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FACULDADE DE CIÊNCIAS AGRÁRIAS E VETERINÁRIAS
CÂMPUS DE JABOTICABAL

RESISTANT GENOTYPES, BIOLOGICAL CONTROL AND SELECTIVE PESTICIDES FOR THE INTEGRATED MANAGEMENT OF *Tetranychus evansi* (ACARI: TETRANYCHIDAE) ON TOMATO

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2022

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Advisor: Prof. Dr. Daniel Júnior de Andrade
Co- Advisor: Prof. Dr. Gilberto José de Moraes

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TÍTULO DA TESE: RESISTANT GENOTYPES, BIOLOGICAL CONTROL AND SELECTIVE PESTICIDES FOR THE INTEGRATED MANAGEMENT OF *Tetranychus evansi* (ACARI: TETRANYCHIDAE) ON TOMATO

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PATRICE JACOB SAVI was born in Ifangni (Republic of Benin) on March 17th, 1991. Son of Martin Savi and Beatrice Lokossou, he was an undergraduate in Agronomy at Université de Parakou (Republic of Benin), where he finished his degree in 2012. He did an undergraduate thesis under the supervision of Prof. Dr. Fabien C.C. Hountondji and worked with pest management approaches in vegetable crops at the Songhai center in Porto-Novo (Republic of Benin). After getting his degree, he got his first job as a Head Agriculturist at "Groupe Vision Plantain Hévea Cacao (GVPHeC)" in Ifangni (South of the Republic of Benin), mostly working with farmers on the control of the banana weevil, *Cosmopolites sordidus* (Coleoptera: Curculionidae), and farm maintenance. From there, he went for an internship of two-month at a non-governmental organization "FUDEL" in Ifangni (South Benin) specializing in vegetable crop production to gain more hands-on experience as a horticulturist. After completing this internship, he got a position as Head horticulturist for two years at Aganmandin Farm in charge of Formation, Research in Horticulture, and Animal Husbandry working with all aspects related to the maintenance, pest, and disease control strategies for vegetable crops. From 2014 to 2016, he was also a Professor at Benin Secondary School. In March 2016, he was awarded a scholarship by Conselho Nacional de Desenvolvimento Científico e Tecnológico-CNPq to join the Master Program in Agricultural Entomology Graduate Program (Agronomy) at São Paulo State University (UNESP) working under the supervision of Prof. Daniel Júnior de Andrade, and Gilberto José de Moraes. While in his Master's, he got an interest in selecting African tomato genotypes resistant to *Tetranychus evansi* (Acari: Tetranychidae). He was immediately accepted to continue as a Doctorate student, soon after the completion of his Master´s program, in early 2018 at the same University, under the supervision of the same advisors, and received a scholarship from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior-CAPES.

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DEDICATION

Every challenging work needs self-efforts as well as the guidance of elders especially those who were very close to our hearts.

My humble effort I dedicate to my

Parents: Béatrice and Martin
Brothers: Simon, David, Élie, and Isaac
Sisters: Agnès and Elisabeth

Whose affection, love, encouragement and prayers of day and night make me able to get such success and honor.

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SUMMARY

Page

GENÓTIPOS RESISTENTES, CONTROLE BIOLÓGICO E PESTICIDAS SELETIVOS PARA O MANEJO INTEGRADO DE *Tetranychus evansi* (ACARI: TETRANYCHIDAE) EM TOMATE

RESUMO - O ácaro vermelho do tomateiro, *Tetranychus evansi* Baker & Pritchard (Acari: Tetranychidae), é uma praga invasora do tomateiro em vários países, com potencial de reduzir a produtividade em até 90% na África. Devido ao alto potencial biótico da praga, o manejo focado no uso de defensivos sintéticos muitas vezes não é eficiente ou insustentável ao longo do tempo, sendo necessária a sua integração com outros métodos de controle. Estudos anteriores encontraram em genótipos selvagens fonte expressiva de resistência (tricomas glandulares) que poderiam ser exploradas para aumentar o nível de resistência de variedades de interesse a esta praga. Além disso, *Phytoseiulus longipes* Evans (Phytoseiidae), encontrado na América do Sul, mostrou-se um promissor ácaro predador de *T. evansi*. No entanto, a integração deste ácaro predador em programas de MIP onde *T. evansi* é um problema sério requer conhecimento detalhado das interações com outras práticas de manejo. Dessa forma, objetivou-se com este trabalho estabelecer um sistema de manejo integrado para *T. evansi* com a aquisição de genótipos de tomateiro resistentes, biopesticides eficientes a *T. evansi*, um genótipo adequado que pudesse otimizar o desempenho do ácaro predador *Phytoseiulus longipes* Evans (Phytoseiidae) e com a definição de agrotóxicos seletivos comumente usado em tomateiro a esse predador. Os estudos foram conduzidos em condições de laboratório e semi-campo. As progênies F1, SPJ-10-2017 e SPJ-05-2018 obtidas cruzando o genótipo selvagem resistente [*Solanum habrochaites*, acesso PI 134417] com *Solanum lycopersicum*, cv TLCV15 [importante genótipo cultivado amplamente cultivado no Benin] herdaram significativos tricomas glandulares tipos I, IV e VI de seu pai resistente (PI 134417). As densidades de tricomas glandulares herdados pelos genótipos da progênie foram capazes de reduzir ou suprimir as infestações causadas por *T. evansi*. No entanto, o genótipo de progênie causou atrasos importantes no crescimento populacional e reduziu significativamente a sobrevivência e o potencial de predação de *P. longipes*. Os genótipos cultivados com maior número de tricomas não glandulares mostraram-se adequados para a implementação do programa IPM que visa otimizar o uso de *P. longipes* como agente de biocontrole. Os resultados demonstraram que o uso de biopesticidas à base de azadirachtin e oxymatrine apresentaram alta atividade contra *T. evansi* e pode ser uma importante alternativa para uso no manejo de *T. evansi* em substituição ou rotação com acaricidas sintéticos. Além disso, azadiractina mostrou-se mais segura ao ácaro predador tanto no controle biológico aumentativo quanto na conservação, enquanto a oximatrina mostrou-se adequada apenas para o controle biológico aumentativo se 10 dias for observado após aplicação. Os agrotóxicos comumente usados no sistema de cultivo do tomateiro como abamectina, propargite, imidacloprid e o fungo entomopatogênico *Hirsutella thompsonii* (Fischer) (Deuteromycetes) são mais compatíveis com o controle biológico aumentativo do que com a conservação se os prazos de segurança adequados forem respeitados antes da liberação. Os inseticidas piretróides (cipermetrina e deltametrina) e organofosforados (dimetoato, clorpirifós) não são compatíveis com o uso de *P. longipes* em programas de MIP. Esses resultados são importantes para o manejo sustentável dessa praga invasora e, ao mesmo tempo, fornecem diretrizes práticas que possibilitam uma melhor forma de uso de agrotóxicos em programas de MIP que visam conservar ou realizar liberações aumentativas do ácaro predador.

Palavras-chave: ácaro vermelho do tomateiro, Manejo Integrado de Pragas, genótipo resistente, Phytoseiidae, potencial de predação, biopesticidas, impacto de agrotóxicos sobre predador

RESISTANT GENOTYPES, BIOLOGICAL CONTROL AND SELECTIVE PESTICIDES FOR THE INTEGRATED MANAGEMENT OF *TETRANYCHUS EVANSI* (ACARI: TETRANYCHIDAE) ON TOMATO

ABSTRACT - The tomato red spider mite, *Tetranychus evansi* Baker & Pritchard (Acari: Tetranychidae), is an invasive tomato pest in several countries, with the potential to reduce yield by up to 90% in Africa. Due to the high biotic potential of the pest, the management focused on the use of synthetic pesticides is often not efficient or unsustainable over time, requiring the integration with other control methods. Previous studies found in wild genotypes expressive source of resistance (glandular trichomes) that could be explored to increase resistance level of varieties of interest to this pest. Furthermore, *Phytoseiulus longipes* Evans (Phytoseiidae), found in South America proved to be a promising predatory mite of *T. evansi*. However, the incorporation of this predatory mite into IPM programs requires detailed knowledge of the interactions with other management practices. Within this context, the objective of the present study was to establish an integrated management system with the acquisition of tomato genotypes resistant to *T. evansi*, a suitable genotype that could optimize the performance of predatory mite *P. longipes* and with the definition of selective pesticides to this predator.The studies were conducted under laboratory and semi-field conditions. Our results indicated that the progenies F1, SPJ-10-2017 and SPJ-05-2018 obtained by crossing the wild-resistant genotype [*Solanum habrochaites*, Knapp e Spooner var *glabratum* access PI 134417] with *Solanum lycopersicum* L., cv. TLCV15 [cultivated genotype widely grown in Benin] have inherited significant glandular trichomes types I, IV and VI from their resistant parent (PI 134417). The densities of these glandular trichomes inherited by progeny genotypes were able to reduce supress the infestation caused by *T. evansi*. However, the bred progeny genotype SPJ-05-2018 caused important delays population growth and reduced significantly a survival, and the predation potential of *P. longipes*. The cultivated genotypes with many non-glandular trichomes proved to be more suitable for the implementation of IPM program that aim to optimize the use of *P. longipes* as biocontrol agent. The results showed that the use of azadirachtin- and oxymatrine based biopesticides had high activity against *T. evansi* and may be an important alternative in the management of the mite in replacement or rotation with synthetic acaricides. Azadirachtin proved to be the safest against the predatory mite toward both augmentative biological control and conservation whereas oxymatrine proved to be suitable only toward augmentative biological control 10 days after application. Other pesticides used in tomato cropping system such as abamectin, propargite, imidacloprid and the enthomopathogenic fungus *Hirsutella thompsonii* (Fischer) (Deuteromycetes) are more compatible with augmentative biological control than conservation if appropriate safety deadlines are respected before release. The insecticides belonging to pyrethroid (cypermethrin and deltamethrin) and organophosphate (dimethoate, chlorpyrifos) groups are not compatible with the use of *P. longipes* in IPM programs. These results are important to sustainably manage this invasive mite pest, and at the same time, provide

practical guidelines to enable a better way of using pesticides in IPM programs that aim to conserve or increase the predatory mite *P. longipes*.

Keywords: tomato red spider mite, Integrated Pest Management, resistant genotype, Phytoseiidae, predation potential, biopesticides, impact of pesticides on predator

CHAPTER 1- General Considerations

Introduction

Tomato (*Solanum lycopersicum* L.) is one of the most widely cultivated and consumed vegetable crops worldwide (Kimura and Sinha, 2008; Sibomana et al., 2016). In sub-Saharan Africa, this crop is commonly planted by small-scale farmers and represents an important source of income (Arah et al., 2015; Sibomana et al., 2016). However, efforts of farmers to increase tomato production often face various problems in the field, such as interference from intruder organisms (pests and diseases) that substantially affect tomato production (Arah et al., 2015; Wakil et al., 2018; Abera et al., 2020). Among pests, the spider mites should be highlighted, as some species are key pests of this crop, requiring considerable investments in their control every year in an attempt to prevent yield losses (Brust and Gotoh, 2018).

In this group, *Tetranychus evansi* Baker & Pritchard (Acari: Tetranychidae), also known as tomato red spider mite, has been considered as one of the most devastating pests for tomato and other solanaceous plants in the world, mainly in Africa (Navajas et al., 2013; Azandémè-Hounmalon et al., 2015; Savi et al., 2019a; Djossou et al., 2020). This mite is considered native to South America, probably from the northeastern part of Brazil, but over the past decades, it has gradually invaded and colonized several tropical and subtropical habitats worldwide (Navajas et al., 2013). Importantly, in most invaded regions, *T. evansi* is a recurrent pest responsible for massive crop losses and significant economic damages, especially to small-scale farmers (Ferragut et al., 2013; Azandémè-Hounmalon et al., 2015; Knegt et al., 2020).

It is presently found in 44 countries in Africa, the Americas, Europe, and Asia (Migeon and Dorkeld, 2006-2019; Fan et al., 2021). The rapidly changing climate, which will result in higher average temperature and lower rainfall in large parts of the world, probably facilitates the ongoing expansion of *T. evansi* into new territories, thus putting progressively larger areas at risk (Meynard et al., 2013; Ximénez-Embún et al., 2016; Migeon and Dorkeld, 2006-2019). *Tetranychus evansi* pierces cell walls to extract their contents. This feeding activity causes chlorotic spots, reducing the photosynthetic capacity and often leading to leaf fall (de Moraes and Flechtmann, 2008; Bensoussan et al., 2018). In the absence of effective management methods, mite-feeding activity can cause yield losses ranging from 56 to 100%, mostly in African countries (Sarr et al., 2002; Boubou et al., 2010; Azandémè-Hounmalon et al., 2015).

In general, spraying acaricides or insecticides with acaricidal properties is the major management strategy for controlling *T. evansi* (Blair, 1989; Toroitich et al., 2014; Gotoh et al., 2011; Azandémè-Hounmalon et al., 2015; Bagaram, 2016). However, the rapid development and high reproductive capacity (Qureshi et al., 1969; de Moraes and Flechtmann, 2008) allow *T. evansi* to reach high population levels, leading producers to conduct several annual pesticide applications for its control (Savi et al., 2019a). Unfortunately, this excessive usage of pesticides usually becomes untenable in the long run because it leads inevitably to the development of resistance, negatively impacting the environment as well as human and livestock health, besides increasing crop production costs (Blair, 1989; Nyoni et al., 2011; Azandémè-Hounmalon et al., 2015). Therefore, the sustainable management of this invasive mite requires the development of ecologically friendly crop protection methods that suit the needs of small farmers. Within this context, the use of acaricides should be subordinated or integrated with other control methods, such as biological control and the use of resistant genotypes. The use of selective acaricides has also gained more credibility in the last decades (Hoy, 2011; Stenberg, 2017; Stout et al., 2018; Duso et al., 2020).

In the search for such methods, the resistance of five cultivated tomatoes widely grown by smallholder farmers in western Africa (Kekefo, TOML4, Akikon, Tounvi, and TLCV15) and two wild relatives [*Solanum habrochaites* Knapp & Spooner (accessions PI 134417 and PI 134418) and *Solanum pennellii* Correll (accession LA-716)] have been evaluated in a previous study. Unfortunately, the tomato varieties grown in Africa and other commercial tomato cultivars have proved to be susceptible to *T. evansi*. In contrast, the wild tomato genotypes have been experimentally shown highly resistant to *T. evansi* (Savi et al., 2019a, b). This difference has been attributed to the presence of a greater number of glandular trichomes and their toxic compounds (as acyl-sugars, methyl ketones, and terpenoids) on wild genotypes, but their absence of great reduction on cultivated tomatoes (Resende et al., 2008; Bleeker et al., 2012; Lucini et al., 2015). These structures proved to entangle or kill phytophagous arthropods through the sticky or toxic exudates that the trichomes produce (Kang et al., 2010; Zhang et al., 2020). They can also serve as repellent barriers to small herbivores, preventing them from feeding freely on the surface of a plant (Zhang et al., 2020). Hence, the introgression of glandular trichomes from wild tomato relatives to cultivated tomatoes, through interspecific crossings, could conceivably be one of the ways to increase the resistance degree of cultivars of interest against *T. evansi* and consequently adapted to sustainable production systems (Savi et al., 2019b). This could also help small-scale farmers who are not well resourced to purchase effective acaricides. That accounts for the first goal of this dissertation.

Biological control using natural enemies is also a major component of any integrated pest management (IPM) program (Van Driesche et al., 2008; Almarinez et al., 2020). In the case of spider mites, predatory mites of the family Phytoseiidae are the most commonly studied and important group of natural enemies considered for their control (McMurtry et al., 2013). Species of this family have been commercialized to suppress pest mite populations (McMurtry et al., 2013). Their feeding preference for phytophagous mites, short life cycles, and feasible large-scale production make them good candidates for pest control (Abad-Moyano et al., 2009). The phytoseiid mite *Phytoseiulus longipes* Evans (Acari: Phytoseiidae) found naturally in association with *T. evansi* in the extreme south of Brazil and northern Argentina proved to be the only promising predator for red spider mite control (Furtado et al., 2007; Silva et al., 2010; Ferreiro et al., 2011). This predatory mite has the potential to control other *Tetranychus* species, given its adaptation to the type I-a lifestyle of McMurtry et al. (2013).

However, the incorporation of predatory mites into IPM programs requires detailed knowledge and understanding of the interactions of these mites with other crop management practices (Fountain and Meed, 2015). The defensive traits that crops exhibit to protect themselves from damage caused by pests and disease can also strongly influence the survival and efficacy of their predators. In the case of tomato, several studies have demonstrated that the performance of most phytoseiid mites tends to be lower compared to other crops (Koller et al., 2007; Sato et al., 2011; Davidson et al., 2016; Paspati et al., 2021). This fact has been attributed to the impact of tomato defenses mediated by the glandular trichomes and their exudates, increasing phytoseiid mortality or phytoseiid prey-searching efficacy if non-glandular and glandular trichomes are above a certain threshold (Castagnoli et al., 1999; Koller et al., 2007; Sato et al., 2011; Paspati et al., 2021). Tomato genotypes can vary highly in trichome density and such variation could influence differently the performance of phytoseiid mites. However, although tomato has been found to affect predatory mites more than other crops, studies about this subject are scarce. Thus, the effect of tomato genotypes with varying levels of susceptibility or resistance to *T. evansi* on *P. longipes* should be investigated. Such knowledge will be fundamental in the choice of tomato genotypes to optimize the use of this phytoseiid mite as a biocontrol agent in the integrated management program of *T. evansi*. Accordingly, this was the second goal of this dissertation.

In the development of Integrated Pest Management Programs, it is also important for farmers to have available pesticides minimally harmful to the natural enemies, or that the effect of these two control measures taken together is greater than the sum of their separate effect (Schmidt-Jeffris and Beers, 2020; Bilbo and Walgenbach, 2020). Several studies on non-target effects on key phytoseiid species have been conducted to address this need. However, knowledge of pesticides non-target effects on *P. longipes* is still scarce. Therefore, investigations should be conducted to determine which pesticides are most selective against this predatory mite in the tomato cropping system, as pest control practices adopted by most Western African tomato producers are dominated by the intensive use of pesticides, with no attention on their possible effect on natural enemies of *T. evansi*.

Thus, this dissertation intends to establish an integrated management system for *T. evansi*, with the acquisition of tomato genotypes resistant to *T. evansi*, a suitable genotype that could optimize the performance of predatory mite *P. longipes*, and with the definition of pesticides with lower risk to this predator. The thesis is organized as follows. Chapter 2 explores the bottom-up effects of progeny genotypes from interspecific crossings of wild and cultivated tomatoes on the behavioral responses and demographic parameters of *T. evansi* to increase resistance degree of a cultivar of interest in the Republic of Benin (West Africa) cropping system against *T. evansi*. Chapter 3 compares *P. longipes* population performance and predation capacity on tomato genotypes with different susceptibility levels to *T. evansi*, to identify the genotype, which should be included in the envisioned IPM program to optimize *P. longipes* as a biocontrol agent. Chapter 4 evaluates the effectiveness of two bio-acaricides (azadirachtin- and oxymatrine-based formulations), which are labeled as selective for bio-control agents for controlling *T. evansi* on tomato crop, explores also their synergistic effect with the predatory mite *P. longipes* to support effective integrated management of *T. evansi*. Chapter 5 assesses the short- and long-term effects of direct exposure as well as the persistence of residual activity of ten pesticides commonly used in the Western African tomato cropping system against *P. longipes* to screen those are suitable toward conservation and/ or preservation of this predatory mite in the field. Chapter 6 presents a summary of overall empirical findings, conclusion, and suggestions resulting from this thesis.

5. Conclusion

Through studies performed under laboratory and screen-house conditions, we have shown that the pesticides commonly used in tomato crop in West Africa greatly differed in their lethal and sub lethal toxicity to *P. longipes*, a new biocontrol agent of *T. evansi.* The results suggest insecticides belonging to pyrethroid (cypermethrin and deltamethrin) and organophosphate (dimethoate, chlorpyrifos) groups are not compatible with both conservation and increase of *P. longipes* population in IPM programs. Azadirachtin is the safest for augmentative biological control, as well as for conservation mostly during periods of low presence of *P. longipes* eggs. Other pesticides (abamectin, propargite, imidacloprid, oxymatrine, *H. thompsonii*) are most suitable for augmentative biological control than conservation if appropriate safety deadlines are respected before release.

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