Influence of nitrogen fertilizer on resistance to fall armyworm (Lepidoptera: Noctuidae) in tropical Honduran landrace sorghum[1]

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Abstract. Host plant resistance studies in the Honduran National Sorghum Breeding Program in cooperation with the United States Agency for International Development, Sorghum and Millet Collaborative Research Support Program (INTSORMIL), and the Panamerican School of Agriculture, Zamorano, Honduras have identified landrace sorghums with some level of antibiosis resistance to fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith). As the nutritional condition of a plant can influence insect growth, survival and fecundity, and it's ability to respond to insect attack, the influence of plant fertilizer on this resistance mechanism in the landrace sorghums was evaluated. San Bernando III, a FAW resistant tropical Honduran landrace sorghum cultivar, and Cacho de Chivo-169, a FAW susceptible sorghum cultivar from Guatemala, were tested in a laboratory biossay for whorl-feeding resistance to FAW. Leaf material from plants fertilized with low or high levels of nitrogen were fed to the larvae. No significant differences were observed in development or mortality of FAW larvae or pupae, female pupal weight, fecundity, net multiplication rate, or intrinsic rate of natural increase when larvae were fed on resistant or susceptible sorghum plant material collected from either of the fertilizer treatments. This study provides information that is useful to entomologists and others in regulating plant nutrition in insect-plant resistance research and suggests that increased nitrogen fertilization may have limited influence on the FAW host plant resistance mechanism expressed as antibiosis in the Honduran landrace sorghums. The small variety nitrogen effects recorded may be meaningful, but this observation needs further critical evaluation.

Key words: Insect development, plant nutrient, *Spodoptera frugiperda*, *Sorghum bicolor*.

Resumen. Estudios de resistencia de plantas hospederas en el programa Hondureño de mejoramiento de sorgo, en cooperación con la Agencia Internacional para el Desarrollo (USAID), el programa colaborativo para la investigación de sorgo y mijo (INTSORMIL) y la Escuela Agrícola Panamericana, Zamorano, Honduras, han identificado variedades criollas de sorgo con cierto nivel de antibiosis al gusano cogollero, *Spodoptera frugiperda* (J.E. Smith). Así como el estado nutricional de la planta puede influir en la habilidad para resistir al ataque de insectos, también afecta el crecimiento, sobrevivencia y fecundidad del insecto plaga. La influencia de fertilizantes en este mecanismo de resistencia en los sorgos criollos fue evaluado en este estudio. San Bernardo III, una variedad hondureña criolla resistente al cogollero, y Cacho de Chivo-169, una variedad guatemalteca susceptible al cogollero, fueron probados en bioensayos a nivel de laboratorio para analizar la resistencia a nivel de la etapa de cogollo de la planta. Las larvas fueron alimentadas con hojas de plantas con bajo y alto nivel de nitrógeno y no se encontró diferencia en el desarrollo o mortalidad de las larvas o pupas, peso de las pupas hembras, fecundidad, tasa de multiplicación neta o tasa de incremento natural de cogollero, cuando las larvas fueron alimentadas con sorgos susceptibles o resistentes en ambos tratamientos de fertilización. Este estudio provee información útil para entomólogos y otros respecto a la regulación del estado nutricional de las plantas en estudios de resistencia de plantas y sugiere que el incrementar el nitrógeno en las plantas tiene poca influencia en los mecanismos de resistencia de las plantas contra cogollero, expresados como antibiosis en las variedades criollas de Honduras. Los pocos efectos encontrados en variedad nitrógeno podrían ser significante, pero necesita más estudios.

Palabras claves: Desarrollo de insectos, nutrientes de insectos, *Spodoptera frugiperda*, *Sorghum bicolor*

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INTRODUCTION

Much has been done to identify insect resistance in crop plants such as cotton, maize and sorghum (Painter 1951, Maxwell and Jennings 1980, Meckenstock 1988, Lin *et al.* 1990 and Meckenstock *et al.* 1991). Efforts have been made to incorporate host plant resistance into integrated pest management strategies because insecticides and other insect control tactics have often been only moderately effective or have failed under high insect pressure (Wiseman and Davis 1979, Wiseman *et al.* 1981, Andrews 1988).

The chemical compounds procyanidin, dhurrin and maysin have been identified to be involved in host plant resistance to insects (Reese 1981, Dreyer *et al.* 1981, Dreyer and Campbell 1983). The relationship of these chemicals with plant resistance to insect pests has been investigated. Guiragossian and Mihm (1985) reported that there was no relationship between HCN content of sorghum and resistance to the fall armyworm. Maysin and 6-MBOA toxins in corn plants were found to have no significant influence on leaf feeding resistance to first instar fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith), because the chemicals were in low concentrations in the whorl (Hedin *et al.* 1990). However, Wiseman *et al.* (1973a) reported that corn plants treated with fertilizer containing nitrogen were more susceptible to FAW than untreated plants. Diawara *et al.* (1992) observed that FAW resistance in sorghum was positively correlated with higher concentrations of total leaf nitrogen. Extreme expression of non-preference, antibiosis, and tolerance to FAW were observed in "Gahi" millet, *Pennisetum typhoides* (Burn), when plants were treated with combinations of NPK fertilizer in greenhouse tests (Leuck 1972). Sorghum plants with dark green leaves, an indication of high nitrogen content, were observed to be preferred over plants with light green leaves by FAW moths for oviposition (Pitre 1979).

Meckenstock *et al.* (1991) reported various levels of antibiosis resistance in Honduran landrace sorghums to FAW larvae. These sorghums generally exhibited intermediate levels of resistance, having a negative influence on developmental times, pupal weight, and fecundity. As nutritional factors are recognized to influence the ability of plants to withstand insect injury, the present study evaluated the effect of nitrogen concentrations on FAW developmental rates when larvae were fed a resistant landrace sorghum cultivar or a susceptible sorghum cultivar reported by Meckenstock *et al.* (1991) to respond differently to FAW attack.

MATERIALS AND METHODS

A laboratory bioassay was conducted using sorghum leaf material obtained from small field plots to determine the effect of nitrogen concentration on sorghum resistance to FAW. The bioassay was conducted in the Department of Plant Protection at the Panamerican School of Agriculture, El Zamorano, Honduras.

San Bernardo III, a resistant tropical Honduran landrace sorghum cultivar, and Cacho de Chivo-169, a susceptible sorghum cultivar from Guatemala, were planted by hand in 5 m rows, spaced 80 cm apart in 1988. Plants were thinned to a density of 8-10 plants/m of row and weeds were removed manually. Treatments were arranged in a split plot design having fertilizer treatments (nitrogen level) as main plots and cultivars as subplots. Fertilizer (18-46-0, NPK) was incorporated into the soil prior to planting. The fertilizer treatment levels included a low concentration (150 kg/ha) and a high concentration (250 kg/ha) at planting and urea (46% N) was applied 20 days after planting at 50 kg/ha and 228 kg/ha in the low and high treatments, respectively. Cultural practices for sorghum production in the region were used.

Fall armyworm neonates were obtained from a colony originally collected from sorghum at El Conchal and La Coyota in southern Honduras. They were maintained in the laboratory at 27± 2° C and 14:10 L:D photoperiod. Twenty-five days after germination, sorghum leaf material was cut from the treatment plots, wrapped in moist paper, placed in paper bags, labeled accordingly, and taken immediately to the laboratory in a refrigerated box to be used in feeding and developmental tests. The basal one-half of the youngest whorl leaves were selected as bioassay food material for larvae. Leaves were first washed with distilled water, then with a 5% hypochlorite solution, and finally with distilled water. The leaf material was placed in 29.6 ml plastic cups containing 5 mm of agar to provide moisture inside the cup. Also, the cup contained 0.28 g of corncob-grit treated with 0.03% griseoflavin, 0.04% phaltan, and 0.03% tetracycline, which was used as an antibiotic to reduce contamination by microorganisms. The plant material was changed daily until FAW larvae reached the pupal stage.

The distal one-half of the source sorghum leaves was used to analyze foliage nitrogen using methods described in Methods of Analysis - A.O.A.C. (1970). The variables measured for antibiosis were larval plus pupal mortality (Pm), developmental times for larvae and larvae plus pupae (Tm), pupal weight (wp), fecundity (F), net multiplication rate (R_o), and the intrinsic rate of natural increase (r).

Fecundity was estimated using the formula reported by Leuck and Perkins (1972):

$$F = 5.33\,wp - 423.23 \qquad [1]$$

where wp = female pupal weight (mg).

The net multiplication rate (R_o), a measure of the rate of increase of a population per generation rather than per unit of time, was calculated using fecundity and survivorship (Birch, 1948):

$$R_o = F \times S \qquad [2]$$

where S = 1 - Pm, with Pm = total percent mortality/100.

The intrinsic rate of natural increase (r_m) was estimated according to Birch (1948):

$$r_m = [\mathrm{Log}_e (R_o)]/ T \qquad [3]$$

where T = generation time (days) measured from egg hatch to adult emergence.

Lower values of r_m represent a lower rate of increase in the FAW population (i.e., reduced fitness) which may be attributed to diet (i.e., antibiosis of the plant). The r_m provides a basis for measuring relative fitness because it is a measure of the rate of growth of a population that incorporates the effects of all factors affecting fitness.

Treatments were arranged in a randomized complete block design using six larvae per treatment and six replications. Cups were placed in plastic holding trays corresponding to the experimental design established in the field. Data were analyzed by analysis of variance (ANOVA) using the GLM procedures of SAS (SAS Institute 1985). The treatment means were separated with Duncan's multiple range test (Duncan 1955) at the 0.05 level. Correlation analyses between the parameters immature development or fecundity and total nitrogen concentration (TNC) were performed using a Pearson correlation analysis (SAS Institute 1985).

RESULTS AND DISCUSSION

When FAW larvae were fed leaf tissue from the basal one-half of whorl leaves, no significant differences were observed among treatments in percent larval or pupal mortality (Table 1). However, fall armyworms fed San Bernardo III experienced a small increase in mortality compared with the susceptible Cacho de Chivo-169, whether fertilized or not. There were no differences in larval developmental times between fertilizer treatments; however, larval plus pupal developmental times were shorter for insects fed San Bernardo III, with both low and high nitrogen concentrations, than for insects fed the susceptible cultivar with the two nitrogen concentration levels (Table 1). No significant differences were observed among treatments for female pupal weight, fecundity, net multiplication rate, or intrinsic rate of natural increase.

Wiseman *et al.* (1973a) and Wiseman *et al.* (1973b) reported that corn plants treated with nitrogen fertilizer were more susceptible to FAW than corn that received no fertilizer, and there was an extremely detrimental effect on the FAW population when plants received no nitrogen, but were treated with zinc. These responses were

Treatment		% Larval + pupal mortality	Developmental time, (d)		Female pupal wt,	Fecundity	Net multiplication rate	Intrinsic rate of Increase
Cultivar	N/ha (kg)	(Pm)	Larva	Larva + Pupa Tm	(mg)	(F)	(R_o)	(r_m)
San Bernardo III	50	44.5 a✤	15.5 a	26.1 a	213.5 a	714.7 a	396.6 a	0.229 a
San Bernardo III	150	50.1 a	15.5 a	26.3 a	206.3 a	676.4 a	337.5 a	0.221 a
Cacho de Chivo	50	41.8 a	16.2 a	26.7 b	210.2 a	697.1 a	405.7 a	0.224 a
Cacho de Chivo	150	36.2 a	15.3 a	26.7 b	211.6 a	704.5 a	449.4 a	0.228 a

✤Means within a column not followed by the same letter are significantly different at the 0.05 level as determined by protected least significant analysis [Snedecor and Cochran, 1980] (SAS Institute, 1985). N=36 larvae/treatment) not observed in the present study, but small differences between cultivars, although not significant, indicated the general tendency for reduced fitness in FAW when fed San Bernardo III, the resistant cultivar, receiving the high level of nitrogen fertilizer. These data suggest that cultivar and nitrogen (fertilizer level) could have a synergistic effect on the overall fitness of the FAW. Additional studies would be required to adequately define this relationship.

There were no significant correlations between parameters of FAW development and total nitrogen concentration (Table 2). The amount of nitrogen fertilizer applied in a field depends on the requirements of the crop plants in order to reach maximum yield potential. Because the amount of nitrogen selected for these studies was very high, the results should not be interpreted for field application. The levels of nitrogen applied to the susceptible and resistance sorghum cultivars in this study had no consistent significant influence on the development of FAW. San Bernardo III, the landrace sorghum with some resistance to FAW and fertilized with a high level of nitrogen, had limited negative effect, although not significant, on the fitness of the FAW. Correlation analyses (Table 2) for parameters of insect development and concentrations of total plant nitrogen revealed no significant correlations. Definitive studies under controlled conditions would be required in order to evaluate the specific levels of nitrogen or other plant physiological components on FAW developmental rates, as well as the effect of other specific nutrients and nutritional indices on the fitness of FAW. Understanding the relationship between plant nutrient status and the plants ability to respond to fall armyworm feeding can assist plant breeders and entomologists in recognizing levels of insect resistance in the host plant and in defining studies required to identify insect resistance mechanisms in sorghum. This study complements other investigations in the Honduran National Sorghum Breeding Program to identify sorghum with resistance to insect pests.

Table 2. Correlations between developmental variables of fall armyworm reared on whorl leaves of two sorghum cultivars and concentrations of total nitrogen (TNC)[a].

Developmental variables	Correlation coefficients r^2 (P)
Larva + pupa mortality	-0.017 (0.96)[b]
Generation time	0.187 (0.65)
Pupal weight	-0.440 (0.27)
Fecundity	-0.441 (0.27)
Net developmental rate	-0.420 (0.30)
Intrinsic rate of increase	-0.237 (0.51)

[a] grams/kg dry matter

[b] Coefficients in table are followed by P values (SAS Institute, 1985).

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