EFFICIENCY OF ATTRACTORS AND TRAPS FOR CAPTURE OF SPOTTED-WING DROSOPHILA IN VINEYARDS¹

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ABSTRACT - Pest monitoring is a practice that enables the estimation of populational density of insect-pests in crops. However, it requires the use of attractors and traps that present high efficiency. The objective of this work was to evaluate the efficiency of food attractors using different trap models for the capture of spottedwing drosophila (*Drosophila suzukii*) insects, and the capture selectivity of these models in vineyards of Cabernet Sauvignon. The experiment was conducted in the 2018-2019 crop season, in São Joaquim, SC, Brazil. A randomized block design was used, in a 2×4 factorial arrangement consisted of two types of traps (PET bottle and plastic container), three attractors (SuzukiTrap[®], Droskidrink, and Yeast), and distilled water as a control. The traps were distributed in a minimum distance of 15 m and fixed in the middle third of the plants. The collections and replacing of attractors were done with 21-day intervals. The data were analyzed through generalized linear models, using the Poisson distribution or Poisson with a dispersion parameter. All attractors tested captured adult *D. suzukii* insects. No significant differences were found between the trap types used. The findings of the present work showed that the trap plastic container with the Droskidrink attractor is the most suitable capture method to be used in monitoring programs for *D. suzukii* insects in commercial vineyards.

Keywords: Drosophila suzukii. Integrated pest management. Selectivity. Food attractors. Cabernet Sauvignon.

EFICÁCIA DE ATRATIVOS E ARMADILHAS NA CAPTURA DE DROSOFILA-DA-ASA-MANCHADA NA CULTURA DA VIDEIRA

RESUMO - O monitoramento é uma prática que permite estimar a densidade populacional dos insetos-praga nos cultivos. Entretanto, torna-se necessário o uso de atrativos e armadilhas que apresentem elevada eficiência. Objetivo deste trabalho foi avaliar a eficácia de atrativos alimentares em diferentes modelos de armadilhas para a captura de *Drosophila suzukii* e a seletividade de captura desses conjuntos em vinhedos da cultivar Cabernet Sauvignon. O experimento foi conduzido durante a safra 2018/2019, no município de São Joaquim, SC. Utilizou-se o delineamento casualizado em bloco no esquema fatorial 2x4, com dois tipos de armadilhas (garrafa PET e pote plástico) e três atrativos (SuzukiTrap[®], Droskidrink e levedura) e água destilada como testemunha. As armadilhas foram distribuídas num distanciamento mínimo de 15 m e fixadas no terço médio das plantas. As coletas e reposição dos atrativos foram realizadas com intervalo de 21 dias. Os dados foram analisados por meio de modelos lineares generalizados, utilizando a distribuição de Poisson ou Poisson com parâmetro de dispersão. Todos os atrativos testados capturaram adultos de *D. suzukii*. Não foi identificado efeito significativo entre o tipo de armadilha utilizado. A partir de todas as informações obtidas nesse trabalho, o uso de armadilha pote plástico com atrativo Droskidrink é o mais recomendável ao uso em programas de monitoramento de *D. suzukii* em vinhedos comerciais.

Palavras-Chave: *Drosophila suzukii*. Manejo integrado de pragas. Seletividade. Atrativos alimentares. Cabernet Sauvignon.

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INTRODUCTION

Spotted-wing drosophila insects (*Drosophila suzukii* (Matsumura, 1931) (Diptera: Drosophilidae)) were identified in 2015 in the region of southern highlands of the state of Santa Catarina, Brazil, and have been caused damages to vine fruits (*Vitis vinifera*) (ARIOLI, BOTTON; BERNARDI, 2015). The South region of Brazil encompasses 90% of the country's grape crop area and production and has thermal indexes that make it favorable for the development of *D. suzukii* (BENITO; SILVA; SANTOS, 2016).

Grape is not a primary host for *D. suzukii*, but larvae of this species have already been found feeding on wine grapes, causing damages by consuming the fruit pulp. In addition, they favor the infection of fruits by diseases and, consequently, the attraction of other animals throughout the crop cycle (ATALLAH et al., 2015; IORIATTI et al., 2015). These losses affect the marketing and quality of fruits, which present low shelf-life time. Damages caused by *D. suzukii* in grapes also affect the quality of the wort, which is the raw material to produce juices and fine wines (IORIATTI et al., 2015; ARIOLI; BOTTON; BERNARDI, 2015).

Pest monitoring is a strategy used for the management of fruit flies (Diptera: Tephritidae) that can be used for management of D. suzukii insects by assisting winemakers in the detection of the pest and, consequently, in the definition of a better time for the pest control. Traps and food attractors may present different efficiency levels to attract and capture insect-pests of the order Diptera in orchards with several species and varieties of fruit trees (AZEVEDO et al., 2012). Their specificity is important to correctly quantify the pest populational density in the field (FREWIN et al., 2017) and make the screening. visualization, and correct identification of captured insects easier (IGLESIAS; NYOIKE; LIBURD, 2014; BURRACK et al., 2015; FREWIN et al., 2017). In addition, choosing specific food attractors and traps for the capture of insectpests is required to prevent beneficial non-target insects from being affected, consequently, causing an ecological imbalance.

The use of baited traps with volatile compounds that release attractive odors to D. suzukii insects can be an efficient tool, since flies of the family Drosophilidae are attracted by substances under fermentation (LANDOLT et al., 2011; ALNAJJAR; COLLINS; DRUMMOND, 2017; KARAGEORGI et al., 2017; CLOONAN et al., Yeast-based attractors 2018). have been recommended for the monitoring of D. suzukii insects in strawberry and raspberry orchards in the South of Brazil (SANTOS, 2014; SANTOS et al., 2016). Homemade formulations containing apple vinegar, wine and/or sugar have been also widely studied and recommended in European and

American countries for several fruit tree orchards (LANDOLT et al., 2011; GRASSI et al., 2014; BERNARDI et al., 2015). However, synthetic ready-to-use attractors have been developed by commercial companies, such as Suzukii Trap[®] (Bioibérica S.A., Barcelona, Spain), which is a specific compound made of peptides and organic acids for the capture of *D. suzukii* insects.

Attraction of adult *D. suzukii* insects to traps presenting dark colors, such as red, black, and purple has been observed, since this pest can separate the spectral composition of a stimulus from its general intensity (BASOALTO; HILTON; KNIGHT, 2013; KIRKPATRICK et al., 2015; CAHENZLI et al, 2018). In addition, several trap models to capture *D. suzukii* insects had been tested in Europe and North America, including plastic cups and traps whose entries are made of screens with different meshes (KANZAWA, 1939; WU et al., 2007; LEE et al., 2012). However, no study in grape vineyards is found for the high-altitude conditions of the South of Brazil.

Considering the importance of *D. suzukii* and its potential damages to the viticulture sector in different producing regions and the scarcity of technical information for the Brazilian conditions, researches that seek efficient strategies for its management are essential. In this context, the objective of this work was to evaluate the efficiency of food attractors using different trap models for the capture of adult *D. suzukii* insects, and the capture selectivity of these models in vineyards of Cabernet Sauvignon in the southern highlands of the state of Santa Catarina, Brazil.

MATERIAL AND METHODS

The experiment was conducted during the 2018-2019 crop season in vineyards of Cabernet Sauvignon with approximately 3.78 ha and 19 years of implementation, in the municipality of São Joaquim, in the southern highlands of the state of Santa Catarina, Brazil (28°14'12.81"S, 50° 4'20.84"W, and altitude of 1400 m). A randomized block design was used, in a 2×4 factorial arrangement consisted of two types of traps, three attractors, and a control treatment containing only distilled water. Four replications were used for each trap-attractor combination. The traps were distributed with a minimum distance of 15 m, as recommended by Mazzetto et al. (2015), and fixed in the middle third of the plants (1.6 m above ground level).

The traps tested were: a) 500-mL transparent PET bottles; each bottle had three holes of 1 cm diameter in its middle third, which is commonly used for capture of flies of the family Tephritidae in the South of Brazil; and b) 1030-mL plastic containers (10 cm diameter \times 14 cm height); 27 holes of 0.2 cm diameter were distributed in three groups of nine holes in the upper third of the container, in which a red band was placed in its middle third.

The PET and plastic container of each trap type were filled with 100 mL and 150 mL of the attractor solution, respectively. The food attractors tested were: a) Suzukii Trap[®] (Bioibérica S.A., Barcelona, Spain) with no dilution; b) Droskidrink (one part of red wine, three parts of apple vinegar, and 20 g L⁻¹ of sugar) (GRASSI et al., 2014); c) Yeast (20 g of biological yeast, 50 g of sugar, and 1000 mL of water); d) distilled water (control).

The attractor was replaced at the time of collections of captured insects, which occurred at two times: 1) end of the grape maturation (21 days after implementation), and 2) at the grape harvest (21 days after the first evaluation). The insects captured were placed in 80-mL bottles containing 70% alcohol and sent to a laboratory for screening, identification, and sex determination. Adult *D. suzukii* insects were identified and other insects were quantified and classified to the order level.

Adult *D. suzukii* insects were identified based on their external morphology and analysis of genitalia, using specific dichotomous keys for *D. suzukii* (VLACH, 2010). Males present a dark spot in the wings and pecten pairs in the tibias of anterior legs; whereas females present a doubly serrated ovipositor. These observations were carried out with the aid of a binocular stereoscopic at $5 \times$ magnification (Eyemag, Carl-Zeiss).

The data were analyzed through generalized linear models, using the Poisson distribution or Poisson with a dispersion parameter. In the cases of significant effects, the means were compared using contrasts. The analyses were carried out using the statistical program R (R Core Team, 2019), considering a 5% significance level.

RESULTS AND DISCUSSION

The attractors and traps had no effect for total number of adult insects of the specie Drosophila suzukii captured, nor for the males captured (Table 1). The attractors had effect for the females captured (Table 1). The capture of insects of the order Diptera was affected by the attractor, with no effect of the interaction between attractor and trap (Table 1). The capture of insects of the order Coleoptera was affected by the interaction between attractor and trap, whereas the capture of insects of the order Hymenoptera was affected by the attractors and traps independently (Table 1). Insects of the order Lepidoptera were not captured in the plastic container due to the size of the entry hole, thus, the comparison between attractors was possible only for the PET bottles.

Table 1. Descriptive level (p-value) of the test of effects of the sources of variation.

Source of Variation	Dr	Drosophila suzukii			Insect Order		
	Male	Female	Total	Diptera	Coleoptera	Hymenoptera	
Block	0.7437	0.1385	0.3485	0.1631	< 0.0001	0.0081	
Trap (T)	0.1694	0.5821	0.5645	0.4831	0.2547	0.0012	
Attractor (A)	0.2312	0.0307	0.8922	< 0.0001	< 0.0001	0.0002	
$T \times A$ interaction	0.1717	0.8458	0.4177	0.9015	0.0115	0.5551	

The control treatment (water) was not used in the analysis because of absence of insects captured.

The use of the Droskidrink attractor resulted in the highest capture of dipterans, regardless of the trap used. The number of Hymenoptera insects captured were greater when using the Yeast attractor, regardless of the trap used. The plastic container resulted in smaller number of Hymenoptera insects, which is desirable, since they are non-target insects in integrated pest management programs (Table 2). Any physical or chemical barrier that prevents the capture of non-target insects is desirable because such captures complicate the trap inspection activity.

The number of males and total adult *D.* suzukii insects captured showed no significant differences between the attractors tested (Table 2). The attractors presented different number of females captured; the Suzukii Trap[®] attractor presented significantly lower capture than the Droskidrink (Table 2).

 Table 2. Mean (± standard error) number of adult *Drosophila suzukii* insects and insects of the orders Diptera and Hymenoptera captured with different food attractors in vineyards of Cabernet Sauvignon.

Attractor -	_	Drosophila suzukii	Diptera	Hymenoptera	
	Male	Female	Total	Total	Total
Suzukii Trap®	100.1±26.6 ^{ns}	89.0±17.9 b	189.1±38.9 ^{ns}	433.5±45.9 b	3.2±1.0 c
Droskidrink	137.0±16.5	157.8±6.9 a	294.8±22.4	804.0±45.3 a	7.5±1.9 b
Yeast	87.7 ±17.5	117.8±21.9 ab	205.6±36.6	732.5±157.1 ab	16.2±4.3 a
Water	0.0 ± 0.0	$0.0{\pm}0.0$	$0.0{\pm}0.0$	0.5±0.5 c	0.0 ± 0.0

Means followed by the same letter are not different from each other by the Tukey's test ($p \le 0.05$).

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The combination of the plastic container trap with the Droskidrink attractor resulted in greater number of insects captured for the order Coleoptera (Table 3). Insects of the order Lepidoptera were significantly more attracted by the Yeast attractor (Table 3). Significant captures were found when using the SuzukiTrap[®] and Droskidrink attractors, which differed from the control treatment (Table 3).

Table 3. Mean (± standard error) number of insects of the orders Coleoptera and Lepidoptera captured with different food attractors in vineyards of Cabernet Sauvignon.

Attractor	Со	leoptera	Lepidoptera
	PET bottle *	Plastic container **	PET bottle
SuzukiTrap®	3.0±1.1 b A	2.0±,40 c A	0.8±0.6 b
Droskidrink	12.7±3.6 a B	21.0±5.2 a A	0.8±0.7 b
Yeast	13.2±3.7 a A	10.5±1.6 b A	2.6±1.2 a
Water	0.0±0.0	0.0±0.0	$0.0{\pm}0.0$

Means followed by same lowercase letter in the columns, or uppercase letter in the rows, are not different from each other by the Tukey's test ($p \le 0.05$). * 500 mL PET bottle with six 1-cm holes in the upper third. ** plastic container with 27 0.3-cm holes in the upper third and a red band in the middle third.

Iglesias, Nyoike, and Liburd (2014) evaluated the efficiency of captures of *D. suzukii* insects in blueberry and blackberry orchards and found higher performance for yeast baits when compared to apple vinegar and wine baits. Contrastingly, the Droskidrink attractor presented similar performance to that of the Yeast attractor in capturing adult *D. suzukii* insects in the vineyards of Cabernet Sauvignon evaluated in the present work.

Significant capture of adult *D. suzukii* using the Droskidrink attractor was also found by Padilha et al. (2016), who reported better performance for this attractor when compared to the CeraTrap[®], Torula[®], and BioAnastrepha[®] attractors, which are commonly used in Brazil to capture flies of the family Tephritidae, but, are not as efficient for capturing *D. suzukii* insects. Wollmann et al. (2019) evaluated commercial orchards of blackberry (*Rubus* sp.) in the state of Rio Grande do Sul, Brazil, and found high attractiveness of the attractor based on wine, apple vinegar, and sugar, with similar proportions to the Droskidrink attractor, to adult *D. suzukii* insects.

The Droskidrink formula was developed by Grassi et al. (2014), who found a higher performance for this attractor when using red traps and indicated that the Droskidrink attractor is significant superior to other two commercial products evaluated. Iglesias, Nyoike and Liburd (2014) also found best results for the Droskidrink attractor when compared to the Suzukii Trap[®] attractor, and reported that baits containing vinegar and wine are attractor that act as excellent preservatives of the collected organisms, thus favoring the laboratorial identification of the insects.

Harmon et al. (2019) evaluated blueberry and blackberry orchards in Florida, USA, and found lower capture indexes for male and female *D. suzukii* insects using the Suzukii Trap[®] attractor, compared to the other attractor tested, regardless of the trap

used. In the present study, the Droskidrink had better performance in capturing females than the Suzukii Trap[®] attractor, which can be due to the release of volatile compounds that are attractive to flies, since Drosophilidae insects are attracted by acid acetic or substrates containing acid acetic for oviposition purposes (JOSEPH et al., 2009). A higher capture of females is desirable, since they are the ones that cause damages to fruits.

According to Harmon et al. (2019), the evaporation of the product is another disadvantage of the attractor Suzukii Trap[®], which turns into a syrup-type thick brown residue within a week. The fast evaporation and formation of a consistent gelatinous residue can probably hinder the capture of insects by using this attractor, which would facilitate their escape from the trap.

The Yeast attractor did not differ from the other attractors tested for the capture of *D. suzukii* insects, despite presenting a trend of lower capture of males (Table 2). A negative aspect of the Yeast attractor was the great difficulty of sampling at the time of collection and evaluation; despite it presented no syrup consistency, it formed a sticky residue throughout the 21 days of exposure in the field, which hindered the visualization of insects and, mainly, the cleaning of the traps. Similar situation was observed by Iglesias, Nyoike, and Liburd (2014), who reported that the yeast baits turned into a high turbidity and sedimented substance over time, making the identification of insects difficult.

Santos et al. (2016) evaluated capture of *D. suzukii* insects in raspberry orchards and found selectivity for the Yeast attractor, which was used single or combined with a commercial mixture, and found higher indexes of capture of adult *D. suzukii* in blackberry orchards when using the trap containing larger hole diameter (7 mm). In the present study, the traps used had different entry sizes (1 and 0.2 cm), but presented no statistical difference for the capture

of adult D. suzukii insects.

Lee et al. (2012) evaluated types and models of traps and found no difference between traps (plastic container) with red bands and transparent traps (PET bottle). However, Renkema, Buitenhuis and Hallett (2014) report that, despite not significantly different, traps with red and black bands tend to capture a greater number of adult *D. suzukii* than light colors traps. The difference between colors of traps is observed when all other physical characteristics are equal (LEE et al., 2013); in the present study, the traps were physically different.

Traps with dark, red, and black colors were attractive to *D. suzukii* in orchards with dark color fruits (RENKEMA; BUITENHUIS; HALLETT, 2014) and in orchards with mature or post-harvest fruits, presenting higher indexes of capture of *D. suzukii* when compared to yellow and transparent traps (LEE et al. 2013). The vineyard of Cabernet Sauvignon used in the present study was between the pre-harvest and harvest periods, with grape berries already presenting dark colors, however, this result was not found, since no significant difference in capture was found between the traps tested.

Other factors that can affect the capture success are the attractor surface area, trap size, and position of entry holes (RENKEMA; BUITENHUIS; HALLETT, 2014). Santos et al. (2019) found higher performance for traps of the Brazilian transparent model for the capture of adult *D. suzukii* insects when compared to the other models tested, and they reported that the performance of this model can be due to the location of the holes (lower third) and the convex shape of the bottle used, which hinder the escape of insects due to phototropism. This model is similar to the PET bottle used in the present study, differing only in location and size of entry holes.

According to Sarto and Sorribas (2011), and Iglesias, Nyoike and Liburd (2014), the type of bait is the main factor for the attraction of D. suzukii insects and not the type and color of the trap. Therefore, Iglesias, Nyoike and Liburd (2014) recommend the use of plastic cup traps, since they are more practical and require less time to build and less changes. Lee et al. (2013) reported that traps should be easy of obtaining, development, maintenance, placing in the plant canopy, handling, and storing by the farmer to facilitate their use in integrated pest managements. Considering only the capture of D. suzukii insects by an attractor and its interaction with the different trap types, the results of the present work indicate that winemakers have alternatives to start the monitoring of this pest in vinevards; however, some issues should be considered when choosing the attractor to be used.

Regarding the selectivity, the Yeast attractor captured large number of insects of the orders Coleoptera, Lepidoptera, and Hymenoptera; according to Azevedo et al. (2012), it can be related to the odor that the yeast releases. The selectivity of the plastic container traps was higher, with no capture of Lepidoptera and low capture of Hymenoptera insects. Selectivity in the capture of insects, such as pollinators and parasitoid species, is essential for fruit production to keep the ecological dynamics and balance in the orchards. The selectivity of the plastic container traps for the capture of Lepidoptera species can be related to the diameter of the entry hole, which was smaller than that of the PET bottle traps.

The recommendation of an attractor with high efficiency depends on the purpose. The use of attractors that present high indexes of capture of adult insects is interesting for the monitoring of insect-pests by presenting accurate data of populational density and high capture selectivity. The Droskidrink attractor presented these characteristics, with good capture of female *D. suzukii*, which is important because they are responsible for damages in fruits, making their control essential.

The use of efficient traps and more selective attractor compounds for the monitoring of insectpests is essential to ensure their capture and monitor the pest fluctuation in commercial vineyards. Further studies are needed to evaluated the number of traps and durability of attractors under field conditions. This information may provide monitoring systems with higher reliability for winemakers regarding the management of *D. suzukii* insects in commercial vineyards.

CONCLUSION

The Droskidrink, Suzukii Trap[®], and Yeast attractors were efficient for the capture of adult *Drosophila suzukii* insects in vineyards of Cabernet Sauvignon.

The Droskidrink attractor captured a greater number of female *D. suzukii* insects.

The traps tested presented no significant difference for total capture of adult *D. suzukii* insects.

The use of plastic container traps with Droskidrink attractor is the most suitable capture method to be used in monitoring programs for *D. suzukii* insects in commercial vineyards.

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