1Original Research Article2DIFFERENT SOURCES AND4CONCENTRATIONS OF 6-BA IN CHEMICAL5THINNING OF POST-FLOWERING IN APPLE6TREES78910ABSTRACT

11

Aims: Evaluate the efficiency of sources and dosages of 6-benzyladenine in the chemical thinning of the apple tree 'Fuji Suprema', as well as possible histological changes in the fruits produced, in the Midwestern region of the state of Santa Catarina. **Study design:** The experiment was arranged in a randomized block design and replicated six replications. **Place and Duration of Study:** The experiment was carried out in the municipality of Caçador, Brazil (latitude 26°46 'S, longitude 51° W, altitude 960 meters), during the growing season of 2016/2017. **Methodology:** Two commercial products based on 6-BA (Benzyladenine), Exilis[®] and Maxcel[®], both with 2% BA in their composition, were tested. Were applied individually in post-flowering period (in fruits of 5-10 mm), which were compared with plants with no thinning and manual thinning. Treatments were as follows: Control (no thinning); manual thinning; Exilis[®] (1.5, 3.0, 4.5 and 6.0 L ha⁻¹) and Maxcel[®] 4.5 L ha⁻¹), using a spray volume equivalent to 1000 L ha⁻¹. The fruit set and the number of fruit per cm² branch, was reduced in treated plants, regardless of the concentration of BA or formulation used. The productivity ranged from 38 to 56.5 t ha⁻¹ in the treatments of Exilis[®], 60.5 t ha⁻¹ in manual scaling and 24.5 t ha⁻¹

in the Maxcel[®] 4.5 L ha⁻¹, characterizing excessive thinning. BA is efficient in fruit thinning in 'Fuji Suprema' apple trees, being the reduction of fruiting, and increase of fruit size, proportional to the applied concentration. There may be differentiated efficiency of the product by its formulation, even though they have concentrations of active ingredient equivalent. Exilis® was efficient in thinning of apple "Fuji Supreme", when applied to fruit 5 to 10 mm in diameter reduced the need for manual thinning, without causing toxicity. BA can promote the increase in the cell density of fruits produced in proportion to the applied concentration.

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13 Keywords: Histology. Productive capacity. Cell density.

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15 **1. INTRODUCTION**

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17 Fruit plants, such as apple trees, generally produce more flowers than are 18 necessary for a commercial crop. In years with abundant flowering and favorable 19 environmental conditions for pollination, fruit set can be excessive, so that there is 20 excess fruit on the plant, making it difficult to obtain large fruits and consequently of high 21 commercial value. If all the flowers are fertilized and develop, the plant may lack 22 photosynthetic resources or structural integrity for adequate fruit ripening [1]. In these 23 conditions, thinning is a necessary and indispensable practice to obtain fruits of better 24 quality (higher caliber and uniform color) and greater market value, as well as to avoid 25 the alternation of production [2,3,4].

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The use of chemical products aimed at fruit thinning is a technique consolidated in the apple tree culture, mainly due to the practicality of application, as it allows an adequate adjustment of the fruit load when applied correctly and at the right moment of the development (stage) of the fruits [5]. As the time of application of the chemical thinners, the formulation, the mode of action of these products, as well as the worked cultivar can determine the efficacy of chemical thinning [6,7,8]. Among the chemical thinners for the culture of the apple tree, stand out: ANA (Acetic Naphthalene Acetic), Carbaryl, Ethephon, Metamitron and BA (Benzyladenine). The results of the application of BA in the thinning can vary, according to the concentration, temperature, humidity, luminosity, volume of syrup applied and stage of fruit development in moment of application [9,10,11]. The knowledge of temperature and light in the three to four days after the application of chemical thinners has made the thinning process more predictable and reliable [12].

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42 BA is a compound of the cytokinins group, which has the ability to stimulate fruit 43 growth and vegetative activity, influences cell division, thus increasing competition for 44 assimilates, reducing the energy available for fruit development, reducing the rate of net 45 CO2 assimilation, which results in an increased fruit fall [13]. Cytokinins increase fruit 46 size in apples, even in the absence of thinning, due to the promotion of cell division in 47 apple tissue [14,10]. According to Yuan and Greene [15], BA is considered a good 48 product because it has low toxicological profile and imitates the biological action of the 49 cytokinin that is synthesized in plants.

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The final size of the fruits is associated with the density and size of the cells present in the fruit, and these factors are determined by the genetic expression, which delimits the duration of the phases of cell division and elongation during fruit growth and development [16,17,18]. The genetic factor will predetermine the final size of the fruits, but may be influenced by the nutritional state of the plants, cultural traits and environmental factors [19].

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Advances in the understanding of the physiological responses of plants and factors involved in the development of fruits, aim to improve technologies for adjusting loads to acceptable levels, with less dependence on manual thinning [20]. Information about possible anatomical and histological interferences caused in fruits by the

62 application of chemicals for load adjustment, or even manual thinning, is still limited. The 63 objective of the present study was to evaluate the efficiency of sources and dosages of 64 6-BA in the chemical thinning of 'Fuji Suprema' apple tree in the Midwest region of the 65 state of Santa Catarina, as well as possible histological changes in the fruits produced.

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67 2. MATERIAL AND METHODS

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The study was conducted in an experimental orchard in the Midwestern region of Santa Catarina, in the municipality of Caçador (latitude 26°46 'S, longitude 51° W, altitude 960 meters), in the 2016/17 season. Apple trees of "Fuji Suprema" cultivar, with Marubakaido / M-9 rootstock and planting density of 2,500 ha⁻¹ plants were used.

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The 'Fuji Suprema' apple tree is a mutation of the 'Fuji' group, with high effective fruiting and fixation of 3 to 5 fruits per inflorescence. It is managed plants in the central leader in driving system, according to the recommendations of the apple production system [21], and management practices in the integrated system of apple production [22].

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80 The experimental design was randomized blocks with seven treatments and six 81 replicates. The treatments were: Control (No thinning); MT - Manual thinning; Exilis® 82 (1.5, 3.0, 4.5 and 6.0 L ha⁻¹, values equivalent to the concentrations of BA 30, 60, 90 and 120 mg L⁻¹) and Maxcel[®] 4.5 L ha⁻¹ (BA 90 mg L⁻¹). The products used were: Exilis[®] SL 83 (20g / L 6-BA), toxicological class I; Maxcel[®] SC (20 g / L 6-BA)) toxicological class II. 84 85 The treatment of manual scaling was performed using scissors, following the criterion of 86 two fruits in terminal buds of brindila (long branches) and one fruit in spur, according to 87 CP - productive capacity of the plants, 5 to 6 fruits per cm² of trunk section area [21]. The 88 application of the treatments with chemical thinners was performed with a motorized 89 costal spray, containing a tip with three D-S spray nozzles, using a volume of syrup 90 equivalent to 1000 L ha⁻¹, up to the point of dripping.

The application of the treatments was performed on October 13, 2016, eighteen days after PF - full bloom, which occurred on September 25, 2016, when fruits were with an equatorial diameter between 5 and 10 mm. In the first days after the application of the treatments a significant increase in the maximum, minimum and average temperatures, and precipitation of 4.8mm was registered in the first day after the application (Table 1). The fruits were harvested at 166 days after PF, on March 10, 2017.

- 98
- 99 Table 1. Meteorological data observed three days before and three days after the

100 application of the different treatments for load adjustment (thinning). Caçador, SC,

- 101 Brazil, 2019.
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Date	Те	Dresinitation (mm)		
Dale	Maximum	Mmnimum	Averege	Precipitation (mm)
1) 10/10/16	23,4	6,8	15,2	19,9
2) 11/10/16	18,6	11,2	13,8	13,2
3) 12/10/16	18,6	11,0	14,9	0,0
4) 13/10/16	22,4	13,2	17,0	0,0
5) 14/10/16	26,0	11,0	18,2	4,8
6) 15/10/16	22,0	16,5	19,1	0,0
7) 16/10/16	31,2	16,4	23,4	0,0

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104 The variables evaluated were: The fruit set (%), determined in two branches per 105 plant, was evaluated in the number of flower bunches during the PF and the number of 106 fruits, 30 days after. In these same branches were counted the number of floral clusters 107 with fruits, resulting in percentage of floral clusters with fruits, and number of fruits per inflorescence. At harvest (March 13, 2017): production (kg plant⁻¹ and fruits plant⁻¹); 108 109 productive efficiency (kg cm⁻² and fruits cm⁻²); average fresh fruit mass (g); classification 110 of fruits by size classes (%); number of seeds per fruit; pulp firmness (lbpol⁻²) and soluble 111 solids (° Brix) according to Scolaro et al. [23].

112

For the anatomical analyses of fruits, two fruits per plant were collected during the cycle, only of terminal twig buds. After collection, samples were fixed in FAA solution (Formalin / acetic acid / ethyl alcohol 1: 1: 8); fractionated and processed; included in

116 historesin; submitted to microtomy, confection and staining of microscopy slides; capture

117 and analysis of images in specific software (ToupView[®]).

118

119 Statistical analysis of data was performed through analysis of variance, and 120 variables whose results revealed significance (p <0.05) were submitted to comparison of 121 means by the Scott-Knott test and, or regression analysis at 5% probability. Statistical 122 analyses were performed by the Sisvar software, version 5.6 [24].

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124 3.1 RESULTS AND DISCUSSION

After the application of the treatments with chemical thinners, a significant reduction of these parameters in the treated plants was observed, independently of the concentration or formulation of BA used (Table 2). The mean number of fruits per inflorescence did not present significant differences.

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130Table 2 – Fruit set (%), mean number of fruits per inflorescence and131number of fruits per cm2 of branch cross section, Fuji Suprema cultivar,132submitted to different treatments for load adjustment (thinning), in the 2016/2017133season, Caçador-SC, Brazil, 2019.

Traction		n° fruits	Fruits per
Treataments	Fruit set (%)	cm ⁻² branch	inflorescence
Control (no thinning)	109.0 a	6.9 a	2.5 ^{ns}
Manual thinning	119.9 a	6.2 a	2.2
$Exilis^{\ensuremath{\mathbb{R}}}$ 1.5 L ha ⁻¹	59.5 b	2.9 b	1.8
Exilis [®] 3.0 L ha ⁻¹	58.3 b	3.3 b	1.8
Exilis [®] 4.5 L ha ⁻¹	76.0 b	3.7 b	2.0
Exilis [®] 6.0 L ha ⁻¹	66.6 b	2.6 b	1.7
Maxcel [®] 4.5 L ha ⁻¹	25.9 b	1.4 b	1.2

Average	73,6	3,9	1,9
VC (%)	33,7	39,3	32,4

135 Means followed by the same letter in the column do not differ from one another by the 136 Scott-Knott test (P=.05). * ns: not significant (P>.05).

137

138 This situation can be alarming, because in some cases, when considering the 139 CP-productive capacity of apple trees, which according to Sezerino [21] would be around 140 five fruits per cm⁻², the reduction in fruiting caused by the raleantes in the crop studied, 141 may have been very severe, leading to a large reduction in orchard productivity. 142 Considering a stand of 2,500 plants ha⁻¹, the treatments of Exilis[®] presented a 143 productivity ranging from 38 to 56.5 t ha-1, with manual thinning of 60.5 t ha-1 and Maxcel[®] treatment 4.5 L ha⁻¹ of 24.5 t ha⁻¹, which is much below the production capacity, 144 145 showing excessive thinning. In addition to the applied concentration, the time and the 146 residual period of the product can help in the understanding and justify the results 147 observed here. According to Ambrožič Turk and Stopar [25], BA can be active as a 148 raleante in a broader period of phenological phases, from the end of flowering to fruits 149 with 20 mm in diameter, not only in fruit between 10 mm, indicated by many reports, and 150 the persistence of the raleante effect of BA can extend for up to 18 days after 151 application. For an efficient chemical thinning with BA, the dose of the product must be 152 taken into account, together with the climatic conditions and the cultivar [8].

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154 Plant production (kg plant⁻¹) was significantly reduced by the application of Exilis[®] 3.0 L ha⁻¹ and Maxcel[®] 4.5 L ha⁻¹ treatments (Table 3). But when we analyzed the 155 number of fruits per plant and the productive efficiency (kg cm⁻²) all treatments with 156 157 chemical thinners differed significantly from the control treatment and manual thinning, 158 however, Maxcel[®] 4.5 L ha⁻¹ was the treatment that caused the highest production 159 efficiency. The mean mass of the fruits presented differences between the treatments, 160 characterized by the significant increase in the mass of the fruits from plants submitted 161 to the treatments of chemical thinning. Regardless of the concentration of the commercial Exilis[®] product applied, the average fruit masses were higher than the 162

treatments of manual thinning and Control, but lower than the treatment of Maxcel[®] 4.5 L ha⁻¹, which can be attributed to a reduction productivity caused by this treatment. It should be noted that the average mass of the fruits harvested in the plants of the treatment Maxcel[®] 4.5 L ha⁻¹ increased in 54,8% in relation to the Control.

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168 Similar behavior was observed in fruit classification, where the largest size class 169 (65mm), which presented the highest commercial value, all treatments of Exilis[®] differed 170 significantly from the control and manual thinning treatments, increasing the percentage of fruits of larger caliber and reducing however, the Maxcel[®] 4.5 L ha⁻¹ was superior to all 171 172 treatments tested. Szot et al., [26] obtained excellent results with the application of BA 173 300mg L⁻¹ (Maxcel[®] 7.5 L ha⁻¹) and BA 300mg L⁻¹ + Extract of algae, characterized by a 174 very uniform crop formed by only two fruit size classes, 75-85 mm and above 85 mm in 175 diameter, such results would be excellent at the producer level, as it would eliminate the 176 need for manual thinning and increase profitability. Petri et al. [9] working with concentrations of 80 and 120 mg L⁻¹ of BA obtained large fruit proportions (<135) greater 177 than 75% and when associated BA 80 mg L-1 + Carbaryl 1000 mg L-1 the results were 178 179 superior to 80% of the fruits in categories of greater commercial value.

180

Marchioretto et al. [27], reported the occurrence of changes in the physicalchemical characteristics of 'Fuji Suprema' apple fruits treated with BA at 60 mg L-1 compared to untreated plants, characterized by an increase in soluble solids concentration and firmness of pulp, as was observed in the present study, in which Exilis® 3.0 L ha-1, Exilis® 4.5 L ha-1 and Maxcel® 4.5 L ha-1 were superior to the other treatments of thinning and plants without control, .

187Table 3 - Production (kg plant⁻¹ and fruits plant⁻¹), MFM (average fresh188mass - g fruits⁻¹), productive efficiency and firmness pulp of fruit 'Fuji Suprema'189apple tree, submitted to different treatments for load adjustment (thinning), in the1902016/2017 season, Caçador-SC, Brazil, 2019.

Treataments Produção / Planta MFM Productive Efficiency

	kg	frutos	(g)	kg cm ²	frutos cm ²
Control (no thinning)	25.2 a	191.5 a	131.4 c	1.3 a	10.0 a
Manual thinning	24.2 a	177.7 a	137.2 c	1.4 a	10.5 a
Exilis [®] 1.5 L ha ⁻¹	22.6 a	142.8 b	158.4 b	1.0 b	6.1 b
Exilis [®] 3.0 L ha ⁻¹	15.2 b	92.8 c	163.5 b	1.0 b	6.1 b
Exilis [®] 4.5 L ha ⁻¹	20.8 a	124.0 c	170.2 b	0.9 b	5.5 b
Exilis [®] 6.0 L ha ⁻¹	20.5 a	118.3 c	173.6 b	1.0 b	6.1 b
Maxcel [®] 4.5 L ha ⁻¹	9.8 b	47.5 d	203.4 a	0.4 c	1.8 c
Average	19,8	127,8	162.5	1,0	6,6
VC (%)	24,0	24,0	10,2	25,4	27,8

		Caliber			Firmness of
Treataments	>65 mm	55-65 mm	Brix°	Pulp	
					(Lib)
Control (no thinning)	29.7 d	48.8 a	21.4 a	11.3 b	16.4 b
Manual thinning	44.5 c	39.2 b	16.2 a	11.8 b	16.5 b
Exilis [®] 1.5 L ha ⁻¹	60.2 b	28.0 c	11.8 b	11.3 b	16.3 b
Exilis [®] 3.0 L ha ⁻¹	59.0 b	31.3 b	9.6 b	12.1 a	18.0 a
Exilis [®] 4.5 L ha ⁻¹	67.4 b	23.3 c	9.3 b	12.3 a	16.0 b
Exilis [®] 6.0 L ha ⁻¹	67.7 b	25.3 c	7.0 b	12.0 b	16.6 b
Maxcel [®] 4.5 L ha ⁻¹	83.4 a	10.2 d	6.4 b	13.1 a	18.4 a
Average	58,8 b	29,5	11,7	12.0	16.9
VC (%)	11,3	17,0	28,3	7,8	8,8

191Means followed by the same letter in the column do not differ from one another by the192Scott-Knott test (P=.05).

193

When analyzing the characteristics of the fruits harvested, the plants treated with Maxcel[®] 4,5 L ha⁻¹ presented the highest values for the variables average fresh mass, height and diameter of the fruits, surpassing all other treatments (Table 4). These results can be related to the great reduction of the effective fructification caused by this

198 treatment, as well as the type of structure that gave origin to these fruits, which in this 199 case were only fruits from brindilas, structures with greater productive capacity and that 200 produce fruits of better quality [28]. Although differences were found in the number of 201 seeds present in the fruits, this did not cause a change in the shape of the fruits 202 harvested. Although there was no difference in cell size (µm²), fruits from plants 203 submitted to manual thinning, Exilis[®] 3.0 L ha⁻¹ and Maxcel[®] 4,5 L ha⁻¹ had cell density of 204 cells per fruit section area) increased by 18.5%, 21.1% and 30.1%, respectively, 205 compared to fruits of plants with no thinning (Control). Several studies corroborate the 206 results observed in the present study, such as Marchioretto et al. [27], who observed an 207 increase in the mean fresh mass, in the height and diameter of the fruits of 'Fuji 208 Suprema' apple trees treated with BA at 60 mg L⁻¹. Szot et al., [26] report the fruit yield of 209 'Jonagold Red Prince' apples with a mean mass of 68.3% higher than the no-rale plants, with the application of BA 300mg L⁻¹ (Maxcel[®] 7.5L ha⁻¹), with fruits with a mean mass of 210 211 304.6 g and 181.0 g, respectively.

212

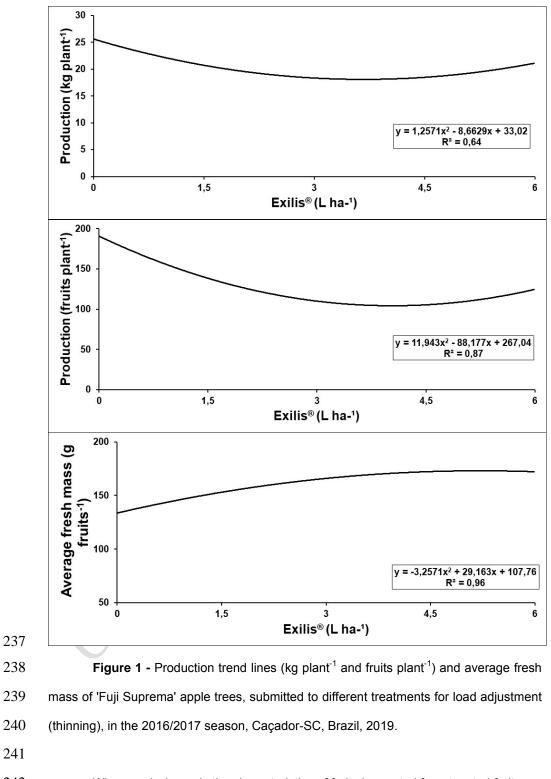
Table 4 - MFM (mean fresh mass - g fruits⁻¹), diameter and height (mm) and height / diameter ratio (A / D), mean number of seeds (NMS), cell area of cells per fruit section area (NCASF) of the fruits of brindilas, harvested from 'Fuji Suprema' apple trees, submitted to different treatments for load adjustment (thinning), in the 2016/2017 season, Caçador-SC, Brazil, 2019.

Treataments	MFM	DØ	Hei.	A/D	NMS	Area cell	NCASF	
Treataments	(g)	m	m		NNIO	(µm²)	NOADI	
Control (no thinning)	189.6 b	75.9 b	64.5 b	0.85 ^{ns}	7.5 a	132844.17 ^{ns}	34607.85 b	
Manual thinning	179.9 b	78.4 b	64.6 b	0.82	5.8 b	118157.49	41010.51 a	
Exilis [®] 1.5 L ha⁻¹	205.4 b	80.1 b	65.8 b	0.82	6.4 b	131140.91	38815.61 b	
Exilis [®] 3.0 L ha ⁻¹	223.3 b	80.8 b	67.2 b	0.83	7.8 a	123031.67	41928.62 a	
Exilis [®] 4.5 L ha ⁻¹	217.0 b	78.6 b	68.6 b	0.87	5.9 b	128621.05	37735.46 b	
Exilis [®] 6.0 L ha ⁻¹	201.9 b	76.7 b	66.2 b	0.86	6.3 b	128990.22	35836.01 b	

Maxcel [®] 4.5 L ha ⁻¹	281.9 a	86.6 a	73.7 a	0.85	6.9 a	131740.23	45032.66 a
Average	214.1	79.6	67.2	0.85	6.7	127789.39	39280.96
VC (%)	16,6	5,7	6,6	6,1	15,2	8,8	11,5

Means followed by the same letter in the column do not differ from one another by the
Scott-Knott test (P=.05). * ns: not significant (P>.05).

222 When analyzing the concentrations of BA applied, more specifically the Exilis® 223 formulation (1.5, 3.0, 4.5 and 6.0 L ha⁻¹, values equivalent to the concentrations of BA 224 30, 60, 90 and 120 mg L⁻¹), a quadratic response (kg plant⁻¹ and fruits plant⁻¹), with maximum reduction of production (kg plant⁻¹ and fruits plant⁻¹) in doses of 3.44 L ha⁻¹ 225 (68.9 mgL⁻¹) and 3.69 L ha⁻¹ (73.8 mgL⁻¹), respectively (Figure 1). However, for the 226 227 average fresh fruit mass, the opposite behavior was observed, with a maximum increase in the mean fruit mass in the dose of Exilis 4,53 L ha⁻¹ (90.6 mgL⁻¹). Similar results are 228 229 presented by other authors, such as Barreto et al. [8], which, working with increasing 230 doses of BA for load adjustment in peach trees, reported behavior similar to those 231 observed in the present study, characterized by an increase in the percentage of 232 abscision and mass of fruits, in response to the increase of applied dosage, excess of fruits with doses of 400 mg L⁻¹. For 'Fuji Suprema' apple trees it has already been proven 233 234 in other studies that the highest concentrations of BA are more effective in the thinning, 235 however, there is some reduction in its effectiveness when applied to fruits with a 236 diameter greater than 10 mm in comparison to the application in fruits [9].



242 When analyzing only the characteristics of fruits harvested from toasted fruits, no 243 changes were observed in fruit shape (height and diameter), but the average fresh fruit mass has a quadratic behavior proportional to the dose increase of Exilis[®] applied, reaching the maximum point yield at the dosage of 3.2 L ha⁻¹, which corresponds to 65.9 mg L⁻¹ BA. Cellular dimensions were not altered by increasing doses of Exilis[®] applied, however, cell density had a similar behavior to fruit mass, with a greater increase in the number of cells per section of the fruits with the dose of 3.04 L ha⁻¹ of Exilis[®], corresponding to 60.8 mg L⁻¹ BA (Figure 2).



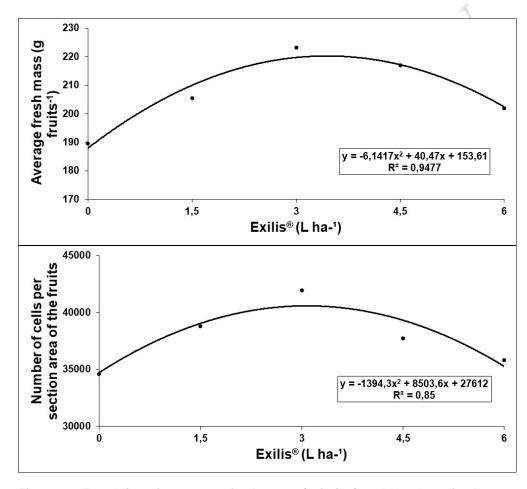


Figure 2 - Trend lines for average fresh mass (g fruit-1) and Number of cells per section area of the fruits of brindilas, harvested from 'Fuji Suprema' apple trees, submitted to different treatments for load adjustment (thinning), in the 2016/2017 season, Caçador-SC, Brazil, 2019.

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257 **4.1 CONCLUSION**

258	
259	The BA is efficient in adjusting fruit load in 'Fuji Suprema' apple trees, reducing
260	fruiting, as well as improving fruit quality and caliber proportional to the applied
261	concentration.
262	There is a differentiated response in the efficiency of the chemical thinners by
263	their formulation, even though both have concentrations of active ingredient equivalent.
264	Exilis ${\ensuremath{\mathbb R}}$ was efficient in the thinning of the 'Fuji Suprema' apple tree, when
265	applied in fruits with 5 to 10 mm of diameter increased the percentage of fruits of greater
266	caliber that has greater commercial value.
267	BA can promote the increase in the cell density of fruits produced in proportion to
268	the applied concentration.
269	
270	COMPETING INTERESTS
271 272 273	Authors have declared that no competing interests exist.
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