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The effects of nitrogen rate and the ratio of $NO_3^-:NH_4^+$ on *Bemisia tabaci* populations in hydroponic tomato crops

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ABSTRACT

Nitrogen fertilization is one of the factors that influences *Bemisia tabaci* (Gennadius) population density. The aim of this study was to determine the effects of three N application rates (75, 205 and 335 mg/l) and three ratios of $NO_3^-:NH_4^+$ ions (92:8, 75:25 and 55:45) in standard nutrient solution (205 mg/l N) on the population density of *B. tabaci*. The experiments were conducted on spring-summer hydroponic crops of tomato. The effect of plant stratum on the whitefly population was also determined. The aggregation of *B. tabaci* adults as well as their oviposition rate was higher at 205 and 335 mg/l N than on plants grown at 75 mg/l N. By the end of the experiment (60 d after infestation), the number of nymphs on plants at 205 mg/l N was higher than on plants at 75 mg/l N. The number of pupae was lowest on plants supplied with 75 mg/l N. An increase in NH_4^+ percentage in standard nutrient solution (from 25% to 45% of the total N) reduced adult population density and oviposition rate. The density of nymphs and pupae, at 60 d after infestation, was lower on the tomato plants grown at 75:25 and 55:45 $NO_3^-:NH_4^+$ ratios compared to the 92:8 ratio. The 75:25 and 55:45 $NO_3^-:NH_4^+$ ratios resulted in a higher incidence of blossom-end rot of tomato fruit, with a lower incidence of disorder at 75:25 than at the 55:45 ratio. Plant stratum influenced adult whitefly distribution in two years of the study. Middle stratum leaves were more attractive to adults in both years. The results demonstrate the effects of N fertilization (N rate and the ratio of $NO_3^-:NH_4^+$) and plant stratum on *B. tabaci* population density.

1. Introduction

Tobacco or sweetpotato whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) is one of the most important economic plant pests worldwide, particularly in greenhouse horticultural crop production. This cosmopolitan species is extremely polyphagous and has been found on more than 500 different plant species, often herbaceous annual plants (Mound and Halsey, 1978, 340 pp.). Adults and nymphs feed on phloem sap and excrete honeydew that supports the growth of sooty moulds and reduces photosynthetic rate and product quality of the plants infested. *B. tabaci* was first recorded in Croatia in 2000 (Žanić et al., 2001) and today the Q biotype is predominant and widespread in the coastal area of Croatia (Žanić et al., 2005; Škaljac et al., 2010).

Plant nitrogen (N) content, as a consequence of N fertilization, is one of the factors influencing whitefly infestations on agricultural crops. According to Tripp et al. (1992), the number of greenhouse whitefly [*Trialeurodes vaporariorum* (Westwood)] adults is positively correlated with percentage of N in tomato leaves. Jauset et al. (1998) reported that the number of *T. vaporariorum* adults and the number of eggs oviposited, was higher on leaves of tomato plants fertilized with high rates of N. In a choice test, the number of *T. vaporariorum* eggs oviposited and the number of adults emerged was higher on chrysanthemum plants treated with high concentrations of N fertilizer (Bentz and Larew, 1992). In choice bioassays on gerbera, the number of eggs and nymphs of *T. vaporariorum* increased significantly as the supply of N increased (Ortega-Arenas et al., 2006). High N fertilization of tomato also increased the egg survival rate, size of pupal exuviae and female tibial length of *T. vaporariorum* (Jauset et al., 2000).

The number of adult and immature individuals of *B. tabaci* biotype B (*Bemisia argentifolii* Bellows and Perring) during population peaks increased with increasing amounts of applied N on cotton under field conditions (Bi et al., 2001). On the contrary, in a field crop of tomato, Leite et al. (2004) found no effect of leaf N on the *B. tabaci* population. Moreover, plant N content had no effect on the developmental time of *T. vaporariorum* (Jauset et al., 2000) and *B. tabaci* (Watson et al., 1994).

The effect of applied N rate on pest populations has been the subject of scientific interest in numerous crops (Bentz and Larew, 1992; Jauset et al., 1998; El-Rafie, 1999; Berlinger and Wermelinger,

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2001; Ortega-Arenas et al., 2006), whereas the effect of the ratio of NO_3^-:NH_4^+ applied has received less attention. In hydroponic cultivation of tomato the standard nutrient solution has a ratio of NO_3^-:NH_4^+ ions of approximately 92:8. According to Siddiqi et al. (2002), percentage of total N had no negative effect on early vegetative growth of tomato, but in a long-term experiment, a higher proportion of NH_4^+ up to 50% of total N resulted in a high incidence of blossom-end rot (BER) disease.

The aim of our study was to determine the influence of three nitrogen rates in nutrient solution and three ratios of NO_3^-:NH_4^+ ions on the population density of *B. tabaci* Q biotype, on tomato, *Lycopersicon esculentum* (Miller), cv. Belle under hydroponic cultivation. We have also studied the effect of plant stratum on pest populations.

2. Material and methods

Two experiments were conducted using tomato cv. Belle, in a hydroponic system during spring-summer of 2008 and 2009. We used a greenhouse at the Institute for Adriatic Crops at Split (43°30′17.17″N, 16°29′49.71″E) in the Mediterranean area of Croatia. The greenhouse roof was 4.5 m high. The mean air temperature during the experiment was 27.3 °C in 2008, and 26.1 °C in 2009. Automatic ventilation was activated at 22 °C.

2.1. *Bemisia tabaci population*

A population of *B. tabaci*, Q biotype, was obtained from a commercial greenhouse in Kaštel Stari (43°32′46.84"N, 16°17′32.07"E), and reared on poinsettia plants (*Euphorbia pulcherrima* Willd, cv. Christmas Star) in a growth chamber (26 °C, 60% RH and 14:10 h light: dark photoperiod). This population was used to infest peppermint plants (*Mentha piperita* L.) in a low tunnel covered by non-woven polypropylene material (Agryl P30, Sodoca) in the greenhouse, under conditions favourable for pest development. Infested peppermint plants were used as sources for the tomato infestation.

2.2. *Tomato cultivation and experiments*

Tomato seeds were sown in an organic substrate (Brill Type 4, Germany) and planted into rockwool cubes (7.5 × 7.5 × 6.5 cm, KRAN-IZOL s.r.o., Check Republic) 25 d after sowing. Seedlings with seven fully expanded leaves and approximately 30 cm high were then transplanted to rockwool slabs (7.5 × 20 × 100 cm). Transplanting was done on the 30 April 2008 and 29 April 2009 at a density of three plants per slab. Tomato plants were fertigated by a drip irrigation system using the standard nutrient solution for cultivation of tomato in rockwool (Sonneveld and De Kreij, 1999).

The experiments on the effects of N fertilization and plant stratum on *B. tabaci* population density included three nitrogen treatments and three strata, and were conducted in a completely randomized experimental design with four replications. Each treatment comprised 24 plants, in four replications of six plants each.

In both years, the pest population densities were assessed on the four innermost plants of each experimental plot, the two marginal plants being excluded from the assessments. In addition, 15 leaves were numbered, started from the top (first fully expanded leaf) to the bottom on each of the four measured plants per plot. The plants were divided into three strata: upper (U) (leaves number 1, 2, 3, 4, and 5); middle (M) (leaves number 6, 7, 8, 9, and 10); and lower (L) (leaves number 11, 12, 13, 14, and 15) part of the plant canopy.

The stem height and diameter, number of leaves per plant, and Hydro N-tester chlorophyll meter (HNT) (Minolta, Japan) measurements were taken in both years. The tomato plants were cut above the third leaf of the eighth cluster on 12 June in both years. In 2009, fruits were harvested as they matured (at a light red colour) in order to measure fruit yield characteristics. Seven fruit harvests were taken during the experiment, starting on 25 June. The average fruit weight, total yield and percentage of fruits damaged by BER were recorded.

2.3. *Treatments applied*

In 2008, the tobacco whitefly was studied on a tomato crop fertigated with three different nutrient solutions, with levels of N at: low (75 mg/l N), standard (205 mg/l N), and high (335 mg/l N) concentrations. The treatments commenced on the 14 May, 14 d after transplanting. Whitefly adults were releasing onto the crop on the 20th day (3 June) when the effect of the N feeding was first determined, based on measurements of the total leaf N measured by Kjeldahl digestion with a Kjeltec System 1026 and relative water content (RWC) (Goreta et al., 2007). HNT was also used as a non-destructive N and chlorophyll diagnostic tool (Ortuzar-Iragorri et al., 2005). All measurements were performed on the youngest fully developed leaf, usually the fourth from the top of the plant.

In 2009, we tested the effect of three ratios of NO_3^-:NH_4^+ ions (92:8, 75:25, and 55:45) on the population density of *B. tabaci* on tomato. The composition of the nutrient solution was the same as the standard applied in 2008, i.e. 205 mg/l N, but with differing NO_3^-:NH_4^+ ratios depending on the treatment. The NO_3^-:NH_4^+ ratio 92:8 was considered as the standard, while the two others had increased proportions of NH_4^+. The proportions of NH_4^+ chosen were selected according to Siddiqi et al. (2002) who showed that proportions of NH_4^+ up to 50% of the total N had no negative effect on early vegetative tomato growth. The treatments were applied from the 6 May, 7 d after transplanting to rockwool slabs. Whitefly adults were released on the 23 May when HNT values at the fourth leaf from the top differed significantly between treatments.

On 3 June 2008 and 23 May 2009, tomato plants were infested with *B. tabaci* reared on peppermint plants. Three highly infested peppermint plants were placed in the greenhouse within each replication of six tomato plants and removed after 2 d.

2.4. *Whitefly sampling*

In both years, sampling of whitefly instars was performed using the same procedure.

Assessments of adult number were conducted 2, 4, 6 and 8 d after infestation (DAI). *B. tabaci* adults were recorded early in the morning (7 a.m.) on the 15 marked leaves of all 16 plants in each treatment, using the leaf-turn method, starting with the youngest leaf. At the last survey (at 8 DAI), adults were removed using a manual aspirator (Gempler's, Madison, WI, USA).

To record the egg density at 10 DAI, the 4th, 9th and 14th leaves were collected from two plants per experimental plot and examined in the laboratory. The total number of eggs per leaf was recorded using a stereo microscope (Stemi 2000, Zeiss). The leaf area was determined using a leaf area meter (LI-COR), and the data were expressed as the number of eggs per 100 cm^2.

The density of nymphs and pupae at 60 DAI, was recorded from 9 cm^2 leaf cuttings taken from the 4th, 9th and 14th leaves of two sampled plants from each plot. The total number of nymphs (of all four instars) and pupae, including empty pupal cases, per 9 cm^2 cutting was recorded using a stereo microscope and expressed as the number of nymphs and pupae per cm^2.

2.5. *Tomato foliage infestation with sooty mould*

At the end of the experiments, 60 DAI, the overall infestation of the tomato plants with sooty mould was estimated on two plants