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Application of pupal parasitoids for the biological control of *Drosophila suzukii* (Diptera: Drosophilidae) in berry fruit production

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Abstract

The spotted-wing drosophila, Drosophila suzukii*, poses a significant threat to European stone fruit and berry production. Conventional management methods often prove partially effective, prompting the need for alternative protection strategies. The "ParaDrosu" project aims to develop a biological control approach utilizing two native German parasitoid species:* Trichopria drosophilae *and* Pachycrepoideus vindemiae*. Key aspects are investigated, including mass rearing, climatic suitability, and application techniques. Here, we report on laboratory studies assessing the impact of organic plant protection products on parasitoid lifespan and parasitization rate, as well as field trials to evaluate parasitoid dispersal and efficacy. Results suggest potential for biological control, highlighting the importance of further optimization and integration into pest management strategies.*

Keywords: *Drosophila suzukii*, biological control, parasitoid wasps, plant protection products, field trials.

Introduction

The invasive spotted-wing drosophila (SWD), *Drosophila suzukii* Matsumura, has become a major pest in stone fruit and protected European berry fruit production since its unintentional introduction in 2008 (Calabria et al., 2012). SWD females possess a unique ability to attack intact ripe and ripening soft-skinned fruit by depositing eggs into the intact fruit skin with its serrated ovipositor (Atallah et al., 2014). The emerging larvae then damage the fruit by feeding on fruit pulp, leading to premature rotting (Walsh et al., 2011). Its host preferences comprise economically important crops such as strawberries, cherries, raspberries, and blueberries among various cultivated host plants (Kirschbaum et al., 2020). This pest's rapid reproductive rate and capacity to infest multiple hosts make it particularly challenging to manage through conventional or organic means (Del Fava et al., 2017). Insecticide applications are commonly employed but often ineffective due to the timing of infestation of ripening or ripe fruits and long harvest periods while having negative effects through insecticide residues when applied close to harvest (Van Timmeren and Isaacs 2013). While exclusion netting can offer some protection, its costliness and occasional inadequacy in controlling emerging populations still cause concerns (Stockton et al., 2020). Fortunately, the native pupal parasitoids *Trichopria drosophilae* Perkins (Hymenoptera: Diapriidae) (TD) and *Pachycrepoideus vindemiae* Rondani (Hymenoptera: Pteromalidae) (PV) have been found to successfully parasitize *D. suzukii* and thus have a great potential as biological control agent that may supplement current measures.

The research project "ParaDrosu," initiated in 2021, aims to develop an innovative biological management strategy against *D. suzukii*. Key aspects under investigation include the

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successful mass rearing of parasitoids, which necessitates suitable host species, optimal climatic conditions, and an appropriate diet for adult parasitoids. Moreover, the rearing system must be both efficient and economically viable. An appropriate parasitoid application technique ensuring optimal protection during transport and release and sufficient dispersal in the crop is developed. Field studies in different berry cropping plantations help to determine the best release times, intervals, and parasitoid numbers. Relevant plant protection products, both used in integrated and organic production, are tested for negative side effects on the parasitoids to define possible incompatibilities and suitable timing for integrated pest management strategies. Semi-field and field trials in Southern Germany will deliver data on the parasitoids' efficiency in controlling *D. suzukii* in berry fruit cultivation.

Results from the described research project should lead to the definition of a biological control agent for mass production and detailed instructions to successfully control *D*. *suzukii* in organic and integrated berry fruit cultivation under Middle European climatic conditions.

Material and Methods

Insect material

Adult *D. suzukii* flies were kept in a BugDorm cage and were allowed to lay eggs in plastic boxes containing an artificial diet, thus providing food for successful larval development. The company Katz Biotech AG (Berlin, Germany) provided the parasitoid wasps *P. vindemiae* and *T. drosophilae*. Parasitoid populations were reared in BugDorm cages containing *D. suzukii* pupae. After the wasps' emergence, adults were fed with honey.

The side-effects of organic plant protection products on TD and PV lifespan and parasitization rate

The side effects of two organic insecticides (SpinTor™ a.i. Spinosad, Neudosan Neu® a.i. potassium soap), two organic fungicides (VitiSan® a.i. potassium hydrogen carbonate, Kumulus WG® a.i. sulfur), and one acaricide (Eradicoat® a.i. maltodextrin) were studied in laboratory conditions. For this purpose, 250 ml of each product was prepared with the specified or, in the case of a specified range, with the maximum water application rate. Tap water was used as a control. Using a SprayLab, the plant protection products were sprayed onto the top and bottom of plastic petri dishes (5.5 cm diameter) (pressure: 2.75 bar; traveling speed: 1.75 km/h). The parasitoids were placed in the petri dishes for a treatment exposure of three hours. Five replicates with 5-6 pairs of one species were used to observe the life span after contact with the fresh spray coating or a 22 h dried coating. Then, parasitoids were placed in a glass vial for 10-11 days to evaluate the mortality at 22°C, 80% RH, and 16:8 L:D. In addition, four replicates were carried out with 10 females of each species, individually placed with 10 *D. suzukii* pupae each for 24 h after contact with the dried spray for three hours to determine a possible negative influence on parasitization rate.

Dispersal of Trichopria drosophilae in protected cultivation

In 2022, a field trial was established in a commercial berry farm in Wiesbaden, Germany, on a plot measuring 130 by 16 meters and completely enclosed with a net. This plot consisted of five rows of raspberries and one row of blackberries (grown in pots). Within the central row of raspberries, TD parasitoids were released at three locations and on three separate dates (September 6th, 13th, and 21st). Each release involved 500 parasitoids. Bait stations, comprising raspberries enclosed in cups with lids to prevent *D. suzukii* entry, were hung at 5-meter intervals along the same row or at similar heights in adjacent rows (with row spacing of 2.80 meters). These stations were set up one week prior to the first release and subsequently on each release date. The raspberries used in the bait stations were harvested from the same area the previous week and stored at 4°C. Each raspberry bait was filled with

five L3 larvae of *D. suzukii* taken from laboratory breeding. The larvae pupated when the bait stations were deployed. After two days in the field, the bait stations were removed, and the pupae were dissected from the berries. The dissected pupae were incubated for six weeks at 23°C and at 70% relative humidity. Following incubation hatched insects and the developmental stages of both *D. suzukii* and parasitoids within the pupae were identified and counted.

Results and Discussion

An assessment of side effects of organic plant protection products revealed that the insecticide Spintor™ and the acaricide Ecaricoat® showed a strong negative effect on the lifespan of both TD and PV when freshly applied as a spray treatment. Same holds true for freshly applied Neudosan Neu® and contact mortality of TD. Contact to a dry coating of Spintor™ resulted in significantly increased mortality of both parasitoids and a significantly lower parasitization rate of PV females compared to a water control. The tested organic fungicides had no negative impact on lifespan and parasitization rate.

Our study of the dispersal of TD parasitoids in the protected berry cultivation systems demonstrated that TD could move up to 15 meters along the same row of raspberries to parasitize SWD pupae present in bait stations. Also, TD could move through the adjacent rows to parasitize the pupae in the bait stations two rows further on. Similar observations were made in field experiments by Rossi Stacconi et al., 2018, as they observed the same patterns of TD dispersal towards multiple directions from the release point up to 40 m away from it. As several environmental factors, such as the main wind direction, the temperature and the humidity gradients, can influence the preferential parasitoid pathways in field or under semi-field conditions, these factors should be included in studies optimizing the release of TD for biological control purposes.

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