EFFECTS OF FOLIAR APPLIED JASMONIC ACID AND SEED APPLIED IMIDACLOPRID ON PHYTOHORMONE EXPRESSION AND TWOSPOTTED SPIDER MITE POPULATIONS IN COTTON

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Abstract

Cotton responds to insect injury by eliciting signal pathways that result in the production of metabolites that have been demonstrated to deter insect feeding, reduce insect fitness, and attract predators. Furthermore, neonicotinoid seed treatments have been shown to suppress some of these natural secondary metabolites increasing the prevalence of phytophagous arthropods on seedling cotton. Therefore, we investigated the effects of selected elicitors on the production of plant defenses using phytohormone analysis and measuring population growth of two-spotted spider mite, *Tetranychus urticae*, (TSSM) on cotton plants grown in a growth chamber after foliar applications of Jasmonic Acid (JA) and seed applied imidacloprid. Our research indicates neonicotinoid seed treatments may alter naturally occurring secondary plant metabolites resulting in significantly more adult spider mites as compared JA treated cotton. However, eggs, motiles and total mites were not affected by the addition of a neonicotinoid seed treatment or exogenous applications of JA. Concentrations of JA were determined to be significantly greater in plots where JA was applied compared to plots that had no JA applications. Jasmonate-isoleucine (JA-Ile) was also determined to be significantly greater in plots sprayed with JA compared to plots that were not sprayed. Concentrations of 12-oxo-phytodienoic acid (OPDA), salicylic acid (SA), abscisic acid (ABA) were not found to be significantly different in sprayed compared to non-sprayed plots. Finally, neonicotinoid seed treatments appeared to have little effect on the population growth or health of TSSM populations in this study.

Introduction

Neonicotinoid insecticides are the fastest growing and one of the most frequently used classes of insecticides in the world (Jeschke 2008). The pervasiveness of these insecticides stems from their extended insecticidal activity; variety of formulations including foliar, drench and seed applied uses in agricultural settings and cost effectiveness. Despite their wide spread use, neonicotinoid insecticides have been associated with outbreaks of several species of spider mites in horticultural and agronomic crops (Szczepaniec et al. 2013). Spider mites often do not respond to applications of neonicotinoids, and elimination of natural enemies after a neonicotinoid application may not be the sole mechanism resulting in large population increases of pest mites. Recent research has demonstrated a negative physiological response in plant defense hormones in cotton, corn and tomato to applications of neonictoinoid insecticide seed treatments (Szczepaniec et al. 2013) Injury by spider mites, specifically the TSSM in Louisiana cotton is severe, and control measures are costly. In 2015, Louisiana producers lost 15,608 bales of cotton to TSSM resulting in 1.0 applications costing $9.36 per acre (Williams 2015).

Therefore, the objective of this study was to investigate the effects of selected elicitors on the production of plant defenses using phytohormone analysis and measuring artificial populations of TSSM on pre-squaring cotton after foliar applications of JA and seed applied imidacloprid.

Materials and Methods

This study was performed at the Macon Ridge Research Station (MRRS) near Winnsboro, LA during 2015. The test area consisted of 48 cages (Fig. 1, 2), arranged in a randomized complete block design and maintained at 28ºC and 14:10 LD configuration. The study consisted of a 3x3 factorial with factor A consisting of an imidacloprid seed treatment, factor B consisting of an infested treatment and factor C consisting of a foliar JA application.

Figure 1. Top and bottom cage used to contain mites.

Figure 2. Top and bottom cage with attached thrips netting, water resistant clay and watering holes

The variety used for all treatments was Phytogen 499 WRF [WideStrike® (WS; Cry1Ac, Cry1F)]. Seed treatments were applied by hand using a small plastic bag (2.5 lbs. seed/bag using a 50% slurry). All seeds were planted in Miracle Gro® potting soil and water was added as needed. Cages were constructed out of 1 gallon clear, plastic PET containers (ULINE®, 2015) for the cage tops with 4 inch holes drilled into each side and top (Figure 1,2). Thrips netting was secured over each hole, with hot glue, to facilitate evapotranspiration by the plants and to reduce condensation accumulation. Cage bottoms were constructed out of 0.5 gallon clear, plastic PET containers. Bottoms were filled with soil to a designated mark and small watering holes were melted into each side of the cage bottom to reduce the amount of disturbance to each cage for watering. Two seeds were planted approximately 1.5 inches in depth and allowed to germinate before being thinned to one. After plants had reached sufficient height, water proof modeling clay (Prima Plastilina®, 2014) was secured around the plant stalk to prevent any mites from escaping the confinement area (Figure 1,2).

Seven days after planting (approximately two true leaves), ten field collected, 1st instar larval stage TSSM were placed on the terminal leaves with a 10/0 fine camel hair paint brush. Larvae were allowed to naturally distribute before cage tops were secured onto bottoms and placed into the growth chamber. Seven days after infestation, foliar JA applications were made using a 2 nozzle per row, 3 liter back pack sprayer calibrated to deliver 10 gallons per acre. Treated cages were gently removed from the growth chamber and randomly arranged in two straight lines approximately 40 inches apart to simulate plants grown in a row on 40 inch centers. Applications of JA were conducted inside of a facility to negate any disturbance from wind and to reduce the possibility of thrips contamination to exposed cotton plants. After application, the spray was allowed to dry for 1 hour and cage tops were resecured and placed back into the growth chamber.

Ten days after application, all leaf tissue was excised and examined under a dissecting microscope for presence of all mite life stages. Random tissue samples were pulled from excised leaves and individual samples from each cage were weighed, recorded and placed into 1ml microcentrifuge tubes (Fisher Scientific®, 2014) before immersion in liquid nitrogen. Samples were packed in dry ice for shipment to the Donald Danforth Plant Institute in St. Louis, MO for phytohoromone analysis.

All data were analyzed using PROC GLIMMIX. . Data analyzed by treatment used random effects of Rep and IST*Rep. Where significant interactions were detected between IST and JA, the SLICEDIFF option of the LSMEANS statement was utilized to determine if a given treatment differed in number of adult, immature, egg, motile, immature + egg and total mites.

Results and Discussion

Adult TSSM were significantly ($P < 0.05$) reduced when JA was applied as compared to the non-treated control (Figure. 8). This demonstrates the ability of JA to possibly reduce adult TSSM survival in pre-squaring cotton plants. By priming or saturating the plant with this phytohormone, cotton may acquire an increased level of natural protection that helps effectively slow or disrupt the life cycle of TSSM populations infesting cotton. However, all other mite life stages did not significantly differ under the presence of JA or the IST treatment (Figure. 8 – 11). Therefore, JA may play an important role in slowing initial colonization by migrating adult spider mites throughout the season.

Furthermore, our research indicates that imidacloprid may not suppress naturally occurring secondary metabolites in a laboratory setting (Figure. 3 – 7). However, significant differences in JA and JA-Ile concentrations were detected in the JA treatments compared to the non-treated black seed plots (Figure. 3 – 4). JA-Ile is a conjugate of jasmonic acid and isoleucine and is an important signaling pathway in plants to herbivore induced responses. This demonstrates the potential for JA to be absorbed into leaf tissue and positively increase JA and JA metabolite concentrations in cotton plants. Other metabolites tested were OPDA, SA and ABA. This suite of plant hormones play an important role in signaling the JA pathway to produce compounds such as protease inhibitors when plants are injured by arthropods. However, the use of JA in agricultural cotton production systems is cost prohibitive. The use of commercially labelled acaricides provide adequate control of TSSM populations and are competitively priced to allow producers the option of controlling damaging mite populations while economically producing cotton.

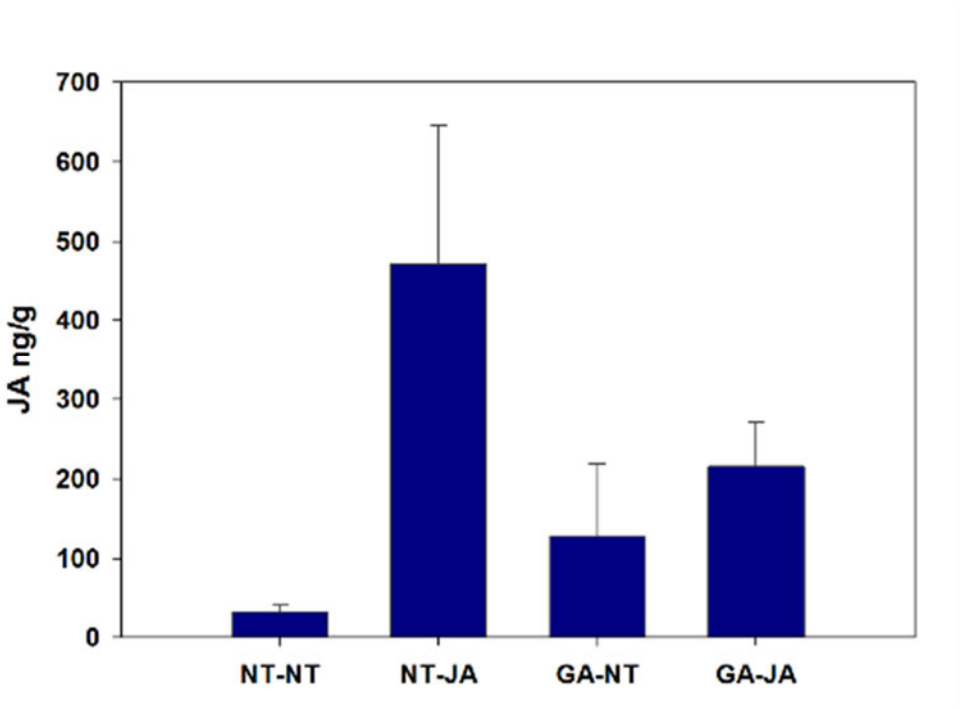

Figure 3. JA concentration in ng/g of fresh weight.

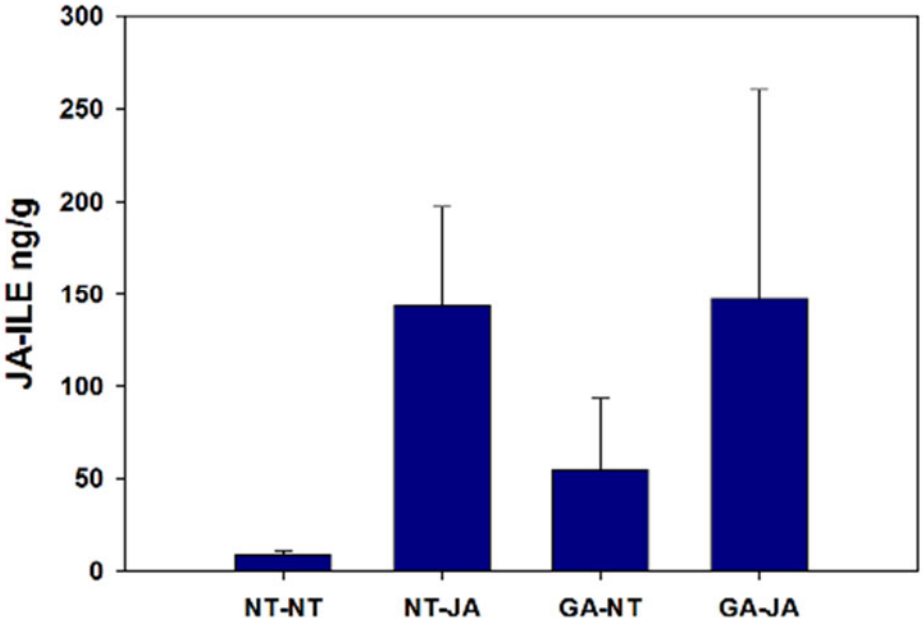

Figure 4. JA-Ile concentration in ng/g of fresh weight.

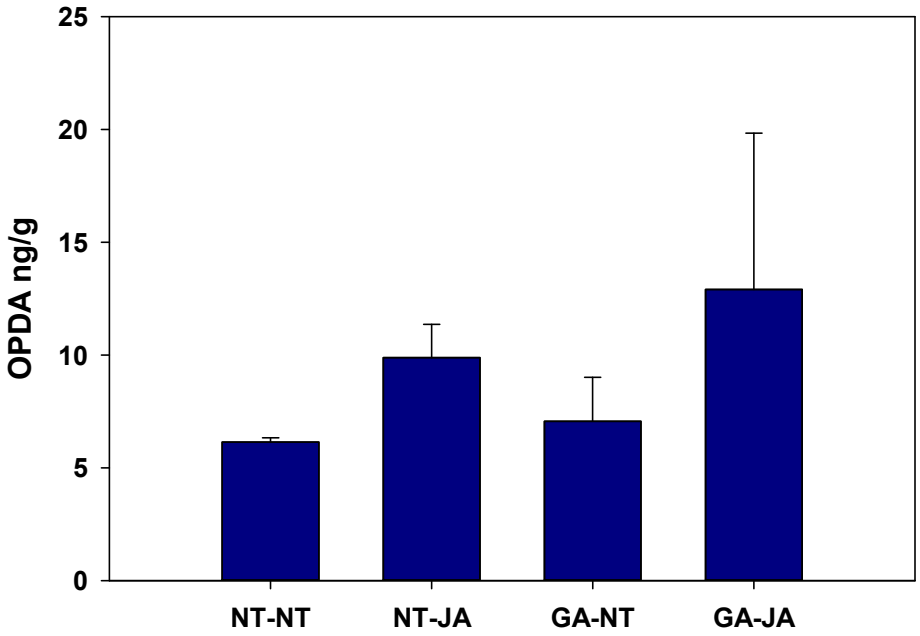

Figure 5. OPDA concentration in ng/g of fresh weight.

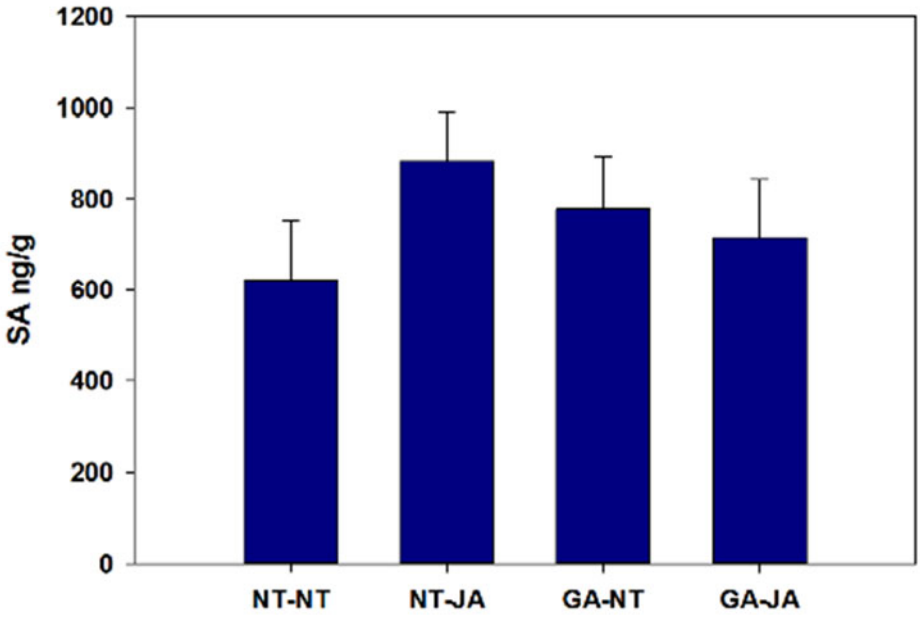

Figure 6. SA concentration in ng/g of fresh weight.

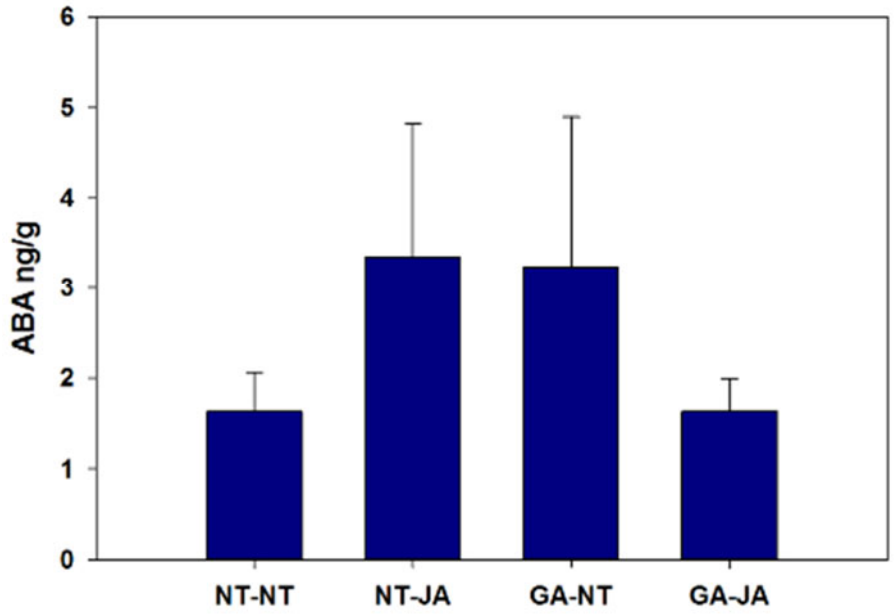

Figure 7. ABA concentrations in ng/g fresh weight.

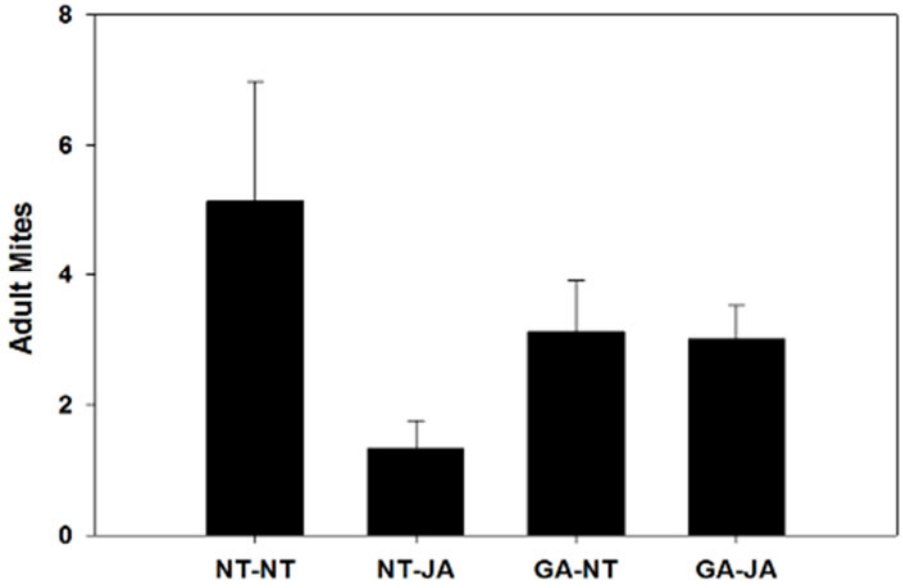

Figure 8. Total adult mites across treatments

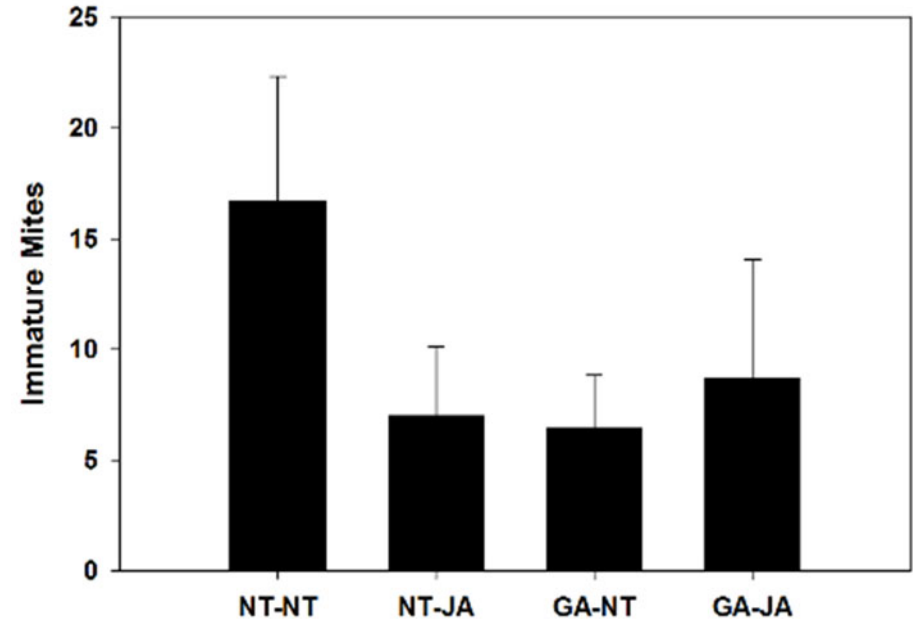

Figure 9. Total immature mites across treatments.

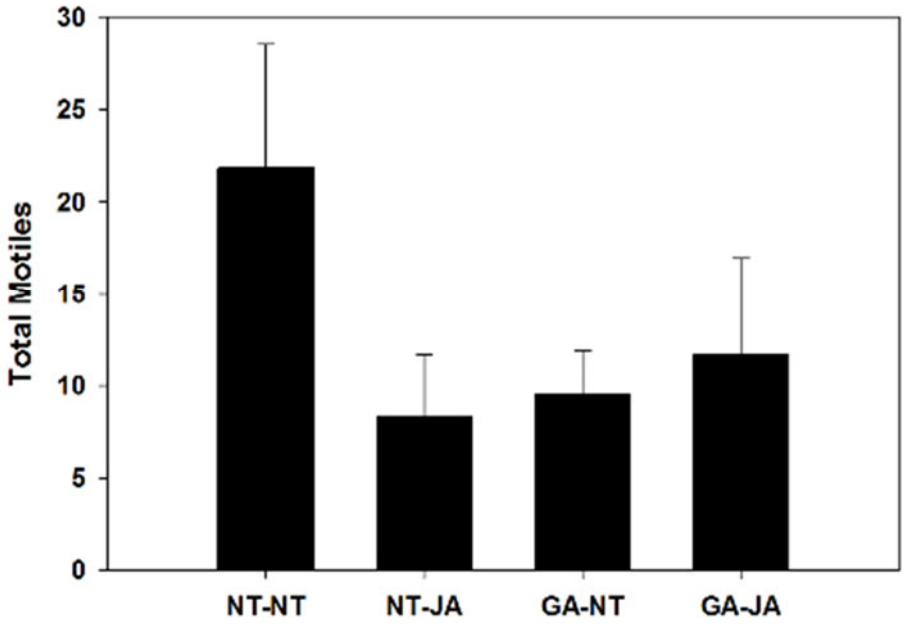

Figure 10. Total motiles across treatments.

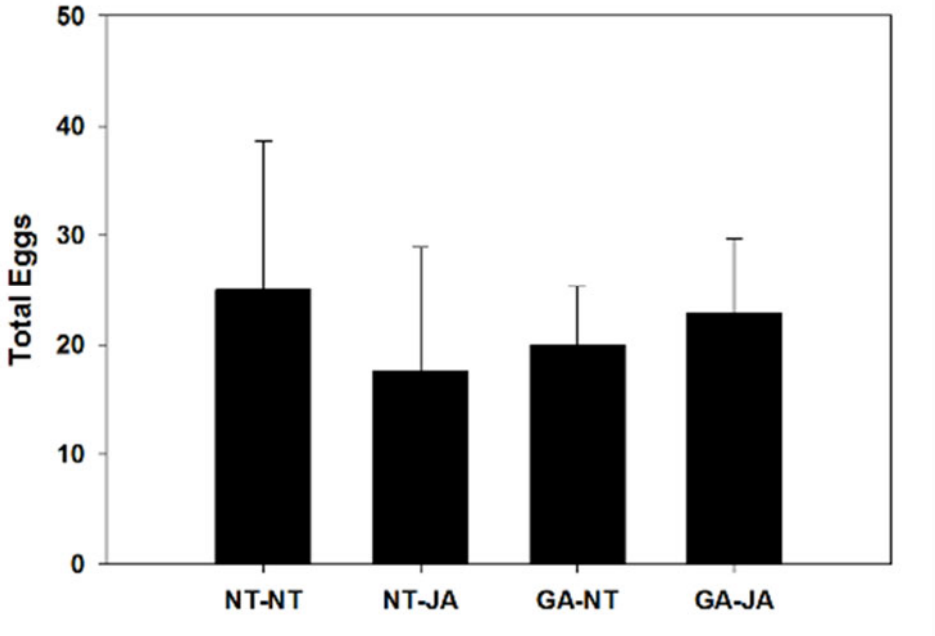

Figure 11. Total eggs across treatments.

References

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Szczepaniec A, Raupp M. J, Parker R. D, Kerns D, Eubanks M. D. (2013) Neonicotinoid Insecticides Alter Induced Defenses and Increase Susceptibility to Spider Mites in Distantly Related Crop Plants. PLoS ONE 8(5): e62620. doi:10.1371/journal.pone.0062620

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