Effects Of Botanical Extracts And A Synthetic Fungicide On Severity Of Cercospora Leaf Spot (Cercospora Sesame Zimm) On Sesame (Sesamum Indicum L.) Yield Attributes Under Screen House Condition In Ardo-Kola, Taraba State, Nigeria.

Tunwari, B.A., Nahunnaro, H

ABSTRACT: Screen house trials consisting five plant extracts including a synthetic fungicide (*Azadirachta indica*, *Jatropha curcas* Linn., *Alium sativum*, *Ocimum gratissimum*, *Chromolaena odorata* and Benlate) were tested at the Taraba State University(TSU) Research Farm centre (Ardokoa: Latitude 08^0 53′N and Longitude 11^0 19′E) in a completely randomized design arrangement. The results of the screen house experiments proved that the plant extracts (Garlic, Ocimum, Chromolaena and Benlate) led to 15.56 to 15.22% reduction of severity respectively, which culminated in to 31.18 to 30.26%, 14.04 to 15.79%, 21.21 to 21.76%, and 12.83 to 20.29% increased in capsules per plant, seeds per capsule, 1000 seeds weight and seeds yield per plant in 2011 and 2012 respectively over the unsprayed control treatment.

Key words: Sesame, *Cercospora sesami*, botanical extracts, growth attributes.

1. INTRODUCTION

Sesame or beniseed called 'Ri'idi' in Hausa, 'Igogo' in Igbo and 'Yanmoti' in Yoruba is probably the most ancient oil seed known and used by man. It is one of the important oil crops and ranked 9th among the top thirteen oil seed crops, which make up to 90% of the world edible oil production (Kafiriti and Deckers, 2001). The world hectarage exceeds 6 million and world output stood at 2.4 million tones (Phillips 1997; Dudley et al., 2000). In Africa, Nigeria is the second largest producer after Sudan. Sesame oil is of good quality. According to Dudley et al. (2000), the oil is used for cooking, baking, candy making, soaps, lubricant, body massage, hair treatment, food manufacture, industrial uses and alternative medicine (blood pressure, stress and tension). Also, Irvine (1970) reported that leaves are used in vegetable soup while seeds are consumed when fried and mixed with sugar in most African countries and stems are used in making paper, fuel wood and source of potash after burning. However, sesame cultivation is faced with several set-backs such as poor yielding potential of the available varieties coupled with constant devastating effects of pests and diseases (Nyanapah et al., 1995).

- *Tunwari B.A is currently pursuing PhD in Plant Pathology, Department of Crop Production and Protection,Federal University Wukari, Katsina - Ala Road, P.M.B. 1020, Wukari, Taraba State, Nigeria. E-mail: adamubilkoya@gmail.com Phone: +2348066043907.*
- *Nahunnaro, H. Senior Lecturer, Department of Crop Protection Modibbo Adama University of Technology, P.M.B. 2076, Yola, Adamawa State, Nigeria E-mail: adamubilkoya@gmail.com Phone: +2348038298393.*

Amongst these, cercospora leaf spot (CLS) has been identified as one of the most prevalent diseases in Nigeria and other parts of Africa of which there is no satisfactory control methods being evolved yet (Poswal ad Misari, 1994; Kolte, 1985; Nyanapah et al., 1995). The Cercospora leaf spot (CLS) disease caused by Cercospora sesami Zimm is a major destructive disease of sesame (Sesamum indicum L..) grown in Nigeria. The disease is endemic in most of the sesame growing areas of Takum, Donga, Wuakeri, Bali, Kurmi, Karim-Lamido in Taraba state and major sesame growing regions of Nigeria which infects the leaves, stem, branches, petiole, pods and causes severe seed yield loss and also deterioration in the quality of the seed. Under severe infections, disease has been reported to cause 22 to 53 per cent loss in seed yield (Enikuomehin et al., 2002). Over time, chemical control has been practiced by farmers for higher gains (Gerken et al., 2001), but these pests and diseases can become resistant to chemical insecticides very quickly. Moreover, the misuse of chemical pesticides in terms of quantity applied or in dangerous combinations (Obeng-Ofori et al., 2002) have created a myriad of problems which include pest resistance, resurgence of pests, pesticide residues, destruction of beneficial fauna and environmental pollution (AVRDC, 2003b). Under such debilitating circumstances, interest in organic farming has been growing and therefore exploring alternative options to control CLS disease of sesame is a fundamental means of supporting the smallholder farmer to diversify into organic production and be able to tap into the high profits associated with organic products.

2. MATERIALS AND METHODS

Pathogen isolation: The causal organism *Cercospora* sp. was isolated from leaves showing typical leaf spot symptomps of *Cercospora* leaf spot. With the aid of forceps flamed and cooled in methylated spirit, the leaves were carefully transferred unto humid chambers made up of petri dishes that were under laid with filter papers at both covers and wetted thoroughly. The whole process was carried out under sterile conditions in a Laminar flow Chamber. The plates were then kept on the bench in a previously fumigated clean room where they could access sunlight at room temperature for 72 hours, and examined for sporulation under Trinocular Microscope.

Purification and Examination: After 72 hours pieces of leaves around the spot areas were cut using sterile needles and transferred unto glass slides containing cotton blue stain. The slides were then covered with lids, pressed tightly with fingers to elimnate air bubbles and mounted on a trinocular microscope. Observed conidia from separate conidiophores were picked using glass needle (that was previously flamed and cooled in methylated spirit) and transferred aseptically in to six different plates containing V8 agar using single spore technique as part of the culture purification process. The process was carried out under sterile condition. The plates were placed in an incubator for 7 days. After that the plates were removed and kept on the bench in a previously fumigated clean room where they could access sunlight at room temperature for 2 -3 months where morphological characteristics of the fungus were observed daily during the growth period until the cultures had fully sporulated. Detailed microscopic examinations were carried out during the growing stages to reveal the organism. In each case, temporary slides were prepared and viewed under x 45 objective lens of a trinocular microscope.

Source and preparation of extracts: Plant extracts from leaves of Azadirachta indica, Jatropha curcas Linn., Alium sativum (bulbs), Ocimum gratissimum(L.), and Chromolaena odorata that were obtained from within and around Modibbo Adama University of Technology, Yola were used for the field trials. These plants were being selected because they were associated with pest management and disease control practices in several parts of Africa (Adesegun, et al., 2012; Essien and Akpan, 2004; Enikuomehin and Peters, 2002; Ogbebor and Adekunle, 2008). The crude extracts were obtained by first of all sterilizing plant parts in 10% Sodium hypochlorite (NaCl) for 1 minute, washed 5 times in distilled water, air dried and later oven dried at 70^0C for 20 minutes according to Akinbode and Ikotun (2008). Thereafter, the plant materials were ground using mortar and pestle and sieved in a 40 mm sieve into a fine powder. To obtain extracts 100 g of the grounded powder (packaged according to plant species) were weighed in to conical flask. After that 100 mls distilled water was added to form a ratio of 1:1 weight over volume basis. This was then corked with rubber brim and shaken well for 20 minutes to mix and allow to stand overnight (24 hours) at room temperature and the content filtered using a muslin cloth. To obtain 10% concentration for spray, 100 mls of the filtered extract suspension was added to 900 mls distilled water to make up to 1 litre. This was then kept in glass bottles until needed.

Pots preparation, planting and maintenance: The effects of the extracts on disease development and spread were determined using potted plants in the screen house in 2011 and 2012. Seeds of sesame sown in 25 cm diameter pots containing 10 kg previously heat-sterilized loam soil. Ten seeds were sown at a depth of 3cm in each pot. Planting was done according to treatment combinations. At two weeks of planting the seedling density was reduced (thinned) to 5 plants /bucket. The pots were arranged according to completely randomized Design (CRD) and replicated four times. After 3 weeks of good establishment, the seedlings were sprayed with spore suspension of $1x10^4$ spores/ml in distilled water of the pathogen using hand sprayer. The plants were then covered with perforated transparent polythene sheets to build relative humidity and symptoms of Cercospora leaf spot were observed as from 4weeks after planting. The plant extracts (10%) was sprayed using hand sprayer (2 litres capacity) from 4 weeks after planting (WAP) and repeated at 6, 8 and 10 WAP. The synthetic fungicide, Benlate was sprayed at the recommended rate of 2 kgha-1 at 4 and 8 WAP, while the control was left unsprayed.

Disease Assessment: Severity was estimated by assessing 5 plants per pot and determining overall score according to percentage area covered using a scale of 1-7 (Table 1) as outlined by Enikuomehin *et al*., (2002) with some modifications. This helped to determine the extend of establishment of the disease. The following formula was used in determining the severity of infections:

$\sum nx / N (7)$ (Chaube and Pundhir, 2005),

where x = grade per leaf;

n = number of leaves per given grade,

N = total number of leaves examined/pot;

7 = the maximum disease grade.

The following formulas were also used to estimate percentage reduction ad yield increase over the control:

$$\text{Disease reduction (\%)} = \frac{\text{severity in control-severity in trt}}{\text{Disease severity in control}} \times 100$$

$$\text{Yield increase (\%)} = \frac{\text{Yield in trt plot} - \text{Yield in control plot}}{\text{Yield in control plot}} \times 100$$

Four replications in a CRD were maintained for each treatment for the 2011 and 2012 screen house experiments. Data collected were subjected to analyses of variance (ANOVA), using the general linear model (GLM) procedure of SAS Version 9 (SAS, 2005). Treatment means separation was carried out with the Duncan's Multiple Range test (DMRT) according to Gomez and Gomez (1984) and the probability of treatment means being significantly different was set at $P < 0.05$.

3. RESULTS

Effect of botanical extracts on severity of Cercospora leaf spot (CLS): Results of the two year screen house experiments in 2011 and 2012 revealed highly significant ($P \leq 0.001$) effects of plant extracts on severity of CLS of sesame compared to unsprayed control. Ocimum, Chromolaena which are significantly at par with Garlic and Benlate recorded the lowest severity of 38 to 39%, which is highly significantly different from the other plant extracts (Neem and Jatropha) tested, while the unsprayed control produced the highest severity of 45 to 46% (Tables 2 and 3) at 12 weeks after sowing (WAS) in 2011 and 2012 respectively.

Effect of plant extracts on Capsules per plant (CPP): Plant extracts had significant effect on the number of capsules per plant (Tables 2 and 3) for the 2011 and 2012 screen house trials. Cold extracts obtained from Garlic, Ocimum, Chromolaena and the synthetic fungicide benlate had significantly ($P \leq 0.001$) more number of capsules per plant of 187 to 192 than the other plant extracts (Neem and Jatropha), while the lowest capsules per plant of 143 to 146 were obtained from the unsprayed control under the 2011 and 2012 screen house trials respectively.

Effect of plant extracts on seeds per capsule (SPC): Extracts obtained from plants of Garlic, Ocimum, Chromolaena and the synthetic fungicide benlate also significantly gave the highest seeds per capsule (65 to 67), while the lowest numbers of seeds per capsule (57 to 58) were obtained from unsprayed control respectively during the 2011 and 2012 screen house experiments Table 2.

Effect of plant extracts on 1000 seeds weight (OTSW): Plant extracts has highly significant effect on the weight of 1000 seeds in 2011 and 2012 screen house trials (Tables 2 and 3). The highest values of 3.10 to 3.17 g were obtained from the extracts (Garlic, Ocimum, Chromolaena and the synthetic fungicide benlate), while the lowest OTSW of 2.57 to 2.55 g were obtainable from unsprayed control in the 2011 and 2012 trials respectively.

Effect of plant extracts on seeds yield per plant (SYPP): Tables 2 and 3 show the mean performance of the effects of plant extracts evaluated for control of CLS as related to yield parameters under screen house condition in 2011 and 2012. Seedlings treated with extracts of Garlic, Ocimum, Chromolaena and the synthetic fungicide benlate were among the plants with the highest seed yield per plant (7.34 to 7.54 g). It was also noted that plots treated with plant extracts (Garlic, Ocimum, Chromolaena, Jatropha and neem) and benlate significantly gave the highest seed yield per plant, while the lowest mean values of 6.56 to 6.22 g respectively were obtained from plots of unsprayed control in 2011 and 2012 during screen house experiments respectively.

Table 1: Disease score for Cercospora leaf spot disease of sesame

Scale	Disease severity (%)	Resistant Category	Rating	Leaf spot characteristics
1.	0 – 14	Immune (I)	No disease	No trace of infection
2.	14.1 – 29	Highly Resistant (HR)	Hypersensitivity	Hypersensitive spot on lower leaves only
3.	29.1 – 43	Resistant (R)	Trace infection	Small lesions on lower leaves only
4.	43.1 – 57	Moderately resistant (MR)	Slight infection	Small lesions on lower and upper leaves and stem
5.	57.1 – 71	Moderately usceptible (MS)	Moderate infection	Advanced lesions[1] on upper and lower leaves, with or without new infections on stem and petiole
6.	71.1 – 86	Susceptible (S)	Severe infection	Advanced lesions on upper and lower leaves, flower, buds, stems and petiole and slight infection of pod[1]
7.	86.1 - 100	Highly susceptible (HS)	Very Severe infection	All features of 6 above with severe infection of pod

[1]Advanced lesion is characterized by a dark to dark-brown spots with a whitish to straw-coloured or perforated centre (Enikuomehin *et al*. , 2002).

Table 2: Effects of some selected plant extracts and a synthetic fungicide on severity of CLS as it affects some yield related parameters of sesame following screen house experiment at Ardokola, Taraba, Nigeria in 2011.

Effect of extracts on severity of CLS on sesame in screen house trials at Ardo-kola in 2011					
Treatments	Cersev 12	CPP	SPC	OTS W	SYPP
Neem	39.52b	172.25b	61.82b	2.79c	6.89b
Jatropha	38.74c	169.44b	61.01b	2.89b	6.95b
Garlic	38.12cd	185.81a	65.19a	3.11a	7.34a
Ocimum	37.77d	189.94a	65.36a	3.10a	7.37a
Chromolaena	37.77d	187.56a	65.40a	3.12a	7.37a
Benlate	38.12cd	189.38a	65.42a	3.13a	7.40a
Control	44.65a	143.44c	57.04c	2.57d	6.56c
Mean	39.24	176.83	63.03	2.95	7.13
CV (%)	2.60	7.47	2.85	2.06	1.72
S.E.	1.02	13.21	1.80	0.06	0.12
p-value	0.001	0.001	0.001	0.001	0.001

Means in the same column followed by the same letter(s) are not significantly different (0.05) using Duncan's Multiple Range Test. Cersev12 = Cercospora leaf spot severity at 12 WAS; CPP = Capsules plant-1; SPC = Seeds capsule-1 ; OTSW = One thousand seed weight (g); SYPP = Seed yield per plant (g).

Table 3: Effects of some selected plant extracts and a synthetic fungicide on severity of CLS as it affects some yield related parameters of sesame following screen house experiment at Ardokola, Taraba, Nigeria in 2011.

Effect of extracts on severity of CLS on sesame in screenhouse trials at Ardo-kola in 2012					
Treatments	Cersev12	CPP	SPC	OTSW	SYPP
Neem	39.52b	172.25b	61.82b	2.79c	40.98b
Jatropha	38.74c	169.44b	61.01b	2.89b	40.88b
Garlic	38.12cd	185.81a	65.19a	3.11a	39.02c
Ocimum	37.77d	189.94a	65.36a	3.10a	38.93c
Chromolaena	37.77d	187.56a	65.40a	3.12a	38.84c
Benlate	38.12cd	189.38a	65.42a	3.13a	38.66c
Control	44.65a	143.44c	57.04c	2.57d	46.26a
Mean	39.24	176.83	63.03	2.95	40.50
CV (%)	2.60	7.47	2.85	2.06	1.78
S.E.	1.02	13.21	1.80	0.06	0.72
p-value	0.001 **	0.001 **	0.001 **	0.001 **	0.001 **

Means in the same column followed by the same letter(s) are not significantly different (0.05) using Duncan's Multiple Range Test. Cersev12 = Cercospora leaf spot severity at 12 WAS; CPP = Capsules plant-1; SPC = Seeds capsule-1 ; OTSW = One thousand seed weight (g); SYPP = Seed yield per plant (g).

4. DISCUSSION

Screening of some selected plant extracts (Garlic, Ocimum, Chromolaena, Jatropha, neem and Benlate) showed less amount of Cercospora leaf spot disease on sesame for in vivo antifungal activity. The results also proved that the plant extracts (Garlic, Ocimum, Chromolaena and Benlate) led to 15.56 to 15.22% reduction of severity respectively, which culminated in to 31.18 to 30.26%, 14.04 to 15.79%, 21.21 to 21.76%, and 12.83 to 20.29% increased in capsules per plant, seeds per capsule, 1000 seeds weight and seeds yield per plant in 2011 and 2012 respectively over the unsprayed control treatment. Similar works by Adesegun *et al*. (2012), Zarafi and Moumoudu (2010); Ogbebor and Adekunle (2008) revealed that in vitro and in vivo experiments using cold water extracts of Ocimum leaf, Chromolaena leaf, garlic bulb, mahogamy seed, ginger rhizome and shear butter leaf could be used successfully as environmentally safe and economical fungicides against fungal diseases. Fungitoxic activities of *Ocimum* species and *A. sativum* against plant pathogens have been reported (Tewari and Dath 1984, Lakshmanan 1990, Tewari 1995, Ogbebor and Adekunle 2005, Ogbebor *et al.* 2007). *In vivo* experiment. Lakshmanan (1990) reported the effectiveness of antifungal properties of *A. sativum.* Ogbebor *et al.* (2007) demonstrated high antifungal properties of aqueous extracts of *O. basilicum* and *A. sativum* on *Colletotrichum gloeosporioides*. Adesegun *et al*. (2012) revealed that *Ocimum gratissimum* aqueous extract gave comparable reduction in mycelia growth of southern blight (*Sclerotium rolfsii*) of Tomato, which is also in agreement with the work of Awuah (1989) who found that extracts of *O. gratissimum* led to 24.6% reduction in radial growth of Rhizopus species and 60% reduction of *Ustilaginoidea virens*. Several studies reported the direct effect of neem leaf and fruit extracts on target pests and pathogens (Eppller 1995; Amadioha, 2000). According to the work of Eman (2011) lemon grass oil was found at all concentrations had strong capacity to reduce the spread of CLS on Okra plants. The works of Wilson *et al*. (1997) on 345 plants extracts tested showed that Allium and Capsicum species were most effective. *Allium sativum* was demonstrated earlier to have good anti-fungal activity and useful as post-harvest treatment (Ark and Thompson, 1959), has not been commercialized.

5. CONCLUSION

This finding can be of some practical value to agricultural extension workers in the affected areas in advising farmers on the expected yield of their crop at various Cercospora leaf spot intensity. It could therefore, be concluded that the plant extracts Garlic, Ocimum and Chromolaena, were comparable to synthetic fungicide (benlate) in reducing the amount of Cercospora leaf spot on sesame. However, further evaluation of the materials using different parts, determination of the economic rate, formulation, packaging, method and frequency of application are needed.

6. REFERENCES

[1]. E.A., Adesegun, E.O. Ajayi, O.S. Adebayo, A.K. Akintokun, and O.A., Enikuomehin, Effect of Ocimum gratissimum (L.) and Aframomum melegueta (K. Schum.) Extracts on the Growth of Sclerotium rofsii (Sacc.). International Journal of Plant Pathology. ISSN 1996-0719/ DOI: 10.3923/JPP.2012.

[2]. O.A. Akinbode, and T. Ikotun, Evaluation of some bioagents and botanicals in in vitro control of Colletotrichum destructivum. African Journal of Biotechnology 7(7): 868-72, 2008.

[3]. A.C. Amadioha, Controlling rice blast in vitro and in vivo with extracts of Azadirachta indica. Crop Pro t e c tion. 19: 287-290, 2000.

[4]. Ark, P.A., and Thimpson, J.P., Control of certain diseases of plants with antibiotics from garlic(Allium sativum L.) . Plant Diseases Reports. 43: 276-282, 1959.

[5]. AVRDC, Effects of natural and synthetic pesticides in controlling Diamond back moth in cabbage. Progress report 2002. Shanhua, Taiwan: AVRDC-the World Vegetable Centre, pp.157-158, 2003b.

[6]. R.T. Awuah, Fungitoxic effects of extracts from some West African plants. Annals of Applied Biology, 115: 145 – 453, 1989.

[7]. J.M., Babu, D. Ahmad and K.. Vinod, Bio-efficacy of plant Extracts to control Fusarium solani f.sp. melongenae incitant of Brinjal wilt.Global Journal of Biotechnology and Biochemistry 3(2): 56-59,2008.

[8]. H. S. Chaube, and V. S. Pundhir, Crop diseases and their management. Prentice- Hall of India, New Delhi.703pp, 2005.

[9]. S.H. Eman, Farrag, First Record of Cercospora Leaf Spot Disease on Okra Plants and its Control in Egypt. Plant Pathology Journal, 2011. ISSN 1812-5387/DOI: 10.3923/ppj, 2011.

[10]. O.A., Enikuomehin,. and O.T. Peters, Evaluation of Crude extracts from Some Nigerian plants for The control of field diseases of sesame (Sesamum indicum L.). TropicalOilseeds Journal, 7: 84-93, 2002.

[11]. O.A., Enikuomehin, V.I.O., Olowe, O.S. Alao, and M.O., Atayese, Assessment of Cercospora leaf spot disease of sesame in different planting dates in South-western Nigeria. Moor Journal Agricultural Research. 3: 76- 82, 2002.

[12]. A. Eppler, Effects on viruses and organisms. In: schmutterer, H. (ed.), The Neem Tree. Sources of Unique Natural products for Integrated Pest Management, medicine Industry and Other Purposes, pp. 93-117. Weinheim, New York, Basel, Cambridge, Tokyo: VCH, 1995.

[13]. J.P. Essien, and E.J. Akpan, Antifungal activity of ethanol leaf extract of Eucalyptus canaldulensis Dehn. against ringworm pathogens. Global Journal of Pure and Applied Science. 10: 37-41, 2004.

[14]. T.S., Dudley, W. James and A. Mccallum, Texas Agricultural Experimental Station, College Station and Yankum. Prepared April 12, 2000. P125, 2000.

[15]. A. Gerken, J.V. Suglo and M. Braun, Crop protection policy in Ghana. Pesticide Policy Project produced by Plant Protection and Regulatory Services Directorate of Ministry of Food and Agriculture, Accra, 2001.

[16]. K.A. Gomez and A.A. Gomez, Statistical procedures for Agricultural Research; 2nd ed. John Willey and Sons: pp 306 – 308, 1984.

[17]. J., Irvine. West African Crops 3rd edition, Oxford University Press, Pp 71-74, 1970.

[18]. E.M. Kafiriti and J. Deckers, Sesame in:Raemnekers, Rlt (ed.) Crop Production in Tropical Africa DGIC, Brussels, Belgium, P345, 2001.

[19]. S.J. Kolte, Diseases of annual edible oil seeds crop. Vol. II Rapeseed – Mustard and Sesame Diseases. CRC Pres, Inc. Boca Raton Florida, pp 83- 122, 1985.

[20]. P. Lakshmanan, Effect of certain plant extracts against Corynespora cassiicola. Ind. J. Mycol. And Plant path. 20 (3): 267- 269, 1990.

[21]. J.O., Nyanapah, P.O. Ayiecho, and J.O. Nyabundi, Evaluation of sesame cultivars for resistance to Cercospora leaf spot. East Africa Agriculture and Forestry Journal 60(3), 115-121, 1995.

[22]. O.N. Ogbebor, and A.T. Adekunle, Inhibition of Drechslera heveae (Petch) M. B. Ellis, causal organism of Bird's eye spot disease of rubber (Hevea brasiliensis Muell Arg.) using plant extracts. African Journal of General Agriculture. Vol. No. 1, 2008.

[23]. O. N., Ogbebor, A. F. Adekunle, and G. A. Evueh, Inhibition of Drechslera haveae (Petch) M.B. Ellis,causal organism of birds' eye spot disease of rubber (Hevea brasiliensis Muell Arg) using plant extracts. A paper presented at 34th Annual conference of the Nigerian Society for Plant Protection(NSPP) held at Nasarawa State University, Keffi, 17- 21st September, 2007.

[24]. D. Obeng-Ofori, E.O. Owusu and E.T. Kaiwa, Variation in the level of carboxyl esterase activity as an indicator of insecticide resistance in populations of the diamond back moth, Plutella xylostella (L.) attacking cabbage in Ghana. J. Ghana. Sci. Assoc., 4(2): 52-62, 2002.

[25]. T.A. Phillips, Agricultural Note Book Series, Longman publishers, London, pp 61-62, 1997.

[26]. M.A.T. Poswal, and S.M. Misari, Resistance of Sesame Cultivars to Cercospora leaf spot induced By Cercopora sesami. Zimm. Discovery and Innovations 6:66-70, 1994.

[27]. SAS, Statistical Analysis Software Package Version 9. SAS Institute Inc., Cary, NC, USA, 2005.

[28]. S.N. Tewari and P. Dath, Effects of some leaf media the growth of three fungal pathogens of rice. Indian Phytopathology, 458– 461pp, 1984.

[29]. S.N., Tewari, Ocimum sanctum L., a botanical fungicide for rice blast control. Trop. Sci. 35: 263-273, 1995.

[30]. C.I., Wilson, J.M., Solar, A.. Ghaouth and M.E. Wisniewski, Rapid evaluation of plant extracts and essential oils for antifungal activity against Botrytis cinerea. Plant Diseases 81: 204-210, 1997.

[31]. A. B. Zarafi and U. Moumoudou, In vitro and in vivo control of pearl millet midrib spot using plant extracts. Journal of Applied Biosciences 35: 2287- 2293 ISSN1997–5902, 2010..