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ECOPHYSIOLOGY OF FUNGUS-RESISTANT GRAPES GROWN AT THREE ALTITUDES IN THE SOUTH OF BRAZIL

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ABSTRACT
Worldwide, climatic conditions change due to latitude, longitude, and altitude. Latitude directly influences the duration of the photoperiod and the intensity of solar radiation received, longitude mainly represents continentality/oceanity and altitude influences changes in climate due to atmospheric pressure. Some winegrowers seek to obtain more favorable conditions for viticulture in higher altitude areas, compensating, to a certain extent, the effect of latitude. Numerous scientific studies confirm the existence of morphological and physiological changes that occurred in the vine plant exposed to certain environmental conditions, typical of most altitude vineyards. Therefore, the objective of this work was to carry out climatic assessments at different altitudes and their interactions with the development of varieties resistant to fungal diseases. The aim of this work was to carry out climatic assessments at different altitudes and their interactions with the development of varieties resistant to fungal diseases. The varieties Aromera, Calardis Blanc, and Felicia

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were studied, white vines resistant to fungal diseases, cultivated in Água Doce (1329 meters), Curitibanos (1000 meters), and Videira (774 meters), cities in the state of Santa Catarina, during 2018/19 and 2019/20. The analyzes included climatic data, net photosynthesis (A), stomatal conductance (gs), transpiration (E), internal carbon (Ci), maximum quantum yield of photosystem II (Fv/Fm), non-photochemical quenching (NPQ), yield effective quantum of photosystem II (ΔF/Fm'), apparent electron transport rate (ETR), leaf area (AF), number and weight of bunches, Ravaz Index, soluble solids (SS) and total acidity (TA). Compared to cultivation at an altitude of 774 meters, the altitude of 1329 meters provided a total average decrease of 4.28ºC in temperatures, 99% in NPQ, 30% in AF, 4.65% in bunch weight, and 15% in ATT; and average total increment of 144.70 W.m-2 in radiation, 13.5Km.h-1 in wind speed, 17% in A, 81% in gs, 9% in E, 8% in Ci, 8% in Fv/Fm, 14% in the ΔF/Fm', 45% in the ETR, 19% in the number of bunches and 9% in the SS. It is concluded that there is an adaptation to growing conditions, but they are strongly regulated by altitude and variations in climate, with higher altitude providing more favorable climatic conditions for the development of the studied varieties.

Keywords: PIWI. Gas exchange. Vigor. Photosynthesis.

INTRODUCTION

Vineyards at higher altitudes benefit from greater thermal amplitude, less mineralization of soil organic matter, and a more significant amount and intensity of solar radiation (Berli *et al.*, 2015). These conditions benefit by extending the crop cycle, influencing plants photosynthetic and respiratory balance, and lowering vegetative growth. These specific characteristics of climatic conditions and development favor the accumulation of energy in the fruits since it is not allocated to the growth of branches. It delays the ripening of the fruits and allows a maturation with a additional accumulation of sugars and aroma precursors (Jiang *et al.*, 2013). From the 2000s onwards, new regions between 900 and 1,400 m altitude, in the states of Santa Catarina and Rio Grande do Sul, began to stand out in the production of grapes and fine wines (Brighenti *et al.*, 2013).

Despite the vine cultivation options at different altitudes, southern Brazil has the limiting climatic factor of excess rainfall and relative humidity, which can induce greater vegetative growth and form a favorable microclimate for the development of fungal diseases, which may cause a reduction in photosynthesis, productivity, and grape quality. Both phytosanitary difficulties and adaptation problems have been efficiently circumvented through the intensive use of fungicides, hormones, and growth regulators. However, given the increase in the market's demand concerning food safety and environmental protection, the concept of quality goes beyond the intrinsic characteristics of the product (Maia *et al.*, 2015). In addition to productivity and quality characteristics, traditionally considered in genetic improvement programs, resistance to diseases and adaptation of cultivars to different producing regions are priority items for Brazilian viticulture (Maia *et al.*, 2015).

The sustainability of production systems is a requirement, imposing commercial barriers to products originating from systems that do not rigorously consider these aspects. Integrated production systems spread to different cultures, and sustainable production is the main objective of agriculture. In this context, genetic improvement plays a crucial role in creating new varieties of vine that are resistant to diseases, adapted to the environmental conditions of the different producing regions, and have the characteristics of productivity and fruit quality that the market requires (Maia *et al.*, 2015).

From this scenario, molecular marker-assisted selection combined with multiple backcrossing with *Vitis vinifera* developed varieties with a percentage of more than 85% of *V. vinifera* in their lineage; these varieties are referred to as "PIWI", from German: *Pilzwiderstandsfähige*, “disease resistant”. They have been recommended as the most suitable choice for organic viticulture and are grown worldwide. These fungus-resistant varieties are being tested and analyzed in countries such as Austria, Brazil, Czech Republic, Denmark, England, France, Germany, Hungary, Italy, Latvia, Luxembourg, Netherlands, Portugal, Slovakia, Spain, South Africa, Switzerland, Thailand, and Turkey. Therefore, the objective of this study was to generate scientific knowledge from climatic assessments at different altitudes in the South of Brazil, and their interactions with the development of fungus-resistant varieties.

MATERIAL AND METHODS

Aromera, Calardis Blanc, and Felicia are white grapes varieties with genes that provide medium to high resistance to downy mildew (*Plasmopara viticola*) and powdery mildew (*Erysiphe necator*). They are cultivated in experimental vineyards in the municipalities of Água Doce (26º43'3" S; 51º30'1" W) at an altitude of 1329 meters, Curitibanos (27º17'20" S; 50º36'17" W) at an altitude of 1000 meter and Videira (27º1'30" S; 51º9'0" W) at an altitude of 774 meters. The vineyards implementation was carried out in 2015, and the plants are on the Paulsen 1103 rootstock under a trellis system. The spacing is 3.0 meters between rows and 1.2 meters between plants. The experimental design was in randomized blocks, consisting of five replications of 10 plants per plot, totaling 150 plants analyzed per variety, during the 2018/19 and 2019/20 harvests.

The meteorological data were analyzed from the winter of 2018 to the autumn of 2020, divided according to the seasons, and obtained from the meteorological stations of the Santa Catarina Environmental Resources and Hydrometeorology Information Center (CIRAM-Epagri) installed close to the vineyards. The variables analyzed were minimum temperatures during spring, autumn, and winter and average temperatures during summer, accumulated precipitation (mm), average relative humidity (%), wind speed ($km.h^{-1}$), and average global solar radiation ($W.m^{-2}$).

The values of net photosynthesis (A_N), stomatal conductance (g_s), transpiration rate (E), and intercellular CO_2 concentration (Ci) were detected using a portable Infrared Gas Analyzer (IRGA; LI-6400XT; LI-COR, Lincoln, NE, USA). The IRGA leaf cuvette (area: 2 cm^2) was set as follows: photosynthetically active radiation (PAR) 800 $\mu mol\ m^{-2}\ s^{-1}$, reference (CO_2) 400 ppm and flow rate 400 $\mu mol\ s^{-1}$. The measurements were performed between 9 to 11 a.m. when the grapes were ripe for harvest on the first fully expanded mature leaf from the middle third of the shoot.

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Maximum quantum yield of photosystem II (Fv/Fm), non-photochemical quenching (NPQ), photosystem II effective quantum yield (ΔF/Fm'), and apparent electron transport rate (ETR) was measured using a portable pulse amplitude fluorometer (Mini PAM, Walz, Effeltrich, Germany), on the same leaf used for gas exchange determinations. Before the measurements, the leaves were dark adapted for 20–30 min, to ensure oxidation of the plastoquinone pool. The initial fluorescence was obtained by switching on the measuring light of 0.1 μmol m^{-2} s^{-1} at 600 Hz and the maximum fluorescence yield (Fm) was estimated by applying saturation pulses of 8 mmol m^{-2} s^{-1} and 0.8 s duration. Measurements were performed during harvest during harvests from 9 to 11 am., with a metal clip (DLC-8) coupled to the MINI-PAM sensor, avoiding the midrib (Maxwell and Johnson, 2000).

The leaf area was evaluated using a CI-202 portable leaf area meter (CID Bio-Science, Inc., Camas, Washington, USA). Was collected 150 whole and healthy leaves of different sizes per variety at each altitude, randomly obtained from different shoots on both sides (east and west) of the rows. Leaves with a midrib length less than 3.0 cm were not evaluated. The yield of the varieties was determined, on the day of harvest, weighing the bunches (kg.$plant^{-1}$). To analyze the balance between vegetative vigor and production, the Ravaz Index was calculated using the ratio between fruit production per plant (kg) and the weight of the pruned material (kg). A weekly collection of 30 berries per variety was carried out, and the musts were extracted to determine the soluble solids content (SS) in °Brix and total acidity (TA) in mEq.L^{-1}. The analyzes were carried out at the Laboratory of Analysis of Beverages and Vinegars of the State of Santa Catarina (Labesc) of the Agricultural Research and Rural Extension Company of Santa Catarina (Epagri).

The data were subjected to analysis of variance, where the same variety represented the sources of variation at different altitudes and by different varieties at the same altitude. According to the F test result, the means were compared by Tukey's test at a 5% error probability.

RESULTS AND DISCUSSION

Meteorological data are presented in Tables I, II, III, IV and V at altitudes of 1329 meters (Água Doce), 1000 meters (Curitibanos), and 774 meters (Videira), from winter 2018 to autumn 2020. Throughout the analyzed period, there is a consistent behavior of minimum temperatures in autumn, winter, and spring and average in summer, always being lower with increasing altitude. During the spring of 2018, the average difference in temperatures

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between the lowest and highest altitude reached 4.28ºC. Significant differences were observed only during spring and summer, seasons from budbreak to harvest (Table I). The altitude has been considered the central differential for elaborating fine wines in Santa Catarina. Its inverse relationship with temperature makes the vegetative cycle of the grapes longer, providing a physicochemical and sensory differential to the wines (Brighenti *et al.*, 2013).

Table I. Temperatures data from winter 2018 to autumn 2020 in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil.

Altitude	Temperature (ºC)			
	Winter 18(1)	Spring 18(1)	Summer 18/19(2)	Autumn 19(1)
774m	10.96a±4.22(*)	15.25a±2.67	22.34a±1.86	15.63a±4.00
1000m	9.49a±4.73	12.91b±3.04	20.40b±1.96	14.46ab±4.15
1329m	9.50a±3.71	10.97c±2.54	18.96c±2.13	14.13b±3.84
Altitude	Winter 19(1)	Spring 19(1)	Summer 19/20(2)	Autumn 20(1)
774m	11.81a±4.72	15.17a±2.61	21.91a±2.05	9.59a±4.30
1000m	10.94a±4.76	13.76b±2.40	19.91b±2.29	9.45a±3.59
1329m	10.27a±4.94	12.75c±2.46	18.58c±2.19	7.43b±5.48

(1)Minimum temperatures. Average temperatures(2). (*)Means followed by the same letters lowercase within same column are not statistically different by Tukey's test ($p>0.05$).

Precipitation and relative humidity did not show a direct relation with altitude (Table II). These variables are below what is considered ideal for vine's development in the regions studied. A vine's total water use over a cool-climate or short growing season ranges from 300 to 600 mm, depending on variety, planting density, canopy size and configuration, and seasonal temperature patterns (Williams, 2014). Water use on the vine in a warm climate with longer growing seasons ranges from approximately 400 to 800 mm throughout the cycle (Williams, 2014). However, the entire Southern Region of Brazil has a rainfall volume above the vine development limit, as could be observed during the spring and summer of 2018/2019, periods in which the combined volume of precipitation reached, on average, 1,328 mm at the lowest altitude and 1,377mm at the highest altitude (Table II). Rainfall can influence the date and occurrence of flowering. Excessive precipitation before the flowering stage can make anthesis difficult. During flowering, it can scatter pollen, causing the flowers to fail to fertilize and their consequent fall, resulting in clusters with berries with a wide variation in size and maturity. The plant sensitivity is high at this stage. Any deviation from the temperature and humidity values necessary for the healthy vine development can prevent the flowering putting at risk the entire phenological cycle (Santos, 2011), in addition to increasing the incidence of fungal diseases.

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Table II. Accumulated precipitation data from winter 2018 to autumn 2020 in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil.

Altitude	Precipitation (mm)			
	Winter 18	Spring 18	Summer 18/19	Autumn 19
774m	279.60c±10.81(*)	614.40a ±12.16	714.90a±16.40	557.40b±14.02
1000m	421.8a±10.93	409.20b±7.86	587.40b±11.69	403.60a±10.82
1329m	346.21b±9.91	689.91a±14.56	688.03a±16.19	484.92a±13.15
	Winter 19	Spring 19	Summer 19/20	Autumn 20
774m	190.40a±6.39	530.40a±9.15	433.20a±13.11	456.60a±17.19
1000m	157.6c±4.43	422.20b±9.15	346.40b±10.11	288.60b±8.59
1329m	172.10b±5.06	345.34c±8.94	429.53a±14.87	430.06a±14.04

*Means followed by the same letters lowercase within same column are not statistically different by Tukey's test ($p>0.05$).

The ideal relative humidity is between 62 and 68% for the healthy vine's development. However, humidity above 75% - average data observed during the entire period analyzed at the three altitudes (Table III) - acting together with high temperatures and precipitation during the vegetative period, favors the incidence of fungal diseases (Shimano and Sentelhas, 2013). In studies on climatic risks for the occurrence of fungal diseases in vines in the South and Southeast of Brazil, these authors found that, when the vine production cycle is concentrated in spring/summer, periods when rainfall is more abundant and temperatures higher, the environment becomes more conducive to the occurrence of fungal diseases, resulting in a higher number of fungicide application. An example was the city of Caxias do Sul/RS, which, according to the historical average, has a climate similar to that of the cities analyzed in the present study and has an average altitude of 817 meters. In this city, the average climatic risk of disease occurrence in September was 54%, reaching 19 fungicide applications during the crop cycle. Resistant or partially resistant hybrids can significantly reduce the application of chemical compounds and, therefore, contribute substantially to the sustainability of viticulture. Treatments may be limited during rainy seasons or crucial phenological stages, such as flowering and the setting of fruits (Bem *et al.*, 2020).

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Table III. Relative humidity data from winter 2018 to autumn 2020 in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil.

Altitude	Relative Humidity (%)			
	Winter 18	Spring 18	Summer 18/19	Autumn 19
774m	79.36b±7.90(*)	76.31b±10.12	81.91b±5.65	86.27b±4.74
1000m	84.10a±11.78	86.69a±8.01	88.34a±5.40	85.87ab±4.99
1329m	85.86a±10.48	84.31a±11.20	86.78a±6.73	87.89a±5.12
Altitude	Winter 19	Spring 19	Summer 19/20	Autumn 20
774m	77.55b±10	75.97c±10.52	76.56b±8.18	79.46b±7.82
1000m	84.42a±9.11	86.08a±7.59	83.69a±7.17	83.11a±9.33
1329m	79.22b±11.41	81.48b±10.15	82.85a±10.09	82.19ab±11.91

*Means followed by the same letters lowercase within same column are not statistically different by Tukey's test ($p>0.05$).

On the other hand, the behavior was inverse for the variables wind speed and radiation, being higher at higher altitudes. The wind speed was significantly different throughout the analyzed period and, on average, 13.5 $km.h^{-1}$ higher at the altitude of 1329 meters compared to the altitude of 774 meters, reaching 19.27±4.50 $km.h^{-1}$ during the spring of 2018 (Table IV). According to Keller (2020), wind speeds above 21.6 $km.h^{-1}$ can cause physical damage to plants and reduce shoot length, leaf size, and stomatal density. The incidence of radiation at the altitude of 774 meters is, on average, 86.78 $W.m^{-2}$ and 144.70$W.m^{-2}$ lower in spring and summer, respectively, compared to the altitude of 1329 meters. According to Vieira *et al.* (2011), the availability and intensity of solar radiation are related to the geographic position and altitude of the place. Thus, these authors recorded a higher amount and intensity of radiation in the region of Campo Belo do Sul (950 meters of altitude), in the Planalto Catarinense, concerning Pech Rouge (1.5 meters of altitude) in France. This factor may favor the more significant sugar accumulation, the high concentration of phenolic compounds, and the greater aromatic complexity.

Table IV. Wind speed and global radiation data from winter 2018 to autumn 2020 in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil.

Altitude	Wind Speed ($Km.h^{-1}$)			
	Winter 18	Spring 18	Summer 18/19	Autumn 19
774m	4.49c±1.80(*)	5.36c±1.63	4.33c±1.05	3.75c±1.23
1000m	9.39b±3.54	10.06b±3.21	8.00b±2.30	6.58b±2.46
1329m	18.12a±5.07	19.27a±4.50	17.28a±4.42	15.77a±4.86
	Winter 19	Spring 19	Summer 19/20	Autumn 20
774m	5.07c±1.87	5.31c±1.59	4.24c±0.92	4.28c±1.57
1000m	8.04b±3.06	8.66b±2.61	6.92b±2.02	6.48b±2.95
1329m	20.92a±9.21	18.42a±4.70	16.16a±3.56	17.62a±5.19
Altitude	Radiation ($W.m^{-2}$)			
	Winter 18	Spring 18	Summer 18/19	Autumn 19
774m	273.82a±61.88	357.58b±87.65	364.30c±78.97	242.14b±52.13
1000m	285.90a±67.65	400.20ab±110.43	430.64b±66.71	265.00ab±60.67
1329m	303.54a±69.70	454.12a±102.97	514.60a±75.77	283.80a±60.16
	Winter 19	Spring 19	Summer 19/20	Autumn 20

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774m	261.78b±55.43	424.12b±79.83	424.68c±68.89	306.80b ±63.65
1000m	285.30b±60.34	459.64ab±89.64	509.20b±68.89	332.20ab ±71.57
1329m	345.74a±65.55	501.14a±95.42	563.78a±86.25	374.28a ±84.26

*Means followed by the same letters lowercase within same column are not statistically different by Tukey's test ($p>0.05$).

Concerning gas exchange, higher net photosynthesis (*A*) values were observed at the highest altitude for the three varieties and in both harvests. In the 2018/19 harvest, the values reached 15.30; 15.21, and 15.00 μmol.m^{-2}.s^{-1} for the Aromera, Calardis Blanc, and Felicia varieties cultivated in the higher altitude, respectively, while at the lowest altitude, the values of *A* measured were, on average, 12.66; 12.76 and 12.40 μmol.m^{-2}.s^{-1} and at the intermediate altitude were, on average, 13.76; 13.15 and 13.45 μmol.m^{-2}.s^{-1} for the same varieties, indicating an average increase of 20.34% and 12.79% at the highest altitude (Table V). According to Kozlowski *et al.* (2012), photosynthesis values vary widely between species and places where plants grow and develop. These variations result from the interaction between leaf age, canopy formation, stomatal conductance, and environmental factors such as incident radiation, temperature, and water availability. The high-altitude region of Santa Catarina is characterized by having longer phenological cycles when compared to other wine-growing regions in Brazil (Brighenti *et al.*, 2013). Associated with the excellent availability of solar radiation and low night temperatures, they allow a greater photosynthetic and respiratory balance of plants, and a complete maturation (Malinovski *et al.*, 2012), producing grapes with oenological quality, especially for their intense color, aroma, and acidity (Malinovski *et al.*, 2012; Borghezan *et al.*, 2014; Marcon Filho *et al.*, 2015).

The results for stomatal conductance (g_s), transpiration rate (*E*), and intercellular CO_2 concentration (Ci) showed significant differences only during the 2019/20 harvest, with the highest values at altitude of 1329 meters, reaching, respectively, 1.05 mol.m^{-2}.s^{-1}; 8.75 mmol.m^{-2}.s^{-1} and 369 μmol.mol^{-1} for the Aromera variety. These results represent an average increment of 425.00% in g_s; 122.08% in *E*, and 30.39% in Ci in comparison to the altitude of 1000 meters. In comparison to the altitude of 774 meters, the increments were 288.89%; 57.09%, and 14.60%, respectively (Table V). Reducing g_s at lower altitudes can be a strategy to avoid heat stress at high temperatures, as it reduces the transpiration rate, ensuring better water management. As Avila *et al.*, (2016) observed, this mechanism can cost plants less nutrient absorption since absorption and transport of these occur by mass

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flow. By reducing stomatal conductance, the entry of CO_2 into the mesophyll is also restricted and, consequently, its concentration in this tissue.

Table V. Gas exchange of the Aromera, Calardis Blanc, and Felicia varieties, grown in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil, during the 2018/19 and 2019/20 harvests.

Altitude		774m	1000m	1329m	774m	1000m	1329m
Harvests	Varieties	Net photosynthesis (*A*) (μmol.m^{-2}.s^{-1})			Stomatal Conductance (g_s) (mol.m^{-2}.s^{-1})		
2018/19	Aromera	12.66Ba(*)	13.76Ba	15.30Aa	0.38Aa	0.35Ab	0.35Ab
	Calardis Blanc	12.76Ba	13.15Ba	15.21Aa	0.45ABa	0.50Aa	0.35Bb
	Felicia	12.40Ba	13.45Ba	15.00Aa	0.41Aa	0.46Aab	0.45Aa
2019/20	Aromera	13.34Ba	8.67Cb	14.32Aa	0.27Bb	0.20Bb	1.05Aa
	Calardis Blanc	11.66Bb	10.05Ca	14.04Aa	0.39Ba	0.28Ba	0.92Aab
	Felicia	11.76Bb	10.21Ca	13.02Ab	0.38Ba	0.19Bb	0.79Ab
Harvests	Varieties	Transpiration Rate (*E*) (mmol.m^{-2}.s^{-1})			Intercellular CO_2 Concentration (Ci) (μmol.mol^{-1})		
2018/19	Aromera	7.33Aa	6.03Ab	6.36Ab	328Aa	293Aa	325Aa
	Calardis Blanc	8.09Aa	7.99Aa	7.23Aab	328Aa	331Aa	325Ba
	Felicia	7.59Aa	7.57Aab	7.40Aa	328Aa	330Aa	326Aa
2019/20	Aromera	5.57Bb	3.94Cb	8.75Aa	322Ba	283Ca	369Aa
	Calardis Blanc	6.96Ba	5.28Ca	8.55Aa	317Ba	297Ca	363Ab
	Felicia	6.96Ba	4.03Cb	8.00Aa	326Ba	290Ca	370Aa

*Means followed by the same letters lowercase within same column and uppercase within same row are not statistically different by Tukey's test (p>0.05).

Considering that fluorescence is one of the pillars of studies on photosynthetic regulation and plant responses to the environment (Stirbet *et al.*, 2018), Table VI shows the results of the analysis of the maximum quantum yield of photosystem II (PSII) (Fv/Fm), non-photochemical quenching (NPQ), effective quantum yield of photosystem II (PSII) (ΔF/Fm') and apparent electron transport rate (ETR). Only the Fv/Fm values found at an altitude of 1329 meters remained within the range of 0.83 to 0.85, according to Kalaji *et al.* (2014), indicating that the plant can overcome stresses, preventing photoinhibition damage to the photosystems. Under optimal environmental conditions, the Fv/Fm ratio is between 0.83 and 0.85 in C3 plants and 0.78 in C4 plants (Kalaji *et al.*, 2014).

According to what was studied by Cambrollé *et al.*, (2015), under stress conditions, *Vitis vinifera* ssp. sylvestris may show a reduction in Fv/Fm, which indicates a reduction in the proportion of open reaction centers. Under these conditions, there is usually an increase in NPQ. Plants dissipate light in heat, thus protecting the leaf from light-induced damage. These results agree with what was observed, at an altitude of 1000 meters, the NPQ values reached 3.02 for Felicia in the 2019/20 harvest; and were, on average, 1.93 higher than at 774 meters and 3.43 higher than at 1329 meters (Table VI). The NPQ

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represents the energy loss to the environment in the form of heat in the xanthophyll cycle, which is not used in the photochemical process of photosynthesis. The leaves do not use all the energy from the radiation, dissipating it, aiming to reduce possible photo-oxidative damage to chlorophylls and the photosynthetic apparatus. The non-photochemical dissipation ranges from zero to infinity, but it usually gives values between 0 and 10 (Kalaji *et al.*, 2014).

The values of ΔF/Fm' did not differ statistically between the varieties at the exact altitudes. However, there was variation between altitudes, with the mean values being 0.66 at 1329 meters, 0.41 at 1000 meters, and 0.58 at 1000 meters (Table VI). According to Kalaji *et al.*, (2014), the value of ΔF/Fm' can vary between zero and the value Fv/Fm. Dos Anjos *et al.*, (2012), when studying the light acclimatization of tree species from the Brazilian Atlantic Forest, found a decrease in ΔF/Fm' values in full sun, an indication that part of the absorbed light was not converted into photochemical energy, not being used, possibly causing its dissipation in the form of heat.

The apparent electron transport rate (ETR) represents the relative number of electrons passing through PSII during steady-state photosynthesis. Therefore, it has a direct relationship with the photosynthetic activity of plants. Significantly different values were observed at the three altitudes, and the altitude of 1329 meters presented, on average, values 1.45 higher than those of the altitude of 774 meters and 2.86 higher than those of the altitude of 1000 meters (Table VI). Cambrollé *et al.*, (2015) and Tiecher *et al.*, (2017) analyzed *V. vinifera* ssp. sylvestris and vine seedlings observed that the decreases in ETR values might be related to the reduction of photosynthetic pigments and damage caused to the photosynthetic apparatus of plants.

Table VI. Chlorophyll *a* fluorescence of Aromera, Calardis Blanc, and Felicia varieties, grown in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil, during the 2018/19 and 2019/20 harvests.

Altitude -		774m	1000m	1329m	774m	1000m	1329m
Harvests	Varieties	Maximum Quantum Yield of Photosystem II (Fv/Fm)			Non-Photochemical Quenching (NPQ)		
2018/19	Aromera	0.79Aa(*)	0.65Bb	0.85Aa	0.82Ba	2.00Aa	0.79Ba
	Calardis Blanc	0.78Aa	0.72Aa	0.84Aa	0.71Ba	1.98Aa	0.70Ba
	Felicia	0.78Aa	0.70Aa	0.84Aa	0.95Ba	1.84Aa	0.75Ba
2019/20	Aromera	0.81Aa	0.60Bab	0.85Aa	1.88Ba	2.89Aa	0.57Cb
	Calardis Blanc	0.80Aa	0.70Ba	0.83Aa	1.83Ba	2.29Aa	0.70Ca
	Felicia	0.75Bb	0.68Aa	0.84Aa	1.85Ba	3.02Aa	0.68Ca
Harvests	Varieties	Effective Quantum Yield of Photosystem II (ΔF/Fm')			Apparent Electron Transport Rate (ETR)		
2018/19	Aromera	0.57Ba	0.43Ca	0.65Aa	61.18Ba	50.78Ca	72.87Aa
	Calardis Blanc	0.59Ba	0.44Ca	0.68Aa	55.15Bb	49.94Ca	63.20Aa

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	Felicia	0.56Ba	0.36Ca	0.68Aa	56.11Bb	48.46Ca	63.96Aa
2019/20	Aromera	0.61Ba	0.39Aa	0.72Aa	109.65Ba	40.89Ca	176.25Aa
	Calardis Blanc	0.58Aa	0.43Ba	0.69Aa	105.50Ba	42.19Ca	205.55Aa
	Felicia	0.56Ba	0.41Ca	0.54Aa	99.98Ba	41.73Ca	164.03Aa

*Means followed by the same letters lowercase within same column and uppercase within same row are not statistically different by Tukey's test ($p>0.05$).

Since, at the intermediate altitude, in both harvests, but more markedly in the 2019/20, there is a decrease in the values of *A*, Fv/Fm, ΔF/Fm', ETR, and an increase in NPQ, it is inferred that varieties are developing under stress. In plants grown in this location, stress was initially observed from the decline in vigor with leaf spots related to the problem in the absorption and transport of water and nutrients, resulting in sudden drought and death of some plants during the 2019/20 harvest. From the visual diagnosis, samples were collected for the pathological analysis, which identified the fungi *Cylindrocarpon* sp., *Fusarium* sp., and *Verticillium* sp. in the roots; *Rosellinia* sp., *Cylindrocarpon* sp., and *Fusarium* sp. on the lap; *Fusarium* sp. in the stem, in addition to the presence of the nematode *Pratylenchus* sp.. From the gas exchange analysis during 2018/19 harvest, the stress caused by the diseases was not perceived visually. Nevertheless, the varieties were probably changing the photosynthetic apparatus since the results of chlorophyll *a* fluorescence show similar responses to those observed in the 2019/20 harvest.

The vines also presented a certain degree of stress at 774 meters, but less accentuated, not reaching the leaves visually. The lower *A*, ΔF/Fm' and ETR values compared to the altitude of 1329 meters indicate that development may be hampered. The lower *A* may have been directly influenced by incident radiation since at an altitude of 774 meters, it is, on average, 86.78 $W.m^{-2}$ and 144.70$W.m^{-2}$ lower in spring and summer, respectively, in comparison to the altitude of 1329 meters. In addition, the altitude of 774 meters has higher temperatures and lower wind speeds, which directly increases the canopy temperature and the varieties vegetative growth.

The average leaf area (LA) at the lowest altitude in the three varieties was 31.13% higher than the highest altitude during the analyzed period (Table VII). Except for the period in which the altitude of 1000 meters suffered from the incidence of diseases, there is an inverse relationship between LA and *A* in both harvests and the three varieties. Teixeira *et al.*, (2018) studied leaf morphoanatomical characteristics in white vine cultivars with different geographical origins. They observed that leaves with smaller individual leaf areas have more efficient control of temperature than larger leaves, as the aerodynamic conductance decreases with the leaf area, directly influencing gas exchange.

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The Aromera, Calardis Blanc, and Felicia varieties produced an average number of bunches per plant of 24.62; 54.86, and 39.44, and an average weight of bunches per plant of 2.27; 4.76, and 6.14 kg, respectively (Table VII), which is approximately equivalent to 7,718; 16,184 and 20,876 kg.ha^{-1}. Schmitt *et al.*, (2020), when analyzing the Cabernet Sauvignon and Chardonnay varieties at an altitude of 1329 meters, found mean values in the number of bunches of 15.67 and 16.6, respectively. While Bender *et al.*, (2021), when analyzing 11 clones of the Bordô variety grown at an altitude of 774 meters, found results ranging from 6.9 to 10 kg for the average weight of bunches per plant. At an altitude of 1000 meters, due to the attack of diseases in the 2019/20 harvest, there was a reduction in the number and weight of bunches per plant in all varieties. Aromera was the variety most affected in terms of the number of bunches, and Felicia was the one that suffered the most with the decrease in bunch weight, both reducing the analyses mentioned above by five times in comparison to the previous harvest (Table VII).

The Ravaz Index (RI) was used to analyze the balance of the vineyards between vegetative growth and productive characteristics under different growing conditions (Table VII). Plants with RI between 3 and 10 can be considered acceptable concerning carbohydrate balance; between 5 and 7 plants represent an ideal carbohydrate distribution condition in the plant; below 3, plants present excess vigor and, above ten, plants have excess production. From these parameters, it is possible to infer that the Aromera variety has excess vigor, as it presented values below 3 in all altitudes and harvests, indicating that pruning in this variety has to be intensified. In contrast, the other varieties and altitudes remained with values between acceptable and ideal about the balance of carbohydrates.

The total acidity (TA) values at the time of harvest of the three studied varieties ranged from 63.7 mEq.L^{-1}, in the Calardis Blanc variety cultivated at the highest altitude in 2019/20, to 129.6 mEq.L^{-1}, in the Aromera variety cultivated at the lowest altitude during the 2018/19 harvest (Table VII). These results corroborate the Brazilian legislation, which stipulates that the physicochemical parameters of TA for fine and sparkling wine must be between 40.0 and 130.0 meq.L^{-1} (Brasil, 2018).

In all wine-growing countries of the world, cluster rot, gray rot, or botrytis is present, which is produced by the development of *Botrytis cinerea* in grapes, a necrotrophic pathogenic fungus responsible for enormous economic losses each year in agriculture, especially in the production of grape and wine (Steel *et al.*, 2013). It is probably responsible for the worst disease affecting viticulture, as it gives rise to several serious problems, such

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as contamination with undesirable microorganisms (Barata *et al.*, 2012; Lleixà *et al.*, 2018), settling and filtration problems (Jadhav and Gupta, 2016), presence of Ochratoxin A (Ponsone *et al.*, 2012), musty odors (Steel *et al.*, 2013) and a deterioration in the *perlage* of sparkling wines (Cilindre *et al.*, 2007). All of this cause the wine to lose its quality (Ky *et al.*, 2012; Lopez-Pinar *et al.*, 2017). However, the damage that laccase causes to wine color is undoubtedly one of the biggest concerns (Ky *et al.*, 2012; Steel *et al.*, 2013; Vignault *et al.*, 2019). In grapevine fungal infections, relative humidity plays a crucial role in the ability to cause disease from pathogens, spore germination, conidiation, mycelial growth, and mycotoxin production (Solairaj *et al.*, 2021). As a result of these more pronounced climatic conditions during the summer at the lowest altitude, during the 2018/19 harvest, the early harvest was carried out to maintain the quality of the grapes for winemaking.

Due to this, the varieties did not reach quality standards at altitudes of 774 and 1000 meters during the 2018/19 harvest, as they presented values from 14.29 to 17.50°Brix. Brazilian legislation requires that the ethylic degree be between 19 and 24°Brix in the fine table and sparkling wines (Brasil, 2018). The varieties reached the ideal levels of soluble solids (SS) and acidity for vinification during the 2018/19 harvest at the highest altitude for Aromera and Calardis Blanc. During the 2019/20 harvest, except for the Calardis Blanc variety at the highest altitude, all varieties at all altitudes reached these levels (Table VII). Despite the damage to the number and weight of bunches, the SS content increased during the 2019/20 harvest at an altitude of 1000 meters. This factor can be explained by the greater sun exposure on the berries caused by the drastic reduction in leaf area results similar to those verified by Gatti *et al.*, (2012). They analyzed the effects of thinning bunches and removing leaves before flowering on growth and grape composition in the Sangiovese variety.

Table VII. Leaf area, number of bunches, bunch weight, Ravaz Index, soluble solids, and total acidity of the Aromera, Calardis Blanc, and Felicia varieties, cultivated in the municipalities of Videira (774 meters), Curitibanos (1000 meters), and Água Doce (1329 meters), Santa Catarina, Brazil, during the 2018/19 and 2019/20 harvests.

Altitude -		774m	1000m	1329m	774m	1000m	1329m
Harvests	Varieties	Leaf Area (cm²)			Number of bunches		
2018/19	Aromera	197Ab(*)	166Ba	148Bb	24.1Aa	25.8Ac	26.4Ab
	Calardis Blanc	206Aa	174Ba	158Bab	47.4Ba	67.3Aa	56.2Ba
	Felicia	206Aa	169Ba	160Ba	33.3Ca	39.0Bb	56.7Aa
2019/20	Aromera	190Ab	44Cb	150Bb	23.3Ab	4.5Bb	23.5Ac
	Calardis Blanc	227Aa	65Ca	169Bab	51.1Aa	29.5Ba	52.3Aa
	Felicia	246Aa	73Ca	194Ba	31.6Ab	11.1Bb	36.6Ab

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Harvests	Varieties	Ravaz Index (Kg cachos.kg lenha de poda^{-1})			Bunches Weight (Kg.planta^{-1})		
2018/19	Aromera	0.62Bc	1.93Ac	1.44Ab	2.16Ab	1.67Ac	2.48Ab
	Calardis Blanc	6.07Ab	6.03Ab	3.93Ba	5.08Aa	4.79Ab	2.26Bb
	Felicia	8.00Aa	8.28Aa	4.93Bb	5.22Ba	6.03Aa	6.81Aa
2019/20	Aromera	2.98Ab	2.27Ab	2.17Ab	2.86Ab	0.55Ba	2.16Ab
	Calardis Blanc	7.85Aa	6.55Aa	7.52Aa	5.75Aa	1.52Ba	5.92Aa
	Felicia	7.94Aa	3.47Bb	7.28Ba	6.45Aa	1.17Ba	6.21Aa
Harvests	Varieties	Soluble Solids (ºBrix)			Total Acidity (mEq.L^{-1})		
2018/19	Aromera	14.29Ba	17.50Aa	18.42Aab	129.6Aa	98.5Ba	90.1Ca
	Calardis Blanc	14.32Ca	17.28Ba	19.10Aa	101.3Ab	92.1Ab	76.7Bb
	Felicia	15.30Ba	16.30ABa	17.42Ab	89.4Ab	92.8Ab	85.9Aa
2019/20	Aromera	18.44Bb	22.32Aa	18.52Ba	94.1Ba	102.6Aa	100.1ABa
	Calardis Blanc	18.52Ab	19.62Ab	17.78Aa	67.0Bc	98.8Ab	63.7Bb
	Felicia	20.72Aa	18.32Bb	18.78Ba	78.5Bb	95.0Ac	73.3Bb

*Means followed by the same letters lowercase within same column and uppercase within same row are not statistically different by Tukey's test ($p>0.05$).

CONCLUSION

The study of climatic conditions found in the southern region of Brazil reinforces the importance of including varieties resistant to the major incident fungal diseases to perpetuated a more sustainable and competitive wine production. From the analysis of the PIWI varieties grown at different altitudes, it was possible to conclude that the higher altitude provided more favorable climatic conditions for developing these varieties. Compared to cultivation at the lowest altitude, the highest altitude provided a total average decrease of 3.35ºC in temperatures during spring and summer, of 99% in NPQ, 23.03% in LA, 4.65% in bunch weight, and 15% in TA; and average total increment of 144.70 W.m^{-2} in radiation, 13.5 Km.h^{-1} in wind speed, 17% in *A*, 81% in g_s, 9% in *E*, 8% in Ci, 8% in Fv/Fm, 14% in ΔF/Fm', 45% in ETR, 19% in the number of bunches and 9% in SS.

It is concluded that the greater vigor observed at a lower altitude, caused by climatic conditions of high temperature, humidity, precipitation, and low wind speed, may have influenced incident radiation, gas exchange, and chlorophyll *a* fluorescence. Therefore, to maintain productivity and oenological quality, the canopy management must be studied and adapted to the planting location, mainly when the varieties are grown in similar, less favorable, or extreme climatic conditions.

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