Effect of 2,4-D formulation on volatility under field conditions

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Abstract
Auxinic herbicides have been commonly used in production systems for broadleaf weed control for many years. One potential negative aspect to their use is their propensity to volatilize and move away from the treated area after application. This research examined three herbicide formulations and their relative amounts of vaporization following application under field conditions in Knoxville, TN, in 2017, 2018, and 2019. Herbicide treatments evaluated included 2,4-D choline, 2,4-D amine, and the diglycolamine (DGA) salt of dicamba. Ten field studies were conducted with major parameters including air sampler height (0.3 and 1.3 m) and applied surface condition (dry wheat stubble or green-plant vegetation). The relative volatility indicated by the study was that dicamba > 2,4-D choline = 2,4-D amine. Detected herbicide concentrations were numerically higher at the 0.3-m sampling height and in the green-plant surface condition. These results confirm that dicamba is more volatile than 2,4-D and that there was no difference in vapor emissions between the amine and choline salts of 2,4-D under field conditions.

Introduction
Interest in the volatility of auxin herbicides is not a new research area, and the relative volatilities of different formulations have been under investigation for decades (Carlsen et al.2006; Dorfler et al. 1991; Johnson et al. 2012). Marth and Mitchell (1949) found that the acid, sodium salt, and amide forms of 2,4-D were not volatile; in contrast, all ester formulations showed volatility based on a tomato (Solanum lycopersicum L.) plant bioassay (Marth and Mitchell 1949). Grover et al. (1972) showed that no significant amount of a dimethylamine formulation of 2,4-D was collected as a vapor; and they reported that the drift potential of the butyl ester was 10 times greater than for the dimethylamine formulation in their tests. It is widely accepted that the ester formulations are more volatile than the 2,4-D amine formulations by wide margins. Grover (1976) reported the relative volatility of the high volatile ester, low volatile ester, and amine salts to be 440:33:1 (Grover 1976). Many of these and later studies utilized dicot plant species as bio-indicators of auxin herbicide response, including tomato (Breeze 1990) and soybeans (Glycine max L.) or cotton (Gossypium hirsutum L.) (Sosnoskie et al. 2004; Sosnoskie et al. 2015). Other researchers have previously shown that various samplers of filter matrices effectively capture 2,4-D residues under field conditions (Cessna et al. 2000). Sosnoskie et al. (2015) reported <5% injury for the amine and choline formulations of 2,4-D at any distance to sensitive cotton plants. They also reported that exposure to the amine and choline formulations did not differentially affect plant heights. Injury ratings of 76%, 13%, and 5% were noted for cotton exposed to the ester, amine, and choline formulations, respectively, when under controlled-environment tunnels for 48 h. Results indicate that the choline formulation of 2,4-D was less volatile and injurious to cotton than the ester under the field conditions in this study.

Although this list of references by no means encompasses the full breadth of the published research on 2,4-D volatility, it does provide a background of reference upon which to base the current studies. Given the new transgenic soybean and cotton varieties that withstand 2,4-D postemergence application, there is much more interest in off-target movement of dicamba and 2,4-D in the current use patterns in the United States. This is the first published report directly comparing the volatility of the amine and choline salts of 2,4-D and dicamba under field conditions using quantitative air sampling followed by chemical analysis. Given the great interest in off-target movement of auxin herbicides, this research should be of relevance to many producers, extension personnel, industry registrants, as well as to regulators in the US Environmental Protection Agency and other governments around the world.

Materials and Methods
This research was conducted in Knoxville, TN, in the summers of 2017, 2018, and 2019 (Table 1). Research methods were largely based on a previously reported method.
(Mueller et al. 2013). All research was conducted at a site with Sequatchie loam soil that had no previous 2,4-D or dicamba use in the preceding 12 mo (34% sand, 48% silt, 18% clay, 1.3% organic matter, pH = 6.2, and cation exchange capacity = 11 mEq g⁻¹). Each individual plot was 100 m diam, and each plot received only one of the respective herbicides.

Herbicide treatments were applied at 140 L ha⁻¹ using a tractor-mounted boom (9.9 m in width) (RM series 200; DEMCO Manufacturing, Boyden, IA) with TTI 110015 nozzles (Teejet Manufacturing, Boyden, IA) operated at 275 kPa and a ground speed of 5.2 km h⁻¹. Each treated plot was 100 m in diameter, so each plot was 0.80 ha and was separated from others by a distance of at least 165 m. Boom height was 60 cm above the top of the plant canopy or soil. Applications were usually made at 8:00 am on the day of application. Each plot was sprayed using the same equipment to reduce the variation between chemical applications. The distance between the samplers, 165 ± 50 m, would allow for small droplets to settle upon the air column, the heights of 0.3 and 1.3 m were selected. Samplers of air samplers available, the authors wish to clearly state that each sampling arrangement, which necessitated the use of substantially higher 2,4-D dosages (e.g., 4X the normal application dose) so as to get a positive detection when lower flow rate samplers were used (Mueller 2015). This research utilized higher volume samplers to increase the sensitivity in our measurements. However, the use of these high-volume samplers also decreased the number of measurements that were possible in the field study because of the limited number of samplers. To provide assessment in the vertical surface, the heights of 0.3 and 1.3 m were selected. Samplers were located in the middle of the treated plot area (Figure 1).

The 2017 research involved two studies, both of which had dry wheat stubble as the surface condition (Table 1). The following 2 yr added the variable of having green vegetation also present in some of the field experiments. Previous research has indicated that the surface condition affects dicamba volatility (Behrens and Lueschen 1979; Mueller and Steckel 2021). Because of the limited number of air samplers available, the authors wish to clearly state that each field experiment was independent and separate from all the other experiments.

The time of application was targeted to be 8 am on the respective day of application. However, in two of the studies the actual application time was early in the afternoon at 1:50 and 12:30 pm (studies 1 and 3 in 2019), respectively. This change was made for operational necessities where various obstacles prevented the

Table 1. Field studies from 2017, 2018, and 2019 in Knoxville, TN, to examine the effect of 2,4-D formulation, surface residue, sampler height, and environmental conditions on herbicide emissions following application under field conditions.

| Year | Study no. | Ground cover | Application date | Time of application | Temperature at 0.3 m height Mean | Relative humidity Mean | Temperature at 1.3 m height Mean | Relative humidity Mean |
|------|-----------|--------------|------------------|---------------------|---------------------------------|-----------------------|---------------------------------|-----------------------|
| 2017 | 1         | Wheat stubble| July 25, 2017    | 8:20 am             | 19.2 47.3 29.9                | 74.0 18.9 40.5 28.4   | 73.1                            |                       |
|      | 2         | Wheat stubble| Aug 16, 2017     | 8:30 am             | 21.8 42.7 28.7               | 83.4 21.3 36.3 26.8   | 83.0                            |                       |
|      | 3 Green   | plants      | July 22, 2016    | 7:40 am             | 19.6 35.2 25.7               | 94.2 18.4 34.8 25.2   | 76.8                            |                       |
|      | 4 Green   | plants      | Aug 3, 2018      | 9:00 am             | 18.6 43.3 25.8               | 84.1 19.3 35.3 24.9   | 80.5                            |                       |
| 2018 | 1 Wheat   | stubble     | July 19, 2018    | 1:50 pm             | 21.3 38.5 27.3               | 78.7 21.3 37.8 27.1   | 77.7                            |                       |
|      | 2 Green   | plants      | Aug 20, 2019     | 7:30 am             | 19.3 42.0 28.5               | 74.8 19.5 39.9 28.9   | 71.9                            |                       |
|      | 3 Wheat   | stubble     | Aug 6, 2019      | 12:30 pm            | 19.7 47.0 27.8               | 73.4 18.6 35.4 25.1   | 75.1                            |                       |
|      | 4 Green   | plants      | Aug 29, 2019     | 7:40 am             | 13.8 37.0 22.6               | 89.0 13.1 32.9 21.4   | 74.1                            |                       |
| 2018, 2019 | Wheat stubble | (Four studies)       | 8:30 am             | 18.1 49.5 25.8               | 76.1 16.6 37.8 25.6   | 75.3                            |                       |
|      | Green plants | (Four studies)       | 8:30 am             | 13.8 42.0 25.8               | 86.8 13.1 39.9 25.7   | 74.9                            |                       |

All 27.3
conductance of the study as previously described. For example, in one instance a wind storm blew over a tree that blocked the road providing access to the field site. In the other, an equipment failure caused us to change the tractor to be used to spray the plots. Although the start time of the two studies was changed, the samplers were still operated for the same amount of time.

Within each plot, a temperature and relative humidity sensor probe was placed onto the soil surface and upon the sampler stand assembly. The temperature samplers (HOBO model PRO V2; Onset Computer Corp, Bourna, MA, USA) were set to operate at 30-min intervals. These units were re-zeroed prior to each study. Temperature and relative-humidity data were time stamped. The start time was synchronized to the initiation of each study. Previous calibrations of the HOBO units showed acceptable accuracy and agreement among the samplers (± 0.1% when tested at 20° C, 30° C, and 40° C, data not shown). The environmental data presented are for the mean over the sampled time interval averaged over the sensors for that plot. Additionally, there was a permanent weather station adjacent to the plots (<200 m) that measured wind speed and rainfall.

Q Model CF-1002BRL-Digital portable high-volume air samplers (Hi-Q.net, San Diego, CA) were utilized for air sampling in all experiments (Figure 1). Key components of the samplers included the air sampler main unit (CF-1002BRL-DIG) that included digital readouts for cumulative airflow and time interval sampling, a microfiber filter paper holder (part no. FHA–4CF), and a polyurethane foam (PUF) sampling module (part no. HIQ-1002-CF). The sampling media used was a 10-cm diam HEPA type high-purity binder-less 99.99% efficiency borosilicate glass fiber filter paper (part no. FPAE–102) and an 8-cm long polyurethane vapor collection substrate (part no. HIQ–3PUF). Additional parts included glass cartridges with stainless-steel screens for the PUF head sampler (part no. HIQ-1009) and the associated single Teflon end caps with silicone O-rings (part no. HIQ-1026).

The samplers operated at 185 L min⁻¹. Sampling intervals for all studies were 0.5 to 24 h after treatment. The exact amount of time was recorded for each sampler (data not shown). The samplers automatically measured the cumulative flow and elapsed time. In 2017 and 2018 there were two air samplers per plot at each sampler height. A nontreated control plot (distance ~ 400 m from treated plots) also contained one air sampler at 0.3 m height to validate a lack of contamination (data not shown, none detected). At the end of a designated sampling interval, the entire microfiber filter and the PUF were removed and stored until later analysis. Contamination between samplers was reduced by changing gloves between each sample change increment and using all new storage media, tubes, and bags. All sampling equipment was thoroughly cleaned between each experiment. Each measurement was an independent assessment of 2,4-D or dicamba emissions at that specific location. Sampling medium was placed directly into a small container upon collection at respective intervals, then into a cooler at 0 C and stored in a −20 C freezer for subsequent chemical analysis.

The microfiber filter papers were extracted with methanol for 1.0 h on a reciprocating shaker operated at 80 cycles min⁻¹. Extraction efficiency was approximately 90% for filter media, and data were not corrected for recovery (recovery data not shown). An aliquot of each extraction was passed through a 0.45-μm filter directly into a 2.0-mL vial for later chemical analysis. Once samples were extracted, they were stored in a dark freezer at −20° C and were analyzed within 3 d. 2,4-D and dicamba detections on the PUF media were inconsistent and problematic and were seldom appreciable compared with the amount collected on the filter papers (<10%, often about 1%). Analysis of PUF sampling media has proven to be problematic for dicamba (Mueller 2015). Data presented are from the filter paper matrix only. Given that the vast majority of the auxinic herbicide was collected on the filter paper, the test system was considered valid. Chemical extraction followed by MS/MS of the cellulosic filter paper had minimum analytical...
Herbicide concentrations were determined using an external standard technique of analytical standards of dicamba and 2,4-D acid (chemservice.com) dissolved in methanol. For 2017 and 2018 samples, an Agilent Liquid Chromatograph (1100 series) in line with an Agilent single quad 6120 mass (Agilent Technologies, Santa Clara, CA) spectrometer was used for analysis. A 25 cm × 4.6 mm C-18 column (phenomenex.com) at 35 C was used to separate components of interest from the matrix. The mobile phase (0.7 mL min⁻¹) used a gradient program of acetonitrile and water. Both components were fortified with 0.1% formic acid. Initial conditions were 50% acetonitrile/50% water, followed by a linear gradient to 95% acetonitrile at 4 min, held constant at 95% acetonitrile for 9 min, and then returned to original conditions for equilibration prior to the next injection.

The ANOVA resulted in a significant environment effect (Table 2). Though environments were significant, the authors pooled the years together and considered each year–location combination an environment sampled at random from a population as described by Carmer et al. (1989). Designating the environments random will broaden the possible inference space to which the experimental results are applicable (Carmer et al. 1989). The two-way interaction of surface-by-sampler height and surface-by-herbicide were significant. The main effects of herbicide and sampler height were also significant (Table 2). Given the numerical differences, the authors were not expecting the surface effect to be insignificant (0.1438), which indicates non significance.

The 10 studies were all conducted in late July or August of the three respective years (Table 1). Most application times were approximately 8 am, but two were in the early afternoon in 2019. This midsummer timeframe in Knoxville, TN, would coincide with the wheat stubble, along with environmental conditions favoring herbicide volatility.

As expected, more herbicide was detected with the sampler at the 0.3-m height (38.1 ng m⁻³) compared to the 1.3-m height (8.8 ng m⁻³). The 4× increase in herbicide at the lower height differences in relative humidity (86.8% compared with 74.9%) when averaged over the four green-plant studies with high relative humidity at the lower sampler height. There was no appreciable difference in average temperature in green plots. Temperatures were 2 degrees higher at the lower sampling height compared with the stubble vs green. The reason can be related to the green-plant vegetation's transpiration activities that have been documented for many years where the process reduces the canopy temperature (Blackman 1905). However, the green or stubble surfaces were examined in different experiments, so any conclusions are only speculative.

Given the 30-min time interval for a 24-h experiment, the environmental datasets are robust. A challenge the authors readily acknowledge is quantifying the relationship of the discontinuous environmental data, temperature and humidity, to the single finite point of an herbicide concentration measured from 0.5 to 24 h. There's no doubt that the environment is playing a major role in our observations, but the authors are uncertain how to best explain and possibly correlate our observations. All samplers had quantifiable herbicide detections at all sampling intervals, indicating adequate sensitivity of the field sampling and lab methods.

There was a two-way interaction between surface variable and height of the sampler (P < 0.0027) (Table 2). Applications made to the green-plant material showed significantly higher concentrations (ng m⁻³) of the 2,4-D formulations and dicamba at the 0.3-m sampler than the 1.3-m sampler (Table 3). However, for the wheat stubble surface there was no significant increase in herbicides detected between the two sampler heights. This may be due to greater surface area present in the green vegetation compared to the wheat stubble, along with environmental conditions favoring herbicide volatility.

| Surface condition | Sampler height m | Concentration ng m⁻³ | Value | Pr > F |
|-------------------|------------------|----------------------|-------|--------|
| Green plants      | 0.3              | 70.0                 | A*    |        |
|                   | 1.3              | 15.9                 | B     |        |
| Wheat stubble     | 0.3              | 15.9                 | B     |        |
|                   | 1.3              | 1.7                  |        |        |

*Means followed by a different letter are significant (P < 0.0027).

Results and Discussion
Therereferences, whereas processing of the PUF media resulted in major extraneous peaks and other operational difficulties.

| Effect                      | Denominator DF | F Value | Pr > F |
|-----------------------------|----------------|---------|--------|
| Environment                 | 48             | 9.40    | <0.0001|
| Herbicide                   | 44             | 6.61    | 0.0041 |
| Surface                     | 8              | 8.35    | 0.1438 |
| Sampler height              | 50             | 13.06   | 0.0005 |
| Height × herbicide          | 50             | 1.08    | 0.1727 |
| Surface × height            | 51             | 9.37    | 0.0027 |
| Surface × herbicide         | 44             | 5.43    | 0.0094 |
| Surface × herbicide × height| 47             | 1.50    | 0.2431 |

*Probability less than 0.05 indicates lack of significance for this effect.

**Table 2. ANOVA over all location and years (environments).**

**Table 3. Herbicide concentration as affected by a two-way interaction between surface and sampler height from 2018 and 2019 (years with both surface conditions).**
Table 4. Herbicide concentration means averaged over all years examining surface and herbicide from 2018 and 2019 (years with both surface conditions).

| Herbicide     | Surface condition | Concentration (ng m\(^{-3}\)) |
|---------------|-------------------|--------------------------------|
| 2,4-D amine   | Green plants      | 17.4 B*                        |
|               | Wheat stubble     | 2.7 B                          |
| 2,4-D choline | Green plants      | 28.9 B                         |
|               | Wheat stubble     | 3.3 B                          |
| Dicamba       | Green plants      | 82.4 A                         |
|               | Wheat stubble     | 28.9 B                         |

*Means followed by a different letter are significant (P < 0.0094).

Table 5. Herbicide concentrations as affected by formulation/active ingredient, sampler height, and surface condition from field studies in 2018 and 2019.

| Herbicide     | Sampler height | Surface condition | Concentration (ng m\(^{-3}\)) |
|---------------|----------------|-------------------|--------------------------------|
| 2,4-D amine   | 0.3            | Wheat stubble     | 4.5                            |
|               | 1.3            | Wheat stubble     | 1.1                            |
| 2,4-D amine   | 0.3            | Green plants      | 37.2                           |
| 2,4-D amine   | 1.3            | Green plants      | 10.9                           |
| 2,4-D choline | 0.3            | Wheat stubble     | 5.3                            |
| 2,4-D choline | 1.3            | Wheat stubble     | 1.4                            |
| 2,4-D choline | 0.3            | Green plants      | 56.9                           |
| 2,4-D choline | 1.3            | Green plants      | 15.5                           |
| Dicamba       | 0.3            | Wheat stubble     | 12.8                           |
| Dicamba       | 1.3            | Wheat stubble     | 2.8                            |
| Dicamba       | 0.3            | Green plants      | 136.7                          |
| Dicamba       | 1.3            | Green plants      | 46.4                           |

LSD 21.2

*Concentrations are in nanograms of herbicide normalized to measured airflow from 0.5 to 24 h after treatment.

was mostly based upon the dicamba treatment, although both 2,4-D formulations had more 2,4-D detected at the lower height.

There was a two-way interaction between the herbicides tested and whether they were applied to wheat stubble or green vegetation (Table 4). The driver for this interaction was the 82.4 ng m\(^{-3}\) of dicamba detected on the green surface compared to 28.9 ng m\(^{-3}\) on the wheat stubble surface. The 28.9 ng m\(^{-3}\) of detected dicamba was similar to what was detected for both 2,4-D formulations across both surfaces. These results would be consistent with Mueller and Steckel (2021), who reported that more dicamba emissions were detected from green plants than dead plants or tilled soil.

Herbicide amounts averaged over all factors were 13.4, 19.8, and 48.6 ng m\(^{-3}\) for 2,4-D amine, 2,4-D choline, and dicamba, respectively. There was no difference between the amine and choline formulations of 2,4-D when averaged over all experimental parameters (P = 0.5292). Sosnoskie et al. (2015) reported similar results with no differences between the amine and choline 2,4-D salts. Both 2,4-D formulations showed numerically lower concentrations than the dicamba treatment. The choline treatment was numerically higher than the amine formulation, which implied that either 2,4-D formulation could be used from a volatility perspective. The 2,4-D treatments having lower volatile emissions compared with dicamba is important, especially noting that the dicamba treatment did not include glyphosate in the spray mixture, which might have increased dicamba vapor emissions (Mueller and Steckel 2019). Concurrently, the dicamba treatment did not include volatility-reducing agents, nor did it contain a formulation prepared to reduce dicamba emissions under field conditions. The DGA dicamba formulation used could be applied prior to soybean or cotton emergence in many production systems.

There were few differences when the emissions data were analyzed across formulation, sampler height, and surface condition (Table 5). This was probably affected by the low number of replications that were necessitated by a small number of available air samplers. The dicamba concentration at the 0.3-sampler height in the green surface condition was greater than from all other treatments. The lowest herbicide concentrations were the 2,4-D amine, 2,4-D choline, and the dicamba treatments at the 1.3-m height in the dry-stubble surface condition (Table 5). The sampler height always had a numerically consistent effect with higher concentrations at the lower sampler depth.

In conclusion, based on this research there was no difference between the 2,4-D amine and the 2,4-D choline treatments from a volatility perspective. Moreover, dicamba was clearly more volatile than either the amine or choline salt 2,4-D formulations when applied on green plants. 2,4-D poses less potential off-target movement via vapors than dicamba under field conditions, although a DGA dicamba formulation was used in this research. Off-target movement under field conditions would not be expected to be affected by 2,4-D formulation within the examined choices of amine compared to choline. The low-volatile ester formulation of 2,4-D was not examined in this report but would be expected to have higher volatility and should not be used where sensitive nontarget vegetation could be affected (Grover 1976).

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