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Short title: Quizalofop and 2,4-D interaction

Interaction of quizalofop-p-ethyl with 2,4-D choline and/or glufosinate for control of volunteer corn in corn resistant to aryloxyphenoxypropionates

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Abstract

Corn resistant to aryloxyphenoxypropionates (FOPs) (Enlist[™] corn) enables the use of quizalofop-p-ethyl (QPE) as a selective postemergence (POST) herbicide for control of glufosinate/glyphosate-resistant corn volunteers. Growers usually mix QPE with 2,4-D choline and/or glufosinate to achieve broad-spectrum weed control in Enlist[™] corn. The objectives of this study were to (1) evaluate the efficacy of QPE applied alone or mixed with 2,4-D choline and/or glufosinate for control of glufosinate/glyphosate-resistant corn volunteers in Enlist[™] corn and (2) determine the impact of application time (V3 or V6 growth stage of volunteer corn) of QPE-based treatments on volunteer corn control as well as Enlist[™] corn injury and yield. Field experiments were conducted at South Central Agricultural Lab, Clay Center, NE in 2021 and 2022. Quizalofop-p-ethyl (46 or 93 g ai ha⁻¹) applied at V3 or V6 growth stage controlled volunteer corn $\ge 88\%$ and $\ge 95\%$ at 14 and 28 d after treatment (DAT), respectively. The QPE (46 g at ha^{-1}) mixed with 2,4-D choline (800 g at ha^{-1}) had 33% less expected control of V3 volunteer corn in 2021, and 8% less than expected control of V6 volunteer corn in 2022 at 14 DAT. Volunteer corn control was improved by 7%-9% using the higher rate of QPE (93 g ai ha^{-1}) in a mixture with 2,4-D choline (1,060 g ae ha^{-1}). The QPE mixed with glufosinate had an additive effect and interactions in any combinations were additive beyond 28 DAT. Mixing 2,4-D choline can reduce QPE efficacy on glufosinate/glyphosate-resistant corn volunteers up to 14 DAT when applied at the V3 or V6 growth stage; however, the antagonistic interaction did not translate into corn yield loss. Increasing the rate of QPE (93 g ai ha⁻¹) while mixing with 2,4-D choline can reduce antagonism.

Nomenclature: 2,4-D choline; glufosinate; quizalofop-P-ethyl; corn, Zea mays L.

Keywords: Antagonism; Enlist[™] corn; glufosinate/glyphosate-resistant; corn production system.

Introduction

Volunteer corn is a problem weed in corn-based cropping systems in the midwestern United States (Chahal and Jhala 2015; Jhala et al. 2021). Volunteer corn is an overwintering F_2 population of corn kernels/ears lost during the previous year or failed corn stands under a corn replanting scenario (Shauck and Smeda 2012). Although grain losses can be limited to < 5% with mechanical harvest (Shauck 2011), adverse weather such as the widespread freezing damage in spring 2007 in Tennessee (Steckel et al. 2009) or the widespread windstorm (Derecho) in August 2020 in Iowa (Jha et al. 2020) led to significant volunteer corn in the following growing season (Rees and Jhala 2018).

In Nebraska, on average, 4.0 million ha of corn is planted compared to 2.2 million ha of soybean, a difference of 1.8 million ha. (USDA-NASS 2017a, 2018, 2019, 2020). In addition, the area of corn planted in Nebraska in recent years increased by 0.26 million ha, while soybean area decreased by 0.20-0.28 million ha (USDA-NASS 2017a, 2018, 2019, 2020). This suggests that growers are shifting toward corn-on-corn cropping systems, especially in South Central Nebraska, due to the high-quality productive soil and irrigation (Striegel et al. 2020). Managing volunteer corn in a corn-corn rotation is challenging due to the lack of selective POST herbicides for adequate control (Jhala et al. 2021). However, the recent commercialization of a multiple herbicide-resistant corn hybrid (i.e., Enlist[™] corn) allows POST applications of guizalofop-p-ethyl (QPE) for control of glufosinate/glyphosateresistant corn volunteers. Enlist[™] corn is resistant to 2,4-D choline, glufosinate, glyphosate, and FOP herbicides (Striegel et al. 2020). As of October 2023, QPE (Assure® II; AMVAC, Newport Beach, CA 92660) is the only herbicide labeled for control of volunteer corn in Enlist[™] corn. QPE is labeled at 41 to 93 g ai ha⁻¹ in Enlist[™] corn for control of volunteer corn (Anonymous 2018). Striegel et al. (2020) reported 99% control of glufosinate/glyphosateresistant corn volunteers in EnlistTM corn in Nebraska with QPE (31 g at ha^{-1}) when applied to volunteer corn at the V3 or V6 growth stages.

Infestations of volunteer corn have increased progressively with the adoption of glyphosate-resistant corn (Davis et al. 2008), causing insect resistance (Krupke et al. 2009), disease survival (Chahal et al. 2016), grain contamination (Marquardt et al. 2012), and grain yield losses (Chahal and Jhala 2015; Chahal and Jhala 2016). Volunteer corn is as competitive as common midwestern weed species such as barnyardgrass [*Echinochloa crus-galli* (L.) P. Beauv.], giant foxtail (*Setaria faberi* Herrm.), and waterhemp [*Amaranthus*

tuberculatus (Moq.) J. D. Sauer] (Alms et al. 2016). Volunteer corn at 8 plants m⁻² can cause up to 23% grain yield loss in corn (Marquardt et al. 2012). Clumps of volunteer corn are more competitive than individual volunteer corn plants and usually cause greater yield losses, as Piasecki and Rizzardi (2019) reported that 0.5 to 12 clumps of volunteer corn m⁻² (7 plants clump⁻¹) can reduce corn yield by 7% to 42% compared to 3% to 34% yield loss with 0.5 to 12 individual volunteer corn plants m⁻² (Piasecki and Rizzardi 2019). Yield losses are greater under corn replant conditions; volunteer corn populations of 0.5 to 1, 2 to 4, and 4 to 8 plants m⁻² can reduce yields by 7% to 20%, 44% to 58%, and 59% to 81%, respectively (Shauck and Smeda 2014). Steckel et al. (2009) documented that 27,000 volunteer corn plants ha⁻¹ can reduce replanted corn yields up to 2,200 kg ha⁻¹.

Managing volunteer corn is a challenge due to the commercial cultivation of multiple herbicide-resistant corn hybrids. Earlier, non-selective herbicides such as glufosinate (Alms et al. 2016) and glyphosate (Andersen et al. 1982; Beckett and Stoller 1988) were applied to manage volunteer corn. Planning crop rotations around glufosinate and glyphosate-resistant hybrids was a viable solution for controlling volunteer corn until stacked glufosinate- and glyphosate-resistant corn was commercialized in 2012. Because of the widespread adoption of glufosinate/glyphosate-resistant corn (Soltani et al. 2014), glufosinate and glyphosate are not effective options for control of glufosinate/glyphosate-resistant corn volunteers. Although tillage, such as inter-row cultivation, is effective for controlling volunteer corn; growers have widely adopted conservation tillage (USDA-NASS 2017b). As a result, growers primarily rely on selective POST herbicides with active ingredients other than the herbicide-resistant traits present in the corn hybrids from the previous year (Steckel et al. 2009).

To save time, labor, and money, growers prefer mixing herbicides for POST applications to achieve broad-spectrum weed control. When herbicides are mixed, their interactions can be additive, antagonistic, or synergistic (Colby 1967; Zhang et al. 1995). However, when grass and broadleaf-killing herbicides are mixed, antagonism occurs more frequently (Damalas 2004; Zhang et al. 1995). Synthetic auxins such as 2,4-D or dicamba have been reported to antagonize the efficacy of graminicide/ACCase inhibitors for grass weed control (Blackshaw et al. 2006; Gomes et al. 2020; Lancaster et al. 2019; Minton et al. 1989; Mueller et al. 1989; Underwood et al. 2016). Similarly, glufosinate, a non-selective herbicide widely used as a foliar-applied broad-spectrum herbicide in glufosinate-resistant corn, may (Burke et al. 2005; Chahal and Jhala 2015; Gardner et al. 2006) or may not

antagonize grass control when mixed with graminicide/ACCase inhibitor (Duenk 2022; Eytcheson and Reynolds 2019).

Growers have complained about the reduced efficacy of QPE for controlling volunteer corn when mixed with 2,4-D choline. Information is lacking on the interaction of QPE with 2,4-D choline and/or glufosinate for control of glufosinate/glyphosate-resistant corn volunteers in EnlistTM corn when applied at different growth stages. The objectives of this study were (1) to evaluate the efficacy of QPE applied alone at different rates and in mixtures with 2,4-D choline and/or glufosinate for control of glufosinate/glyphosate-resistant corn volunteers, and (2) to evaluate the effect of time of application (the V3 or V6 growth stage of volunteer corn) on interaction of QPE with 2,4-D choline and/or glufosinate for volunteer glufosinate for volunteer glufosinate-resistant corn control, injury, and yield of EnlistTM corn.

Materials and Methods

Site Description. Field studies were conducted in 2021 and 2022 at University of Nebraska South Central Agricultural Laboratory, Clay Center, NE (40.57°N, 98.13°W). The experimental site had Hastings silt loam soil (montmorillonitic, mesic, Pachic Argiustolls) with 6.5 pH, 3.0% organic matter, 17% sand, 58% silt, and 25% clay. The field had been under corn-soybean rotation for more than six years and irrigated through a center-pivot irrigation system.

Field Experiments. Treatments were arranged in a split-plot design, with the growth stage of volunteer corn (V3 or V6) as the main factor and herbicides as the sub-plot factor. Herbicide treatments consisted of QPE, 2,4-D choline, and glufosinate applied alone or mixed at various rates and combinations (Table 1). Nontreated volunteer corn and weed-free plots were included for comparison. Nontreated volunteer corn plots had volunteer corn but no other weeds, while weed-free plots were free of volunteer corn as well as other weeds. A total of 16 herbicide treatments were evaluated and appropriate adjuvants were added following each herbicide label recommendation (Table 1). Treatments were replicated in three complete blocks. The size of an individual experimental unit was 27 m²; 3 m wide and 9 m long consisting of four corn rows spaced 0.76 m apart.

The experimental field was no-tilled in 2021 and roto-tilled before planting corn in 2022. To mimic volunteer corn, glufosinate/glyphosate-resistant corn seeds (Dekalb DKC60-87RIB) harvested from 2020 and 2021 were planted 4.5 cm deep in rows perpendicular to the crop rows and spaced at 0.76 m at 50,000 seeds ha⁻¹ on May 7, 2021 and June 22, 2022. Corn

planting was delayed in 2022 due to hail and windstorm events on June 7 that resulted in significant plant stand loss and damage; therefore, the study was re-planted. The Enlist[™] corn (Hoegemeyer 8097 SXETM) was planted 4.5 cm deep at 87,500 seeds ha⁻¹ on May 11, 2021, and June 22, 2022. Both volunteer corn and Enlist[™] corn hybrids had relative maturity of 110 days. For controlling broadleaf and grass weeds, atrazine/bicyclopyrone/mesotrione/Smetolachlor (Acuron[®]; Syngenta Crop Protection, LLC, Greensboro, NC) at 2.4 kg ai ha⁻¹ plus glyphosate (Roundup[®] PowerMAX; Bayer Crop Science, St. Louis, MO) at 1,260 g ae ha⁻¹ was applied preemergence (PRE) to the experimental field on May 13, 2021, while for the 2022 season, dimethenamid-P/saflufenacil (Verdict[®]; BASF Co., Research Triangle Park, NC) at 790 g ai ha⁻¹ plus atrazine (Aatrex[®] 4L; Syngenta Crop Protection, LLC, Greensboro, NC) at 1,134 g ai ha⁻¹ was applied on June 24, 2022. The weed-free control received an additional POST application of glyphosate at 868 g ae ha⁻¹ plus dicamba (DiFlexx[®]; Bayer Crop Science, St.Louis, MO) at 456 g ae ha⁻¹ plus acetochlor (Warrant[®]; Bayer Crop Science, St. Louis, MO) at 839 g ai ha⁻¹. Glyphosate at 868 g ae ha⁻¹ was applied POST to the experimental field on May 26 and June 9, 2021, for control of grass and broadleaf weeds. For the V3 growth stage of volunteer corn, the QPE, 2,4-D choline, and glufosinate-based treatments were applied on June 16, 2021 and July 12, 2022. For the V6 growth stage, these treatments were applied on June 24, 2021 and July 26, 2022. Herbicide treatments were applied using a CO₂-pressurized backpack sprayer fitted with five AIXR 110015 flat-fan nozzles (TeeJet Spraying Systems Co., Wheaton, IL) calibrated to apply 140 L ha⁻¹ of spray solution at 276 kPa.

Control of volunteer corn was visually assessed at 14, 28, and 56 d after treatment (DAT) using a scale of 0% to 100%, where 0% stands for no control and 100% stands for complete plant death. A similar scale of 0% to 100% was used to assess EnlistTM corn injury at 14 and 28 DAT. At 28 DAT, volunteer corn density was determined by counting plants in a 3 m length row. At 28 DAT, the aboveground shoot biomass of volunteer corn was collected by randomly placing two 0.5-m² quadrats across the middle two corn rows and hand-harvesting the volunteer corn plants from this area. Biomass was oven-dried at 70 C to a constant weight and then weighed. Grain yield of EnlistTM corn was recorded by harvesting the middle two rows of each plot with a small plot combine, then adjusting grain yield to 15.5% moisture content. Percent reduction (relative to nontreated volunteer corn control) in volunteer corn density and biomass was calculated using Equation 1 (Striegel et al. 2020):

$$Y = \left[\frac{c-B}{c}\right] \times 100$$
^[1]

where C denotes volunteer corn density or biomass from the nontreated volunteer corn control and B denotes volunteer corn density or biomass from the treated plots.

Statistical Analysis.

Data were analyzed using R software ver. 4.2.2 (R Core Team 2019). Interactions of herbicide, volunteer corn growth stage, and year were analyzed, and if they were found to be significant, data for each year were analyzed separately. For individual year models, volunteer corn growth stage-by-herbicide interaction was considered as a fixed effect, while replication and replication-by-volunteer corn growth stage were considered as random effects.

The ANOVA assumptions of normality and equal variances were checked with Shapiro-Wilk and Bartlett's test, respectively (Kniss and Streibig 2019) using the performance package (Lüdecke et al. 2022). The data for volunteer corn control, density, and biomass reduction, and EnlistTM corn injury were non-normal, whereas data for EnlistTM corn yield were normal with homogeneity of variance. The non-normal data were analyzed with generalized linear mixed models with beta error distribution (link = "logit") (Stroup 2015) using the glmmTMB package (Brooks et al. 2022). These models were checked for overdispersion using the DHARMa package (Hartig and Lohse 2022). Nontreated volunteer corn and weed-free controls were excluded due to a lack of variance among replicates (Sarangi and Jhala 2018). The data fulfilling ANOVA assumptions were analyzed with linear mixed-effects model using the *lme4* package (Bates et al. 2022). ANOVA table was calculated using the car package (Fox et al. 2022). After performing ANOVA, the estimated marginal means for treatments were calculated using emmeans (Lenth et al. 2022) and multcomp package (Hothorn et al. 2022). Treatment means were separated according to Tukey's method for p-value adjustments, and back-transformed for presentation for glmmTMB models.

To evaluate herbicide interactions, expected values for volunteer corn control or density/biomass reduction of herbicide mixtures were calculated using Colby's (1967) equations. Equations 2 and 3 were used to calculate expected values for two-way and three-way herbicide mixtures, respectively:

$$E = (X+Y) - \left(\frac{XY}{100}\right)$$
[2]

where E denotes expected volunteer corn control or density/biomass reduction for a two-way herbicide mixture (A+B), and X and Y denote observed volunteer corn control or density/biomass reduction with individual herbicide applications of A and B, respectively, and:

$$E = (X + Y + Z) - \left(\frac{XY + XZ + YZ}{100}\right) + \frac{XYZ}{10,0000}$$
[3]

where E denotes expected volunteer corn control or density/biomass reduction for a threeway herbicide mixture (A + B + C), and X, Y, and Z denote observed volunteer corn control or density/biomass reduction with individual herbicide application of A, B, and C, respectively (de Sanctis and Jhala 2021).

A two-tailed t-test was used to compare observed and expected treatment means of herbicide mixtures (de Sanctis and Jhala 2021). If observed control or density/biomass reduction was significantly more than expected, the interaction was considered synergistic; if observed control or density/biomass reduction was less than expected, the interaction was considered antagonistic; and if observed and expected treatment means had no statistical difference, the interaction was considered additive (Colby 1967).

Results and Discussion

Field Experiment

Volunteer Corn Control

Volunteer corn growth stage-by-herbicide interactions were observed for control assessments in both years; therefore, interaction means are reported separately for 2021 (Table 2) and 2022 (Table 3). In 2021, QPE applied at 46 and 93 g ai ha⁻¹ to V3 volunteer corn provided 88% and 97% control 14 DAT, respectively (Table 2). These results are consistent with Chahal and Jhala (2015), who previously reported 95% control of glyphosate-resistant corn volunteers with QPE at 40 g ai ha⁻¹ 15 DAT. As expected, glufosinate and 2,4-D choline did not provide any control, as volunteer corn was glufosinate-resistant and 2,4-D choline is selective for broadleaf weeds. Control ratings from 3% to 22% were assigned to glufosinate and 2,4-D choline, primarily due to lodging and volunteer corn damage that occurred due to an early-season windstorm in 2021. Compared to QPE alone, the mixture of QPE (46 or 93 g

ai ha⁻¹) with 2,4-D choline (800 or 1,060 g ae ha⁻¹) applied to the V3 growth stage provided 48% to 57% control of volunteer corn (Figure 1). The Colby's analysis further indicated antagonism in a mixture of QPE and 2,4-D choline because the observed control was significantly less (33% to 41% reduction) than the expected control (81% to 98%). Underwood et al. (2016) reported a 20% reduction in volunteer corn control at 28 DAT when QPE (24 g ai ha⁻¹) was mixed with dicamba (600 g ae ha⁻¹). The QPE in a mixture with glufosinate had an additive effect (96%-97%). Similarly, Duenk (2022) reported additive interaction of QPE (24 g ai ha⁻¹) and glufosinate (500 g ai ha⁻¹) with 95% to 98% control of glufosinate/glyphosate-resistant corn volunteers.

The three-way mixtures had the lowest control; mixing the lower rate of QPE (46 g ai ha^{-1}) with both the lower (880 + 656 g ha^{-1}) and higher rates (1.060 + 880 g ha^{-1}) of 2.4-D choline + glufosinate had 7% and 12% control of volunteer corn, which was 74% and 69% less than expected, respectively, based on Colby's analysis. Duenk (2022) also reported a 77% reduction in control of glufosinate/glyphosate-resistant corn volunteers with a mixture of QPE (24 g ai ha⁻¹) and 2,4-D choline (817 g ae ha⁻¹) compared to QPE applied alone (89%). Researchers have reported that reduced efficacy of graminicide herbicide mixed with broadleaf herbicides may be improved by increasing the rate of graminicide application. Underwood et al. (2016) noted a 22% increase (90% vs 68%) in control of volunteer corn when dicamba at 600 g ae ha⁻¹ was mixed with quizalofop at 36 vs 24 g ai ha⁻¹. In this current study, the QPE at 93 g ai ha⁻¹ increased efficacy of the three-way mixtures for volunteer corn control from 7%-12% to 61%-79%. Similarly, at 28 DAT, the higher rate of QPE in mixtures increased control up to 98%. Thus, higher rates of QPE can be used to improve volunteer corn control and overcome antagonism when used in mixtures with broadleaf herbicides. At 56 DAT, all the herbicide interactions were additive for both the V3 and V6 stage of volunteer corn.

In 2022, the QPE controlled 95% to 98% of volunteer corn at 14 DAT irrespective of application time and rate (Table 3). This was consistent with Striegel et al. (2020), who reported 98% control of glufosinate/glyphosate-resistant corn volunteers with QPE at 31 g ai ha⁻¹. The interaction of QPE with 2,4-D choline or glufosinate was additive for both the V3 and the V6 growth stage. Among the three-way mixtures, the QPE at 93 g ai ha⁻¹ applied to V6 volunteer corn in a mixture with the higher rates of 2,4-D choline and glufosinate did not improve control (77%) at 28 DAT compared with the lower rate of QPE (82%). This indicates that increasing the rate of graminicide may not be effective in improving grass

control if the rates of broadleaf herbicides are also increased. In the current study, the rate of 2,4-D choline was not constant; therefore, future experiments should mix varying rates of quizalofop with a fixed rate of 2,4-D choline to reveal the actual contribution of the increased rate of quizalofop for eliminating or minimizing antagonism. At 56 DAT, control was similar among all herbicide interactions when applied to V6 volunteer corn.

Volunteer Corn Density and Biomass Reduction

Interaction means were presented for volunteer corn density and biomass reduction 28 DAT because volunteer corn growth stage-by-herbicide interaction was significant, except for biomass reduction in 2022 (Table 4 and 5). In 2021, the QPE applied to V3 volunteer corn reduced volunteer corn density and biomass by 99% compared to the nontreated volunteer corn control (5 plants m^{-2} and 90 g m^{-2}) (Table 4). Several researchers have previously reported similar results with \geq 90% reduction in volunteer corn density and biomass with QPE at 24-36 g ai ha^{-1} (Duenk 2020; Soltani et al. 2006; Underwood et al. 2016). The observed density (71%) and biomass reduction (68%) were 28% and 31% lower than expected (99%) with a mixture of QPE (46 g ai ha^{-1}) and 2,4-D choline (800 g ha^{-1}). Similarly, Duenk (2020) documented 30% and 58% less than expected reductions in volunteer corn density and biomass with a mixture of QPE with 2,4-D choline (24 + 817 g ha⁻¹), respectively. Among the three-way mixtures, the QPE at 46 g ai ha⁻¹ in a mixture with 2,4-D choline (1,060 g ha⁻¹) and glufosinate (880 g ha⁻¹) had 13% to 49% less reduction in density (50%-73%) and biomass (53%-86%) of V3 or V6 volunteer corn than QPE alone (99%) in both 2021 and 2022 (Table 4 and 5). However, increasing the QPE rate to 93 g ai ha^{-1} improved density and biomass reduction by 6% to 45%, providing the expected reduction of 99% in some cases. Underwood et al. (2016) reported similar results where the higher rate of QPE (30 g ai ha^{-1}) mixed with dicamba (300 g ai ha^{-1}) had greater reduction in volunteer corn density (2 vs 6 plants m^{-2}) and biomass (21 vs 72 g m^{-2}) compared with the lower rate of QPE (24 g ai ha⁻¹). Thus, increasing the rate of QPE when mixed with 2,4-D choline and glufosinate may minimize or avoid antagonism by providing an expected reduction in volunteer corn density and biomass.

Enlist[™] Corn Injury

Little to no injury on $\text{Enlist}^{\text{TM}}$ corn was observed in 2022 (data not shown), though some injury was observed in treatments applied at the V6 growth stage of volunteer corn in 2021 (Table 4; Figure 2). At 28 DAT, the lowest corn injury of 3% was observed with glufosinate

at 656 g ai ha⁻¹ and the highest injury of 27% was observed with glufosinate at 880 g ai ha⁻¹ mixed with QPE at 46 g ai ha⁻¹ and 2,4-D choline at 1,060 g ae ha⁻¹. In additional glufosinate-based treatments, 13% to 18% injury was observed on EnlistTM corn. Glufosinate injury likely occurred due to the late-season application at the V6 growth stage in 2021. Glufosinate is recommended up to the V6 growth stage of glufosinate-resistant corn (Anonymous 2019), but in 2021, the EnlistTM corn was about at the V8 growth stage at the time of the V6 stage of volunteer corn application. In addition, relative humidity was high (86%) at the time of application, which has been found to increase glufosinate translocation that may result in injury (Anderson et al. 1993; Coetzer et al. 2001; Ramsey et al. 2002).

Corn Yield

Volunteer corn growth stage (V3 or V6)-by-herbicide interaction was significant for Enlist[™] corn yield in 2021; therefore, interaction means are presented in Table 6, while yield data for 2022 was combined for volunteer corn growth stages. In 2021, corn yield was similar across treatments when herbicides were applied to volunteer corn at the V3 growth stage. The nontreated volunteer corn control had 13,620 kg ha⁻¹ grain yield, while grain yield in the weedfree control was 15,060 kg ha⁻¹. The 2,4-D choline and glufosinate alone treatments had similar grain yield of 13,740 to 14,270 kg ha⁻¹, probably because volunteer corn plants harvested alongside Enlist[™] corn from the space between two middle rows of the plot may have added some additional yield to these treatments. For the V6 stage of volunteer corn, all treatments had similar yields, except those containing glufosinate. Enlist[™] corn injury due to glufosinate appears to be the most probable cause for yield loss in glufosinate-containing treatments (Table 4 and 6). Similarly, injury in the weed-free control plots (Table 4) led to similar yield (12,180 kg ha⁻¹) as the glufosinate-containing treatments (9,890 to 12,910 kg ha⁻¹; Table 6). In 2022, all treatments had similar corn yield as the weed-free control (11,680 kg ha⁻¹). The reported antagonism of mixing 2,4-D choline with QPE might not be reflected in the yield because antagonism did not occur for longer than 14 or 28 DAT, and eventually volunteer corn was completely killed.

Practical Implications

The only labeled herbicide for selective control of glufosinate and/or glyphosateresistant corn volunteers in EnlistTM corn is QPE (Striegel et al. 2020). Results of this study and previous findings suggest that QPE at 24 to 46 g ai ha⁻¹ can provide \geq 94% control of glufosinate/glyphosate-resistant corn volunteers 28 DAT (Chahal and Jhala 2015; Duenk 2020; Streigel et al. 2020). It was observed in this study that 2,4-D choline can antagonize volunteer corn control for at least the first two weeks when applied at the V3 or V6 growth stage of volunteer corn. Therefore, caution should be taken while applying QPE + 2,4-D choline. Results of this study indicate that increasing the rate of QPE (higher vs. lower labeled rate; 93 vs. 46 g ai ha⁻¹) can overcome antagonism caused by 2,4-D choline. If volunteer corn control is unacceptable, a second application of QPE can be made more than 7 days after the first application (Anonymous 2018). It must be noted that a maximum of 93 g ai ha⁻¹ QPE can be applied per year as a single- or two-split applications when EnlistTM corn is at the V2 to V6 growth stage (Anonymous 2018). For the best control of volunteer corn, QPE should be applied alone or sequentially with broadleaf herbicides (Anonymous 2018; Gomes et al. 2020; Underwood et al. 2016). Mixing glufosinate with QPE resulted in an additive effect for control of glufosinate/glyphosate-resistant corn volunteers regardless of application time (V3 or V6 volunteer corn growth stage). However, glufosinate should be applied before the V6 growth stage in glufosinate-resistant corn, or else drop nozzles should be used when corn is up to 86.4 cm (36 inches) to avoid corn injury (Anonymous 2019; Figure 2).

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Zhang J, Hamill AS, Weaver SE (1995) Antagonism and synergism between herbicides: trends from previous studies. Weed Technol 9:86–90 **Table 1:** Herbicide treatments, rates, products, and adjuvants used for control of glufosinate/glyphosate-resistant corn volunteers in EnlistTM corn in field experiments conducted at Clay Center, NE, in 2021 and 2022.

Herbicide ^a	Rate ^b	Trade name	Manufacturer ^c	Adjuvants ^b
	g ai or ae ha ⁻¹ -			
Nontreated volunteer corn control	868 fb 868	Roundup [®] PowerMAX fb	Bayer Crop Science	AMS 3% v/v
		Roundup [®] PowerMAX		
Weed-free control	456 + 868 + 839	DiFlexx [®] + Roundup [®]	Bayer Crop Science	NIS 0.25% v/v
		PowerMAX + Warrant®		
Quizalofop-p-ethyl	46	Assure [®] II	AMVAC	COC 1% v/v
Quizalofop-p-ethyl	93	Assure [®] II	AMVAC	COC 1% v/v
2,4-D choline	800	Enlist One [®]	Corteva	
2,4-D choline	1,060	Enlist One [®]	Corteva	
Quizalofop-p-ethyl + 2,4-D choline	46 + 800	Assure [®] II + Enlist One [®]	DuPont, Corteva	COC 1% v/v
Quizalofop-p-ethyl + 2,4-D choline	93 + 1,060	Assure [®] II + Enlist One [®]	DuPont, Corteva	COC 1% v/v
Glufosinate	656	Liberty [®] 280 SL	BASF	AMS 3% v/v
Glufosinate	880	Liberty [®] 280 SL	BASF	AMS 3% v/v
Quizalofop-p-ethyl + glufosinate	46 + 656	Assure [®] II + Liberty [®] 280	AMVAC, BASF	COC 1% v/v + AMS 3% v/v
		SL		
Quizalofop-p-ethyl + glufosinate	93 + 880	Assure [®] II + Liberty [®] 280	AMVAC, BASF	COC 1% v/v + AMS 3% v/v
		SL		

Quizalofop-p-ethyl	+	2,4-D	choline	+	46 + 800 + 656	Assure [®] II + Enlist $One^{\mathbb{R}}$ +	AMVAC, Cortev	a, COC 1% v/v + AMS 3% v/v
glufosinate						Liberty [®] 280 SL	BASF	
Quizalofop-p-ethyl	+	2,4-D	choline	+	93 + 800 + 656	Assure [®] II + Enlist One [®] +	AMVAC, Cortev	a, COC 1% v/v + AMS 3% v/v
glufosinate						Liberty [®] 280 SL	BASF	
Quizalofop-p-ethyl	+	2,4-D	choline	+	46 + 1,060 +	Assure [®] II + Enlist $One^{®}$ +	AMVAC, Cortev	a, COC 1% v/v+ AMS 3% v/v
glufosinate					880	Liberty [®] 280 SL	BASF	
Quizalofop-p-ethyl	+	2,4-D	choline	+	93 + 1,060 +	Assure [®] II + Enlist $One^{®}$ +	AMVAC, Cortev	a, COC 1% v/v + AMS 3% v/v
glufosinate					880	Liberty [®] 280 SL	BASF	

^aAtrazine/bicyclopyrone/mesotrione/*S*-metolachlor (Acuron[®]; Syngenta Crop Protection, LLC, Greensboro, NC) at 2.4 kg ai ha⁻¹ plus glyphosate (Roundup[®] PowerMAX; Monsanto Co., St. Louis, MO) at 1,260 g ae ha⁻¹ was applied PRE to the entire experimental field on 13 May 2021, while for the 2022 season, dimethenamid-P/saflufenacil (Verdict[®]; BASF Co., Research Triangle Park, NC) at 790 g ai ha⁻¹ plus atrazine (Aatrex[®] 4L; Syngenta Crop Protection, LLC, Greensboro, NC) at 1,134 g ai ha⁻¹ was applied on 24 June 2022.

^bAbbreviations: ai, active ingredient; ae, acid equivalent; AMS, Ammonium sulfate (N-Pak[®] AMS Liquid; Winfield United, LLC, St. Paul, MN); COC, Crop oil concentrate (Agri-Dex[®]; Helena Chemical Co., Collierville, TN).

^cAMVAC, Newport Beach, CA 92660; Bayer Crop Science, St. Louis, MO 63167; Corteva AgriScience LLC, Indianapolis, IN 46268; BASF Corporation, Research Triangle Park, NC 27709.

Table 2: Control of glufosinate/glyphosate-resistant corn volunteers with quizalofop-p-ethyl, 2, 4-D choline, and glufosinate interaction treatments applied at the V3 and V6 volunteer corn stage in $\text{Enlist}^{\text{TM}}$ Corn at Clay Center, NE in 2021.

Herbicide	Rate	Volu	unteer	corn co	ntrol ^a														
		14 E	DAT					28 E	DAT					56 D	DAT				
		V3 s	stage		V6 s	stage		V3 s	stage		V6 s	stage		V3 s	stage		V6	stag	e
		Obs	erved	Expe	Obs	erv	Ex	Obs	erv	Ex	Obs	erv	Ex	Obse	erv	Ex	Obs	ser	Exp
				cted ^b	ed		pec	ed		pec	ed		pec	ed		pec	ved		ecte
							ted			ted			ted			ted			d ^b
							b			b			b			b			
	g ai or ae ha ⁻¹									9	<i>б</i>								
Nontreated volunteer corn control	-	0		-	0		-	0		-	0		-	0		-	0		-
Weed-free control	-	99		-	99		-	99		-	99		-	99		-	99		-
Quizalofop-p-ethyl	46	88	ab	-	96	а	-	99	а	-	99	a	-	99	a	-	99	a	-
Quizalofop-p-ethyl	93	97	a	-	96	а	-	99	а	-	99	a	-	99	a	-	99	a	-
2,4-D choline	800	3	e	-	3	e	-	18	d	-	12	d	-	16	d	-	18	d	-
2,4-D choline	1,060	3	e	-	3	e	-	19	d	-	10	d	-	18	d	-	17	d	-
Quizalofop-p-ethyl + 2,4-D choline	46 + 800		d	81* ^c	96	а	97	91	b	99	99	a	99	98	a	99	99	a	99
		48													b				
Quizalofop-p-ethyl + 2,4-D choline	93 + 1,060		cd	98*	96	a	97	98	a	99	99	a	99	98	a	99	99	a	99
		57							b						b				

Glufosinate	656	4	e	-	3	e	-	22	d	-	12	d	-	16	d	-	18	d	-
Glufosinate	880	3	e	-	3	e	-	12	d	-	12	d	-	12	d	-	15	d	-
Quizalofop-p-ethyl + glufosinate	46 + 656	96	a	81	96	a	97	99	a	99	99	a	99	99	a	99	99	a	99
Quizalofop-p-ethyl + glufosinate	93 + 880	97	a	98	97	a	97	99	a	99	99	a	99	99	a	99	99	a	99
Quizalofop-p-ethyl + 2,4-D choline	46 + 800 + 656		e	81*	96	a	97	76	c	96*	98	a	98	93	b	97	99	a	96
+ glufosinate		7										b			c				
Quizalofop-p-ethyl + 2,4-D choline	93 + 800 + 656		abc	98	97	a	97	96	a	96	99	a	98	98	a	97	99	a	96
+ glufosinate		79							b						b				
Quizalofop-p-ethyl + 2,4-D choline	46 + 1,060 + 880		e	81*	97	a	97	79	c	97*	99	a	98	92	c	97	99	a	97
+ glufosinate		12																	
Quizalofop-p-ethyl + 2,4-D choline	93 + 1,060 + 880		bcd	98*	97	a	97	95	a	97	99	a	98	98	a	97	99	a	97
+ glufosinate		61							b						b				
P-value		< 0.0	001	•				< 0.0	001		•			0.00	2				

^aTreatment means with the same letters within the column are statistically similar according to Tukey's method for p-value adjustments and

Sidak confidence-level adjustments.

^bExpected values for herbicide mixtures were calculated using Colby's (1967) equations.

^cAsterisks(*) indicate that observed and expected values are significantly different as per t-test (P < 0.05), suggesting antagonistic interactions.

Table 3: Control of glufosinate/glyphosate-resistant corn volunteers with quizalofop-p-ethyl, 2, 4-D choline, and glufosinate interaction treatments applied at the V3 and V6 volunteer corn stage in $\text{Enlist}^{\text{TM}}$ Corn at Clay Center, NE in 2022.

Herbicide	Rate	Volu	inteer	corn c	contro	ol ^a													
		14 E	DAT					28 D	DAT					56 E	DAT				
		V3 s	stage		V6	stage		V3 s	stage		V6 s	tage		V3 s	stage		V6 s	stage	
		Obs	erve	Ex	Ob	serv	Ex	Obse	erv	Ex	Obse	erv	Ex	Obse	erv	Ex	Obse	erv	Exp
		d		pec	ed		pec	ed		pec	ed		pec	ed		pec	ed		ecte
				ted			ted			ted			ted			ted			d ^b
				b			b			b			b			b			
	g ai or ae ha ⁻¹										%								
Nontreated volunteer corn control	-	0		-	0		-	0		-	0		-	0		-	0		-
Weed-free control	-	99		-	99		-	99		-	99		-	99		-	99		-
Quizalofop-p-ethyl	46	98	a	-	95	ab	-	99	a	-	99	a	-	99	a	-	99	a	-
						c													
Quizalofop-p-ethyl	93	98	a	-	98	a	-	99	a	-	99	a	-	99	a	-	99	a	-
2,4-D choline	800	2	e	-	2	e	-	1	e	-	1	e	-	1	c	-	1	c	-
2,4-D choline	1,060	2	e	-	2	e	-	1	e	-	1	e	-	1	c	-	1	c	-
Quizalofop-p-ethyl + 2,4-D	46 + 800	96	abc	99	88	ab	96	99	a	99	96	a	99	99	a	99	98	a	99
choline						c						b							
Quizalofop-p-ethyl + 2,4-D	93 + 1,060	96	abc	99	95	ab	99	98	a	99	99	a	99	98	a	99	99	a	99

choline						c													
Glufosinate	656	2	e	-	2	e	-	1	e	-	1	e	-	1	с	-	1	c	-
Glufosinate	880	2	e	-	3	e	-	1	e	-	1	e	-	1	с	-	1	c	-
Quizalofop-p-ethyl + glufosinate	46 + 656	98	a	99	97	ab	95	99	a	99	99	a	99	99	a	99	99	a	99
Quizalofop-p-ethyl + glufosinate	93 + 880	98	a	99	96	ab	99	99	a	99	99	a	99	99	a	99	99	a	99
						c													
Quizalofop-p-ethyl + 2,4-D	46 + 800 + 656	89	abc	99	84	bc	96	98	a	99	90	b	99	98	a	99	95	a	99
choline + glufosinate												c						b	
Quizalofop-p-ethyl + 2,4-D	93 + 800 + 656	91	abc	99	90	ab	99	97	a	99	97	a	99	98	a	99	99	a	99
choline + glufosinate						c			b			b							
Quizalofop-p-ethyl + 2,4-D	46 + 1,060 +	61	d	99	80	cd	96	76	d	99	82	c	99	89	b	99	97	a	99
choline + glufosinate	880			*c						*		d						b	
Quizalofop-p-ethyl + 2,4-D	93 + 1,060 +	97	ab	99	58	d	99	99	a	99	77	d	99	99	a	99	95	a	99
choline + glufosinate	880						*						*					b	
P-value		< 0.	001			1		< 0.0	001					< 0.0	001	1			1

^aTreatment means with the same letters within the column are statistically similar according to Tukey's method for p-value adjustments and Sidak confidence-level adjustments.

^bExpected values for herbicide mixtures were calculated using Colby's (1967) equations.

^cAsterisks(*) indicate that observed and expected values are significantly different as per t-test (P < 0.05), suggesting antagonistic interactions.

Table 4: Enlist^{$^{\text{TM}}$} corn injury, density, and biomass reduction of glufosinate/glyphosate-resistant corn volunteers 28 DAT with quizalofop-pethyl, 2, 4-D choline, and glufosinate interaction treatments applied at the V3 and V6 volunteer corn stage in Enlist^{$^{\text{TM}}$} Corn at Clay Center, NE in 2021.

Herbicide	Rate ^b	En	list™	۱ د	corn	Den	sity red	uction ^a				Bior	nass rec	luction ^a	l		
		inj	ury ^a														
		V3		V6		V3 s	stage		V6 s	stage		V3 s	tage		V6 s	tage	
		sta	ge	stag	e	Obse	erved	Expec	Obse	erv	Exp	Obse	erved	Exp	Obse	erve	Exp
								ted ^b	ed		ecte			ecte	d		ecte
											d ^b			d ^b			d ^b
	g ai or ae ha ⁻¹										%						
				-													
Nontreated volunteer corn control	-	0	c	0	c	0		-	0		-	0		-	0		-
Weed-free control	-	0	c	10	b	10		-	10		-	10		-	10		-
						0			0			0			0		
Quizalofop-p-ethyl	46	0	c	0	c	99	a	-	99	a	-	99	a	-	99	ab	-
Quizalofop-p-ethyl	93	0	c	0	c	99	a	-	99	a	-	99	a	-	99	ab	-
2,4-D choline	800	0	c	0	c	18	fg	-	22	fg	-	18	ef	-	16	def	-
2,4-D choline	1,060	0	c	0	с	26	fg	-	14	g	-	20	ef	-	15	def	-
Quizalofop-p-ethyl + 2,4-D	46 + 800	0	c	0	c	71	cde	99* ^c	99	а	99	68	bcde	99*	99	ab	99
choline																	
Quizalofop-p-ethyl + 2,4-D	93 + 1,060	0	c	0	c	97	ab	99	99	a	99	93	abc	99	99	ab	99

choline																	
Glufosinate	656	0	c	3	c	30	efg	-	18	fg	-	24	ef	-	7	f	-
Glufosinate	880	0	c	15	b	22	fg	-	21	fg	-	22	ef	-	14	def	-
Quizalofop-p-ethyl + glufosinate	46 + 656	0	c	16	b	99	a	99	99	а	99	99	a	99	99	ab	99
Quizalofop-p-ethyl + glufosinate	93 + 880	0	с	14	b	99	a	99	99	а	99	99	a	99	99	ab	99
Quizalofop-p-ethyl + 2,4-D	46 + 800 +	0	c	13	b	50	defg	94*	99	a	94	53	cdef	99*	97	abc	99
choline + glufosinate	656																
Quizalofop-p-ethyl + 2,4-D	93 + 800 +	0	c	17	b	93	abc	94	99	a	94	82	abc	93	99	ab	99
choline + glufosinate	656																
Quizalofop-p-ethyl + 2,4-D	46 + 1,060 +	0	c	27	a	55	def	95*	99	a	99	60	bcde	94**	99	ab	99
choline + glufosinate	880												f				
Quizalofop-p-ethyl + 2,4-D	93 + 1,060 +	0	c	18	b	74	bcd	95*	99	a	98	78	abcd	94	99	ab	99
choline + glufosinate	880																
P-value		< 0	0.001	1		< 0.0	001	1	1			< 0.0	001		1		

^aTreatment means with the same letters within the column are statistically similar according to Tukey's method for p-value adjustments and Sidak confidence-level adjustments.

^bExpected values for herbicide mixtures were calculated using Colby's (1967) equations.

^cAsterisks(*) indicate that observed and expected values are significantly different as per t-test (*P < 0.05, **P < 0.10), suggesting antagonistic interactions.

Table 5: Density and biomass reduction of glufosinate/glyphosate-resistant corn volunteers 28 DAT with quizalofop-p-ethyl, 2, 4-D choline, and glufosinate interaction treatments applied at the V3 and V6 volunteer corn stage in EnlistTM Corn at Clay Center, NE in 2022.

Herbicide	Rate ^b	Dens	sity rec	luction ^a				Bion	nass re	duction ^a
		V3 s	tage		V6 s	tage		V3 +	V6 st	age
		Obse	erved	Expected ^b	Obse	erved	Expected ^b	Obse	erved	Expected ^b
	g ai or ae ha ⁻¹						%			
Nontreated volunteer corn control	-	0		-	0		-	0		-
Weed-free control	-	100		-	100		-	100		-
Quizalofop-p-ethyl	46	99	a	-	99	a	-	99	a	-
Quizalofop-p-ethyl	93	99	a	-	99	a	-	99	a	-
2,4-D choline	800	3	d	-	3	d	-	14	b	-
2,4-D choline	1,060	3	d	-	6	d	-	14	b	-
Quizalofop-p-ethyl + 2,4-D choline	46 + 800	99	a	99	89	abc	99	94	a	99
Quizalofop-p-ethyl + 2,4-D choline	93 + 1,060	97	ab	99	99	a	99	99	a	99
Glufosinate	656	12	d	-	5	d	-	15	b	-
Glufosinate	880	6	d	-	5	d	-	9	b	-
Quizalofop-p-ethyl + glufosinate	46 + 656	99	a	99	99	a	99	99	a	99
Quizalofop-p-ethyl + glufosinate	93 + 880	99	a	99	99	a	99	99	a	99
Quizalofop-p-ethyl + 2,4-D choline +	46 + 800 + 656	97	ab	99	79	bc	99*	90	a	96

glufosinate														
Quizalofop-p-ethyl	+	2,4-D	choline	+	93 + 800 + 656	97	ab	99	97	ab	99	95	a	96
glufosinate														
Quizalofop-p-ethyl	+	2,4-D	choline	+	46 + 1,060 + 880	54	c	99* ^c	73	bc	99*	86	a	97*
glufosinate														
Quizalofop-p-ethyl	+	2,4-D	choline	+	93 + 1,060 + 880	99	a	99	55	с	99*	92	a	98
glufosinate														
P-value						< 0.0	01		•	•		< 0.0	01	

^aTreatment means with the same letters within the column are statistically similar according to Tukey's method for p-value adjustments and Sidak confidence-level adjustments.

^bExpected values for herbicide mixtures were calculated using Colby's (1967) equations.

^cAsterisks(*) indicate that observed and expected values are significantly different as per t-test (P < 0.05), suggesting antagonistic interactions.

Table 6: EnlistTM corn yield as influenced by quizalofop-p-ethyl, 2, 4-D choline, and glufosinate interaction treatments applied at the V3 and V6 volunteer corn stage in 2021 and 2022 at Clay Center, NE.

Herbicide	Rate	Corn yie	ld ^a				
		2021				2022	
		V3 stage		V6 stage		V3 + V6	5 stage
						b	
	g ai or ae ha ⁻¹			k§	g ha ⁻¹	I	
Nontreated volunteer corn control	-	13,620	abcdef	15,170	abc	10,520	ab
Weed-free control	-	15,060	abcde	12,180	cdef	11,680	ab
Quizalofop-p-ethyl	46	15,180	abcd	15,900	ab	11,650	ab
Quizalofop-p-ethyl	93	16,200	abc	15,670	ab	11,340	ab
2,4-D choline	800	13,740	abcdef	17,110	a	11,150	ab
2,4-D choline	1,060	14,270	abcde	14,820	abcd	11,240	ab
Quizalofop-p-ethyl + 2,4-D choline	46 + 800	15,150	abcd	16,340	a	10,510	ab
Quizalofop-p-ethyl + 2,4-D choline	93 + 1,060	16,310	abc	15,180	abc	12,350	a
Glufosinate	656	14,010	abcdef	16,010	ab	11,210	ab
Glufosinate	880	13,990	abcdef	11,590	def	9,160	b
Quizalofop-p-ethyl + glufosinate	46 + 656	15,090	abcde	12,910	bcdef	12,650	a
Quizalofop-p-ethyl + glufosinate	93 + 880	14,950	abcde	9,970	f	11,590	ab

Quizalofop-p-ethyl + 2,4-D choline + glufosinate	46 + 800 + 656	14,320	abcde	11,870	def	10,990	ab
Quizalofop-p-ethyl + 2,4-D choline + glufosinate	93 + 800 + 656	14,950	abcde	10,910	ef	11,690	ab
Quizalofop-p-ethyl + 2,4-D choline + glufosinate	46 + 1,060 + 880	14,680	abcde	9,890	f	11,080	ab
Quizalofop-p-ethyl + 2,4-D choline + glufosinate	93 + 1,060 + 880	14,510	abcde	9,970	f	12,840	a
P-value		< 0.001				0.0260	
V3 vs V6 (2021) ^b		14,751 v	s 13,467 (I	P-value = ().1196)		
V3 vs V6 (2022) ^b		11,720 v	s 10,985 (I	P-value = ().1408)		

^a Treatment means with the same letters within the column are statistically similar according to Tukey's method for p-value adjustments and Sidek confidence level adjustments

Sidak confidence-level adjustments.

^bThe estimated marginal means for the main effects of volunteer corn stage in 2021 and 2022.



Figure 1: Volunteer corn control 14 DAT: **a**) non-treated for volunteer corn, **b**) quizalofop at 93 g ai ha⁻¹ + 2,4-D choline at 1,060 g ae ha⁻¹, and **c**) quizalofop at 93 g ai ha⁻¹ applied at the V3 growth stage of volunteer corn.



Figure 2: Glufosinate injury symptoms in glufosinate-containing treatments applied when corn volunteers were at the V6 growth stage and $\text{Enlist}^{\text{TM}}$ corn was at the V8 growth stage (an off-label treatment, as glufosinate is labelled up to the V6 growth stage).