Agrosearch(1999) 5, 1 &2 62 - 69

EFFECT OF PLANT AGE AND INJURY ON THE PATHOGENICITY OF *CHOANEPHORA CUCURBITARUM* IN OKRA *ABELMOSCHUS ESCULENTUS MOENCH.*

O. S. BALOGUN AND J.O. BABATOLA
Department of Crop Production,
University of Ilorin, Ilorin, Nigeria.

Abstract

Experiments were carried out using potted plants to study the effect of inoculating okra, Abelmoschus esculentus at various stages of growth with Choanephora cucurbitarum spores in suspension of water, and also to evaluate the role of injury to plant seeds, foliage, blossom and fruits in the pathogenicity of the fungus. Inoculation of plants at planting, seedling emergence or 10 days after planting caused leaf infection significantly earlier than inoculations made at later stages of growth. Whereas inoculation alone, regardless of its frequency was found to have no significant reduction effect on plant height as at first harvest, the frequency of injury appeared to influence height. Wounding plants, at least once, followed by inoculation with fungal spores (s) significantly reduced height compared to the control, indicative of the predispositional effect of injury to infection. Inoculations or injuries alone regardless of the frequency did not significantly affect the number of days to flowering. However, inoculations at older stages of seedling growth and at flowering caused significantly higher number of rotted buds and fruits, hence higher abortion than earlier inoculations. Consequently, lesser number of fruits and total edible fruit weight were recorded on such plants. On the other hand, higher intensities of wounding combined with inoculations not only significantly delayed flowering, they also resulted in significantly higher percentage rot and subsequent yield reductions than early wounds with or without inoculation.

Key words: inoculation, infection, rots and abortion, edible fruit yield.

INTRODUCTION

Okra, *Abelmoschus esculentus* Moench an important fruit vegetable widely grown for the edible mucilaginous pod (Gbile, 1981), is a source of vitamins (Sai, 1965; Oyolu, 1981) and proteins (Purseglove, 1969) in most tropical countries, including Nigeria.

The crop is susceptible to attack by many pests and disease-causing organisms that can prevent the optimization of yield. Among the pests are flea beetles, *Podagrica* spp and other defoliators, which perforate the leaves reducing photosynthetic area and may predispose the plant to pathogenic attack. Fungal attack is a major cause of disease in okra. It may occur in complex with nematodes such as in the case of *Rhizoctonia solani* and *Meloidogyne incognita* (Golden and Van Gundy, 1975) or alone. Of importance is *Choanephora cucurbitarum* a fungus, which induces premature fruit rot and abortion (Adebanjo and Dede, 1985). According to Barnett and Lilly (1955), *C. cucurbitarum* thrives best at 100 % relative humidity and 25 °C and it is stimulated to produce large sporangia at 31°C. Warm wet conditions in densely planted crops favours its attack (Holliday, 1980). According to Tai Luang et al (1975), *C. cucurbitarum* attack could lead to as much as 20% yield loss in the field.

As at now, there is still a dearth of information on the host-pathogen interactions, especially in the aspect of time and mode of inoculation, as well as predispositional factors, vis-à-vis fungal infectivity and susceptibility of the host plant. Among the objectives of this study, therefore, were to determine the influence of mechanical injury, such as caused by insect pests, to seeds prior planting, seedlings, foliage, blossom and fruits on the pathogenicity of the fungus under varying intensity of inoculation. This is with a view to determining that which could bring about the infection that is most detrimental to growth and yield.

MATERIALS AND METHODS

Cultivar NHAe 47-4, a short duration, dwarf cultivar of okra obtained from the National Horticultural Research Institute, Ibadan Nigeria was used in two sets of experiments carried out in a screen house. Plants were raised on steam-sterilized sandy loam soil inside 5-litre plastic buckets.

In experiment 1, inoculation of the plants in randomly selected sets of five pots, per inoculation day, was done immediately after planting (day 0), and then 5, 10, 15, 25, and 45 days after planting respectively. Plants were kept under moistened polythene chamber immediately after inoculation on each occasion for 24 hours to provide an atmosphere conducive to germination of the spores. Five plants inoculated with water only served as control.

In experiment 2, four levels each of two factors were factorially combined to obtain 16 treatments. The factors were (A) mechanical injury whereby A0 = no injury; A1= seed only injured; A2 = seed + 35 day old seedling injured; and A3 = seed + 35 day old seedling + flowers/fruits injured). The other was Inoculation of *C. cucurbitarum* (B) whereby B0 = not inoculated; B1= seed only inoculated; B2= seed + 35 day old seedling inoculated; and B3= seed + 35 day old seedling + flowers/fruits inoculated). The treatment combinations were as follows: A0B0, A0B1, A0B2, A0B3, A1B0, A1B1, A1B2, A1B3, A2B0, A2B1, A2B2, A2B3, A3B0, A3B1, A3B2 and A3B3. Each treatment combination was replicated 3 times giving 48 observations altogether.

Wounding preceded inoculation in treatments where they were combined. Wounding of seeds was by puncturing 3 holes on each, while the four uppermost leaves of each injured plant received ten 5 mm diameter holes by cock- boring. Flower buds, blossom, and fruits were wounded by puncturing 10 random holes with pin, simulating insect damage. Inoculation was done by spraying with a hand spray gun, about 1.0 x 10^4 fungal spores suspended in 100 ml sterile distilled water, per pot. The fungal spores were obtained from a culture isolated from infected flowers of okra. NPK 15:15:15 fertilizer was applied at the rate of 5 g per pot three weeks after germination in both experiments. Mature edible pods were harvested by hand picking 10 days after flower opening on each plant.

Records of plant height on each inoculation day (in Experiment 1 only) and at first harvest (in both experiments), number of days to infection of leaves, and number of leaves infected after inoculation were made. Other parameters recorded include number of buds prior flower opening per plant and the percentage of such that got rotten, days to flower opening, fresh weight and number of edible pods per plant, average pod weight and pod length. All data were subjected to analysis of variance.

RESULTS AND DISCUSSION

Emerging as well as young seedlings of okra succumbed to infection of the foliage earlier than the older plants. Only the young leaves of the older plants were found susceptible while old leaves succumbed only if they had been wounded before inoculation. *Tai Luang Juan et al (1975)* had also reported such with seed isolates of the fungus.

In experiment 1, as regards the number of days it took symptoms to manifest on the foliage, inoculations on day 5 i.e. at germination, apparently led to the earliest infection with a mean of 3.6 days post inoculation (dpi). This was not significantly different from day 10 (4.2 dpi) but different from that on day 0 (i.e. at seeding), which was 6.4 days after inoculation or 1.4 days after germination. This was not surprising since the emerging tender hypocotyls and primary leaves may have been exposed to the fungal spores while still inside the soil. The manifestation of symptom of infection in seedlings inoculated on day 15 took significantly longer than the preceding groups but was significantly lower than those done on days 25 (14.0 dpi) and day 45 (14.6 dpi), which did not differ significantly between each other (Table 1).

In experiment 2, young and old leaves that were injured before inoculation manifested the symptoms as early as 3 days after inoculation. By day 15, the unwounded but inoculated seedlings had also shown symptoms of infection. Only the initial 2 leaves were infected in both wounded and unwounded inoculated seedlings (Table 2).

Inoculation alone, regardless of its frequency was found to have no significant reduction effect on plant height as at first harvest. The frequency of injury, however, appeared to influence height. For instance, in experiment 1, infection did not significantly reduce the plant height at harvest compared to the control (Table 1). As shown on Table 2, in experiment 2 infections per se, regardless of injury level (i.e. A0B1, A0B2 and A0B3) also did not significantly reduce the plant height.

On the other hand, whereas injury alone of the seed before planting did not significantly reduce the height of plant measured at harvest, two or three levels of injury alone (i.e. A2B0 and A3B0) while not being significantly different from one level of injury (i.e. A1B0), significantly reduced height compared to the control (i.e. A0B0). Injuring plant, at least once, followed by inoculation (s) significantly reduced height compared to the control, and that is indicative of the predispositional effect of injury to infection.

Linear regression analysis showed a correlation (r= 0.93), between the incidence of rotted and aborted buds and the age of plants at inoculation (Fig. 1). The highest mean percentage, 51%, in experiment 1, was in plants inoculated at 45 days after planting, when most plants were already in full bloom. This however, was not significantly different from that made on day 25, which was towards late seedling stage (Table 1). In experiment 2, the apparently highest percentage, 67 %, was recorded in plants with maximum intensities of injury and inoculation (i.e. a_3b_3), although this too was not significantly different from those of plants with at least two levels of inoculation with or without injury.

Fungal inoculations alone i.e. without injury, did not cause any significant delay in the number of days to flowering in observed in both experiments. However, the control plants bloomed significantly earlier than plants that were wounded at least twice followed by inoculations. Both buds and blossoms, especially senescing ones, including immature and wounded fruits were found to have succumbed to infection after inoculation. Adebanjo and Dede (1985) had shown Choanephora to be a soft rot organism, which attack flowers and fruits of okra leading to their abortion.

Consequent upon the higher incidence of rots and abortion in plants that were lately inoculated (e.g. day 25 and 45 in experiment 1) (Table 1), or that were lately inoculated alone (e.g. A0B2 and A0B3) or inoculated after being wounded (e.g. A2B2, A2B3, A3B2 and A3B3 in experiment 2) (Table 2), significantly lower quantities of edible fruits were produced per plant compared to the control. The total edible fruit weight was also significantly lower than in plants either inoculated alone or wounded alone at the earlier stages of growth and the control. The negative relationship (r= - 0.88), between percentage rots/ abortion and the total edible fruit weight per plant is shown in

Figure 2. The average weight and length of pod did not differ significantly among treatments. This was due to the fact that all edible fruits were those that virtually escaped infection and were harvested the same number of days (10 days) after flower opening in any one case.

From the results of this study, we have been able to establish that inoculations of plants at the later stages of growth i.e. just before and during flowering had the greatest damaging effect on yield. Mechanical injury, as may also be inflicted naturally by insect pests, can predispose seedlings' foliage to infection by the fungal pathogen. Such wound spots may in turn serve as source of inoculum for later infection of the buds and flowers. Therefore, the use of suitable fungicide and insecticide would be appropriate at these stages to forestall serious yield loss especially in areas where other such environmental conditions that favour high incidence of the disease, as earlier identified by Barnett and Lilly (1955) and Holliday (1980), occur during the cropping season.

Table 1: Effect of varying time of inoculation of *Choanephora cucurbitarum* on disease development and some growth and yield parameters in cultivar NHAE 47 4 okra

Treatments (Inoculation time in days after planting)	Time of infection after inoculation (days)	Height before Inocu-lation (cm)	Height at first harvest 60 DAP (cm)	Mean no. of buds before flowering per plant	% rots per plant	No. of days to Flowering	Edible fruit weight per pot (g)	Mean pod weight (g)	Mean pod length (cm)	Mean no. of fruits per plant
Control	*	*	42.1	5.2	0a	45.6	104.1a	18.0	6.2	5.8a
Day 0	6.4a	0a	40.5	5.0	15.7ab	45.2	90.3ab	17.7	6.0	5.4a
Day 5	3.6b	2.8b	39.8	4.6	13.0ab	45.4	86.1b	16.6	6.0	5.2a
Day 10	4.2ab	6.4c	39.6	4.8	13.0ab	46.1	88.5b	16.5	6.1	5.4a
Day 15	9.8c	11.8d	40.6	4.6	21.0b	45.0	86.6b	16.1	5.8	5.4a
Day 25	14.0d	24.8e	41.5	4.4	50.0c	46.2	72.2c	17.3	6.1	4.2b
Day 45	14.6d	34.6f	44.0	5.4	50.7c	44.8	65.0c	18.2	6.2	3.6b
			N.S	N.S		N.S		N.S	N.S	

Figures within columns followed by the same letter(s) do not differ significantly at P= 0.05 using the New Duncan's Multiple Range Test

N.S - Not significant

Table 2: Effect of varying combinations of injury and inoculation of *C. choanephora* on plant height, severity of rots/abortion and fruit yield in cultivar NHAE 47-4 okra

Treatment combinations	Mean no of. leaves/plant infected at 15 DAP	Plant height at day 60 (1st harvest) (cm)	Mean no. of buds before flowering	Mean % rots per plant at 1st harvest	No.of days to flowering	Weight of edible fruits per pot (g)	Mean fruit weight (g)	Mean no. Mean no. of fruits per plant	Mean pod length (cm)
a0b0*	0.0 a	41.5 a	4.7	6.7 a	49.3 a	111.9 f	17.7	6.3 g	6.3
a1b0	0.0 a	37.8 abc	4.3	6.7 a	50.7 a	101.3 ef	18.0	5.7 fg	6.3
a2b0	0.0 a	32.6 cd	4.0	21.7 ab	53.0 abc	75.4 d	17.4	4.3 de	6.3
a3b0	0.0 a	32.9 cd	4.3	25.0 ab	53.7 abc	68.0 bcd	17.0	4.0 cd	6.1
a0b1	0.7 a	39.7 ab	4.0	6.7 a	51.0 a	101.4 ef	17.9	5.3 f	6.3
a1b1	2.0 b	35.3 bcd	4.0	19.4 ab	53.0 abc	89.8 e	17.9	5.0 ef	6.2
a2b1	2.0 b	32.3 cd	3.7	15.0 ab	57.7 de	71.9 cd	17.1	4.3 de	6.6
a3b1	2.0 b	34.5 cd	4.7	31.8 b	57.0 cde	71.0 cd	17.0	4.0 cd	6.3
a0b2	0.3 a	41.6 a	4.3	41.7 bc	49.7 a	74.5 cd	17.2	4.3 de	6.4
a1b2	1.7 b	35.1 bcd	4.3	38.3 bc	52.0 ab	69.5 cd	17.4	4.0 cd	6.4
a2b2	1.7 b	33.0 cd	4.3	55.6 bc	55.7 bcd	63.0 bcd	17.1	3.7 bcd	6.2
a3b2	0.7 a	30.8 d	4.0	47 8 bc	60.3 e	53.1 ab	17.7	3.0 ab	6.6
a0b3	1.3 b	39.7 ab	5.0	53.3 bc	50.3 a	69.7 cd	17.4	4.0 cd	6.2
a1b3	1.3 b	36.3 bc	4.3	55.6 bc	52.3 ab	63.8 bcd	17.4	3.7 bcd	6.2
a2b3	1.7 b	32.0 cd	4.3	55.6 bc	57.0 cde	58.5 bc	17.7	3.3 bc	6.3
a3b3	2.0 b	31.9 cd	4.0	67.2 c	57.3 cde	40.8 a	17.6	2.3 a	6.2
			N.S				N.S		N.S

Figures within columns followed by the same letter(s) do not differ significantly at P= 0.05 , New Duncan's Multiple Range Test.

* a_0 : No injury; a_1: Seed only injured; a_2 : Seed + 35 day old seedling injured; a_3 :Seed + 35 day old seedling + flowers/fruits injured.

b0 : Not inoculated; b1: Seed only inoculated; b2: Seed + 35 day old seedling inoculated; b3: Seed + 35 day old seedling + flowers/fruits inoculated. Treatments were derived by factorial combination.

N.S means Not significant

* Not applicable to the control

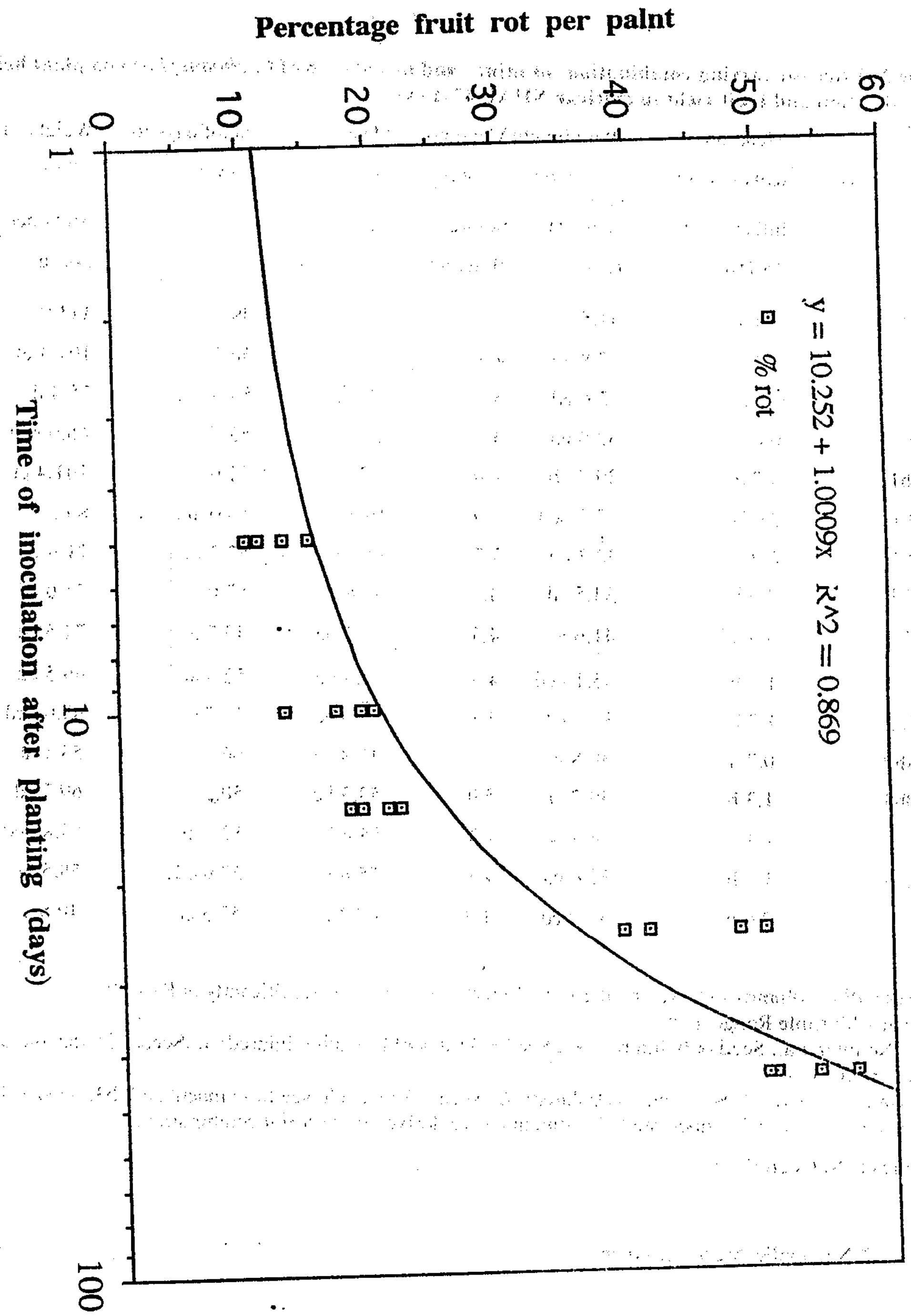

Fig 1: Relationship between age of plants at inoculation and incidence of fruit rots and abortion in okra, cv NHAe 47-4

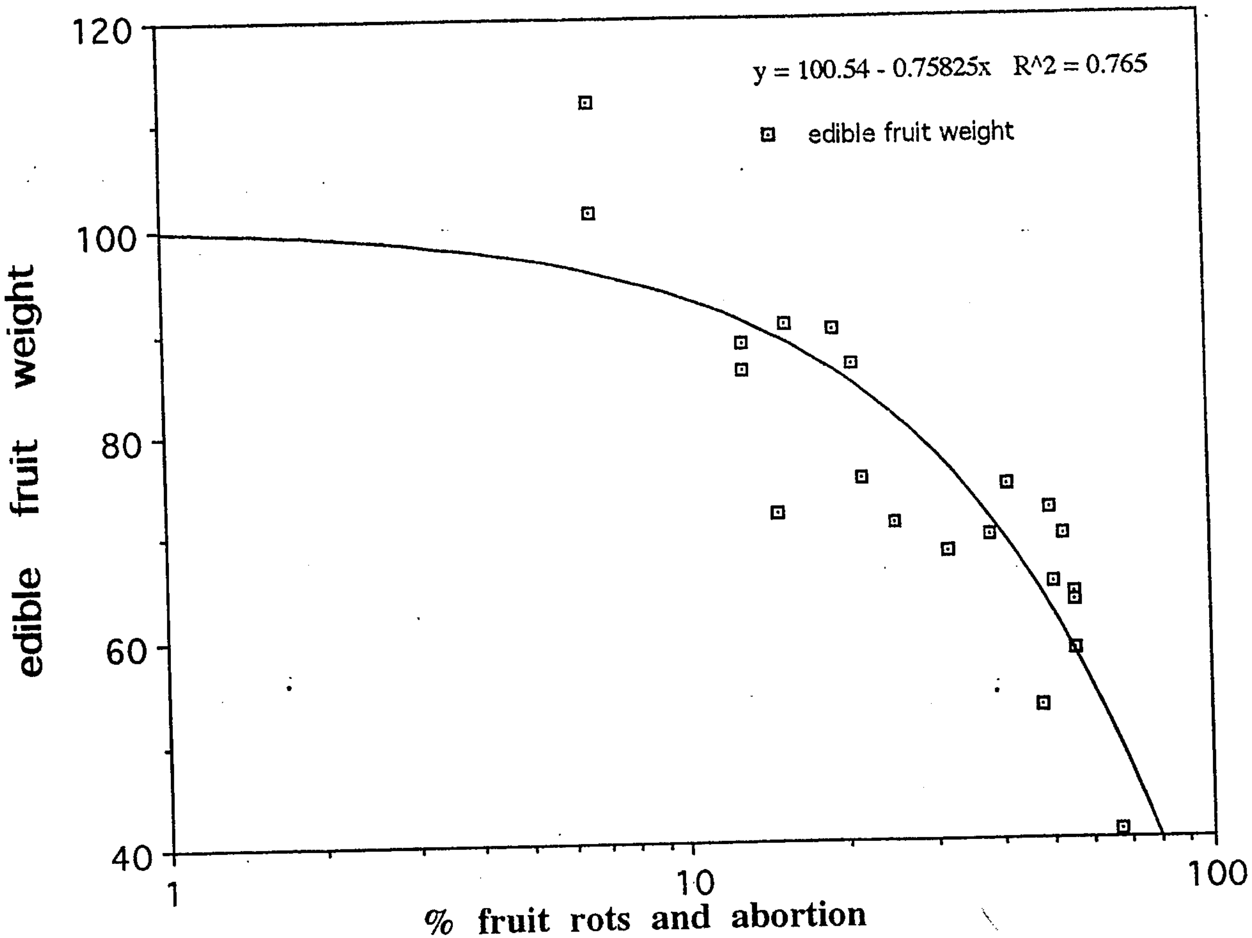

Fig 2: Relationship between severity of rots and the total edible fruit weight in okra infected with C. choanephora