

# Hydrogen Cyanamide Application Accelerates Vegetative Bud Break and Causes Earlier Yield in ‘Optimus’ and ‘Colossus’ Southern Highbush Blueberry

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**Abstract.** Southern highbush blueberry (*Vaccinium corymbosum* interspecific hybrid) cultivation is a major industry in subtropical regions where low winter temperatures are infrequent and inconsistent. In Florida and other subtropical areas, growers use hydrogen cyanamide (HC) applications during endodormancy to mitigate the negative effects of low chill accumulation. Hydrogen cyanamide is a synthetic plant growth regulator that increases and expedites dormancy release and budbreak. However, southern highbush blueberry cultivars differ in their sensitivity to HC. Optimus and Colossus are two recently released cultivars from the University of Florida blueberry breeding program. The effects of HC in these cultivars are unknown. This research aimed to describe responses to HC applications at different rates for these new varieties. Experiments took place in a commercial farm in Waldo, FL, on 3- to 4-year-old deciduous blueberry bushes. HC was applied at rates of 3.8 g·L<sup>-1</sup> (0.38%), 5.1 g·L<sup>-1</sup> (0.50%), and 6.4 g·L<sup>-1</sup> (0.63%) in ‘Optimus’ and 3.8 g·L<sup>-1</sup> (0.38%), 5.1 g·L<sup>-1</sup> (0.50%), 6.4 g·L<sup>-1</sup> (0.63%), and 7.7 g·L<sup>-1</sup> (0.75%) in ‘Colossus’. In both cultivars, the control treatment was not sprayed. Vegetative bud count, and flower bud development, flower bud mortality, and yield were determined. HC application thinned reproductive buds and increased vegetative budbreak. Although seasonal yield was not increased, HC advanced fruit ripening early in the season.

Southern highbush blueberry (SHB, *Vaccinium corymbosum* interspecific hybrid) is a popular fruit crop in the southeastern United States. In temperate climates, SHB plants typically drop their leaves and enter a distinct period of rest (endodormancy) that protects them from harsh environmental conditions during winter. SHB emerges from dormancy after exposure to a critical amount of cold ( $\leq 7^{\circ}\text{C}$ ) temperature (chill accumulation). However, in subtropical climates, SHB often exhibits insufficient and inconsistent chill accumulation. Low chill accumulation causes delayed budbreak and bloom and may result in delayed or reduced yields (Dozier et al. 1990).

SHB growers rely on dormancy-breaking agents, such as hydrogen cyanamide (HC), to overcome insufficient chilling and induce dormancy release (Wang et al. 2021; Williamson et al. 2001, 2002). HC is a synthetic plant growth regulator and restricted-use pesticide with potential phytotoxic effects on blueberry. When applied after some natural chilling accumulation (~300 h) and before flower buds progress past stage 2 (Williamson et al. 2001), HC can stimulate reproductive and vegetative budbreak in HC-responsive cultivars (Wang et al. 2021; Williamson et al. 2002), but also has negative effects (primarily flower bud injury) on crop yield when not properly applied. A typical HC rate for tolerant SHB cultivars is 0.75% HC (Harmon et al. 2022), but higher and lower application rates are not uncommon. SHB cultivars vary in their sensitivity to injury and responsiveness to HC applications. Therefore, new cultivars’ responses to HC need to be evaluated.

The objective of this study was to identify HC application rates that benefit two new SHB cultivars: Optimus and Colossus. On the basis of previous studies with SHB (Williamson et al. 2002), we hypothesized that higher rates of HC result in 1) higher rates of reproductive bud mortality, 2) higher rates of vegetative budbreak, and 3) earlier harvest. We tested these hypotheses in a multiyear experiment at a commercial farm.

**Experimental site and plant material.** The experiments were conducted on a commercial farm near Waldo, FL, during the 2018–19 and 2019–20 growing seasons. Plant material consisted of 3- to 4-year-old ‘Optimus’ and ‘Colossus’ SHB. The bushes were planted in raised beds of sandy soil amended with pine bark. There were 3188 bushes per hectare with 0.9 m between plants and 3.7 m between rows. Bushes were irrigated with drip irrigation and protected from frost with overhead irrigation. Pest, disease, and fertilizer management were done in accordance with current recommendations for Florida (Harmon et al. 2022; Phillips and Williamson 2020).

Five HC rates (0%, 0.38%, 0.50%, 0.63%, and 0.75% v/v a.i.) were tested for ‘Colossus’ and four rates (0%, 0.38%, 0.50%, and 0.63% v/v a.i.) were tested for ‘Optimus’. HC was applied using the commercial formulation Dormex™ (AlzChem, Trostberg, Germany). A nonionic surfactant (Ad Spray 80; Helena Agri-Enterprises, Collierville, TN, USA) was used at 0.25% v/v. Treatments were applied using a Jacto Arbus 1000 airblast sprayer at 935.4 L·ha<sup>-1</sup>. Plants were sprayed on 2 Jan 2019 and 31 Dec 2019. For both years, ~168 chill hours (hours  $\leq 7.2^{\circ}\text{C}$ ) had been accumulated at the time of application. Spray applications were made before a significant proportion of flower buds had developed past stage 2, which is a standard industry practice (Harmon et al. 2022) to minimize spray injury to flower buds. Average temperatures in the 7 d before spraying were 19.9°C in 2019 and 13.9°C in 2020. Average temperatures in the 7 d after spraying were 15.2°C in 2019 and 14.0°C in 2020.

**Experimental design and data collection.**

Given the constraints of the commercial planting, each cultivar was analyzed using a separate randomized complete block design with six replications. Treatments were applied to groups of 15 contiguous bushes. Each group was a replication. To avoid contamination, two representative plants from the center of each group were selected for data collection. Adjacent rows were not used to avoid contamination. Data collected (from four canes per plant) included vegetative bud counts, reproductive bud development, reproductive bud damage, and reproductive bud mortality. Reproductive buds were scored using a qualitative scale from 1 to 7 based on Spiers (1978), where 1 represents a compact dormant bud and 7 represents the stage after corolla drop for a fertilized flower. Actively growing vegetative buds were counted weekly in ‘Optimus’ and ‘Colossus’ from 18 Jan to 27 Feb in 2019 and from 12 Jan to 12 Mar in 2020. Vegetative buds were also scored using a qualitative scale from 1 to 6 based on Nesmith et al. (1998), where stage 1 is a dormant vegetative bud with no sign of emergence and stage 6 is a mature elongating vegetative bud with the first three to four leaves completely unfolded. Bud mortality was determined visually based on oxidation, lack of development, and/or abscission. Yield was determined in 2019 by harvesting two representative plants at the center of each group.

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Plants from both cultivars were harvested twice per week for the duration of the harvest period and weekly yields were computed. Yield was not determined in 2020 due to limitations imposed by the COVID-19 pandemic.

**Data analysis.** Reproductive bud development and reproductive bud mortality rate data were arcsine square root transformed due to nonnormality. Transformed data were analyzed with analysis of variance, linear regression, and quadratic regression. An effect was considered significant at  $P \leq 0.05$ . Where the mean and variance of control treatments were equal to zero, data were not used in statistical analysis for specific dates. All analyses were conducted in R (Version R version 4.2.0; R

Core Team, Vienna, Austria). Where appropriate, means were separated using Tukey honestly significant difference.

## Results

**Reproductive bud development.** There were 46 and 53 d between bloom and fruit maturation in ‘Colossus’ and ‘Optimus’ in 2019, respectively. HC application did not affect reproductive bud development in ‘Optimus’ in 2019. However, HC application occasionally delayed reproductive bud development in 2020 (Fig. 1). There was a positive linear relationship between HC application rate and reproductive bud development

stage on the final data collection in 2019 for ‘Colossus’. For most of the 2020 bud development period, ‘Colossus’ exhibited strong positive linear and quadratic relationships between HC rate and reproductive bud development stage (Fig. 2).

**Vegetative bud development.** HC-treated plants exhibited earlier and more abundant vegetative bud growth and developed canopies earlier than controls in both cultivars. In ‘Optimus’, there was nearly no vegetative bud-break in the nonsprayed control until 8 Feb 2020 (Table 1). Data from the control treatment were left out of the analysis until this date (see Data Analysis section). However, after that date, ‘Optimus’ exhibited a strong positive

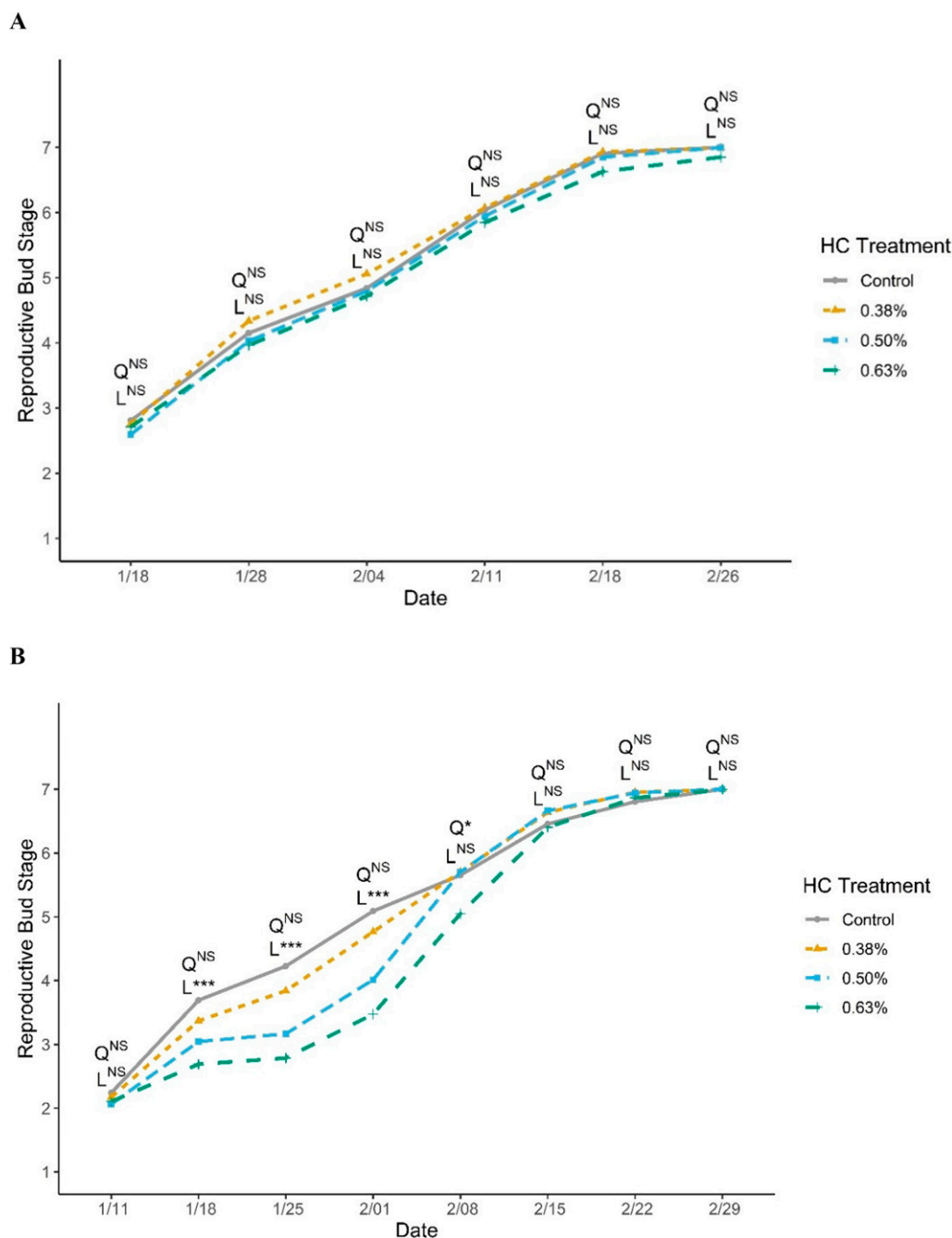


Fig. 1. ‘Optimus’ reproductive bud development as affected by different hydrogen cyanamide (HC) rates in (A) 2019 and (B) 2020. L and Q represent linear and quadratic regressions, respectively. NS indicates a  $P \geq 0.05$ . \*, \*\*, and \*\*\* indicate  $P < 0.05$ , 0.01, and 0.001, respectively.

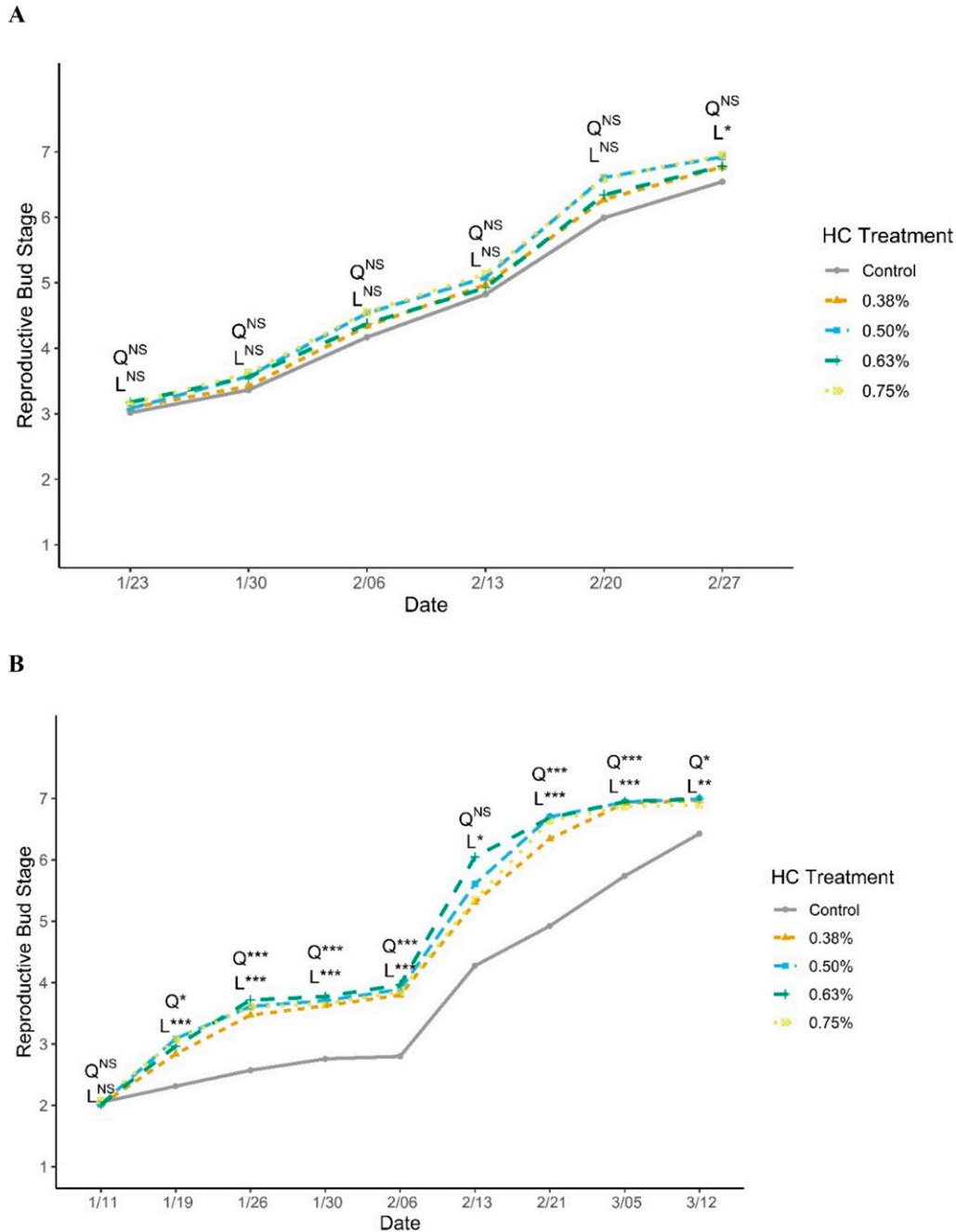


Fig. 2. ‘Colossus’ reproductive bud development as affected by different hydrogen cyanamide (HC) rates in (A) 2019 and (B) 2020. L and Q represent linear and quadratic regressions, respectively. NS indicates a  $P \geq 0.05$ . \*, \*\*, and \*\*\* indicate  $P < 0.05$ , 0.01, and 0.001, respectively.

linear relationship between HC rate and vegetative bud number for the rest of the growing season (Table 1). In ‘Colossus’, data from the

control treatment were left out of the analysis until 13 Feb 2020 (Table 2). There was a linear relationship between HC rate and vegetative bud

number throughout the growing season. For ‘Optimus’, all HC treatments had more actively growing vegetative buds than the control (Table 1).

Table 1. ‘Optimus’ southern highbush blueberry vegetative bud development in plants treated with different rates of hydrogen cyanamide in 2020.

Treatment	No. of actively growing vegetative buds/node on growing date							
	16 Jan	21 Jan	28 Jan	8 Feb	15 Feb	22 Feb	29 Feb	7 Mar
0.63%	0.329	0.329	0.355	0.377 a	0.380 a	0.382 a	0.420 a	0.422 a
0.50%	0.251	0.251	0.263	0.281 a	0.352 a	0.376 a	0.382 a	0.390 a
0.38%	0.255	0.255	0.276	0.299 a	0.335 a	0.351 a	0.361 a	0.375 a
Control	0.001	0.001	0.001	0.001 b	0.145 b	0.155 b	0.182 b	0.200 b
ANOVA $P$ value	NS	NS	NS	***	***	***	***	***
Linear $R^2$	0.11 NS	0.11 NS	0.11 NS	0.765***	0.685***	0.657***	0.627***	0.608***
Quadratic $R^2$	0.62 NS	0.62 NS	0.63 NS	0.778 NS	0.714 NS	0.710 NS	0.642 NS	0.629 NS

Within a column, means followed by the same letter are not significantly different according to Tukey’s honestly significant difference,  $P \leq 0.05$ . The control was not included in the analysis from 16 Jan to 28 Jan because the variance was zero. ANOVA = analysis of variance. NS = indicates a  $P > 0.05$ . \*, \*\*, and \*\*\* indicate  $P \leq 0.05$ , 0.01, and 0.001, respectively.

Table 2. ‘Colossus’ southern highbush blueberry vegetative bud development in plants treated with different rates of hydrogen cyanamide in 2020.

Treatment	No. of actively growing vegetative buds/node on growing date								
	12 Jan	19 Jan	26 Jan	30 Jan	6 Feb	13 Feb	21 Feb	27 Feb	5 Mar
0.75%	0.199 a	0.285 a	0.286 a	0.294 a	0.299 a	0.305 a	0.306 a	0.330 a	0.332 a
0.63%	0.098 b	0.198 ab	0.205 ab	0.216 ab	0.227 ab	0.234 ab	0.248 a	0.270 ab	0.272 ab
0.50%	0.104 ab	0.152 bc	0.160 bc	0.160 bc	0.166 bc	0.169 bc	0.196 ab	0.217 ab	0.222 ab
0.38%	0.045 b	0.072 c	0.79 c	0.080 c	0.081 c	0.102 cd	0.126 bc	0.152 bc	0.173 b
Control <sup>i</sup>	0.000	0.000	0.000	0.000	0.000	0.037 d	0.057 c	0.070 c	0.156 b
ANOVA <i>P</i> value	**	***	***	***	***	***	***	***	**
Linear <i>R</i> <sup>2</sup>	0.43***	0.64***	0.62***	0.63***	0.64***	0.65***	0.64***	0.61***	0.39***
Quadratic <i>R</i> <sup>2</sup>	0.43 NS	0.62 NS	0.60 NS	0.61 NS	0.62 NS	0.70*	0.66 NS	0.62 NS	0.47*

Within a column, means followed by the same letter are not significantly different according to Tukey’s honestly significant difference,  $P \leq 0.05$ . ANOVA = analysis of variance. NS indicates  $P > 0.05$ . \*, \*\*, and \*\*\* indicate  $P \leq 0.05$ , 0.01, and 0.001, respectively.

<sup>i</sup> The control was not included in the analysis from 12 Jan to 6 Feb because the variance was zero.

For ‘Colossus’, the lowest HC concentration did not affect vegetative budbreak compared with the control (Table 2). By the start of harvest, HC-treated plants of both cultivars had nearly twice as many actively growing vegetative buds as plants in the control treatment. In both cultivars, the proportion of mature (stage 6) vegetative buds in HC-treated plants was higher at the beginning of harvest compared with the controls (data not shown).

**Yield and harvest date.** In ‘Optimus’, harvest began on 4 Apr 2019 and ended on 16 May 2019. In ‘Colossus’, harvest began on 4 Apr 2019 and ended on 23 Apr 2019. Plants produced on average between 1750 and 2500 g of fruit per bush by the end of the harvest season in ‘Colossus’ and 4500 to 5500 g of fruit per bush in ‘Optimus’ (Figs. 3 and 4). There was a positive linear relationship between HC rate and amount of fruit harvested for both cultivars early in the season (4 Apr in ‘Colossus’; 4 Apr 2020 and 11 Apr 2020 in ‘Optimus’). These trends continued later in the season but results from the regression analyses were not significant. Total cumulative yield was unaffected by HC treatment.

**Reproductive bud damage and mortality.** HC application had no effect on the number of

florets per reproductive bud in either cultivar in 2020 (Table 3). There were 3.87 to 4.66 berries per reproductive bud in ‘Optimus’ and 3.11 to 3.91 berries per reproductive bud in ‘Colossus’. No effect on reproductive bud mortality was observed in ‘Colossus’ where percent bud mortality ranged from 7.6% to 12.8% in 2019 and 6.3% to 17.5% in 2020 (Table 4). However, higher HC rates caused more bud mortality in ‘Optimus’ in 2019 but not in 2020. The 0.75% HC treatment exhibited the highest mortality rate at 34.1% (Table 4). In 2020, high bud mortality rates were observed in the control treatments, suggesting that the field was suffering from gall midge (*Dasineura oxycoccana*) infestation.

### Discussion

Blueberries produced in Florida are the first domestic blueberry fruit to enter the American market each year. Growers rely on early-season cultivars to target the market window in March and April when prices are highest. For example, a kg of blueberries cost \$12.19 on 4 Apr 2019, when harvest began. By 15 May 2019 (only five weeks later) prices had fallen to \$5.62 per kg (Agronomics 2022). Early production makes growing

blueberries profitable in Florida and other low-chill production areas. However, there is a downward trend in chill hour accumulation over the past 2 decades (Fraise and Karrei 2020). This can delay and reduce harvests. Therefore, HC remains an important tool for blueberry growers because it can result in earlier, more profitable harvest.

Hydrogen cyanamide application can help overcome suboptimal chill accumulation and advance harvests dates for some SHB varieties (Wang et al. 2021; Williamson et al. 2001, 2002). HC application rate is key to a favorable response. A typical rate for HC-tolerant cultivars under fully dormant conditions are 0.75% HC (Harmon et al. 2022). However, effective rates for newly developed cultivars (which are expected to be more sensitive to HC due to changes in the breeding methods; Munoz, personal communication) are unknown. Rates that are too high can cause severe damage to flower buds, which can reduce yield but may advance harvest date (Williamson et al. 2001). As indicated by our bud mortality results, even ideal rates can cause mild phytotoxicity. In this experiment, high HC application rates hindered reproductive bud development at the start of the 2019–20 season. This apparent delay was

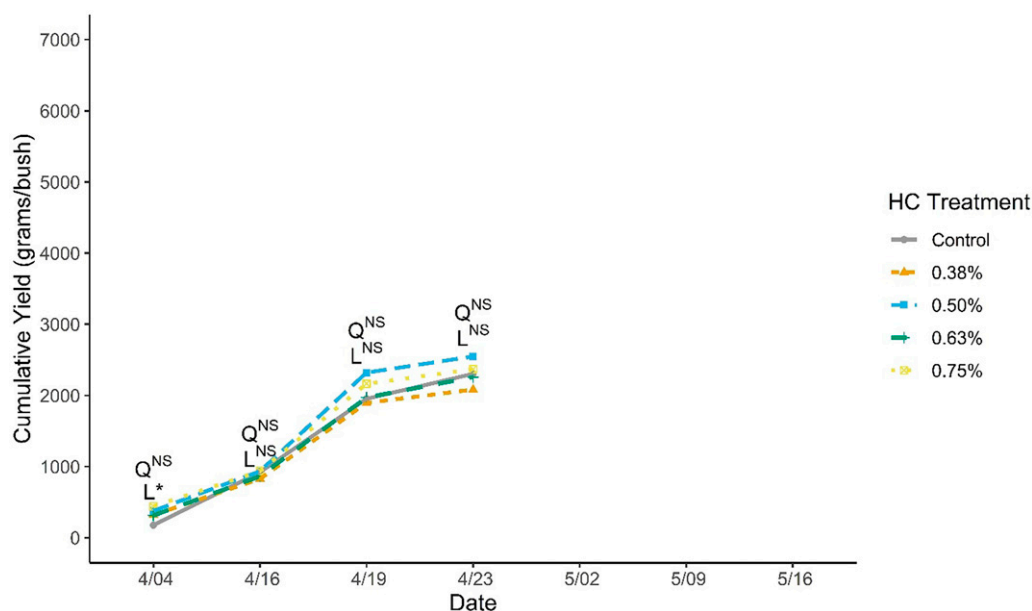


Fig. 3. ‘Colossus’ cumulative yield (grams/bush) over the 2019 harvest season in plants treated with different hydrogen cyanamide (HC) rates. L and Q represent linear and quadratic regressions, respectively. NS indicates a  $P \geq 0.05$ . \*, \*\*, and \*\*\* indicate  $P < 0.05$ , 0.01, and 0.001, respectively.

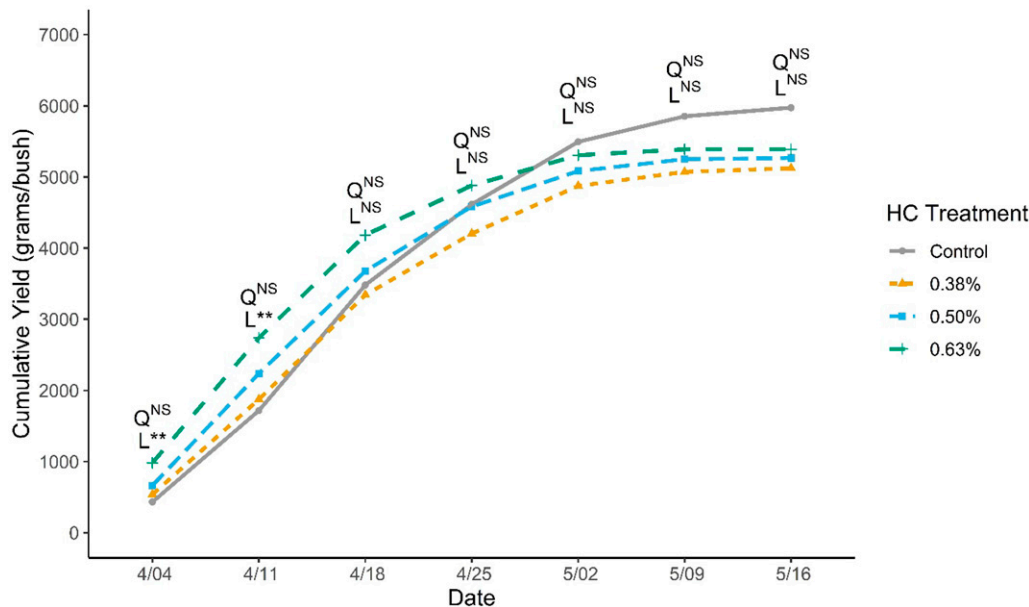


Fig. 4. ‘Optimus’ cumulative yield (grams/bush) over the 2019 harvest season in plants treated with different hydrogen cyanamide (HC) rates. L and Q represent linear and quadratic regression, respectively. NS indicates a  $P \geq 0.05$ . \*, \*\*, and \*\*\* indicate  $P < 0.05$ , 0.01, and 0.001, respectively.

probably caused by reproductive bud damage. Damaged buds are not immediately apparent, and their development scores were likely stagnant for 1 or more weeks after HC application. Therefore, HC had a partial inflorescence thinning effect. HC has been shown to also have thinning effects in apple and plum (Fallahi et al. 1992). Later in the season, reproductive budbreak in HC-treated plants was equal to or greater than the controls.

Hydrogen cyanamide application also affected vegetative budbreak. ‘Optimus’ and ‘Colossus’ exhibited more and earlier vegetative budbreak in HC-treated plants compared with the controls. The observed increase in vegetative budbreak agrees with previous studies in blueberry and other fruit crops (Cook et al. 2001; Williamson et al. 2002; Stringer et al. 2003). Photosynthesis in vegetative organs likely served as carbohydrate sources for reproductive bud development. Thus, accelerated reproductive growth might be related to the availability of carbohydrates to support reproductive bud development.

As expected, HC rate had a positive linear relationship with weekly yield in both ‘Optimus’ and ‘Colossus’ early in the harvest season. This suggests that both cultivars can potentially

benefit from HC application. In previous studies, HC applications led to higher total yields in blueberry (Lin and Agehara 2021; Wang et al. 2021; Williamson et al. 2002) and other woody perennial crops (Ghrab and Mimoun 2014). Here, HC-treated ‘Optimus’ or ‘Colossus’ did not exhibit higher seasonal yields than controls. For ‘Optimus’, all HC treatments showed a nonsignificant trend for lower seasonal yields compared with the control. Previous research suggests that phytotoxic effects of using HC may include damage to reproductive bud structures resulting in fewer berries per inflorescence or death of reproductive buds resulting in lower berry yields (Williamson et al. 2001). It is possible that ‘Optimus’ reproductive buds were more advanced than ‘Colossus’ reproductive buds at the time of HC application and therefore were damaged to a greater extent. Nevertheless, considering the high farmgate prices in early spring, HC application in ‘Optimus’

might still be more profitable than production without this PGR.

In the current study, ‘Optimus’ berry yields at the first two harvest dates increased linearly with increasing HC rate. HC applied at 0.63% had the highest yield at these early dates, but higher rates were not tested because previous observations suggested ‘Optimus’ was highly sensitive to HC. A similar linear response was observed for ‘Colossus’ at the first harvest date only. Although significant ( $P \leq 0.05$ ), the magnitude of the yield differences for ‘Colossus’ were small and may not be practically relevant. HC application when ~168 chill hours had been accumulated increased vegetative budbreak and thinned reproductive buds. Previous research in blueberry has established that the ratio of vegetative organs (leaves) to flower buds affects fruit development rates (Maust et al. 1999). Thus, this change in source–sink relations might have led to accelerated reproductive bud development and achieved higher early season yields. Because this is the period when harvest is most profitable, our results suggest that HC application could be beneficial for SHB growers in Florida who grow the higher yielding ‘Optimus’. Our results support the hypotheses that higher HC rates cause 1) more flower bud injury and 2) higher rates of vegetative budbreak. However, berry ripening and harvest were only slightly accelerated. The risk of reproductive bud thinning from HC should be considered, especially when the increase in early harvest is minimal for an already early-season, light-cropping cultivar as was observed with ‘Colossus’.

Table 4. Reproductive bud mortality (%) in ‘Optimus’ and ‘Colossus’ southern highbush blueberry plants treated with different rates of hydrogen cyanamide during the 2019 and 2020 seasons.

Treatment	Cultivar			
	Optimus		Colossus	
	2019	2020	2019	2020
0.75%	NA <sup>1</sup>	NA	11.7 a	8.4 a
0.63%	34.1 a	59.8 a	10.7 a	6.1 a
0.50%	18.9 b	50.0 a	12.8 a	9.8 a
0.38%	7.0 c	23.4 a	7.6 a	6.3 a
Control	3.0 c	24.2 a	12.1 a	17.5 a
ANOVA $P$ value	**	NS	NS	NS
Linear $r^2$	***	**	NS	NS

Within a column, means followed by the same letter are not significantly different according to Tukey’s honestly significant difference,  $P \leq 0.05$ . NA = not applied. NS indicates  $P > 0.05$ . \*, \*\*, and \*\*\* indicate  $P \leq 0.05$ , 0.01, and 0.001, respectively.

Table 3. Number of berries produced per reproductive bud in ‘Optimus’ and ‘Colossus’ southern highbush blueberry plants treated with different rates of hydrogen cyanamide in 2020.

Treatment	Cultivar	
	Optimus	Colossus
0.75%	NA	3.31
0.63%	4.66	3.72
0.50%	3.87	3.47
0.38%	3.87	3.11
Control	4.22	3.91
Linear $R^2$	NS	NS

NA = not applied. NS indicates  $P > 0.05$ . \*\*\*, \*\* and \* indicate  $P \leq 0.05$ , 0.01, and 0.001, respectively.

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