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Isolation and Identification of Fungi Associated with Rot of Cucumber (*Cucumis sativus* L.) in Jimeta, Yola North Local Government Area, Adamawa State

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ABSTRACT

Cucumber fruit (*Cucumis sativus* L.) is believed to have originated from Southern Asia and fourth most important vegetable after tomato, cabbage and onion. The objective is to isolate and identify fungi responsible for rot of cucumber (*Cucumis sativus* L.) in the study area. Potato dextrose agar was used as growth medium. Three fungal pathogens (*Aspergillus flavus, Rhizopus stolonifer* and *Aspergillus brasiliensis*) were identified through pathogenicity test to be causing rot of Cucumber fruit in Jimeta markets (Gwari Market, Jimeta Modern Market and Jimeta Shopping Complex Market), Yola North of Adamawa state. Isolation and identification of pathogens responsible for cucumber fruit rot in Jimeta Markets were carried out, where *Rhizopus stolonifera* had the highest (25-48%) frequency of occurrence while *Aspergillus flavus* was the least (22-38%). From the study, it is concluded that *Aspergillus brasiliensis*, *Rhizopus stolonifer* and *Aspergillus flavus* were found out to be the fungal pathogen that caused rot of Cucumber (*Cucumis sativus* L.) fruit in the study area. The vulnerability of Cucumber fruit to fungal rot is not far from the poor way and manner at which the fruits are conveyed, stored and traded in the study area. It is recommended to farmers and fruit vendors to be conveying perishable fruits in soft bags and stored in cool dry places.

INTRODUCTION

Cucumber (*Cucumis sativus* L.) is believed to have originated in Southern Asia [9]. Cucumber fruit is a warm season annual plant cultivated for its fleshly fruit, eaten as a salad vegetable or used for pickling [10]. It is popularly used for skin treatments and for natural beautification. Cucumber has various benefits associated with it - right from skin to body and overall health [16]. It is a nutritious and a naturally cool fruit [17]. It is an important vegetable and one of the most popular members of the Cucurbitaceae family [16], it is a creeping vine that bears fruits that is cylindrical in shape. Squash, Melon and pumpkins are other vegetables that belong to the Cucurbitaceae family [5]. Cucumbers have cooling properties and are extremely good for bringing relief to the eyes in summers [10]. The fruit has high water content, about 96%water [16]. Other than its use as a salad vegetable, cucumber fruit extracts are often incorporated as a primary ingredient in many topical skin preparations [10]. Its deep cleansing action stems from its natural chemical constituent of glycolic, lactic and salicylic acids [11]. These acids are known as organic or fruit acids because they contain one or more carboxyl radicals (COOH) in their structure [11]. Cucumber seeds possess similar properties to those of the allied pumpkin (*Cucurbita pepo*) which are distinctly diuretic, but mainly employed as a very efficient taeniacide and can be used as anemetic substance [10]. Fruits and vegetables including cucumber are highly perishable products; the quality is affected by post-harvest handling, transportation, storage and marketing [2]. [12] were able to isolate and identify post-harvest fungi that were responsible for rot in cucumber as *Aspergillus niger, Cladosporium cladosporioides, Rhizopus stolonifer, Fusarium solani, Aspergillus flavus* and *Geotrichum candidum*; the fruits turn white and the interior of the fruits are heavily colonized by white mycelium of fungi, infected fruits initially show water-soaked lesions and eventually shrivel and rot.

Cucumber fruits are seriously affected by post-harvest diseases which can cause losses of up to 30% of the total yield of crops [2]. The need to determine and ascertain the fungal organisms responsible for fruit rot especially cucumber in the study area cannot be overemphasized so that fungi responsible for rot can be ascertain and documented.

The aim of the study is to isolate and identify fungi associated with post-harvest cucumber fruit rots in the study area.

MATERIALS AND METHODS

Description of Study Area

Determination of incidence and severity of cucumber postharvest rots were carried out within the months of July-September, 2021 in selected markets in Yola North local government area of Adamawa State, according to [1]. Yola North lies between latitude 9° 11' to 9° 19' N and longitude 12° 20' to 12° 30'E, covering a total area of 1,213.30 km^2 (**Fig. 1**). The area has a tropical climate marked by dry and rainy seasons. The rainy season commences around May and ends in October. The rainfall is characterized by a mean total of 1,113.3mm, August and September being the wettest months with about 25% of the total annual rainfall. The dry season starts in late October and ends in late April.

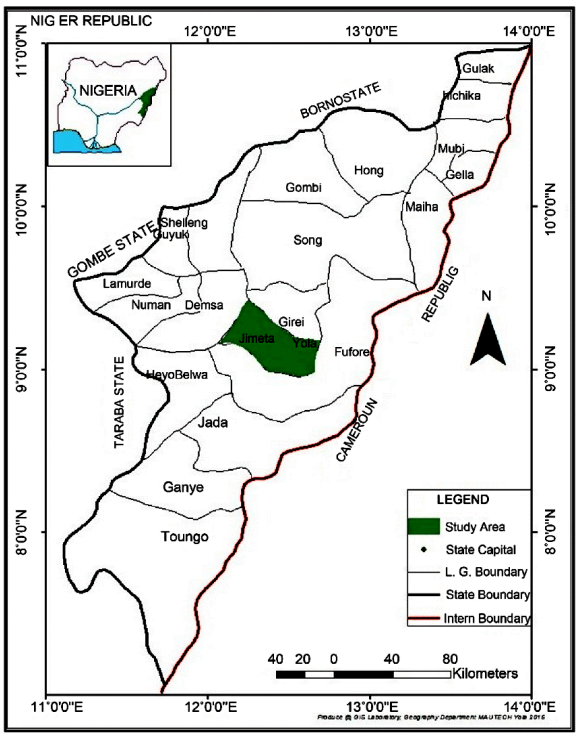

Fig. 1. Map of Adamawa state showing the study area.

Collection of Sample

Fifty rotted and healthy cucumber (*Cucumis sativus* L.) fruits were randomly examined from each market namely; Jimeta Modern Market, Jimeta bye-pass market (Gwari market) and Jimeta shopping complex market, which resulted to a total of one hundred and fifty fruits examined and the rotted samples were collected in a clean separate polythene bags and were brought to the Department of Plant Science Laboratory of Modibbo Adama University, Yola for further studies.

Sterilization of Glass wares

The Petri-dishes used for solid media preparation were sterilized in an oven at 160 °C for 6 hours in a sterilizing can. Needle and Cork borer used for inoculation were sterilized by flaming and cooled by dipping into methylated spirit according to [18].

Preparation of Potato Dextrose Agar (PDA)

Thirty-nine (39 g) of potato dextrose agar (PDA) was dissolved in 1 litre of distilled water. The PDA was poured into conical flask, then covered with cotton and wrapped with aluminum foil and autoclaved at 121°C for 15 minutes at 10 lbs pressure. Six (6) millilitres (0.1%) of streptomycin was added to the sterilized media, just before pouring into the Petri dishes to prevent bacterial growth and allowed to cool and solidify [18].

Isolation and Identification of Fungal Pathogens

With the help of sterilized knife, diseased cucumber fruits were peeled and thereafter cut into small pieces of about 5 mm and sterilized in 0.01% mercury chloride for 30 seconds, rinsed in five (5) different changes of distilled water, and dried on a sterilized filter paper before plating at the center of 9cm diameter Petri dish containing 20 mL solidified medium. Cultures were incubated at room temperature ranging from 27-32 °C for three (3) days when new colonies immerged (**Fig. 2**). These were sub-cultured promptly on fresh sterilized medium using [18] method. With the aid of a sterile needle, small circular portion (2 mm) of the pure culture were taken to a sterilized slide; this was stained with lacto-phenol cotton blue and examined under x 10 magnifications of the microscope [8]. The morphological and cultural characteristics observed were compared with structures in organisms' identification guide of [8]

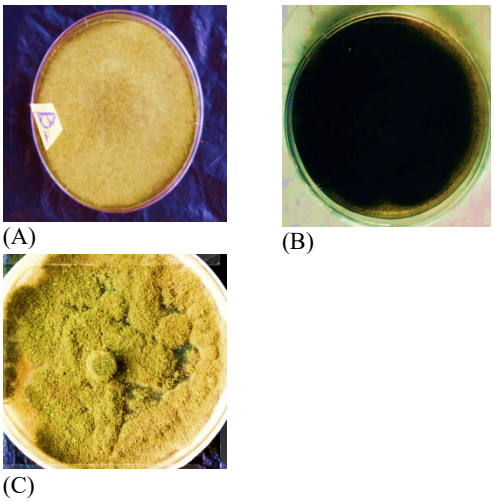

Fig. 2. Pure culture of (a) *Rhizopus stolonifer* (X 10) (b) *Aspergillus brasiliensis* (X 10) and (c) *Aspergillus flavus* (X 10).

Determination of Fungal Rot of Cucumber

Determination of fungal rot of cucumber (**Fig. 3**) was determined by sampling out the number of cucumber fruits having rot out of the total number of healthy ones in each of the market. These were expressed in percentage using the formula:

$$\frac{\text{Number of rotted cucumber fruits}}{\text{Total number of cucumber fruits}} \text{ x } 100\ \%$$

Pathogenicity Test

Pathogenicity test was carried out according to the method of [13]. Healthy cucumber fruits of similar size and weight were obtained, and surface sterilized by dipping in 0.01% mercury chloride for 60 seconds and rinsed in three (3) different changes of distilled water. One (1 cm) holes were made in each fruit aided with a sterilized cork borer, then 10 mm disc portion of the pure-culture of organisms were inserted and sealed with vaseline to avoid entry of other organisms and labeled using masking tape.

Fruits were stored in polythene bags and were incubated at room temperature for one (1) week to observe whether the isolates are the original cause of rot to the cucumber fruit. Similar set up was made as control where distilled water was used instead of the fungal inocula.

(a)

(b)

Fig. 3. Healthy (a) and diseased (b) cucumber.

RESULTS AND DISCUSSION

Rhizopus stolonifer, Aspergillus brasiliensis and *Aspergillus flavus* were the three different fungi found associated with postharvest deterioration of Cucumber (*Cucumis sativus* L.) in Jimeta Markets of Yola North Local Government Area of Adamawa State (**Table 1**), which is in agreement with the work of [12], who revealed nine (9) different species of fungi in vegetables and fruits among which *Aspergillus niger, Rhizopus stolonifer* and *Aspergillus flavus* were inclusive. It is also in consonance with the work of [7], who reported *Aspergillus niger, Rhizopus stolonifer* and *Fusarium oxysporum.* Similarly, [3] reported seven (7) fungi responsible for Guava fruit spoilage namely *Pseudollescheria boydii, Fusarium oxysporum, Rhizopus stolonifer, Aspergillus parasiticus, Neosartorya pseudofischeri, Aspergillus niger,* and *Aspergillus fumigatus.* [19] also reported *Rhizopus stolonifera* among the phytopathogenic fungi associated with Cucumber (*Cucumis sativus* L.) revealed in his research. In the same manner [4] found eight different fungi responsible for rots of fruits, these include *Aspergillus flavus, Aspergillus candidus, Aspergillus glaucus, Candida tropicalis, Aspergillus niger, Aspergillus tereus, Pseudallescheria boydii* and *Aspergillus carbonarius.*

Table 1. Frequency of occurrence of fungi isolated from cucumber fruits in Jimeta markets.

Pathogen	Frequency of Pathogen (%)		
	GWM	JMM	JSC
Rhizopus stolonifera	48.00	40.00	25.00
Aspergillus brasiliensis	43.00	32.00	22.00
Aspergillus flavus	38.00	30.00	22.00
LSD (0.05)	9.88	0.37	1.31

Key:
GWM = Gwari Market
JMM = Jimeta Modern Market
JSC = Jimeta Shopping Complex
LSD: Least Significant Difference

The result is contrary to the work of [6] which revealed *Fusarium oxysporum, Geotrichum candidum* and *Mucor species* as the fungi found which may be due to the differences in climatic factors of the study areas. Incidence of rot observed in the study showed Gwari Market had highest fungal disease prevalence followed by Jimeta modern market and Jimeta Shopping complex respectively (**Table 2**). This is attributed to poor hygiene condition of the markets. This is in agreement with the work of [14] in Pakistan who reported the incidence of *Rhizopus specie* and *Aspergillus niger* on mango fruit rot. [15] reported *Rhizopus specie, Aspergillus specie, Saccharomyces species, Penicillium species* and *Fusarium species* to be responsible for spoilage of Cucumber, Mango and Pineapple fruits. Future research is needed to inhibit fruit and vegetable spoilage due to pathogens, perhaps using plants and herbal bioactive compounds themselves that had been demonstrated in several instances [20-22].

Table 2. Virulence of fungi isolated from cucumber fruit in Jimeta markets.

Visual scale	*R. stolonifera*	*brasiliensis*	*A. flavus*
Low virulent group	-	-	-
Moderately virulent group	-	-	-
High virulent group	-	-	+++
Very high virulent group	-	-	-
Totally virulent group	+++++	+++++	-

Key:
- : No virulence
+ : 1-20% = low virulent group
++ :21-40% = moderately virulent group
+++ : 41-60% = high virulent group
++++ : 61-80% = very high virulent group
+++++ : More than 80% = totally virulent group

CONCLUSION

The results from this study showed that several microscopic pathogens are associated with cucumber fruit rot in the study area which is evident as three pathogens of two genera are associated with rot of Cucumber (*Cucumis sativus* L.) fruits in Jimeta markets, Yola North of Adamawa State of Nigeria. These are *Aspergillus brasiliensis, Rhizopus stolonifer* and *Aspergillus flavus.* The frequency of occurrence showed *Rhizopus stolonifer* to be 48.00 % followed by *Aspergillus brasiliensis* 43.00 % while 30.00 % was the least in place of *Aspergillus flavus*. Virulence of pathogens isolated from Cucumber fruit showed that *Aspergillus brasiliensis* and *Rhizopus stolonifer* to be 'totally virulent' while *Aspergillus flavus* was the least with 'high virulent group'. The research further showed that cucumber fruits must be properly sanitized before consumption.

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REFERENCES

1. Adebayo, A.A and Tukur, A.L. Adamawa State in Maps, Paraclette Publishers (A Division of Paraclette and Sons) Paraclette Publishing House, P.O. Box 5448, Yola Nigeria. 1999; Pp 32-33.
2. Agrios, G.N. Plant Pathology 5th Edition Academic Press, New York; 2005
3. Amadi, J.E., Nwaokike, P., Olahan, G. S. and Garuba. T. Isolation and Identification of Fungi involved in the Postharvest Spoilage of Guava (Psidium guajava) in Awka Metropolis. J Eng Appl Sci. 2014;4(10): 21-43.
4. Anjorin, S.T., Adebayo, S.E. and Hussaini, A. M. Control of Toxigenic Fungi and Mycotoxins with Phytochemicals: Potentials and Challenges. J Food Saf Develop Ctries. 2013; 2 (3): 45-67.
5. Bello A.O., Oladipo O.T. and S. A. Saheed. Numerical Taxonomic Study of some Solanum L. Species (Solanaceae) Using Vegetative and Floral Morphological Characters. Ife J Sci. 2013; Vol. 15, no. 3 524
6. Chiejina, N.V. Mycoflora of some salad vegetables, Bio-Research. 2008; 6(2): 392-395.
7. Chiejina, N.V., and Onaebi, C.N. In-vitro Fungicidal Activity of Two Plant Extracts against five Phytopathogenic Fungi of Cucumber (Cucumis sativus L.) fruit. International J Appl Nat Sci. 2013; 2(3): 61-68.
8. Fawole, M.O. and Oso, B.A. Laboratory manual of Microbiology. 5th Edition, Spectrum Books Limited, Ibadan, 2007; 22-23
9. Lerner, B.R. and Dana, M.N. Growing Cucumbers, Melons, Squash, pumpkins and Gourds; 2001 Purdue University Cooperative Extension Services.
10. Mateljan, G. The World's healthiest foods-essential guide for the healthiest way of eating 1st Edition. World's Healthiest Foods, 2006; 880 p
11. McGraw. Hill Dictionary of Chemistry: 3rd Edition. McGraw Hill Book Co. New York, 223 pp. 1984
12. Naureen, F., Humaira, B., Viqar, S., Jehan, A. and Syed, E. Prevalence of Postharvest rot of vegetables and fruits in Karachi, Pakistan. Pak J Bot. 2009; 41 (6): 3185 – 3190.
13. Okigbo, R.N., Agbata, C.A., Echezona, C. E. Effects of leaf extracts of Azadirachta indica and Chromolaena odorata on postharvest spoilage of fungi rot of yams in storage. Curr Res J Biol Sci. 2010; 2 (1): 9 – 12.
14. Pernezny, K. and Simone, G.W. Proposed lists of names for diseases of Mango (Mangifera indica L.). Phytopathol News. 2000;34(2): 25-26.
15. Sani, M.A, Usman N., Kabir F., Kutama A.S. The Effect of Three Natural Preservatives on the Growth of Some Predominant Fungi Associated with the Spoilage of Fruits (Mango, Pineapple and Cucumber). Glob Adv Res J Agri Sci. 2015; 4(12):923-928.
16. Simons, A. J. The New Vegetable Growers Handbook. Pergium Books Ltd, Middlesex, pp 166-7; 2003.
17. Valenzuela, H., Hamasaki, R.T. and Fukuda, S. (2005). Field cucumber production guidelines for Hawaii. Httpwww.Extentohawii.edu/kbase/reportscucumber production.html. Accessed 22nd September, 2011.
18. Zakari, B. G, Zakari S. S and Talba, Usman. Potential of aqueous extract of Jatropha curcas on the control of post-harvest of sweet orange (Citrus sinensis L.) fruits in Wukari. Int J Agri Biol Sci. 2019;3(3):92-99.
19. Zandstra, B. Weed Control Guide for Vegetable Crops. East Lansing, Michigan, Michigan State University; 1999 E-433
20. Uba G, Yakubu A, Baba AM. The effect of the plant *Adiantum philippense* extracts on biofilms formation and adhesion to *Shigella flexneri*: A Predictive Modelling Approach. J Biochem Microbiol Biotechnol. 2021;9(2):34-9. Available from:
21. Ummu Rabiâu R, Saâidu H, Labaran HS, Olanrewaju SA, Muntari M, Ibrahim SA. Phytochemical Screening and antibacterial activity of stem bark of Baobab tree (*Adonsonia digitata*) on some clinical isolate. J Biochem Microbiol Biotechnol. 2020;8(2):21-4.
22. Uba G, Ginsau MA, Aujara KM. Biofilm Formation, Adhesion with *Staphylococcus aureus* against food borne pathogen: A mathematical modeling on the effects of *Adiantum phillippense*. J Biochem Microbiol Biotechnol. 2020;8(2):25-9.