


Influence of hard water on 2,4-D formulations for the control of dandelion

Geoffrey P. Schortgen¹ and Aaron J. Patton² 

Research Article

Cite this article: Schortgen GP and Patton AJ (2021) Influence of hard water on 2,4-D formulations for the control of dandelion. *Weed Technol.* **35**: 371–379. doi: [10.1017/wet.2020.135](https://doi.org/10.1017/wet.2020.135)

Received: 28 September 2020
Revised: 18 November 2020
Accepted: 20 November 2020
First published online: 9 December 2020

Associate Editor:

Patrick E. McCullough, University of Georgia

Nomenclature:

2,4-D; dandelion; *Taraxacum officinale* G.H. Weber ex Wiggers; MCPP; MCPA

Keywords:

amine; antagonism; emulsifiable concentrate; emulsion; ester; hard water; water-soluble; water-insoluble

Author for correspondence:

A.J. Patton, Department of Horticulture and Landscape Architecture, Purdue University, 625 Agriculture Mall Drive, West Lafayette, IN 47907 Email: ajpatton@purdue.edu

¹Extension Educator, Agriculture & Natural Resources, Wabash County Extension, Purdue University, Wabash, IN, USA and ²Professor, Department of Horticulture and Landscape Architecture, Purdue University, West Lafayette, IN, USA

Abstract

The herbicide 2,4-D is used in a variety of cropping systems, especially in grasses because it is a selective postemergence broadleaf herbicide. However, the most common formulation (2,4-D dimethylamine) is antagonized when mixed in hard water. The objective of this research was to determine which formulations of 2,4-D or premixes of various formulations of synthetic auxin herbicides are subject to hard water antagonism. Formulations surveyed for hard water antagonism in the first experiment included 2,4-D dimethylamine, 2,4-D diethanolamine, 2,4-D monomethylamine, 2,4-D isopropylamine salt, 2,4-D choline salt, 2,4-D isooctyl ester, and 2,4-D ethylhexyl ester. Synthetic auxin formulation types in the second experiment included water-soluble, emulsifiable concentrates and emulsion-in-water. All formulations were mixed with both soft and hard water (600 mg CaCO₃ L⁻¹) and applied to dandelions to determine whether antagonism occurred in hard water. Water-soluble (amine and choline) 2,4-D formulations were antagonized by hard water, but water-insoluble (ester) 2,4-D formulations were not antagonized. Similar results were found by formulation type with water-soluble synthetic auxin premixes antagonized but emulsifiable concentrates not antagonized. Furthermore, water-soluble salt formulations were not antagonized when formulated in premixes with other synthetic auxin herbicides as an emulsion-in-water. This research demonstrates that all 2,4-D water-soluble formulations and water-soluble premixes with phenoxyacetic acid herbicides are subject to hard water antagonism. Formulations of 2,4-D containing emulsifying agents protect against antagonism by the water-insoluble nature of ingredients in their formulation.

Introduction

The herbicide 2,4-D has been formulated in many ways since its development in the 1940s (Peterson 1967). In the pure acid form, 2,4-D has low water solubility and does not readily go into solution (Peterson et al. 2016). As such, 2,4-D acid is commonly reacted with bases to form salts or with alcohols to form esters (Peterson et al. 2016). Several of the salts that were historically used shortly after 2,4-D was developed included sodium, potassium, ammonium, lithium, and several amine salts (Anonymous 2007; Ross and Lembi 2009). Currently, common 2,4-D formulations include water-soluble amine salts, a water-soluble choline salt, and emulsifiable concentrate esters (Jervais et al. 2008; Peterson et al. 2016).

The most common 2,4-D amine formulation manufactured today is the dimethylamine salt (Jervais et al. 2008; Peterson et al. 2016). Choline salts are a new, water-soluble, 2,4-D formulation that reduce volatilization and eye irritation (Eytcheson et al. 2012; Stagg et al. 2013). Water-soluble formulations prepared in water with pH equal to the pK_a will result in half of the 2,4-D amine molecules being dissociated (i.e., 2,4-D acid no longer bonded to the amine salt; Tan and Crabtree 1994). The pK_a of 2,4-D is 2.64, which is its dissociation constant (Bekbölet et al. 1999). Because spray water pH is higher than the pK_a of 2,4-D, typically between 7 to 8 (Hem 1985), a water-soluble formulation is dissociated (deprotonated) when in spray solution (Tan and Crabtree 1994). When 2,4-D is dissociated, it interacts with divalent cations in the spray solution such as Ca, Fe, Mg, Mn, and Zn (Devkota and Johnson 2016; Nalewaja et al. 1990, 1991; Patton et al. 2016; Roskamp et al. 2013; Schortgen and Patton 2020).

Recent papers describe the hard water antagonism that can occur from 2,4-D dimethylamine (Patton et al. 2016; Roskamp et al. 2013; Schortgen and Patton 2020), but those researchers did not evaluate other amine formulations. Past research by Nalewaja et al. (1990) demonstrated that 2,4-D diethanolamine is also antagonized by hard water. Later, Nalewaja et al. (1991) noted that 2,4-D antagonism of the diethanolamine formulation likely meant that other 2,4-D salt formulations and MCPA ([4-chloro-2-methylphenoxy]acetic acid) would also be antagonized. However, data on MCPA and other 2,4-D formulations was not published.

Commonly used low-volatile ester formulations with longer carbon chains include butoxyethyl (also called butoxyethanol) and 2-ethylhexyl (also called isooctyl; Thompson 1986). Esters, which are oil soluble, are more effective at entering the waxy plant cuticle, but have a

high potential for drift due to a low vapor pressure (Grover et al. 1972; Jervais et al. 2008; Peterson et al. 2016). Ester products of 2,4-D are formulated as emulsifiable concentrates and should not be influenced by hard water cations since emulsifiable concentrates do not go directly into solution, dissociate, or come into contact with the divalent cations present in hard water (Müller 2000). However, little published data exists describing the efficacy of 2,4-D ester formulations applied in hard water (Müller 2000; Nalewaja et al. 1991). Two reports described that “2,4-D ester” is not antagonized but no data on the ester formulation was presented and the ester formulation(s) tested were not named (Nalewaja et al. 1990, 1991).

A report by Devkota and Johnson (2016) based on their research on 2,4-D choline efficacy was the first on hard water antagonism of the choline formulation. Their research was with a water-soluble 2,4-D choline salt formulation (GF-2654, U.S. Environmental Protection Agency [EPA] registration number [reg. no.] 62719-634; Montague 2017) that was later improved for increased stability and performance in hard water based on Devkota and Johnson’s (2016) research. The GF-2654 formulation is different than Enlist™ One (2,4-D choline salt, GF-3335, EPA reg. no. 62719-695; Montague 2017), which is formulated to form an emulsion upon dilution (Li et al. 2013; Shao and Tank 2015). Additional research is needed on the influence of water hardness on commercially available formulations.

Several of the above-mentioned papers tested 2,4-D as a stand-alone ingredient but few experiments have tested potential hard water antagonism of commercially available premixes. Kelly (1953) and Peterson et al. (2016) noted that proprietary ingredients such as sequestering agents are often added to improve the efficacy of 2,4-D formulations. Despite this, Devkota and Johnson (2016) reported that a commercially available premix formulation of 2,4-D choline + glyphosate is antagonized by hard water.

Many commercial formulations of 2,4-D (Table 1) are currently available, but past research has focused on hard water antagonism of the 2,4-D dimethylamine formulation. Furthermore, information is scant on potential antagonism of formulations with emulsifiers or formulations of synthetic auxin premixes. The objective of this research was to determine which formulations of 2,4-D or premixes of various formulations of synthetic auxin herbicides are subject to hard water antagonism.

Materials and Methods

Water Hardness and Adjuvant Inclusion Impact on 2,4-D Formulations

Two experiments were conducted at Purdue University to address the experimental objective. Experiment 1 was conducted in the Purdue University Horticulture greenhouses, West Lafayette, IN (40.421°N, 68.914°W) to determine the influence of water hardness on the efficacy of various 2,4-D formulations. Dandelion was used to evaluate the effects of hard water on 2,4-D formulation. Dandelion seed was collected locally in Lafayette, IN. Dandelions were germinated in 16 × 12 × 6 cm flats (Hummert International, Earth City, MO 63045) containing potting soil (Fafard growing mix, Sun-Gro Horticulture, Agawam, MA 01001) in the greenhouse. After germination and emergence, a single seedling was transplanted into 3.8 cm-diameter cone-tainers (Ray Leach SC-10 Super Cell Cone-tainers, Stuewe & Sons, Tangent, OR 97389) containing a 1:2:1 mixture of potting soil, sand, and a Whitaker silt loam soil (fine-loamy, mixed, active, mesic Aeric Endoaqualf). After

transplanting, plants were irrigated with municipal tap water as needed and fertilized biweekly with two water-soluble fertilizers (3:1 mixture of 15 N–2.2 P–12.5 K and 21 N–2.2 P–16.6 K, respectively; The Scotts Co., Marysville, OH 43040) to provide the following (in mg L⁻¹): 200 N, 26 P, 163 K, 50 Ca, 20 Mg, 1.0 Fe, 0.5 Mn and Zn, 0.24 Cu and B, and 0.1 Mo. Seventy-six percent of the nitrogen provided was in the nitrate form.

Two experimental runs were conducted in the greenhouse between April 2016 and June 2016, each arranged as a randomized complete block design on the greenhouse benches with four blocks in each run. Temperature and photosynthetically active radiation (PAR) from herbicide application to the conclusion of the experiment were collected with a mini-weather station (WatchDog 2475 Plant Growth Station, Spectrum Technologies, Plainfield, IL 60585). Temperatures for experimental run 1 averaged 24.4 C with an average daily light integral of 20 mol m⁻² d⁻¹. Temperatures for experimental run 2 averaged 25.3 C with an average daily light integral of 38 mol m⁻² d⁻¹.

The experiment was conducted to evaluate the effect of solution hardness on the performance of 2,4-D formulations. Formulations included a 2,4-D dimethylamine (DMA) with and without proprietary ingredients (DMA w/o prop). Other water-soluble formulations (amines) included 2,4-D diethanolamine (DEA), 2,4-D isopropylamine (MIPA), and 2,4-D dimethylamine salt plus monomethylamine (MMA) salt, and a choline salt formulation (Table 1). Water-insoluble or emulsifiable concentrate formulations of 2,4-D included 2,4-D 2-ethylhexyl ester (2-EHE) and 2,4-D butoxyethyl ester (BEE; Table 1). The same acid equivalent of each formulation was added to mixtures to achieve an herbicide rate of 1.60 kg ae ha⁻¹ (1.42 lb ae ha⁻¹, the maximum single application rate to ornamental turf) in both soft and hard water. Soft water with a hardness levels of 0 mg L⁻¹ was achieved by using distilled water and hard water was created with CaCl₂ 2[H₂O] L⁻¹ to achieve a hardness level of 600 mg CaCO₃ L⁻¹ following the methods described by Schortgen and Patton (2020). Calcium chloride was used to mimic a naturally occurring hard water from calcium carbonate based on a report by Nalewaja et al. (1991) since 1) carbonates and chloride salts are similarly antagonistic, 2) calcium chloride is antagonistic to 2,4-D, and 3) artificially created hard water from calcium chloride is similarly antagonistic to naturally occurring hard water from wells. Water hardness is reported here in milligrams per liter of equivalent calcium carbonate per standard convention (Boyd 2015). “The total hardness of water results from divalent cations—mainly from calcium and magnesium—expressed as equivalent calcium carbonate. The total hardness equivalence of 1 mg/L calcium is 2.5 mg/L, while 1 mg/L magnesium equates to 4.12 mg/L” (Boyd 2015, p.179).

Treatments were applied to 5- to 12-cm-diameter dandelion rosettes. Applications were made on April 4, 2016 in experimental run 1 and May 11, 2016 in experimental run 2. Calcium solutions were prepared prior to adding the herbicide. After the herbicide was added, treatments were mixed thoroughly and applied immediately afterward. The pH of each treatment was determined using a meter (VWR B40PCID symphony benchtop multi parameter meter, VWR International, Inc., Radnor, PA 19087) at each step of mixing to ascertain the influence of formulation on spray carrier pH. Treatments were applied using compressed air in a track spray chamber (Generation III Research Sprayer, DeVries Manufacturing, Hollandale, MN 56045) calibrated to deliver 815 L ha⁻¹, a common spray volume used in the lawn care industry (Christians et al. 2017), using a TeeJet® 8004EVS nozzle (TeeJet Technologies, Spraying Systems

Table 1. 2,4-D formulations or formulation combinations tested for efficacy in hard water in Experiment 1.

Common name (abbreviation)	Trade name and manufacturer	CAS No. ^a	Solubility ^b	Notes on use
			mg L ⁻¹ at 25 C, pH = 7	
2,4-D dimethylamine salt (DMA)	Weedar® 64, Nufarm Americas Inc., Alsip, IL 60803	2008-39-1	729,397	Most commonly used amine formulation
2,4-D dimethylamine salt (without proprietary ingredients) (DMA w/o prop)	N/A, Nufarm Americas Inc.	2008-39-1	729,397	Partially formulated without proprietary ingredients such as adjuvants
2,4-D diethanolamine salt (DEA)	N/A, PBI-Gordon Corp., Kansas City, MO 64101	5742-19-8	806,000	Often formulated in mixture with 2,4-D DMA
DMA + 2,4-D monomethylamine salt (MMA)	Amicide® Advanced 700, Nufarm Australia Ltd., Laverton North Victoria 3026	2008-39-1+ 51173-63-8	729,397	Mixture registered for use in Australia
2,4-D isopropylamine salt (MIPA)	N/A, Nufarm Americas Inc.	5742-17-6	657,000	Also called monoisopropylamine and often formulated in mixture with glyphosate
2,4-D choline salt (Choline)	GF-2654, Dow AgroSciences LLC, Indianapolis, IN 46268	1048373-72-3	Unknown	A different choline formulation than tested here is an ingredient in Enlist Duo (along with glyphosate) herbicide with Colex-D® technology
2,4-D 2-ethylhexyl ester (2-EHE)	Weedone® LV4, Nufarm Americas Inc.	25168-26-7	1.0*	Also known as 2,4-D isooctyl ester
2,4-D butoxyethyl ester (BEE)	Shredder™ E-99, Winfield Solutions LLC, St. Paul, MN 55164	1929-73-3	1.0*	Also called butoxyethanol ester

^aChemical Abstract Services number.

^bSolubility data from Jervais et al. 2008. Those solubility values followed by asterisk (*) are formulated as emulsifiable concentrates.

Co., Wheaton, IL 60187) at 275 kPa. Plants were not irrigated for 24 h after application.

Weed epinasty was assessed 1 and 2 wk after application (WAA) on a 0% to 100% scale, where 0% was no epinasty and 100% represented complete epinasty with all leaves exhibiting symptoms including twisting or bending of stems and curling of leaves. Visual estimates of percent weed control were recorded at 3 and 4 WAA on a scale of 0 (no control) to 100 (complete plant death). Digital images were taken of the weeds 4 WAA using a camera and light box and analyzed for percent green coverage using ImageJ (v. 1.48v, National Institutes of Health, Bethesda, MD 20892; Schneider et al. 2012) as described by Patton et al. (2018). At 4 WAA, plants were destructively harvested by washing the roots free of soil and dissecting into an aboveground shoot tissue and belowground root tissue fraction. Next, leaf fresh weight was measured and then tissues were placed into a forced-air dryer at 60 C for 3 d before dry weights were measured. All measurements (fresh and dry weight, green leaf area) collected during destructive harvest were compared to the nontreated control within block of each respective run to calculate the percent reduction. Data were converted to percent reduction in each block as $\{[1 - (\text{treatment mass or coverage/mass or coverage in nontreated cone-tainer})] \times 100\}$ prior to analysis.

Analysis of variance was performed using the PROC GLIMMIX procedure in SAS 9.4 (SAS Institute Inc., Cary NC 27513). When *F*-tests were significant at $P \leq 0.05$, Tukey's HSD test ($\alpha = 0.05$) was used for mean separation. Correlation coefficients between measurements were determined using the PROC CORR procedure. Additionally, heat maps of the data were created using Prism software (GraphPad Software, Inc., La Jolla, CA 92037) to assist with the visualization of hard water antagonism and comparison of treatments.

Synthetic Auxin Premix Efficacy Is Dependent on Formulation and Water Hardness

A second experiment was conducted in the Purdue University horticulture greenhouses to determine the influence of water hardness

on the efficacy of different commercially available premixes of synthetic auxin herbicides with 2,4-D or MCPA (Table 2). Herbicides were selected to include three groups: water-soluble formulations (i.e., soluble liquids), water insoluble emulsifiable formulations (i.e., emulsifiable concentrates, EC), and emulsion formulations (i.e., emulsion-in-water, EW; microemulsion, ME; Mulqueen 2003; Tu et al. 2001). Two commercially available herbicides in each group with a different set of ingredients were selected (Table 2). The efficacy of each of the six herbicides was tested in two water hardness levels: distilled water or water with 600 mg CaCO₃ L⁻¹ equivalents for a total of 12 treatment combinations. Spray solutions were prepared as described previously and weed propagation, transplanting, and plant culture were also similar to the first experiment.

The experiment was initiated in the greenhouse in autumn 2017 and repeated in winter 2018 with each arranged as a randomized complete block design with four blocks. Treatments were applied to 5- to 12-cm-diameter dandelion rosettes. Applications were made on October 31, 2017 in experimental run 1 and February 20, 2018 in experimental run 2 using the same equipment and application parameters described in Experiment 1. Temperatures for experimental run 1 averaged 23.5 C with an average daily light integral of 14 m⁻² d⁻¹. Temperatures for experimental run 2 averaged 23.8 C with an average daily light integral of 23 mol m⁻² d⁻¹.

Weed epinasty, control, digital images, fresh weights, and dry weights were collected in the same manner as the first experiment. A nontreated control was included in this experiment and prior to analysis all measurement data were converted to percent reduction in the same manner described in the first experiment. ANOVA was conducted in two different ways with the PROC GLM procedure in SAS 9.4. Both the ANOVA of formulation type by water hardness and ANOVA of commercially available herbicide premixes by water hardness were conducted. Main effects were formulation type (three levels: water-soluble formulations, emulsifiable formulations, and emulsions) and hardness (two levels) in the first ANOVA and commercially available herbicide premixes (six herbicides) by hardness (two levels) in the second ANOVA. For data sets where main effects or interactions were significant at $P \leq 0.05$,

Table 2. Variable formulations of commercially available premixtures containing 2,4-D tested for their efficacy in soft and hard water in Experiment 2.

Ingredients	Trade name and manufacturer	Herbicide applied	EPA registration no.	Ingredient formulation	Product formulation type ^a
2,4-D + MCPP + dicamba	Triplet® SF, Nufarm Americas Inc., Burr Ridge, IL 60527	1.16 + 0.31 + 0.11 ^b	228-312	dimethylamine salt	water-soluble
MCPA + triclopyr + dicamba	Horsepower®, Nufarm Americas Inc., Burr Ridge, IL 60527	1.33 + 0.13 + 0.13	228-313	dimethylamine salt (2,4-D and dicamba) or trimethylamine salt (triclopyr)	water-soluble
2,4-D + MCPP + dicamba + cafentrazone	SpeedZone®, PBI/Gordon Corporation, Kansas City, MO 64101	0.85 + 0.27 + 0.08 + 0.028	2217-833	2-ethylhexyl ester (2,4-D)	emulsifiable formulation ^c
MCPA + triclopyr + dicamba	Cool Power®, Nufarm Americas Inc., Burr Ridge, IL 60527	1.26 + 0.13 + 0.13	228-317	2-ethylhexyl ester (2,4-D) or butoxyethanol ester (triclopyr)	emulsifiable formulation
2,4-D + fluroxypyr + halauxifen-methyl	GameOn™, Dow AgroSciences LLC, Indianapolis, IN 46268	0.96 + 0.19 + 0.01	62719-724	Choline salt (2,4-D), 1-methylheptyl ester (fluroxypyr), methyl ester (halauxifen)	emulsion (emulsion-in-water)
2,4-D + fluroxypyr + dicamba	Escalade® 2, Nufarm Americas Inc., Burr Ridge, IL 60527	1.0 + 0.14 + 0.14	228-442	dimethylamine salt (2,4-D), 1-methylheptyl ester (fluroxypyr)	emulsion (microemulsion, ME)

^aThree formulation types tested included: water-soluble, emulsifiable, and emulsion.

^bApplication rate for each ingredient in the commercial premix. The application rate selected for each herbicide was the high label rate.

^cThe emulsifiable concentrate (EC) formulation was used in this experiment. The formulation of SpeedZone was changed in 2020 from EC to emulsion-in-water (EW).

Fisher's protected LSD test ($\alpha = 0.05$) was used for mean separation. No significant treatment by experimental run interactions were present, so data were analyzed across experimental runs. Additionally, heat maps of the data were created similar to Experiment 1 to assist with the visualization of hard water antagonism by formulation type.

Results and Discussion

Water Hardness and Adjuvant Inclusion Impact on 2,4-D Formulations

To ensure that mixture pH was not a confounding factor, measurements were taken at each step of preparation for all 2,4-D formulation mixtures at both 0 mg CaCO₃ L⁻¹ and 600 mg CaCO₃ L⁻¹ (data not shown). The pH readings for all mixtures were above the pK_a (2.64) of the 2,4-D in its acid form and ranged from 3.52 (for 2-EHE) to 9.39 (for MMA). The lowest pH of a water-soluble formulation was 4.61, which is 1.97 pH units above the pK_a (2.64) of 2,4-D. At a pH of 4.64, 99% of the 2,4-D molecules are predicted to become dissociated (Tan and Crabtree 1994). Thus, we can infer that all water-soluble formulations (amines and choline) were dissociated and able to interact with Ca²⁺ in the hard water solutions (Tan and Crabtree 1994). Herbicides formulated as emulsifiable concentrates do not have a pK_a value because they do not dissociate in solution, or come into direct contact with cations that cause hard water antagonism. This applies to all water-insoluble ester formulations of 2,4-D (Peterson et al. 2016).

No significant treatment by experimental run interactions were present, so data were analyzed across experimental runs. The main effect of 2,4-D herbicide formulation and the main effect of water hardness were both significant for all observed ratings and calculated percent reductions ($P < 0.001$, data not shown). ANOVA also revealed 2,4-D formulation-by-water hardness interactions for all ratings and measurements ($P < 0.025$, data not shown) and discussion of our results were based on these interactions.

Dandelions treated with 2,4-D BEE exhibited more epinasty than amine and choline formulations at 1 WAA but there were no differences in epinasty between 2,4-D applied in soft water to those applied in hard water (Table 3). The increased efficacy of ester formulations continued throughout the duration of the experiment (Tables 3 and 4), and was consistent with faster uptake of this formulation (Peterson et al. 2016). Treatments with the lowest epinasty rating 2 WAA were amines and a choline formulation of 2,4-D mixed in hard water. There was a separation between soft and hard water treatments with a reduction in epinasty for several of the water-soluble formulations (DMA w/o prop, MMA, and choline) of 2,4-D mixed with hard water (Table 3). One observation from this study was that hard water antagonism is difficult to diagnose the first week after 2,4-D applications, as there is sufficient herbicide uptake to cause epinasty, but insufficient time to evaluate weed control.

Formulation by water hardness interactions for control ratings 3 and 4 WAA revealed similar results to earlier 2 WAA epinasty ratings. Ester formulations continued to provide the highest dandelion control of all the 2,4-D formulations tested. Furthermore, the water-insoluble ester formulations were not influenced by water hardness at any rating (Table 3). The improved control from the ester formulations is largely attributed to increased leaf absorption of the ester compared to amine formulations (Richardson 1977). Differences in control 3 and 4 WAA between water hardness treatments revealed that hard water reduced the efficacy of nearly all water-soluble amine formulations when mixed with hard water. The only water-soluble formulation that did not have a significant reduction in control 3 and 4 WAA was 2,4-D DEA, which was attributed to the poor dandelion control observed in this experiment in both soft and hard water (Table 3).

The destructive harvesting and calculation of percent fresh shoot weight reduction, dry shoot weight reduction, dry root weight reduction, and green leaf area reduction revealed a similar story about the 2,4-D formulation by water hardness interaction. Measurements collected 4 WAA were highly correlated with one another ($r = >0.96$, $P < 0.0001$). The highest reduction in

Table 3. Percent epinasty 1 and 2 WAA and percent control 3 and 4 WAA following applications of each respective 2,4-D formulation at 1.6 kg ae ha⁻¹ in Experiment 1 as influenced by 2,4-D formulation and water hardness.^{c,d}

2,4-D Formulation	Epinasty 1 WAA	Epinasty 2 WAA	Control 3 WAA	Control 4 WAA
	%			
DMA				
soft ^a	29 b-e ^b	31 c-f	42 c	42 bc
hard ^a	14 cde	14 f	4 ef*	5 e*
DMA w/o prop				
soft	29 b-e	50 bc	70 ab	59 b
hard	13 cde	14 f*	4 ef*	4 e*
DEA				
soft	25 b-e	23 def	12 def	14 de
hard	10 e	8 f	4 ef	3 e
MIPA				
soft	37 a-e	42 bcd	46 bc	51 bc
hard	20 b-e	16 ef	6 ef*	6 e*
MMA				
soft	28 b-e	41 b-e	30 cde	38 bc
hard	12 de	15 f*	8 def	6 e*
Choline				
soft	34 b-e	21 b-e	33 cd	34 cd
hard	13 cde	11 f*	3 f*	3 e*
2-EHE				
soft	40 a-d	62 ab	96 a	100 a
hard	41 a-c	58 ab	91 a	94 a
BEE				
soft	46 ab	61 ab	87 a	87 a
hard	65 a	77 a	91 a	94 a

^aDistilled water with 0 mg CaCO₃ L⁻¹ was used as the soft water source. Hard water was prepared by adding calcium chloride (CaCl₂ 2[H₂O] L⁻¹) at 887.1 mg L⁻¹ to achieve a water hardness level of 600 mg CaCO₃ L⁻¹.

^bMeans followed by the same letter within columns type are similar according to Tukey's HSD test ($\alpha = 0.05$). Each mean value is representative of eight observations. Means followed by an asterisk (*) within a column and within a formulation type indicate statistical separation from the distilled (soft) water carrier and the occurrence of hard water antagonism.

^cAbbreviation: WAA, weeks after application. See Table 1 for additional abbreviations.

^dData were combined across experimental runs.

dandelion tissue weight or green leaf area occurred when dandelions were treated with either the 2-EHE or BEE water-insoluble ester formulations of 2,4-D (Table 4). Furthermore, the water insoluble ester formulations performed in similar ways when used in both soft and hard water mixes (Table 4; Figure 1). This observation supports statements on the indifference of using hard or soft water when mixing 2,4-D esters for applications (Müller 2000; Nalewaja et al. 1990, 1991). The 2,4-D ester formulation is protected against hard water antagonism since the 2,4-D remains dissolved in the oil, whereas the emulsifying agent disperses the droplets (Peterson et al. 2016).

The only amine (water-soluble) treatments that achieved similar results to those of the ester formulations were those applied with soft water (Table 4; Figure 1). All water-soluble 2,4-D formulations had reduced efficacy when applied with hard water at 600 mg CaCO₃ L⁻¹ in this experiment (Figure 1). This is consistent with earlier reports on older standalone 2,4-D amine formulations such as 2,4-D diethanolamine, 2,4-D triethanolamine, and the previously used alkali salt formulations (Kelly 1953; Müller 2000; Nalewaja et al. 1990, 1991; Ross and Lembi 2009; Szabo and Buchholtz 1961). This is the first report of hard water antagonism of 2,4-D MMA and MIPA, although antagonism of DEA and DMA was previously reported (Nalewaja et al. 1990, 1991; Patton et al. 2016; Roskamp et al. 2013). Our results are consistent with unpublished results reported by Nalewaja et al. (1991) who indicated that other 2,4-D salt formulations were subject to hard water antagonism similar to the 2,4-D DEA formulation. Kelly (1953) noted

Table 4. Percent dandelion tissue weight reduction 4 WAA and percent green leaf area reduction following applications of each 2,4-D formulation at 1.6 kg ae ha⁻¹, as influenced by 2,4-D formulation and water hardness.^{d,e}

2,4-D Formulation	Fresh shoot weight	Dry shoot weight	Dry root weight	Green leaf area
	% reduction			
DMA				
soft ^a	49 bcd ^b	49 abc	66 ab ^b	64 a-d
hard ^a	19 ef*	28 cde	15 de*	-7 c-f ^c
DMA w/o prop				
soft	47 bcd	49 abc	62 ab	55 a-e
hard	12 ef*	21 de*	13 de*	-11 def
DEA				
soft	30 cde	35 b-e	29 cd	15 b-f
hard	7 f*	15 e	-3 e*	-42 f
MIPA				
soft	49 bcd	48 abc	68 ab	67 abc
hard	9 f*	18 de*	20 de*	-4 c-e
MMA				
soft	46 bcd	43 a-d	58 bc	60 a-d
hard	14 ef*	21 ed	11 de*	-19 ef*
Choline				
soft	38 cde	35 b-d	61 b	54 a-e
hard	14 ef	21 de	5 de*	-27 f*
2-EHE				
soft	85 a	62 a	93 a	99 a
hard	70 ab	52 abc	89 ab	96 a
BEE				
soft	64 abc	52 abc	87 ab	90 ab
hard	73 ab	58 ab	88 ab	96 a

^aDistilled water with 0 mg CaCO₃ L⁻¹ was used as the soft water source. Hard water was prepared by adding calcium chloride (CaCl₂ 2[H₂O] L⁻¹) at 887.1 mg L⁻¹ to achieve a water hardness level of 600 mg CaCO₃ L⁻¹.

^bMeans followed by the same letter within columns type are similar according to Tukey's HSD test ($\alpha = 0.05$). Each mean value is representative of eight measurements. Means followed by an asterisk (*) within a column and within a formulation type indicate statistical separation from the distilled (soft) water carrier and the occurrence of hard water antagonism.

^cMeans with a negative value indicate the plants were larger (higher mass or leaf area) than the nontreated control.

^dAbbreviation: WAA, weeks after application. See Table 1 for additional abbreviations.

^eData were combined across experimental runs.

that 2,4-D formulations were designed to perform well in 1,000 mg CaCO₃ L⁻¹ through the addition of sequestering agents in the formulation to protect against hard-water antagonism. However, this experiment demonstrated that current water-soluble 2,4-D formulations contain little protection against hard water (600 mg CaCO₃ L⁻¹) antagonism.

Based on a report by Devkota and Johnson (2016) and preliminary research by Schortgen (2017) that demonstrated hard water antagonism of the 2,4-D choline formulation, Dow AgroSciences improved the 2,4-D choline formulation to have greater stability and performance in hard water. The GF-2654 formulation of 2,4-D choline salt used in this experiment is different from that of Enlist™ One (2,4-D choline salt, GF-3335, EPA reg. no. 62719-695; Montague 2017), which is formulated to form an emulsion upon dilution (Li et al. 2013; Shao and Tank 2015). (Note: The GF-2654 formulation of 2,4-D choline salt is available as Freelexx® (EPA reg. no. 62719-634) and Embed® (EPA reg. no. 62719-634).)

Synthetic Auxin Premix Efficacy Is Dependent on Formulation and Water Hardness

Data were analyzed across experimental runs since there were no significant treatment by experimental run interactions. ANOVA for 2,4-D treated dandelions revealed a hardness-by-formulation interaction for control, shoot fresh weight, dry weight, green leaf

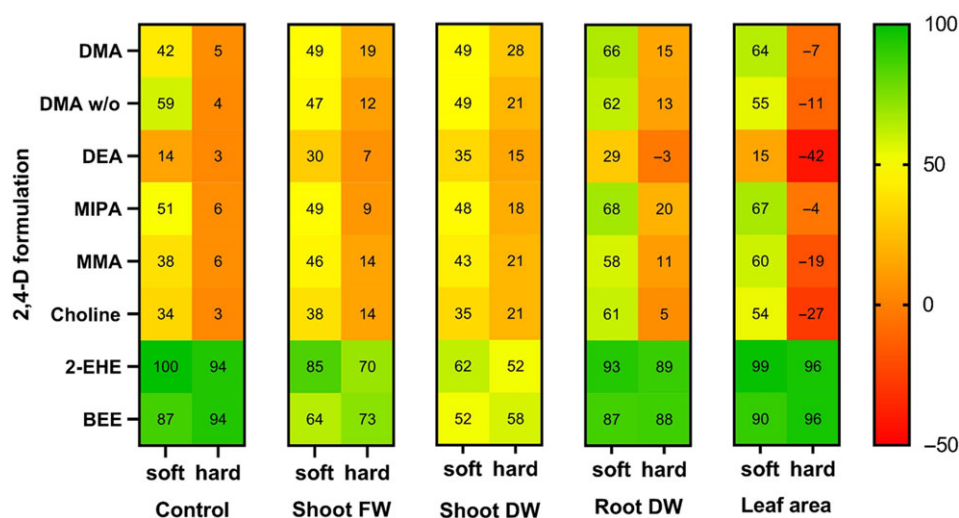


Figure 1. Visualization of hard water antagonism using a heat map with the relative performance of each 2,4-D formulation at 1.6 kg ae ha⁻¹ 4 wk after application, as influenced by 2,4-D formulation and water hardness. Mean values appear in Tables 3 and 4. A four-color scale with green indicating highest weed control, yellow indicating intermediate control, and orange or red indicating poor control. A shift in heat color within a formulation from soft (0 mg CaCO₃ L⁻¹) to hard water (600 mg CaCO₃ L⁻¹) is indicative of hard water antagonism. A change in color by formulation is indicative of efficacy differences. See statistical comparisons provided in Tables 3 and 4. Abbreviations: DW, dry weight; FW, fresh weight.

coverage, and regrowth (Table 5). Epinasty 1 WAA was less than 16% for all treatments with less than 5% difference between formulations with no difference in epinasty between treatments 2 WAA (data not shown). Subtle differences occurred between premix formulation type applied in distilled water. Most notable was that treatments of water-soluble premix formulations in hard water (600 mg CaCO₃ L⁻¹) were less effective at controlling dandelions compared to other premix formulations, revealing reduced efficacy due to hard water antagonism (Table 6; Figure 2). Dandelions treated with water-soluble formulations of synthetic auxin herbicides in hard water (600 mg CaCO₃ L⁻¹) had higher fresh and dry shoot weights, green leaf area, and regrowth following a harvest 4WAA compared with water-soluble formulations applied in distilled water (Table 6; Figure 2).

ANOVA with herbicide premix as the main effect (Table 7) also demonstrated hard water antagonism of water-soluble herbicides regardless of their synthetic auxin ingredients (Table 8). Herbicides formulated as esters (emulsifiable concentrates) and newer emulsion formulations (emulsion-in-water or microemulsion) were not antagonized by hard water when evaluated 4 WAA (Table 8). Although the formulations were not tested as single ingredients, results from these soluble premixes (2,4-D + MCPP + dicamba; MCPA + triclopyr + dicamba) are consistent with other reports that water-soluble formulations of MCPA are subject to hard water antagonism similar to that of 2,4-D (de Villiers et al 2000; Nalewaja et al. 1991; Nalewaja and Matysiak 1993). We were unable to find any previously published information on the potential for hard water antagonism of MCPP or triclopyr. Previously, Roskamp et al. (2013) reported that dicamba diglycolamine salt was not subject to hard water antagonism when tested as a standalone ingredient at 280 g ae ha⁻¹ and applied at 140 L ha⁻¹. Our results show that premixes with dicamba dimethylamine salt (applied here at 108–133 g ae ha⁻¹) are subject to hard water antagonism. Possible explanations are that 1) instead of dicamba being antagonized by hard water, the phenoxyacetic acids 2,4-D, MCPA, and MCPP in these premixes (Table 2) are being antagonized, thereby leading to a reduced efficacy; or 2) lower rates of dicamba dimethylamine tested here at higher application volumes (i.e.,

more calcium cations in solution) are subject to hard water antagonism. The potential for dicamba to be antagonized at lower rates by hard water is supported by Nalewaja and Matysiak (1993) who reported dicamba dimethylamine salt antagonism when applied at 70 g ae ha⁻¹ in a spray volume of calcium chloride solution at 160 L ha⁻¹.

The 2,4-D-containing emulsion formulation types (emulsion-in-water or microemulsion) tested in this experiment were not antagonized by hard water (Tables 6 and 8; Figure 2). In Experiment 1 presented here and a previous report by Devkota and Johnson (2016), the water-soluble 2,4-D choline salt was antagonized by hard water. However, in Experiment 2, the premix of 2,4-D choline + fluroxypyr + halauxifen-methyl (GameOn™) was not antagonized due to a formulation difference. Li (2015) detailed a new method of combining the water-soluble 2,4-D choline salt with water-insoluble formulations of other synthetic auxin herbicides, which provides a stable, transparent, aqueous solution formulation that forms an oil-in-water emulsion upon dilution. The GameOn™ formulation tested here combines the 2,4-D choline salt with fluroxypyr meptyl 1-methylheptyl ester and the methyl ester of halauxifen-methyl (Anonymous 2019). The Escalade® 2 formulation, which also was not subject to antagonism, is a microemulsion formulation (Anonymous, 2020), and combines the 2,4-D dimethylamine salt with fluroxypyr meptyl 1-methylheptyl ester and dicamba acid (Anonymous 2016). As such, formulating water-soluble 2,4-D using water-based dispersion technology such as oil-in-water emulsions appears to be an effective method to prevent hard water antagonism. Water-based dispersion technology has advantages over conventional emulsifiable concentrates in that less solvent is needed, lower skin and eye toxicity is achieved, formulations are more compatible with containers, and particle drift can be reduced (Li et al. 2013; Mulqueen 2003).

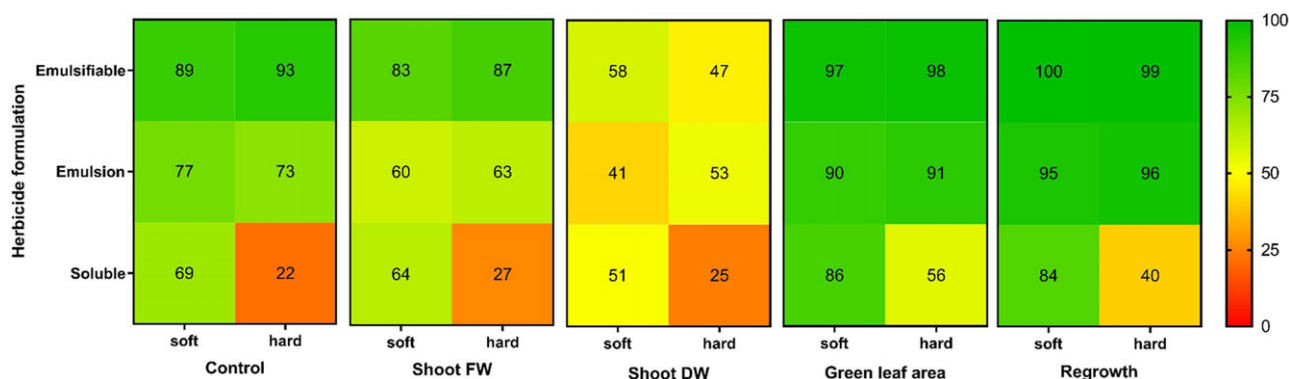
Previous research demonstrated that 2,4-D dimethylamine was subject to hard water antagonism (Patton et al. 2016; Roskamp et al. 2013; Schortgen and Patton 2020). This research demonstrates that all 2,4-D water-soluble formulations and water-soluble premixes with phenoxyacetic acid herbicides are subject to

Table 5. Formulation solubility type ANOVA for epinasty, visual control, shoot fresh weight reduction, shoot dry weight reduction, and percent green leaf area reduction, and regrowth dry shoot weight reduction with P-values for the main effects of formulation solubility type, water hardness, and the interaction between hardness and formulation solubility type in Experiment 2.^a

	Epinasty 1 WAA	Epinasty 2 WAA	Control 3 WAA	Control 4 WAA	Percent fresh shoot weight reduction 4 WAA	Percent dry shoot weight reduction 4 WAA	Percent green leaf area reduction 4 WAA	Percent regrowth dry shoot weight reduction 8 WAA
	P-value							
Formulation ^b	0.0012	0.0659	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Hardness	0.3350	0.0916	0.3000	0.0008	0.0707	0.1986	0.0022	0.0010
Formulation by hardness	0.8252	0.0673	0.0002	<0.0001	0.0005	0.0042	<0.0001	<0.0001

^aAbbreviation: WAA, weeks after application.^bThree formulation types tested included: water-soluble, emulsifiable, and emulsion.**Table 6.** Influence of herbicide formulation type and water hardness level on dandelion control efficacy in experiment two.^{a,b,c}

Formulation type ^d	Hardness ^e mg CaCO ₃ L ⁻¹	Control 4 WAA	Control 4 WAA	Fresh shoot weight 4 WAA	Dry shoot weight 4 WAA	Green leaf area 4 WAA	Regrowth dry shoot weight 8 WAA
		%		% reduction			
Emulsifiable	0	71 a	85 ab	78 ab	54 ab	96 a	100 a
Emulsifiable	600	77 a	91 a	85 a	61 ab	97 a	99 a
Emulsion	0	52 b	77 abc	60 c	41 b	90 ab	95 ab
Emulsion	600	65 ab	73 bc	63 bc	53 ab	91 ab	96 ab
Soluble	0	52 b	69 c	64 bc	51 ab	86 b	84 b
Soluble	600	18 c*	22 d*	27 d*	25 c*	56 c*	40 c*

^aAbbreviation: WAA, weeks after application.^bData were combined across experimental runs.^cMeans followed by the same letter within columns type are similar according to Tukey's HSD test ($\alpha = 0.05$). Each mean value is representative of eight measurements. Means followed by an asterisk (*) within a column and within a formulation type indicate statistical separation from the distilled (soft) water carrier and the occurrence of hard water antagonism.^dThree formulation types tested included: water-soluble, emulsifiable, and emulsion. Products described as emulsion-in-water and microemulsion are included in the emulsion group (Table 2).^eDistilled water with 0 mg CaCO₃ L⁻¹ was used as the soft water source. Hard water was prepared by adding calcium chloride (CaCl₂ 2[H₂O] L⁻¹) at 887.1 mg L⁻¹ to achieve water hardness levels of 600 mg CaCO₃ L⁻¹.**Figure 2.** Visualization of hard water antagonism using a heat map with the relative performance of each formulation type 4 wk after application (and 8 wk after application for regrowth), as influenced by formulation type and water hardness. Mean values appear in Table 6. A three-color scale with green indicating highest weed control, yellow indicating intermediate control, and red indicating poor control. A shift in heat color within a formulation from soft (0 mg CaCO₃ L⁻¹) to hard water (600 mg CaCO₃ L⁻¹) is indicative of hard water antagonism. A change in color by formulation is indicative of efficacy differences. See statistical comparisons provided in Table 6. Abbreviations: DW, dry weight; FW, fresh weight.

hard water antagonism. Formulations of 2,4-D containing emulsifying agents can protect ingredients against antagonism by the water-insoluble nature of the ingredients in their formulation.

Applicators can prevent hard water antagonism of water-soluble formulations by using high quality water low in calcium, magnesium, and iron cations or by mixing with a water conditioning agent such as ammonium sulfate or urea ammonium

nitrate (Schortgen 2017; Schortgen and Patton 2020). A common communication problem is that manufacturer labels of water-soluble formulations of 2,4-D do not include instructions on how to mix it with hard water or precautions against applying it using hard water (Schortgen and Patton 2020). Schortgen and Patton (2020) suggest that manufacturers of 2,4-D dimethylamine should consider adding statements to the label about the use of ammonium

Table 7. Commercially available herbicide ANOVA for epinasty, visual control, shoot fresh weight reduction, shoot dry weight reduction, and percent green leaf area reduction, and regrowth dry shoot weight reduction with P-values for the main effects of commercially available herbicide premixes, water hardness, and the interaction between hardness and commercially available herbicide premixes in Experiment 2.^a

	Epinasty 1 WAA	Epinasty 2 WAA	Control 3 WAA	Control 4 WAA	Percent fresh shoot weight reduction 4 WAA	Percent dry shoot weight reduction 4 WAA	Percent green leaf area reduc- tion 4 WAA	Percent regrowth dry shoot weight reduc- tion 8 WAA
	P-value							
Herbicide ^b	0.0004	0.1436	<0.0001	<0.0001	<0.0001	0.0156	<0.0001	<0.0001
Hardness	0.3006	0.0966	0.2955	0.0004	0.0698	0.5498	0.0002	0.0004
Herbicide by hardness	0.0799	0.3176	0.0003	<0.0001	0.0021	0.0103	<0.0001	<0.0001

^aAbbreviation: WAA, weeks after application.

^bThree commercially available herbicide premixes tested are listed in Table 2.

Table 8. Efficacy of herbicide premixes and water hardness level on dandelion control efficacy in Experiment 2.^{a,b}

Herbicide ^c	Hardness ^d	Formulation type	Control 4 WAA	Fresh shoot weight 4 WAA	Dry shoot weight 4 WAA	Green leaf area 4 WAA	Regrowth dry shoot weight 8 WAA
	mg CaCO ₃ L ⁻¹		%	% reduction			
Triplet® SF	0	Soluble	81 ab ^e	70 abc	58 a	89 ab	87 ab
Triplet® SF	600		20 d*	24 d*	23 d*	42 d*	33 c*
Horsepower®	0	Soluble	57 c	57 c	44 a-d	84 b	80 b
Horsepower®	600		24 d*	29 d*	26 cd	70 c*	48 c*
SpeedZone®	0	Emulsifiable	77 abc	70 abc	47 abc	94 ab	100 a
SpeedZone®	600		94 a	88 a	59 a	99 a	98 ab
Cool Power®	0	Emulsifiable	93 a	87 a	62 a	99 a	100 a
Cool Power®	600		87 ab	82 ab	64 a	97 a	100 a
GameOn™	0	Emulsion	83 ab	61 bc	34 bcd	92 ab	98 ab
GameOn™	600		86 ab	73 abc	55 ab	94 ab	97 ab
Escalade® 2	0	Emulsion	72 bc	58 c	48 ab	88 ab	92 ab
Escalade® 2	600		59 c	53 c	51 ab	89 ab	95 ab

^aAbbreviation: WAA, weeks after application.

^bData were combined across experimental runs.

^cHerbicide ingredients and rates are listed in Table 2.

^dDistilled water with 0 mg CaCO₃ L⁻¹ was used as the soft water source. Hard water was prepared by adding calcium chloride (CaCl₂·2[H₂O] L⁻¹) at 887.1 mg L⁻¹ to achieve water hardness levels of 600 mg CaCO₃ L⁻¹.

^eMeans followed by the same letter within columns type are similar according to Tukey's HSD test ($\alpha = 0.05$). Each mean value is representative of eight measurements. Means followed by an asterisk (*) within a column and within a formulation type indicate statistical separation from the distilled (soft) water carrier and the occurrence of hard water antagonism.

sulfate similar to the statement on the glyphosate label. Furthermore, manufacturers may need to modify sequestering agents in the formulation to better protect against hard-water antagonism (Kelly 1953; Peterson et al. 2016).

Another solution to preventing hard water antagonism is to use formulations with emulsifying agents. Applications of esters when environmental conditions permit or the use of advanced formulations, such as emulsion-in-water formulations, will prevent hard water antagonism. While these emulsion-in-water, microemulsion, or other advanced emulsion formulations are shown here to protect against hard water antagonism, practitioners often struggle to know the formulation type they are using because the information is often difficult to find. For example, neither the Escalade® 2 or GameOn™ labels (Anonymous 2016, 2019) indicate their formulation type or that they form an emulsion-in-water. Conversely, both the Cool Power® and SpeedZone® labels note that the material forms an emulsion-in-water (Anonymous 2003, 2009). None of these four labels contain a statement that inform the applicator that the products are formulated for effective weed control when mixed with hard water. Manufacturers should amend labels

of all 2,4-D-containing herbicides to assist applicators in the prevention of hard water antagonism.

Acknowledgments. We thank the Midwest Regional Turf Foundation for funding this project and Dow AgroSciences LLC, Nufarm Americas Inc., PBI-Gordon Corp., and Winfield Solutions LLC for providing 2,4-D formulation samples or commercially available herbicides for testing. This work was supported by Hatch project 1004762 from the U.S. Department of Agriculture–National Institute of Food and Agriculture. No conflicts of interest have been declared. We also thank Drs. Steve Weller, Fred Whitford, and Bryan Young for their helpful review of this manuscript; and express our appreciation to Katherine Wierzbowski, Joe Ikely, Dan Weisenberger, and Julie Young for their assistance with this experiment.

References

- Anonymous (2003) Cool Power® herbicide product label. Burr Ridge, IL: Nufarm Americas Inc. 5 p
- Anonymous (2007) 2,4-D. Pages 332–328 in Senseman SA, ed. Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America
- Anonymous (2009) SpeedZone® herbicide product label. Kansas City, MO: PBI/Gordon Corporation. 5 p

- Anonymous (2016) Escalade® 2 herbicide product label. Burr Ridge, IL: Nufarm Americas Inc. 7 p
- Anonymous (2019) GameOn™ herbicide product label. Indianapolis, IN: Dow AgroSciences LLC. 6 p
- Anonymous (2020) Escalade® 2 - US - Turf. <https://nufarm.com/usturf/product/escalade2/>. Accessed August 10, 2020
- Bekbölet M, Yenigün O, Yücel I (1999) Sorption studies of 2,4-D on selected soils. *Water Air Soil Pollut* 111:75–88
- Boyd CE (2015) *Water Quality: An Introduction*. 2nd ed. Auburn, AL: Springer. 357 p
- Christians NE, Patton AJ, Law QD (2017) *Fundamentals of turfgrass management*. Hoboken, NJ: John Wiley & Sons
- de Villiers BL, Smit HA, Lindeque RC, Smit JJ (2000) Optimizing MCPA (K-salt) activity with adjuvants. *S Afr J Plant Soil* 17:63–65
- Devkota P, Johnson WG (2016) Effect of carrier water hardness and ammonium sulfate on efficacy of 2,4-D choline and premixed 2,4-D choline plus glyphosate. *Weed Technol* 30:878–887
- Eytcheson AN, Reynolds D, Irby J, Steckel L, Walton L, Haygood R, Ellis D, Richburg J (2012) Volatility of 1713 GF-2726 as compared with other auxin herbicides. Beltwide Cotton Conference. Orlando, FL, January 3–6, 2012
- Grover R, Maybank J, Yoshida K (1972) Droplet and vapor drift from butyl ester and dimethylamine salt of 2,4-D. *Weed Sci* 20:320–324
- Hem JD (1985) Study and interpretation of the chemical characteristics of natural water. 3rd ed. Water-Supply Paper 2254. Alexandria, VA: U.S. Geological Survey. Office of Chemical Safety and Pollution and Prevention. Pp 63–64
- Jervais JA, Luukinen B, Nuhl K, Stone D (2008) 2,4-D Technical fact sheet. Corvallis, OR: National Pesticide Information Center, Oregon State University extension services. <http://npic.orst.edu/factsheets/archive/2,4-DTech.html> Accessed: December 23, 2020
- Kelly JA (1953) Present methods of formulation. *J Agric Food Chem* 1:254–257
- Li M, inventor; Dow Agrosciences, assignee (2015) Aqueous herbicidal concentrates. U.S. Patent Application No. 14/511,288
- Li M, Tank H, Kennedy A, Zhang H, Downer B, Ouse D, Liu L. (2013). Enlist Duo herbicide: a novel 2, 4-D plus glyphosate premix formulation with low potential for off-target movement. *In Pesticide Formulation and Delivery Systems: 32nd Volume, Innovating Legacy Products for New Uses*. West Conshohocken, PA: ASTM International
- Montague KV (2017) Notice of Pesticide Registration, GF-3335. Washington, DC: U.S. Environmental Protection Agency. https://www3.epa.gov/pesticides/chem_search/ppls/062719-00695-20170131.pdf Accessed: January 16, 2019
- Müller F (2000) *Agrochemicals; composition, production, toxicology, application*. 1st ed. Weinheim Germany: Wiley VCH
- Mulqueen P (2003) Recent advances in agrochemical formulation. *Adv Colloid Interface Sci* 106:83–107
- Nalewaja JD, Matysiak R (1993) Spray carrier salts affect herbicide toxicity to kochia (*Kochia scoparia*). *Weed Technol* 7:154–158
- Nalewaja JD, Wozinca Z, Manthey FA (1990) Sodium bicarbonate antagonism of 2,4-D amine. *Weed Technol* 4:588–591
- Nalewaja JD, Wozinca Z, Matysiak R (1991) 2,4-D amine antagonism by salts. *Weed Technol* 5:873–880
- Patton AJ, Weisenberger DV, Johnson WG (2016) Divalent cations in spray water influence 2,4-D efficacy on dandelion (*Taraxacum officinale*) and broadleaf plantain (*Plantago major*). *Weed Technol* 30:431–440
- Patton AJ, Weisenberger DV, Schortgen GP (2018) 2,4-D-Resistant Buckhorn Plantain (*Plantago lanceolata*) in Managed Turf. *Weed Technol* 32:182–189
- Peterson GE (1967) The discovery and development of 2,4-D. *Agr Hist* 41: 243–254
- Peterson MA, McMaster SA, Riechers DE, Skelton J, Stahlman PW (2016) 2,4-D past, present, and future: a review. *Weed Technol* 30:303–345
- Richardson RG (1977) A review of foliar absorption and translocation of 2,4-D and 2, 4, 5-T. *Weed Res* 17:259–272
- Roskamp JM, Chahal GS, Johnson WG (2013) The effect of cations and ammonium sulfate on the efficacy of dicamba and 2,4-D. *Weed Technol* 27:72–77
- Ross MR, Lembi CA (2009) *Applied weed science*. 2nd ed. West Lafayette, IN: Purdue University. Pp 138–141
- Schneider CA, Rasband WS, Eliceiri KW (2012) NIH Image to ImageJ: 25 years of image analysis. *Nat Methods* 9:671–675
- Schortgen GP (2017) Enhancing weed control by reducing hard water antagonism of 2,4-D in spray tank mixtures. M.S. thesis. West Lafayette, IN: Purdue University
- Schortgen GP, Patton AJ (2020) Weed control by 2,4-D dimethylamine depends on mixture water hardness and adjuvant inclusion but not spray solution storage time. *Weed Technol* 34:107–116
- Shao H, Tank H, inventors; Dow Agrosciences, assignee (2015) Pesticide emulsion concentrates containing natural or petroleum derived oils and methods of use. U.S. Patent Application No. 14/535,455
- Stagg N, Blewett T, Tank H, Li M, Liu L, inventors; Dow Agrosciences, assignee (2013) October 22. Aqueous herbicidal concentrates of auxinic carboxylic acids with reduced eye irritation. U.S. Patent No. 8,563,473 B2
- Szabo SS, Buchholtz KP (1961) Penetration of living and non-living surfaces by 2,4-D as influenced by ionic additives. *Weeds* 9:177–184
- Tan S, Crabtree GD (1994) Cuticular penetration of 2,4-D as affected by interaction between diethylene glycol monooleate surfactant and apple leaf cuticles. *Pestic Sci* 41:35–39
- Thompson WT (1986). *Agricultural Chemicals, Book II, Herbicides*. Fresno, CA: Thompson Publications
- Tu M, Hurd C, Randall JM (2001). *Weed Control Methods Handbook*. Arlington, VA: The Nature Conservancy. <https://www.invasive.org/gist/products/handbook/methods-handbook.pdf>. Accessed: October 16, 2020