

Evolution of the grapevine bud dormancy under different thermal regimes

Evolução da dormência de gemas de videira sob diferentes regimes térmicos

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Abstract

Fluctuations in winter chilling availability impact bud dormancy and budburst. This study aimed to quantify the thermal requirements during dormancy for 'Italia' grape, under different thermal regimes. Cuttings of grapevines 'Itália' were collected in Veranópolis-RS, on April/2017, with zero chilling hours (CH). The cuttings were exposed to constant (7.2°C) or alternating (7.2 and 18°C for 12/12h, 12/12h or 18/6h) temperatures, or yet, a constant temperature (7.2°C) or alternating (7.2 and 18°C for 12/12h), combined with one or two days a week at 25°C. Periodically, part of the cuttings was transferred to 25°C for daily budburst evaluation. The induction of the endodormancy (dormancy induced by cold) occurred with 200 CH, independent of the thermal regime, and the overcoming with 300 HF, at 7.2°C. The alternating heat of 18°C in the middle of the cold did not affect the process of overcoming endodormancy. Heat waves during endodormancy resulted in an increased CH to overcome the bud dormancy. The negative effect of high temperature depended on the exposure time. Chilling was partly cancelled during dormancy when the heat wave lasted 36 continuous hours or more. These evidences serve as basis for new model adjustments for budburst prediction, especially for regions with mild and irregular winters, such as those of Southern Brazil.

Key words: Budburst. Chilling hours. Endodormancy. *Vitis vinifera*.

Resumo

Flutuações na disponibilidade de frio hibernal afetam a dormência e brotação de gemas. Objetivou-se quantificar os requerimentos térmicos durante a dormência de gemas de videira 'Itália' sob diferentes regimes térmicos. Estacas de videiras 'Itália' foram coletadas em Veranópolis-RS, em abril/2017, com zero horas de frio (HF). As estacas foram submetidas à temperatura constante (7,2°C) ou alternada (7,2 e 18°C, por 6/18h, 12/12h ou 18/6h), ou ainda, temperatura constante (7,2°C) ou alternada (7,2 e 18°C, por 12/12h), combinadas com um ou dois dias por semana a 25°C. Periodicamente, parte das estacas de cada tratamento foi transferida para 25°C, para avaliação diária da brotação. A entrada em endodormência (dormência controlada pelo frio) ocorreu com 200 HF, independentemente do regime térmico, e a saída com 300 HF, a 7,2°C. O calor alternado de 18°C em meio ao frio não se mostrou prejudicial à superação da endodormência. Ondas de calor de 25°C na endodormência resultaram em aumento de HF para superação do processo. O efeito negativo das altas temperaturas dependeu do tempo de exposição. O calor anulou parcialmente o frio após um período de 36 horas contínuas no período de dormência. Tais evidências servem de base para ajustes de modelos para predição da brotação, principalmente para regiões com frio hibernal ameno e irregular, como do Sul do Brasil.

Palavras-chave: Brotação. Endodormência. Horas de frio. *Vitis vinifera*.

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Introduction

In temperate and subtropical climates, grapevines go through a bud dormancy period during autumn and winter, during which temporary suspension of visible plant growth occurs. According to Lang et al. (1987), there are three types of dormancy: paradormancy, endodormancy, and ecodormancy. While in paradormancy, the lack of bud development is the result of the influence of another plant organ, for instance, the apical dominance. In endodormancy, sprout inhibition results from a series of biochemical and physiological events at the meristematic level or in nearby tissues. These are triggered by the perception of an environmental stimulus, usually low temperatures, changes in photoperiod, or both. This type of dormancy can have different lengths and intensities (depth), being overcome with the accumulation of a certain number of chilling hours $\leq 7.2^{\circ}\text{C}$ (CH) during the autumn and winter. This type of dormancy varies with species and cultivar. After overcoming endodormancy, budburst becomes dependent on the environmental conditions of the spring, mainly temperature and water availability. This stage is called ecodormancy.

According to Biasi et al. (2010) and Marafon et al. (2011), in a productive system, failure to supply the needed chilling during endodormancy can cause serious phenological problems, including insufficient and/or uneven plant budburst and flowering. Poor or uneven budburst may compromise both yield and branch distribution of the fruit plants, whereas poor and uneven flowering may lead to loss of pollination, and consequently, loss of efficiency in orchard production.

In southern Brazil, it is normal to experience significant variation between years regarding the availability of cold during autumn and winter. As such, most cycles require the use of techniques to artificially overcome endodormancy, and chemical products, including hydrogen cyanamide, are used to “compensate” for the lack of winter cold (BOTELHO et al., 2010). Chemical management

to overcome endodormancy presents substantial limitations concerning the technical tools used to make decisions regarding the need to exercise the control at the beginning of each cycle. These limitations force technicians and producers to apply controls indiscriminately to induce budburst every year because of the fear of loss. The compounds available for overcoming dormancy are highly toxic to humans, as well as the environment. In addition, these controls are frequently used without the proper technical knowledge or adequate safety criteria of the producer (FELIPPETO et al., 2013; ANZANELLO et al., 2014a). In Italy, the high risk of poisoning by hydrogen cyanamide, which is the main product used for overcoming dormancy worldwide including Brazil, led to the temporary suspension of its sale, as well as a review of regulation by the European Union authorities (SETTIMI et al., 2005). In Brazil, given there are no recommendations regarding chemical treatments to overcome dormancy, only small variations in doses between cycles are suggested, based on the number of chilling hours for each winter period. Nevertheless, the calculation of chilling hours is primarily based on dormancy experiments and models that were conducted under weather conditions with genotypes that are different from that of southern Brazil.

The models used to quantify the cold accumulated each year to overcome dormancy have been in development since the 1930s. They allow for the characterization of thermal requirements of genotypes and aid in decisions regarding sprout inducing treatments of fruit plants (NIGHTINGALE; BLAKE, 1934). The most well-known and used models are Chilling Hours $\leq 7.2^{\circ}\text{C}$ (WEINBERGER, 1950), the Utah Model (RICHARDSON et al., 1974), and the North Carolina Model (SHALTOUT; UNRATH, 1983). These models are adjusted to North American climatic conditions, which are characterized by constant and regular autumns and winters, and for the most part, were developed for peach and apple

tree crops (WEINBERGER, 1950; RICHARDSON et al., 1974; SHALTOU; UNRATH, 1983). For the climatic conditions of southern Brazil (the main production hub for grapes), where significant thermal fluctuations occur during the autumn and winter, these models are unreliable and mostly imprecise (FELIPPETO et al., 2013; ANZANELLO et al., 2014a). Therefore, under these conditions, heat and cold fluctuations affecting the onset and overcoming of dormancy should be better studied; in particular, the effects of characterization of alternating heat during the winter period should be addressed. This will enable the adjustment and/or development of models more adapted to the prediction of Brazilian crop budburst potential.

The problems with dormancy are aggravated by the expansion of grapevine cultivation areas into largely marginal regions (CAMPOY et al., 2011). Additionally, there is the possibility of increasing global temperature because of the intensification of the greenhouse effect, and a progressive downward trend in the availability of chilling hours in the State of Rio Grande do Sul (CARDOSO et al., 2012). The change in climate could directly affect the endodormancy stage, as well as the budburst ability of grapevines, as well as other temperate fruit species.

Given the importance of grape production in southern Brazil and the lack of technical and scientific information regarding the influence of the local climate on the dormancy process, it is crucial to generate basic information to create more effective models to predict the budburst responses of the grapevine crop. Starting with a precise model, producers and technicians will have access to an important tool to make decisions with respect to budburst management techniques, thereby decreasing expenses and increasing the efficiency of chemical treatments, in terms of both dosage and environmental impact.

The goal of this study was to characterize the thermal needs of grapevines (*Vitis vinifera* 'Italia')

buds during the dormancy period under different thermal regimens, to aid in the adjustment and/or development of a new model to predict crop budburst.

Material and Methods

The experimental material collected for the dormancy progress assessment was composed of branches formed during the vegetative stage of 'Italia' grapevines. The plant cuttings were collected from the middle of the branches and measured 40 to 60 cm in length, were approximately 1 cm in diameter, and contained 5 buds per cutting, without leaves. When selecting the material to be collected, bud maturity (well closed buds) and the health and vigor of the cuttings were considered, prioritizing those with intermediate growth.

The cuttings were sampled in a commercial vineyard located in the municipality of Veranópolis - RS, in the Serra Gaúcha, during winter 2017, with zero CH in the field. After removing the branches, they were wrapped in bundles using newspaper, moistened, placed in plastic bags, and transported to the Departamento de Diagnóstico e Pesquisa Agropecuária (DDPA), of the Secretaria da Agricultura Pecuária e Desenvolvimento Rural do Estado do RS (SEAPDR), in Veranópolis-RS, to evaluate bud dormancy under controlled conditions. The cuttings went through a cleaning process, as per the methodology proposed by Anzanello et al. (2014b).

After disinfestation, the cuttings were distributed in bundles containing 10 units, packed using black plastic film, and subjected to different thermal regimens in Electrolab heated incubator chambers, model EL202 in the dark. Cortázar-Atauri et al. (2009) claimed that photoperiodism does not present a specific function in bud endodormancy in grapevines, given that the process is exclusively dependent on temperature. The task of characterizing the thermal needs was subdivided into two stages.

The first consisted of evaluating different periods of bud exposure under a constant cold temperature, whereas the second stage tested temperature (cold/hot) variation, with different amplitudes and time lengths.

The study was subdivided into three experiments. In experiment 1, the goal was to determine the cold needed to induce and overcome endodormancy. The 'Italia' grapevine buds were subjected to one cold temperature (7.2 °C) and seven exposure times (0, 100, 200, 300, 400, 500, and 600 CH). Then, they were subjected to a temperature of 25 °C to evaluate budburst chronology. In experiment 2, the goal was to evaluate the effects of oscillating temperatures (cold/hot) over the endodormancy period. The temperature consisted of daily cycles of 6/18 h, 12/12 h, or 18/6 h, ranging between 7.2 °C and 18 °C. The buds went through these three treatments over increasing periods (0, 100, 200, 300, 400, 500, and 600 CH), and were then subjected to a temperature of 25 °C for budburst chronology assessment. The calculation of CH was completed for chilling hours maintained at 7.2 °C. In experiment 3, the goal was to evaluate the effect of different heat waves over the endodormancy period. Four treatments (thermal regimens) were used, with a constant (7.2 °C) or alternating (7.2 °C and 18 °C, for 12/12 h) temperature, combined with one (24 h) or two days (48 h) per week at 25 °C. The buds were subjected to the treatments of increasing periods (0, 100, 200, 300, 400, 500, and 600 CH), and were then exposed to a constant temperature of 25 °C to evaluate budburst chronology. The calculation of CH was completed for chilling hours maintained at 7.2 °C.

To experience the heat (25 °C) for the induction and evaluation of budburst, the cuttings were placed in heated incubator chambers, with a photoperiod of 12 h light and 12 h darkness. The cuttings were processed by cutting one beveled end, approximately 1 cm above the bud, and the other approximately 7 cm below the first cut, resulting in a single

node cutting (with only one bud). The cuttings were planted in pots with wet phenolic foam. The experimental design consisted of a randomized block design to control the effect of differences in air circulation inside incubator chambers. For each temperature combination and exposure time, there were three repetitions (3 pots with 10 cuttings).

Cutting irrigation inside the incubator chambers was conducted by adding water every 48–72 h. The water was replenished enough to saturate the phenolic foam, avoiding the accumulation of free water. During the period of budburst induction and evaluation, the emergence of diseases was prevented by using the defensive chemicals pyrimethamine and tebuconazole (systemic) and iprodione and captan (contact), which were sprayed at a dosage of 1.5 to 2.0 ml L⁻¹, except for tebuconazole, for which the dosage was 1.0 ml L⁻¹. The application was conducted every 14 to 21 days, switching between contact and systemic products.

Budburst evaluation at a constant temperature of 25 °C was performed every 2–3 days until day 35, and the budburst date for each bud at the green tip stage was recorded (CARVALHO et al., 2010). The data regarding final budburst rate (percentage of sprouted buds), precocity (number of days before the first bud sprouted), and uniformity (number of days between the first and last bud sprouted) were subjected to variance analysis. The results yielded significant differences according to the F-test, when their means were compared by the Tukey test, at a significance level of 5%.

Results and Discussion

The induction of bud endodormancy under controlled conditions, signaled by the decrease in the budburst capacity at different temperatures and exposure times, occurred with 200 CH for all thermal regimens (constant and oscillating) (Figure 1). This indicated that, for grapevine buds to trigger endodormancy, a few hours in the cold every

day is sufficient. The effectiveness of oscillating temperatures in endodormancy induction was reported by Cook et al. (2005) and Alldermann et al. (2011), who claimed that the effect of low temperatures in the middle of mild autumn temperatures provides the signal that triggers the bud dormancy mechanism. This effect generates changes in the bud meristematic tissues, which conditions their ability to withstand cold. Anzanello et al. (2014a) also noticed a positive effect of alternating cold and hot (3 to 15 °C) temperatures on the buds of apple cultivars Castel and Royal Gala when they entered endodormancy, with an efficiency similar to that of a constant temperature of 3 °C.

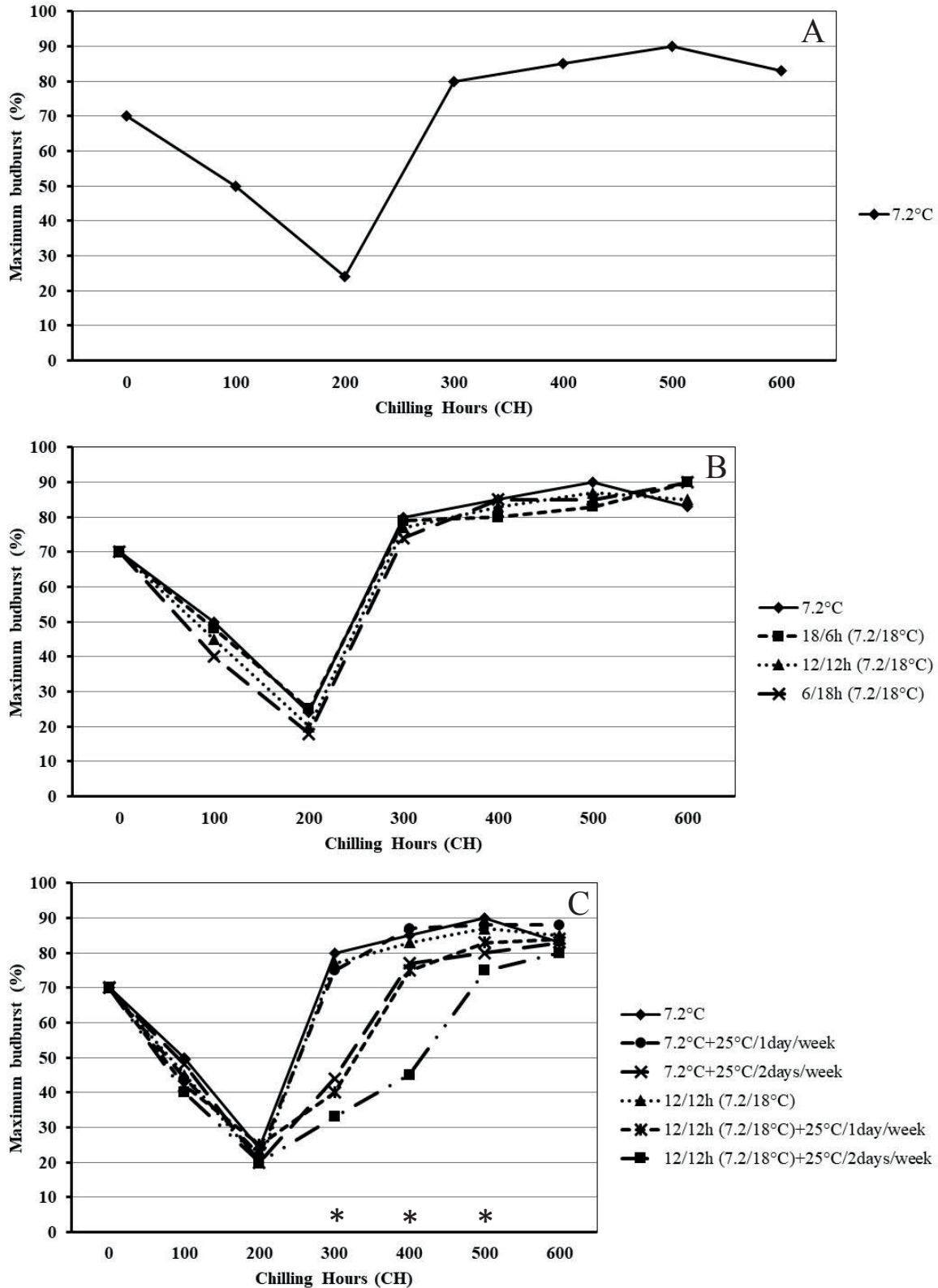
Overcoming endodormancy corresponded to the post-induction period of the process (minimum budburst) until the time at which budburst levels were suitable again (above 70%), signaling the metabolic trigger for plant growth after the winter period. By computing the cold, 300 CH were required for the 'Italia' grapevine to overcome endodormancy at a constant temperature (7.2 °C) (Figure 1A). Under the alternating temperature regimen of 7.2/18 °C for 6/18 h, 12/12 h, and 18/6 h, the cold requirement for the 'Italia' grapevine did not change, and dormancy was also overcome with 300 CH (Figure 1B). Based on these results, it was confirmed that the alternating heat of 18 °C for 6, 12, or 18 h in the middle of the cold season was not harmful to the process of overcoming endodormancy.

Heat waves of 25 °C during endodormancy resulted in an increase in the number of CH needed to meet the requirements of the buds to overcome

endodormancy, yielding an effect that varied with the thermal regimen. In constant cold conditions of 7.2 °C, exposures of 24 h at 25 °C per week did not increase the number of CH needed to overcome endodormancy. However, exposures of 48 h at 25 °C per week increased the cold requirement of the crop up to 100 h (Figure 1C). Under alternating temperatures of 7.2/18 °C and exposures at 25 °C for 24 and 48 h per week, the cold needed to overcome endodormancy increased by approximately 100 and 200 h, respectively. Three groups were formed among the treatments (Figure 1C). Thermal regimens at a constant 3 °C, with alternating 3/15 °C (12/12 h) and 7.2 °C once per week at 25 °C did not result in a different requirement, on average, of 300 CH to overcome dormancy. Treatments at 7.2 °C with two days per week at 25 °C and the alternating regimen 7.2/18 °C (12/12 h) with one day per week at 25 °C increased the number to 400 CH to break dormancy. The alternating treatment 7.2/18 °C (12/12 h) with two days per week at 25 °C required approximately 500 CH to break dormancy.

According to Luedeling and Brown (2011), temperature fluctuations made it necessary to increase the cold to overcome dormancy in fruit plants in temperate climates. Erez and Lavee (1971) confirmed that the negative effect of high temperatures depended on their intensity and duration. Exposure from 2 to 4 h at 21 °C was not harmful. Nevertheless, when they lasted longer than 8 h, they had a nullifying effect on the chilling hours. At higher temperatures (24 °C), 2 h of exposure could yield a canceling effect.

Figure 1. Maximum budburst in ‘Italia’ grapevines subjected to a constant temperature of 7.2 °C (A), alternating temperatures of 7.2/18 °C (B), and heat waves in the middle of the cold during the dormancy period (C). Veranópolis, 2017. Significant differences in maximum budburst within each chilling period, according to the results of Tukey’s test ($p < 0.05$), are marked with a (*).



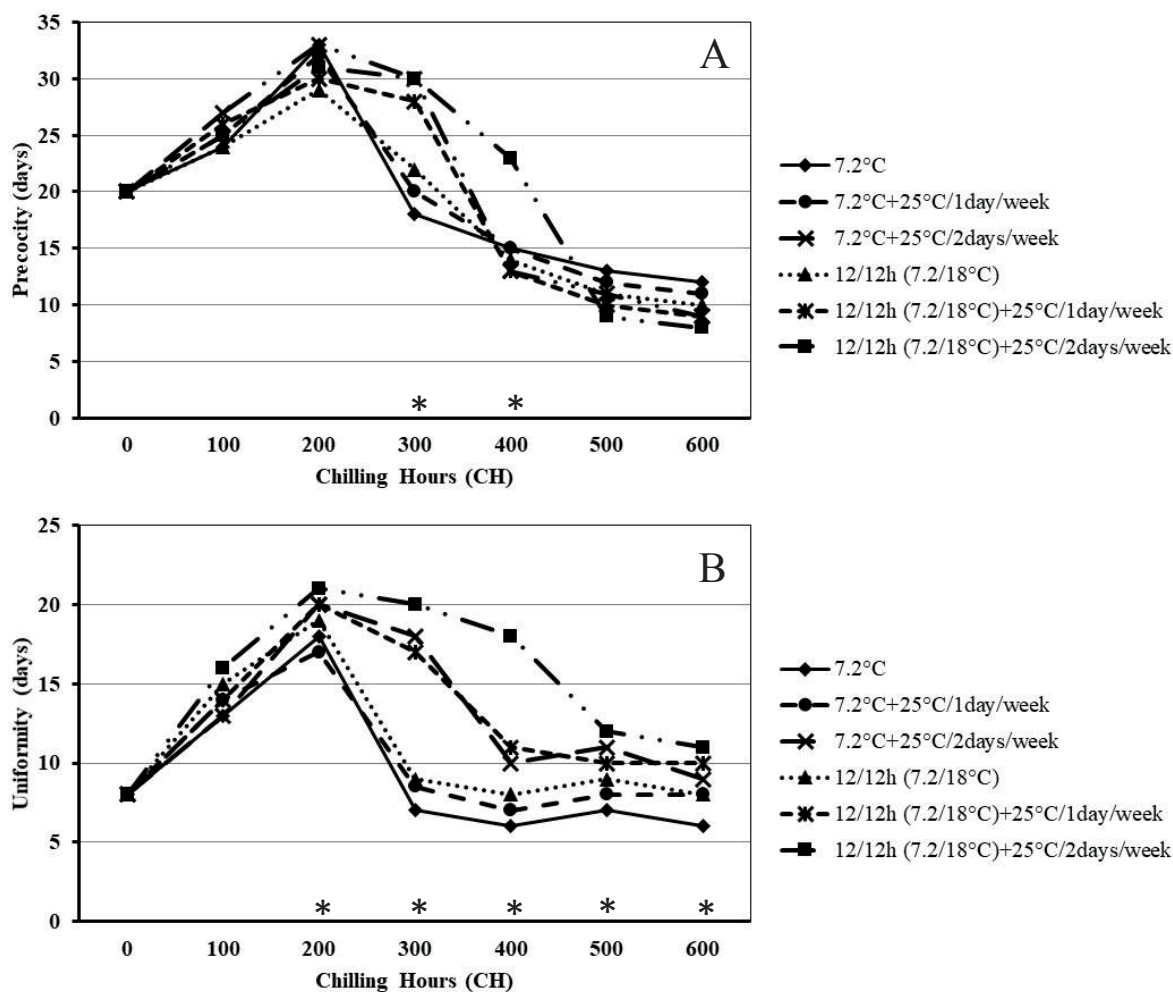
Based on the results obtained in this study, the heat nullified part of the effect of the accumulated cold after continuous heat exposure for 36 h or more during endodormancy. In the cyclic temperature regimen, 24 or 48 h at 25 °C were always accompanied by 12 h at 18 °C, totaling 36 or 60 h with a lack of cold. These conditions reversed the dormancy process and increased the crop cold requirements (Figures 1A and 1C), which points to the need for adjustments to the models used for the prediction of budburst. These include the Utah (RICHARDSON et al., 1974) and North Carolina (SHALTOUT; UNRATH, 1983) models, which consist of hourly conversions of high temperatures to negative chilling units (UF). As such, a model that partially cancels the effect of the cold only after 36 h or more of heat exposure should be proposed. This would break the dormancy process except with the influence of high temperatures before this time period. This result also clashes with the results of the Modified North Carolina and Utah Models, which were proposed by Ebert et al. (1986), according to whom high temperatures result in negative cold accumulation only for a few days (4 days), after the last positive cold unit is recorded. Results similar to those of this study were obtained by Anzanello et al. (2014a), who, while working with apple trees, found that heat waves (≥ 15 °C) that lasted more than 36 h during endodormancy resulted in an increase in the number of chilling hours required to overcome dormancy. This evidence serves as a basis for adjusting models to predict budburst, mainly in regions with mild and irregular winter cold, such as southern Brazil.

According to Haverroth et al. (2009), budburst time is related to the depth of the bud endodormancy state. This trend can be observed in budburst precocity for the different thermal regimens (Figure 2B). Up to the period of maximum endodormancy and/or maintenance of this stage, the number of days before

budburst increased, decreasing the rate at which endodormancy was overcome. It was also observed that budburst precocity increased with higher cold duration, mainly after overcoming endodormancy (Figure 2B). Legave et al. (2008), while working with apple trees, reported that the need for heat units for the beginning of the vegetative stage was lower with a greater number of hours of accumulated cold, confirming the obtained results. While working with a crop of Carrick pears, under controlled conditions, Herter et al. (2001) also observed a decrease in the average budburst time with an increase in the time exposed to cold. For oscillatory regimens or heat waves, budburst precocity was higher when compared to the treatment with a constant of 7.2 °C, regardless of the time exposed to cold, after meeting the cold requirements during endodormancy. This fact was justified by the oscillatory and heat wave regimens, which provide a higher thermal sum for achieving budburst when compared to the regimen with a constant cold temperature (7.2°C) in the endodormancy period.

Regarding budburst uniformity, the response was different from the evolution of cold during endodormancy (Figure 2B). Throughout the dormancy-breaking process, the uniformity of values showed greater variability. After this period, the budburst appeared more uniform and regular. Generally, the behavior of the crop demonstrated the importance of low temperatures during the dormancy period to overcome it and ensure uniformity in budburst during the spring. According to Campoy et al. (2011) and Marafon et al. (2011), meeting the cold requirements during endodormancy is essential for avoiding phenological disorders, including insufficient and/or uneven budburst and flowering. Carvalho and Biasi (2012) reported that, with the increase in cold duration, the buds present a more limited (uniform) budburst time, which is in agreement with the results of this study.

Figure 2. Precocity (A) and budburst uniformity (B) of the ‘Italia’ grapevine buds subjected to a constant temperature of 7.2 °C, alternating temperatures of 7.2/18 °C, and heat waves in the middle of the cold during the dormancy period. Veranópolis, 2017. Significant differences in precocity and uniformity within each chilling period according to the Tukey test ($p < 0.05$) are marked with a (*).



The obtained results displayed greater uniformity and a shorter average budburst time; the increase of cold accumulation was also achieved by other authors (BIASI et al., 2010; BRUCKNER et al., 2010) working with different fruit species in temperature climates. However, this information is not considered by budburst prediction models, making research necessary to increase the effectiveness and applicability of field models.

Conclusions

The induction of endodormancy for ‘Italia’ grapevines occurred at 200 CH, regardless of the thermal regimen used over the dormancy period. Daily cycles of fluctuations between cold (7.2 °C) and mild (18 °C) temperatures were not harmful to the dormancy process. Heat waves that lasted more than 36 h during dormancy resulted in an increase in the number of chilling hours required to overcome dormancy. Overcoming endodormancy for ‘Italia’ grapevines occurred after 300 CH (7.2 °C

or alternating 7.2/18 °C) or after 400/500 CH under thermal regimes with heat waves that lasted longer than 36 h during dormancy. Periods with longer exposure to cold during dormancy favor the precocity of budburst. Uniformity in budburst is more regular after supplying the cold required for the genotype.

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