

Short Title: Off-Target Movement of Dicamba

Off-Target Movement Assessment of Dicamba in North America

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Abstract

Six experiments were conducted in 2018 on field sites located in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin to evaluate the off-target movement (OTM) of dicamba under field-scale conditions. The highest estimated dicamba injury in non-dicamba-resistant (DR) soybean was 50, 44, 39, 67, 15, and 44% injury for non-covered areas and 59, 5, 13, 42, 0, and 41% injury for covered areas during dicamba application in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin, respectively. The level of injury generally decreased

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exponentially as the downwind distance increased under covered and non-covered areas at all sites. There was an estimated 10% injury in non-DR soybean at 113, 8, 11, 8, and 8 m; and estimated 1% injury at 293, 28, 71, 15, and 19 m from the edge of treated field downwind when plants were not covered during dicamba application in Arkansas, Indiana, Michigan, Ontario and Wisconsin, respectively. Filter paper collectors placed from 4 up to 137 m downwind from the edge of the sprayed area suggested that the dicamba deposition reduced exponentially with distance. The greatest injury to non-DR soybean from dicamba OTM occurred at Nebraska and Arkansas (as far as 250 m). Non-DR soybean injury was greatest adjacent to the dicamba sprayed area but, injury decreased with no injury beyond 20 m downwind or any other direction from the dicamba sprayed area in Indiana, Michigan, Ontario, and Wisconsin. The presence of soybean injury under covered and non-covered areas during the spray period for primary drift suggests that secondary movement of dicamba was evident at five sites. Further research is needed to determine the exact forms of secondary movement of dicamba under different environmental conditions.

Nomenclature: Dicamba; soybean, *Glycine max* (L.) Merr.

Key Words: Crop injury, dicamba-resistant soybean, primary drift, secondary drift, sensitivity.

Introduction

Dicamba is a Group 4, benzoic acid herbicide that has been in use as an integral part of weed management programs in North America in corn (*Zea mays* L.) and cereals for over 50 years (Cao et al. 2011; Hartzler 2017). This herbicide was discovered in 1958 and was subsequently registered for annual, biennial, and perennial broadleaf weed control in 1962 in the United States

(Hartzler 2017). The rapid increase in the evolution of herbicide-resistant (HR) weeds in North America has resulted in renewed interest in dicamba (Behrens et al. 2007; Heap 2019). Development of transgenic HR crops, specifically glyphosate- and dicamba-resistant soybean (Xtend[®] technology, Bayer CropScience, St. Louis, MO) and cotton has provided a new weed management tool to control HR weeds (Byker et al. 2013). These crop cultivars have transgenes that confer resistance to glyphosate through an insensitive enolpyruvyl shikimate 3-phosphate synthase (CP4 EPSPS) enzyme and to dicamba through metabolism by dicamba monooxygenase (Byker et al. 2013). Dicamba is an efficacious, cost-effective, broad-spectrum, broadleaf herbicide with minimal risks to the environment (Shaner 2014). It is currently labelled for weed management in dicamba-resistant (DR) soybean, corn, DR cotton (*Gossypium hirsutum* L.), small grains, and pasturelands.

Research conducted in North America has shown that when timely and accurately applied, dicamba alone or in tank-mixtures with other herbicides can control key glyphosate-resistant (GR) broadleaf weeds such as Palmer amaranth (*Amaranthus palmeri* (L.) Wats.), waterhemp (*Amaranthus tuberculatus* L.), common ragweed (*Ambrosia artemisiifolia* L.), giant ragweed (*Ambrosia trifida* L.), and horseweed (*Conyza canadensis* (L.) Cronq.) (Byker et al. 2013; Johnson et al. 2010; Spaunhorst and Bradley 2013; Spaunhorst et al. 2014; Vink et al. 2012). For example, according to Nebraska (93%) and Wisconsin (66%) growers, weed management significantly improved with adoption of dicamba products in soybean (Werle et al. 2018).

In 2019, 22 million hectares of DR soybean were grown in the United States (Unglebee 2019). In 2017, 2018 and 2019 in eastern Canada 13, 31 and 50% of soybean fields were seeded to DR soybean cultivars, respectively (P.H. Sikkema, personal communication, August 8, 2019). The availability of DR crops and concomitant increase in the use of dicamba has increased the

potential for injury to sensitive plants in adjacent areas due to off-target movement (OTM) of dicamba (McCowan et al. 2018).

OTM can be related to droplet size and nozzle selection (particle drift), product formulation (vapor drift), and meteorological conditions (temperature, relative humidity, and wind speed). Particle drift occurs when droplets are carried by the wind during application away from the target area. Even the recommended nozzles for dicamba that produce extremely coarse and ultra-coarse droplets yield at least 1% fines (Bish et al. 2019a, b). Conversely, volatility occurs when the herbicide reaches the intended target, but due to the inherent high vapor pressure of the herbicide combined with certain meteorological conditions, the herbicide can volatilize. Another important factor affecting OTM is air temperature inversion, that is characterized by a cool air layer above the soil surface which limits vertical air mixing causing small suspended droplets/vapor to remain close to the ground and move laterally in a concentrated cloud. It generally occurs in the evening through the early morning during summer months (Bish et al. 2019a, b). It has been recommended that applicators should not spray pesticides during such conditions (Bish et al. 2019a, b). In a survey of Nebraska growers, 69% reported that the main cause of dicamba injury in neighboring non-DT soybean fields was due to volatilization, 23% physical drift, and 8% temperature inversion (Werle et al. 2018).

In 2017, two new formulations of dicamba were registered for use in DR soybean and cotton in North America, which included XtendiMax[®] (Anonymous 2018a) or FeXapan[®] (Anonymous 2018b) with VaporGrip technology from Monsanto and Dupont, respectively, and Engenia[®] (Anonymous 2019a) from BASF. In 2019, Syngenta registered Tavium[™] for use in soybean and cotton, which includes S-metolachlor for residual weed control and dicamba with VaporGrip technology (Anonymous 2019b). XtendiMax[®], FeXapan[®], and Tavium[™] are all diglycolamine

(DGA) salts and have been engineered to have reduced volatility. Engenia[®] is a N,N-Bis-(3-aminopropyl) methylamine (BAPMA) salt that has reduced volatility risk by strengthening the bond between dicamba acid and base within the formulation (Anonymous 2019a). The above formulations are reported to reduce the formation of dicamba acid and therefore dicamba volatilization (Anonymous 2018a, b; 2019 a, b). To further reduce injury to sensitive plants due to OTM of dicamba, numerous restrictions have been added to the dicamba labels including nozzle type, approved mixtures, exclusion of ammonium sulfate, carrier volume, boom height, application speed, wind speed and direction, and buffer zones.

Research in Arkansas, Missouri, Tennessee, Nebraska, and Indiana has reported that under certain environmental conditions, these new formulations of dicamba can still volatilize and move to non-target areas, even when applied according to the manufacturers' recommendations (Jones et al. 2019; Norsworthy et al. 2018). A national survey conducted by the University of Missouri in 2017 reported soybean injury on 1.3 million hectares in the United States (Bradley 2017). Complaints about extensive injury in non-target crops by growers prompted Missouri and Arkansas to regulate dicamba sales in those states in 2017 (Gray 2017). These concerns have also prompted the Environmental Protection Agency (EPA) to announce additional restrictions for continued use of dicamba in the United States (EPA 2018a).

Therefore, the objective of this research was to (1) quantify the amount of dicamba due to primary (particle drift) and secondary movement (particle + vapor drift) from applications made in different environmental conditions, geographies and/or landscapes; (2) and evaluate the effects of primary and secondary movement of dicamba on symptomology of non-DR soybean adjacent of sprayed areas located at six different regions in North America.

Materials and Methods

Experimental Methods

Six experiments were conducted on field sites located in Arkansas (Farm, Proctor, AR 35° 06' 50.16" N, 90° 22' 2.94" W), Indiana (Farm, Montezuma, IN 39° 47' 34" N / 87° 22' 15" W), Michigan (Farm, Fowlerville, MI 42° 39' 19.08" N, 84° 1' 6.96" W), Nebraska (Burnside Farm Co., Stapleton, NE 41° 25' 36.09" N, 100° 29' 39.82" W), Ontario (Farm, Dresden, ON 42° 38' 31.398" N, 82° 11' 34.1628" W) and Wisconsin (Farm, Arlington, WI 43° 19' 32.6" N, 89° 19' 43.3" W) during the 2018 growing season (Figure 1). The treated area (hectareage for each site is listed in Table 1) was planted to DR soybean while the surrounding area was planted to a glyphosate-resistant (GR) non-DR soybean cultivar of similar maturity group. Applications were made when plants were at V3 growth stage (Fehr and Caviness 1977) in Michigan, Nebraska, and Wisconsin, and at R1 growth stage in Arkansas, Indiana and Ontario. A tank-mixture of dicamba at 613 g ae ha⁻¹ (XtendiMax[®] with VaporGrip[®] Technology, Bayer Co., St. Louis, MO) plus glyphosate at 1,334 g ae ha⁻¹ (Roundup PowerMax[®], Bayer Co., St. Louis, MO) plus drift-reducing adjuvant at 0.5% v v⁻¹ (Intact[™], Precision Laboratories, Waukengan, IL) was applied in an application volume of 140 L ha⁻¹. Additionally, acetochlor at 1,050 g ai ha⁻¹ (Warrant[®], Bayer CropScience, St. Louis, MO) was added to the dicamba mixture in Arkansas for residual control of Palmer amaranth. This mixture is permitted per the XtendiMax[®] label. Attempts were made to follow the label, but the application conditions used represent the normal use pattern for dicamba (Table 1).

Environmental conditions (wind speed, wind direction, air temperature, and relative humidity) at each site were collected using weather stations positioned outside of the sprayed area (Table 2; Figure 2, 3). In IN, NE, ON, and WI, the sensors were positioned at 0.33, 0.56,

0.89, and 1.50 m above the crop canopy. In AR and MI, temperature and relative humidity sensors were positioned only at 1.50 m above canopy. Conditions were recorded during the application time until the drift sampling was completed.

Spray Particle Drift Evaluation

Before the applications, 125 mm diameter filter papers (Whatman no. 1, Whatman, Maidstone, UK) were attached to a 15 x 15 cm cardboard sheet placed horizontally at the soybean canopy height outside of the treated area just prior to herbicide application to determine particle drift. The filter papers were collected 30 min after application and placed in individual 50-mL centrifuge tubes (Sarstedt AG & Co., Nümbrecht, DEU). Samples were stored in coolers containing dry ice until transfer to storage at -20°C prior to analysis.

Downwind and Upwind Samples

Filter papers were placed at several downwind distances from the field line (sprayed area). The field line was defined as the edge of the sprayed area from the furthest downwind nozzle on the boom. The distances were 15, 30, 46, 61, 76, 91, 107, 122, 137 m (Arkansas); 4, 7, 16, 30, 45, 60, 75, 90 m (Indiana); 4, 8, 16, 31, 45, 60, 75, 90, 105, 120 m (Michigan); 4, 8, 16, 31, 45, 60, 75, 90, 105 m (Nebraska and Ontario); and 4, 8, 16, 31, 45 m (Wisconsin). Three lines of sample collectors were used at each site (except Nebraska where two lines were used) spaced 15 m apart as appropriate for the test site and local landscape, with the center line located at the midpoint of the sprayed area. Additionally, three filter papers were placed at 30 m from the upwind edge of the application area at all sites. Upwind samples were collected by a person that did not previously collect samples from the downwind deposition area to avoid any cross contamination (Figure 1).

Field Air Samples

A set of air pumps (SKC Inc., AirChek 224-52, Eighty-Four, PA, USA) with rechargeable batteries (Anker Innovations, Powercore+ 20100 USB-C, Shenzhen, Guangdong, CHN) and polyurethane foam (PUF) (SKC Inc., Cat. no. 226-92, Eighty-Four, PA, USA) were positioned on a horizontal stand at each height. Batteries continuously used for 48 hours were replaced for charged one. The airflow rate of air pumps and PUFs were calibrated between 2.9 and 3.1 L min⁻¹ (Check-mate Calibrator, SKC Inc., Eighty-Four, PA, USA). Samples were collected, placed in uniquely labeled screw cap tube containers (Sarstedt AG & Co., Nümbrecht, DEU), and stored in coolers containing dry ice until transfer to storage at -20°C prior to analysis.

Pre-application Samples

Two pre-application air samples were collected at 0.56 m above canopy level using air sampling equipment placed near the center of the sprayed area. The samples were collected within 24 h prior to the dicamba application. The pre-application air monitoring event lasted for approximately 6 h. These samples were used to determine the background level of dicamba in the air prior to study initiation.

Post-Application Samples

A mast was erected in the middle of the sprayed area and air samplers were positioned at 0.33, 0.56, 0.89, and 1.50 m above the crop canopy. Sampling periods in hours after application (HAA) were 0.5-4.5, 4.5-17.5, 17.5-28.5, 28.5-41.5, 41.5-52, 52-64.5 HAA in Arkansas, 0.5-6, 6-18, 18-29.5, 29.5-41.5, 41.5-54, 54-65.5, 65.5-78 HAA in Indiana, 0.5-4, 4-8, 8-20, 20-32, 32-44, 44-56, 56-68 HAA in Michigan, 0.5-5, 5-18, 18-30, 30-42, 42-54, 54-66 HAA in Ontario, 0.5-10.5, 10.5-22.5, 22.5-34.5, 34.5-46.5, 46.5-56.5 HAA in Nebraska, and 0.5-4, 4-8, 8-20.5, 20.5-32, 32-44, 44-55 HAA in Wisconsin.

Sample Analysis

All samples were shipped overnight in coolers containing dry ice at -20°C to Mississippi State University Laboratory (Mississippi State Chemical Laboratory, Mississippi State, MS, USA) for analysis. The dicamba was extracted using 30 mL of methanol containing $^{13}\text{C}_6$ labeled dicamba (CAS #: 1173023-06-7, Sigma Aldrich, St. Louis, MO, USA) as an internal standard. The PUF samples were homogenized with a SPEX SamplePrep Geno/Ginder® (OPS-Diagnostics, Lebanon, NJ). The supernatant was concentrated with a TurboVap to 1 mL, filtered, evaporated, and solvent exchanged to an appropriate volume of 25% acetonitrile in water solution so that the samples were concentrated 50X. Quality control samples included a blank matrix sample (PUF or Filter) that was devoid of dicamba and a spiked matrix sample that was fortified with a known concentration of dicamba. The spiked matrix sample was used to determine the efficiency of the extraction for every batch: recoveries ranged between 80 and 120%, and the level of detection for PUFs and filters were both 3 ng/PUF or filter. All samples were carefully managed in a manner to avoid the potential for cross contamination and stored at -20°C until analysis.

LC-MS/MS Method

The dicamba was quantitated using an Agilent 1290 liquid chromatograph coupled with an Agilent 6460 C triple quadrupole mass spectrometer (Agilent Technologies, Santa Clara, CA). Chromatographic separation was performed using an Agilent Zorbax Eclipse Plus 100 mm column. The mobile phases consisted of 0.1% formic acid in water for the aqueous phase (A) and 0.1% formic acid in acetonitrile as the organic phase. The flow rate 0.3 mL/min with the following gradient program: 0 to 0.5 min of 25% B, 0.5 to 1 min of 50% B, and 1 to 4 min of 60% B. The ionization of dicamba was performed using electrospray ionization (ESI) in negative mode with an auxiliary gas (N_2), source temperature of 200°C , and a gas flow rate of 10 L m^{-1} .

Plant Effects

OTM on non-DR soybean was assessed with visual estimation of injury along covered and non-covered transects downwind and perpendicular to the sprayed area. Covered and non-covered plants were rated starting at 15.2 m and every 15.5 m out to 259 m (Arkansas), 1.5 m and every 1.5 m out to 13.5 m (Indiana), 0.8 m and every 0.8 m out to 15.2 m (Michigan), 4.6 m and every 1.5 m out to 15.2 m (Nebraska), 5, 10, 15, 20, 30, 45, 60, 75, and 90 m (Ontario), and 0.5 m and every 0.5 m out to 10 m (Wisconsin). Plants were covered just before the applications using tarps with dimensions of 16 x 3 x 1.5 m elevated off the soybean canopy by a PVC pipe frame in all sites except Arkansas (Figure 1). In Arkansas, the tarps were 7.6 x 3 x 1.5 m beginning 3 m from the sprayed area in three downwind transects. The tarp at this site was rested on the plants to ensure no risk for physical drift. In Arkansas, 19 L buckets covering 3 non-DR soybean plants were used similar to that used in other dicamba research (Jones et al. 2019).

Plant injury ratings were collected at 28 days after application (DAA) in Wisconsin and 21 DAA in all other sites. Three soybean plants at each distance in the covered and non-covered areas were randomly selected and visually rated on a 0 to 100 scale, with 0 representing no crop injury and 100 representing complete plant death.

In Arkansas, the periphery of 5% injury was mapped using a global positioning unit at 21 DAA. The area on non-DR soybean injured by dicamba to a 5% or greater level was determined in Google Earth (Google, Mountain View, CA, USA).

Statistical Analysis

The three-parameter log-logistic model (*drm* function) of the *drc* package in R statistical software (Ritz et al. 2015) was fitted to the dataset of dicamba injury (%) and dicamba deposition (ng filter⁻¹) on non-DR soybean:

$$Y(x) = \frac{d}{1 + \exp(b(\log(x) - e))} \quad \text{Equation 1}$$

where Y is the Non-DT soybean injury (%) or dicamba deposition ($\eta\text{g cm}^{-2}$), d is the upper limit of Y , and e (inflection point) represents 50% Y reduction relative to d . The parameter b is the relative slope around the e , and x is the distance (m) from the DR soybean treated area. This was the top model based on log likelihood of the function *mselect* in the *drc* package of R software. The *drc* package *ED* function estimated the distance from the dicamba application block area that caused 1% (D_1), 10% (D_{10}) and 20% (D_{20}) dicamba injury (%) on non-DR soybean plants (Ritz et al. 2015).

Model goodness of fit. Root mean squared error (RMSE) and modelling efficiency (ME) were calculated and used to test the goodness-of-fit of three-parameter log-logistic and linear models (Ritz and Streibig, 2008; Mayer and Butler, 1993):

$$RMSE = \sqrt{\frac{RSS}{n-p-1}} \quad \text{Equation 3}$$

$$ME = 1 - \left[\frac{\sum_{i=1}^n (O_i - P_i)^2}{\sum_{i=1}^n (O_i - \bar{O}_i)^2} \right] \quad \text{Equation 4}$$

where the RSS is the residual sums of squares; n is the number of data points; p is the number of model parameters; O_i is the observed, P_i is the predicted and \bar{O}_i is the mean observed value. The ME values range from $-\infty$ and 1, with values closer to 1 indicating better predictions.

Flux Calculations

The dicamba flux of each field site was calculated using the Aerodynamic (AD) and Integrated Horizontal Flux (IHF) Methods as recommended by EPA at Guideline OCSPP 835.8100 (EPA 2018b). Calculations were made using Excel 2016 worksheets (Microsoft Corporation, Redmond, WA, USA) as provided by EPA (EPA 2018b). The AD method requires a minimum fetch of 100 times greater than the highest height of the air sampler, whereas the IHF method requires a minimum fetch of 20 m (Majewski et al. 1990).

Aerodynamic Method

The dicamba flux was calculated according to the Equations 5 and 6 (Majewski et al. 1990):

$$P = \frac{-(0.42)^2(c_{ztop}-c_{zbottom})(u_{ztop}-u_{zbottom})}{\Phi_m\Phi_p[\ln(\frac{z_2}{z_1})]^2} \quad \text{Equation 5}$$

where P is the flux ($\mu\text{g m}^{-2}\cdot\text{s}^{-1}$), c_{ztop} ($\mu\text{g m}^{-3}$) is the concentration at the top sampler adjusted according to the regression of concentration vs. \ln (height), $c_{zbottom}$ ($\mu\text{g m}^{-3}$) is the concentration at the bottom sampler adjusted according to the regression of concentration vs. \ln (height), u_{ztop} (m s^{-1}) is the wind speed at the top sampler adjusted according to the regression of wind speed vs. \ln (height), $u_{zbottom}$ (m s^{-1}) is the wind speed at the bottom sampler adjusted according to the regression of wind speed vs. \ln (height), Φ_m and Φ_p (dimensionless) are the Internal Boundary

Layer (IBL) stability correction terms determined according to the following conditions based on the calculation of the Richardson number, R_i :

$$R_i = \frac{(9.8)(c_{ztop} - c_{zbottom})(T_{ztop} - T_{zbottom})}{\left[\left(\frac{T_{ztop} + T_{zbottom}}{2}\right) + 273.16\right] + (u_{ztop} - u_{zbottom})^2} \quad \text{Equation 6}$$

where R_i (dimensionless) is Richardson number, T_{ztop} ($^{\circ}\text{C}$) is the temperature at the top sampler adjusted according to the regression of temperature vs. \ln (height), $T_{zbottom}$ ($^{\circ}\text{C}$) is the temperature at the bottom sampler adjusted according to the regression of temperature vs. \ln (height).

If $R_i > 0$ (for Stagnant/Stable IBL):

$$\Phi_m = (1 + 16R_i)^{0.33} \text{ and } \Phi_p = 0.885 (1 + 34R_i)^{0.4}$$

If $R_i < 0$ (for Convective/Unstable IBL):

$$\Phi_m = (1 - 16R_i)^{-0.33} \text{ and } \Phi_p = 0.885 (1 - 22R_i)^{-0.4}$$

Integrated Horizontal Flux Method

The dicamba flux was calculated according to the Equations 7 and 8 (Majewski et al. 1990):

$$P = \frac{1}{x} \sum_{z_0}^{z_p} (A - \ln(z) + B) * (C * \ln(z) + D) dz \quad \text{Equation 7}$$

where P is the flux ($\mu\text{g m}^{-2} \cdot \text{s}^{-1}$), z (m) is the height above ground level, A is the slope of the wind speed regression line by $\ln(z)$, B is the intercept of the wind speed regression line by $\ln(z)$, C is the slope of the concentration regression by $\ln(z)$, D is the intercept of the concentration regression by $\ln(z)$. Z_p was determined using the Equation 8:

$$Z_p = \exp \left[\frac{(0.1 - D)}{C} \right] \quad \text{Equation 8}$$

Results and Discussion

Arkansas

The greatest occurrence of injury to non-DR soybean from OTM of dicamba occurred in Arkansas. After waiting 6 d at the field site because of insufficient winds to make a labeled application, the dicamba-containing mixture was applied beginning at 2:58 pm on July 16, 2018, with the entire application requiring approximately 45 min. A fire started adjacent to the field indicated there was no inversion present during application based on rapid dispersion of smoke. Meteorological data collected at a 0.33 and 1.50 m height several kilometers from the test site indicated absence of an inversion during application. Air temperature, relative humidity, and wind speed at boom height during application were 32°C, 66.5%, and 1.3 m s⁻¹, respectively. Wind during the application was predominantly from the west/northwest direction but following application wind was from all 360 degrees of the treated field over the next 24 h (Figure 2).

OTM at the test site was predominately in the form of secondary drift, based on 1) a similar level of injury for covered and non-covered soybean plants (Table 3; Figure 4), 2) the upper limit of dicamba deposited downwind being only 60 ng filter paper⁻¹ (Table 4; Figure 5), and 3) the presence of damaged soybean on all four sides of the field. By 22 days after application, soybean covered with tarps during application on all sides of the treated field were injured at least 40%, with a similar level of injury for covered and non-covered plants (Supplemental Table 1). The average wind speed of 1.3 m s⁻¹ at height of the boom during application contributed to less physical drift of dicamba during application compared with other sites, except Indiana (Table 4).

An estimated 10% injury to soybean occurred at 84 m from the treated field when plants were covered during application, and 1% injury to soybean was estimated beyond the field edge

(approximately 250 m) for covered and non-covered plants (Table 3). Furthermore, at 22 days after application, 24.0 ha of non-DR soybean exhibited 5% or more injury from dicamba when only 15.6 ha of DR soybean were treated (Supplemental Table 1). Hence, 1.5x the treated area was injured at least 5% by the dicamba application. A logical question becomes “why the extensive movement in all directions and greater injury in Arkansas than at other sites?”

It is well established that air temperature directly influences dicamba volatility (Behrens and Leuschen 1979), and volatilization of the dicamba formulation tested in this experiment increases substantially at temperatures above 30°C (Mueller and Steckel 2019a). Temperatures during and following treatment frequently exceeded 30°C at this test site over a three-day period. However, high temperatures alone do not adequately explain the extensive injury at the Arkansas site, especially considering post-application daily temperatures exceeded 30°C at Indiana and Nebraska. Slow moving stable air in combination with high temperatures sufficient for dicamba volatility following application may need consideration (Bish et al. 2019b). Furthermore, the addition of glyphosate to dicamba may have also contributed to secondary movement of the auxin herbicide, depending upon pH of the spray solution. The addition of the potassium salt of glyphosate to the dicamba formulation applied in these field trials results in a pH drop of 1.0 to 2.1 units, depending on the water source (Mueller and Steckel 2019b), which can cause the volatility of dicamba to more than double (Mueller and Steckel 2019a). Unfortunately, the pH of the water source and spray solution was not measured.

Another factor that may need consideration is the presence or absence of dew following exposure. The presence of dew and rewetting of dicamba on leaves at the Arkansas site is unknown, but other research on metolachlor has found gaseous losses to increase when the application surface (soil in this instance) was moist during application (Prueger et al. 2017). Dew

is a common occurrence in the mid-southern United States during the summer months and it may be possible that rewetting of soybean leaves following previous exposure facilitates conversion of the dicamba salt to its respective acid form.

The Integrated Horizontal Flux method was a poor indicator of the risk for injury to soybean from dicamba based on the flux being lowest in Arkansas, yet the most extensive injury was observed at this location (Figure 6). These results are not surprising considering recent research in the mid-southern United States has found volatile dicamba concentrations as low as $1 \text{ ng m}^{-3} \text{ d}^{-1}$ to be sufficient to cause symptomology in soybean (Brabham et al. 2019), a level much lower than that once thought to elicit injury to the crop.

Indiana

Dicamba was applied to soybean on August 9, 2018 at 1:00 pm, 38 days after planting. The average wind speed during the herbicide application was 0.6 m s^{-1} at the height of the spray boom, which is below the label requirement of sustaining average wind speeds between 1.3 and 4.5 m s^{-1} (Table 2). However, wind gusts up to 3.1 m s^{-1} were observed during the application (Figure 2). In addition, a smoke bomb was released just prior to the herbicide application and provided a visual confirmation that an air temperature inversion did not exist.

Less soybean injury was observed for sensitive soybean that were protected from primary drift (plastic cover) during the application compared with plants that had no cover (Figure 4). The difference in soybean injury with and without the plastic cover during the period for primary drift indicates that secondary herbicide movement of dicamba was evident. However, secondary movement of dicamba was relatively minor compared with the primary drift at this site. Soybean injury along the transects reached beyond 30 m downwind from the application (Supplemental

Table 1) and was associated with dicamba deposition on the filter paper in amounts greater than 15 ng (Figure 5). No soybean injury or deposition of dicamba on filter paper was observed for any upwind samples (Supplemental Table 1 and 2). Data and subsequent modeling of the air samples for dicamba describe very little secondary movement of dicamba (Figure 6) which supports the soybean injury data.

Michigan

Dicamba was applied to soybean on June 12, 2018 between 10:18 and 11:00 am, 37 days after planting. At the time of application, average air temperature was 19°C, relative humidity was 76.5%, and average windspeed was 1.6 m s⁻¹, which was in the label wind speed range of 1.3 and 4.5 m s⁻¹ (Table 2). Wind speeds did not fall below or exceed label recommendations within the 10 h following application (Figure 2).

To account for the SE wind direction during application, downwind tarps and transects were placed on the W and N sides of the sprayed area (Figure 1). Greater dicamba injury to soybean occurred on the W side (Tarp A). Maximum injury from primary dicamba drift (non-covered) to soybean was 60%, 3 m downwind outside of Tarp A (Supplemental Table 1). Soybean injury levels declined as the downwind distance increased, however there was still 15% injury 15.2 m downwind. Less soybean injury occurred on the N side of the sprayed area, accounting for overall less injury from the combined downwind transects (Figure 4). Primary dicamba drift plus secondary movement, averaged across the measurements outside the three tarps, caused at least 10% soybean injury 11 m (± 3 m) downwind (Table 3).

Secondary dicamba movement was also detected on soybean that were kept covered during and up to 1 h after the application. Soybean injury was as much as 13% ($\pm 1\%$), and injury at 1%

could be detected as far out as 7 m downwind (Table 3; Figure 4). Secondary dicamba movement to a much lesser extent also occurred on the upwind side of the sprayed area. At one of the three transects, 5% soybean injury was detected at the 8 m transect on the E side (Supplemental Table 1). No injury was detected 30 m away from the sprayed area or at any distance on the S side. Whereas, soybean injury (3%) was found on the E side where tarps were placed at about 10 m, during and up to 1 h after dicamba application. Soybean injury on the upwind side was only apparent 21 DAT.

Higher amounts of dicamba deposition in the downwind direction closely followed soybean injury. The maximum amount of dicamba detected was 1180 ng filter⁻¹, 4 m away from the sprayed area on the W side (Transect 1) (Supplemental Table 2). Maximum dicamba deposition on the N side was 14.9 and 275 ng filter⁻¹, for Transects 2 and 3, respectively. Dicamba deposition was reduced with distance from the edge of the application area, even though there was as much as 15 ng of dicamba filter⁻¹ detected as far as 120 m away from the application area. The 50% reduction of dicamba deposition downwind was estimated to be 2 m (± 1 m) from the edge of the application area (Table 4; Figure 5). In comparing dicamba deposition to soybean injury, it appears that a minimum of 50 ng filter⁻¹ of dicamba may be needed to elicit a response in soybean.

Similar to other locations, the highest amount of dicamba detected using the PUFs was within the first 6 h after application (Figure 6). Flux calculations followed a diurnal cycle with higher dicamba fluxes during the day and lower fluxes at night. No additional dicamba was captured at the Michigan location at 48 HAA.

Nebraska

Dicamba was applied to soybean on July 10, 2018 at 9:30 am, 46 days after planting. During the application, the air temperature, relative humidity, and wind speed were 23°C, 80%, and 3.2 m s⁻¹, respectively. In Nebraska, wind speed was up to 4.5-fold higher than the other sites, especially Indiana (4.5-fold) and Arkansas (3.2-fold). During the air samplings after application, temperature data suggests that inversions occurred overnight with stronger gradients in the two nights following the application.

Visible injury on non-DR soybeans that were either covered or non-covered during the application was observed at 21 DAA, showing injury levels up to 50% on covered and up to 75% on non-covered plants (Figure 4). These results suggest that secondary movement of dicamba occurred causing injury on soybean under the tarps, probably due to the high temperatures (over 30°C) and suggested temperature inversions on the next two days after application. These outcomes would agree with Mueller and Steckel (2019a), who reported that temperature appears to be a major contributor of dicamba secondary movement, with greater dicamba detections in the air at higher temperatures. The injury level decreased as the downwind distance increased, reaching 31 and 50% of injury on covered and non-covered plants, respectively, at 15 m from the sprayed area.

Higher amounts of dicamba on filter papers were detected in Nebraska when compared to the other sites (Figure 5), which may be explained by the higher wind speed during the application. The amount of dicamba detected on filter papers decreased exponentially as downwind distance increased, with greater slope up to 15 m where approximately 1514 ng filter⁻¹ of dicamba were detected, which resulted in 50% of injury on non-DR soybean.

Although a higher amount of dicamba was detected using the PUFs in Nebraska when compared with some of the other sites (Figure 6), flux calculations from all sites had a similar tendency for detecting greater dicamba flux during the days and lower flux during the nights. Higher amounts of dicamba detected during the day is probably due to higher air temperatures and wind speeds and lower air relative humidity compared with night conditions. Interestingly, dicamba was detected in the air samples up to 56 h after application, not only in Nebraska but also in Indiana and Wisconsin. Although very fine droplets may remain suspended in the air under low wind speed conditions (Miller and Stoughton 2000), it is unlikely that those droplets remained suspended in the air because calm wind speed conditions had frequency of 0 to 2% (Figure 2), suggesting that other sources of secondary movement such as vapor and dust may be associated to the results.

These results may help to understand the reasons for having 6,164 ha out of a 46,515 ha (13% of survey respondents) of non-DR soybean injured by dicamba in 2017 in NE according to Werle et al. (2018). The authors attributed that the primary suspected causes would be tank contamination, application during a temperature inversion, and/or secondary movement. Indeed, secondary movement happened (otherwise no injury should have been observed on soybean under the tarps); however, primary movement was also important for resulting up to 25% more injury on non-covered soybeans when compared with covered soybeans. Regardless of cause, farmers and applicators should be cautious of nearby sensitive crops and weather conditions during and up to 56 h after dicamba applications in order to mitigate spray drift and its consequences.

Ontario

In Ontario, the application was made to soybean on July 25, 2018 between 10:45 to 11:30 am (Table 2). At the time of application, the temperature was 25°C with a relative humidity of 61% and wind speed of 5.4 km h⁻¹ (1.5 m s⁻¹) from the north/northwest.

Dicamba levels in the air was below levels to cause any injury symptomology outside of the sprayed area (Figures 4 and 6). Dicamba captured in the air by PUFs occurs mostly during the daytime periods with warmer temperatures and lower relative humidity. There was minimal amount of dicamba captured in the air sample during the night time, except at 24-hr. There was an increase in dicamba concentration in the air from the upwind PUFs at 36 h after dicamba application which the authors attributed to the strong southwest winds (Figure 2).

There were no dicamba injury symptoms in the sensitive soybean in upwind areas under tarped or non-tarped areas (Supplemental Table 1). There were no dicamba injury symptoms in the sensitive soybean in downwind direction under the tarped areas, there was dicamba injury symptoms in the sensitive soybean in the non-tarped areas after dicamba application (Table 3; Figure 4). There was 525 ng filter⁻¹ of dicamba on filter papers placed downwind adjacent to the dicamba application block (Table 4). The dicamba deposition was reduced with distance from the edge of the application block. The 50% reduction of dicamba deposition downwind was estimated to be 7 m from the edge of dicamba application block (Table 4). At 4 m downwind from the sprayed area, dicamba caused 15% (± 1) soybean injury. However, at 16 m downwind from the sprayed area, dicamba did not cause any soybean injury (Supplemental Table 1).

Based on dicamba concentrations from the filter paper and PUF samples, the amount of dicamba captured was minimal and correlates with injury expected in dicamba-sensitive soybean. Dicamba was still detectable in some PUFs samples 57 h after application. However,

there was no detectable dicamba in the PUFs at 69 h after application. As expected, soybean injury was greatest adjacent to the dicamba sprayed area but, soybean injury decreased rapidly with no injury beyond 20 m downwind or any other direction from the dicamba sprayed area.

Wisconsin

Dicamba was applied to soybean during the morning of July 11, 2018. At the time of application (11:00 AM), wind speed was 1.4 m s^{-1} out of south, air temperature at 25°C and 47% relative humidity. Application was completed within 30 min. The dicamba application was made following the label directions to reduce dicamba OTM. Smoke bombs were used to indicate wind direction and no temperature inversion during application (rapid dispersion of smoke).

At 28 DAT, dicamba injury on non-DR soybean was observed in the north side (downwind) of the DR soybean block, and no injury was observed in the south side (upwind; Supplemental Table 1). Despite covering the non-DR soybean area during and until an hour after dicamba application, injury occurred downwind in both cover and no cover areas of non-DR soybean in Wisconsin. The highest dicamba injury on non-DR soybean was 44% (± 1) and 41% (± 1) injury without and with cover, respectively (Table 3). The dicamba injury rapidly decreased as distance from the dicamba application area increased (Figure 4). Minimal to no visible injury was observed on non-DR soybean after 19 m downwind. Dicamba application in Wisconsin happened before both the DR and non-DR soybeans reached the reproductive stage. According to Kniss (2018) meta-analysis investigating soybean response to dicamba exposure, the likelihood for non-DT soybean injury is higher when the crop is exposed to dicamba at flowering stages (R1 to R2) when compared to vegetative stages (V1 to V7).

The dicamba deposition reduced with distance from the application block (Figure 5; Table 4). A $1,684 \text{ ng filter}^{-1}$ of dicamba was detected adjacent to the dicamba application block (Table

4). The 50% reduction of dicamba deposition was estimated at 0.6 m from the dicamba application block. Similar to other locations, the highest amount of dicamba detected using the PUFs was detected within the first 6 h after application (Figure 6). Flux estimations followed a diurnal cycle with higher dicamba concentrations measured during the day and lower concentrations at night. Dicamba was still detected up to 56 h after application in Wisconsin.

Dicamba injury on non-DT soybean in the downwind direction in Wisconsin was likely caused by both primary and secondary dicamba OTM. Calm winds blew towards the same direction for 72 h after application at this site, likely carrying secondary dicamba vapor/particles towards the downwind direction.

In summary, the greatest injury to non-DR soybean from OTM of dicamba occurred in Nebraska and Arkansas followed by Michigan, Wisconsin, Indiana and then Ontario. The level of injury generally decreased as the downwind distance increased under covered and non-covered plants at all sites. The highest estimated dicamba injury on non-DR soybean was 50, 44, 39, 67, 15, and 44% injury for non-covered areas and 59, 5, 13, 42, 0, and 41% injury for covered areas in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin, respectively. The upper limit of dicamba deposition on non-DR soybean plants (based on dicamba deposition on filter paper placed downwind adjacent to the dicamba application block) was 60; 31; 4,931; 20,471; 525; and 1,684 ng filter paper⁻¹ in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin, respectively. The dicamba deposition diminished with distance from the application block.

Non-DR soybean injury was greatest adjacent to the dicamba sprayed area; the soybean injury due to OTM of dicamba rapidly decreased as distance from the dicamba application area increased. There was no injury beyond 20 m downwind or any other direction from the dicamba

sprayed area in Indiana, Michigan, Ontario, and Wisconsin. This study cannot conclude that all soybean injury was solely the result of primary drift. Soybean injury was sometimes evident in directions that was not always downwind from the application. The difference in soybean injury with and without the plastic cover during the spray period for primary drift (particle) also indicates that secondary herbicide movement of dicamba (particle + vapor) happened at five out of six locations. Temperature seems to be an important factor in dicamba behavior under some field conditions. Slow moving stable air in combination with high temperatures sufficient for dicamba volatilization following application may contribute to off-site movement. Further research is needed to determine the exact mechanism of secondary movement of dicamba when applied according to the manufacturer's labelled directions. This research reemphasizes the importance of maintaining appropriate stewardship to avoid injury to sensitive plants and crops besides soybean and preserve the use of dicamba for weed management in North America.

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Table 1. Field and application information for experiments conducted in six different locations to evaluate dicamba off-target movement (OTM) during the 2018 growing season.

Location	Sprayed area	Nozzle type	Carrier volume	Boom height	Boom width	Travel speed	Nozzle spacing	Sprayer	Variety 1 – DR ^a	Variety 2 - non-DR ^a	Planting date	Soybean population	Row spacing	Soil type	Soil pH
	ha		L ha ⁻¹	m	m	m s ⁻¹	m					seeds ha ⁻¹	m		
Arkansas	15.6	UR 11006	140	0.6	30.5	5.4	0.51	Case 3230	AG47X6	P47T89RR	6/1/2018	345,935	0.96	Sharkey silty clay	6.3
Indiana	8.1	TTI 11004	140	0.6	30.5	5.4	0.51	AGCO Rogator	Channel 3417R2X	Channel 3509R2	7/1/2018	456,950	0.19	Fox loam	6.2
Michigan	21.4	TTI 11004	140	0.6	36.6	3.6	0.51	JD 4930	AG26X8	AG4034RR2Y	5/6/2018	345,935	0.76	Loam/sandy loam	6.8
Nebraska	12.1	TTI 11004	140	0.6	36.9	4.7	0.38	JD R4038	AG24XYRR2X	AG2431RR2Y	5/25/2018	444,773	0.25	Holdrege silt loam	6.1
Ontario	16.9	TTI 11004	140	0.5	30.5	3.6	0.51	JD R4045	P21A28X	P22T69R	6/1/2018	457,128	0.38	Sandy loam	6.8
Wisconsin	2.8	TTI 11004	140	0.6	13.7	2.9	0.51	Demco	AG21X7	AG2035	6/5/2018	345,935	0.76	Plano silt loam	6.7

^a DR: dicamba-resistant

Table 2. Meteorological data during dicamba applications in six different locations in 2018 growing season.^a

Location	Application date and time	Soybean growth stage ^b	Meteorological data during application			
			Air temperature C	Relative humidity %	Wind speed ^c m s ⁻¹	Wind direction degrees
Arkansas	07/16/2018; 2:58 pm	R1	33.1 ± 0.4	65.2 ± 1.2	1.0 ± 0.5	221 ± 43
Indiana	08/09/2018; 1:00 pm	R1	29.2 ± 0.1	64.3 ± 0.2	0.6 ± 0.1	182 ± 13
Michigan	06/12/2018; 10:30 am	V3	19.0 ± 0.5	76.5 ± 1.5	1.6 ± 0.3	94 ± 18
Nebraska	07/10/2018; 09:30 am	V3	22.6 ± 0.4	79.9 ± 2.0	3.2 ± 0.7	163 ± 64
Ontario	07/25/2018; 11:26 am	R1	25.0 ± 0.2	60.5 ± 0.3	1.5 ± 0.2	218 ± 23
Wisconsin	07/11/2018; 11:00 am	V3	24.8 ± 0.3	46.7 ± 0.6	1.4 ± 0.2	123 ± 14

^aAveraged data from all four heights measured at each site.

^bGrowth stage as defined by Fehr and Caviness 1977.

^cWind speed in Arkansas and Indiana were lower than labeled during applications.

Table 3. Estimated parameters (*b*, *d* and *e*) and downwind distance (m) where 1% (D_1), 10% (D_{10}) and 50% (D_{50}) dicamba injury were observed on covered and non-covered non-DR soybean at six sites.

State/ Province	Cover	Estimated parameter ^a			Distance ^b		
		<i>b</i> (±SE) m	<i>d</i> (±SE) %	<i>e</i> (±SE) m	D_1 (±SE) ----- m -----	D_{10} (±SE)	D_{50} (±SE)
Arkansas	Non-covered	2.6 (0.3)	50.1 (1.4)	66.6 (3.8)	293 (26)	113 (4)	7 (1)
	Covered	1.7 (0.2)	59.2 (7.1)	33.5 (6.9)	347 (40)	84 (8)	13 (3)
Indiana	Non-covered	2.2 (0.4)	37.9 (6.8)	5.2 (1.1)	28 (4)	8.3 (1.0)	-
	Covered	4.6 (3.5)	3.3 (1)	6.5 (1.1)	8 (3)	-	-
Michigan	Non-covered	1.4 (0.7)	39.1 (8.7)	4.9 (2.6)	71 (52)	11 (3)	-
	Covered	1.7 (0.3)	13.0 (1.5)	1.7 (0.5)	7 (7)	1 (1)	-
Nebraska	Non-covered	16.4 (27.2)	65.1 (6.1)	6.6 (0.9)	-	-	-
	Covered	5.1 (7.7)	47.1 (6.1)	9.9 (2.9)	-	-	-
Ontario	Non-covered	3 (0.9)	15.8 (3.1)	10.7 (0.7)	15 (2)	8 (2)	-
	Covered	-	-	-	-	-	-
Wisconsin	Non-covered	2.9 (0.2)	44.0 (0.7)	5.2 (0.2)	19 (1)	8 (0.1)	-
	Covered	2.4 (0.2)	40.9 (1.2)	3.1 (0.2)	15 (1)	5 (0.1)	-

^a *b*, the slope; *d*, the upper limit; and *e*, the inflection point relative to the upper limit (50% injury reduction related to the upper limit). SE, standard error.

^b D_1 is the distance of 1% injury; D_{10} is the distance of 10% injury; and D_{50} is the distance of 50% injury.

Table 4. Estimated parameters (*b*, *d* and *e*) of dicamba deposition on non-DR soybean plants (based on dicamba deposition on filter papers (ng filter paper⁻¹) placed downwind adjacent to the sprayed area at six sites.

State	Estimated parameter		
	<i>b</i> (±SE)	<i>d</i> (±SE)	<i>e</i> (±SE)
	----- ng filter paper ⁻¹ -----		
Arkansas	4.1 (2.7)	59.9 (19.3)	81.9 (14.9)
Indiana	1.5 (0.8)	31.4 (17.5)	31.8 (25.7)
Michigan	2.1 (1.9)	4931.5 (4371.4)	1.4 (0.6)
Nebraska	2.4 (0.9)	20471.0 (14187.0)	5.5 (4.1)
Ontario	2.3 (1.2)	524.8 (548.4)	6.4 (6.1)
Wisconsin	1.5 (0.4)	1684.2 (417.9)	0.6 (0.3)

^a *b*, the slope; *d*, the upper limit; and *e*, the inflection point relative to the upper limit (50% dicamba deposition reduction related to the upper limit). SE, standard error.

Figure legends

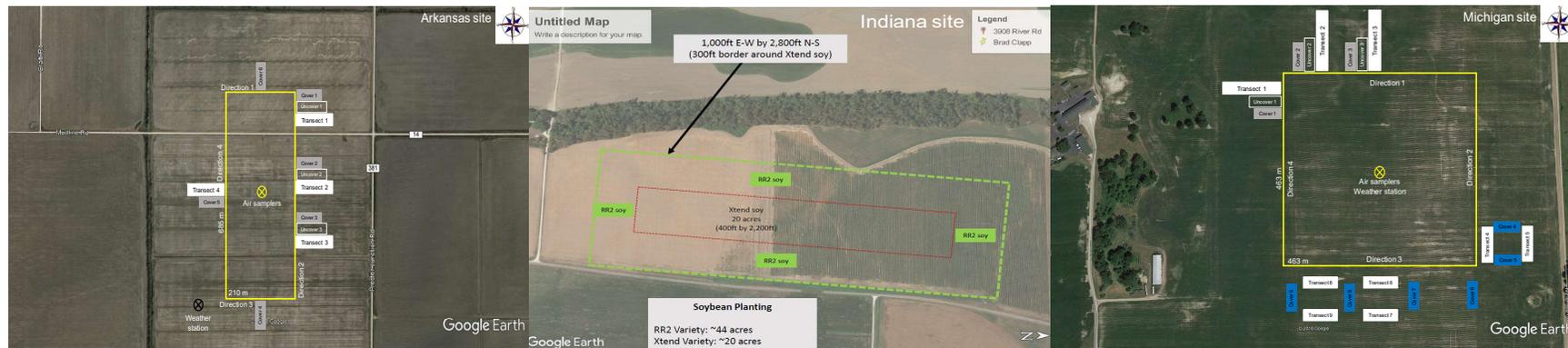
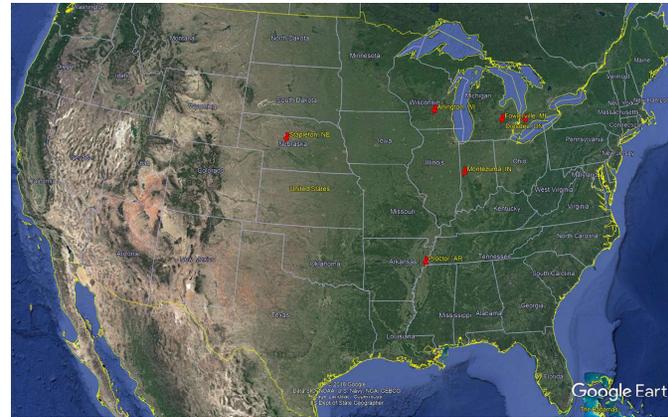




Figure 1. Field plot layout for six experiments conducted in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin to evaluate off-target movement of dicamba from applications in 2018 growing season.

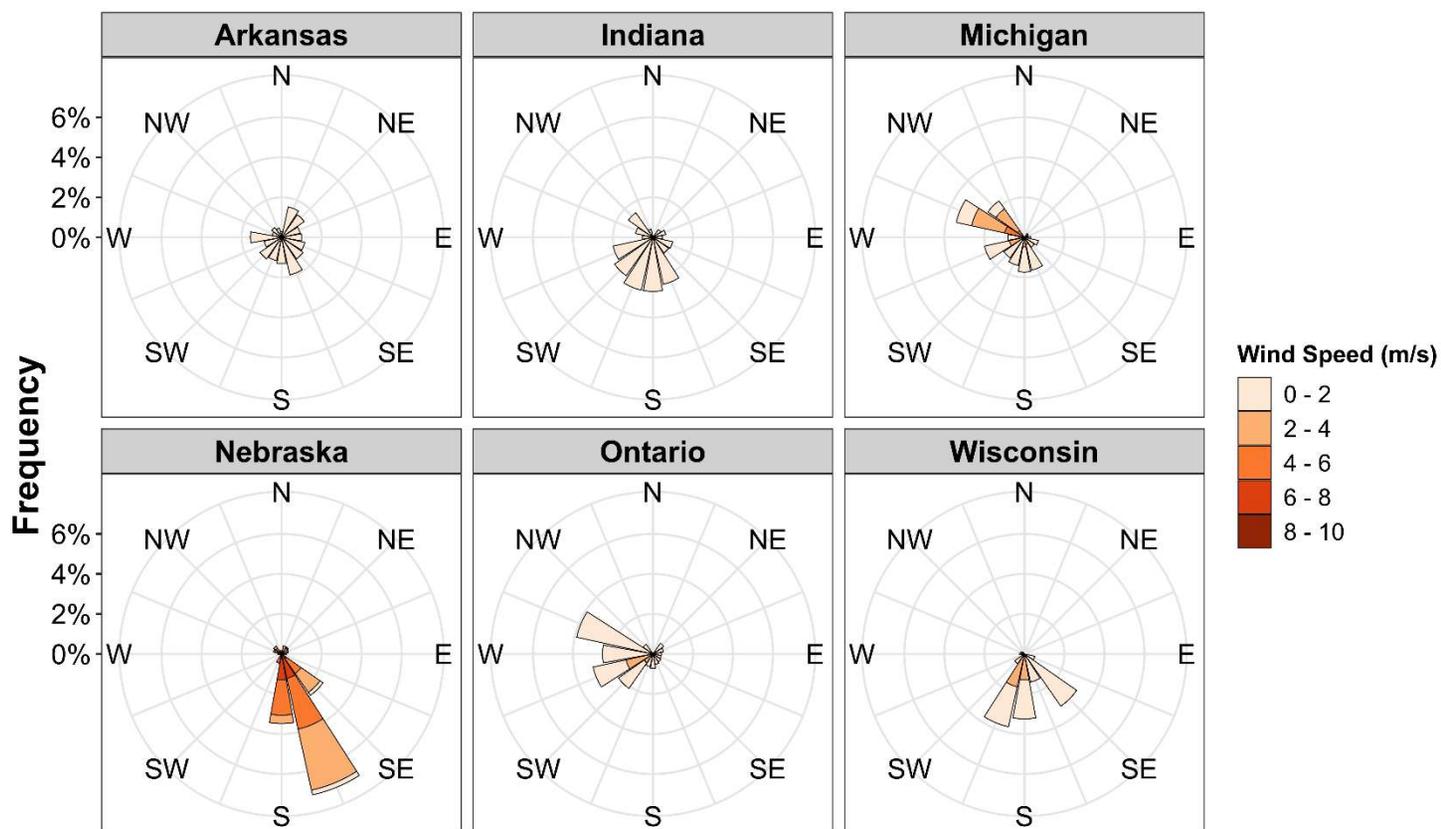


Figure 2. Wind rose plots demonstrating the average wind frequency, speed and direction during air sampling period after dicamba application for six experiments conducted in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin in 2018.

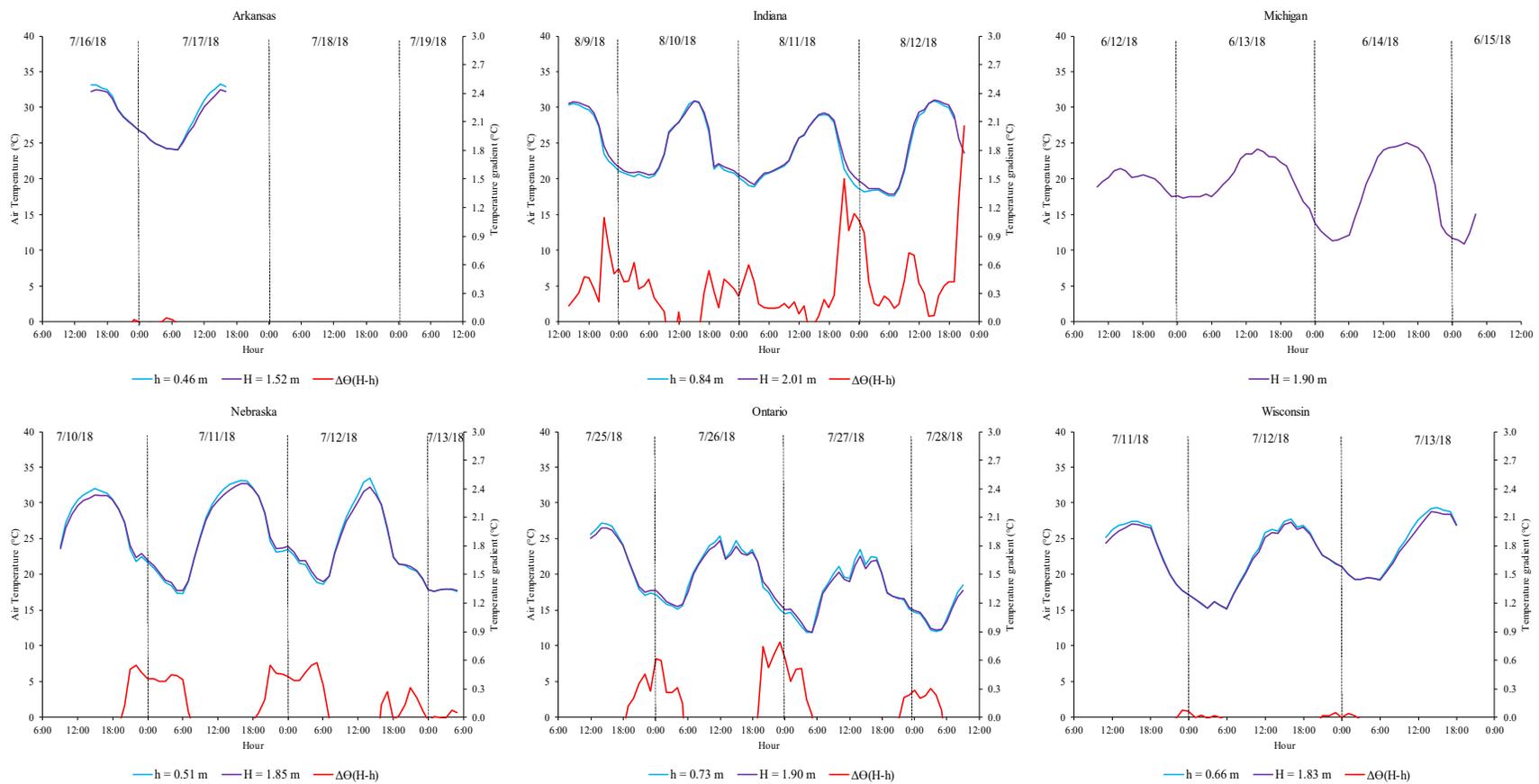


Figure 3. Temperature fluctuations and inversions during air sampling period after dicamba application for six experiments conducted in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin in 2018.

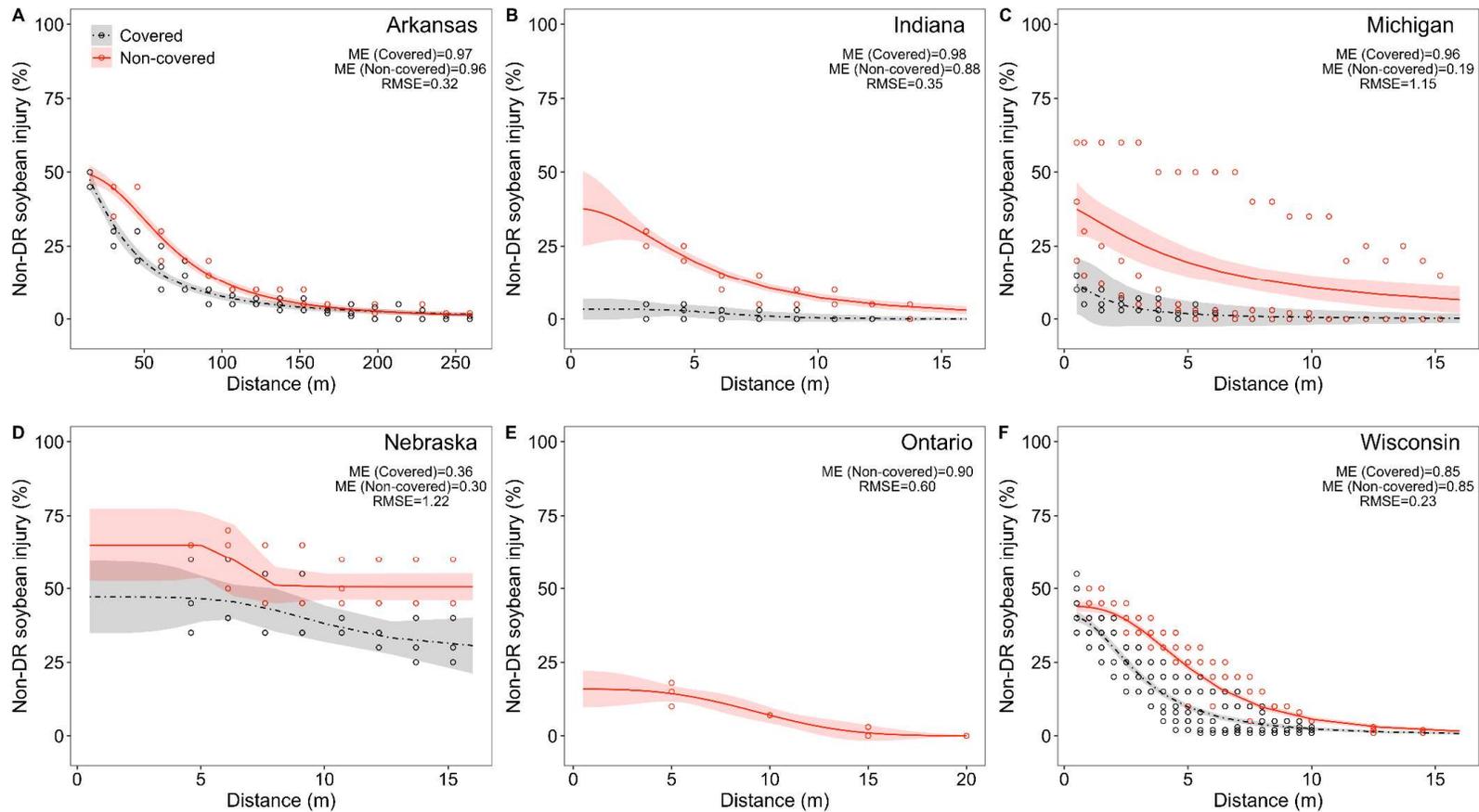


Figure 4. Non-dicamba-resistant soybean injury at various distances from the dicamba-treated areas (covered and non-covered) in the downwind direction in Arkansas (A), Indiana (B), Michigan (C), Nebraska (D), Ontario (E), and Wisconsin (F) at 28 days after application (DAA) in Wisconsin and 21 DAA in the other sites. Shading areas represent 95% confidence interval.

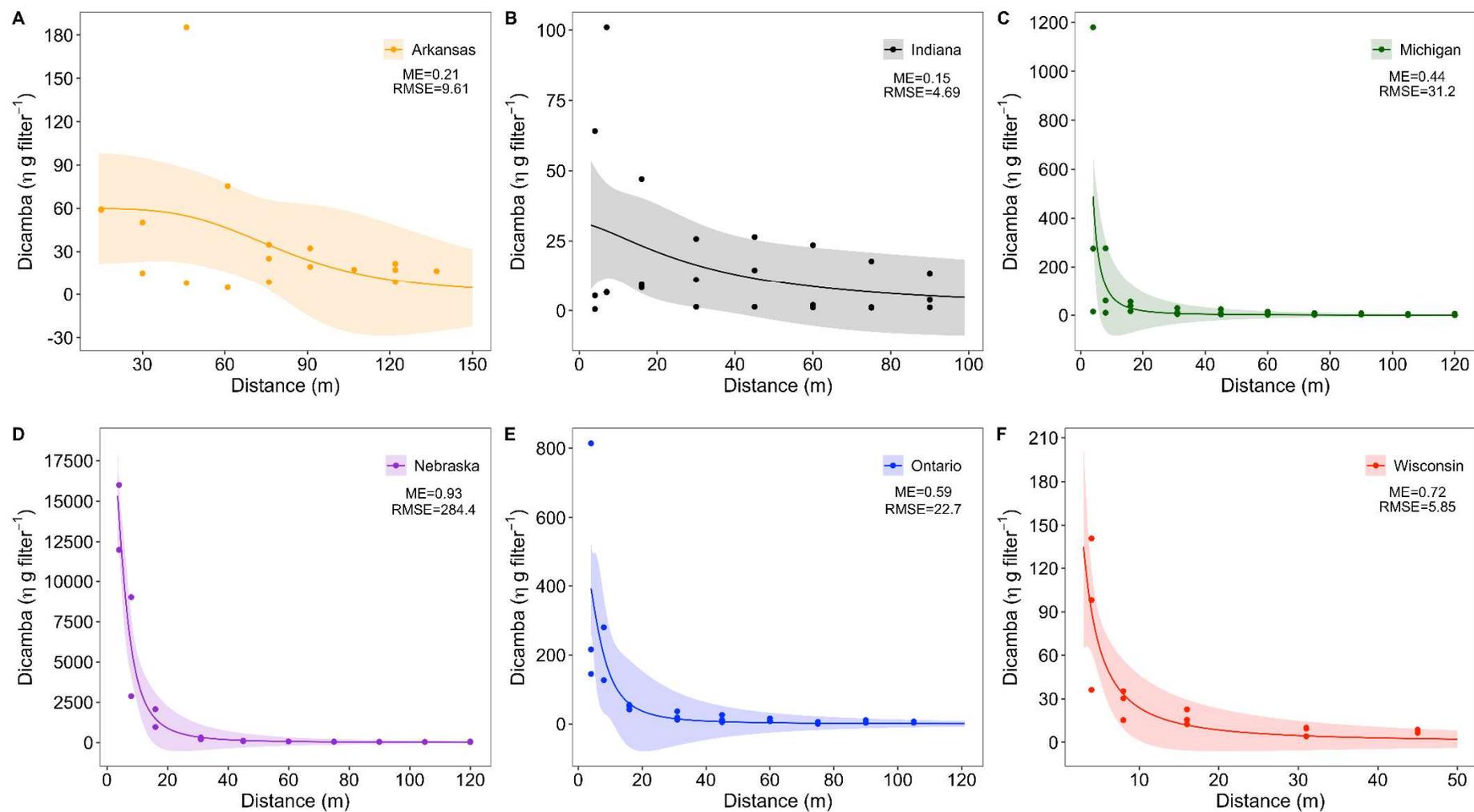


Figure 5. Dicamba deposition at various distances from the dicamba-treated area in the downwind direction in Arkansas (A), Indiana (B), Michigan (C), Nebraska (D), Ontario (E), and Wisconsin (F). Shading areas represent 95% confidence interval.

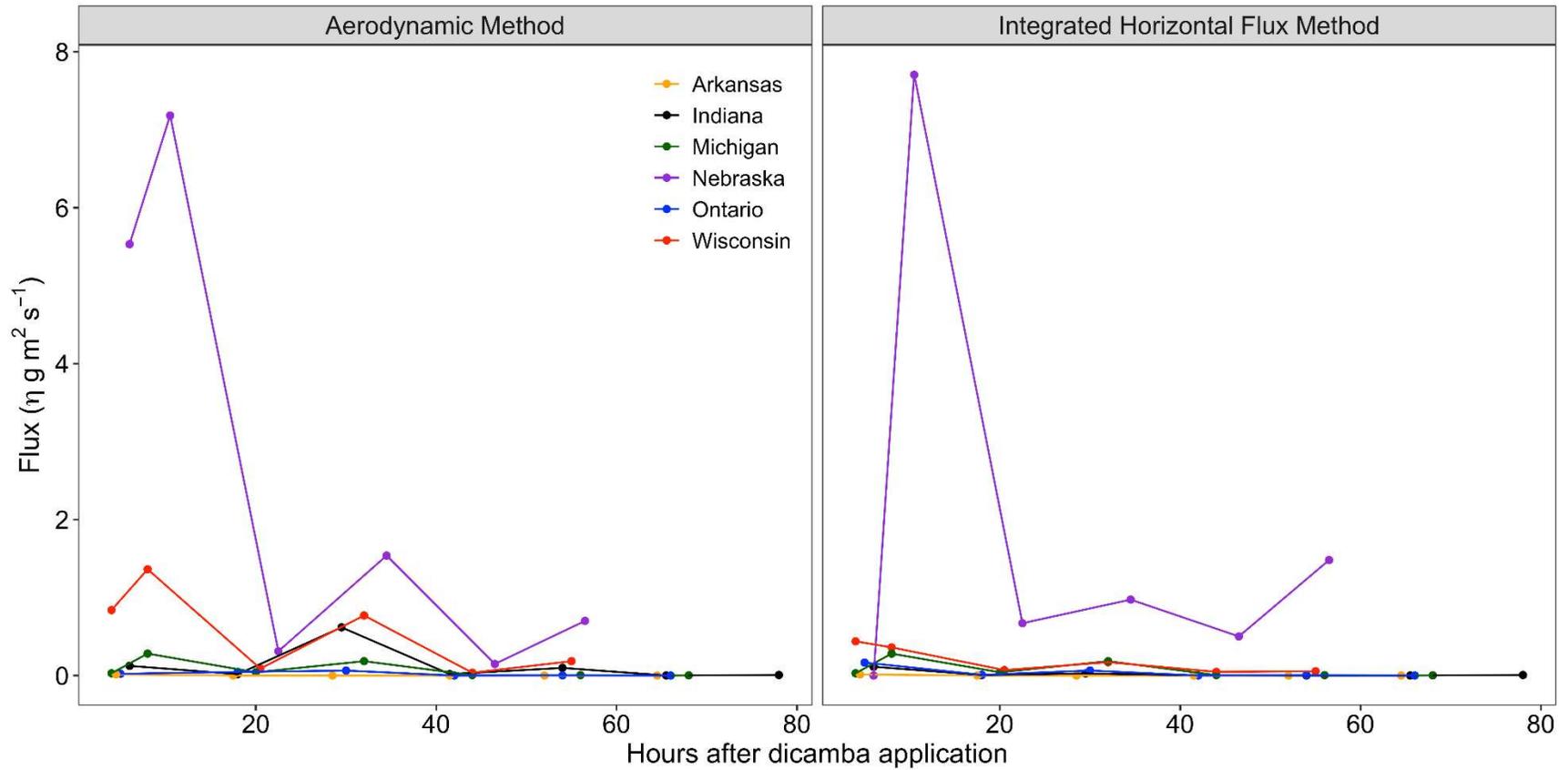


Figure 6. Dicamba flux from the treated area estimated using Aerodynamic and Integrated Horizontal Flux methods up to 78 hours after application in Arkansas, Indiana, Michigan, Nebraska, Ontario, and Wisconsin.