 7.00ab |
| <i>Trichoderma harzianum</i>    | 10.00c                               | 10.00b | 8.00ab | 7.00b | 0.00c  |
| <i>Colletotrichum fruticola</i> | 0.00d                                | 0.00d  | 0.00c  | 0.00c | 0.00c  |
| <i>Aspergillus niger</i>        | 14.00a                               | 12.00a | 10.00a | 9.00a | 0.00c  |
| <i>Aspergillus fumigatus</i>    | 12.00b                               | 10.00b | 10.00a | 8.00a | 8.00a  |
| <i>Aspergillus tubingensis</i>  | 12.00b                               | 8.00c  | 0.00c  | 0.00c | 0.00c  |
| <i>Aspergillus nidulans</i>     | 10.00c                               | 0.00d  | 7.00b  | 0.00c | 0.00c  |

Values are mean ± standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher's protected LSD at  $p \leq 0.05$ ). Key: Control Nil

Table 2. The highest inhibitory effect of aqueous extract of *C. inophyllum* was recorded on *A. fumigatus* and *A. nidulans* which had the highest zone of inhibition of (15.00%) at (1.0g/ml) and was also active at all concentrations.

**Table 1: Antifungal activities of *Calophyllum inophyllum* aqueous extract on fruit rot fungal pathogens.**

| Isolates                        | Diameter of zones of inhibition (mm) |         |        |        |       |
|---------------------------------|--------------------------------------|---------|--------|--------|-------|
|                                 | Concentrations (g/mL)                |         |        |        |       |
|                                 | 1.0                                  | 0.8     | 0.6    | 0.4    | 0.2   |
| <i>Aspergillus flavus</i>       | 12.20bc                              | 10.00cd | 9.50a  | 0.00c  | 0.00c |
| <i>Rhizopus sp</i>              | 14.70a                               | 11.20bc | 9.50a  | 0.00c  | 0.00c |
| <i>Trichoderma harzianum</i>    | 12.10bc                              | 10.70c  | 8.50b  | 8.00ab | 0.00c |
| <i>Colletotrichum fruticola</i> | 13.20b                               | 12.00ab | 7.00c  | 0.00c  | 0.00c |
| <i>Aspergillus niger</i>        | 14.00ab                              | 11.60b  | 10.80a | 0.00c  | 0.00c |
| <i>Aspergillus fumigatus</i>    | 15.00a                               | 13.10a  | 10.70a | 8.90a  | 7.20b |
| <i>Aspergillus tubingensis</i>  | 12.00                                | 11.40b  | 8.00b  | 0.00c  | 0.00c |
| <i>Aspergillus nidulans</i>     | 15.00a                               | 13.00a  | 10.80a | 9.10a  | 8.00a |

Values are mean ± standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher's protected LSD at  $p \leq 0.05$ ). Key: Control Nil

Table 3 shows that aqueous extract of *Z. officinale* at 1.0g/mL was found to have more inhibitory effect on the rot fungi than 0.8g/mL, 0.6g/mL, 0.4g/ml, and 0.2g/mL of the same aqueous extract of *Z. officinale*, this indicates that higher concentration of extract promotes higher inhibition of fungal growth.

The highest and lowest inhibitory effect of 1.0g/mL aqueous extract of *Z. officinale* was recorded against *A. nidulans* (26.20%) and *C. fruticola* (22.00%) respectively.

**Table 3: Antifungal activities of ginger rhizome (*Zingiber officinale*) aqueous extract on fruit rot fungal pathogens**

| Isolates                        | Diameter of zones of inhibition (mm) |        |         |        |         |
|---------------------------------|--------------------------------------|--------|---------|--------|---------|
|                                 | Concentrations (g/mL)                |        |         |        |         |
|                                 | 1.0                                  | 0.8    | 0.6     | 0.4    | 0.2     |
| <i>Aspergillus flavus</i>       | 22.40c                               | 20.20d | 18.00d  | 16.20c | 14.00b  |
| <i>Rhizopus sp</i>              | 24.50bc                              | 22.20b | 21.00b  | 11.00f | 10.20e  |
| <i>Trichoderma harzianum</i>    | 22.20c                               | 21.50c | 20.20c  | 15.00d | 11.00d  |
| <i>Colletotrichum fruticola</i> | 22.00cd                              | 21.00c | 20.00c  | 15.00d | 12.00cd |
| <i>Aspergillus niger</i>        | 26.00a                               | 23.10a | 22.00a  | 20.10a | 19.20a  |
| <i>Aspergillus fumigatus</i>    | 22.10c                               | 22.00b | 21.00b  | 12.00e | 10.90de |
| <i>Aspergillus tubingensis</i>  | 25.10b                               | 21.20c | 20.00c  | 18.00b | 14.00b  |
| <i>Aspergillus nidulans</i>     | 26.20a                               | 22.40b | 21.10ab | 20.00a | 11.00d  |

Values are mean ± standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher's protected LSD at  $p \leq 0.05$ ). Key: Control Nil

Table 4 shows highest inhibitory effect of aqueous extract of *H. suaveolens* was recorded on *T. harzianum* and *A. fumigatus* at (18.00%) in (1g/ml) concentration it was also observed that the plant extracts was active in most of the concentrations.

**Table 4: Antifungal activities of *Hyptis suaveolens* aqueous extract on fruit rot fungal pathogens**

| Isolates                        | Diameter of zones of inhibition (mm) |        |         |         |        |
|---------------------------------|--------------------------------------|--------|---------|---------|--------|
|                                 | Concentrations (g/mL)                |        |         |         |        |
|                                 | 1.0                                  | 0.8    | 0.6     | 0.4     | 0.2    |
| <i>Aspergillus flavus</i>       | 15.00c                               | 12.00d | 12.00d  | 10.00b  | 10.00b |
| <i>Rhizopus sp</i>              | 12.00d                               | 10.00f | 8.00f   | 0.00d   | 0.00d  |
| <i>Trichoderma harzianum</i>    | 18.00a                               | 15.70b | 14.20bc | 13.00ab | 11.20a |
| <i>Colletotrichum fruticola</i> | 17.00b                               | 12.00d | 10.50e  | 9.50c   | 7.00c  |
| <i>Aspergillus niger</i>        | 10.00e                               | 10.00f | 0.00f   | 0.00d   | 0.00d  |
| <i>Aspergillus fumigatus</i>    | 18.00a                               | 17.00a | 17.40a  | 13.80a  | 10.00b |
| <i>Aspergillus tubingensis</i>  | 12.00d                               | 11.70e | 10.00e  | 0.00d   | 0.00d  |
| <i>Aspergillus nidulans</i>     | 14.50c                               | 13.00c | 11.00de | 10.00b  | 0.00d  |

Values are mean ± standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher's protected LSD at  $\leq 0.05$ ). Key: Control Nil

Table 5 shows *Aspergillus flavus* (18.00%) and *Aspergillus niger* (10.00%) had the highest and lowest zone of inhibition with concentration aqueous extract of *K. grandifoliola* (1.0g/mL) respectively. It was also observed that different concentrations of the plant extract were active and inhibitive against the rot pathogens of pineapple. Higher concentration of extract promotes higher inhibition of fungal growth.

**Table 5: Antifungal activities of *Khaya grandifoliola* aqueous extract on fruit rot fungal pathogens**

| Pathogens                       | Diameter of zones of inhibition (mm) |         |        |        |        |
|---------------------------------|--------------------------------------|---------|--------|--------|--------|
|                                 | Concentrations (g/mL)                |         |        |        |        |
|                                 | 1.0                                  | 0.8     | 0.6    | 0.4    | 0.2    |
| <i>Aspergillus flavus</i>       | 18.00a                               | 12.00c  | 10.00b | 8.00c  | 8.00a  |
| <i>Rhizopus sp</i>              | 13.00d                               | 10.00d  | 0.00d  | 0.00d  | 0.00b  |
| <i>Trichoderma harzianum</i>    | 15.00c                               | 14.00ab | 13.00a | 11.20a | 8.00a  |
| <i>Colletotrichum fruticola</i> | 12.00d                               | 10.00d  | 9.00bc | 7.00bc | 0.00ba |
| <i>Aspergillus niger</i>        | 10.00e                               | 0.00e   | 0.00d  | 0.00d  | 0.0b   |
| <i>Aspergillus fumigatus</i>    | 17.00b                               | 15.10a  | 13.00a | 10.00b | 8.00a  |
| <i>Aspergillus tubingensis</i>  | 12.00d                               | 10.00d  | 8.00c  | 0.00d  | 0.00b  |
| <i>Aspergillus nidulans</i>     | 15.00c                               | 13.00bc | 10.00b | 0.00d  | 0.00b  |

Values are mean ± standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher’s protected LSD at ≤ 0.05). Key: Control Nil

Highest inhibitory effect of aqueous extract of *D. stramonium* at 1.0g/ml was recorded on *A. tubingensis* (24.00%) while *Colletotrichum fruticola* (10.00) was most resistant to the same concentration of *D. stramonium* at 1.0g/ml. *Trichoderma harzianum* and *Colletotrichum fruticola* were most resistant to low concentration of *D. stramonium*.

Table 6 revealed the proximate analysis of the rotten pineapple, the nutritional composition of infected pineapple by individual isolated causal organism, the physiological effect of *Aspergillus flavus*, *Rhizopus sp*, *Trichoderma harzianum*, *Colletotrichum fruticola*, *Aspergillus niger*, *Aspergillus fumigatus*, *Aspergillus tubingensis* and *Aspergillus nidulans* on the pineapple fruit was observed as they were all observed to cause reduction in the ash content, fat content, crude fiber, crude protein and carbohydrate content of the fruit while they were observed to cause the increase in the level of moisture content and dry matter content of the pineapple fruit. The changes caused by the organisms make the fruit unsafe for consumption as reported by Amusa *et al.* (2003).

**Table 6: Result of Proximate composition of pineapple fruit infected with fungal rot isolate**

| Parameter (%)          | Sample codes |         |        |         |         |         |         |        |         |
|------------------------|--------------|---------|--------|---------|---------|---------|---------|--------|---------|
|                        | Control      | AF      | RS     | TH      | CF      | AN      | AFM     | AT     | AND     |
| Moisture content       | 85.25a       | 88.17a  | 86.08a | 86.86a  | 87.59a  | 86.60a  | 87.90a  | 90.10a | 89.85a  |
| Dry matter ash content | 15.10a       | 15.90b  | 15.12a | 16.06a  | 16.01a  | 15.75ab | 16.05a  | 16.10a | 15.99a  |
| Crude fibre            | 6.20a        | 5.90a   | 5.86a  | 5.26ab  | 5.72ab  | 5.91a   | 5.87a   | 5.05ab | 5.50ab  |
| Fat content            | 42.02a       | 42.79ab | 42.50b | 43.32a  | 43.03a  | 43.90a  | 42.74ab | 43.62a | 42.98ab |
| Crude protein          | 3.35a        | 2.60a   | 2.21b  | 2.21b   | 2.87a   | 2.30ab  | 2.56ab  | 2.57ab | 2.90a   |
| Carbohydrate           | 5.04a        | 4.81a   | 4.67a  | 4.60a   | 3.08b   | 4.61ab  | 4.19ab  | 4.88a  | 4.97a   |
|                        | 83.03a       | 80.07ab | 76.67b | 79.76ab | 78.98ab | 78.45ab | 79.91ab | 80.03a | 78.97ab |

Values are mean  $\pm$  standard error of the mean for bioassay conducted. Means followed by the same letter(s) are not significantly different (multivariate analysis, Fisher's protected LSD at  $\leq 0.05$ ).

Key: Healthy pineapple as control

AF = *Aspergillus flavus*

RS = *Rhizopus sp*

TH = *Trichoderma harzianum*

CF = *Colletotrichum fruticola*

AN = *Aspergillus niger*

AFM = *Aspergillus fumigatus*

AT = *Aspergillus tubingensis*

AND = *Aspergillus nidulans*

#### 4. DISCUSSION AND CONCLUSION

The physiological and morphological characteristics of the fungal isolates were also shown in Table 1. The isolated fungi in this study agreed with fungal species associated with post-harvest rot of pineapple fruits in Maiduguri, Nigeria as reported by Akinmusire (2011). In this study, bioactivity of the test plant could be ascribed to fungitoxic compounds present. This report is in consonance with the earlier report of several researchers that used different fungal organisms and plants as bioagents (Ranasinghe *et al.*, 2003, Amadioha, 2002, Obagwu and Korsten, 2003, Sanzani *et al.*, 2009 and Amare, 2002). Extract of *D. stramonium*, *Z. officinale*, *K. grandifoliola*, *H. suaveolens* and *C. inophyllum* in (0.2, 0.4, 0.6, 0.8, and 1.0g) were found to exhibit inhibitory effects on most of the fungi at varying concentrations with the exception of *Z. officinale*, which was found to have inhibitory effect on all the fungi isolated at all tested concentrations.

The growth inhibition zone against *Escherichia coli* and *Staphylococcus aureus* has been reported (Sreenivasa *et al.*, 2012). Fresh ginger juice showed inhibitory action against *Aspergillus niger*, *Saccharomyces cerevisiae*, *Mycoderma sp.* and *Lactobacillus acidophilus*. The benzene extract of *Zingiber officinale* rhizome showed highest antibacterial activity against drug resistant *P. aeruginosa* isolated from wound and pus samples. Ansari *et al.*, (2016) reported prophylactic and therapeutic activities rhizome of ginger. Jainu *et al.*, (2016) reported antimicrobial activity of ginger against *Escherichia coli*, *Proteus vulgaris*, *Staphylococcus aureus*, *Streptococcus pyrogenes* and *Salmonella typhi*.

Anti-dermatological effects of *H. suaveolens* volatile oil against some fungal and bacterial diseases have been reported (Okonogi *et al.*, 2005).

The plant extracts used in the research exhibited preservative potential on pineapple fruit from fungal rot fungi. Moreover; it has revealed the potential of aqueous and ethanolic extracts of *D. stramonium*, *Z. officinale*, *K. grandifoliola*, *H. suaveolens* and *C. inophyllum* as alternative or complementary to chemicals fungicides in controlling pineapple rot because of their fungitoxic activities on the test organisms. The utilization of nutrients by fungi for growth caused nutrient deficiency of the infected pineapple fruit, if consumed; it can lead to health problem such as inability to decrease severity of respiratory tract infection. Peri-follicular hemorrhage which can lead to scurvy, retarded growth, muscle wasting, low metabolic activities and reduction in energy (Diribe and Elom, 2002). Post-harvest pineapple fruit rot is a threat to food security and economic stability of various key players in agribusiness. Aqueous extracts of the test plants were observed to exhibit inhibitory effects on the isolated microorganisms. More also, aqueous

extracts of *Zingiber officinale*, *Khaya grandifoliola*, *Hyptis suaveolens*, *Calophyllum inophyllum* could be applied on pineapple fruits to extend their shelf lives.

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