UNIVERSIDADE FEDERAL DE LAVRAS

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PLANTING DENSITY FOR CHARDONNAY GRAPEVINES IN THE SOUTH OF MINAS GERAIS

LAVRAS - MG

2017

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Dissertação apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Agronomia/Fitotecnia, área de concentração em Produção Vegetal, para a obtenção do título de Doutor.

Dr. Murillo de Albuquerque Regina

Orientador

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Ficha catalográfica elaborada pelo Sistema de Geração de Ficha Catalográfica da Biblioteca Universitária da UFLA, com dados informados pelo(a) próprio(a) autor(a).

de Almeida, Luciana Wilhelm.
Planting Density for Chardonnay Grapevines in the South of Minas Gerais / Luciana Wilhelm de Almeida. - 2017.
49 p. : il.

Orientador(a): Murillo Regina de Albuquerque.

Dissertação (mestrado acadêmico) - Universidade Federal de Lavras, 2017.
Bibliografia.

1. Planting density. 2. Shoot thinning. 3. Sparkling wines. I. Universidade Federal de Lavras. II. Título.

LUCIANA WILHELM DE ALMEIDA

PLANTING DENSITY FOR CHARDONNAY GRAPEVINES IN THE SOUTH OF MINAS GERAIS

DENSIDADE DE PLANTIO PARA VIDEIRA CHARDONNAY NO SUL DE MINAS GERAIS

Dissertação apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Agronomia/Fitotecnia, área de concentração em Produção Vegetal, para a obtenção do título de Doutor.

APROVADA em 10 de agosto de 2017.

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LAVRAS - MG
2017

AGRADECIMENTOS

À Universidade Federal de Lavras, especialmente ao Departamento de Agricultura, pela oportunidade de realização deste curso de mestrado em Fitotecnia.

Ao Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPQ) pela concessão da bolsa de estudos.

À Fundação de Amparo à Pesquisa de Minas Gerais (FAPEMIG) pelo auxílio financeiro na execução do projeto de pesquisa.

À Empresa de Pesquisa Agropecuária de Minas Gerais (EPAMIG) pela infraestrutura cedida para realização do experimento e avaliações.

Ao pesquisador Murillo de Albuquerque Regina, meu orientador, pelos ensinamentos em vitivinicultura que foram essenciais para o desenvolvimento deste trabalho.

À pesquisadora Cláudia Rita de Souza pelos ensinamentos de ecofisiologia da videira, pelas discussões intermináveis em fisiologia, densidade e desbrota em videira, pelas sugestões e correções realizadas para melhoria deste trabalho, e pelo apoio e incentivo na pesquisa.

À pesquisadora Renata Vieira da Mota, por sua ajuda e ensinamento sobre qualidade da uva e do vinho, pelas correções e sugestões realizadas ao trabalho, pelas caronas e apoio durante a elaboração do trabalho.

Aos meus pais Cleide Wilhelm de Almeida e Waldemar de Almeida Junior e minha irmã Anna Flora pelo apoio, incentivo, dedicação, amizade e amor durante todos os anos da minha vida.

Ao meu namorado Frederico pelo apoio, incentivo e carinho e por todas as conversas intermináveis sobre viticultura.

Muito obrigada!

ABSTRACT

Brazilian southeast has potential to produce sparkling wines from Chardonnay variety in sites above 900 m. Sparkling wine quality is relate to vineyard climate and management, since they affect the vine development and grape, must and base wine compositions. In this study five planting densities, with and without shoot thinning, were compared in order to optimize field productivity and based wines quality. Chardonnay vines were subjected to five planting densities (2,000, 2,667, 4,000, 5,333 and 8,000 pl ha^{-1}) combined to not thinning and thinning treatments, for each planting density vines were thinned to 20, 15, 10, 8 and 5 shoots per plant, respectively. Vegetative vigor, yield and grape components were analyzed for two years and must and base wine composition for one year. Increasing planting density, lower bud fruitfulness, cluster number and yield were accessed and a positive linear relation was found between grape, must and wine acidity and shading effect induced by denser treatments. Shoot thinning increased bud fruitfulness, enhancing productivity only for the highest density, and no differences were found among not thinning planting densities varying from 2,667 pl ha^{-1} to 8,000 pl ha^{-1}. Planting densities from 2,667 pl ha^{-1} to 5,333 pl ha^{-1}, not thinnined, are recommended to southeastern region of Brazil, since they showed better productivities and necessary acidity to provide freshness in final sparkling wines.

Keywords: Planting density. *Vitis vinífera*. Shoot thinning. Canopy management. Grape quality.

RESUMO

Regiões do Sudeste brasileiro situadas acima de 900 m de altitude apresentam grande potencial para o plantio de Chardonnay para produção de vinhos espumantes. A qualidade do vinho espumante é afetada pelo clima e pelas práticas culturais adotadas no vinhedo, uma vez que ambos afetam o desenvolvimento da videira e composição da uva, do mosto e do vinho. No presente trabalho cinco densidades de plantio, com e sem desbrota de ramos, foram comparadas a fim de otimizar a produtividade por área, e o impacto na qualidade do vinho base. Videiras da variedade Chardonnay foram submetidas a cinco densidades de plantio, 2,000, 2,667, 4,000, 5,333 e 8,000 pl ha^{-1} e a desbrota ou não de ramos, de maneira que em cada densidade de plantio as plantas foram desbrotadas a 20, 15, 10, 8 e 5 ramos por planta, respectivamente. O vigor vegetativo, a produção e a composição da uva foram analisados por dois anos e a composição do mosto e do vinho base foi analisada somente por um ano. Ao aumentar a densidade de plantio, foi observado menor fertilidade de gemas, número de cachos e produção por planta, e uma relação diretamente proporcional foi observada entre acidez (da uva, do mosto e do vinho base) e o sombreamento causado pelas maiores densidades. A desbrota aumentou a fertilidade de gemas, elevando a produtividade somente para o tratamento mais denso, de 8,000 pl ha^{-1}. Entre densidades de 2,667 e 8,000 pl ha^{-1}, não submetidas à desbrota. Não foram encontradas diferenças em relação à produtividade. Diante disso, o indicado para regiões do Sudeste brasileiro, é a escolha de densidades de plantio entre 2,667 pl ha^{-1} e 5,333 pl ha^{-1} uma vez que apresentaram melhores produtividades e acidez necessária para proporcionar frescor na bebida final.

Palavras-chave: Densidade de plantio. *Vitis vinífera.* Desbrota. Acidez. Viticultura.

SUMMARY

CHAPTER 1 GENERAL INTRODUCTION

1 INTRODUCTION

Sparkling wine is the fastest growing segment of the wine market, presenting an increase in its production of more than 40% in the last ten years, which is due to the high global demand and consumption (INTERNATIONAL ORGANISATION OF VINE AND WINE - OIV, 2014). The most traditional sparkling wines producers and exporters are France, Italy and Spain; however, the expensive cost of these traditional sparkling wines reflects a low market competition being therefore inconsistent with the growing worldwide demand. Thus, regions that produce this kind of wine at lower prices, such as Brazil, that have been growing their importance in the sparkling wine market, due to better market prices. The enological industry in Brazil is internationally recognize by its sparkling wine quality, and is located mainly in southern Brazil. The quality is directly related to the climate, which has proved to be ideal for such products.

The quality of a sparkling wine is influenced mainly by its acid content, hence benefiting from berries with high acidity and low sugar concentration and pH (JONES et al., 2014). In south and southeast of Brazil, the climate provides such needed features, since the maturation period of the vines coincides with the rainy season, forcing the harvest of grapes that have not yet maturated. This guarantee not only the freshness of the sparkling wine but also the lightness since the phenolic maturation does not take place. Not only the climate affect and interfere in grapes composition but also vineyard managements can improve the necessary acidity, sugar and pH levels.

As South region of Brazil, Minas Gerais, a southeastern state of Brazil, presents the same climate for regions above 900m, enabling the production of sparkling wines from Chardonnay variety in these highlands site as reported by Regina et al. (2010). However Minas Gerais main research regards to regions below 900m that produce still wines from the double-pruning management (FÁVERO et al., 2011). Relating to sparkling wines Mendonça et al. (2016), studying different pruning methods, recommended Cordon Royat as most appropriate for Chardonnay vines in southeastern regions. Besides this research, there are no scientific verifications for Chardonnay in southeastern Brazil. Indeed there are few worldwide published studies relating vineyard management and its influence in sparkling wine quality (JONES et al., 2014).

Viticultural practices, like fertilization, planting density, clones, pruning method, shoot and cluster thinning, affects vine performance, development, and consequently grape composition. The planting density affects the level of exploitation of the environment by the vine, since it can provides wider spaces allowing individually larger vines or it can provides narrower spaces restricting the individual vine growth but allowing higher number of vines per area, hence greater exploitation per area. This will also affects the competition between vines at underground level by the root system for water and nutrients, and at aboveground level by the vegetative part for light (CARBONNEAU; DELOIRE; JAILLARD, 2007).

The vineyard load can be manage by pruning, shoot thinning or cluster thinning. It affects the crop load, productivity and the source-sink balance within the vine, improving most importantly the vine balance, and consequently the grape quality. Shoot thinning however can be more effective in achieving the balance between vegetative growth and yield, reducing the canopy density and improving canopy microclimate (JUNQUERA et al., 2011; REYNOLDS et al., 1994a, 1994b; SMART et al., 1988). The ideal vineyard load or shoot number per area depends directly of the cultivar and vineyard site climate (REYNOLDS; MOLEK; SAVIGNY, 2005).

The choice of planting density for a commercial vineyard is extremely important, since it influences directly the initial investment and the final productivity for the winegrower. As few reports regarding planting density effects on vine, grape and wine have been published, even more considering for sparkling wines production, many winegrowers do this choice empirically. In addition, studies reporting the effect of planting density and vineyard load for Brazilian conditions have not yet been performed. Making it evident the need to debate in such matter. Therefore, this work proposes to compare five vine densities with and without shoot thinning and its effects on vine performance and on grape and wine quality, in order to recommend the most appropriate management for Chardonnay variety to produce sparkling wines, in southeastern Brazil.

2 BIBLIOGRAPHIC REVIEW

2.1 Sparkling wine production in the world

Vines are widely cultivated in all continents of the world and considering wine production, Italy, France, Spain and United States stand out, producing more than half of the total for 2016, which was 259 million of hectoliters (OIV, 2016). The sparkling wines represent 7% of world wine production, that is 18,13 million of hectoliters, mostly boosted for France, Italy, Germany and Spain They produce together 74% of the total sparkling wine produced in the world. In the last ten years, an increase of more than 40% in the production of this type of wine was reached, mainly to its high global demand and the growing consumption (OIV, 2014).

The most traditional country in sparkling wines production is France with the Appelation d'Origine Controlée of Champagne. This region, located in the north of France, suffers from constant rainfall, a low seasonal temperature range and strong winter frosts, which conditions are not ideal for still wine production (AOC Champagne). These variations of climate, combined with the difficulty they had in removing the carbon dioxide bubbles produced, and the British nobility preferences for these sparkling wines, in the late 1600s, led to the creation of one of the most glamorous drinks in the world (JONES et al., 2014).

The varieties used in AOC Champagne to produce the champagne sparkling wine are Chardonnay, Pinot Meunier and Pinot Noir. Per year, are produced an average of 309 millions of bottles, profiting 4,7 billions of Euros, and more than 50% of it regards to exportation. Indeed, France alone is responsible for 53% of the total exports in terms of value in the world, due to high product prices (COMITÉ CHAMPAGNE, 2017; OIV, 2014).

For the production of quality sparkling wine, the grapes must be yield with relatively low pH, of 3.20, high titratable acidity, up until 12 g L^{-1} of tartaric acid, and a low soluble sugar, ranging from 16º to 19º brix, in order to have a fresh beverage and with a final low alcohol content of 10% to 13%. Therefore, the grapes must be harvested before reaching full maturity, when compared to table/still wines, which will ensure the acidity and lower alcohol potential (FAVERO et al., 2006). Hence, for sparkling wines production the cultivated area should enhance such particularities.

Since the 2008 crisis, alternatives for Champagne wines have been gaining space on the market, mainly due to their lower prices and good quality. This search is mostly in United States and United Kingdom, which are the biggest importers of sparkling wines.

Consequently, countries that produce this type of product, at lower prices, have benefited, for instance, Spain with Cava and Italy with its Prosecco sparkling wines (OIV, 2014). Others countries, Brazil included, have grown their importance in sparkling wine market, both for increasing in production and for their high quality sparkling wines.

2.2 Brazilian regions for sparkling wine

Brazilian viticulture occupies 85,000 hectares distributed in three regions: South, Southeast and Northeast of Brazil. The South region is where 76% of the country vineyards are, and where is also the mainly Brazilian oenological industry, the Northeast region concentrates 12.2% of the vineyards, while the Southeast 11% (ANUÁRIO…, 2015). Therefore, the South region, mainly, the Rio Grande do Sul state, is responsible for more than 90% of the Brazilian wine production, sparkling wines included.

2.2.1 South Region

The South region comprises two major poles in the production of sparkling wines, which are the Serra Gaúcha and Serra do Sudeste regions. The first region (Serra Gaúcha), is a very rugged terrain, has 32.000 hectares of vineyard, which less of 20% is cultivated with *Vitis vinifera* varieties and the rest is mainly cultivated with American varieties, the average annual rainfall is 1,700 mm, 17.2 ºC for average temperatures and relative humidity of 76%. The second region has topography less hilly than the Serra Gaucha region, allowing mechanization, in addition to lower annual rainfall (average of 1,000mm) providing better climatic conditions, and fewer incidences of diseases. Such conditions lead many wineries from Serra Gaucha to invest and cultivate grapes in this region (PROTAS; CAMARGO; MELLO, 2006).

For these regions, as well as the entire South hemisphere, the maturation and harvest period occur from December to the end of January and the beginning of February. The high rainy season affecting the sugar accumulation, the reduction of organic acids and also the phenolic maturation of the grapes which lead to poor full-bodied wines and little varietal expression. Despite such particularities are not desired for still wine production, they are not an obstacle to the production of sparkling wines (FAVERO et al, 2006; MANDELLI et al., 2003; REGINA et al., 2010).

The varieties used at these two regions are Chardonnay, Pinot Noir, Riesling Italic, Moscato Giallo and Moscato Branco. For base wines destined to Champenoise or Charmat method, the first three varieties are most commonly used, while the Moscato group grapes are designed for Asti method (muscat sparkling wine) (CALIARI; ROSIER; LUIZ, 2013). The management consists in an earlier harvest, avoiding therefore fungus attack and maintaining the acidity and low sugar content desired for a base wine.

Due to these climatic characteristics in addition to the high oenological technology in the South of Brazil, Brazilian sparkling wines are recognized for their freshness and high quality. Including winning many awards worldwide and being listed as one of the top ten sparkling wines in the world along with great Champagnes, according to Effervescents du Monde Confrontation Internationale des Meilleurs Vins Effervescents of 2016.

The recognition and elevated quality of the national sparkling wine led to an increase of its commercialization and consumption. The domestic market commercialized in 2016, 16.8 million of liters of sparkling wine, of which 12.4 million liters provided by Champenoise or Charmat method and 4.4 million of Asti method. Even though the muscat sparkling wines have a minor volume of commerce, considering trades since 2011, an increase of 49.7% was registered for this segment. Which means more than double of the increase for traditional sparkling wines, that was 21.6%. Comparing to the still wine market, that decreased 1.7% in the same period, both sparkling wines indicate opportunity in a rising market (UNIÃO BRASILEIRA DE VITIVINICULTURA - UVIBRA, 2017).

2.2.2 Southeast Region

Brazilian southeast region involves São Paulo and Minas Gerais as the representative producer states. Their viticulture for fine wine production are relatively young, having less than 20 years of development. The first projects and works developed in order to prove its viability took place in Minas Gerais with Syrah variety for winter harvest, and with Chardonnay and Pinot for summer harvest (AMORIM; FAVERO; REGINA, 2005; FAVERO et al., 2008; MOTA et al., 2010; REGINA et al., 2010). In consideration of the few years of advance and that is still in an establishment phase, it is certain that there is a high need to generate a lot of knowledge and technologies for this growing viticulture.

In Brazilian southeast region, there are two segments for viticulture for fine wines. In general, the first is for areas located up to 1000m high, where the double pruning management is applied for “Syrah” variety. Therefore, the grapes are harvested in winter season, when we have the lowest index of rainfall in this region, ensuring better climatic conditions for grapes maturation. These grapes produced by this method are destined for still wines production. (DIAS et al., 2012; FAVERO et al., 2011; MOTA et al., 2010; REGINA et al., 2010; SOUZA et al., 2015).

The second segment is for areas above 1000m high, where the double-pruning management does not apply. These high-altitude regions are destined for cultivation of Chardonnay variety for sparkling wine production (REGINA et al., 2010). High altitudes regions are successful regions for cultivate *Vitis vinifera* varieties, due to the altitude variation influence, as take place in the highlands of Santa Catarina state (FAVERO et al., 2006; MALINOVSKI et al., 2012; ROSIER, 2006). The altitude variation in the South of Minas Gerais influences, mostly, the average temperature, reducing them. This mild temperature retains the acidity as less acid malic degrades, this is important because it ensures the freshness required for sparkling wines (REGINA et al., 2010).

Another important feature of these high regions is the similarity with the South of Brazil climate. As already discussed, the maturation and harvest season occur in summer, which is the month of most rainfall for this region. In order to escape from the rains and avoid diseases, the grapes are harvested before full maturation, with high titratable acidity, low pH and soluble solids, which are the ideal for sparkling wines production. In addition, the closest proximity of South of Minas Gerais to the major consumer's centers as São Paulo, Rio de Janeiro e Belo Horizonte cities is more advantageous than the South region proximity (REGINA et al., 2010). That is an enormous potential for the production of sparkling wines.

The production in Champagne region is regulated by the Appelation d'Origine Controlée (AOC). Therefore planting and viticulture practice such as varieties, vine spacing, pruning and yield per hectare are all regulated. The same occurs for Cava wines, produced in Spain. But for others regions in the world, like Southeast of Brazil, the vine management and the winemaking process its decided by the wine-grower (JONES et al., 2014). Many viticulture practices can be applied in order to produce quality grapes and wines, but little is known about which techniques and practices are most appropriate for Brazilian Southeast high-altitude areas. Regina et al. (2010), found that the Chardonnay variety had satisfactory soluble solids and total acidity balance and was therefore indicated for the production of sparkling wines for Caldas, a high altitude city of South of Minas Gerais. As to the pruning management, Mendonça et al. (2016) testing two types of pruning, Cordon Royat and Guyot, in Divinolândia city at 1.280m of altitude, affirms that Cordon Royat had the best results for vegetative vigor and productivity in Chardonnay grapes, without impair its quality.

Nevertheless, it is still necessary to study others variables such as clones, training systems, rootstocks and planting density, in order to create an effective management for the production of Chardonnay in highlands of the Southeast of Brazil.

2.3 Chardonnay cultivar

Chardonnay is a white cultivar, originally from Burgundy, France. It is the fifth cultivar most planted in the world, covering 190,126 ha worldwide, with United States, France and Australia presenting the largest planted areas (ANDERSON; ARYAL, 2013). This clearly characterizes Chardonnay as a high phenotypic plasticity cultivar (GAVRILESCU; BOIS, 2016). Even though it is grown on different regions in the world, it responds better to mild temperatures, typical of Northern regions, as its homeland, leading viticulture in tropical countries to seek areas with these particularities.

In Brazil, the cultivar was first implanted in São Roque in the state of São Paulo, but it was in Rio Grande do Sul state (introduced in 1948), that it best adapted and developed producing quality wine, mostly as base wines for sparkling wines (CAMARGO, 2003; RIZZON; MIELE; SCOPEL, 2009). For Brazilian Southeast its cultivation is recent, taking place preferably in high altitudes lands, due to the drop in average temperature, allowing the Chardonnay cultivar to develop properly, which due to their Northern origin adapted well to these highlands conditions.

Chardonnay cultivar is an early budding variety and presents compact clusters with small and round berries, and also both vine and berries are predisposed to powdery mildew and bunch rot (JACSKON, 2008), which are the two diseases that bring difficulties in Brazilian production, both in South and Southeast of Brazil (BEM et al., 2016). For being an early budding cultivar it is liable to damage due to late frosts (spring frosts), very usual in Serra Gaucha (CAMARGO et al., 2003).

According to Jackson (2008), Chardonnay grapes produce wines with fruity notes, including apple, peach and melon, and provide finesse and elegance to sparkling wines, in addition to a capacity for aging. However, sensorial as physicochemical characteristics vary according to soil, climate and management of the vineyard winemaking process, that is the *terroir*, also to the yeasts used during the fermentation and the in-bottle lees contact duration.

In 1995, this cultivar was introduced in Minas Gerais state by EPAMIG (SOUZA et al., 2002). According to Regina et al. (2010), the high lands present ideal edaphoclimatic conditions for producing Chardonnay for sparkling wines, since these areas present mild temperatures.

2.4 Planting and Vine density

2.4.1 Planting density

The planting density is one of the components of the conduction system, being an important factor that modifies the structure of the vine and therefore its development, however it must not be considered as a solo factor (CARBONNEAU; DELOIRE; JAILLARD, 2007; HIDALGO, 1999).

The planting density refers to the number of vines per hectare, which depends on the space each vine occupies (HIDALGO, 1999), therefore it is directly related with the exploitation by the plant for the environment (CARBONNEAU; DELOIRE; JAILLARD, 2007). This exploitation of the environment comprehend the colonization of the soil by the root system and the colonization of the air by the aboveground part of the plant, that is the trunk, the arms, the shoots and leaves (CARBONNEAU; DELOIRE; JAILLARD, 2007; CHAMPAGNOL, 1984; HIDALGO, 1999). Each vine will achieve different developments due to the different planting densities that they may be subjected, thus affecting the plant physiology of this vines (TODA, 1991).

2.4.1.2 Inter-row and in-row effects

The plant density can present different combinations of plant spacing and row spacing that is different inter-row and in-row distances, and each of them has a different effect on the vine when increasing it or decreasing it. According to Carbonneau, Deloire and Jaillard (2007) the inter-row spacing permits the mechanization of the vineyard and yet presents a minor competition between neighboring plants, even for high densities. The most important effect of inter-row spacing regards the interception of light, that is an effect on the aboveground part; if the distance between the rows are too wide, the interception of radiation by the plants will decrease as more and more light will reach the soil more than it will reach the vines, being a disadvantage. Considering the root system, wider inter-row spacing presents a lower root density with roots concentrated on soil surface. The in-row spacing, differently from inter-row spacing, induces a significant competition between neighboring plants. Increasing the in-row distance, plants will be more distant from each other, empty spaces on the foliage can form causing a misuse of the conduction system and reducing the radiation interception; on the other hand, by decreasing the in-row distance, therefore with closer plants, an overlap of foliage can occur that may cause shading to varying degrees. As for the root system, higher will be the competition for water and nutrients as narrower the in-row distance is. The inter-row distance commonly used vary from 1 m to 4 m and for in-row distance it varies from 1 m to 2 m considering lower densities, and 0,5 m to 0,9 m considering higher densities (CARBONNEAU; DELOIRE; JAILLARD, 2007).

2.4.1.3 Density limits

At first the advantages of planting more plants per hectare that is higher planting densities, are more obvious than planting less plants per hectare (lower densities), since more vines per area will increase the productivity of the vineyard. Nevertheless, it is important to consider the consequences of extreme low number of vines per hectare and extreme high number of vines per hectare.

In general, the ideal planting density when considering lower densities is the one that do not present empty spaces on the vegetative canopy of the espalier, which can be defined as minimum density. When considering higher densities, and characterizing the maximum density, the ideal planting density is the one that despite the competition between plants, it will not reduce the vigor in an extreme and/or negative way, decreasing the profitability of the vineyard (CARBONNEAU; DELOIRE; JAILLARD, 2007; CHAMPAGNOL, 1984). According to Carbonneau, Deloire and Jaillard (2007) the minimum densities vary from 800 to 1,000 vines per hectare for vigorous vineyards, 2,000 vines per hectares for most of the vineyards, 3,000 vines per hectare weak vineyards; the maximum densities vary from 10,000 vines per hectare for most of the vineyards and 8,000 vines per hectare for vineyards subjected to drought.

The planting density creates a confinement on the root system, which can change the soil exploitation by the root system, yet its morphological and biochemical composition, therefore affecting the performance of the vine (HUNTER, 1998; RICHARDS, 1983). Soil conditions together with climate and conduction system determine the most proper planting density.

2.4.1.4 Soil conditions

The maximum expansion and exploitation of the plant's root system are limited by soil fertility (CHAMPAGNOL, 1984). Once the development of the root system affects the aboveground part of the vine, in fertile soil conditions more and more water and nutrients will be distributed by the roots for the aboveground parts, therefore increasing vigor. On the contrary, in poor soils as the roots will not expand as much as in fertile soils, less water and nutrients they will collect and distribute to aboveground level (CHAMPAGNOL, 1984). Thus, the soil fertility, and its water and nutrients availability determine the ideal planting density selected for establishing the vineyard.

Regions with no water deficiency that is high water availability, a high planting density is thought to stimulate root competition for both the nutrients and water, therefore reducing the vegetative vigor and creating a proper condition to reach grape maturity (COPPOLANI, 1994).

For Champagnol (1984), when vineyards are located on hot and sunny climate regions and in non-limiting soil conditions, lower planting densities are recommended. Adopting wider spacing the control of the excess of foliage caused by excess of vigor is fulfilled, therefore the ideal density would be of 1,000 to 2,000 vines/ha. However, for Carbonneau, Deloire and Jaillard (2007) the control of the vigor can be reached by reducing the in-row spacing and stimulating the exposed leaf surface for a maximum value, in this way there would not be a density limit. This narrowing would stimulate root competition for water and nutrients, and by reducing the vigor would create a proper condition to reach grape maturity (CARBONNEAU; DELOIRE; JAILLARD, 2007; COPPOLANI, 1994). The opposite idea is applied to regions with limiting soils conditions. Since both water and nutrients are scarce, the ideal is not to create a competition between plants therefore low planting densities are recommended.

The traditional idea that in non-limiting soil conditions the high plant density stimulates root competition therefore controlling plant vigor is widely adopted in Mediterranean regions. Gallet (1993) presents that from North to South of France the tendency of vine density follows the water availability. For example, in Champagne region as the planting density and many others managements are controlled by INAO (National Institute of Appellation of Origin), the sum of the distance inter-row and in-row must be less than 2.5 m, resulting in an high vine density, of 7,500 to 10,000 vines per hectare, due to the high annual rainfall in this region (COPPOLANI, 1994). The same applies in Spain, planting density is established according to the water regime of each region, so that in regions where the rainfall is limited and soils are poorer the planting density is lower (wider vine spacing), avoiding competition between vines. Accordingly, planting densities in Spain alter from 1,500 to 4,000 vines per hectare (HIDALGO, 1999).

Nevertheless, Bernizzoni et al. (2009) testing different in-row spacing for Barbera cultivar, did not observed the suggested competition due to decreased vine spacing, once did not impaired the expected higher shoot vigor from the lower number of nodes retained at pruning. So, for his conditions the recommended spacing was the narrower one, of 0.9 m between plants, since provided maximum cropping with no adverse effects on grape composition. Yet Hunter (1998) testing different planting densities for Pinot Noir cultivar, concluded that densities between 2,222 vines/ha and 2,500 vines/ha performed better and more balanced than the lower density of 1.111 vines/ha and the higher ones of 5,000, 10,000 and 20,000 vines/ha.

In addition, Reynolds et al. (2004) discusses that the widely carried out idea that grapevines that produce high yields compromise high wine quality and the view that closely spaced grapevines are critical for high-quality wines, are widespread misconception that limit adaptation of new viticultural technology. Moreover many have confirmed the advantages of wider vine spacing, mostly of them performed at new world viticultural countries (HEUVEL et al., 2013; KLIEWER et al., 1988; MORRIS et al., 1984; REYNOLDS; WARDLE; NAYLOR, 1995; REYNOLDS et al, 1996; SHAULIS et al., 1982; SMART, 1988; SMART; SHAULIS; LEMON, 1982a, 1982b).

2.4.2 Vineyard load control

The vineyard load refers to the number of buds per hectare, and its selection must be calculated considering the production objectives of the vineyard, but most importantly its potential. The load most commonly adopted varies from 40,000 to 80,000 buds per hectare (CARBONNEAU; DELOIRE; JAILLARD, 2007). The control is done in order to obtain branches with adequate vigor. The minimum vigor allows the buds a sufficient floral induction and lignified shoots in order to buds survive during winter season and sprout in the following spring season. The maximum vigor is the one that presents imbalance between vegetative growth and productive development that is floral induction; such unbalanced can lead to improper maturation of grapes. The adequate vigor is above minimum vigor and below maximum vigor (TODA, 1991). Another reason to do the control of load is to achieve the adequate balance between vegetative growth and yield, that is known to be directly related to the grape composition and hence the wine quality (BRAVDO et al., 1985; KLIEWER; DOKOOZLIAN, 2005).

The vineyard with a low load will not have its vegetative potential totally exploited, resulting in insufficient leaf area as well as insufficient yield. On the other hand, if the load is high, the vegetative potential will be totally exploited with a maximum of the leaf area, however, with an excessive yield, mainly for the fertile cultivars; therefore the level of reserves will be lower, and it may also impair berry composition. The ideal load is, therefore, the one that allows the vegetative and yield potential of each cultivar without compromising its perenniality. If the load is too low, secondary buds may sprout, which is not ideal since their fertility is reduced; and if the load is excessive, most of the buds may no develop and yet sprout in an ununiformed and weak way in the next season (CHAMPAGNOL, 1984).

The load must be determined by the current vigor and number of branches. When vigor is considered appropriate, with an average length and diameter (for vertical position system, 1.30 m of shoot length and minimum of 1.0 cm of shoot diameter), the number of buds left by winter pruning will be the same as the number of shoots; when vigor is considered excessive, with shoots too long, the number of buds can be increased and if vigor is too low, the number of buds must be diminished (CHAMPAGNOL, 1984).

Regarding to the interaction between vineyard load and planting density, Champagnol (1984) describes that as planting density decreases the effective load per hectare also decreases, despite of the theoretical vineyard load that was left during winter pruning. This decrease in the vineyard load will consequently reduce the number of branches per hectare, due to this an increase in vigor is verify which will provoke higher leaf areas, not being favorable to grape quality. In general, the insufficient effective load, the alteration in leaf area / fruit weight ratio, the overlap of vegetation, are problems faced mostly by vineyards conducted in lower densities and long pruning, reinforcing the importance of adopting an adequate pruning method.

The number of buds retained on a vine depends on some degrees, vine vigor, spacing and training system. The managements that define these numbers of buds (that is the load) are winter pruning and shoot thinning, being the last given as shoot density as well (REYNOLDS et al., 1994). Nonetheless, the control applied per winter pruning not always is sufficient in achieving the balance between vegetative growth and yield (JUNQUERA et al., 2011).

The shoot thinning is applied in order to reduce canopy density, helping to establish balance in the grapevine, by turning it more efficient at radiation interception. As more light reaches the canopy, higher bud fruitfulness, and improved fruit composition it will have. On the other hand, the excess of shoot density can create shade canopies compromising the bud fruitfulness, photosynthesis and grape composition (REYNOLDS et al., 1994; REYNOLDS; MOLEK; SAVIGNY, 2005). Yet, if the shoot thinning is too intense and vigor increases will affect the flower differentiation due to excess of vigor (DRY, 2000).

Most research performed in shoot thinning in order to achieve the best shoot density concluded that 15 to 25 shoots per meter of row had better improvements in the canopy microclimate and in the wines sensory attributes (MURISIER; ZIEGLER, 1991; REYNOLDS et al., 1994a, 1994b; REYNOLDS; MOLEK; SAVIGNY, 2005; SMART, 1988). The shoot thinning can be applied in different phenological growth stages as well, yet when applied after bloom may be superior to early season thinning (after budburst and before bloom). However, the ideal vineyard load or shoot density depend directly on the cultivar and characteristic of the vineyard site (REYNOLDS; MOLEK; SAVIGNY, 2005).

2.4.3 Effects on quality

Planting density among others viticultural practices, as row orientation, conduction system and pruning, affect quality of sparkling wines, once they control cluster exposure to light and vine vigor, which affects fruit flavor, acid content and sugar levels (HANCOCK, 1994; REYNOLDS et al., 1986; RIBÉREAU-GAYON et al., 2007a).

The control of vineyard load is important since it regulates the vigor and the leaf area / fruit weight ratio and therefore the hormonal balance and sugar content on berries. The load favorable to quality is the one that allows an exploitation of the vegetative potential by branches with moderate vigor and a rentable yield with high content of soluble solids on berries and reconstitution of the starch deposit (CHAMPAGNOL, 1984). In addition, this balance is easier to achieve in low or moderate fertile soils with high planting densities than in high fertile soils, in which the vigor must be controlled by forcing a moderate yield volume and reducing the overlap of the crop (CHAMPAGNOL, 1984) that can be achieved by applying shoot thinning.

The higher or lower incidence of light on cluster region, affects the composition of the grapes, most importantly when considering sparkling wine production is the acid malic content, as it is degraded by the increase of light and temperature. So if less light get into the cluster region, due to the shading caused by the overlap of leaves by the higher planting density or higher number of shoots, lower will be the temperature and exposure of cluster, and consequently higher concentration of malic acid in the grapes and its must (KLIEWER; LAKSO, 1975; REYNOLDS et al., 1986). Which were confirmed by Kliewer (1971) who showed that low light increased concentrations of titratable acidity, malic acid and tartaric acid. Therefore, possibly a certain degree of shading may be beneficial for producing base wine for sparkling wines (JONES et al., 2014). Yet, for still wines adopting high densities can be advantageous, since it restricts potassium imports therefore maintain a good acidity level in wines elaborated by these grapes (RIBÉREAU-GAYON et al., 2007b).

The same can be discussed about the light incidence on the canopy. Higher or lower incidence will have an impact on photosynthesis; hence, the sugar accumulation and its supply to mature the clusters, and its storage, that is, starch concentrations (HUNTER et al., 1995). Yet, the shading provoked by smaller vine spacing or excess of shoots per plant can affect inflorescence induction and differentiation, reducing bud fruitfulness for the next cycle (SÁNCHEZ; DOKOOZLIAN, 2005).

Bernizzoni et al. (2009) examining the impacts of in-row spacing for Barbera cultivar concluded that despite the discussed, the in-row vine spacing had no interference in must composition and by increasing the in-row distance of 0.9 m to 1.5 m low effect in modifying vine performance was observed. Reynolds et al. (2004) and Heuvel et al. (2013); stated that fruit composition was not affected by vine spacing as mentioned before.

The shoot thinning applied in Pinot Noir and Cabernet Franc improved canopy microclimate improving leaf and cluster exposure but had limit effects on yield components for both cultivars (REYNOLDS; MOLEK; SAVIGNY, 2005), following the same trend as Riccardi (2008), in which Noiret cultivar composition was not affected by shoot thinning. However, in Chancellor cultivar this same management improved soluble solids by 5% over a 3-year average (MORRIS; MAIN; OSWALD, 2004). Reynolds et al. (1994a) reported that soluble solids was more affected by shoot thinning than titratable acidity for increased shoot densities, due to the higher yields on those treatments, confirming the effect shoot thinning has on vine balance. As for sensory attributes, in higher shoot densities hexenols compounds seem to increase, yet monoterpenes seem to decrease (REYNOLDS et al., 1994b). Reynolds (1994b) reinforce the recommendation of 20 to 25 shoots per meter of row for Riesling cultivar and similar medium-clustered *V. vinifera* (‘Chardonnay’, ‘Pinot Blanc, etc).

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CHAPTER 2: Planting density for Chardonnay grapevines in the South of Minas Gerais

1 INTRODUCTION

In Brazil studies carried out about planting densities and its influence on yield, grape and wine quality have not been published yet. The adopted densities are empirically applied and based most of the time on the training system used (REGINA et al., 1998). This situation evidences the need to carry out studies on planting densities and its effects on grape and wine production and quality, mainly for new viticulture regions such as the Southeast of Brazil, once the choice of the vine density influences not only final production and quality, but also the initial investment by the wine-grower.

The conditions for producing quality wines in Southeast of Brazil are propitious, not only for still wines but also for sparkling wines. The research developed by EPAMIG 'NUTEV-Núcleo Tecnológico Uva e Vinho" regarding still wines elaborated from "Syrah" grapes by double-pruning method are more than consolidated, both in performance in field and vinification methods and aging potential. The success was not only established by the proper climate conditions but also by public-private partnerships that have been taking place since 2001. The successful implementation of a technological package for the production of sparkling wines should follow the same steps. The interest from private public is already a reality, where many producers aim for information on the implementation of vineyards for producing sparkling wines, since it is an increasing market worldwide. Therefore, the need to expand and improve research in potential cultivars, clones, rootstocks, conduction systems, planting density, winemaking methods and enological features, and yet wine marketing features for the sparkling wines market must be fulfilled.

The development in sparkling wine production however is not only about technical features in order to produce quality wines, but also about economical features concerning the profitability of the vineyard, that is making it a sustainable activity. Within these parameters, the planting density adopted has high impact not only in vine performance and berry quality but also in the initial cost, since regards number of plants per area. Since there is no published works about vine density in Brazil, the observation and discussion is based on the traditional thought that in non-limiting soils, that is without restriction of water and nutrients, higher planting densities can be adopted since the competition between plants reduce the individual vigor and increases the productivity without impairing the quality, if a canopy management is applied, like a vineyard load control, which was adopted in this work. Yet, the load control applied can modify the radiation interception by the canopy, reducing the shading effect and modifying the vine balance, therefore impact yield, and mostly grape and wine components hence its quality. The aim of this work was to evaluate the impact of planting densities and shoot thinning on vine performance and on grape and wine quality, and yet recommend the best plant density for Chardonnay cultivar in the highlands of Southeast of Brazil.

2 MATERIAL AND METHODS

2.1 Vine site and experimental design

The vineyard was established in the city of Caldas in the south of Minas Gerais, Brazil (altitude of 1,150m). The vines were seven years old Chardonnay clone 76 -ENTAV-INRA grafted onto 1103 Paulsen. The minimum and maximum annual average temperatures are 12.1ºC and 25.1ºC, the annual average precipitation is 1,500mm most of it during the summer season (December to March) (TONIETTO; VIANELLO; REGINA, 2006).

The first two years, 2014 and 2015, the experiment consisted in five planting densities, with 8 replications, completely randomized. The five vine densities had the same row spacing of 2.5 m, and between plants the distance varied in 2.0 m, 1.5 m, 1.0 m, 0.75 m and 0.50 m, resulting in 2,000 (T1), 2,667 (T2), 4,000 (T3), 5,333 (T4) and 8,000 (T5) plants per hectare, respectively. Evaluations for these initial cycles consisted on leaf layer number, yield components (except for berry weight and number) and grape pH, total soluble solids and acidity.

Based on initial results of the previous cycles, a shoot thinning treatment was also applied in 2016 and 2017 seasons. So that the experiment design changed for a 5 x 2 subdivided plot, with the five planting densities as the main plot and as subplot, the shoot thinning treatment. Part of the vineyard was thinned to the same number of shoots per hectare, stipulated at 40,000 shoots per hectare, and part was unthinned. In order to reach the 40,000 shoots per hectare, all plants of each planting density were thinned to different numbers of shoots per plant. So that, vines in 2,000 pl ha^{-1}were thinned to 20 shoots per vine, in 2,667 pl.ha^{-1} to 15 shoots per vine, in 4,000 pl.ha^{-1} to 10 shoots per vine, in 5,333 pl.ha^{-1} to 8 shoots per vine and in 8,000 pl.ha^{-1} to 5 shoots per vine, as showed in table 1. Each plot was one replication, thus one plant per replication.

Table 1 - Planting density, in-row spacing, area per plant and number of shoots per hectare for not thinned and thinned treatments, for Chardonnay variety, Caldas - MG, 2017.

Planting density	In-row spacing (m)	Area per plant (m²)	Vineyard bud load		Shoot number per plant	
			Unthinned	Thinned	Unthinned	Thinned
2,000 (T1)	2.00	5.00	74,000	40,000	37	20
2,667 (T2)	1.50	3.75	79,980	40,000	30	15
4,000 (T3)	1.00	2.50	80,000	40,000	20	10
5,333 (T4)	0.75	1.87	95,994	40,000	18	8
8,000 (T5)	0.50	1.25	120,000	40,000	15	5

The vines were trained in a vertical shoot positioning trellis system, with bilateral cordons and double spur pruned. The shoot thinning was done by removal of shoots after budburst and before the shoots achieved the first wired. All cultural practices were applied equally for all treatments, in accordance with standard commercial practices for Chardonnay cultivar.

In October 2015 a hail rain occurred impairing 2015 production and the development of vines for the subsequent cycle, of 2016. For this reason, the present work addresses to 2014 and 2017 cycles.

2.2 Vine vigor analysis

Leaf area per plant and per row meter was measured before shoot trimming. For measuring the leaf area per plant, four vines per treatment were used and for measuring the leaf area per row meter, six random linear meter were used in each treatment. Single leaf area was estimated by the equation $y = -14,68 + 1,73x + 0,30\ x^2$ (REGINA et al., 2000), in which 'y' is the estimated single leaf area and 'x' the sum of the lengths of the two main lateral veins. The average shoot leaf area was calculated by multiplying the total number of leaves per shoot by the average single leaf area. Average single leaf area was obtained from eight to ten leaves per shoot and the average shoot leaf area from four shoots per vine/liner meter. The total leaf area per vine was calculated by multiplying the average shoot leaf area by the total number of shoots per vine or per meter.

Leaf layer number (LLN) was measured after *véraison*, as described by Smart and Robinson (1991). Sixteen plants per treatment were used, with four insertions (two in each cordon) per plant, totalizing in sixty-four insertions per treatment. For quantifying the vegetative and reproductive balance, leaf area/fruit weight ratio ($m^2\ kg^{-1}$) was calculated. In order to quantify the plant storage compound, starch was quantified on dried and powered samples of trunk as described in Souza et al. (2015). Samples were collected two months after harvest.

2.3 Yield components

The bud fruitfulness was obtained during the full bloom. It was calculated, dividing the number of inflorescence per number of branches, of eight plants for each treatment.

At harvest, yield and total cluster number were recorded for eight plants for each treatment. The productivity per hectare was estimated multiplying the average yield of the eight plants, for the total number of vines per hectare for each treatment

2.4 Grape and must composition

Berries were crushed by hand, and its must analyzed for pH, titratable acidity (TA) and soluble solids (ºBrix). For measuring pH, TA and soluble solids a digital potentiometer (Micronal, B472), titration with 0.1N NaOH with phenolphthalein as an indicator and a portable digital refractometer (ATAGO Pal 1) were used, respectively.

A sample of must was collected to quantify the tartaric and malic acids. Both were determined in the acid fraction obtained after passage of the sample in an anion exchange resin Bio-Rex 5 (Bio Rad Labs) (MCCORD; TROUSDALE; RYU, 1984). As internal standard was used 10 µL of Isobutyric acid (2,5 µg/ µL) for each 1 mL aliquot of the sample, being added before the sample was passed through the resin. A 5 µL aliquot of the filtered acid fraction (Millex 0,45 µm filter) was injected into an Agilent 1260 Infinity liquid chromatograph equipped with a SupelcoGel C-610H column (Supelco, 30 cm x 7.8 mm) adjusted to 30ºC, and diode array detector (DAD) at 210 nm. The isocratic run was performed with aqueous 0,5% phosphoric acid solution as the mobile phase. The identification and quantification of the chromatograms were based on calibration curves with standard solution of tartaric and malic acids

2.5 Winemaking

After harvest, the grapes were stored in a cool chamber for 24h at 10ºC. For each treatment, 25.2 kg of grapes were destemmed by hand and thereafter crushed and its must was separated from the solids part and placed in 13,25 L Pirex® glass carboy, and then added 2 g per hectoliter of pectolitic enzyme COAPECT VRC® and 100 mg SO_2 kg^{-1} for all must. The cold settled occurred at 10ºC for 48h, following by the racking for a 13,25 L Pirex® glass carboy and for all treatments must presented an average density of 1,0693. The clear must was inoculated with rehydrated wine yeast Maurivin PDM® 20 $g.hl^{-1}$ (*Saccharomyses cerevisiae bayanus*), corrected with glucose to achieve 11% alcohol and added with 20 $g.hl^{-1}$ of fermentation activator Actimax Vit®.

The density was measured daily during alcoholic fermentation at 17ºC. When density reached constantly 990 and the residual sugar were below 3 g.L^{-1} the wine was racked to remove lees and added with 50 mg.L^{-1} potassium metabisulfite and 80 g.hl^{-1} of bentonite Microcal Alpha®. The wines stayed 12 days in cold stabilization and then racked to remove the clarification lees, still not limpid, more 30 g.hl^{-1} of bentonite Microcal Alpha® was added followed by 12 days of clarification. The final racking was on January 22.

2.6 Base wine analysis

The base wine analyses were performed after the final racking. Total acidity (g L^{-1} tartaric acid), volatile acidity (g L^{-1} acetic acid), pH, alcohol(%), crude extract (g L^{-1}), residual sugars (g L^{-1}) and ashes (g L^{-1}) analyses were also performed in order to detect if the wine was within standards of Brazilian law (AMERINE; OUGH, 1980; BRASIL, 1986).

2.7 Statistical analysis

All data were tested by analysis of variance (ANOVA), for 2014 cycle with one factor (planting density) and for 2017 cycle with two factors (planting density and shoot thinning). The treatments mean values were compared by Scott-Knott test at 5% significance level.

3 RESULTS AND DISCUSSION

3.1 Vine vigor

In not thinning treatments, the lowest density presented higher values for leaf area per plant, and lower values were presented by the denser treatment of 8,000 (figure 1a). The highest values of the lowest density treatment can be explained by the longest arms of its vines, since the in-row spacing is larger, vines have more growing space and more shoots, hence more leaves per plant. The opposite explain the lower leaf area per plant for the 8,000 vines ha^{-1} treatment. Kliewer, Wolpert e Benz (2000), found similar effect by reducing the in-row spacing of Cabernet Sauvignon cultivar vines from 3 and 2 meters to 1 meter, resulting in less shoots per vine. The highest planting density showed higher LLN for 2014 and 2017 cycle (figure 1c and table 2), and both years showed the same tendency of lower LLN values for lowest densities (2,000 and 2,667 pl ha^{-1}). In addition, the leaf area per row meter (figure 1b) presented similar results to LLN, with the 8,000 pl ha^{-1} treatment showing the higher area, and lower LLN values for the lowest densities. The increase of LLN and leaf area per row meter as the planting density increases is due to larger numbers of vines per meter and larger number of shoots per meter with overlapping of foliage. These results suggest shaded canopy, mainly observed in the 8,000 plants ha^{-1} treatment. In addition, the lower values of LLN and leaf area per row meter when distributed through the larger arms of the vines in the 2,000 pl ha^{-1} treatment, may contribute to form gaps in the vegetative canopy, since this vines have less shoots per meter.

When thinning treatment was applied the leaf area per plant (figure 1a) presented same tendency to not thinning treatments, with higher values showed by the lowest planting density and lower values showed by the higher density treatment. In addition, the thinning treatment had no effect on leaf area per row meter and little variation in LLN. The decrease showed in leaf area per plant and LLN from 4,000 to 8,000 pl ha^{-1}, and in leaf area per row meter for 8,000 pl $ha^{-}1$, can be explained by the reduced number of shoots per plant and reduced number of leaves per plant, induced by the thinning treatment. When the management of shoot thinning is applied, the excessive increase of shoot number per meter induced by the higher number of vines per hectare is attenuated, as observed in the leaf area per row meter and LLN results under thinning treatment, which suggest that these vines present less shading canopies.

These results agrees with Reynolds et al. (1994a) that by increasing number of shoots, denser canopies were observed and yet, lowering the number of shoots more gaps on the canopy were detected. According to Carbonneau, Deloire and Jaillard (2007) the minimum density is the one that does not present empty spaces on the vegetative canopy, below this limit although those blank spaces are observed impairing light interception by the plant. Results showed by 2,000 pl ha^{-1} may indicate that this treatment is below minimum density limit.

The leaf area: yield (m^2 kg^{-1}) ratio (figure 1D) was only affected by planting density treatment. The ratio was significantly increased by higher planting densities (5,333 and 8,000). The leaf area:yield ratio is a way to measure vine balance, and accordingly to Kliewer and Dokoozlian (2005) the values indicating adequate balance between vegetative and reproductive part range from 0,8 to 1,2 m^2 kg^{-1} of fruit, which would indicate that for this work all treatments present unbalanced vines. Higher values can indicate excessive shading (JACKSON, 2008) which is observed for 8,000 pl ha^{-1} treatment as discussed for leaf area per meter of row and LLN. However, this ratio has shown controversy results for many authors (DIAS et al., 2017; KELLER et al., 2005; MARCON FILHO et al., 2016), suggesting that it varies considerably with cultivar (JACKSON, 2008), soil conditions and mostly by climate.

It is well known that starch is the main carbohydrate stored in perennial structures (trunk and roots) of grapevine and is used to support the initial shoot development during the spring period (BATES; DUNST; JOY, 2002; ZAPATA et al., 2004). The starch storage can be influenced by many factors such as climatic conditions, canopy management, crop load and planting density as showed by several studies (HUNTER, 1998; SMITH; HOLZAPFEL, 2009; WEYAND; SCHULTZ, 2006; ZUFFEREY et al., 2012). The planting density and shoot thinning did not affect starch content in trunk samples (figure 2). In this study, the increasing of leaf area:fruit weight ratio from 2.0 (mean values of both thinning treatments of 2,000, 2,667 and 4,000 pl ha^{-1}) to 3.2 (mean values of both thinning treatments of 5,333 and 8,000 pl ha^{-1}) was not enough to compromise the trunk starch accumulation. These results suggest that source-sink relation, expressed by leaf area:fruit weight ratio, was not unbalanced by treatments. Similar response was also verified in Pinot Noir and Chasselas vines, which trunk starch was not decreased by reducing in half the in-row distance (HUNTER, 1998; ZUFFEREY et al., 2012). Moreover, these authors also showed that starch concentration can be compromised only by extremely high planting densities (20.000 vines per hectare) and phenological stage.

Figure 1 - Leaf area per plant (A), leaf area per meter of row (B), leaf layer number (C) and leaf area yield^{-1} ratio (D) of field grown Chardonnay variety in different planting densities and shoot thinning treatments, Caldas-MG, 2017. Means followed by the same letter, uppercase for thinning and lowercase for not thinning treatment, were not significantly different (p < 0.05) by Scott-Knott test. '*' indicates a significantly difference (p > 0.05) between thinned and not thinned treatments within the planting density and 'NS' and 'ns' for not significantly different (p < 0.05) by Scott-Knott test.

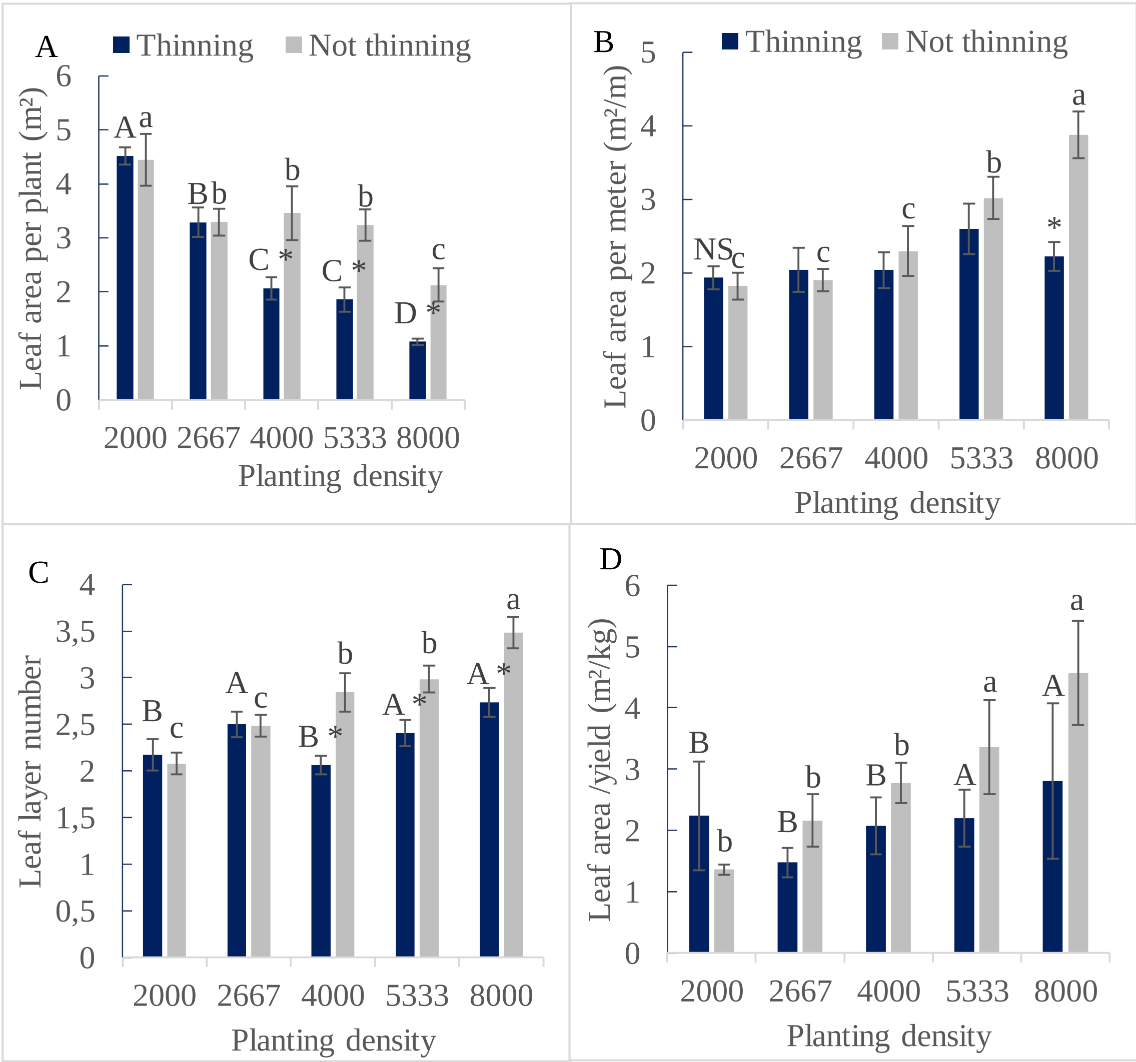
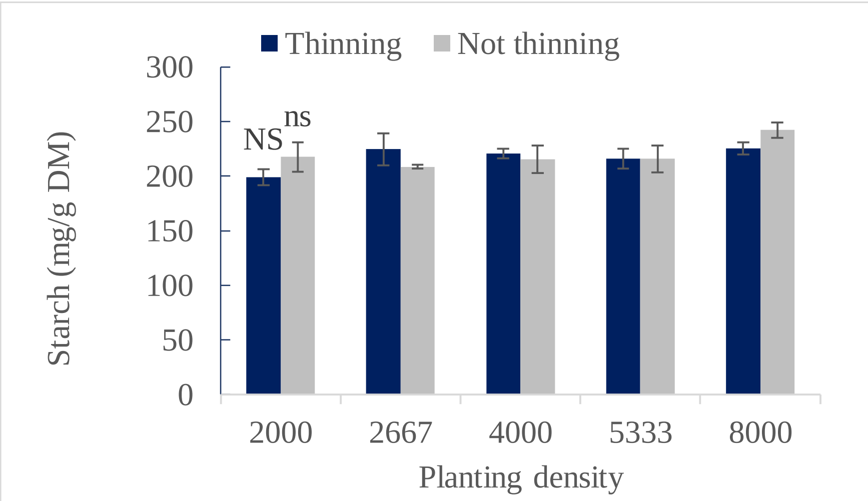


Table 2 – Leaf layer number of field grown Chardonnay in different planting densities, Caldas-MG, 2014.

Planting density (pl ha^{-1})				
2,000	**2,667**	**4,000**	**5,333**	**8,000**
Leaf Layer Number				
2.33 b	2.44 b	2.41 b	2.78 a	3.19 a

The means followed by same letter in same line were not significantly different at 5% of probability by Scott-Knott test.

Figure 2 - Starch of field grown Chardonnay variety in different planting densities and shoot thinning treatments, Caldas-MG, 2017. 'NS' and 'ns' indicates not significantly different ($p < 0.05$) by Scott-Knott test, for thinning and not thinning treatment, respectively.



3.2 Yield components

In not thinning treatments, in 2014 cycle the highest bud fruitfulness and cluster number were showed by planting densities from 2,000 to 4,000 pl ha^{-1}. For 2017 cycle, there was no interaction between planting density and thinning treatments on bud fruitfulness and the highest value was showed by 2,000 and 2,667 pl ha^{-1}. The highest cluster number was showed by 2,000 pl ha^{-1} and the lowest by 8,000 pl ha^{-1}. The bud fruitfulness is related to cluster number and is directly influenced by environmental and agronomical conditions of the previous cycle. The highest densities presented shaded canopies due to the overlapping of foliage and the lowest density presented higher light input due to gaps in the canopy (figure 1c and table 2). Similar results was also observed in the previous cycle of 2016 (data not shown). The shading and gaps in the canopy probably affected the latent bud exposure to light and consequently the inflorescence induction and differentiation, reducing the bud fruitfulness for 2017 cycle in higher densities and increasing it in lower densities (figure 3B), as already mentioned by Sánchez and Dokoozlian (2005). Buttrose (1974) reported that besides light incidence, temperature is also a dominant factor for inflorescence primordia formation. This suggest that not only light may have affected bud fruitfulness and consequently cluster number, but also temperature, since it may change due to increased shading or increased light incidence. However, Bernizzoni et al. (2009) observed no effect on bud fruitfulness when reducing in-row distance in Barbera cultivar. The difference in leaf area per row meter from the wider treatment (1.5 m) to narrower treatment (0.9 m) was considerably lower in this study (1 m²) compared with the difference in the leaf area per row meter from the wider (2.0 m) to narrower (0.5 m) in-row distance reported here for Chardonnay (2 m²), suggesting that Chardonnay vines were more shaded than Barbera vines.

The yield in not thinning treatment, in general, was reduced by increasing planting density in 2014 and 2017 (E and F figure 3), as also observed in Barbera and Riesling cultivars (BERNIZZONI et al., 2009; REYNOLDS et al., 2004). The highest yield was showed by densities from 2,000 to 4,000 pl ha^{-1} in 2014 cycle (figure 3E). Yet in 2017, the highest yield was showed by the lowest planting density and the lowest yield by the highest planting density (figure 3F). These results are probably due to cluster number influence (C and D figure 3), and reflect the contrast in vine size induced by wider in-row distance of 2 m in 2,000 pl ha^{-1} and narrower in-row spacing of 0,5 m in 8,000 pl ha^{-1}.

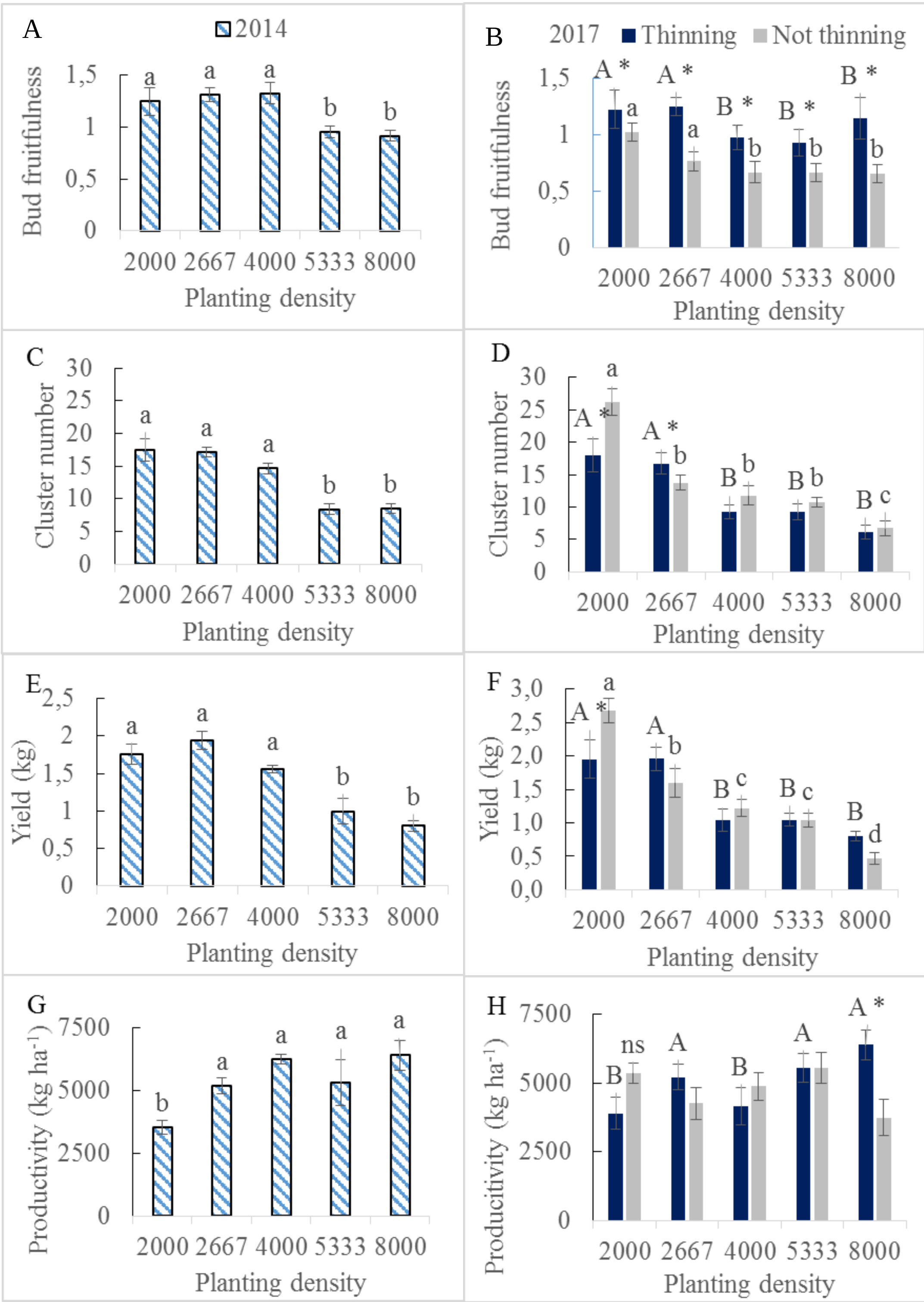
Under thinning treatment, planting densities showed same results among planting densities for bud fruitfulness (figure 1B), cluster number (figure 1D) and yield (figure 1F), with densities of 2,000 and 2,667 pl ha^{-1} showing higher values.

The increase in bud fruitfulness in densities from 4,000 to 8,000 pl ha^{-1} was probably due to shoot thinning treatment that reduced LLN (figure 1C), and the increase in 2,000 and 2,667 pl ha^{-1} bud fruitfulness is due to the thinning treatment combined to the gaps provided by wider distance between shoots, suggesting that it may have increased the incidence of light and maybe temperature on latent buds of the previous cycles. The management of shoot number per area has shown to improve bud fruitfulness as many others factors in grape (SMART, 1988); however the influence of smaller in-row distance between vines was not overcame by shoot thinning for densities from 2,000 to 5,333 pl ha^{-1}, as observed in the lower values of the higher densities in all yield components. The exception was for the 8,000 pl ha^{-1} planting density, which the increase in bud fruitfulness was crucial for the increase in productivity under shoot thinning management. As observed by Reynolds et al. (1994a) for Riesling variety, where high shoot density led to low bud fruitfulness and number of cluster due to shaded canopies. The reduced cluster number and yield in the 2,000 pl ha^{-1} thinned treatment may be due to removal of fertile shoots, and the increased in cluster number in the 2,667 pl ha^{-1} thinned treatment probably due to increase in bud fruitfulness.

Other yield components (berry weight and cluster weight) showed little or no difference between planting densities in both years, and in both thinning treatments in 2017. The 2017 average, for all treatments, was 1,86 g per berry and 112 g per cluster; and in the 2014 cycle treatments presented an average of 1,67 g per berry and 102 g per cluster. So is possible to assume that cluster number had more effect on yield than the others components did, as also observed by Clingeleffer et al. (2001) and Dias et al. (2017).

In 2014, the lowest productivity was showed by 2,000 pl ha^{-1} (figure 3G). In 2017, productivity was only affected by planting density under shoot thinning treatment, and the highest productivities were presented by 2,667, 5,333 and 8,000 pl ha^{-1} (figure 3H). The 2017 results for thinning treatments are probably explained by the increase in number of plants per hectare and in bud fruitfulness. The increase in bud fruitfulness was probably the main factor that increased the productivity 8,000 pl ha^{-1} under thinning treatment, since it presented a significant difference between thinning and not thinning treatments. In not thinning planting densities, the increase from 2,667 vines to 8,000 vines per hectare was not enough to overcome lower bud fruitfulness induced by shading in 2014 and 2017. Bernizzoni et al. (2009), Hedberg and Raison (1982) and Reynolds et al. (2004) also observed high productivity induced by high number of vine per area.

Figure 3 - Bud fruitfulness (A and B), cluster number (C and D), yield (E and F) and productivity (G and H) of field grown Chardonnay variety in different planting densities and shoot thinning treatments, in 2014 and 2017 cycles, Caldas-MG. Means followed by the same letter, uppercase for thinning and lowercase for not thinning treatment, were not significantly different ($p < 0.05$) by Scott-Knott test. '*' indicates a significantly difference ($p > 0.05$) between thinning and not thinning treatments within the planting density and 'NS' and 'ns' for not significantly different ($p < 0.05$) by Scott-Knott test.



3.3 Grape, must and base wine composition

The means of grape pH and soluble solids in both years presented little differences among treatments. In 2014, all treatments presented an average of 3.31 pH and 18.53 ºbrix and in 2017, not thinning planting densities presented an average of 3.19 pH and 16.45 ºbrix, while thinning planting densities presented an average of 3.19 pH and 16.40 ºbrix. Despite little differences, all treatments were within ranges for sparkling wines production (COPPOLANI, 1994; FÁVERO et al., 2006).

Tartaric and malic acid are the main acids on grape and both are synthetize on leaves and berries, and most of their content is produced before *véraison*. After *véraison*, however, tartaric acid concentration remains relatively constant while malic acid decreases, reaching different concentrations on harvest time, an average of 5.3 g L^{-1} of tartaric acid and 4.21 g L^{-1} of malic acid (JACKSON, 2008; REGINA et al., 2010). The malic acid concentration decreases mainly by light exposure and higher temperatures by cellular respiration (JACKSON, 2008). In general, malic and tartaric acid content increased as the planting density increased (figure 4). The highest content of malic acid showed in not thinning 5,333 and 8,000 pl ha^{-1}, and in thinning 8,000 pl ha^{-1} treatments, was probably due to shaded canopy, and the lowest content showed in not thinning 2,000 and 2,667 pl ha^{-1}, probably for the opposite effect. The shoot thinning treatment attenuated the differences in malic acid content and reduced it in 4,000 pl ha^{-1} density, probably by allowing greater light input in the canopy. These results relate to vigor parameters discussed (figure 1B and 1C). Many others had also observed lower malic acid content related to higher temperatures and light exposure (KLIEWER, 1971; KLIEWER; LAKSO, 1975; KLIEWER; SMART, 1989; TARDAGUILA et al., 2010; TODA; BALDA, 2014).

The highest content of tartaric acid in all thinned vines relates to results found by Poni et al. (2006) and Tardaguila et al. (2010) in which increased content of tartaric acid was found in treatments defoliated. Kliewer and Schultz (1964) observed that in sun exposed berries higher amounts of $^{14}CO_2$ were incorporated into tartaric acid rather than for shaded berries. However, differences in genotype and variation in canopy porosity, hence affecting light exposure and temperature in the fruit zone may also interfere in results (BULOLA et al., 2012; TARDAGUILA et al., 2010).

In not thinning treatments for both years, 2017 and 2014, the grape must, in general, presented an increase in acidity values as the planting density also increased. The lowest densities, of 2,000 and 2,667 pl ha^{-1}, showed a grape acidity average of 7.78 g L^{-1} of tartaric acid in 2014 and 9.25 g L^{-1} of tartaric acid in 2017, while higher densities, from 4,000 to 8,000 pl ha^{-1}, showed higher acidities, of 8.5 g L^{-1} and 9.99 g L^{-1} of tartaric acid in 2014 and 2017, respectively. This can be explained by tartaric and malic acid content in grape (a and b figure 4), that are the main acids responsible for grape acidity, and by the shaded canopy the higher densities presented (figure 1c and table 2), indicating that the malic acid was less degraded in those conditions (figure 4a).

When thinning treatment was applied, the difference in grape acidity among planting densities attenuated, as also observed for malic acid content. The average acidity for densities from 2,000 to 5,333 pl ha^{-1} was 9.03 g L^{-1} and for the highest density was 9.85 g L^{-1}. This suggests that greater incidence of light induced by the shoot thinning treatment had a positive effect on grape acidity, since it degraded more malic acid in those conditions. However, this degradation was not high enough to impair the acidity levels desired in grape must.

The acidity was affected mostly by LLN and its shading effect, as figure 5 indicates. Leaf layer number was positively correlated with grape, must and wine acidity, indicating that acidity was linearly increased by shading. Similar results were found relating denser canopies to high wine total acidity for Riesling cultivar (REYNOLDS et al., 1994b), high cluster exposure to low acidity in Merlot berries (SPAYD et al., 2002) and in grape, must and wine of Chardonnay Musqué (REYNOLDS et al., 2007). These results indicate that shading preserve acidity in berries, must and wine, which is positive for sparkling wine production as suggested by Jones et al. (2014). However, the maximum difference of acidity observed between treatments was below minimum level for tasting recognition of mouth-feel sensation of sourness, which is 2.67 g L^{-1} of tartaric acid (JACKSON, 2009).

The base wine, in all treatments presented an average of 0.14 g L^{-1} of acetic acid for volatile acidity, 0.99 for density at 20ºC, 20.66 g L^{-1} for crude extract, 11.65% (v/v) for alcohol content, pH 3.18, 2.22 g L^{-1} for residual sugars and 1.43 g L^{-1} for ashes. All parameters show that the base wines are suitable for sparkling wine production (FÁVERO et al., 2006; RIZZON; MIELE; SCOPEL, 2009). Moreover, all means found were similar to Chardonnay base wines produced in Serra Gaúcha and Encruzilhada do Sul (POERNER et al., 2010), and to sparkling wines from Santa Catarina (CALIARI et al., 2014). Results (figure 1d) suggest that the leaf area:yield ratio reported by Kliewer and Dokoozlian (2005) as ideal for fruit composition and wine quality may not apply for Chardonnay grown in highlands of Brazilian Southeast for sparkling wine production. Minimal differences found on grape, must and wine composition and on starch content agrees with Bernizzoni et al. (2009), Heuvel et al.

(2013), and Reynolds et al. (2004), whom have concluded that planting density has minimal effect or no effect in grape and must composition.

Figure 4 - Tartaric acid (A) and malic acid (B) content of field grown Chardonnay grapes in different planting densities and shoot thinning treatments, Caldas-MG, 2017. Means followed by the same letter, uppercase for thinning and lowercase for not thinning treatment, were not significantly different (p < 0.05) by Scott-Knott test. '*' indicates a significantly difference (p > 0.05) between thinned and not thinned treatments within the planting density by Scott-Knott test.

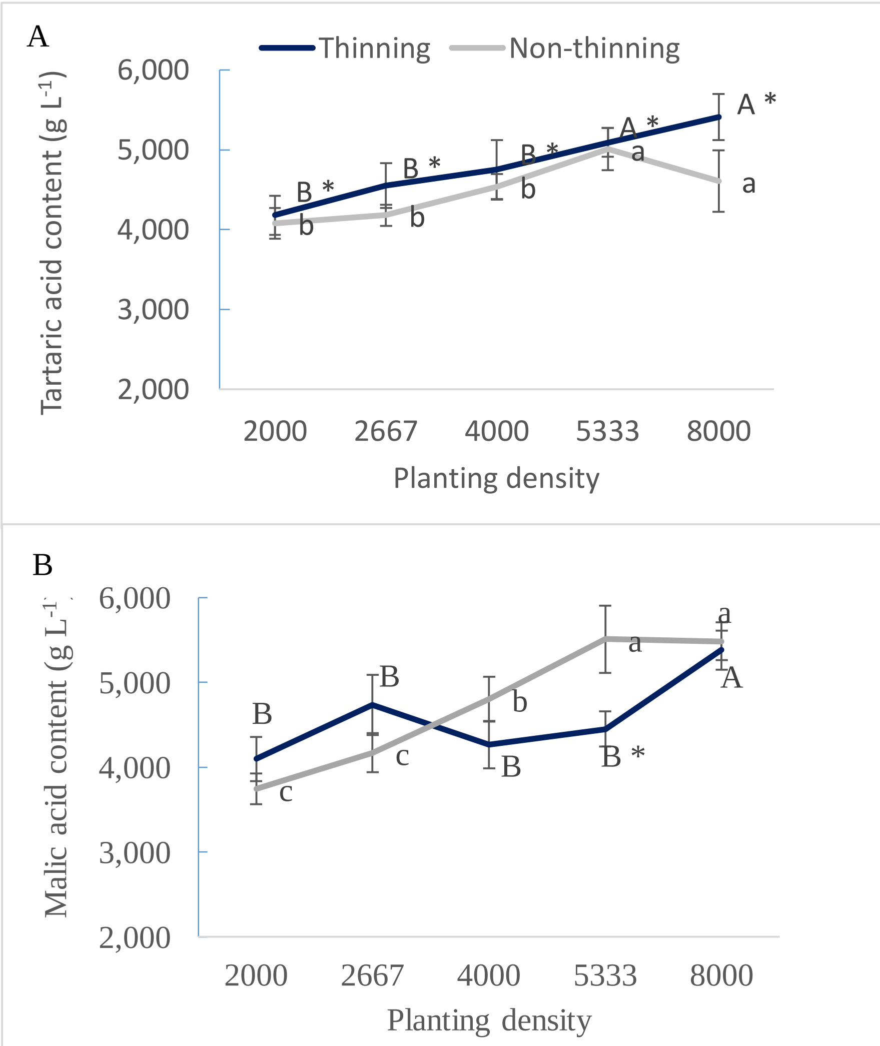
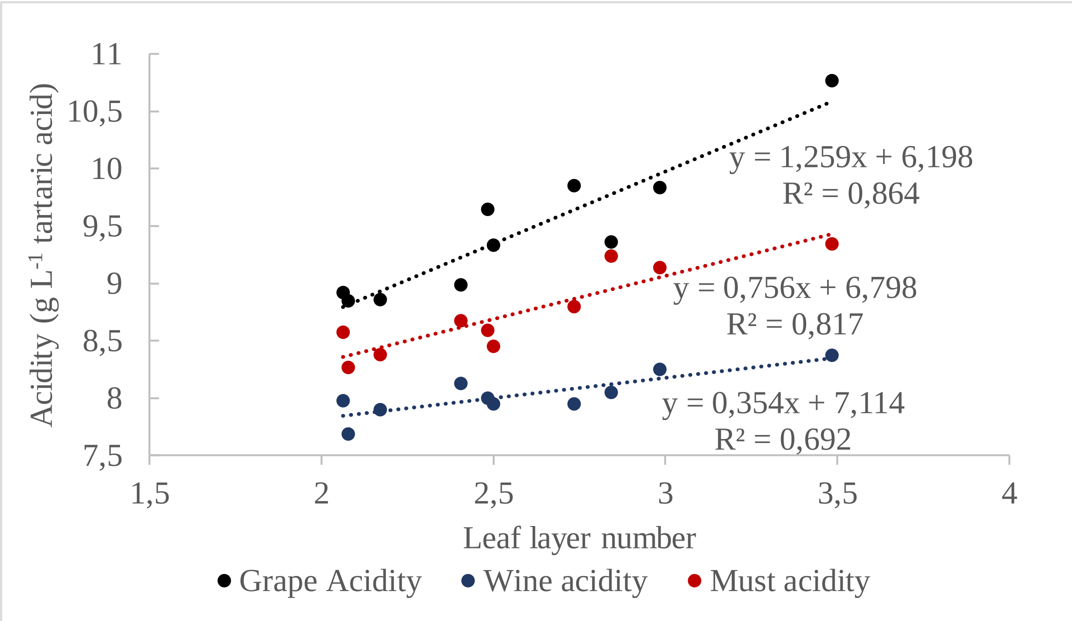


Figure 5 - Regression equation for acidity and leaf layer number of Chardonnay grapes, must and wine, in different planting densities. Using means of all ten treatments, Caldas-MG, 2017.



4 CONCLUSION

Increasing planting density for not thinning vines, leaf area per meter of row and LLN, also increase, indicating denser canopies and shaded clusters; it also diminish bud fruitfulness and consequently number of clusters and yield. The highest number of plants per hectare is not enough to overcome the low bud fruitfulness and increase productivity in planting densities from 2,667 pl ha^{-1} to 8,000 pl ha^{-1}.

The planting density and shoot thinning have little effect on grape, must and base wine composition, with exception to acidity levels. Higher planting densities preserve acidity levels in berries and must, guarantying freshness in base wines for sparkling wines. The maintaining of the acidity levels is mainly due to tartaric and malic acid content and LLN, that increase as planting density increases.

For Brazilian Southeastern highlands, we recommend planting densities from 2,667 to 5,333 vines per hectare, without shoot thinning. The planting density of 8,000 vines per hectare is only justified when applying shoot thinning; however, it presents higher costs of establishment and management.

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GENERAL REVIEW

The Brazilian southeast vitiviniculture has been growing and reaching high quality standards in their wines, produced under double-pruning method. The production of sparkling wines is an advantageous activity to diversify the products offered by the region, as it has an increasing market in Brazil and worldwide. To benefit the sparkling wine production in this region, vineyard managements must be studied, in order to raise yield without impairing the grape quality, and consequently the sparkling wine quality.

The planting density is an important management decision since it affects the initial investment and the final profit by the winegrower. The average productivity for a high-altitude Chardonnay vineyard in the region varies from 8 to 10 t ha^{-1}, when in a 4,000 pl ha^{-1} density. The average productivity reached in this work was considerably below this range, in 2014 vines were still young (4 years old), explaining this reduced productivity, yet for 2017, the hail rain that occurred in October of 2015, may have damaged the buds for 2016 and 2017 harvest and affecting the crop. The recommended planting density vary from 2,667 pl ha^{-1} to 5,333 pl ha^{-1}, depending mainly on the vineyard site characteristics. In sites with cooler temperatures, lower densities can be adopted (2,667 pl ha^{-1}), which is an advantage for the winegrower due to its low initial cost, and in sites with higher temperatures, higher densities can be adopted (5,333 pl ha^{-1}), so it conserves acidity.

However, if the vineyard had not been affected by the hail rain, the little differences found among grape, must and wine quality and tendencies found in productivity values among treatments, could be enhanced. Furthermore, with vines perfectly healthy, the productivity for vineyards with 8,000 pl ha^{-1} under thinning management, would reach values that could justify the additional activity of shoot thinning.

The success of wines from southeast region was not only established by the proper climate conditions but also by public-private partnerships that have been taking place since 2001. This partnership allowed the development of a technological package for double-pruning management. The implementation of a technological package for the production of sparkling wines should follow the same steps. The interest from private public is already a reality, where many producers aim for information on the implementation of vineyards for producing sparkling wines, since it is an increasing market worldwide. Therefore, the need to expand and improve research in potential cultivars, clones, rootstocks, conduction systems, winemaking methods and enological features, and yet wine marketing features for the sparkling wines market must be fulfilled.