Crop Protection 29 (2010) 34–38

Contents lists available at ScienceDirect

Crop Protection

journal homepage: www.elsevier.com/locate/cropro

Manihot flabellifolia Pohl, wild source of resistance to the whitefly *Aleurotrachelus socialis* Bondar (Hemiptera: Aleyrodidae)

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ARTICLE INFO

Article history:
Received 11 April 2009
Received in revised form
24 August 2009
Accepted 30 August 2009

Keywords:
Host plant resistance
Wild species
Manihot esculenta
Aleurotrachelus socialis

ABSTRACT

Aleurotrachelus socialis is one of the most important pests of cassava (*Manihot esculenta* Crantz) in the Neotropics. In Colombia, high whitefly populations can reduce crop yields by 79%; and although the farmers intensify the use of insecticides, this practice is highly contaminating, costly and leads to the development of resistance in the insect. An alternative for managing whitefly populations is to develop genetically resistant varieties. Wild parents of *Manihot* are a useful source of genes against pests for the cultivated species of cassava. Based on prior research that showed the existence of moderate-to-high levels of resistance to *A. socialis* in *Manihot flabellifolia*, a wild species of cassava, this study was proposed to characterize this new source of resistance, evaluating the biology and demographics of *A. socialis* on eight accessions of *M. flabellifolia*, a susceptible check (CMC-40) and a resistant (MEcu72) check. The averages of *A. socialis* longevity and fecundity on the accessions were not significantly different to MEcu72, but different from CMC-40 ($P < 0.05$). Development time was not significantly different, ranging from 35–40 days on accessions and MEcu72 and 33.5 days on CMC-40 ($P < 0.05$). In contrast, the population growth rate (r_m) was significantly lower on the *M. flabellifolia* accessions, with Fla 61 standing out with a growth rate 98 and 99% less than that obtained on MEcu72 and CMC-40, respectively. Once the resistant levels have been identified to *A. socialis* on the *M. flabellifolia* accessions, interspecific crosses of *M. esculenta* subsp. *M. flabellifolia* and backcross programs could be developed to incorporate the desirable characteristics from the wild relatives into elite progenitors of *M. esculenta*.

1. Introduction

Cassava (*Manihot esculenta* Crantz) is the only one of the 98 *Manihot* species described that has economic importance as a crop. Since its introduction from South America to the tropical countries in Africa and Asia, it has contributed to the food security and improvement of living standards for more than a billion people including processors and merchants around the world (Balagopalan, 2002; Hillocks, 2002). Approximately 65% of world cassava production is concentrated in humid and subhumid lowland agroecosystems of Latin America, Africa and Asia (Henry and Gottret, 1995); however, the most significant damage caused by arthropod pests and diseases occurs from 1000–1400 masl (meters above sea level), especially where there are extended dry periods of more than three months (Bellotti et al., 1999). In the Neotropics the prolonged growth cycle (up to 2 years) and tolerance to drought are agronomic traits that favor the presence of arthropod pests in cassava. South America, which is its center of origin, is also noted for the great diversity of arthropod pests that feed on this crop (Bellotti et al., 1994).

Among the more than 200 species of arthropod pests that can cause moderate-to-severe losses to the crop (Bellotti, 2002), the whiteflies (Hemiptera: Aleyrodidae) stand out. This is due to their proven effectiveness as vectors of viruses, together with the damage their direct feeding and excretion of honeydew can cause (Oliveira et al., 2001; Brown et al., 1995). Among the whitefly species of greatest economic importance are *Aleurotrachelus socialis* (Bondar), *Trialeurodes variabilis* (Quaintance), *Bemisia tuberculata* (Bondar), *Bemisia tabaci* (Gennadius) and *Aleurothrixus aepim* (Goeldi) (Bellotti et al., 1999). To date, *A. socialis* appears to be specific to cassava and predominant in Colombia, Venezuela and Ecuador; while high populations of *A. aepim* are found in northeastern Brazil, where they attack cassava as well as other host plants.

In these countries as well as other neotropical countries, the control measures in cassava are based on the use of costly

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doi:10.1016/j.cropro.2009.08.014

insecticides. In addition, the fact that cassava is grown in marginal soils, poor in nutrients, exposes the crop to stress, increasing its susceptibility to attack from these pests and making its production at all scales and economic levels unprofitable (Calatayud et al., 2002).

Although host plant resistance represents a practical, rational alternative that can keep whitefly populations low, reduce yield losses and is a valuable tool in an integrated management program, it is little known and/or promoted. This technique dates back to the work of Vavilov (1940), who first called attention to the potential of wild parents as a source of genes for crop improvement; e.g. the wild species of *Manihot* are known sources of genes for resistance to cassava pests and diseases, as well as to postharvest physiological deterioration (PPD). Recent studies have identified the only known source of delay in PPD in an interspecific hybrid between *M. esculenta* and *Manihot walkerae* (Croizat). In addition, some findings have provided the bases for believing that *Manihot glaziovii* Muell. Arg., *Manihot catingae* Ule. and *Manihot carthaginensis* (Jacq.) Mull. Arg., represent potential sources of genes for tolerance to drought. Likewise, studies of damage and population levels showed from moderate-to-high levels of resistance in *Manihot flabellifolia* (Burbano et al., 2003) for the cassava green mite *Mononychellus tanajoa* (Bondar), the mealybug *Phenacoccus herreni* (Cox-Williams), and the whitefly *A. socialis*. These results point to the need for conducting studies to determine the total impact of resistance on the demographics of the target species, in this case *A. socialis*.

The purpose of this study was to quantify the effects of the levels of resistance in eight accessions of *M. flabellifolia* on the life history and demographic parameters of *A. socialis*, the most important whitefly that affects cassava in northern South America.

2. Materials and methods

This study was conducted during 2007 in glasshouses of the International Center of Tropical Agriculture (CIAT), located at 1000 masl, in the municipality of Palmira, Cauca Valley Province, Colombia (27 ± 2°C, 60–70% RH).

2.1. Plants and whiteflies

In the tissue culture lab of the Cassava Genetics Program at CIAT, 20 seedlings of each *M. flabellifolia* (Pohl) Cifferi accession (Fla 15, Fla 19, Fla 21, Fla 25, Fla 33, Fla 52, Fla 61, Fla 75), the genotype of *M. esculenta* CMC-40 (susceptible check) and MEcu72 (resistant check) were multiplied *in vitro*. Then the accessions and genotypes were planted in sterile soil in 1-kg plastic pots and kept in the glasshouse at 30 ± 2°C and 50–60% RH. For the trials of longevity, fecundity and demographic parameters, ten 40-day-old plants of each accession, CMC-40 and MEcu72 were selected. The remaining plants were used in the trials to determine preference for oviposition. For both experiments the *A. socialis* adults were taken from a colony established at CIAT in 1992 (Bellotti and Arias, 2001).

2.2. Longevity and fecundity

Forty pairs of recently emerged *A. socialis* adults coming from CMC-40, were placed in individual clip cages (diam. = 2.5 cm; height = 2 cm) using a manual aspirator (impregnated with wax from the wings of adult whiteflies to reduce mortality that could result from their handling). They were placed on the underside of the leaves of the *M. flabellifolia* accessions (Fla 15, Fla 19, Fla 21, Fla 25, Fla 33, Fla 52, Fla 61, Fla 75), CMC-40 and MEcu72. Every 48 h the adults were moved to a new area of the leaf until the natural death of the females. Fecundity was estimated as the average number of eggs laid by the female every 48 h; and longevity as the maximum time (days) that a female lived.

2.3. Development time, survival rate and proportion of females

Fifty adults (males and females), two days of age from CMC-40 plants, were placed on the underside of the leaves of *M. flabellifolia* accessions (Fla 15, Fla 19, Fla 21, Fla 25, Fla 33, Fla 52, Fla 61, Fla 75), CMC-40 and MEcu72, using a manual aspirator in clip cages. After 6 h the adults were removed, and 200 eggs were selected at random and the rest were removed with the help of needle and a fine brush. The egg-adult development time, the survival rate of the immature stages, and the sex ratio were recorded.

2.4. Demographic parameters

Life tables were generated by combining data on the development and reproduction time (l_x-m_x), calculating the demographic parameters defined by Price (1975) such as (1) net reproduction rate (R_o) or average number of descendents that one female had in one generation; (2) generation time (T), equivalent to the period between the emergence of the parents and that of the progeny; and (3) intrinsic growth rate of the population (r_m), estimated using the equation (Carey, 1993):

$$\sum \exp(-r_m X) l_x m_x = 1$$

where: X = age of the female; l_x, = specific age of survival; m_x, = the proportion of females from the progeny of one female age x.

In calculating the values for r_m, the corrected age ($X + 0.5$) was used. The equation ln $2/r_m$ was used to estimate the days required for the population to double in number (Carey, 1993).

2.5. Free choice preference for oviposition

Antixenosis was studied in free choice trials with respect to oviposition under glasshouse conditions. The plants from the *in vitro* crop were established initially in bags and later transplanted to plastic pots. For the evaluations three leaves were selected, enumerated in descending order, starting with the first open leaf. Plants from *M. flabellifolia* accessions (Fla 15, Fla 19, Fla 21, Fla 25, Fla 33, Fla 52, Fla 61, Fla 75), the susceptible genotype CMC-40 and resistant genotype MEcu72 were introduced into five cages made of cotton mesh and wood (1 m^3) and randomly distributed at an equal height in a circle. The plants were infested with 500 *A. socialis* adults, 24 h postemergence. The insects were released onto a plastic tray covered with black cardboard in the center of the cage, using a manual aspirator. The whiteflies were removed 48 h later and collected from each host plant, breaking the data down by leaf number. Oviposition was estimated as the average number of eggs laid per female on the three leaves of each accession, susceptible and resistant genotype. This was done with the aid of a stereo microscope.

2.6. Statistical analyses

The Kaplan-Meier statistical package, which includes the Cox-Mantel, Peto-Wilcoxon and Gehan-Wilcoxon statistical tests, was used to compare the survival of females on the host species based on mean survival times (Lee, 1992) (Statistix 8.0). Differences among the mean values for longevity, fecundity, oviposition rate, development time and antixenosis for oviposition were analyzed using one-way ANOVA. Student-Newman-Keuls was used for the multiple-comparison tests. The survival rates (the immature stage values) were compared using the chi square test (SAS Institute,