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Phenotypic screening of cowpea (*Vigna unguiculata*) genotypes in response to parasitic weed *Alectra vogelii*

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Abstract. *Ugbaa M, Omoigui L, Bello L. 2020. Phenotypic screening of cowpea (*Vigna unguiculata*) genotypes in response to parasitic weed* Alectra vogelii. *Asian J Agric 4: 14-17.* Cowpea (*Vigna unguiculata* (L.) Walp) production is constrained by several abiotic and biotic factors. Among the biotic constraints, the parasitic flowering plant *Alectra vogelii* Benth. is one of the most formidable limitations in the dry savannas of West and Central Africa, a region that accounts for over 64% of world production. *Alectra* causes yield losses estimated between 41 and 100% in susceptible cultivars. Several control measures have been suggested for the control of the parasite. These include cultural practices, application of ethylene chemicals, and host plant resistance. Among these control measures, the use of resistance cultivars appears to be the most attractive option to the resource poor farmers in sub-Saharan Africa. This study was designed to revalidate and determine the reaction status of some improved and local cowpea genotypes to *Alectra vogelii*. Two screening experiments using pot culture technique and arranged in a Completely Randomized Design, were carried out in the screen house. Pot culture was comprised of sand and topsoil mixed in a 2:1 ratio and inoculum of 5000 *Alectra vogelii* seeds. To enhance effective parasite seed germination, the pot culture was watered twice a day for seven (7) days before planting of test cowpea genotypes. At 30 days after planting (DAP) *A. vogelii* shoots emerged from pots planted to susceptible cowpea genotypes, although some had delayed emergence up to 40 DAP. Susceptible cowpea showed leaf chlorosis, stunted growth, and partial leaf senescence. Some developed symptoms but *A. vogelii* shoots did not emerge. In both experiments, cowpea genotypes of B301, IT98K-573-1-1 and IT98K-205-8 were consistently resistant. They showed no attachment or emergence of the parasite. The absence of attachment on resistant cowpea genotypes suggests hyperactive mechanism of resistance to *A. vogelii*. This is a localized necrotic response that killed off attached parasite at the point of contact, a form of programmed cell death (PCD). This response strongly indicates dominant action in the resistance to *A. vogelii* in these genotypes. Resistant genotypes can be used as sources of resistance genes to develop improved resistant cowpea varieties.

Keywords: *Alectra*, cowpea, pot culture, resistance, screening

INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp) is an important food legume grown in tropical and subtropical regions of the world, primarily in sub-Saharan Africa. The largest areas under cultivation are in the savannas of West and Central Africa (WECA) where cowpea is an important crop. Cowpea is grown on about 12, 577, 845 million ha worldwide, with an annual grain production of about 7, 407, 924 tons (FAO 2017). Nigeria is the largest cowpea producer in the world and accounts for over 3.4 million tons of grain production from an estimated 3.7 million ha land area (FAO 2017). Nigeria is also the largest consumer of cowpea (FAO 2017) with consumption per capita is about 25-30 kg annually (FAO 2010).

Cowpea forms a major staple in the diet in Africa and Asian continents and is an important source of nutritious food and fodder in West Africa (Awe 2008). Cowpea contains 23-35% protein in its grains, about twice the protein content of most cereals, and is a rich source of B-vitamins, 62% soluble carbohydrates, small amounts of other nutrients, and relatively free from anti-nutritive factors (Brader 2002). Their amino acid also complements those of cereals (Asumugha 2002). Islam et al. (2006) emphasized that all parts of the plant that are used as food are nutritious, providing both protein and vitamins. Immature pods and peas are used as vegetables while several snacks and main dishes are prepared from the grains (Bittenbender et al. 1984). The use of cowpea haulms as fodder is attractive in mixed crop/livestock systems where both grain and fodder can be obtained from the same crop (Tarawali et al. 1997). The crop is also a valuable and dependable commodity that produces income for many smallholder farmers and traders in sub-Saharan Africa (Langyintuo 2003).

Despite the importance of cowpea, yield on farmers' field is still low and significant national production deficit exists due to a variety of abiotic and biotic stresses that constrain its production and productivity. These constraints include insect pests, diseases, parasitic weeds nematodes, drought, and heat (Omoigui et al. 2007). One of the serious biological constraints of increasing cowpea productivity in semi-arid regions of West and Central Africa (WECA) is the threat of being attacked by parasitic weeds *Strigages nerioides* and *Alectra vogelii* (Ehlers and Hall 1997). Fields infested by these parasitic weeds are difficult to clean due to considerable number of seeds produced and the dormancy mechanisms, which enable seeds to survive in the soil for several years. *A. vogelii* for instance, can produce as many as 600,000 seeds per plant (SP-IPM 2003). In an infested field, up to 75% of the crop damage occurs underground before shoots emerge above ground

(Emechebe and Singh 1989). According to Lagoke et al. (1991), several cultivated lands have been abandoned due to high infestations with these noxious parasitic weeds. Although attacks by *Alectra* are less severe than that of *Striga*, total yield loss is not uncommon in fields heavily infested. Yield loss resulting from *A. vogelii* attack ranges from 41-100% on susceptible cultivars (Lagoke et al. 1997). Development and use of cowpea varieties resistant to *A. vogelii* are one of the viable options to curb crop losses caused by this parasite. But equally important to prevent the spread of the parasite to new regions and be affordable for poor farmers and small-holder farming systems prevalent in Africa. Resistant varieties are particularly needed in the northern Guinea savannah zone of Nigeria where *A. vogelii* has become a major challenge in cowpea production (Mangani et al. 2008). Therefore, this study was designed to phenotype selected cowpea genotypes for their response to *A. vogelii* infestation.

MATERIALS AND METHODS

Planting materials/inoculum source

The plant materials used in this study were obtained from the International Institute of Tropical Agriculture (IITA), Kano Station, and the Molecular Biology Laboratory of the University of Agriculture, Makurdi, Nigeria. The eleven cowpea genotypes and their resistance/susceptibility phenotypes previously reported are presented in Table 1. IT03K-338-1 and IT98K-205-8 are advanced breeding lines developed at IITA earlier reported having resistance to *Striga* and *Alectra*. IT97K-499-35 and IT98K-573-1-1 are improved varieties developed also by IITA. B301 is a landrace from Botswana previously reported to be resistant to *Striga* and *Alectra*. Borno Brown, BOSADP brown, Golam white, Banjar, Kanannando Brown, and Yamisra are local cultivars grown in the Northeast, and previously reported to be susceptible to *Alectra*. Seeds of the parasitic weed *Alectra vogelii* Benth. used in this research were obtained from Borno State, through the Molecular Biology Laboratory of the University of Agriculture, Makurdi, Nigeria.

Experimental location and design

The plant materials were screened to re-validate their response to *Alectra vogelii*. Plants were grown with the pot-culture technique. Plastic pots with size of 15 cm in diameter and 14 cm in length containing about 1.8 kg of soil were used. Each pot was filled with a mixture of top-soil and sand in a ratio of 2:1 and inoculated with five thousand *Alectra vogelii* seeds. The experiment was carried out in the screen house of the Molecular Biology Laboratory, Department of Plant Breeding and Seed Science, University of Agriculture Makurdi (Lat 7.8°N, Long 8.6° E, and Alt 115 m above sea level). The experiment was laid out in Completely Randomized Design (CRD) with three replications. Each cowpea genotype was planted to two (2) pots in each replication. 5000 *Alectra* seeds were inoculated into each pot. Estimation of 5000 *Alectra* seeds was done as shown below.

Estimation of *Alectra* seeds

One thousand (1000) *Alectra* seeds were counted using a compound microscope (Brunel Microscope 0723219. X400 magnification) and weighed with a digital sensitive balance (Denver Instruments TP-3002). The weight of 5000 *Alectra* seeds was extrapolated as shown below.

Weight of 1000 seeds = 0.125g

Therefore, weight of 5000 seeds = 0.125g × 5 = 0.625g

Inoculated pot culture was conditioned prior to plant the test materials by watering for seven days to enhance germination of *Alectra* seeds.

Cultural practices

Three cowpea seeds were sown per pot at a depth of 2 cm and later thinned to two at 14 days after planting (DAP). One gram of NPK (15:15:15) fertilizer was applied per pot at 7 DAP at a rate of 15kg ai/ha. Weeds were hand-pulled while still tender.

Data collection and scoring

At about 30 DAP, pots were examined for *Alectra* shoot emergence. Thereafter, *Alectra* shoot count was done on a daily basis until the experiment finished at 65 DAP. At the end of experiment, the soil was washed off the plant root after submerging each pot in a 20-L bucket of water for about 5 min. The roots of each plant were gently separated and carefully freed from any remaining soil and examined for *Alectra* tubercle attachment. Plants allowing attachment and emergence of *Alectra* were classified as susceptible. Those without any attachment and free of infestation were categorized as resistant.

Table 1. Plant materials used and their resistance response to *Alectra*.

Cowpea cultivars/landraces/lines	Pedigree	Response to *Alectra*
IT03K-338-1	IT87D-941-1 × IT90K-59	R
IT97K-499-35	IT93K-596-9-12 × IT93K-2046-1	MR
IT98K-573-1-1	IT93K-596-9-12 × IT86D-880	R
IT98K-205-8	IT93K-596-9-12 × IT93-2046-1	R
B301	Landrace from Botswana	R
Borno Brown	Commercial cultivar in Borno	S
Golam white	Commercial cultivar in Borno	S
BOSADP Brown	Commercial cultivar in Borno	S
Banjar	Commercial cultivar in Borno	S
Kanannado Brown	Commercial cultivar in Borno	S
Yamisra	Commercial cultivar in Borno	S

Note: S: susceptible, R: resistance, MR: moderately resistant (Omoigui et al. (2012)

RESULTS AND DISCUSSION

At about 30 DAP, *A. vogelii* started emerging from the susceptible cowpea plants. While some genotypes had delayed emergence until 8 weeks. The susceptible cowpea plants showed leaf chlorosis, stunted growth, and partial leaf senescence. Some plants developed the symptom but *A. vogelii* did not emerge from the soil.

In both experiments, the cowpea cultivars: B301, IT98K-573-1-1, and IT98K-205-8 were consistently resistant to *A. vogelii* (Table 2). These cowpea lines showed no attachment or emergence of the parasite and were classified as resistance while the remaining cowpea lines either demonstrated attachment or emerged *Alectra* and were classified as susceptible (Figure 1).

It has been shown by researchers that studies on parasitic weeds and their interactions with host plants can be successfully carried out in screen house using pot trials. For example, screening for varietal resistance, conditioning trials, nutritional interrelationships of host and parasite, effect of herbicides, germination stimulants, and other aspects have been approached effectively using pot and buried seed studies (Sand et al. 1990). One of the major advantages of pot trials is that they can be carried out year-round, while field experimentation is limited to one cycle per year. In cowpea, pot screening using pot culture inoculated with parasite seeds has been successfully employed in screening for varietal resistance to *Striga* (Omoigui et al. 2012) and *Alectra* (Kureh et al. 1994; Magani et al. 2008).

In the present study, pot screening of cowpea under screen house condition using pot mixture of topsoil and sand (2:1 vol/vol) with inoculum of 5000 *Alectra vogelii* seeds proved a reliable method for assaying resistance to the parasitic weed. Magani et al. (2008) who reported similar success also stated that chances of success improve with increased density of parasite seeds in pot mixture.

This study showed that cowpea genotypes of B301, IT98K-573-1-1, and IT98K-205-8 were resistant to *Alectra vogelii* as they supported no attachment of the parasite tubercle across two experiments. This result confirms the work of previous researchers who also reported resistance of B301 (Atokple et al. 1995; Omoigui et al. 2012) and IT98K-573-1-1 and IT98K-205-8 (Omoigui et al. 2012) to *Alectra vogelii.*

Table 2. Response of cowpea cultivars to *Alectra vogelii* averaged across two (2) experiments

Cowpea genotype	No. of *Alectra* count/plant	Response
Borno Brown	11	S
BOSADP Brown	12	S
Golam white	8	S
Banjar	12	S
Kanannado brown	10	S
Yamisra	11	S
IT03K-338-1	1	MR
B301	0	R
IT97K-499-35	1	MR
IT98K-573-1-1	0	R
IT98K 205-8	0	R

Note: R: Resistant, S: Susceptible, MR: Moderately resistant

Figure 1. Chlorosis and partial leaf senescence of susceptible cowpea plant due to Alectra infestation

Figure 2. A. *Alectra vogelii* infected plants showing numerous hairy attachments. B. *Alectra vogelii* shoots and attachments on susceptible plant exposed after washing away the pot culture.

The absence of attachment on the cowpea genotype suggests an active mechanism of resistance that prevented successful attachment of parasite. This response seems to be a localized necrotic response that killed off attached parasites at the point of contact (hyperactive response, HR), a form of programmed cell death (PCD) in the host (Zhang and Zaitlin 2008). This implies that resistant cowpea cultivars did not allow the parasite haustorium to penetrate and establish connection with its vascular system to derive nutrients for its survival. This response strongly indicates dominance interaction in the resistance to *Alectra* in these genotypes. Berner et al. (1993) stated that the mechanism of resistance of cowpea to *Alectra* appears to be post-attachment mortality of the parasite with no reduction in parasite seed germination. Borg (1999) also considers that low induction of germination of parasite seeds, which seems to be the resistance response to parasitic plants in some other crops, plays a little role in resistance of *Orobanche* in legumes. In *Nicotiana langsdorffii*, one of two species of *Nicotiana* known to express an incompatible interaction with the oomycete *Perono sporatabacina* (the causal agent of tobacco blue mold disease), Zhang and Zaitlin (2008) showed that incompatibility was due to the hypersensitive response (HR), and plants expressing the HR are resistant to *P. tabacina* at all stages of growth. Resistance was due to a single dominant gene in *N. langsdorffii* accession S-4-4 named *NlRPT*.

The resistance of B301, IT98K-573-1-1, and IT98K-205-8 to *Alectra vogelii* also implies that these genotypes can be cultivated in Borno State where the *Alectra vogelii* seeds used in this work were obtained. Omoigui et al. (2012), who conducted a two-year field screening in this region, stated that even though some of the genotypes, especially B301, were low yielding the genotypes would be useful sources of cowpea *Alectra* resistance genes for incorporation into high yielding and adapted cultivars for host-specific race in the region. The use of resistant cultivar integrated with other cultural practices appears to be the only viable management option for effective control of *Alectra* infestation on cowpea.

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