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Short title: Spray drone pattern, pt 1

Agricultural spray drone deposition, Part 1: Methods for high throughput spray pattern analysis

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Abstract

Effective pesticide application is dependent on precise and sufficient delivery of active ingredients to targeted pests. Water-sensitive papers (WSP) have been used to estimate the stain coverage, droplet density, droplet size, total spray volume, and other spray quality metrics by analyzing deposit stains using image analysis software. However, due to their expensive cost, WSPs are typically distributed along unidimensional transects at intervals of 0.5 m or more, which comprises 0.5% or less of the total treated area. This might limit the ability to accurately represent the deposition of agricultural sprayers with irregular patterns, such as agricultural drone sprayers in their early development stage. This study introduces a novel approach utilizing white Kraft paper and a blue colorant proxy for assessing spray deposition. A custom Pythonbased image analysis tool, SprayDAT (Spray Droplet Analysis Tool), was developed and compared to traditional image analysis software, DepositScan. Both models showed increased accuracy in detecting larger objects, with SprayDAT generally performing better for smaller droplets. DepositScan underestimated the total deposited spray volume by up to 2.7 times less compared to the colorant extraction assessed via spectrophotometry and the predicted output based on flow rate, coverage, and speed. Accuracy of software-estimated spray volume declined with increasing total stain coverage, likely due to overlapping stain objects. Droplet density exhibited a Gaussian trend with peak density at approximately 22% stain cover, offering evidence for overlapped stains for both DepositScan and SprayDAT as stain cover increased. Both models showed exponential growth in volumetric median diameter (VMD) with increasing stain cover. SprayDAT is freely accessible through an online repository. It features a userfriendly interface for batch-processing large sets of scanned images and offers versatility for customization to meet individual needs, such as adjusting spread factor, updating the standard curve for spray volume estimation, or modifying the stain detection threshold.

Keywords: deposition analysis, droplet size, image analysis, SprayDAT, spray pattern analysis, UAV application.

Introduction

Pesticide efficacy is dependent on the sufficient delivery of active ingredients to targeted pests (Creech et al. 2015; Hislop 1987). According to Knoche (1994), foliar-applied herbicide performance was inversely related to droplet sizes in 71% of 159 experiments. However, small droplets (<100µm) are prone to drift (Frank et al. 1991), therefore, droplet spectral analysis is used to manage the conflict between pesticide performance and nontarget drift (Al Heidary et al. 2014; Makhnenko et al. 2021; Matthews et al. 2014). With ground-based agricultural sprayers, droplet spectra of a given spray tip are classified under controlled, indoor conditions with highspeed cameras, laser diffraction, or phase doppler particle analysis (Anonymous 2023; Nuyttens et al. 2007; Sijs et al. 2021). Spray tips are then oriented to ensure uniform deposition along the length of an equipment-mounted boom. Since these systems are designed to deliver inherently uniform spray deposition, methods employed to assess in-field spray deposition have focused on crop canopy penetration (Chen et al. 2020; Huang et al. 2023; Wang et al. 2019) or windmediated drift effects (Fritz et al. 2011; Wang et al. 2020). Since particle analysis systems are impractical for field use, researchers have sought to estimate droplet sizes and overall deposition coverage based on stains deposited on water-sensitive paper (WSP) or other media (Cunha et al. 2012; Fritz et al. 2011; Hewitt et al. 2002; Li et al. 2021a, 2021b; Martin et al. 2019; Panneton 2002; Salyani and Fox 1999; Salyani et al. 2013; Wang et al. 2020; Woldt et al. 2018; Zhu et al. 2011).

There are several limitations of using WSP to assess spray deposition. Most WSP products comprise a sampling area that is less than 30 cm^2 and are spaced along unidimensional transects at intervals of 0.5 m (Wen et al. 2019; Zhang et al. 2021), 1 m (Ahmad et al. 2020; Qin et al. 2016) or more (Bonds et al. 2023; Fritz et al. 2019). Previously reported research involving WSP-assessed spray deposition sampled between 0.01 and 0.5% of the treated area (Qin et al. 2016; Wang et al. 2019). Such discrete sampling may be sufficient for systems with inherently uniform spray delivery, but may be too low to adequately assess deposition uniformity of an agricultural spray drone. However, Richardson et al. (2020) used Gaussian modeled deposition on steel plates and compared this to modeled deposition estimates based on fluorescence detected on continuous string and found similar slopes across sampling resolutions of 0.25 to 1.0 m. In preliminary research (Koo et al. 2021), our continuous deposition analysis revealed multiple peaks near the center of the spray drone flight path at near 2-m height that were not

resolved or not reported in the Richardson et al. (2020) paper involving the same model of spray drone. Spray drones are designed to achieve efficient delivery of low-volume spray and typically use rotor-induced wind to disperse small droplets in an effort to increase spray coverage (Qin et al. 2016; Zheng et al. 2018). These design elements are likely to cause changes in deposition patterns depending on drone height and ambient wind (Hunter et al. 2020; Richardson et al. 2020).

Further limitations of WSP are caused by physical characteristics of the bromophenolblue coated paper which varies in response to ambient moisture (Turner and Huntington 1970). Droplets smaller than 50 µm in diameter do not have enough moisture to create stains on WSP (Hoffmann and Hewitt 2005), and small droplet stains that are created may not be resolved by scanners. At relative humidity greater than 85%, WSP stains can increase in size or occur spontaneously (Anonymous 2002; Franz et al. 1998). An overlap of droplet stains also can generate erroneous data since image analysis software will detect the overlapped droplets as a single droplet. This phenomenon has not been characterized, but has been shown to increase with increasing spray volume (Cunha et al. 2012; Zhu et al. 2011). Previous researchers have suggested that utility of WSP to estimate deposition patterns is limited when \geq 20% of the WSP area is covered by droplet stains (Cunha et al. 2012; Zhu et al. 2011). Ellipse-shaped stains also cause an issue since image analysis software assumes that droplets have circular shapes to calculate droplet sizes (Deveau 2021b). This is another source of error that will be exacerbated with the advent of agricultural drone sprayers, as these sprayers cause a large percentage of elongated stain objects or streaks due to wind shear (Fritz et al. 2019). Detected objects derived from stains on WSP are identified based on differential hue and saturation, then detected pixel area is converted to an estimated stain diameter assuming all objects are circular (Cunha et al. 2012; Zhu et al. 2011). Estimated stain diameters are converted by a spread factor, specific to each type of WSP (Deveau 2021a), to represent actual droplet size (Ahmad et al. 2020; Anonymous 2002; Cunha et al. 2013; Salyani and Fox 1994, 1999; Zhu et al. 2011). Given that WSP does not allow for extraction of spray deposits and further estimation of mass per unit area of spray-delivered compounds, relationships of, and errors associated with stain coverage on WSP are often limited to relative comparisons that vary by type of WSP (Deveau 2021c) rather than quantifiable deposition. Mylar cards or other plastic and glass-based samplers are methods that enable pesticide quantification. Spray solutions typically contain dye or

fluorescent tracers that are used as a proxy for pesticides. Tracer-treated Mylar cards are removed from the field, washed with ethanol, and analyzed by a spectrofluorometer. The fluorescence value can be converted to a mass dye per area using pre-determined standard curves (Fritz et al. 2011; Wang et al. 2020). Much like Mylar cards, extracts of pesticide-sprayed filter papers were analyzed by liquid, gas, or mass chromatography (Hewitt et al. 2002; Li et al. 2021a, 2021b). Chromatography and fluorometry are robust methods to quantify pesticide delivery, though their costs usually restrict sampling area below that needed to assess highly variable deposition patterns typical of agricultural spray drones. Cotton string and monofilament line also were utilized by researchers using similar extraction and analysis techniques to Mylar card and filter paper assessment (Fritz et al. 2011; Martin et al. 2019; Wang et al. 2020; Woldt et al. 2018). String samplers can generate continuous data, albeit on a narrow, unidimensional transect. This technique is useful for assessing spray drift, but not for characterizing multidimensional spray patterns.

Thus, the development of a new analysis method that can characterize spray deposition on continuous and/or multidimensional areas with spatially referenced, mass-to-area assessment of extracted pesticide or tracer dye is needed. Furthermore, assessing accuracy of digitally analyzed stain coverage on paper media to detect droplet deposition quantity and spectral relationships requires spatial reference of stain images and mass-to-area quantification of extracted compounds. To achieve this goal, we utilized spectrophotometric analysis of tracer dye extracted from digitally imaged Kraft paper. A custom python-based image analysis method, SprayDAT, was compared to the ImageJ-based image analysis software, DepositScan, to estimate total spray deposition and droplet spectral characterization based on actual droplet sizes. SprayDAT offers two advantages over DepositScan that are necessary to deal with the thousands of images generated by continuous sampling of economical Kraft paper. The first is batchprocessing of images and the second is that code is easily modified depending on the needs of users. To realize these software advantages, SprayDAT must be compared with DepositScan for stain cover and droplet spectral estimation. Since our method allows for both stain imaging and colorant extraction from the same surface, this paper will be the first to report a direct comparison between extracted colorant and digitally analyzed stain objects per unit area. The first objective of this study was to assess the accuracy of digitally imaged stains analyzed by two computer software packages to estimate total spray deposited and droplet spectral characteristics

compared to measured spray delivery rates, manufacturer-reported droplet spectral characteristics, and extracted blue colorant quantified via spectrophotometric analysis. Our second objective was to assess the validity of a novel computational method for assessing stain objects as a measure of spray deposition quantity and droplet spectral characterization.

Materials and Methods

Sampler and Proxy

White Kraft paper (Oren International, Pensacola, FL) and colorant (Blazon blue spray pattern indicator, Milliken, Spartanburg, SC) were selected as the sampler and pesticide proxy. This combination provides high-contrast stains like WSP but offers affordable, scalable spatial sampling and easy water extraction and spectrophotometric analysis of the proxy. Since the Kraft paper and dye method costs 0.2 cents for each USD spent on WSP, larger sampling areas are feasible. Preliminary studies were conducted to evaluate various solvents and volumes to assess the extraction efficiency of the proxy colorant. Colorant was extracted from a 528 cm² area of Kraft paper at 99% efficiency (data not shown) using 100 ml tap water in 125 ml screw-top jars shaken at 250 rpm with a G2 Gyratory Shaker (New Brunswick Scientific Co., Edison, NJ) for 10 min. A 0.4 ml aliquot was removed, mixed with 1.6 ml of tap water, and absorbance was read at 650 nm with a Genesys 5 spectrophotometer (Thermo Fisher Scientific, Waltham, MA). The equation below describes the second-order polynomial regression $(R² = 0.99)$ that relates colorant absorbance value (a) at 650 nm and known amount of colorant (c, ul colorant 100 ml^{-1} tap water).

$$
c = 1.5629 \ a^2 + 10.776 \ a \tag{1}
$$

Sample preparation

Known doses of colorant were applied to 20.3 cm by 30.5 cm Kraft paper using a spray boom equipped with a single flat-fan nozzle [TeeJet XR11001VS (Spraying Systems Co., Wheaton, IL)] operated at varying speeds at a pressure of 120 kPa, resulting in a flow rate of 4.2 ml s⁻¹. This flow rate and nozzle was selected to mimic the manufacturer-supplied parameters of a DJI MG-1P agricultural spray drone (DJI, Shenzhen, China) that has been evaluated in other experiments. The colorant was mixed 1:1 with water and sprayed in a custom laboratory spray system. A rigid steel frame was prepared to maintain the spray height at 61 cm above the Kraft paper. Four Kraft papers were aligned, and the locations corresponding to the leading and trailing edges of each paper were marked on the steel frame. Thirty unique doses were applied by

varying the nozzle speed, which was confirmed via high-speed video (Edgertronic, Sanstreak Corp, Campbell, CA) at 1500 frames s^{-1} . The time required for the spray nozzle to traverse the distance of the paper was measured by the number of frames it was present in the video, and the speed was calculated. Since all other factors were held constant, the application volume per unit area and the associated rate of colorant were linearly related to speed. All treated Kraft papers were scanned at 23.6 dots mm^{-1} (600 dpi) with a Ricoh MP C307 color scanner (Ricoh, Tokyo, Japan). Colorant was extracted from a 619 cm² area of Kraft paper using 100 ml tap water in 125 ml screw-top jars shaken at 250 rpm with a G2 Gyratory Shaker (New Brunswick Scientific Co., Edison, NJ) for 10 min. A 0.4 ml aliquot was removed, mixed with 1.6 ml of tap water, and absorbance was read at 650 nm with a Genesys 5 spectrophotometer (Thermo Fisher Scientific, Waltham, MA).

Development of a new image analysis method, SprayDAT (Spray Droplet Analysis Tool) Image conversion

Computational models were generated using Python 3.9 and tested to measure deposition coverage based on colorant stains and to estimate droplet size. Scanned images were converted to grayscale images, and a selected threshold of 154 within the grayscale range of 0 to 255 was applied to create a binary mask of black droplet stains and white backgrounds. The threshold was found to best encompass all of the detected droplets without highlighting potential noise in the scanned images, such as dust or dirt particles. The outline of each stain was detected using the contour detection algorithm proposed by Suzuki and Abe (1985), and the areas of each stain were calculated based on Green's theorem. Each stain was then considered as a circle, and the diameter of each stain was estimated. The proportion of stain area and the number of stain objects were used to calculate spray coverage and droplet density respectively.

Reference stain detection

To test the accuracy of stain detection by SprayDAT, nominal spot sizes of 100, 200, 300, 500, 1000, and 2000 µm in diameter on the reference card (ORBITRANSIT, China) were analyzed. The reference card was scanned at 23.6 dots per mm (600 dpi) using a Ricoh MP C307 color scanner (Ricoh, Tokyo, Japan). The diameters of the nominal-sized spots on the scanned images were analyzed by SprayDAT and compared to the results of DepositScan. *Spread factor*

To estimate the actual droplet sizes from the Kraft paper sampler, the spread factor between the stain size and actual droplet sizes needed to be determined (J. P. A. R. Cunha et al. 2012; M. Cunha et al. 2013; Zhu et al. 2011). The spread factor between Kraft paper and blue colorant, which has not been reported by previous researchers, was determined across a range of discrete droplet diameters, using methods similar to Smith et al. (2000). A custom-built droplet generator consisting of a 2.5 cm spinning disc was operated via a brushless motor at 3750 rpm. Droplets of discrete size were ejected through a 0.5 cm by 1.0 cm aperture in an enclosure around the spinning disc at 46 cm above the sampling location. A camera slider (GVM GT-60D, Great Video Maker, Philadelphia, PA) was fitted with a horizontal platform that contained a 5 cm petri dish filled with a bi-phasic solution of polydimethylsiloxane (PDMS) (MicroLubrol®, Type 200 Silicone Oil, MicroLubrol, LLC, Clifton, NJ), and a 25 cm² sheet of Kraft paper. The lower 1 mm of the dish contained PDMS of 12,500 cSt viscosity, while the upper 2 mm layer contained PDMS of 100 cSt viscosity. Droplets are suspended in the oil solution at the interface between the two PDMS layers of varying viscosity allowing for accurate imaging on a narrow plane. Both PDMS-filled petri dishes and Kraft papers were placed at 20, 32, 43, 55, 66, 77, and 89 cm from the droplet generator to achieve droplet diameters of 112, 143, 183, 218, 246, 275, and 315 µm, respectively, with a standard deviation less than 10% in all cases.

After treatment with different sized droplets, the petri-dishes were carefully transferred to a white translucent panel illuminated underneath with an LED light (Craftsman 4500LM 46W LED Work Light, Craftsman, Towson, MD) and photographed with a digital camera (Canon EOS 5D, Canon, Tokyo, Japan) at an effective resolution of 157 dots mm⁻¹ (2400 dpi). Kraft papers were scanned at 23.6 dots mm^{-1} (600 dpi) with a Ricoh MP C307 color scanner (Ricoh, Tokyo, Japan). The droplet sizes and stain diameters were measured by counting pixels and converted based on known pixel to distance relationships using the binarization threshold of 154 within the grayscale range of 0 to 255. The spread factor was calculated as the difference between droplet diameters in the PDMS solution and stain diameters on Kraft paper.

Droplet spectra analysis

A total of 120 scanned sample images were digitally analyzed via SprayDAT and compared to analyses from ImageJ-based 'DepositScan' software developed by USDA-ARS (Zhu et al. 2011). Detected droplet stains on each scanned image were converted to actual droplet sizes based on the calculated spread factor. Afterwards, the volume of each droplet was

calculated based on the following equation, where V_i is the estimated droplet volume based on d_i , the estimated actual droplet diameter from equation 2.

$$
V_i = \frac{\pi d_i^3}{6}, i = 1, ..., N
$$
 [2]

Cumulative volume was calculated by summing the volume of each droplet, and $D_{v0.1}$, $D_{v0.5}$ (VMD), and $D_{v0.9}$ were calculated using a similar interpolation method described by Zhu et al. (2011). Since DepositScan generates $Dv_{0,1}$, VMD, and $Dv_{0,9}$ based on a spread factor (*d* = 0.95 $d_s^{(0.910)}$ originating from work on WSP, output from DepositScan was adjusted using the Kraft-paper-specific spread factor described in equation 3.

Spray deposition estimation

In addition to comparing computational models for droplet spectra, the total deposition of blue colorant from different computational models was assessed. Firstly, the reference spray deposition was estimated based on the application speed of each sample, as the spray volume could be calculated from the known flow rate and spray width. An extraction-based method was also employed to quantify total colorant extracted from each piece of Kraft paper that could then be used to quantify spray deposition. This method utilized a standard curve correlating colorant dosage with spectrophotometric absorbance values, which were then converted into deposition volumes (μ l colorant cm⁻²), accounting for the surface area of the sample. The colorant deposited per unit area estimated by SprayDAT or DepositScan was based on the total volume of all droplets based on measurements of each discrete stain object via digital imagery of sprayed Kraft paper adjusted for the ratio of colorant to water.

Results and Discussion

Spread factor calculation

The difference between droplet diameters in the PDMS solution and stain diameters on Kraft paper were subjected to a nonlinear two-parameter power regression $(R^2=0.9)$ where *ds* and *d* are stain and actual droplet diameters in μ m, respectively.

$$
d = 0.83 \, d_s^{0.79} \tag{3}
$$

Compared to the spread factor $(d = 0.95 d_s^{0.91})$ of the WSP described in Zhu et al. (2011), equation 3 indicated that blue colorant proxy left more stains on white Kraft paper than water on the WSP.

Object detection and measurement by two computational models

Table 1 describes the trend of object detection when two computation models (SprayDAT and DepositScan) assessed objects of known nominal sizes ranging from 100 to 2000 µm on reference cards (ORBITTRANSIT, China). Both SprayDAT and DepositScan exhibited a trend of increased accuracy in diameter detection as object size increased (Table 1), as has been reported for similar object size estimation (Brandoli et al. 2021; Zhu et al. 2011). SprayDAT enlarged 100 µm droplets by 17%, but all other objects were measured accurately to within a 4% error rate. DepositScan, in contrast, enlarged 100, 200, and 300 μ m objects 51, 22, and 10%, respectively while accurately measuring objects that were 500 to 2000 µm within a 5% error rate. When 120 Kraft-paper samples were analyzed for spray deposition stains, total cover of all stain objects estimated by DepositScan and SprayDAT were linearly related with a 0.99 coefficient of determination (Figure 1). The slope of 0.99 that relates total stain cover between the computational models indicates that SprayDAT is estimating stain objects to be slightly smaller than that of DepositScan (Figure 1), in agreement with observations made when estimating known objects on reference cards where DepositScan enlarged objects as much as 51% (Table 1). Across the 120 Kraft-paper samples sprayed in the laboratory, application speeds ranged from 0.4 to 8.4 m/s, resulting in 3.8% to 33.4% SprayDAT-assessed stain cover (Figure 2). The relationship between application speed and percentage stain cover followed an inverse first-order polynomial regression ($R² = 0.95$) and deviated only slightly in trend from predicted application volume based on variable speeds given a constant flow rate of 4.2 ml s⁻¹ (Figure 2). *Droplet density relationships*

The density of individual stain objects on Kraft paper is referred to as "droplet density" and expressed as the estimated number of droplets landing in each $cm²$ of Kraft paper. The relationship of droplet density as measured by SprayDAT and DepositScan was linear with a 0.93 coefficient of determination. The slope of 1.13 indicates that SprayDAT is estimating a higher number of droplets per unit area compared to DepositScan (Figure 3). The likely reason for increased droplet density assessed by SprayDAT is the higher accuracy of SprayDAT in detecting smaller droplets (Table 1). Detecting overlapping multiple droplets as a single large droplet has been a widely reported problem in image analysis techniques for paper-based samplers such as WSPs (Brandoli et al. 2021; J. P. A. R. Cunha et al. 2012, M. Cunha et al. 2013; Fox et al. 2003; Özlüoymak and Bolat 2020; Zhu et al. 2011), but none of these reports characterized the relationship between total stain cover and droplet density. In our study, the

number of unique stain objects exhibited a Gaussian trend as total stain cover increased with peak droplet densities of 129 and 138 droplets cm^{-2} for DepositScan and SprayDAT, respectively (Figure 4). These peak droplet densities were reached at 22.7 and 22.2% stain cover, respectively, after which, droplet density declined (Figure 4). The decline in droplet density after 22% stain cover is evidence that additional droplet stains increasingly overlap as more droplets are added. These overlapping stains either merge into larger stain formations or are concealed by pre-existing stains, resulting in a complex pattern of layered stains. This phenomenon was reported but not characterized by other researchers (Brandoli et al. 2021; Fox et al. 2003; Zhu et al. 2011). Fox et al. (2003) estimated arbitrarily that WSPs or Kromekote® cards with stain coverage exceeding 20% would be of limited value for estimating total volume of spray per unit area. This 20% estimate aligns well with our Gaussian relationship between total stain coverage and droplet density (Figure 4). While neither computational method can resolve overlapping stain objects, SprayDAT tended to more accurately detect small stains (Table 1) and slightly conserve droplet density (Figures 3, 4) compared to DepositScan.

Estimating droplet spectra based on stain objects

The average VMD estimated by SprayDAT and spread-factor-corrected (Equation 3) DepositScan across 120 Kraft-papers sprayed at a constant flow rate with variable speed, was 108 µm and 121 µm, respectively (Table 2). The manufacturer of the TeeJet XR11001VS nozzle characterizes the spray output at the utilized operating pressure of 120 kPa (Anonymous 2023) as 'Fine' based on the American Society of Agricultural and Biological Engineers (ASABE) Standard 572.3 suggesting the VMD ranges from 106 to 235 μ m (Anonymous 2020; Grisso et al. 2019). Therefore, average VMD estimations from SprayDAT and DepositScan with adjusted spread factor were in range of expected values, although DepositScan estimated a significantly larger VMD compared to SprayDAT. Excluding objects that were outside the bounds of 40 to 300 or 40 to 400 µm did not significantly impact VMD, $DV_{0.1}$, or $DV_{0.9}$ (Table 2). These data in Table 2 appear to contrast with reports of overlapping and enlarged stain objects as total stain coverage increases (Brandoli et al. 2021; Fox et al. 2003; Zhu et al. 2011) since bounding did not alter VMD estimation by SprayDAT (Table 2). It should be noted, however, that over 60% of our 120 Kraft-paper samples had total stain coverage of less than 10% and only 11 of the 120 samples exceeded total stain coverage of 20% (Figure 2). Thus, our average spectral analysis across all 120 samples was less subject to the errors associated with excessive

total stain coverage. When we evaluate VMD as a function of total stain cover, the data fit an exponential growth curve (Figure 5) in all cases for DepositScan, DepositScan with adjusted spread factor, and SprayDAT. Since all factors were held constant with the exception of speed, the VMD in this laboratory experiment should be constant. The increase in VMD associated with increased stain cover in the computational models is evidence of overlapping or merged deposition stains that occur in increasing frequency as total stain cover increases. These data in Figure 3 also highlight the importance of spread factor accuracy. The original output from DepositScan is largely deviant compared to SprayDAT or spread-factor-adjusted DepositScan. Since spread factors have been reported to deviate with varying WSP products (Deveau 2021a), we see this as a significant limitation of DepositScan. The method required to adjust DepositScan output as spread factor changes (Zhu et al. 2011) requires numerous manual calculations with limited ability for automation. In SprayDAT, images are batch processed and users would simply replace one equation at a specified place in the code if a different spread factor were desired.

Accuracy of predicting deposition volume via spray deposit stains

Despite numerous papers evaluating WSP or other methods to digitally assess spray deposit stains (Brandoli et al. 2021; Cunha et al. 2012; 2023; Ferguson et al. 2016; Özlüoymak and Bolat 2020; Zhu et al. 2011), and spray volume per unit area estimates based on deposit stains computed by available software (Brandoli et al. 2021; Zhu et al. 2011), the accuracy of spray volume estimates based on stain-object-to-droplet-volume relationships remains unreported. A non-linear two parameter power model described the relationship between total stain cover on Kraft paper and estimated spray volume deposited based on speed and flow rate of the sprayer, extracted colorant quantified by standardized spectrophotometric absorbance, and deposit stains measured by DepositScan or SprayDAT (Figure 6). The extraction method underestimated spray deposition approximately 9% compared to speed-based calculations, which are considered the reference (Figure 6). The underestimation might be attributed to losses during the extraction process, but preliminary experiments indicate our extraction method accounts for over 99% of colorant (data not shown).

Both computational models that relied on deposit stains were divergent with the actual predicted spray volume of calibrated equipment and estimates of spray volume based on extracted colorant. The divergence between these methods increased with increasing total stain cover such that DepositScan and SprayDAT estimated 2.7 and 3.2 times less spray volume than speed-based predictions when total stain coverage was 30% (Figure 6). The complimentary trends between all four methods is testament that estimates based on stain objects can appropriately reflect relative differences in spray deposition, but these data are the first to characterize how inaccurately deposit-stain-based methods estimate spray volume compared to colorant extraction or predicted spray output based on equipment calibration. This inaccuracy likely stems from declining droplet density as total stain coverage increases (Figure 4), a problem inherent to estimates based on deposit stains. When the x axis of Figure 4 was truncated to 10% total stain cover, a linear trend ($R^2 = 0.97$) with slope 13.2 was predicted (data not shown) that extrapolates at 30% total stain cover to 397 drops cm^{-2} , a difference of 2 times magnitude similar to that between model estimates and predicted deposition (Figure 6).

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Competing Interests

The authors declare no conflicts of interest.

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Table 1. Comparison of diameters measured by SprayDAT and DepositScan for nominal spot diameters of 100, 200, 300, 400, 500, 1000, and 2000 µm on a size reference card (ORBITRANSIT, China).

Note: The reference card was scanned at 23.6 dots mm⁻¹ with the Ricoh MP C307 color scanner (Ricoh, Tokyo, Japan).

Table 2. Volume median diameter (VMD; $Dv_{0.5}$), $Dv_{0.1}$, and $Dv_{0.9}$ measured by ImageJ-based DepositScan and Python-based SprayDAT for stain objects detected on 120 Kraft paper samples following spray of a blue colorant solution. Original DepositScan output was compared to that with an adjusted spread factor, and SprayDAT was executed with or without bounding to exclude objects smaller than 40 µm or larger than 300 to 400 µm that were deemed erroneous.

Note: All samples were scanned at 23.6 dots mm⁻¹ with a Ricoh MP C307 color scanner (Ricoh, Tokyo, Japan). Data were subjected to SAS (SAS Institute Inc., Cary, NC) 9.4 PROC

GLM procedure, and means were separated by Fisher's Protected LSD (α =0.05)

^aOriginal software uses a spread factor to relate stain size to estimated droplet size based on work with water sensitive paper.

^bSpread factor (eq.2) was adjusted to reflect relationship between blue stains on craft paper and actual size of droplets suspended in oil solution.

Figure	Model	Equation	Equation	Parameters	R^2
		name			
$\overline{4}$	SprayDAT	Gaussian	$y = y_0 + ae^{-0.5 (x-x0)/b^2}$	$a = 138$, $b = 17.3$, 0.73	
				$x0 = 22.2$	
$\overline{4}$	DepositScan	Gaussian	$y = y_0 + ae^{-0.5 (x-x0)/b^2}$	$a = 129$, $b = 17.7$, 0.64	
				$x0 = 22.7$	
5	DepositScan	Exponential	$y = ae^{(bx)}$	$a = 170.06$,	0.97
		growth		$b = 0.0456$	
5	DepositScan	Exponential	$y = ae^{(bx)}$	$a = 75.38$,	0.97
	adjusted	growth		$b = 0.034$	
5	SprayDAT	Exponential	$y = ae^{(bx)}$	$a = 68.6$,	0.98
		growth		$b = 0.039$	
6	Speed	Power	$y = ax^b$	$a = 0.0051$,	0.94
	estimation			$b = 1.263$	
6	Extraction	Power	$y = ax^b$	$a = 0.0045$,	0.99
				$b = 1.279$	
6	Stain object -	Power	$y = ax^b$	$a = 0.0014$,	0.99
	DepositScan			$b = 1.259$	
6	Stain object -	Power	$y = ax^b$	$a=0.0009$,	0.99
	SprayDAT			$b = 1.348$	

Table 3. Nonlinear regression equations with parameters utilized for this study.

Description of equations used

Figure 1. The relationship between percentage stain cover of 120 scanned Kraft paper samples assessed by SprayDAT and DepositScan.

Figure 2. The relationship between the application speed and SprayDAT-assessed stain cover on 120 scanned Kraft paper samples. Predicted spray volume as calculated based on constant flow rate and variable speed is provided as a reference.

Figure 3. The relationship between droplet density (droplets cm⁻²) of 120 scanned Kraft paper samples assessed by SprayDAT and DepositScan.

Figure 4. The relationship between percentage stain cover and droplet density (droplets cm⁻²) of 120 scanned Kraft paper samples assessed by SprayDAT and DepositScan.

Figure 5. The relationship between the percentage stain cover and volume median diameter (μ m) of 120 scanned Kraft paper samples assessed by DepositScan, DepositScan with adjusted spread factor, and SprayDAT.

Figure 6. The relationship between percentage stain cover and estimated total colorant deposited cm^{-2} predicted by speed calculation, colorant extraction, and stain-object-based volume calculated by SprayDAT and DepositScan.