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Assessment of antibacterial capacity of *Vernonia amygdalina* against Post harvest fruit rot organisms of okra (*Abelmoshus esculentus* L moench)

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ABSTRACT: *Okra is an economically important vegetable crop cultivated in the tropical and subtropical parts of the world. Advocacy for consumption of local vegetables like okra could help to enhance food security and combat malnutrition in developing countries. Okra is a multipurpose crop due to the various uses of its fresh leaves, buds, flowers, capsules, seeds and seeds. The focus of the study is to evaluate the antimicrobial effects of aqueous leaf extracts of V. amygdalina against post harvest bacterial associated with okra. Fresh leaves of the test plant were collected, air dried, and pulverized. Hundred grams of pulverized leaves of the test plant was mixed with 200ml of cold water a room temperature and left overnight. This was later filtered and the filtrate served as extract. Five bacterial strains were isolated from the the okra viz: Bacillus subtilis, B. panthotenticus, B. cereus, Psedumonas chlororaphis, Aeromonas hydrophila. Agar well diffusion test method was used to determine the antibacterial capacity of the test plant. 20% aqueous extract of the test plant most inhibited Bacillus subtilis, Pseudomonas chlororaphis, Aeromonas hydrophila, B. cereus and B. panthotenticus by 0.83, 1.65, 1.19, 1.51 and 1.43% respectively. It is shown from the result that higher concentrations of aqueous extract favoured higher inhibition of bacterial growth.*

KEYWORDS: *V. amygdalina*, antibacterial, okra.

INTRODUCTION

Okra **(***Abelmoshus esculentus* L Moench**)** originated from Ethiopia (Getachew, 2001; Simmone *et al*., 2004, Dandema, 2010, Sathish and Eswar, 2013). It was later propagated in North Africa, in the Mediterranean, Arabia and India by the 12th century (Nzikou *et al*., 2006). Okra is one of the most widely known and consumed species of okra of the famiy Malvaceae (Naveed *et al*., 2009). Okra was previously included in the genus hibiscus; it was later designated to *Abelmoshus* (Aladele *et al*., 2008). It is an economically important vegetable crop grown in both tropical and sub tropical parts of the world (Oyelade *et al*., 2003, Andras *et al.*, 2005, Saifullah and Rabanni, 2009). It is mainly grown for its tender green capsule and leaves which are cooked and commonly consumed as boiled vegetable (Ndunguru and Rajabu, 2004, Chattopadhyay *et al*., 2011, Mihretu *et al*., 2014). Okra mucilage is used as plasma replacement or blood volume expander and as well as

binder of cholesterol and bile acid carrying toxins dumped into it by the liver (Madison, 2008 and Maramag, 2013). Okra seeds can be roasted and ground to form a caffeine-free substitute for coffee (Calisir and Yildiz, 2005). It is also a source of confectionery (Adetuyi, *et al*., 2011). With these various importances of okra, little attention has been paid to its improvement programme as it is considered a minor crop (Sanjee *et al*., 2010). The total commercial production of okra worldwide was put at 4.8million tons in which India and Nigeria are predominant producers (Gulsen *et al*., 2007). Various pre and post harvest diseases such as viral, bacterial and fungal have been competing with okra thereby hampering global production of okra. Therefore, the purpose of his study is to evaluate the antimicrobial efficacy of varied concentration of aqueous leaf extracts of *V. amygdalina* against post harvest bacterial associated with okra fruits.

MATERIALS AND METHODS

Preparation and Sterilization of laboratory wares

Glass wares used for this research were washed using detergents and water, these were allowed to air dry before sterilization. The inoculating chamber and other working surfaces were sterilized by swabbing with 70% alcohol. Preparation and Sterilization of media were done according to the manufacturer's indication.

Sample collection and identification

Fresh leaf samples of *V. amygdalina* were collected from a vegetation site in Ado Ekiti. The identity of the collected plant was authenticated by the herbarium unit of Ekiti State University, Ado Ekiti. The leaf samples were air dried at room temperature for two weeks before pulverizing. The powdered samples were kept in a clean air tight container in the laboratory prior to use.

Preparation of media

Twenty eight (28g) of powdered nutrient agar was weighed on Melter weighing balance and dispensed into 100ml beaker. The medium was dissolved by boiling in a water bath in order o homogenize. This was later removed an stand to cool o room temperature before dispensing into sterile MacCartney bottles before it was autoclaved at 121^0C for 15min.

Preparation of plant extracts

Hundred grams (100g) of the powder of test plant was weighed into 200ml of distilled water and this was left overnight at room temperature. This was filtered using Whatman No. 1 filter paper and the filtrate stored in the sterile bottle at 4^0C.

Isolation of bacteria

After extraction, 1ml of the broth of infected okra was taken using syringe and dispensed into 9ml of sterile water. His process was serially diluted and the final diluents were kept in the test tube and corked using cotton wool to avoid contamination.

Phytobacteria Pathogenicity Tests

The pathogenicity test is the main criterion for the identification of bacteria suspected of being the aetiological agents of a plant disease. This involves reproduction of lesions following artificial infection of suitable hosts under greenhouse conditions. Occasionally, pathogenicity tests may be performed under controlled laboratory conditions on okra fruits. The difficulty of choosing susceptible varieties of the host and suitable conditions indicates the necessity to use additional and practical means of identification of pathogenic bacteria. Inoculated okra fruits were placed in individual "moist chambers" consisting of glass tubes lined on, the bottom with moist cotton wool. After which lesions were observed as marks of infection.

Determination of antibacterial activity of the test plant

Pour plate method was used to determine antimicrobial property of the test plant. Molten nutrient agar was dispensed into sterile Petri dishes and this was allowed to cool down to 45^0C and the bacterial inoculum was streaked on the medium. Wells were punched into the agar using 4mm cork borer and the wells were filled with 1ml of the test plant extracts. The plates were incubated at 37^0C for 24hr. The antibacterial activity of the test plant was determined by measuring the diameter of the zone of inhibition using metre rule. Distilled water was taken as control.

RESULTS

Table 1 showed the effects of varied concentrations of aqueous extracts of *V. amygdalina* against post harvest bacteria of okra viz: *A. esculentus* that *B. subtilis*, *A. hydrophila*, *B. cereus*, *B. panthotenticus*, *P. chlororaphis*. These bacteria were cultured as surface contaminants from okra fruits. All the concentrations (5 to 20%) of the test plant mostly inhibited *B. cereus*, followed by *A. hydrophila* while the list inhibited was *B. subtilis.*

Table 1**: Effects of different concentrations of cold aqueous extracts of *V. amygdalina* against post harvest bacteria of okra fruits.**

Concentration of extracts %	Bacterial Isolates				
	B. subtilis	*P. chlororaphis*	*A. hydrophila*	*B. cereus*	*B. panthotenticus*
Control	10.43a	5.89e	6.60c	9.70c	10.23b
5	0.33e	0.54d	0.77c	1.08b	0.92b
10	0.50e	0.90c	1.11b	1.41a	1.04c
15	0.68e	1.03d	1.12c	1.46a	1.18b
20	0.83e	1.65a	1.19d	1.51b	1.43c
LSD	0.51	0.07	0.12	0.07	0.94

Values followed by the same letter are not significantly different at ($p<0.05$ at Fischer's LSD)

Table 2 showed the effects of varied concentrations of hot extracts of *V. amygdalina* against post harvest bacteria of okra viz: *A. esculentus* that *B. subtilis*, *A. hydrophila*, *B. cereus*, *B. panthotenticus*, *P. chlororaphis*. All the concentrations (5 to 20%) of the test plant inhibited the

bacterial isolates. However, 10% of the hot extract of the test plant mostly inhibited *A. hydrophila* (1.06) followed by 20% against *B. panthotenticus (0.97),* while the list inhibited was 5% extract concentration against *B. subtilis (0.26)*

Table 2: Effects of different concentrations of hot water extracts of *V. amygdalina* against post harvest bacteria of okra fruits.

Concentration of extracts %	Bacterial Isolates				
	B. subtilis	*P. chlororaphis*	*A. hydrophila*	*B. cereus*	*B. panthotenticus*
Control	10.43a	5.89e	6.60c	9.70c	10.23b
5	0.26e	0.34d	0.73c	0.95a	0.87b
10	0.31e	0.39c	1.06c	0.82c	0.91a
15	0.51c	0.43d	0.90b	0.91b	0.94a
20	0.61ce	0.42d	0.59b	0.59b	0.97a
LSD	4.15	2.28	2.36	3.64	3.85

Values followed by the same letter are not significantly different at ($p<0.05$ at Fischer's LSD)

DISCUSSION

Overcoming food and nutritional insecurity among women, pregnant and lactating mothers, and children under 5 years of age, remain a daunting challenge in many developing countries in sub-Saharan Africa (Andersen *et al*., 2003; Tchientche *et al.*, 2013). The importance of foodstuffs as a source of dietary nourishment depends on both major content and the level of minor nutrient constituents in the diet. Therefore, increase in cultivation and consumption of okra as nutrient-rich indigenous Okra pods that will help to supplement/formulate the diets should be encouraged as additional source of minerals to the diet of the indigenous people in order to alleviate the problems associated with malnutrition in the developing countries (Habtamu, *et al*., 2016). Therefore, Okra pods could be engaged in fortification, formulation and supplementation of other food materials. The result of this study showed that the use of plant extract as one of the main means of curing okra superficial contaminants mostly in developing countries with minimal adverse effect, this corroborated the finding of Chethana, *et al.*, (2012). The use of pesticides globally has portent environmental and health challenge to man and animal. Hence, there is a need to explore biological means of controlling harmful microbes. Leaves of *V. amygdalina* serve as soup delicacy to stimulate digestive system.

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