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EARLY LEAF REMOVAL AS STRATEGY TO REDUCE *Botrytis* BUNCH ROT ON CHARDONNAY GRAPEVINE IN HIGH ALTITUDE REGION OF SANTA CATARINA STATE

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ABSTRACT - The objective of this work is to compare different periods of leaf removal in the control of this disease in Chardonnay grown in high altitude regions of Santa Catarina - Brazil. The experiments were conducted in São Joaquim city, in 2018 and 2019 growing seasons. The treatments consisted of leaf removal at the phenological stages of full bloom, buckshot berries, pea-sized berries, veraison, and the control treatment, in which the plants without leaf removal. Epidemiological variables were calculated from: beginning of symptom appearance; times to reach maximum disease incidence and severity; maximum disease incidence and severity; and areas under the incidence and severity disease progress curves. The results of the present work show the importance of early leaf removal of Chardonnay in management of *Botrytis cinerea*, it is recommended to be carried out at the phenological stages of full bloom, buckshot berries and pea-sized berries, as it provides a reduction in the maximum severity of B. cinerea and a reduction of AUSDPC. The rate of disease progression demonstrated that B. cinerea infection and development was faster in plants not submitted to leaf removal. the management of early leaf removal (performed before veraison) is more efficient to prevent or suppress the development of B. cinerea in Chardonnay in edaphoclimatic conditions of high altitude regions of Santa Catarina (Brazil).
Keywords: *Vitis vinifera* L., summer pruning, gray mold, integrated disease management.

DESFOLHA PRECOCE COMO ESTRATÉGIA PARA REDUÇÃO DA OCORRÊNCIA DE BOTRYTIS NA VIDEIRA CHARDONNAY EM REGIÃO DE ELEVADA ALTITUDE DE SANTA CATARINA

RESUMO - O objetivo deste trabalho comparar diferentes épocas de desfolha no controle da doença na variedade Chardonnay cultivada em regiões de altitude de Santa Catarina. Os experimentos foram conduzidos em um vinhedo comercial no munícipio de São Joaquim, SC, sul do Brasil, nas safras 2018 e 2019. Os tratamentos consistiram na desfolha nos estádios fenológicos de plena florada, baga chumbinho, baga ervilha, veraison, e o tratamento controle foram plantas não submetidas a desfolha. Avaliou-se a incidência e severidade da doença semanalmente, após o aparecimento dos primeiros sintomas. Com os dados, variáveis epidemiológicas foram calculadas a partir do: início do aparecimento dos sintomas; tempo para atingir a máxima incidência e severidade da doença; incidência e severidade máxima da doença; e área abaixo da curva de progresso da incidência e severidade da doença. Os resultados do presente trabalho evidenciam a importância da desfolha precoce da videira no manejo integrado da B. cinerea, na qual recomenda-se sua realização nos estádios fenológicos plena florada, baga chumbinho e baga ervilha, por proporcionar redução da severidade máxima da B. cinerea, e redução da AACPSD. A taxa de progresso da doença demonstrou que a infecção e o desenvolvimento da B. cinerea foi mais rápido em plantas não submetidas ao manejo da desfolha. O manejo da desfolha precoce (realizada antes da veráison) é mais eficiente para prevenir o desenvolvimento de B. cinerea na videira Chardonnay nas condições edafoclimáticas das regiões de altitude de Santa Catarina (Brasil).
Palavras-chave: *Vitis vinífera* L., poda verde, podridão cinzenta, manejo integrado de doenças.

INTRODUCTION

The high altitude region of Santa Catarina State has the characteristic of presenting longer phenological cycles when compared to other viticultural regions in Brazil (BRIGHENTI et al., 2013), presenting a higher availability of solar radiation and lower night temperatures in the final stage of maturation, consequently, the grapes have greater oenological potential (MALINOVSKI et al., 2016) and higher levels of aromatic compounds (WURZ et al., 2017a), especially in white varieties. However, this region has favorable climatic conditions with for the occurrence of many fungal diseases that can significantly reduce the yield and grape quality (DE BEM et al., 2015). One of the main factors limiting grape production is the occurrence of gray

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mold or *Botrytis* bunch rot caused by the fungus *Botryotinia fuckeliana* (de Bary) Whetzel, a sexual form of *Botrytis* cinerea Persoon ex Fries (ELLIS, 1971). *Botrytis* bunch rot tends to be more difficult to control in cultivars that produce compact clusters, such as Chardonnay (HED et al., 2011).

Currently the control of B. cinerea is based on the sprays of synthetic organo fungicides (WURZ et al., 2020). However, the single dependence on this control method is not sustainable, due to the emergence of fungicides resistance in populations of B. cinerea (LEROCH et al., 2011). This fungal infection is more likely to occur in less exposed and compact clusters (DE BEM et al., 2015).

The grapevine leaf removal is a traditional management practice, reduces the canopy density, increases air circulation, provides less humidity in the cluster zone and allows better coverage of fungicides in the clusters (LOHITNAVY et al., 2010; TARDAGUILA et al., 2012; MOLITOR et al., 2014; WURZ et al., 2017b), therefore, it is a cultural practice to be used in the integrated management of B. cinerea in viticulture. However, according to Wurz et al. (2020), there is little information about the effect of cultural practices in high altitude wine producing regions, especially Brazil, Argentina and Chile.

Considering that Chardonnay is the main white variety grown in high altitude region of Santa Catarina (VIANNA et al., 2016), and that it is highly susceptible to the occurrence of B. cinerea, the objective of this work was to compare the efficiency of different leaf removal periods to control *Botrytis cinerea* in the cv. Chardonnay grown in São Joaquim, Santa Catarina State, Southern Brazil.

MATERIAL AND METHODS

The experiment were carried out in a commercial vineyard located in São Joaquim municipality (28°17’39” S, 49°55’56” W), at an altitude of 1,230 m above sea level, in Santa Catarina State, Southern Brazil, during the 2018 and 2019 growing seasons. The region’s climate is classified as Köppen-Geiger classification, and displays 1,714 heliothermic index, 1,621 mm average annual rainfall, and 80% relative humidity (TONIETTO; CARBONNEAU, 2004). The soils of the region are classified as Cambissolo Húmico (Inceptisol), Neossolo Litólico (Entisol), and Nitossolo Háplico (Ultisol), developed from riodacite and basalt rocks (SANTOS et al., 2018).

Daily rainfall, relative humidity, and temperatures were recorded at the Santa Catarina Hydrology and Environmental Resources Center (Epagri) from August to March of the 2018 and 2019 growing seasons, which corresponded to the interval of phenological stages from bud break (August) to harvest (March) (Figure 1).

The vineyard consisted of approximately 1,000 vines (10 rows of approximately 100 vines) of 10-year-old *V. vinifera* cv. Chardonnay grafted onto ‘Paulsen 1103’ rootstock. The vineyard was trained in vertical shoot positioning trellis at distances of 3.0 m x 1.5 m, arranged along a single North/South-oriented row, covered with anti-hail net and pruned to a double directional horizontal cordon at 1.2 m in height.

The cultivar Chardonnay is susceptible to *Botrytis* bunch rot and the disease has been present in the vineyard in previous years. The fungicides pirimetanil/Mythos SC (anilinopyrimidine 300 mL of active ingredient (a.i) L^{-1} , 150 g of commercial product (c.p) ha^{-1}; mancozeb/Dithane NT (dithiocarbamate, 800 mL a.i L^{-1}, 350 g c.p ha^{-1}); fenamidone/Censor SC (imidazole, 500 mL a.i L^{-1}, 30 mL c.p ha^{-1}), thiophanate methyl/Cercobin 700 WP (benzimidazole, 700 mL a.i/ L^{-1}, 70 mL c.p ha^{-1}), triazol/Folicur PM (tebuconazole, 100 g i.a L^{-1}, 250 g c.p ha^{-1}), Clorothalonil/Bravonil 720 (Tetrachloroisophthalonitrile, 123 g i.a L^{-1}, 150 g c.p ha^{-1}) and ditianone/Delan WP (quinone, 750 g i.a L^{-1}, 125 g c.p ha^{-1}).

Leaf removal treatment was carried out manually at the phenological stages of full bloom, buckshot berries, pea-sized berries and veraison. In treated plots, four basal leaves and lateral shoots were removed from the nodes opposite the cluster and from the first node above and below the cluster, in accordance with the scale of Baillod and Baggiollini (1993). In control plots, leaves were not removed. The field experiment was conducted using a completely randomized block design with four replicate plots for each treatment, and each replicate plot comprised ten adjacent vines that formed a continuous canopy.

Botrytis bunch rot incidence was evaluated by counting diseased clusters, and severity was determined by counting the number of rotted berries per cluster according to the diagrammatic scale of Hill et al. (2010), based on 12 levels of disease severity: 1%, 5%, 10%, 15%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, and 90%. For each plot, 30 random clusters were evaluated in each treatment. The evaluations took place from 1/16/2018 to 2/21/2018 and from 1/4/2019 to 2/8/2019, with weekly evaluations, during 2018 and 2019 growing seasons, respectively.

Botrytis bunch rot progress curves were constructed and epidemics were compared in both growing seasons using four epidemiological measures: beginning of symptom appearance (BSA), times to reach maximum disease incidence and severity (TRMDI and TRMDS, respectively), maximum disease incidence and severity (I_{max} and S_{max}, respectively), and areas under the incidence and severity disease progress curves (AUIDPC and AUSDPC, respectively). AUDPC was calculated as $[(Y_i + Y_{i+1})/2]$ $(t_{i+1} t_i)$, where Y = disease intensity (incidence or severity), t = unit of time, and i = cumulative number of evaluations. This area represented the trapezoidal integration value of severity (CAMPBELL; MADDEN, 1990).

The non-normally distributed data of *Botrytis* bunch rot incidence and severity were subjected to arcsine square root transformation to establish criteria of normality (residuals and homogeneity of variance). Tests for significance were performed using analysis of variance, and post hoc comparisons were performed using the Scott-Knott test ($p = 0.05$).

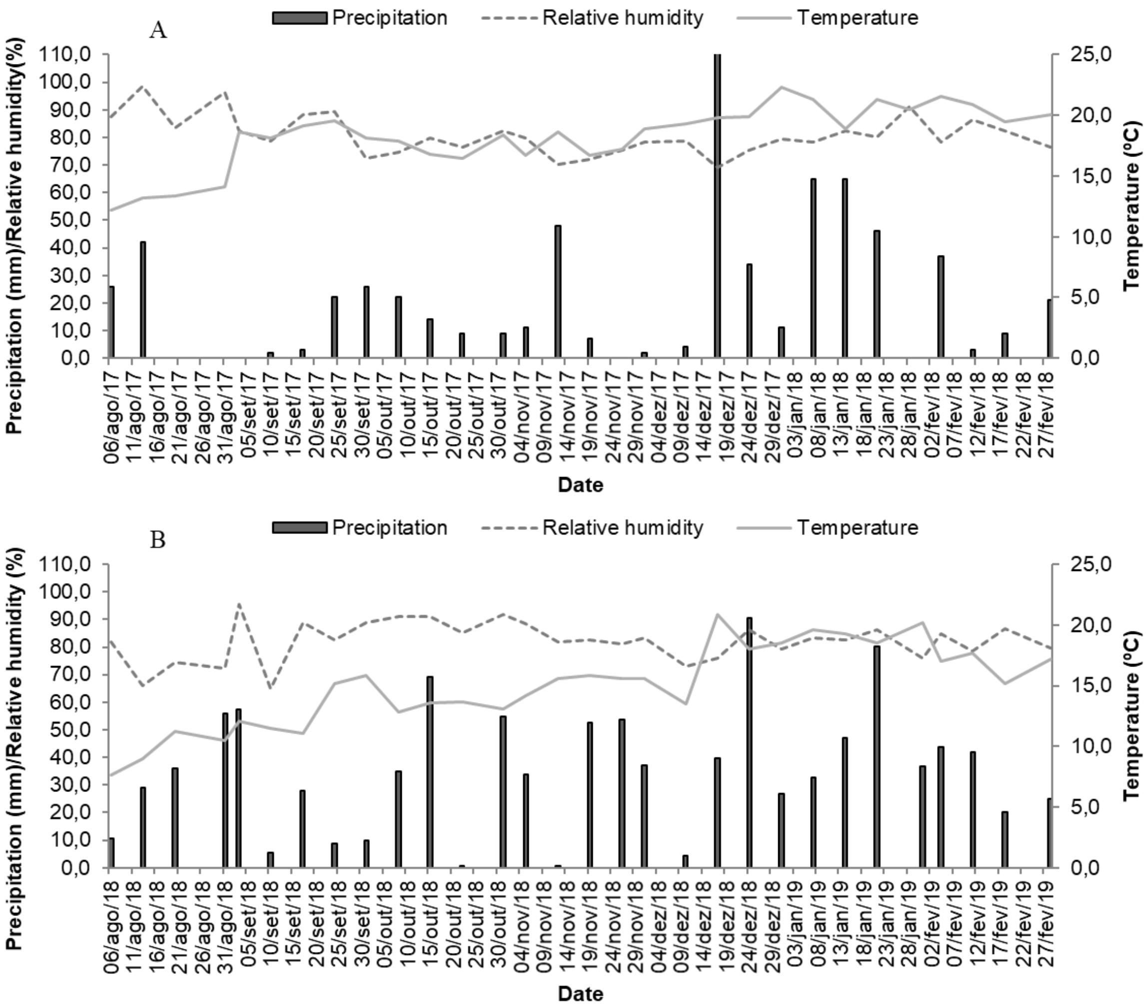

FIGURE 1 - Accumulated rainfall (mm), relative humidity (%) and mean temperature (ºC) in São Joaquim/SC (Brazil) during the 2017/2018 (A) and 2018/2019 (B) growing seasons.

RESULTS AND DISCUSSION

The mean temperature, relative humidity and the volume of rainfall during the vegetative cycle of Chardonnay (August to February) were different in the two evaluated growing seasons, presenting averages of 18.3ºC, 80.8% and 649 mm in the 2017/2018 and 15.0ºC, 82.1% and 1066.7 mm in the 2018/2019 cycle, respectively (Figure 1). The combination of high rainfall and high relative humidity are factors that favor the appearance of *Botrytis cinerea*, especially in spring and summer (BEM et al, 2015). In addition, temperature range between 15-23ºC is defined as optimal for the disease development (HED et al., 2009).

The fungus can attack almost all floral organs, and the attacks on clusters, during maturation, assume greater severity. In these periods, between full bloom and maturation, high relative humidity were observed, associated with the occurrence of precipitation and the optimum temperature for occurrence of B. cinerea. For Wurz et al. (2017b), favorable environmental conditions and biological conditions are fundamental for the disease development and have fundamental importance in determining the strategies for disease control.

For the variables beginning of symptom appearance (BSA) and time to reach maximum disease incidence (TRMDI), there was no effect of different leaf removal timings on both growing seasons (Table 1). The beginning of symptom appearance occurred 12 and 7 days after the first evaluation, in the 2018 and 2019 seasons, respectively. In 2018, the maximum disease incidence was found 35 days after the first evaluation, and in 2019, the maximum disease incidence ranged from 25 to 32 days, with no significant differences between leaf removal timing. A previous work carried out by Wurz et al. (2017b), did not found effect of different leaf removal timing on the variables BSA and TRMDI of B. cinerea on Cabernet Sauvignon.

The variable time to reach maximum disease severity (TRMDS) was influenced by the different leaf removal timings in 2019, the TRMDS was greater when leaf removal was carried out in full bloom, buckshot berries and veraison, with values of 35, 32.3 and 35 days, respectively. While plants not submitted to leaf removal reached the maximum disease severity with 27 days, indicating, therefore, that early leaf removal delays B. cinerea epidemic. In 2018, it took 35 days for maximum disease severity, for all leaf removal timings.

The observed values of maximum disease incidence and maximum disease severity were lower in 2019. The leaf removal timing influenced the maximum severity of B. cinerea, but did not influence the maximum incidence values, as observed in Table 2. In 2018, values from 86.6 to 96.6% of maximum disease incidence were observed, and in 2019, values between 73.3 and 83.3% of maximum incidence were observed, with no significant differences between the different timings of leaf removal.

TABLE 1 - Beginning of symptom appearance (BSA) (days after the first evalution), time to reach maximum disease incidence and severity (TRMDI and TRMDS) (days) of Chardonnay subjeted to different timings of leaf removal in high altitude regions of Santa Catarina State in 2018 and 2019.

Leaf removal timing	BSA (days)		TRMDI (days)		TRMDS (days)	
	2018	2019	2018	2019	2018	2019
Control	12 ns	7 ns	35 ns	30 ns	35 ns	27 b*
Full bloom	12 ns	7 ns	35 ns	32 ns	35 ns	35 a
Buckshot berries	12 ns	7 ns	35 ns	25 ns	35 ns	32 a
Pea-sized berries	12 ns	7 ns	35 ns	30 ns	35 ns	30 b
Veráison	12 ns	7 ns	35 ns	25 ns	35 ns	35 a
CV(%)	0.2	0.1	0.5	25.5	0.5	8.5

*Means followed by different letters, in the columns, differ by Scott-Knott test (p≤0.05). ns = not significant for analysis of variance (ANOVA).

However, for the variable maximum disease severity, there was an influence of leaf removal timing. When this management was performed early, in the stages full bloom, buckshot berries and pea-sized berries resulted in the lowest values of maximum disease severity; with observed values of 9.2, 9.7, 8.6% and 3.8, 2.5, 4.1% in 2018 and 2019, respectively. For Wurz et al. (2020), the grapevine leaf removal influences the microclimate in cluster zone, and consequently, reduces the development of B. cinerea. These results confirm other studies relating the performance of leaf removal management with the reduction of B. cinerea (CONIBERTI et al., 2013; KOMM; MOYER, 2015; WURZ et al., 2017b).

As with maximum disease incidence, there were no statistically significant differences between the different leaf removal timings times for the area under the incidence disease progress curve (AUIDPC), in both evaluated growing seasons. However, an effect was observed between the different leaf removal timings for the area under the severity disease progress curve (AUSDPC), in both evaluated growing seasons (Table 2). Regarding AUSDPC, the lowest values observed in 2018 were 128.6, 127.6 and 126.6 for leaf removal performed in full bloom, buckshot berries and pea-sized berries, respectively; in 2019, it was observed values of 53.7 and 51.7, for leaf removal carried out in full bloom and buckshot berries, respectively. In both growing seasons, the highest AUSDPC values were observed in clusters of plants not submitted to leaf removal, with values of 235.7 and 102.3, in 2018 and 2019, respectively.

TABLE 2 - Maximum disease incidence and severity (I_{max} and S_{max}), and areas under the incidence and severity disease progress curves (AUDIPC and AUDSPC) of Chardonnay subjeted to different timings of leaf removal in high altitude regions of Santa Catarina State in 2018 and 2019.

Leaf removal timing	I_{max} (%)		S_{max} (%)		AUIDPC		AUSDPC	
	2018	2019	2018	2019	2018	2019	2018	2019
Control	96.6 ns	76.6 ns	17.5 a	5.1 a	1631.6 ns	1795.0 ns	235.7 a	102.3 a*
Full Bloom	96.6 ns	73.3 ns	9.2 c	3.8 b	1655.0 ns	1741.1 ns	128.6 c	53.7 c
Buckshot Berries	86.6 ns	76.6 ns	9.7 c	2.5 c	1518.3 ns	1986.6 ns	127.6 c	51.7 c
Pea-Sized Berries	90.0 ns	83.3 ns	8.6 c	4.1 b	1436.3 ns	1948.5 ns	126.6 c	79.5 b
Veráison	93.3 ns	73.3 ns	13.3 b	4.7 a	1880.0 ns	1855.0 ns	178.2 b	79.4 b
CV(%)	13.7	14.2	7.5	6.5	18.7	19.5	12.5	11.2

*Means followed by different letters, in the columns, differ by Scott-Knott test (p ≤ 0.05). ns = not significant for analysis of variance (ANOVA) (p≤0.05).

The data observed in the present study corroborate those observed by Wurz et al. (2020), when evaluating the effect of leaf removal on the control of B. cinerea on Sauvignon Blanc, in which the removal of four basal leaves, before the phenological stage veraison, reduces the severity and AUSDPC of B. cinerea, in regions of high altitude. The values of severity and AUSDPC found in the present study can be considered low, when compared with previous studies carried out by and, with the Cabernet Sauvignon (WÜRZ et al., 2017b) and Sauvignon Blanc (WÜRZ et al., 2020), in high altitude regions of Southern Brazil.

During the growing season 2017/2018, there was an increase in disease incidence in the first thirty days of evaluation, after which there was a stabilization for seven days, and after 02/14/2018, there was an increase in AUIDPC again (Figure 2), for all leaf removal timings. The only exception occurred when leaf removal was performed on the the stage of pea-sized berries, which showed an increase in AUIDPC throughout the evaluation period, however, at harvest time, there were no significant differences between the leaf removal timings, as previously described in Table 2. During the growing season 2018/2019, occurred an increase in AUDIPC values in the first twenty days of evaluation, and after January 31, 2019, there was almost a stabilization of AUIDPC.

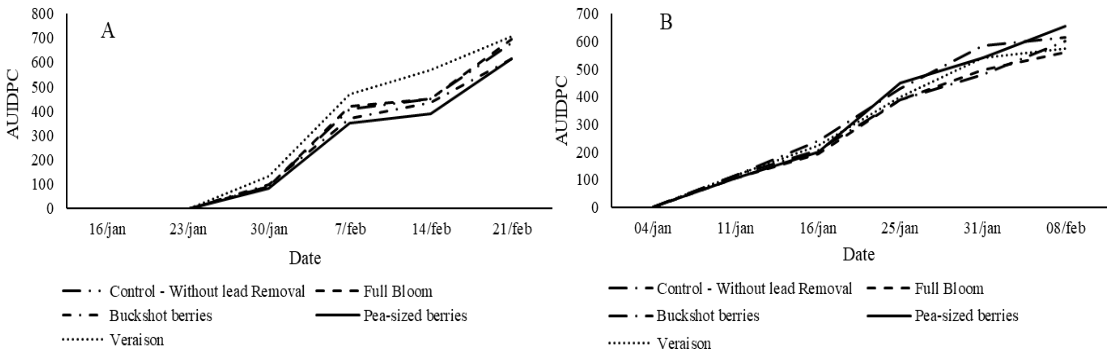

FIGURE 2 - Area under the incidence disease progress curves (AUDIPC) of Chardonnay subjeted to different timings of leaf removal in high altitude regions of Santa Catarina State (Brazil) in 2018 (A) and 2019 (B).

Regarding the severity disease progress curves, in 2017/2018 growing season, there was an increase, not very significant until 01/30/2018, after that date it was observed a significant increase in AUSDPC, mainly on plants not submitted to leaf removal, which showed the greatest increase in AUSDPC values (Figure 3).

For the 2018/2019 growing season, from 11/01/2019 to 01/25/2019, there was an increase in AUSDPC values for all different leaf removal timings, then the values stabilized until 02/01/2019; subsequently, the AUSDPC values increased, with the highest values observed for plants not submitted to leaf removal. It is known that full blomm is the most important phenological stage for B. cinerea infection, due to the susceptibility of floral parts (KELLER et al., 2003), the infections remains latent and then increases the infection exponentially from veraison until harvest (BERESFORD et al., 2006; SADRAS; MORAN, 2013).

When comparing Figures 2 and 3 with climatic data, it is observed that periods with no increase in values of AUIDPC and AUSDPC, coincide with periods of lower rainfall, and low relative humidity, evidencing the climatic influence, in epidemiological variables of B. cinerea. For Holz et al. (2013), the leaf removal directly influences the increase in solar radiation and wind speed in cluster zone, having an effect in reducing the progress of B. cinerea. In addition, in situations with no leaf removal, the remaining leaves in cluster zone become a physical barrier, preventing the applied fungicides to control B. cinerea from reaching the biological target (BERESFORD et al., 2006; WÜRZ et al., 2017b).

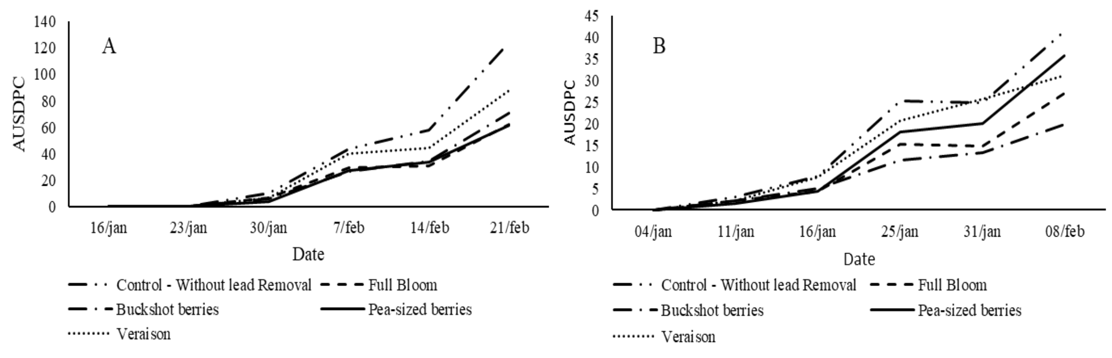

FIGURE 3 - Area under the severity disease progress curves (AUDSPC) of Chardonnay subjected to different timings of leaf removal in high altitude regions of Santa Catarina State (Brazil) in 2018 (A) and 2019 (B).

Normally, in cool climate regions (as the studied location), the harvest time is more often determined by clusters disease than grape maturation itself (MOLITOR et al., 2012). The delay in the epidemic caused by the early leaf removal allows a temporal benefit at maturation time. In general, wines from grapes harvested late and at the appropriate time of maturation, have higher quality and are often preferred in tastings (SPRING, 2004). Early leaf removal carried out in full bloom, buckshot berries and pea-sized berries phenological stages, are an efficient strategy for disease reduction, and provide grapes with superior oenological quality, with greater accumulation of soluble solids, total polyphenols and anthocyanins (WÜRZ et al., 2017b; WÜRZ et al., 2017c).

With the accomplishment of the present work, it was verified the importance of the early leaf removal management of of grapevine, being an indispensable management for the culture, and future works should explore the effects of this management on the chemical and sensorial quality of the wines.

CONCLUSIONS

The results of the present work show the importance of early leaf removal in grapevine integrated management of B. cinerea, in which it is recommended to be carried out in full bloom, buckshot berries and pea-sized berries phenological stages, as it provides a reduction in the maximum severity of B. cinerea and reduction of area under the severity disease progress curves (AUSDPC) values.

The disease progression rate showed that B. cinerea infection and development was faster when the plants were not subjected to leaf removal management.

Thus, the management of early leaf removal (performed before veraison) is more efficient to prevent or suppress the development of B. cinerea in Chardonnay in edaphoclimatic conditions of high altitude regions of Santa Catarina (Brazil).

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