

1.3 Pathogenicity test

Spore suspension of *Lasiodiplodia* was used to inoculate fresh fruits 1mm deep at the equator and incubated at $28\pm 2^{\circ}\text{C}$ and 75% relative humidity inside sterilized desiccators. The disease symptoms were noted and re-isolation from infected fruit tissue was performed on fresh sterile MEA plate and its cultural characteristics were compared with the original isolate.

1.4 Preparation of extract of *Tridax procumbens*

Detached *Tridax procumbens* leaves were air dried for 14 days. The dried leaves were pulverized with a blender to a smooth powdery form. About 28 g of the pulverized leaves were dissolved in 1 L of water, thoroughly shaken together and later filtered with a muslin cloth to obtain the crude /stock solution. Varying concentrations (5, 10, 15 and 100%) of the aqueous extract were then prepared from the stock / crude extract with appropriate volumes of water.

1.5 Inoculation and Treatment of orange fruits with *Tridax procumbens* extract

The orange fruits were artificially infected with spores (6.50×10^4 cfu/ml) of *Lasiodiplodia* and later dipped separately into each concentration (5%, 10%, 15% and 100%) of the prepared extract for 5 min while benlated (fungicide) and untreated orange fruits served as positive and negative control respectively. Each set up consisted of 3 fruits. After treatment, the fruits were then placed inside a sterilized Petri dish and transferred into desiccators, and stored at $28\pm 2^{\circ}\text{C}$ and 75% relative humidity and assessed daily for disease severity

1.6 Assessment of rot severity

Assessment of disease severity was done using the scale of (Arekemase and Oyeyiola, 2007) but with slight modification where 1=disease free, 2=slight rot /decay up to 10% of the fruit, 3=moderate rot /decay up to 25% of the fruit, and 4=severe rots/decay $\geq 35\%$ of the fruit surface. Rot/decay was recognized by light brown discolouration on the fruit or by appearance of mycelium on the fruit surface.

1.7 Phytochemical screening of the aqueous extract of *Tridax procumbens*

The phytochemical screening of the aqueous extract was done according to the method described by (Trease and Evans, 2004). The phytochemicals screened for were tannin, saponin, phlobatannin, flavonoid, alkaloid and cardiac glycosides.

1.8 Gas chromatography-mass spectrophotometry (GC-MS) of *Tridax procumbens* extract

The *Tridax* extract was subjected to chromatographic analysis using a Varian 3800/4000 gas chromatograph mass spectrometer equipped with an Agilent Technologies and a BP5 (30 m $\times$ 0.25 mm $\times$ 0.25 microns) capillary column. Nitrogen was used as a gas carrier. 1.0 μL volumes of the extract were injected using a splitless mode at an injector temperature of 270°C . The oven temperature was ramped from 80 to 200°C (1 min hold) at a rate of $5^{\circ}\text{C}/\text{min}$. The oven temperature was held at 280°C for 6 min following each analysis. The total run time for each sample was approximately 45 min. The GC-MS interface temperature was set to 280°C . The peaks of the organic compounds in the samples were identified in Wiley's NIST 08 Mass Spectral Library and expressed in terms of its balance and retention time.

1.9 Statistical analysis

The data obtained for the disease severity were subjected to analysis of variance and where significant, the means were compared at 5% level of probability using Tukey's Test (SPSS version 20).

2 Results

2.1 Effects of different concentrations of *Tridax procumbens* extracts on disease severity in orange fruits pre- inoculated with *Lasiodiplodia* sp and stored at $28\pm 2^{\circ}\text{C}$ and 75 % relative humidity

All treated and control fruits recorded mean disease severity values of 1.00 ± 0.00 by day 5 of storage (Figure 1), indicating that they were all disease free. As storage duration progressed till day 10, except the control fruits that had shown slight rottenness as evident by their mean disease severity values of 2.00 ± 0.52 , all treated fruits were still disease free (healthy), though with mean disease severity values that were not significantly different ($p\geq 0.05$)