Doi: http://dx.doi.org/10.1590/1809-4430-Eng.Agric.v40n1p69-77/2020

INFLUENCE OF GLYPHOSATE CONCENTRATIONS ON SPRAY SOLUTION PHYSICOCHEMICAL CHARACTERISTICS AND DRIFT POTENTIAL

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KEYWORDS
surface tension, contact angle, droplets, MVD, Congo grass.

ABSTRACT
The herbicide glyphosate is sprayed with varying application volumes that result in different concentrations used in the spray solutions. Thus, this study aimed to evaluate the effect of glyphosate concentrations on the potential risk of droplet drift, surface tension of spray solution, and contact angle of droplets on surfaces. Three experiments were conducted in a completely randomized design. Drift potential was evaluated by median volumetric diameter (MVD), percentage volume of droplets with diameter smaller than 100 µm (%V<100 µm), and coefficient of uniformity (Span), with 12 replications. The surface tension of spray solutions and contact angle of droplets were evaluated on Urochloa ruziizensis leaves, paraffin film, and glass, with four replications. Changing glyphosate concentrations in the spray solution changes the droplet spectrum and contact angle on different surfaces due to changes in spray solution physicochemical characteristics, such as surface tension. The increased glyphosate concentrations in the spray solution lead to increased drift potential of droplets, reducing the surface tension and contact angle of droplets on surfaces.

INTRODUCTION
Glyphosate is a widely used herbicide worldwide, alone or in combination with other plant restricted translocation herbicides (Almeida et al., 2015). This herbicide inhibits the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSPs), which is systemic rather than selective (Rodrigues & Almeida, 2018).

Due to the optimization of herbicide spraying operation, the used application volumes have been getting lower, resulting in increasingly concentrated spray solutions. However, an increased concentration may interfere with the effectiveness (Almeida et al., 2014) and safety of applications due to the quality of spray solution distribution and deposition.

Spray solution physicochemical characteristics need to be considered to evaluate application efficiency, as they can interfere with the generated droplet spectrum and deposition, as well as the absorption and penetration of droplets by leaves. Among the main properties evaluated for herbicides are droplet size, surface tension, and contact angle, as they may influence the interaction of the spray solution with the target surface (Marques et al., 2012; Costa et al., 2017; Cunha et al., 2017).

Surface tension is the result of differences in intermolecular interactions of liquid-air interphase molecules with molecules within the liquid, i.e., surface tension is a reflection of cohesive forces in a liquid (Adamson & Gast, 1997; Behring et al., 2004; Kronberg et al., 2014). In general, the lower the surface tension is, the higher the ability of a liquid to spread over a surface

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Area Editor: Renildo Luiz Mion
Received in: 6-26-2019
Accepted in: 11-28-2019
(Decaro Junior et al., 2015; Calore et al., 2015; Vechia et al., 2016). Spreading can be measured by the contact angle formed between a droplet and the surface where it has been deposited. However, the surface characteristics must be taken into account, as spreading is also a function of interfacial characteristics (liquid and surface component substances), as well as surface tension (Iost & Raetano, 2010; Oliveira et al., 2019). Thus, spray solution spreading on plants depends on both surface tension of the liquid and anatomical constituents and biochemical composition of the epidermis (Kissmann, 1998; Behring et al., 2004; Barreto et al., 2017; He et al., 2019), which may also interfere with the differential penetration of products deposited on plants (Marques et al., 2012).

Thus, this study aimed to evaluate the effect of glyphosate concentrations in the spray solution on the drift potential of sprayed droplets, surface tension of spray solution, and contact angle of droplets on Urochloa ruziziensis leaves, paraffin film, and glass surfaces.

MATERIAL AND METHODS

The studies were carried out in the Nucleus of Study and Development in Application Technology (NEDTA) of the Department Fitossanidade - UNESP, Campus of Jaboticabal - SP, Brazil.

The effect of five acid equivalent (a.e.) concentrations of glyphosate was evaluated on spray solution characteristics and drift potential of sprayed droplets. For this, three experiments were carried out under laboratory conditions in a completely randomized design. The first and second experiments evaluated droplet size and surface tension of spray solutions of five glyphosate concentrations (2.44, 4.88, 9.75, 19.5, and 39 g a.e. L⁻¹ of spray solution) (Roundup Ultra®, 650 g a.e. kg⁻¹, WG, Monsanto), with 12 and 4 replications, respectively. The third experiment evaluated the contact angle of droplets in a 5 × 3 factorial scheme (glyphosate concentrations in the spray solutions vs. surfaces), with four replications. The evaluations were carried out on two artificial surfaces (glass coverslips and paraffin film) and U. ruziziensis leaves.

The five acid equivalent concentrations of glyphosate (2.44, 4.88, 9.75, 19.5, and 39 g L⁻¹ of spray solution) correspond to application volumes of 400, 200, 100, 50, and 25 L ha⁻¹, respectively. A dose of 975 g a.e. ha⁻¹ of glyphosate is sufficient to desiccate a cover plant such as U. ruziziensis (Silva et al., 2013; Timossi et al., 2016).

Experiment 1: Evaluation of droplet size and drift potential

Spray solution jet quality and of drift potential attributes (MVD, %V<100 µm, and Span), evaluated by droplet production by hydraulic energy spray nozzles in the treatments, are named in the text as droplet spectrum. A MagnaJet AD-1A 110007 air-induction flat fan spray tip, commonly used by farmers to apply glyphosate at a working pressure of 300 kPa, providing a flow rate of 0.29 L min⁻¹, was used in this experiment. Spraying was driven with compressed air, and the pressure was maintained constant at 320 kPa in the spray tip by a precision pressure regulator to obtain a flow rate of 0.300 L min⁻¹. The distance between the spray nozzle and the point of droplet interception by the laser was 0.4 m (FAO, 1998). The laser intercepted droplets produced across the full range of the jet (50%), moving the nozzle transversely. The time taken to move from side to side of the applied jet was set to three seconds. Twelve replications were obtained with each glyphosate concentration in the spray solution (2.44, 4.88, 9.75, 19.5, and 39 g a.e. L⁻¹ of spray solution), being four readings in three copies of the spray tip model.

Droplet diameter was determined by the laser diffraction method during the sprayed droplet passing through the sampling region of a particle size analyzer (Mastersizer S, Malvern Instruments Co.). In this method, the degree of diffraction that the light ray undergoes is inversely proportional to the particle size (Fernandes et al., 2007). Data decoding, according to the algorithm developed for droplet diameter characterization by laser diffraction, was processed and tabulated directly by the program integrated with the particle size analyzer (Mastersizer S®, version 2.19).

The median volumetric diameter (MVD), coefficient of uniformity of sprayed droplets (Span), and percentage volume of droplets with diameter smaller than 100 µm (%V<100 µm) were measured in the emitted spray solution. Span was determined by [eq. (1)]:

\[
\text{Span} = \frac{Dv0.9 - Dv0.1}{Dv0.5}
\]

Where:

- \(Dv0.1\) is the droplet diameter such that 10% of the sprayed volume is composed by droplets of smaller diameter;
- \(Dv0.5\) is the droplet diameter such that 50% of the volume of the sprayed liquid consists of droplets larger or smaller diameter, and
- \(Dv0.9\) is the droplet diameter such that 90% of the sprayed volume is composed by droplets of smaller diameter (Matthews et al., 2016).

Spraying was driven by compressed air, and the pressure was maintained constant with a precision pressure regulator. The distance between the spray nozzle and the point of droplet interception by the laser was maintained at 0.4 m. The laser intercepted droplets produced across the full range of the jet, moving the nozzle transversely. The time taken to move from side to side of the applied jet was set to three seconds.

Experiments 2 and 3: Determination of surface tension of spray solutions and contact angle of droplets

The surface tension of spray solutions at five acid equivalent concentrations (a.e.) of glyphosate (2.44, 4.88, 9.75, 19.5, and 39 g a.e. L⁻¹ of spray solution) was evaluated with four replications. The contact angle of droplets was evaluated in a 5 × 3 factorial scheme (glyphosate concentrations in the spray solution vs. surfaces), with four replications. The evaluations were carried out on two artificial surfaces (glass coverslips and paraffin film) and U. ruziziensis leaves.

Surface tension and contact angle were determined using an automatic tensiometer (Contact Angle System OCA 15-Plus, Dataphysics®). The equipment was equipped with a digital camera with a 752 x 582-pixel and temporal resolution of 30 images s⁻¹. Images were captured at a distance of 0.1 m and processed using the software SCA20®. The variables were determined with the
droplets formed by a 500-µL precision syringe (Hamilton®). Each droplet had a 5-µL volume of spray solution.

Surface tension was determined by the pendant drop method, in which the image of the droplet formed at the tip of the needle is captured by the digital camera and analyzed by axis asymmetry. Surface tension is calculated based on the Yang-Laplace equation as a function of the deformation of droplets emitted in each sampling (Adamson & Gast, 1997; Ferreira et al., 2013).

Three surfaces with different characteristics were used for the analysis of the contact angle of droplets: U. ruziziensis leaf, glass cover slip (one new coverslip per treatment), and paraffin film (a resistant mixture of plastic paraffin with waterproof paper – Parafilm M®). After the droplet (5 µL) is formed, it hung on the tip of the needle. Thus, for the droplet to settle on the surfaces, their bases hanging on the needle was touched to them. The syringe was rapidly withdrawn upwards using a hand-held conveyor after applying the droplet, followed by measurements. The contact angle has been used to characterize surfaces for affinity to aqueous spray solution droplets and their spreading over the surface. The surface is called hydrophilic (high spread) when this angle is lower than 90°, otherwise it is called hydrophobic (Tang et al., 2008).

Plants of the species U. ruziziensis were grown for four months at the full sun in pots filled with substrate composed of sand, soil, and cattle manure in a ratio of 3:1:1, with daily irrigation until close to field capacity. During this period, three plant cuts were carried out at 0.05 m from the soil. After each cut, maintenance fertilization was performed according to technical recommendations for the crop (Sousa & Lobato, 2004). Leaf harvesting was carried out after U. ruziziensis was grown for 45 days after the last cut.

Considering the genetic variability among U. ruziziensis genotypes, which may consequently change the morphology and anatomy of plant leaves (Santos et al., 2013), the penultimate leaves from the base to the upper part with visible ligule were harvested to determine the contact angle of spray solutions. They were sectioned into longitudinal rectangles of approximately 0.05 × 0.01 m. These sections were arranged horizontally in individual stretchers to reduce undulation that compromised leaf surface structure and image capture for contact angle readings. Images were evaluated every second for 60 seconds after each droplet was deposited individually on the leaf surface. The analysis of surface tension and contact angle considered four evaluation times (5, 15, 30, and 60 seconds) after droplet formation (surface tension) or deposition (contact angle). The mean temperature and relative humidity during the readings were 26.9±2 °C and 47±2%, respectively.

Data analysis

The results obtained in the evaluations were submitted to analysis of variance by the software Assistat 7.7 to detect the significance of glyphosate concentrations in the spray solution for the variables of the droplet spectrum, surface tension, contact angle, and the interaction between factors (surfaces vs. concentrations).

In the event of a significant interaction of the contact angle results between the studied factors, the concentrations were sliced within each surface by nonlinear regression. Models were selected considering the significance of the analysis of variance of the regression, coefficient of determination (R²), and the significance of their parameters using the Student’s t-test.

The values of droplet spectrum (MVD, %V<100 µm, and Span) and surface tension of spray solutions were adjusted to a quadratic model (Equation 2), using the software Sigma Plot 12.3:

\[ y = c + a.x + b.x^2 \]

Where:

- \( y \) is the dependent variable;
- \( x \) is the independent variable, and
- \( a, b, \) and \( c \) are estimated model parameters.

The values of contact angle as a function of glyphosate concentrations in the spray solution on the adaxial surface of U. ruziziensis, paraffin film, and glass coverslips were fit to hyperbolic (Equation 3), quadratic with inverse function (Equation 4), and exponential models (Equation 5), respectively, using the software Sigma Plot 12.3.

\[ y = \frac{a.b}{(b+x)} \]
\[ y = \left(\frac{a}{x^2}\right) + \left(\frac{b}{x}\right) + c \]
\[ y = a.exp(-b.x) + c \]

Pearson’s linear correlation was calculated between MVD, %V<100 µm, and Span, surface tension of spray solutions and MVD, and surface tension of spray solutions and contact angle of droplets, separately for each surface at 60 seconds. The significance of correlations was calculated by the Student’s t-test.

RESULTS AND DISCUSSION

The evaluations of the effect of glyphosate concentrations on spray solution characteristics and drift potential of the sprayed droplets showed differences in the droplet spectrum (Table 1). By increasing glyphosate concentrations in the spray solution (Figures 1A to 1C), MVD, %V<100 µm, and Span responded quadratically with significance ≤ 0.06.

| Variable       | Droplet spectrum |          |          |          |
|----------------|------------------|----------|----------|----------|
|                | MVD (µm) | %V<100 µm | Span     |
| F for droplet spectrum | 118.8497 ** | 76.6068 ** | 21.3106 ** |
| CV (%)         | 6.06    | 12.67    | 11.47    |

TABLE 1. F-values and coefficients of variation (CV%) applied to the means of median volumetric diameter (MVD), percentage volume of droplets smaller than 100 µm (%V<100 µm), and coefficient of uniformity (Span).
**Significant at 1% significance.**

Increasing herbicide concentrations from 2.44 to 19.5 g a.e. L\(^{-1}\) reduced MVD from 548 to 369 µm, respectively, while a spray solution at a concentration of 39 g a.e. L\(^{-1}\) resulted in an MVD of 372 µm. The response of %V<100 µm and Span to glyphosate concentrations in the spray solution was inverse to MVD.

![Graph](image)

**FIGURE 1.** (A) Median volumetric diameter of droplets, (B) percentage volume of droplets smaller than 100 µm, and (C) coefficient of uniformity (Span) provided by concentrations of 2.44, 4.88, 9.75, 19.5, 39 g L\(^{-1}\) of acid equivalent (a.e.) of glyphosate with the spray tip ADIA 110007.

Considering the droplet size classification established by the standard S572.1 (ASABE, 2009), prepared by droplet spectrum analysis generated by reference spray tips, glyphosate concentrations in the spray solution provided MVD classified as extremely thick (2.44 and 4.88 g a.e. L\(^{-1}\) of spray solution), very thick (9.75 g a.e. L\(^{-1}\) of spray solution), and thick (19.5 and 39 g a.e. L\(^{-1}\) of spray solution).

Herbicide concentration in the spray solution acts with different intensities on MVD for different spray tip models. Almeida et al. (2016) observed that the increased concentration of the herbicide diquat from 2 to 4 g a.i. L\(^{-1}\) of spray solution resulted in a reduction in the size of droplets generated by air-induction tips from 599 to 590 µm, respectively, while tips that generate droplets without air induction increased MVD from 166 to 190 µm.

Yu et al. (2009a; 2009b) found that deposit formation by droplets on waxy and hairy leaf surfaces is influenced by droplet size associated with spray solution characteristics. In this sense, Xu et al. (2010) found that 300-µm droplets had a spreading time (2.6 s) 9.7 times faster than 600-µm droplets (25.3 s) for the wet area in the interveinal surface area of the adaxial surface of Syringa reticulata leaves. From the biological point of view, changes in droplet size associated with glyphosate concentrations in the spray solution may influence the relationship between glyphosate absorption and translocation differently in different plant species and, consequently, their control effectiveness and/or intoxication (Prasad & Cadogan 1992; Liu et al., 1996; Feng et al., 2003).

Pearson’s correlation coefficient was negative between MVD and %V<100 µm and Span in the order of −0.99, with significance levels lower than 1% by the t-test. Thus, there is a proportional increase in %V<100 µm and Span as MVD is reduced, leading to a higher drift potential.

Glyphosate concentrations in the spray solution influenced the surface tension at all evaluated times (Table 2), possibly due to the proportional increase in the concentration of surfactants in the inert of the commercial product formulation containing glyphosate used in this research (Oliveira et al., 2015). Thus, surface tension values as a function of concentrations fit a quadratic model (Figures 2A to 2D), with significances of the F-test of regression of 0.05, 0.11, 0.11, and 0.09 at 5, 15, 30, and 60 seconds, respectively.

**TABLE 2.** F-values and coefficients of variation (CV%) applied to the means of surface tension (mN m\(^{-1}\)) of the spray solutions.

| Variable | Time after droplet formation | \(F_{\text{calculated}}\) for surface tension | CV (%) |
|----------|-----------------------------|---------------------------------------------|--------|
|          | 5 s                         | 15 s                                       | 30 s   | 60 s   |
|          | 19.6916 **                  | 25.2160 **                                 | 37.5053 ** | 28.1776 ** |
|          | 1.21                        | 0.95                                       | 0.71   | 0.79   |

**Significant at 1% probability level.**

The quadratic fit of the surface tension (Figures 2A to 2D) was similar to MVD (Figure 1A). Thus, at 5, 15, 30, and 60 seconds, spray solutions with 2.44 to 19.5 g a.e. L\(^{-1}\) provided a reduction in surface tension, while the concentrated spray solution (39 g a.e. L\(^{-1}\)) showed an increase compared to 19.5 g a.e. L\(^{-1}\).
The positive correlation between surface tension and MVD was 0.79, with a significance level lower than 5% by the t-test. It indicates that MVD decreases (Figure 1A) as glyphosate concentrations reduce spray solution surface tension (Figures 2A to 2D). On the contrary, higher surface tension results in increased droplet size. A better droplet uniformity is also observed, with a reduction in the values of %V<100 µm (Figures 1B to 1C).

The results of this research corroborate those of Oliveira et al. (2015), who found an inversely proportional correlation between surface tension and MVD using different concentrations of associated phytosanitary products. Therefore, reducing the volume of application and maintaining the dose of phytosanitary products will increase the concentration in the spray solution, which results in the alteration of its physicochemical characteristics, such as surface tension and droplet spectrum, leading to a higher drift potential (Griesang et al., 2017).

The contact angle showed a significant interaction (p>0.01) between glyphosate concentrations and surfaces (U. ruiziensis leaves, paraffin film, and glass) at the four analyzed times (Table 3). The interactions indicate that contact angle is dependent on target surface characteristics, such as polarity and physicochemical characteristics of spray solutions, such as surface tension (Kissmann, 1998; Behring et al., 2004; He et al., 2019; Iost & Raetano, 2010; Decaro Junior et al., 2015; Barreto et al., 2017; Oliveira et al., 2019).

The values of contact angle as a function of glyphosate concentrations in the spray solution on the adaxial surface of the U. ruiziensis leaf, paraffin film, and glass coverslips were adjusted to hyperbolic, quadratic inverse, and exponential models, respectively. The significance of the F-test of regressions was lower than 0.04, with coefficients of determination higher than 0.81 (Table 4 and Figure 3).
Table 4. Parameters and coefficient of determination of models adjusted to the values of contact angle as a function of glyphosate concentrations in the spray solution on the adaxial surface of U. ruziziensis leaves, paraffin film, and glass coverslips.

| Surface           | Time (s) | Adjusted model       | Parameter  | Coefficients of determination |
|-------------------|----------|----------------------|------------|-------------------------------|
|                   |          |                      |  $\alpha$  |  $\beta$  |  $\gamma$  | R² (adjusted) |
| U. ruziziensis    | 5        | Hyperbolic           | 1120.3422  | 45.9328  |            | 0.81         |
|                   | 15       | Hyperbolic           | 1121.6473  | 40.4663  |            | 0.82         |
|                   | 30       | Hyperbolic           | 1120.0308  | 35.5429  |            | 0.85         |
|                   | 60       | Hyperbolic           | 1119.9706  | 34.1261  |            | 0.85         |
| paraffin film     | 5        | Inverse quadratic   | 1116.0009  | 135.5550  | 37.5144    | 0.99         |
|                   | 15       | Inverse quadratic   | 1119.9827  | 137.1265  | 73.7168    | 0.99         |
|                   | 30       | Inverse quadratic   | 1116.1813  | 15.5122   | 72.2157    | 0.99         |
|                   | 5        | Exponential         | 1118.6952  | 8.3196    | 71.4982    | 0.99         |
|                   | 15       | Exponential         | 1164.7339  | 0.1621    | 75.6767    | 0.95         |
|                   | 30       | Exponential         | 1165.9952  | 0.1940    | 5.4116     | 0.96         |
|                   | 5        | Exponential         | 1166.7183  | 0.2295    | 15.1496    | 0.92         |
|                   | 15       | Exponential         | 1158.6852  | 0.2303    | 14.8171    | 0.92         |

†Roman numerals represent the p-value of the significance of parameters by the t-test.

The behavior of deposited droplets, represented by the contact angle, resulted from the interaction between surface and concentrations (Figure 4). Glyphosate concentrations in the spray solutions promoted a reduction in the contact angle on the adaxial surface of U. ruziziensis leaf in the four evaluated times. On the other hand, artificial surfaces (paraffin film and glass coverslip) showed a stabilization in the reduction of contact angle at all analyzed times from the glyphosate concentration of 9.75 g a.e. L⁻¹ of spray solution (Figure 4).

![Graph](image_url)

FIGURE 3. Contact angle on three surfaces as a function of concentrations of 2.44, 4.78, 9.75, 19.5, and 39 g L⁻¹ of acid equivalent (a.e.) of glyphosate in the spray solution.

Contact angles of droplets on the adaxial surface of U. ruziziensis leaf were high compared to those obtained on glass coverslips for all concentrations and evaluated times (Figure 3). It is due to leaf characteristics, such as high wax content in the epicuticular layer and pilosity (Figure 4). The contact angle of droplets on the adaxial surface of U. ruziziensis leaf was high compared to the paraffin film only at concentrations of 2.44, 4.88, and 9.75 g a.e. L⁻¹ (Figures 3 and 4). In other words, the increase in glyphosate concentration resulted in higher spreading on the leaf, which is explained by the high correlation with surface tension. This droplet behavior in the glass coverslip, which resulted in the lowest values than the other surfaces (Figures 3 and 4), is due to the possible hydrophilic characteristic of the glass. This behavior was not observed on paraffin film due to its hydrophobic characteristics.
Droplets of spray solutions sprayed on hydrophilic surfaces provide their higher spreading, with lower contact angle and, consequently, higher interfacial contact, which may result in a higher spray solution absorption and evaporation rate than hydrophobic surfaces. Both the rate of absorption and evaporation are naturally dependent on the specific characteristics of the leaf surface, liquid, and weather conditions (Iost & Raetano, 2010; Decaro Junior et al., 2015; Barreto et al., 2017; Oliveira et al., 2019; He et al., 2019). Because of the high contact angle, *U. ruziziensis* leaf surface may have small areas wetted by droplets (Decaro Junior et al., 2015), thus reducing the evaporation rate under the weather at application time and after it (Yu et al., 2009a; Yu et al., 2009b; Xu et al., 2010; Xu et al., 2011). Therefore, it may allow longer leaf wetness by droplets to affect the penetration of phytosanitary products directly in the epicuticular layer and their absorption and translocation (Liu et al., 1996).

A positive correlation was observed between the contact angle of droplets and surface tension of spray solution in the order of 0.46, 0.83, and 0.87 for *U. ruziziensis* leaves, paraffin film, and glass, respectively. Their respective levels of significance were 23, 4, and 2% by the t-test. It shows that the contact angle for the leaf surface of *U. ruziziensis* has limitations when estimated indirectly through values of spray solution surface tension. Thus, measuring the contact angle directly with appropriate image analysis criteria is important to avoid variable results due to the presence of pilosity, which may confuse the interpretation of the formed angle in the digitally captured image with computerized and automated analysis.

Based on the results of this research, changing glyphosate concentrations in the spray solution leads to changes in the droplet spectrum due to alterations in the physicochemical characteristics of the spray solution, such as surface tension. The contact angle has a different pattern between surfaces and is responsive to glyphosate concentration in the spray solution. Therefore, because the contact angle of droplets is positively correlated with spreading, they may influence target coverage, spray solution retention (Decaro Junior et al., 2015), evaporation, absorption, and translocation. In this sense, special attention is required in night applications, even at low application volumes, as the presence of free water on the leaf surface of *U. ruziziensis* from dew or wetting caused by positive root pressure, or even due to precipitation events, as there may be dilution of the spray solution on the leaf surface, which may lead to dripping, compromising control effectiveness of applications.

**CONCLUSIONS**

Increasing glyphosate concentrations in the spray solution influence droplet spectrum (MVD and %V<100 µm), surface tension, and contact angle of droplets with the surfaces *U. ruziziensis* leaf, paraffin film, and glass. Increasing glyphosate concentrations increase the potential risk of droplet drift and reduce the contact angle of droplets interacting with the surfaces *U. ruziziensis* leaf, glass coverslips, and paraffin film.

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