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Effects of 2,4-D choline on fruiting in sensitive cotton

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Research Article

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Abstract

With the increase in hectares planted to auxin-resistant cotton, the number of preplant, at-plant, and postplant applications of dicamba and 2,4-D choline to aid in the control of troublesome broadleaf weeds, including glyphosate-resistant Palmer amaranth, has increased. More dicamba and 2,4-D choline applications mean an increased risk of off-target movement. Field studies were conducted in 2019 to 2021 at the Texas Tech University New Deal Research Farm to evaluate dicamba-resistant cotton response to various rates of 2,4-D choline when applied at four growth stages (first square [FS] + 2 wk, first bloom [FB], FB + 2 wk, and FB + 4 wk). Applications of 2,4-D choline were applied at 1,060 (1X), 106 (1/10X), 21 (1/50X), 10.6 (1/100X), 2.1 (1/500X), and 1.06 (1/1000X) g ae ha^{-1} to Deltapine 1822 XF cotton. Relative to the nontreated control, yield losses were observed in all years at FS + 2 wk and FB from rates of 2,4-D choline ≥ 1/100X. At the FB + 4 wk application, only the 1X rate of 2,4-D choline resulted in a yield reduction in all three years. Micronaire, fiber length, and uniformity were negatively influenced by the 1/10X and 1X rates of 2,4-D choline at various timings in 2019, 2020, and 2021. In addition, short fiber content, neps, and seed coat neps increased where micronaire, fiber length, and uniformity were negatively impacted.

Introduction

Upland cotton was planted on more than 5 million hectares in the United States in 2022 (USDA-NASS 2022). Weeds produce the highest potential for yield loss at 34% compared to other agronomic pests (Oerke 2005). Broadleaf weed control prior to the release of glyphosate-resistant cotton cultivars was achieved using a combination of residual herbicides applied preplant, preemergence (PRE), and postemergence (POST) in addition to postemergence-directed applications and tillage (Keeling and Abernathy 1989; Keeling et al. 1989; Keeling et al. 1991; Snipes and Mueller 1992). The release of glyphosate-resistant cotton cultivars drastically shifted weed control to a more chemical approach using a single mode of action, rather than relying on multiple residual herbicides or mechanical weed control (Dill et al. 2008; Norsworthy et al. 2007). As a result, glyphosate resistance in Palmer amaranth was first reported in 2005 in Georgia and has spread across the Cotton Belt in years following (Culpepper et al. 2006; Heap 2022; Sosnoskie et al. 2012). Older chemistries, as well as physical and mechanical weed control practices, are now being reevaluated to combat the growing problem of glyphosate-resistant weed populations.

Since its release in the 1940s, 2,4-D, a synthetic auxin herbicide, has been used to control broadleaf weeds in small grains and monocot crops (Cast 1975). Prior to the release of Enlist® cotton (Corteva Agriscience™, Indianapolis, IN, USA), which can metabolize certain HRAC Group 4 herbicides due to the insertion of gene *AAD-12*, which enables the plant to metabolize certain herbicides to an inactive molecule, 2,4-D could be used only as a preplant burndown or a POST harvest application to control broadleaf weeds (Baker 1993; Everitt and Keeling 2007; Keeling et al. 1989; Wright et al. 2010). Applications of 2,4-D over the top in cotton have shown to be effective in controlling Palmer amaranth in recent years (Manuchehri et al. 2017; Merchant et al. 2014).

An increased risk of off-target movement of auxin herbicides has occurred following the release of both technologies. Off-target movement of auxin herbicides, depending on rate and timing, can lead to delays in maturity as well as decreases in yield and fiber quality (Buol et al. 2018, 2019; Byrd et al. 2015; Everitt and Keeling 2009; Manuchehri et al. 2019; Russell et al. 2020). Byrd et al. (2015), Manuchehri et al. (2019), and Buol et al. (2019) reported that early-season applications of 2,4-D prior to bloom are more injurious than postbloom applications.

Although information exists on the influence of 2,4-D on lint reductions and fiber quality measured using the High Volume Instrument (HVI; Uster Technologies, Uster, Switzerland) (Buol et al. 2019; Manuchehri et al. 2019), effects on fiber quality measured using the Advanced Fiber Information System (AFIS; Uster Technologies) at different rates of 2,4-D choline applied to different growth stages of cotton are poorly understood. The objectives for this study were to determine the effects 2,4-D choline rate and timing of application on changes to boll distribution and retention, yield, and fiber quality measurements.

Materials and Methods

Experimental Design and Management Practices

Field experiments were conducted in 2019, 2020, and 2021 at the Texas Tech University New Deal Research Farm (33.44°N, 101.43° W), equipped with subsurface drip irrigation. Deltapine 1822 XF (Bayer CropScience, St. Louis, MO, USA) cotton was planted at 101,300 seeds ha^{-1} on May 16, 2019, May 18, 2020, and June 5, 2021. Plots, four rows spaced 1.02 m apart by 9.1 m, were arranged as a randomized complete block with 24 treatments plus 1 nontreated control (NTC) in a factorial arrangement. Treatments were replicated 4 times, and plots were kept weed-free throughout the growing seasons. In 2019 and 2020, 32-0-0 fertilizer at 70 kg ha^{-1} was applied through the drip irrigation. Due to increased rainfall and delayed planting date, 40 kg ha^{-1} of nitrogen was applied in 2021.

Factor 1 was an application rate of 2,4-D choline (Enlist One®, Corteva Agriscience) at 1,060 (1X), 106 (1/10X), 21 (1/50X), 10.6 (1/100X), 2.1 (1/500X), and 1.06 (1/1,000X) g ae ha^{-1} (Table 1). Factor 2 was an application timing at first square (FS) + 2 wk, first bloom (FB), FB + 2 wk, and FB + 4 wk (Table 2). Treatments were applied using a CO_2-pressurized backpack sprayer calibrated to deliver 140 L ha^{-1} equipped with TTI 11002 nozzles (TeeJet® Technologies, Glendale Heights, IL, USA), which produced ultra-coarse droplets to minimize off-target movement. Accumulated growing degree days (GDD15.6), computed as the average of the daily maximum and minimum air temperatures minus a base temperature of 15.6 C (Hake et al. 1990; Peng et al. 1989), were calculated from data collected from a weather station 200 m from the study (Model GRWS100, Campbell Scientific, Logan, UT, USA) (Table 3). Defoliation applications occurred for all plots when the NTC reached 70% open boll in all three years.

Box Mapping

Prior to machine harvest, a 1.5-m^2 plant sample was harvested from a center row of each plot. Fruit were removed by fruiting site from each plant and placed in a grid box representing node and fruiting position in a method described by Bednarz et al. (2005) and Ritchie et al. (2011). Fruit was further grouped by flowering date by combining first-position fruit with second-position fruit two nodes lower on the plant and third-position fruit four nodes lower on the plant, as described by Schaefer et al. (2017). A weighted average smoothing function was used for each plot, with 20% of each value coming from each adjacent node and 60% coming from the central node for each fruiting site, based on Ritchie et al. (2011).

Cotton Seed Yield

Immediately after box mapping, the residual plots were machine harvested using a two-row John Deere 7445 cotton stripper equipped with load cells (Rusty's Weigh, Lubbock, TX, USA). To determine lint weight and fiber quality values, samples were ginned and submitted for HVI and AFIS testing to the Texas Tech University Fiber and Biopolymer Institute in Lubbock, TX.

Statistical Analysis

Statistical analysis was conducted using the generalized linear mixed model (GLIMMIX) procedure in SAS 9.4 (SAS Institute, Cary, NC, USA). The interaction of year with treatment factors was found to be significant, so treatment analysis was evaluated within year. The combination of rate and timing was treated as a fixed treatment effect, and replicate was considered a random effect (Littell et al. 2006). Treatment differences that were significant using a Type III test of fixed effects were tested for differences in mean using Fisher's protected least significant difference at $\alpha = 0.05$. Boll distribution was tested by node, and 95% confidence intervals are presented for the control (Figure 1). Boll numbers by node for each treatment that fell outside these confidence intervals were considered significant, provided the Type III tests were significant. The interaction of herbicide rate and timing was tested for significance among yield, HVI, and AFIS fiber quality parameters (length, strength, etc.) by year. The rate × timing interactions in each year were significant ($P_{critical} = 0.05$), so although main effects were also significant in most cases, further analysis was based on the combination of rate and timing, rather than on the main effects of rate and timing separately.

Results and Discussion

Environment by Year

During the three years these trials were conducted, vastly different environmental conditions were experienced. In 2019, cotton was planted on May 16, and the FS + 2 wk application was applied 61 d after, whereas in 2020, cotton was planted on May 18 and first sprayed 45 d later, on July 2 (Table 2). Differences in the amount of time required to reach FS + 2 wk can be attributed to the cooler weather in 2019, when the accumulated growing degree days were approximately half of those experienced during the 2020 growing season (Table 3). In addition to heat units, although all three years were abnormally dry, 2019 received more than double the amount of rainfall in May and September than was received in 2020.

The 2021 growing season received 120 mm of rainfall in May, which resulted in planting being delayed until June 5 (Table 3). Due to the delay in planting, only 40 kg ha^{-1} of 32-0-0 could be applied relative to the 70 kg ha^{-1} applied in both 2019 and 2020. Though not tested, the delayed planting, in addition to the decreased nitrogen fertilizer application, likely explains the lint production and fiber quality differences observed between years.

Boll Production and Reduction

First Square + 2 wk

At FS + 2 wk, the 1X rate of 2,4-D choline resulted in compete boll loss in all three years (Figure 1). The 1/10X rate resulted in substantial boll losses above node 9. In all years, boll production

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Table 1. Rates of 2,4-D used for off-target movement applications.

2,4-D choline rate	Relative rate to standard POST application of 2,4-D choline
g ae ha^{-1}	
1,060	1X
106	1/10X
21	1/50X
10.6	1/100X
2.1	1/500X
1.06	1/1,000X

Table 2. Cotton growth stage and timings of applications.[a,b]

Cotton growth stage	Application date		
	2019	2020	2021
FS + 2 wk	16 Jul (61)	2 Jul (45)	27 Jul (52)
FB	22 Jul (67)	17 Jul (60)	2 Aug (58)
FB + 2 wk	5 Aug (81)	31 Jul (74)	16 Aug (72)
FB + 4 wk	19 Aug (95)	14 Aug (88)	30 Aug (86)

[a]Cotton growth stages were in agreement with Ritchie et al. (2004). Days after planting are in parentheses.
[b]Abbreviations: FB, first bloom; FS, first square.

Table 3. Heat units, rainfall, and irrigation by month in 2019, 2020, and 2021 at the Texas Tech University Research Farm, New Deal, TX.

Year	Month	Accumulated heat units[a]	Rainfall	Irrigation
			——— mm ———	
2019	May	119	108	76
	Jun	265	45	141
	Jul	360	26	138
	Aug	412	29	134
	Sep	282	223	66
	Oct	43	39	0
	Cumulative	*1,481*	*470*	*555*
2020	May	205	39	122
	Jun	332	36	103
	Jul	429	60	106
	Aug	395	7	144
	Sep	149	77	10
	Oct	76	24	0
	Cumulative	*1,586*	*243*	*485*
2021	May	119	120	0
	Jun	299	74	26
	Jul	309	48	76
	Aug	326	79	77
	Sep	265	2	37
	Oct	98	0	0
	Cumulative	*1,416*	*323*	*216*

[a]Computed as the average of the daily maximum and minimum air temperatures minus a base temperature of 15.6 C for each month (Hake et al. 1990; Peng et al. 1989).

above node 11 decreased following both the 1/50X and 1/100X rates of 2,4-D choline. At the 1/500X and 1/1,000X rates, boll losses were observed above node 16 in 2019, above node 15 in 2020, and above node 16 in 2021. Similar results were observed by Buol et al. (2019), who saw seed cotton yield decrease from first-position fruiting sites following 8.3 g ae ha^{-1} of 2,4-D when applied to cotton at first full square.

First Bloom

At FB, the 1X rate of 2,4-D choline resulted in complete boll loss in 2019 and severe boll loss in both 2020 and 2021 (Figure 1). Following the 1/10X and 1/50X rates, boll reductions were observed at and above node 9 in all three years. The 1/100X rate resulted in a reduction in boll production starting at node 9 in 2019 and at node 10 in both 2020 and 2021. When the 1/500X rate of 2,4-D choline was applied, boll reductions were observed at node 11 and above in 2019, between nodes 15 and 21 in 2020, and between nodes 10 and 13 in 2021. At the 1/1,000X rate, boll reductions were observed at node 11 and between nodes 14 and 18 in 2019 and at node 11 in 2021. In 2020, an increase in boll production was observed from nodes 12, 13, and 14 relative to the NTC. Excluding the 1/1,000X rate in 2020, when an increase in boll production was observed, these results are similar to those reported by Manuchehri et al. (2019), who saw boll retention decrease at higher rates of 2,4-D (18.3 and 183 g ae ha^{-1}) when applied to cotton at FB.

First Bloom + 2 wk

The 1X rate of 2,4-D choline reduced boll production in all three years, but complete boll loss was not observed at FB + 2 wk compared to the applications made at FS + 2 wk and FB. Boll reductions were observed above node 6 in 2019 and above node 8 in both 2020 and 2021 from the 1X rate. Following the 1/10X rate of 2,4-D choline, boll reductions were observed above node 9 in both 2019 and 2021. In 2020, boll reductions were observed above node 11 when compared to the NTC. At the 1/50X rate, boll reductions were observed above node 11 in 2019, above node 13 in 2020, and above node 10 in 2021. When the 1/100X rate was applied at FB + 2 wk, boll reductions were observed from nodes 11 to 18 in 2019, from nodes 13 to 18 in 2020, and on nodes 15 and 16 in 2021. In 2019, boll reductions were observed from nodes 11 to 18 at the 1/500X rate and from nodes 15 to 17 at the 1/1,000X rate. These data are in agreement with those reported by Byrd et al. (2015), who reported a decrease in total boll number in all three location groups following applications of 40 g ae ha^{-1} of 2,4-D applied at FB + 2 wk. In both 2020 and 2021, no significant boll reductions were observed at any node, which is similar to observations by Byrd et al. (2015) and Buol et al. (2019), who observed no losses following applications of 2,4-D at 2 g ae ha^{-1} and 8.3 g ae ha^{-1}, respectively, applied at FB + 2 wk.

First Bloom + 4 wk

In 2019, only bolls produced above node 15 experienced reductions. Few to no differences were observed between any of the treatments, regardless of rate, at this timing. In both 2020 and 2021, only the 1X rate of 2,4-D choline resulted in boll reductions relative to the NTC. No differences in boll production were observed from 2,4-D rates ≤1/10X compared to the NTC. These data show that as cotton becomes more mature, its tolerance to 2,4-D increases. Similar results were observed by both Byrd et al. (2015) and Buol et al. (2019), who reported that cotton is less sensitive to 2,4-D choline after the FB growth stage.

Lint Yield

Unlike individual bolls on a plant, which are affected differently at a given spray rate and timing based on maturity, both lint yield and fiber quality are measurements of impact on the entire plant. In the case of off-target herbicide applications, it is important to determine the prospective impact based on stage of growth and rate of application. Therefore both rate and timing impacts are reported for both lint yield and fiber quality within each year.

In 2019, lint reductions were observed from all treatments, except the 1/1,000X and 1/500X rates of 2,4-D choline, at FB + 2

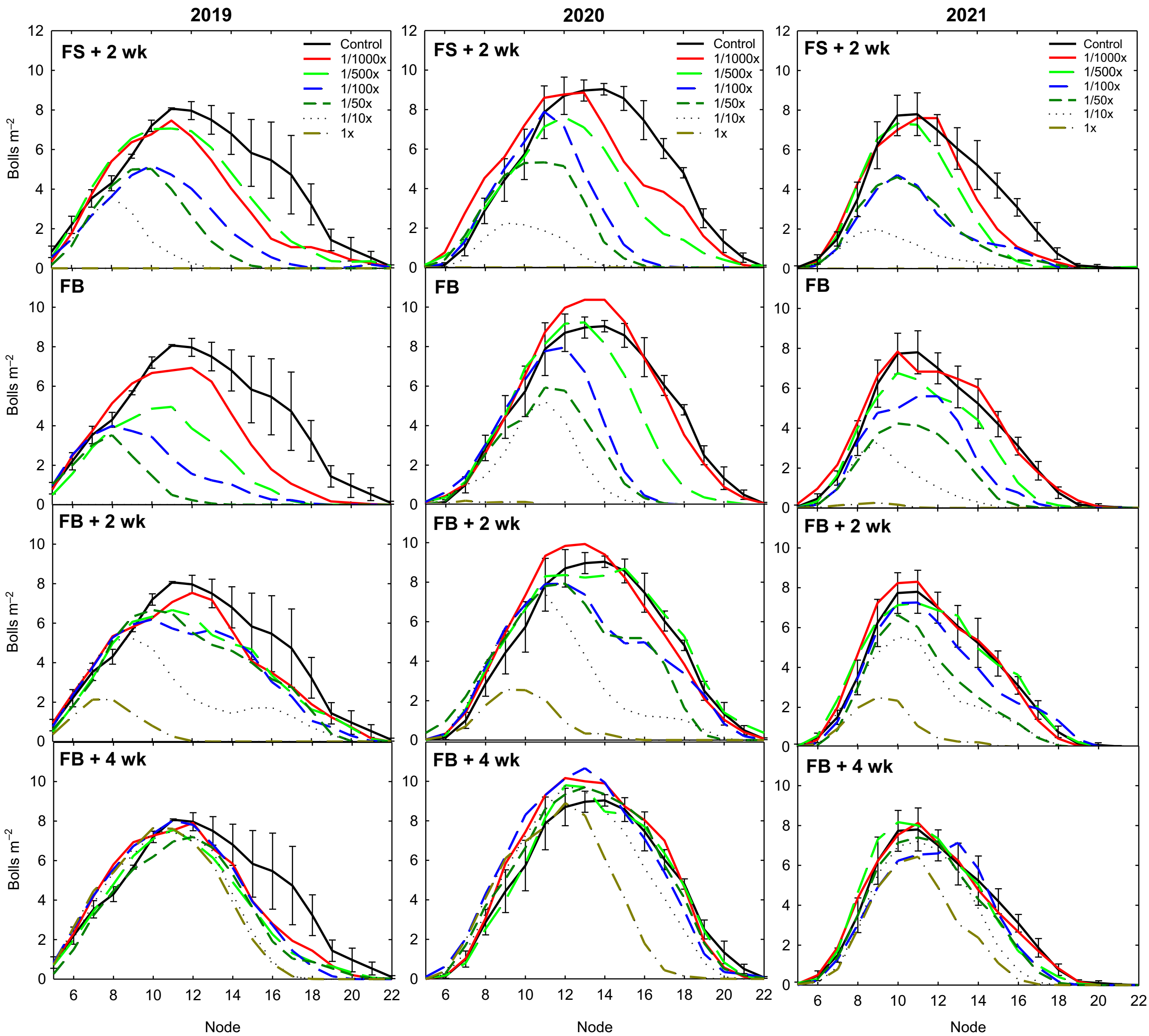

Figure 1. Box mapping boll distribution by rate and timing in 2019, 2020, and 2022. Error bars on the nontreated control represent the 95% confidence intervals based on a generalized linear mixed model. FB, first bloom; FS, first square.

wk (Table 4). The 1/100X, 1/50X, 1/10X, and 1X rates resulted in the greatest lint reductions when applied at FS + 2 wk and FB. At FB + 2 wk and FB + 4 wk, lint was less affected by these higher rates of 2,4-D choline, indicating that cotton is less sensitive when it is more mature.

During the 2020 growing season, no lint reductions were observed following 2,4-D choline at the 1/1,000X or 1/500X rate, regardless of application timing (Table 5). Although lint reductions were observed following the 1/100X and 1/50X rates at FS + 2 wk and FB, no reductions were observed at the FB + 2 wk and FB + 4 wk application timings. At the 1/10X rate of 2,4-D choline, losses were observed at FS + 2 wk, FB, and FB + 2 wk; however, no differences were observed at FB + 4 wk. When the 1X rate of 2,4-D choline was applied to susceptible cotton, complete plant death or significant yield reductions were observed at all application timings.

In 2021, 2,4-D choline at the 1/1,000X rate did not reduce lint production at any of the application timings (Table 6). When the 1/500X rate was applied, only the FS + 2 wk application timing resulted in a lint yield reduction. At both the 1/100X and 1/50X rates, lint reductions were observed at FS + 2 wk and FB. No lint reductions were observed following the 1/100X and 1/50X rates at the FB + 2 wk and FB + 4 wk application timings. When 2,4-D choline was applied at the 1/10X and 1X rates in 2021, lint reductions were reported at all application timings.

During the 2019, 2020, and 2021 growing seasons, applications of 2,4-D choline applied at FS + 2 wk were more injurious than those applied at FB + 2 wk and FB + 4 wk, indicating that cotton is less susceptible to yield losses as it becomes more mature (Tables 4 to 6). These data are in agreement with data reported by Everitt and Keeling (2007), Buol et al. (2019), and Byrd et al. (2015), who reported that applications of 2,4-D prior to FB are more injurious

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Table 4. Least-square means of yield and fiber quality parameters of 25 treatments (rate × timing) applied in 2019.[a,b,c,d]

Rate	Timing	Yield	Micronaire	Length	Uniformity	Strength
		kg ha^{-1}		mm	%	kN m kg^{-1}
NTC	—	1,362 a	4.46 b–e	26.9 ab	80.3 ab	253.8 d–f
1/1,000X	FS + 2 wk	1,095 b–d	4.53 a–d	27.1 a	80.0 a–c	257.7 b–f
	FB	1,095 b–d	4.42 b–e	26.6 a–e	79.8 a–d	259.7 a–e
	FB + 2 wk	1,169 a–c	4.41 c–e	27.0 ab	80.3 ab	261.7 a–d
	FB + 4 wk	1,105 b–d	4.70 a	26.2 c–g	79.4 b–e	250.9 e–g
1/500X	FS + 2 wk	1,049 b–d	4.45 b–e	26.8 a–d	79.8 a–d	259.7 a–e
	FB	1,057 b–d	4.52 a–d	26.4 a–f	79.5 b–e	252.8 d–f
	FB + 2 wk	1,225 ab	4.38 d–f	26.6 a–e	79.8 a–d	255.8 c–f
	FB + 4 wk	1,031 b–d	4.61 a–c	26.0 e–g	79.7 b–d	251.9 d–f
1/100X	FS + 2 wk	758 e	4.35 d–g	27.1 a	79.8 a–d	254.8 d–f
	FB	675 ef	4.38 d–f	26.3 b–f	78.6 d–f	239.1 f–h
	FB + 2 wk	964 d	4.46 b–e	26.5 a–e	79.6 b–e	254.8 d–f
	FB + 4 wk	1,092 b–d	4.64 ab	26.9 a–c	81.0 a	264.6 a–c
1/50X	FS + 2 wk	507 fg	4.34 d–g	26.5 a–f	78.3 ef	238.1 i
	FB	462 g	4.16 fg	25.9 e–g	77.8 f	240.1 hi
	FB + 2 wk	964 d	4.27 e–g	26.2 c–g	78.8 c–f	249.9 fg
	FB + 4 wk	1,033 b–d	4.64 ab	26.1 d–g	79.6 b–e	255.8 c–f
1/10X	FS + 2 wk	152 h	3.51 i	25.5 g	76.0 g	227.4 j
	FB	195 h	3.75 h	25.8 fg	77.8 f	241.1 g–i
	FB + 2 wk	587 e–g	4.15 g	27.1 a	80.1 a–c	269.5 a
	FB + 4 wk	999 cd	4.49 a–e	26.8 a–d	80.5 ab	266.6 ab
1X	FS + 2 wk[e]	0 —	—	—	—	—
	FB[e]	13 h	—	—	—	—
	FB + 2 wk[e]	27 h	—	—	—	—
	FB + 4 wk	403 g	2.58 j	24.4 h	74.5 h	206.8 k

[a]Yield was determined through mechanical harvest from residual plot following box mapping.
[b]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[c]Abbreviations: FB, first bloom; FS, first square; NTC, nontreated control.
[d]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[e]Treatment not included in analysis of variance.

Table 5. Least-square means of yield and fiber quality parameters of 25 treatments (rate × timing) applied in 2020.[a,b,c,d]

Rate	Timing	Yield	Micronaire	Length	Uniformity	Strength
		kg ha^{-1}		mm	%	kN m kg^{-1}
NTC	—	1,587 a–c	4.13 c–g	29.5 a–c	80.9 b–e	310.7 b–e
1/1,000X	FS + 2 wk	1,594 a–c	4.33 ab	29.0 c–f	80.1 c–g	303.8 c–f
	FB	1,732 a	4.15 c–g	29.1 c–f	80.5 b–g	310.7 b–e
	FB + 2 wk	1,508 a–c	4.04 fg	29.0 c–f	80.5 b–g	308.7 b–e
	FB + 4 wk	1,579 a–c	4.21 a–f	28.9 c–f	80.6 b–f	301.8 c–f
1/500X	FS + 2 wk	1,420 bc	4.21 a–f	29.0 c–f	79.8 e–g	301.8 c–f
	FB	1,512 a–c	4.27 a–d	28.7 ef	79.4 gh	292.0 f
	FB + 2 wk	1,611 a–c	4.01 gh	29.0 c–f	80.4 b–g	307.7 b–e
	FB + 4 wk	1,508 a–c	4.15 c–g	29.5 a–c	80.9 b–d	313.6 bc
1/100X	FS + 2 wk	891 d	4.34 a	29.2 c–e	80.5 b–g	308.7 b–e
	FB	821 de	4.18 a–f	28.8 d–f	79.7 fg	306.7 b–e
	FB + 2 wk	1,444 a–c	4.15 b–g	29.5 a–c	81.3 ab	312.6 b–d
	FB + 4 wk	1,628 ab	4.21 a–f	29.1 c–f	80.9 b–e	311.6 b–d
1/50X	FS + 2 wk	717 de	4.12 c–g	28.6 ef	79.8 d–g	297.9 ef
	FB	557 e	4.1 d–g	29.0 c–f	79.8 e–g	296.0 ef
	FB + 2 wk	1,324 c	4.15 c–g	29.8 ab	81.0 bc	321.4 ab
	FB + 4 wk	1,484 a–c	4.22 a–e	29.3 b–d	81.0 bc	306.7 b–e
1/10X	FS + 2 wk	136 f	3.84 hi	28.6 ef	79.5 gh	301.8 c–f
	FB	236 f	3.67 ij	28.9 c–f	79.5 f–h	310.7 b–e
	FB + 2 wk	810 de	4.08 e–g	30.0 a	82.3 a	331.2 a
	FB + 4 wk	1,411 bc	4.29 a–c	29.5 a–c	81.1 bc	312.6 b–d
1X	FS + 2 wk[e]	0 —	—	—	—	—
	FB[e]	0 —	—	—	—	—
	FB + 2 wk	121 f	2.72 l	27.9 g	78.4 hi	297.9 d–f
	FB + 4 wk	891 d	3.48 k	29.0 c–f	79.8 d–g	312.6 b–d

[a]Yield was determined through mechanical harvest from residual plot following box mapping.
[b]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[c]Abbreviations: FB, first bloom; FS, first square; NTC, nontreated control.
[d]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[e]Treatment not included in analysis of variance.

Table 6. Least-square means of yield and fiber quality parameters of 25 treatments (rate × timing) applied in 2021.[a,b,c,d]

Rate	Timing	Yield	Micronaire	Length	Uniformity	Strength
		kg ha^{-1}		mm	%	kN m kg^{-1}
NTC	—	1,152 a	4.05 ab	28.4 b–d	79.6 a–g	296.9 b–f
1/1,000X	FS + 2 wk	1,017 ab	4.11 ab	28.6 a–c	80.0 a–d	298.9 a–d
	FB	1,006 ab	4.02 a–c	28.3 cd	78.8 g–i	296.9 b–f
	FB + 2 wk	1,187 a	4.08 ab	28.6 a–c	79.5 a–g	295.0 c–h
	FB + 4 wk	1,113 a	4.06 ab	28.4 b–d	79.8 a–f	292.0 d–i
1/500X	FS + 2 wk	905 b	4.19 a	28.5 a–d	79.3 c–h	289.1 d–i
	FB	1,097 ab	4.06 ab	27.8 de	79.2 d–h	286.2 e–j
	FB + 2 wk	1,118 a	4.08 ab	28.4 b–d	79.5 b–g	297.9 a–e
	FB + 4 wk	1,177 a	4.05 ab	28.4 b–d	79.9 a–e	297.9 a–e
1/100X	FS + 2 wk	384 d	4.02 a–c	28.6 a–c	78.5 hi	282.2 i–k
	FB	613 c	3.84 c–e	27.8 de	79.4 b–g	293.0 d–i
	FB + 2 wk	1,072 ab	3.92 b–d	29.0 a–c	80.2 a–c	307.7 ab
	FB + 4 wk	1,091 ab	4.08 ab	28.6 a–c	80.0 a–d	298.9 a–d
1/50X	FS + 2 wk	261 de	3.80 de	28.6 a–c	78.9 f–i	284.2 g–j
	FB	650 c	3.53 fg	27.8 de	78.1 i	287.1 d–j
	FB + 2 wk	1,017 ab	3.84 c–e	29.1 ab	79.5 a–g	309.7 a
	FB + 4 wk	1,086 ab	4.09 ab	28.7 a–c	80.3 ab	306.7 a–c
1/10X	FS + 2 wk	135 ef	3.65 ef	28.5 a–d	78.1 i	283.2 h–k
	FB	163 ef	2.95 h	26.4 g	76.5 j	275.4 jk
	FB + 2 wk	680 c	4.07 ab	29.2 a	80.4 a	306.7 a–c
	FB + 4 wk	917 b	4.14 a	28.4 b–d	80.0 a–d	296.0 c–g
1X	FS + 2 wk[e]	0 —	—	—	—	—
	FB	45 f	3.04 h	27.1 e–g	77.2 j	275.4 jk
	FB + 2 wk	208 d–f	3.43 g	27.1 fg	79.0 e–i	286.2 f–j
	FB + 4 wk	623 c	2.70 i	27.6 ef	77.0 j	271.5 k

[a]Yield was determined through mechanical harvest from residual plot following box mapping.
[b]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[c]Abbreviations: FB, first bloom; FS, first square; NTC, nontreated control.
[d]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[e]Treatment not included in analysis of variance.

than those applied after. In both 2020 and 2021, the 1/50X rate of 2,4-D choline applied at FS + 2 wk resulted in yield losses, whereas the same rate when applied at FB + 4 wk did not affect yield (Tables 5 and 6).

Fiber Quality

The impact of 2,4-D choline on fiber quality varied across years, rate, and growth stage. During the 2019 growing season, lower rates of 2,4-D choline had an inconsistent impact on fiber quality. When 2,4-D choline was applied at the 1/1,000X rate at FB + 4 wk, micronaire increased while length decreased relative to the NTC (Table 4). A reduction in length also was observed when 2,4-D choline was applied at FB + 4 wk at the 1/500X rate. At the 1/100X rate of 2,4-D choline, uniformity decreased while short fiber content (SFC) increased when applied at FB (Tables 4 and 7). When 2,4-D choline at 1/100X was applied at FB + 4 wk, fiber strength increased.

Most of the significant deleterious effects of the 2,4-D choline treatments on fiber quality occurred at rates above 1/50X in 2019. The 2,4-D choline treatment at the 1/50X rate applied at FS + 2 wk negatively impacted uniformity, strength, neps per gram, SFC, and seed coat neps (SCN). At FB, all fiber quality measurements that were evaluated were negatively influenced at the same rate of 2,4-D. Fiber length, uniformity, and SFC were negatively influenced at FB + 2 wk following the 1/50X rate. When the 1/50X rate was applied at FB + 4 wk, only fiber length was negatively impacted.

When the 1/10X rate was applied at FS + 2 wk and FB, all fiber quality measurements evaluated were negatively impacted. At the FB + 2 wk timing, micronaire, SFC, and SCN were negatively influenced. Fiber strength increased relative to the NTC at both FB + 2 wk and FB + 4 wk following the 1/10X rate of 2,4-D choline. HVI and AFIS testing could not be evaluated following the 1X rate at FS + 2 wk, FB, and FB + 2 wk due to overall plant injury and insufficient lint collected for testing (Table 4). The FB + 4 wk timing was the only timing to produce sufficient lint, and all fiber quality measurements were negatively influenced at this rate and timing.

In 2020, micronaire increased at FS + 2 wk following the 1/1,000X rate of 2,4-D choline (Table 5). No other fiber quality measurements were influenced by the 1/1,000X rate, regardless of timing (Tables 5 and 8). At the 1/500X rate, length, uniformity, and strength were negatively influenced at the FB timing. Similar to the 1/1,000X rate at FS + 2 wk application, the 1/100X rate increased micronaire at FS + 2 wk. When the 1/100X rate was applied at FB, length, uniformity, neps, and SCN were negatively impacted. The 1/50X rate at FS + 2 wk negatively influenced fiber length and SCN. At the FB application timing, neps and SCN were negatively influenced by the 1/50X rate of 2,4-D choline.

Rates above 1/10X more broadly affected fiber quality in 2020. Micronaire, length, uniformity, SCN, and maturity ratio were all negatively impacted when the 1/10X rate was applied at FS + 2 wk. At the FB timing, the 1/10X rate negatively influenced micronaire, uniformity, neps, SFC, and SCN. The 1/10X rate applied at FB + 2 wk resulted in uniformity and fiber strength increases, while negatively influencing SCN. A decrease in the total number of SFC was observed at the FB + 4 wk timing following the 1/10X rate of 2,4-D choline. Similar to 2019, the 1X rate of 2,4-D choline resulted in little to no lint production at the FS + 2 wk and FB application timings. At the FB + 2 wk application timing, micronaire, length, uniformity, neps, SFC, and SCN were all negatively influenced by the 1X rate. At FB + 4 wk, micronaire, neps, and SCN were negatively influenced.

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Table 7. Least-square means of fiber quality parameters measured using AFIS of 25 treatments (rate × timing) applied in 2019.[a,b,c]

Rate	Timing	Neps g^{-1}	Short fiber content	Seed coat neps	Maturity ratio
			n%	count g^{-1}	
NTC	—	307 a–c	28.9 ab	12.3 ab	0.858 a–d
1/1,000X	FS + 2 wk	293 ab	29.8 a–c	14.8 a–c	0.858 a–d
	FB	323 a–d	29.3 a–c	14.8 a–c	0.858 a–d
	FB + 2 wk	338 a–d	29.6 a–c	15.3 a–c	0.85 c–e
	FB + 4 wk	324 a–d	30.0 a–c	14.3 ab	0.863 a–c
1/500X	FS + 2 wk	309 a–c	29.4 a–c	15.5 a–c	0.863 a–c
	FB	292 ab	28.2 a	10.8 a	0.863 a–c
	FB + 2 wk	334 a–d	30.0 a–c	13.5 ab	0.853 b–d
	FB + 4 wk	306 a–c	29.5 a–c	13.3 ab	0.855 a–d
1/100X	FS + 2 wk	370 b–d	32.6 b–e	16.5 b–d	0.855 a–d
	FB	389 c–e	34.1 d–f	15.8 a–c	0.855 a–d
	FB + 2 wk	322 a–d	30.4 a–d	17.3 b–e	0.858 a–d
	FB + 4 wk	266 a	26.8 a	12.3 ab	0.868 a
1/50X	FS + 2 wk	408 de	36.6 fg	22.0 de	0.845 de
	FB	475 e	38.9 g	22.5 e	0.838 e
	FB + 2 wk	377 b–d	33.1 c–f	17.3 b–e	0.853 b–d
	FB + 4 wk	301 a–c	28.2 a	15.3 a–c	0.865 ab
1/10X	FS + 2 wk	875 g	52.8 i	45.3 h	0.788 g
	FB	667 f	45.2 h	37.5 g	0.808 f
	FB + 2 wk	402 cd	34.8 ef	20.0 c–e	0.848 de
	FB + 4 wk	343 a–d	30.0 a–c	15.5 a–c	0.858 a–d
1X	FS + 2 wk[d]	—	—	—	—
	FB[d]	—	—	—	—
	FB + 2 wk[d]	—	—	—	—
	FB + 4 wk	859 g	47.8 h	30 f	0.770 h

[a]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[b]Abbreviations: AFIS, Advanced Fiber Information System; FB, first bloom; FS, first square; NTC, nontreated control.
[c]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[d]Treatment not included in analysis of variance.

Table 8. Least-square means of fiber quality parameters measured using AFIS of 25 treatments (rate × timing) applied in 2020.[a,b,c]

Rate	Timing	Neps g^{-1}	Short fiber content	Seed coat neps	Maturity ratio
			n%	count g^{-1}	
NTC	—	340 a–d	31.6 b–f	11.2 a	0.886 a–d
1/1,000X	FS + 2 wk	323 a	31.7 b–f	14.3 ab	0.913 a
	FB	368 a–e	31.8 b–f	13.3 ab	0.880 cd
	FB + 2 wk	415 c–e	31.5 b–e	16.5 ab	0.903 a–c
	FB + 4 wk	376 a–e	32.2 b–f	17.8 a–c	0.888 a–d
1/500X	FS + 2 wk	342 a–d	30.4 a–c	13.3 ab	0.863 d
	FB	407 b–e	32.5 b–f	17.5 a–c	0.875 d
	FB + 2 wk	416 c–e	33.4 d–f	17.5 a–c	0.880 cd
	FB + 4 wk	375 a–e	31.5 b–e	15.5 ab	0.908 ab
1/100X	FS + 2 wk	360 a–e	30.2 ab	16.3 ab	0.873 d
	FB	435 ef	31.6 b–e	21.0 bc	0.862 d
	FB + 2 wk	394 a–e	31.1 a–e	17.3 a–c	0.855 de
	FB + 4 wk	328 ab	30.0 ab	17.5 a–c	0.885 b–d
1/50X	FS + 2 wk	423 d–f	32.6 b–f	22.8 bc	0.873 d
	FB	517 g	33.7 ef	22.8 bc	0.893 a–d
	FB + 2 wk	363 a–e	30.8 a–e	17.5 a–c	0.893 a–d
	FB + 4 wk	328 ab	29.8 ab	17.0 a–c	0.880 cd
1/10X	FS + 2 wk	555 c	34.8 f	30.8 de	0.817 e
	FB	658 h	39.3 g	25.2 c–e	0.870 d
	FB + 2 wk	426 d–f	30.7 a–d	23.3 b–d	0.875 d
	FB + 4 wk	337 a–c	28.3 a	19.0 a–c	0.903 a–c
1X	FS + 2 wk[d]	—	—	—	—
	FB[d]	—	—	—	—
	FB + 2 wk	807 i	39.4 g	36.2 e	0.898 a–d
	FB + 4 wk	503 fg	33.2 c–f	21.0 bc	0.863 d

[a]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[b]Abbreviations: AFIS, Advanced Fiber Information System; FB, first bloom; FS, first square; NTC, nontreated control.
[c]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[d]Treatment not included in analysis of variance.

Table 9. Least-square means of fiber quality parameters measured using AFIS of 25 treatments (rate × timing) applied in 2021.[a,b,c]

Rate	Timing	Neps g^{-1}	Short fiber content	Seed coat neps	Maturity ratio
			n%	count g^{-1}	
NTC	—	328 a	32.4 a–c	8.75 ab	0.863 bc
1/1,000X	FS + 2 wk	370 a–c	34.5 c–f	10.50 a–f	0.858 b–d
	FB	354 a–c	31.2 ab	10.30 a–e	0.860 b–d
	FB + 2 wk	332 ab	33.7 a–e	10.80 a–f	0.863 bc
	FB + 4 wk	309 a	31.7 a–c	8.50 a	0.863 bc
1/500X	FS + 2 wk	326 a	33.2 a–d	11.30 a–f	0.865 ab
	FB	345 a–c	33.9 b–f	9.25 a–c	0.858 b–d
	FB + 2 wk	337 ab	32.6 a–c	12.00 a–g	0.863 bc
	FB + 4 wk	329 ab	31.7 a–c	10.00 a–d	0.860 b–d
1/100X	FS + 2 wk	423 b–d	36.1 d–g	16.00 f–j	0.860 b–d
	FB	435 cd	36.8 fg	15.80 e–j	0.850 de
	FB + 2 wk	369 a–c	34.6 c–f	12.00 a–g	0.853 c–e
	FB + 4 wk	309 a	31.9 a–c	10.30 a–e	0.860 b–d
1/50X	FS + 2 wk	530 ef	36.7 e–g	22.30 kl	0.850 de
	FB	575 ef	42.9 h	17.50 g–k	0.823 f
	FB + 2 wk	355 a–c	33.5 a–d	14.50 c–h	0.853 c–e
	FB + 4 wk	282 a	30.8 a	10.30 a–e	0.868 ab
1/10X	FS + 2 wk	623 f	37.9 g	24.80 l	0.843 e
	FB	866 h	51.1 i	26.00 l	0.788 h
	FB + 2 wk	371 a–c	32.6 a–c	15.00 d–i	0.860 b–d
	FB + 4 wk	313 a	31.1 ab	14.30 b–h	0.875 a
1X	FS + 2 wk[d]	—	—	—	—
	FB	871 h	49.2 i	21.30 j–l	0.803 g
	FB + 2 wk	511 de	33.9 b–f	20.50 i–l	0.818 f
	FB + 4 wk	732 g	41.4 h	18.30 h–k	0.790 h

[a]The rate of 2,4-D choline is 1,060 g ae ha^{-1}.
[b]Abbreviations: AFIS, Advanced Fiber Information System; FB, first bloom; FS, first square; NTC, nontreated control.
[c]Means within the same column and followed by a common letter are not significantly different at the 0.05 level of significance.
[d]Treatment not included in analysis of variance.

Neither the HVI nor the AFIS measurements detected changes in fiber quality from the 1/1,000X or 1/500X rates of 2,4-D choline during the 2021 growing season, regardless of application timing (Tables 6 and 9). At the 1/100X rate of 2,4-D choline applied at FS + 2 wk, uniformity, strength, neps, SFC, and SCN were all negatively affected. At the FB timing, micronaire, neps, SFC, SCN, and maturity ratio were all negatively influenced by the 1/100X rate. When the 1/100X rate of 2,4-D choline was applied at FB + 2 wk and FB + 4 wk timings, no differences were observed. The 1/50X rate negatively influenced micronaire, strength, neps, SFC, SCN, and the maturity ratio at the FS + 2 wk application timing. Similar results were observed at the FB timing, when micronaire, neps, SFC, SCN, and maturity ratio were negatively influenced; however, uniformity also decreased at this timing. When the 1/50X rate of 2,4-D choline was applied at FB + 2 wk, both fiber length and SCN increased. No differences in fiber quality measurements were observed following the 1/50X rate applied at FB + 4 wk.

The 1/10X rate negatively influenced all fiber quality measurements, except fiber length, at the FS + 2 wk and FB application timings. When 2,4-D choline was applied at FB + 2 wk, fiber length increased, as did the number of SCN. At FB + 4 wk, the maturity ratio increased following the 1/10X rate. No other fiber quality differences were observed at the FB + 4 wk application timing at the 1/10X rate. Similar to 2019 and 2020, no lint sample was collected from the 1X rate at FS + 2 wk due to complete plant death. At the FB and FB + 4 wk timings, all reported fiber quality measurements were negatively influenced by the 1X rate of 2,4-D choline. When the 1X rate was applied at FB + 2 wk, no differences were observed in uniformity and fiber strength. All other fiber quality measurements were negatively influenced at the FB + 2 wk timing by the 1X rate of 2,4-D choline.

As reported in this trial and others, 2,4-D choline treatments have a significant impact on HVI fiber properties (Buol et al. 2019; Manuchehri et al. 2019). This can be detected at rates as low as 1/100X, depending on the year. Although the HVI is the primary marketing tool used by the U.S. Department of Agriculture Agricultural Marketing Service (USDA-AMS) and can be used to capture the potential impact of 2,4-D drift on the marketing parameter, it does not capture the full extent of the potential damage caused by 2,4-D choline drift. In addition to the standard HVI measurements, the USDA-AMS uses hand classification to make a seed coat fragment call. The presence of seed coat fragments can result in a discount because the fibrous portion of the contaminant makes it difficult to remove. Seed coat fragments in the yarn cause imperfections, resulting in discounts for the spinning mill. The AFIS SCN measurement indicates that 2,4-D may increase the occurrence of seed coat fragments, a problem that would not be captured with HVI testing alone.

The AFIS measurements, neps, SFC, and maturity ratio also are negatively affected by 2,4-D choline drift (Tables 7 to 9). These metrics reflect quality concerns that can affect the efficiency of sample processing at the mill and the quality of the spun yarn but are not included in USDA-AMS reports. Although these results reveal the potential need for fiber quality considerations beyond standard marketing parameters provided by HVI, all fiber quality parameters are affected by the type of processing, and the research samples used in this experiment were not subjected to industry-scale cleaning and ginning used in commercial production. Although the results reveal that off-target movement of 2,4-D choline has a negative impact on many fiber qualities, the level of impact is expected to be different in the types of samples classed by USDA-AMS.

Off-target movement of 2,4-D choline influences cotton boll production, lint yield, and fiber quality measurements. As cotton becomes more mature, it becomes less susceptible to lint reductions and changes to fiber quality. Changes to boll production and positioning have the potential to influence fiber quality. In general, off-target movement of 2,4-D choline negatively influences fiber quality, which can result in discounts to the producer, processing problems at the mill, and imperfections in the yarn. The impact on fiber quality depends on many factors, such as the year, timing of the application, and the rate of 2,4-D to which the plants are exposed. Although HVI fiber quality testing can reveal when there is a problem, additional testing, such as AFIS, is needed to reveal the full extent of the problem. More research is needed to better understand how 2,4-D choline interferes with fiber development, resulting in a degraded fiber quality. These data show that both timing and rate of 2,4-D choline are important factors when determining boll distribution and production, lint yield, and fiber quality following off-target movement of 2,4-D choline.

Practical Implications

Effective control of troublesome weeds, such as Palmer amaranth, remains a top priority for cotton producers. Enlist® cotton will allow the use of 2,4-D choline at-plant and postemergence to aid in the control of troublesome weeds, and on-target application must be a priority to avoid damage of sensitive plants in close proximity. Accurate information regarding plant response to off-target movement of 2,4-D should be a priority. Cotton plants at early growth stages are more sensitive to 2,4-D than more mature plants. Off-target movement of 2,4-D to susceptible cotton can negatively influence lint yield, micronaire, length, uniformity, strength, neps, SFC, seed coat fragments, and maturity ratio.

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