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***Drosophila suzukii* population dynamics in vineyards and wine cultivars susceptibility**

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Abstract: Grapes are not considered a good host for the spotted-wing drosophila, *Drosophila suzukii*. However, its damage results in qualitative and quantitative losses to the wine grapes berries. Knowing the *D. suzukii* population dynamics in vineyards and surrounding areas, as well as the wine cultivars susceptibility, can help winegrowers to carry out the management. The objective of this study was evaluate the *D. suzukii* population dynamics in the field, on the forest border present around the vineyard and in its interior and evaluate the susceptibility of the main wine cultivars for São Joaquim municipality to the attack of *D. suzukii* in laboratory. In the vineyard, *D. suzukii* was monitored from November 2018 to May 2021 with weekly inspections. Plastic pot traps with a red stripe baited with the Droskidrink attractant were used, distributed among the vines and around the vineyard. Monitoring data were correlated with regional climate variables. In laboratory, through non-choice and free-choice test, the susceptibility to *D. suzukii* damage was evaluated in Chardonnay, Sauvignon Blanc, Sangiovese, Cabernet Franc, Cabernet Sauvignon and Merlot berries. Intact and artificially injured berries were offered to adults in plastic screened cages for 24h. Afterwards, the number of punctures, eggs, emerged adults and the period of egg-adult development were evaluated. The data were analyzed in the R environment. Through monitoring, it was found that the presence of *D. suzukii* adults was higher in the forest border (762) in the surroundings compared to inside of the vineyard (128). The precipitation reduced the population of *D. suzukii* inside the vineyard, while the average and maximum temperatures promoted an increase in the population. In free choice tests, the cultivar Sangiovese was susceptible to the attack and development of *D. suzukii*. The cultivar Cabernet Sauvignon showed reduced egg-adult development time compared to the other tested cultivars in no-choice test.

Index terms: monitoring, spotted-wing-drosophila, *Vitis vinifera*.

Dinâmica populacional de *Drosophila suzukii* em vinhedo e susceptibilidade de cultivares viníferas

Resumo: A uva não é considerada hospedeiro primário para a drosófila-da-asa-manchada, *Drosophila suzukii*. Contudo, seus danos resultam em perdas qualitativas e quantitativas das bagas das uvas viníferas. Conhecer a dinâmica populacional da *D. suzukii* nos vinhedos e nas áreas circundantes, bem como a susceptibilidade das cultivares viníferas, pode auxiliar os viti-vinicultores na tomada de decisão sobre o manejo. O objetivo deste estudo a campo foi avaliar a dinâmica populacional de *D. suzukii* nas bordas de mata nativa presentes no entorno do vinhedo e em seu interior, e avaliar a suscetibilidade das principais cultivares viníferas para o município de São Joaquim, ao ataque de *D. suzukii* em laboratório. No vinhedo, foi realizado o monitoramento de *D. suzukii*, de novembro de 2018 a maio de 2021, com vistorias semanais. Utilizou-se de armadilhas de pote plástico com faixa vermelha iscadas com o atrativo Droskidrink, distribuídas entre as vinhas e no entorno do vinhedo. Os dados de monitoramento foram correlacionados com variáveis climáticas regionais. Em laboratório, através de teste com e sem chance de escolha, avaliou-se a suscetibilidade aos danos de *D. suzukii* às bagas das cultivares Chardonnay, Sauvignon Blanc, Sangiovese, Cabernet Franc, Cabernet Sauvignon e Merlot. Bagas intactas e com injúria artificializada foram ofertadas a adultos em gaiolas plásticas teladas, durante 24h. Após, avaliaram-se o número de puncturas, de ovos, de adultos emergidos e o período de desenvolvimento ovo-adulto. Os dados foram analisados no ambiente R. Através do monitoramento, verificou-se que a presença de adultos de *D. suzukii* foi superior nas bordas de mata nativa (762), no entorno, em comparação ao interior do vinhedo (128). A pluviosidade reduziu a população de *D. suzukii* no vinhedo, ao passo que as temperaturas média e máxima promoveram o aumento da população. Em testes de livre escolha, a cultivar Sangiovese apresentou-se suscetível ao ataque e ao desenvolvimento de *D. suzukii*. Em testes sem chance de escolha, a cultivar Cabernet Sauvignon apresentou tempo de desenvolvimento ovo-adulto reduzido, frente às demais cultivares testadas.

Termos para indexação: monitoramento, drosófila-da-asa-manchada, *Vitis vinifera*.

Introduction

The Brazilian winegrowing has been highlighting due to the internal consumption and export of wines and sparkling wines, resulting from the singular 'terroir' of the regions where the grapevines are cultivated (MELLO; MACHADO, 2020). Brazil main winegrowing region is from the temperate climate, traditional in the South and in the Southeast altitude regions (São Paulo and Minas Gerais). The temperate climate winegrowing is characterized by an annual cycle, followed by a dormancy period induced by low winter temperatures (CAMARGO et al., 2011). Within this winegrowing region, São Joaquim mu-

nicipality is located, in Santa Catarina highland region (CAMARGO et al., 2011), center of one of the country's youngest productive poles. In the year of 2021, Santa Catarina highland region, which is considered the coldest and highest altitude of the south of Brazil, obtained the seal of "Geographical Indication of Santa Catarina Altitude Wines", bringing prominence at national level and aggregating value to the wines produced in this region.

The hygiene of the vines as well as the quality of the grape bunches is important to ensure high production and the wine's quality. The *Drosophila suzukii* species (Matsumura, 1931) (Diptera: Drosophilidae) has recently

been detached for causing damage to the vine fruits, compromising the production and the wine's quality produced in the Santa Catarina highland region. Originally from Japan, *D. suzukii* is a polyphagous quarantine pest, popularly known as the Spotted Wing *Drosophila* – SWD (BERRY, 2012) and is in constant global expansion. Since 2008, it has caused significant economic damage in small fruits and fruits with soft skin, in European and North American countries (HAUSER et al., 2009; GOODHUE et al., 2011; CALABRIA et al., 2012; SCHLESENER et al., 2015). In Brazil, *D. suzukii* was identified initially occurring in the Southern and Southeast states (RAMIREZ et al., 2013; DEPRÁ et al., 2014), causing damage in strawberry, blueberry, peach, plum and feijoa (SANTOS, 2014; NUNES et al., 2014; GEISLER et al., 2015; SOUZA et al., 2017).

D. suzukii male adults are small (3 mm), being identified mainly by the characteristic black spot on their wings. The females do not present spots on the wings and measure 4 mm in length. They present serrated ovipositor, which differentiates them from the other *Drosophila* species (SCHLESENER et al., 2015; BERNARDI et al., 2015). *D. suzukii* is a polyphagous pest that has a wide range of hosts (WALSH et al., 2011). Its serrated ovipositor enables the endophytic posture in fresh fruits and soft skin that still are in the pre-harvest period (MITSUI et al., 2010; HAUSER, 2011).

In the grape berries, the damage results both by the endophytic oviposition puncture, as by the larvae that consume the pulp (KANZAWA, 1939, IORIATTI et al., 2015). Recent studies have shown that in white table grapes ('Thompson'), *D. suzukii* has reduced oviposition, elongated biological cycle and lower survival, compared to the larvae that develop in other fruits, such as raspberries and cherries (LEE et al., 2011). Although both table and wine grapes are not considered preferred hosts, Bellamy et al., (2013) and Wang et al., (2019) reported that *D. suzukii* can complete the life cycle in table and wine grapes berries.

In wine grapes, *D. suzukii* damage present negative impact such as berry crack, the penetration of pathogenic agents, the attraction of other animals during the harvest period due to juice extravasation, besides the presence of larvae in the must during processing (ATALLAH et al., 2015; IORIATTI et al., 2015). Also, studies by Barata et al., (2012) and Ioriatti et al., (2018) showed that *D. suzukii* has the ability to disseminate phytopathogenic agents that cause sour rot epidemics or acid rot in vineyards, which depreciate the chemical composition and the quality of the must and wine.

The phenological cycle duration can influence the susceptibility of the wine grapes to *D. suzukii* damage. Late cycle cultivars can be at greater risk of *D. suzukii* oviposition since the populations in the end of the summer are higher than in the beginning (SHRADER et al., 2018). The intrinsic physicochemical characteristics of each grape cultivar, also contribute to the susceptibility to *D. suzukii* attack. Recent studies in the laboratory reveal that the skin hardness and the soluble solids content influence the *D. suzukii* oviposition indices (SHRADER et al., 2018; BASER et al., 2018; TONINA et al., 2020).

The climatic conditions characteristic of Brazil temperate climate region also favor the development of spotted wing *Drosophila*, so much which has also been identified in the vineyards of São Joaquim, Santa Catarina, Brazil (PADILHA et al.; 2016; BENITO et al., 2016). Because it is a recent pest in the Southern region, as in Brazil itself, there is lack of technical information that assist the farmer in the *D. suzukii* control. Some preliminary studies were performed as the susceptibility of grape cultivars in Rio Grande do Sul (ANDREAZZA et al., 2016a) and food attractions for the capture of *D. suzukii* in commercial vineyards in Santa Catarina, Brazil (PADILHA et al., 2016). Due to the importance, there is the need to perform new researches, which together, can assist in the organization of an efficient management plan for the spotted wing *Drosophila*.

In temperate climate vineyards of Santa Catarina highland area in the south of Brazil, the *D. suzukii* population dynamics and the susceptibility of the main cultivated wine grapes are unknown. Understanding the role of the non-agricultural habitats in the *D. suzukii* population dynamics, as well as the differences in the preference and susceptibility of the wine grapes, can assist in the optimization of the pest management techniques. Thus, the aim of this study was to evaluate the *D. suzukii* population dynamics in the native forest border present in the vineyard surroundings and in its interior as well as to evaluate the susceptibility of the main vine cultivars for São Joaquim municipality to the *D. suzukii* attack in the laboratory.

Material and Methods

Drosophila suzukii monitoring

The study was carried out from November 2018 to May 2021 in 3.78 ha vineyard (*in vivo* germplasm bank) of the São Joaquim Experimental Station (EESJ), one of the research units of the Santa Catarina Agricultural Research and Rural Extension Company (EESJ – EPAGRI). The vineyard is located in São Joaquim, Santa Catarina, Brazil (1440 m, 28°14'12.81"S 50° 4'20.84"O), it is composed of 54 wine grapes cultivars (*Vitis vinifera*) with approximately 20 years of implantation, conducted in espalier system.

Part of the vineyard surround corresponds to forest composed of native forest, characteristic of the Montana Mixed Ombrophila forest. These (North and South side) are mixed with eucalyptus plantation (*Eucalyptus* spp.) and Cypress (*Cupressus* spp.) used as windbreaker (Figure 1). In the East and West sides is fruit orchards such as apple, pear, feijoa (Figure 1), which are part of the agricultural activity of São Joaquim, Santa Catarina, Brazil.

D. suzukii monitoring was performed through the installation of eight flytraps, four installed in the interior of the vineyard and another four in the forest border. The "flytraps" were composed of a plastic pot of approximately 1,000 ml with lid, containing

in the wall a red band of 17.5 cm. In the middle third of the trap, three groups of 9 holes of 0.2 cm in diameter were located, totaling 27 entry points for the insects. During the monitoring, the traps were filled with 150 mL of Droskidrink food attractiveness (one part of red wine, three parts of apple vinegar and 20g of sugar L⁻¹) (GRASSI et al., 2014). It was added to the attractive solution two drops (approximately 20µl) of neutral detergent to decrease the liquid surface tension and ensure the trapping of the captured insects, according to Schlesener et al. (2015) recommendation.

In the vineyard, the traps containing the attractive were arranged in shaded site in the middle third of the plants (approximately 1.6 m high) randomly respecting the minimum of 15 m away from the border (MAZZETTO et al., 2015; FUNES et al., 2018). The traps installed outside the vineyard were positioned in the north and south points (inside the forest), respecting the minimum distance of 15 m of the vineyard edge. Weekly, the traps were inspected by collecting the captured insects with the aid of a fine mesh sieve. The food attractiveness was replaced during the collection of the insects.

The captured insects were placed in 80 mL plastic containers containing 70% alcohol, properly labeled. Then, this material was forwarded to the Entomology laboratory of the Center for Agroveterinary Sciences at Santa Catarina State University (CAV/UDESC) for the screening and counting of the captured *D. suzukii* male and female adults.

The identification of the *D. suzukii* adults was performed based on the external morphology and genitalia analysis, following the specific key for *D. suzukii* (VLACH, 2010). For the males, it was observed the dark spot in the wings and the pairs of combs in the front legs. In females, a serrated ovipositor was observed. These observations were performed with the aid of a stereoscopic binocular magnifier with 5x of increase (Eyemag, Carl-Zeiss). After the screening and identification, *D. suzukii* adults were separated by sex and computed for each trap.



Figure 1. Satellite image locating the experimental vineyard at the São Joaquim Experimental Station of the Santa Catarina Agricultural Research and Rural Extension Company, and the surrounding plant composition.

Source: Google Earth, 2023.

Susceptibility of wine grape cultivars to *D. suzukii* damage

The bioassays were conducted in the Entomology laboratory of the Center of Agroveterinary Sciences (CAV) at Santa Catarina State University (UDESC), under controlled conditions with a temperature of 25 ± 2 °C, RH $65 \pm 10\%$ and 12 h photophase, following the methodology described by Ioriatti et al. (2015). The insects used in the bioassays were derived from the breeding of *D. suzukii* kept in the laboratory under the same conditions as the test was carried out. *D. suzukii* population was maintained in plastic screened cages (28 x 28 x 28 cm). The adults were fed with artificial diet based on cornmeal (80 g), yeast (40 g), sugar (100 g), agar (8 g), propionic acid (3 ml), Nipagin (8 ml, in a 10% alcohol dilution) and 1 L of distilled water (MATSUBAYASHI et al., 1992) according to Schlesener et al. recommendation. (2017).

In the field, during the phenological stage, bunches of the tested wine grapes were bagged with 'TNT' tissue bags (non-woven fabric) in order to prevent *D. suzukii* natural infestation. In the harvest, a sample of bunches without bagging was collected for the observation of natural infestation. Throughout the grape maturation period, it was not performed insecticides application in the vineyard.

To test the susceptibility of the wine grape cultivars to *D. suzukii* damage were performed three bioassays: i) non-choice with berry without artificial damage; ii) non-choice with berry with artificial damage and iii) free-choice with berry without artificial damage. In all the bioassays, the insects were deprived of artificial diet for a period of 12h before the release.

Bioassay non-choice – The grape cultivars selected for the bioassays non-choice were

Chardonnay and Sauvignon Blanc (white cultivars) Sangiovese, Cabernet Franc and Cabernet Sauvignon (red cultivars). The experimental units of the tests non-choice consisted of transparent plastic pots (500 mL), containing hollow bottom and closed with voile tissue and capsized in a plastic cover (11 cm ø) lined with filter paper.

For the bioassay non-choice using berries without damage, ten intact grape berries of the respective varieties were offered in each cage containing four *D. suzukii* females and one male. The bioassay was performed in a completely randomized design, with ten repetitions per treatment. For the bioassay non-choice using berries with artificial damage, six berries were used with a cut of approximately 1cm by a sterilized scalpel in 99% ethanol. The bioassay was performed in a completely randomized design, with six repetitions per treatment.

Bioassays free-choice – eight *D. suzukii* females and two males and six berries of each cultivar, composing the experimental unit that consisted of plastic cage (28 x 28 x 28 cm). The grape cultivars tested were Chardonnay, Sauvignon Blanc, Cabernet Franc and Sangiovese. The bioassay was performed in a randomized blocks design with six repetitions.

In all the bioassays, *D. suzukii* adults were removed after 24 h of exposure and the punctures and eggs deposited within the berries were counted with the aid of a stereoscopic binocular magnifier with 5x of increase (Eyemag, Carl-Zeiss). The berries containing *D. suzukii* eggs were stored in translucent plastic containers of 7 cm x 5 cm (diameter x height) and sealed with micro perforated cover to allow gas exchange. They were observed daily for the registration of adult emergency for 25 days. We evaluated the embryonic viability, the period of egg-adult development and the sexual ratio. The sexual ratio was calculated using the following formula: number of females/(number of males + number of females) emerged (SILVEIRA-NETO et al., 1976).

Concomitant to the bioassays, the physico-chemical characteristics of the wine grapes cultivars tested were evaluated. For each cultivar was used samples composed of 20 berries. We evaluated the soluble solids content (°Brix), total titratable acidity, penetration resistance and color patterns. The soluble solids content (°Brix) was measured with the aid of a bench portable digital refractometer. The total titratable acidity was obtained by the titration of grape juice sample, diluted in distilled water, and titrated with NaOH 0.1 N solution, with the aid of an automatic titrator. The penetration resistance was determined in newton (N), with the aid of a digital penetrometer, with a tip of 2 mm and penetration of 10 mm, in the equatorial zone of the fruit. The skin background color was determined by an electronic colorimeter, through the realization of readings in the equatorial region of the fruits and expressed in terms of hue angle (h°) (0° = red; 90° = yellow; 180° = green and 270° = blue) and luminosity (L) that refers to the scale that varies from black (0) to white (100).

Climate Variables

The climatic data were collected daily by the meteorological station located in EESJ – EPAGRI, close to the vineyard, compiled and made available by the climate monitoring system of the Environmental Resources and Hydrometeorology Information Center of Santa Catarina (CIRAM) from EPAGRI. The climate variables observed were: maximum, average and minimum temperature daily; total precipitation monthly; and average relative humidity monthly.

Data analysis

The data obtained during the monitoring period were synthesized by descriptive statistics presenting the weekly values of insects captured. For the comparison of the insect number in the forest border and in the vineyard, and for comparison among the locations in each year (native forest edges and interior of the vineyard) it was used the Wilcoxon nonparametric paired test. For comparison among the season, it was used

the unpaired test (MONTGOMERY; RUNGER, 1943). The multiple regression model was used to describe the total number of insects in the forest border and inside the vineyards according to the climate variables. The selection of the explanatory variables in the model was performed by Stepwise method based on the value of the Akaike information criterion (VENABLES; RIPLEY, 2002).

The variance analysis model was used to analyze the cultivars susceptibility to *D. suzukii* attack data. Thus, the assumptions of normality and variance homogeneity were verified, and when one of the assumptions was not met, we used the transformation with Box-Cox optimal lambda (VENABLES; RIPLEY, 2002). All the analyses were performed in the R environment (R Core Team, 2021) considering the 5% level of significance.

Results

D. suzukii monitoring

During the monitoring period of *D. suzukii* in the EESJ-EPAGRI vineyard, the population peaks in the years of 2019 and 2020 occurred among the months of April and May and September. In 2021, the popula-

tion peak occurred in February (Figure 2). Throughout the monitoring period, it was verified that the population of *D. suzukii* in the forest border proved to be superior to the one observed inside the vineyard. Only in February of 2019, it was possible to observe that the population of *D. suzukii* inside the vineyard was superior to the forest border (Figure 2).

The population of males was superior to the females during the largest population peaks observed in each year of monitoring for the forest border and vineyard (Figure 2). However, it was observed that in the population peaks that occurred in September of 2019 and between September and December of 2020, the number of females captured was superior to the number of males in the forest border (Figure 2). In the interior of the vineyards, only in the population peak in September of 2019 the number of females captured exceeded the males in the same way that was observed in the forest border (Figure 2). The largest population peaks of *D. suzukii* inside the vineyard correspond to the period of the wine grapes fruiting (Figure 2).

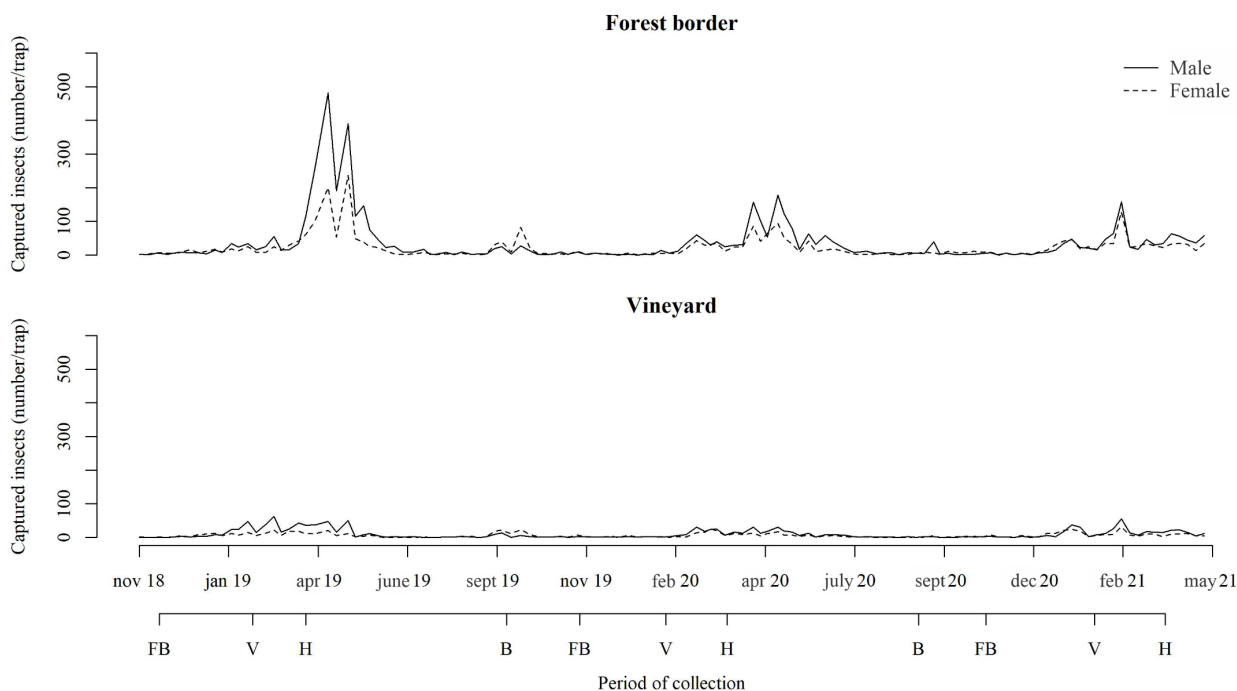


Figure 2. *Drosophila suzukii* population fluctuation inside of the vineyard and in surrounding forest border. São Joaquim, SC, November 2018 to May 2021. Data are weekly catch averages.

FB: full bloom; V: 'veraison' (50%); H: harvest; and B: budburst.

The data obtained through the weekly collections were synthesized according to the grape season, which corresponds to the period of July to June (Table 1). Only in the 2018/19 season, every week, *D. suzukii* adults were captured in the forest border in the surrounding of the vineyards, place where the highest values of *D. suzukii* adults

captures were recorded (Table 1). For inside the vineyard, the higher indices of captures occurred in the 2020/21 harvest (Table 1). In both monitored locations, forest border and inside the vineyard, the 2019/20 season was the one in which the lowest indices of *D. suzukii* adult captures was recorded (Table 1).

Table 1. Descriptive statistics related to the total number of *Drosophila suzukii* adults collected in forest border of the vineyard surroundings and inside of the vineyard, during three monitored season from November of 2018 to May of 2021. São Joaquim, Santa Catarina, Brazil.

	Season	Minimum	Maximum	Average	Median	SD ¹
Forest border	2018/19	3	762	102.50	37.0a	167.44
	2019/20	0	320	046.04	16.5a	063.17
	2020/21	0	402	048.27	28.0a	060.69
	General	0	762	059.05	24.0	96.53
Inside the vineyard	2018/19	0	103	24.26	15.5b	24.80
	2019/20	0	64	13.66	06.0b	15.86
	2020/21	0	128	17.94	10.0b	22.63
	General	0	128	17.81	09.0	21.37

¹ Standard deviation. Medians followed by equal letters, in the season, do not differ according to the Wilcoxon test.

The sex ratio consists of the proportion of females regarding to the total number of *D. suzukii* adults. We observed significant difference among the sex ratio obtained in the forest border and inside of the vineyard only for the 2020/21 season, where the value in the forest border was superior to the vine-

yard (Table 2). When comparing the sex ratio among the season within the collection sites, it was possible to observe significant difference only in the forest border, where the 2020/21 season the sex ratio value was superior and differed only from the 2018/19 season (Table 2).

Table 2. Descriptive statistics and Wilcoxon paired test results for the *Drosophila suzukii* adult sex ratio captured in the forest border of the vineyard surroundings and inside of the vineyard during the period of November of 2018 to May of 2021. São Joaquim, Santa Catarina, Brazil.

Season	Sex ratio (±SD ¹)		Median	Confidence interval limits		p-value
	Forest	Vineyard		Inferior	Superior	
2018/19	0.41±0.19b	0.40±0.25 ^{NS}	0.0019	-0.0583	0.0603	0.9618
2019/20	0.46±0.22ab	0.46±0.25	-0.0178	-0.0707	0.0382	0.5155
2020/21	0.49±0.20a	0.44±0.27	0.0548	0.0020	0.1019	0.0410
General	0.46±0.21	0.44±0.26	0.0172	-0.0143	0.0484	0.2973

¹ Standard deviation. Sex ratio followed by equal letters, in the season, do not differ according to the Wilcoxon test. ^{NS} not significant.

The positive and negative values of the multiple linear regression model estimate, explain how each climate variable influenced the *D. suzukii* population in the forest border and inside the vineyard. The explained variability shows, in percentage, how much of the variability observed in the *D. suzukii* population can be attributed to each variable analyzed considering the sequential model. The ex-

plained variability was of approximately 23% both for the forest border as for the inside of the vineyard (Table 3). This shows that, there is still a percentage of approximately 77% of other factors that influence and affect *D. suzukii* population. Rainfall was the climate variable that negatively influenced *D. suzukii* population both in the forest border as inside of the vineyard (Table 3).

Table 3. Estimates of the multiple linear regression model of the variables evaluated according to climate variables for São Joaquim Experimental Station vineyard of the Agricultural Research Company and Rural Extension of Santa Catarina. São Joaquim, Santa Catarina, Brazil. November of 2018 to May of 2021

Variables evaluated	Climate variables	Estimates ($\pm EP$)	Explained variability (%)	p-value
Forest Border	Maximum temperature	732.44 \pm 75.96	11.61	2.5166x10 ⁻¹⁹
	Rainfall	-000.04 \pm 00.01	12.46	1.7864x10 ⁻¹¹
	Average temperature	192.08 \pm 19.75	15.73	1.3701x10 ⁻¹⁹
Vineyard	Rainfall	-000.01 \pm 00.00	02.43	2.2748x10 ⁻⁰⁸
	Relative humidity	-034.54 \pm 06.90	06.34	9.5972x10 ⁻⁰⁷

Regarding the climate variables, it was observed that in the year of 2019 in June and August the precipitation values were below 50 mm, and May presented precipitation value close to 350 mm (Figure 3). In the year of 2020, March and July presented the lowest and highest value for precipitation, respec-

tively. In the year of 2021, January presented precipitation close to 300 mm and April presented precipitation below 50 mm. The relative humidity oscillated during the period analyzed between 65 and 95%, presenting maximum values in April of 2019 and minimum in August of 2020 (Figure 3).

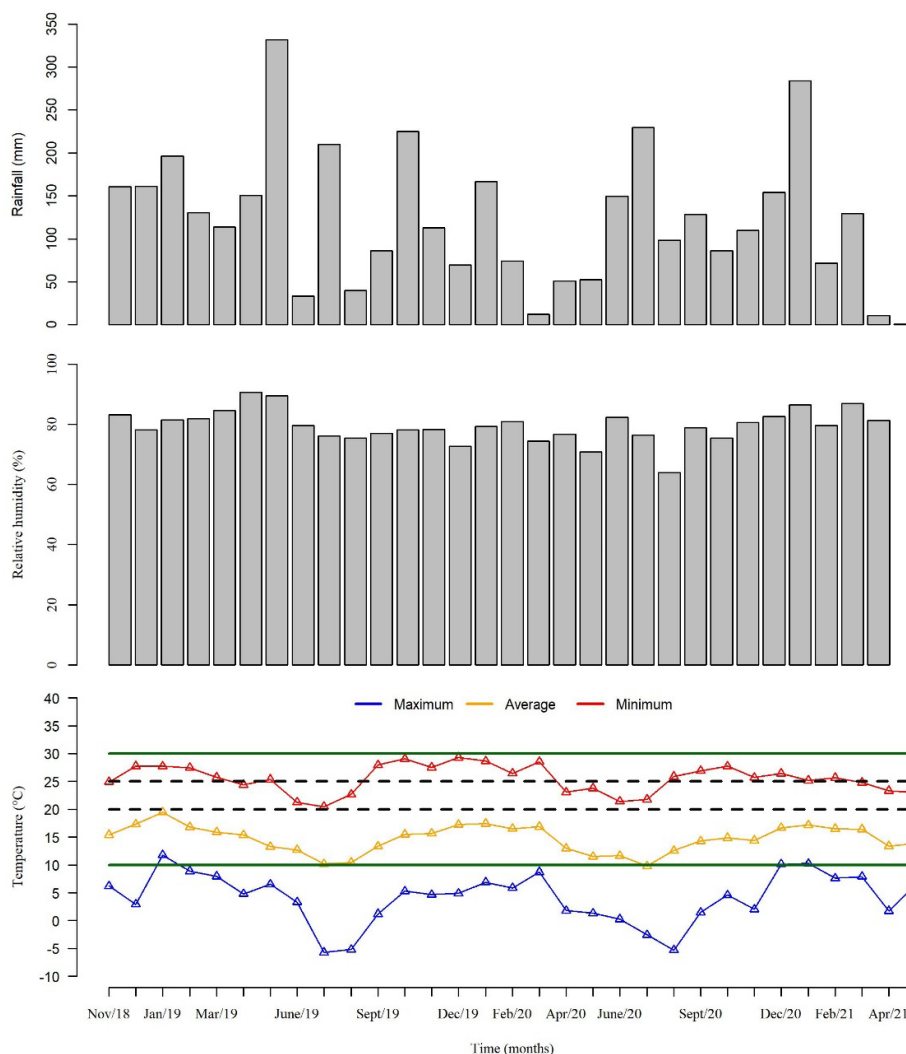


Figure 3. Climate variables during the period from November 2018 to May 2021. São Joaquim, Santa Catarina, Brazil

Observation: The range composed by the dashed lines corresponds to the ideal temperature for *Drosophila suzukii* development. The range composed by the solid green line corresponds to the temperature limits for *Drosophila suzukii* development.

In the EESJ-EPAGRI, throughout the monitoring period, the minimum temperature presented negative values in July and August for the years of 2019 and 2020 (Figure 3). The maximum temperature oscillated between 20 and 30 °C, and in June, July and August of 2019, April, May, June and July of 2020, and April and May of 2021, remained within the range of 20 to 25 °C which corresponds to temperature for the optimal *D. suzukii* development (Figure 3). The average temperature varied, approximately, between 10 and 20 °C remain-

ing within the temperature limits (10 to 30 °C) for the development of *D. suzukii*.

Wine cultivars susceptibility

In the bioassays performed, the berries of all cultivars tested showed susceptibility to the attack, because all presented punctures and *D. suzukii* eggs. It was also possible to observe that the berries of all cultivars tested provided favorable nutritional conditions for the larval development resulting in the emergence of adults. However, there were differences among the cultivars for the variables analyzed (Table 4).

Table 4. Mean values (± standard error) per cultivar for the 2020/2021 season of São Joaquim Experimental Station vineyard of the Agricultural Research Company and Rural Extension of Santa Catarina. São Joaquim, Santa Catarina, Brazil

Berry with no damage and bioassay with non-choice							
Cultivar	Punctures g ⁻¹	Eggs g ⁻¹	Females g ⁻¹	Males g ⁻¹	Embryonic viability (%)	Egg-Adult Time (days)	Sex ratio
Sauvignon Blanc	0.80±0.23 ^{NS}	0.44±0.15 ^{NS}	0.13±0.05 ^{NS}	0.10±0.04 ^{NS}	44.42±13.23 ^{NS}	14.43±01.27 a	0.63±0.07ab
Chardonnay	1.05±0.31	1.03±0.39	0.18±0.11	0.07±0.05	11.04±04.74	16.80±01.30 ab	0.74±0.08ab
Cabernet Franc	1.15±0.34	0.47±0.15	0.04±0.03	0.02±0.01	16.27±09.83	17.75±02.99 b	0.56±0.21ab
Sangiovese	0.78±0.19	0.66±0.15	0.10±0.02	0.02±0.01	21.46±06.14	16.12±02.83ab	0.79±0.07a
Cabernet Sauvignon	1.41±0.22	0.70±0.09	0.10±0.02	0.12±0.02	35.44±07.04	14.78±01.56 a	0.45±0.03b
p-value	0.3083	0.2511	0.2404	0.0499	0.3535	0.0059	0.0310
Berry with no damage and bioassay with free-choice							
Sauvignon Blanc	1.14±0.37 ab	0.66±0.25 b	0.01±0.01 b	0.03±0.03 b	04.16±03.22 b	12.00±0.0 **	0.33±0.00**
Chardonnay	0.34±0.20 b	0.37±0.15 b	0.02±0.01 b	0.02±0.01 b	04.76±03.68 b	16.00±0.0	0.50±0.00
Cabernet Franc	0.66±0.11 ab	0.37±0.14 b	0.00±0.00 b	0.02±0.02 b	02.38±01.84 b	18.00±0.0	0.00±0.00
Sangiovese	1.69±0.19 a	2.20±0.30 a	0.34±0.06 a	0.72±0.19 a	40.70±08.14 a	15.40±01.81	0.38±0.08
p-value	0.03952	0.0008	0.0004	0.0124	0.0021	-	-
Berry with damage and bioassay with non-choice							
Sauvignon Blanc	-	5.44±0.86 ^{NS}	1.47±0.23 ^{NS}	1.66±0.05 bc	64.21±22.17 ab*	14.00±00.48 ab	0.62±0.05 ^{NS}
Chardonnay	-	4.45±0.80	0.93±0.22	1.72±0.04 b	35.76±06.67 ab	14.16±00.46 ab	0.66±0.05
Cabernet Franc	-	7.02±1.29	1.56±0.31	2.28±0.11 a	41.14±04.42 ab	15.33±00.38 b	0.56±0.09
Sangiovese	-	5.07±0.68	0.82±0.21	1.30±0.10 c	20.87±03.13 b	15.83±00.42 b	0.80±0.06
Cabernet Sauvignon	-	5.27±0.42	1.83±0.20	2.01±0.12 ab	66.91±08.32 a	13.00±00.00 a	0.55±0.04
p-value		0.3194	0.0479	<0.0001	0.0392	0.0043	0.0733

Averages followed by the same letter, in the column in each bioassay, do not differ from themselves according to the Tukey test at 0.05% of significance.

*The comparisons were performed in the Box-Cox transformed scale (λ = 0.06).

**Due to the low number of insects emerged was not possible to perform the comparison.

Among the variables analyzed in the bioassay with berries without damage and in bioassays non-choice, it was possible to observe significant difference only for the sex ratio of

the emerging adults and egg-adult development time. Sangiovese grape presented superior index, however differed only from the Cabernet Sauvignon grape for the sex ratio

(Table 4). When we observe the egg-adult development time, the Sauvignon Blanc and the Cabernet Sauvignon grapes favored the development of *D. suzukii*, when compared to the Cabernet Franc.

In the bioassay with berries without damage, where the flies had the possibility to choose the grape for oviposit, significant difference was observed for several variables analyzed (Table 4). Sangiovese grape obtained superior indices in the number of puncture g^{-1} of grape, differing only from the Chardonnay grape. For the number of eggs g^{-1} of grape, females emerged g^{-1} of grape, male emerged g^{-1} of grape and embryonic viability the Sangiovese grape presented superior indices and differed from the other cultivars (Table 4).

When an incision in the grape was made, causing an injury and offering the flies in bioassays non-choice, it was possible to observe difference among the treatments for the males emerged g^{-1} of grape, embryonic viability and egg-adult development period. The Cabernet Franc grape presented the highest value of males emerged g^{-1} of grape but did not differ from the Cabernet

Sauvignon grape. For the embryonic viability, the Cabernet Sauvignon grape proved to be superior to the others, differing significantly only from the Sangiovese grape (Table 4). Regarding the egg-adult period the Cabernet Sauvignon grape was the one that presented the lowest indices differing significantly from the Sangiovese and Cabernet Franc grapes (Table 4).

Regarding the grape chemical analysis, the °Brix and total titratable acidity values varied from 20.90 to 17.50, and 131.96 to 96.91 (Table 5). The pH values remained close to 3.0. Representing the physical analysis, the penetration resistance expressed in newtons (N) varied from 5.68 to 3.26 (Table 5). For the color patterns, the grapes for red wine presented *hue* angle values (h°) close to 270° that corresponds to the blue/purple color, and the grapes for white wines presented values between 90° and 180° that correspond to the yellow and green colors, respectively. Regarding the luminosity, the grapes for red wine are closer to 0, which corresponds to the black/dark, than the grapes for white wine (Table 5).

Table 5. Physicochemical analyses per wine grapes cultivar for the 2020/2021 season of São Joaquim Experimental Station of the Agricultural Research Company and Rural Extension of Santa Catarina. São Joaquim, Santa Catarina, Brazil

Cultivar*	°Brix	TTA	pH	PR (N)	h°	L
Sauvignon Blanc	19.50	119.16	3.07	5.49	106.61	39.14
Chardonnay	17.50	96.91	3.00	3.77	100.58	40.74
Cabernet Franc	20.90	131.96	3.07	5.68	285.46	27.19
Sangiovese	19.30	108.76	3.16	3.23	257.90	27.31
Cabernet Sauvignon	19.40	126.02	3.27	3.26	267.72	27.68

TTA: total titratable acidity; pH: potential hydrogen; PR: penetration resistance; h° : *hue* angle; L: luminosity.

*It was not possible to perform the comparison of the cultivars, because only one compound sample was collected.

Discussion

Drosophila suzukii monitoring

Although grape is not considered a good host, it is possible to observe through the results obtained in this study, the constant presence of *D. suzukii* adults inside the vineyard and in the forest border that surround it. Similar results were also observed in vine-

yards in France (DELBAC et al., 2017; ROUZÉS et al., 2012), Italy (MAZZETO et al., 2020), Germany (BRIEM et al., 2018; WEIßINGER et al., 2019) and Turkey (KASAP; ÖZDAMAR, 2019). During the period in which the monitoring in the vineyard located in the São Joaquim, Santa Catarina, Brazil, it was possible to verify that the *D. suzukii* activity was higher during the summer and early au-

tumn, where the population peaks occurred, as well as Briem et al. (2018) observed in vineyards located in Germany. This may have been influenced by the climate conditions favorable to the development of the spotted wing drosophila, besides correspond to the maturation and harvest of the wine grapes.

Corroborating with the results obtained in this study Mazetto et al. (2020) observed in vineyards located in Italy that in the beginning of the summer the *D. suzukii* captures were low, however after the flowering the population increased rapidly until the grape harvest period. Kasap and Özdamar (2019) observed in vineyards in Turkey that the *D. suzukii* population increases during the grape harvest, but the largest population peaks occur after the harvest, indicating that the *D. suzukii* feeds on fruits remaining in the vineyard or close to and continues its life cycle feeding on fruits such as nectarine and plum.

Small *D. suzukii* population peaks observed in September inside the vineyard and its surroundings can be related to the spring beginning. In hypothesis, the adults go into a diapause during the winter, when the climate conditions are not favorable to its development and return their activities in the spring (HAMBY et al., 2016). Females can enter in already mated diapause and still have greater ability to pass successfully through this period when compared to the males (DALTON et al. 2011; WIMAN et al. 2014), favoring the reproduction and maintenance of the species in the vineyard. This explains the fact of the September population peak was greater for females than for males. Rossi-Stacconi et al. (2016) highlight the importance of monitoring in the beginning of the winter, since the number of females that enter in diapause is an earlier predictor of the population size in the following summer.

When we observe the abundance of *D. suzukii* adults, we realize that the forest border that surround the vineyard is the preferred habitat when compared to the interior of the vineyards. Briem et al. (2018) observed high captures of *D. suzukii* adults in the forest border of the vineyards compared to inside of

the vineyards, where the number of adults captured at the peak season was above 1000 exemplars in the forest border and of 50 to 100 in the vineyards. Weißinger et al. (2019) also collected fewer *D. suzukii* adults inside the vineyards than in the forest border with the presence of blackberries. Still affirm that the microclimate conditions of the forest border and the greater susceptibility of native berries compared to grape berries can explain the differences in the capture rate.

Some factors can favor the permanence of the adults in the forest border such as the microclimate, the availability of native and wild fruits, and diversity of hosts. Briem et al. (2018) and Mazetto et al. (2020) affirm that the areas that surround the vineyards can provide a habitat with favorable conditions for reproduction and development of *D. suzukii*, increasing the captures inside the vineyards. Drummond et al. (2019) report that native fruits serve as hosts, maintaining *D. suzukii* population while there are no cultivated plants available in the field, until a susceptible culture is present, facilitating the seasonal fly migration.

The sex ratio indicated that the proportion of females in the total of adults captured was greater in the forest border than inside the vineyards. These results draw attention to the protection of the vines located close to the vineyard border, since they are under the greater attack risk of the *D. suzukii* females, which use the forest border as shelter. Kehrlí et al. (2022) observed in the Switzerland vineyards that *D. suzukii* females place the largest number of eggs in berries in the interior of grape bunches close to the forest border, orchards and gardens. This highlights the importance of the forest border as a source of survival and reproduction for the *D. suzukii*.

The temperature is a climate variable that contributes positively to *D. suzukii* population dynamics in orchards of several fruit plants (WOLLMANN et al., 2019). In this study, we observed that the maximum temperatures (average 20 °C) and average (average 15 °C) contributed to the increase of the

D. suzukii population in the forest border and inside the vineyard, respectively. However, Benito et al. (2016) affirm that the maximum temperatures (>30°C) are limiting for *D. suzukii* propagation in Brazil. In this study, the average and maximum temperature were not superior to 29°C, remaining within the limit for the development of *D. suzukii* proving that, in the region of São Joaquim, Santa Catarina, Brazil, the temperature regime favors, in much *D. suzukii* development.

Precipitation affected the population negatively, as well as Manko et al. (2021) also observed in areas without commercial crops in Spain, that the precipitation combined with low temperature resulted in no capture of *D. suzukii*. According to Benito et al. (2016), the main factor that can limit the establishment of *D. suzukii* in Brazil is the relative humidity. The results indicate that for the site in study the relative humidity also disfavored the *D. suzukii* population. It may be that in this study there was an effect of the interaction of climate variables on the *D. suzukii* population, however, the interaction and correlation among the variables were not observed.

Wine cultivars susceptibility

Red wine grape cultivars are susceptible to *D. suzukii* attack when compared to white wine grape cultivars (IORIATTI et al., 2015; LINDER et al., 2014). In this study, both the red wine grape cultivars as the white wine grape cultivars proved susceptible to *D. suzukii* damage. However only in the test with free-choice was possible to observe significant difference among the cultivars, where the Sangiovese red wine grape presented itself susceptible to the *D. suzukii* attack, when compared to the others. The Sangiovese grape susceptibility can be related to its intrinsic physicochemical characteristics.

D. suzukii egg posture level in grape berries depends, mainly, on the skin penetrating resistance (ENTLING et al., 2019; IORIATTI et al., 2015). Our results indicate that the Sangiovese grape presents resistance to distinct penetration of the Chardonnay grape, since they differed significantly in the num-

ber of punctures performed by the *D. suzukii* females. Although all cultivars received punctures, and differ among themselves, the Sangiovese grape was the one that presented superior index regarding the number of eggs deposited in the berries. According to Atallah et al. (2014) *D. suzukii* females can perform punctures without actually occurring the oviposition. This observation draws attention to the management of *D. suzukii* in all cultivars, since the damage caused by the puncture results in cuticular lesions in the berries, which can facilitate the penetration of pathogens and yeasts responsible for the deterioration (IORIATTI et al., 2015).

Subtle differences in the chemical composition of the fruits influence the development of the larvae (WEIßINGER et al., 2019). In hypothesis, the superior performance related to the number of females and males emerged as well as the embryonic viability and sex ratio obtained in the tests with the Sangiovese grape can be related to the chemical characteristics of the berries (°Brix, total titratable acidity and pH). These characteristics can provide adequate nutritional substrate for the larval development and consequently the emergence of adults. However, when we observe the performance of the Sangiovese grape in bioassays with non-choice and with the berries with artificial damage, the result is opposite, providing lower emergence of adults and embryonic viability, indicating that the cultivar can have different behavior depending on its physical integrity.

In grape susceptibility studies (wine and table grapes) cultivated in Rio Grande do Sul highland region, Andreatza et al. (2016a) observed that the Cabernet Sauvignon grape received *D. suzukii* punctures, however, did not present emergence of adults. In this study, the Cabernet Sauvignon grape received punctures and provided the development of immature phases of *D. suzukii* resulting in the emergence of adults. These results indicate that although both Rio Grande do Sul and Santa Catarina highland regions belong to the temperate region of grape production, the intrinsic geoclimatic conditions of each sub re-

gion, besides result in distinct 'terroir' for the wines, can also influence in the cultivar susceptibility to the attack of pests.

When we observe the egg-adult development time of *D. suzukii* in bioassays with non-choice and without injury in the berry, in Cabernet Sauvignon grape, the life cycle was shorter (13 to 14 days) while in bioassays with Cabernet Franc grape was longer (15 to 17 days). According to Andreatza et al. (2016 b), the egg-adult life cycle of *D. suzukii* in artificial specific diet for this species is 11 days. Although the Cabernet Sauvignon grape provided shorter egg-adult development time, still promoted prolongation in the *D. suzukii* life cycle. Again, the time differences (days) among the wine cultivars for the egg-adult development time can be related to the intrinsic chemical characteristics of the cultivars. These results draw attention to the management of the remaining fruits in the vineyards, since the fly manages to oviposit and complete its biological cycle in the wine grapes cultivated in São Joaquim municipality.

Sample of bunches without grapes bagging submitted to the bioassays were collected in order to observe natural infestation. However, for the 2020/21 season was not observed the emergence of *D. suzukii* adults. Saguez et al. (2013) collected samples of bunches of Marechal Foch, Gamay and Seyval Noir wine cultivars in Quebec, Canada, and also observed that there was no emergence of *D. suzukii* adults. Similarly, Rouzés et al. (2020) did not observe *D. suzukii* infestation in wine grape bunches collected in Sauterner and Barsac vineyards in France, but the presence of *D. melanogaster* and *D. simulans*. In this study was also observed the emergence of other drosophilids, but not identified at the species level.

Knowledge of the wine cultivars susceptibility to *D. suzukii* attack is essential for the development of integrated management programs. These results, even if preliminary, can assist the winemakers of São Joaquim municipality, SC, in the management of *D. suzukii*, indicating which cultivar the farmer should

have greater attention with phytosanitary management. However, it is necessary to develop studies correlating physicochemical characteristics of the grapes with the *D. suzukii* preference, for better understanding the behavior of this pest in the vineyards of São Joaquim municipality, SC.

The constant presence of *D. suzukii* adults in the captures performed in this study, suggest that the population is already established in the vineyard. The high rates of adults capture in the forest border that surround the vineyard, indicate that this habitat is preferred by flies, compared to the vineyard. Although *D. suzukii* adults prefer the forest border, it is not recommended, in no way, the removal of native vegetation or forests in the surroundings of the orchards, since they can serve as shelter for numerous species of beneficial insects and natural enemies, besides contributing to the conservation of plant and animal species (TSCHARNTKE et al., 2008).

Despite the impossibility of measuring the natural infestation in the wine grape cultivars evaluated, the fact that all the berries offered to *D. suzukii* adults in the laboratory, in bioassays with free-choice and non-choice, presented puncture damage and egg posture, suggests that all cultivars are under the risk of *D. suzukii* attack. Thus, it is highlighted the need to develop field studies aimed at the integrated management of the spotted wing drosophila. Techniques such as toxic bait and mass trapping should be studied to evaluate their viability and employability in the management of *D. suzukii* in the vineyards, since they aim to control the insect-pest without the use of insecticides in covering, presenting themselves as sustainable alternatives.

Conclusion

D. suzukii population peaks occurred in the summer and in the beginning of the autumn.

Precipitation reduced *D. suzukii* population in the vineyard and the average and maximum temperature provided the population increase.

Sangiovese cultivar presented itself susceptible to the attack and development of *D. suzukii* in free-choice bioassay against the Sauvignon Blanc, Chardonnay and Cabernet Franc.

Cabernet Sauvignon cultivar presented rapid egg-adult development time in bioassays non-choice; it can contribute to the *Drosophila suzukii* population increase in the vineyards.

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