

EFFICACY OF GLYPHOSATE ON *POPULUS TREMULOIDES* AS AFFECTED BY DROPLET SIZE AND SPRAY VOLUME

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ABSTRACT

A low-volume laboratory research spray system and the spinning disk atomisers Flak, Herbi, and Micromax were used to study the effect of droplet size and spray volume on glyphosate efficacy on trembling aspen (*Populus tremuloides* Michx.). Five droplet sizes, each with a narrow droplet spectrum, ranging from 177 to 1589 μm VMD were used. Spray volumes from 15 to 120 ℓ/ha were tested. The phytotoxicity of glyphosate increased as carrier volume was reduced (thereby increasing herbicide concentration), but was not affected by changes in droplet size. There was no significant interaction between droplet size, spray volume, and active ingredient (a.i.) rate per hectare. It is possible that the concentration gradient between the droplet and leaf, rather than the droplet coverage, is important for glyphosate phytotoxicity.

Keywords: droplet size; spray volume; glyphosate; spinning disk atomiser; spread factor; controlled droplet atomiser; *Populus tremuloides*.

INTRODUCTION

Environmental, economic, and social concerns impel researchers to look at ways to improve herbicide efficacy, especially in light of the fact that so few herbicides are registered for forestry use in Canada. Research has focused on the effect of droplet size and spray volume on herbicide activity. Small spray droplets are more prone to off-target drift than larger droplets, resulting in damage to non-target species and reduced on-target efficacy. Large droplets have lower risk of drift but involve a reduction in coverage, which may lower efficacy. Low spray volumes allow more hectares to be treated per aircraft per spray session, which in turn can lead to greater overall productivity within the narrow window in which maximum efficacy and crop tolerance occur. Application costs would also be reduced. In forestry, the delivery systems currently being used operationally produce droplet volume median diameters (VMDs) from 400 to 2000 microns. The carrier volume for operational aerial applications of glyphosate ranges in rate from 17 to 100 ℓ/ha . The rationale for using a particular droplet size and for using either high or low volume rates has not been established scientifically.

There is evidence to suggest that lower volume rates are more effective for glyphosate in agricultural applications (Buhler & Burnside 1983, 1987; Kudsk 1988; O'Sullivan *et al.* 1981; Sandberg *et al.* 1978; Stahlman & Philips 1979). When glyphosate was applied as individual droplets, its efficacy increased with increasing herbicide concentration (Ambach & Ashford 1982; Merritt 1982). With respect to the droplet size effect, Prasad & Cadogan (1992) found that smaller droplets were more efficacious than larger ones, whereas Kudsk (1988) and Merritt (1982) found no effect of droplet size on glyphosate efficacy.

Much of the research to date has dealt separately with the possible impacts of spray variables such as droplet size, carrier volume, and a.i. rates. In such studies the role of droplet size on efficacy could not be distinguished from the possible influence of the change in coverage (number of droplets) or volume rate. As a more complicated interaction may possibly be occurring between droplet size, volume rate, and a.i. rate, these three factors should be examined together.

The objective of the present study was to examine the influence of droplet size, spray volume, and a.i. rate, and their interactions on the phytotoxicity of glyphosate on trembling aspen, a major competitor of conifer plantations in Canada (Campbell 1990).

MATERIALS AND METHODS

Spray System Calibration

Spread factor determination

Measuring droplet diameters directly is not routinely practical when analysing the droplet spectrum of a given applicator. Instead, using a dyed spray solution, droplets are collected as stains on Kromekote cards. The droplet diameter is then derived by dividing the diameter of the stain by its spread factor. Spread factors may vary with droplet size and spray-mix concentration. To derive these spread factors two methods were employed to estimate droplet diameter. For droplets from 326 to 977 µm, a radioactive method was used and for droplets from 985 to 1684 µm, a syringe method was used.

Radioactive method: Radiolabelled glyphosate (Monsanto Agricultural Products Co., St. Louis, Missouri) with *N*-phosphonomethyl carbon labelled (^{14}C) and an activity level of $3.44 \times 10^5 \text{ Bq}/\text{mmol}$ was used. Droplets from 550 to 977 µm were generated using a microapplicator, Model M0130 (ISCO Instrumentation Specialties Company, Lincoln, NB, USA). Those from 326 to 549 µm were produced using an acoustic droplet generator (National Aeronautical Establishment, Ottawa, Ontario, Canada). Both devices are capable of producing a range of individual mono-sized droplets. Four droplet-sizes were used ranging from 326 to 977 µm. Solutions of formulated glyphosate (Vision®*) in a series of concentrations were spiked with a known activity of ^{14}C -glyphosate. Using these solutions, from 5 to 50 droplets of each size were collected in vials and their radioactivity was measured. Droplet volume was calculated from the radioactivity of the original solution, the radioactivity of the collected droplets, and the number of droplets in each vial. The corresponding droplet diameter was derived from the formula for the volume of a sphere.

* This product produced by Monsanto is identical in composition to Roundup® (isopropylamine salt of glyphosate + the ethoxylated tallow amine surfactant MON 0818)

Immediately after the deposition of droplets into the vial, 20 droplets of the same size were deposited on a Kromekote card. The diameter of these stains was determined under a microscope. Spread factors were calculated for the different droplet sizes by dividing the diameter of the stain by the diameter of the droplets. Regression analysis was conducted to determine the relationship between droplet size and spread factor.

Microsyringe method: A 5.0- μl syringe (Hamilton Company, Reno, Nevada, USA) was used to create droplets of 0.5 μl (985 μm), 1.0 μl (1241 μm), 1.5 μl (1420 μm), 2.0 μl (1563 μm), and 2.5 μl (1684 μm) onto the Kromekote cards. The stain sizes were measured and spread factors were determined as above.

Spray droplet size calibration

The Greenhouse Low-volume Laboratory Research Spray System or "cart sprayer" (Campbell *et al.* 1994) was designed to deliver sprays of uniform droplets in chosen sizes by using one of several controlled-droplet atomisers. The 177- μm droplets were created using a Flak spinning disc atomiser (Micron Sprayers Ltd, Bromyard, Herefordshire, UK). The 353- μm droplets were produced by using a Micron Herbi 77 spinning disc atomiser (Micron Sprayers Ltd, Bromyard, Herefordshire, UK). By varying the voltage to the nozzle, droplets of 394 to 1589 μm were generated using a Micromax spinning disc atomiser (Micron Corporation, Houston, Texas, USA).

Sprayer calibration is normally based on collecting a desired dosage rate on a two-dimensional target (i.e., litres per hectare), and is therefore concerned only with the vertical vector of the emitted spray. Spinning disc atomisers, though, produce droplets by centrifugal force, and these droplets are emitted in a lateral trajectory. For small droplets (i.e., less than 400 μm), this initial horizontal component is quickly overcome by friction before gravity begins to take over. The momentum of larger heavier droplets, however, causes them to maintain their inertia in the face of opposing frictional forces and thus travel farther in a lateral trajectory than smaller droplets emitted in the same way. This allows the downward force of gravity to come into play, thus creating a diagonal vector for the droplet trajectory. Since the idealised shape of target plants is that of upright cylinders separated in space, these large droplets would impinge on the plants in three-dimensional space rather than the two-dimensional surface on which sprayer calibrations were based. The result would be a higher-than-expected dose per plant.

With the spray system used here, it was not practical to raise the nozzle high enough to allow the lateral trajectory of the droplet to dissipate before it reached the target. Instead, during spraying the long axes of individual plants were shielded inside open-ended cylinders (1.2 m tall \times 0.35 m diameter) made from 0.35-cm corrugated fibreglass sheets. Any shading of the canopy top by the cylinder rim as the front half of the spray cone passed over would be compensated for by reverse-directed spray of the back half of the cone. Sprayer calibrations were conducted using the upper mouth of the cylinder as the target surface.

Kromekote cards were again used to collect the spray. More than 160 stains were measured for each application. Droplet sizes were calculated by dividing the stain diameter by the spread factor. Droplet size uniformity was expressed by the standard deviation or by plotting the droplet spectrum.

Sprayer walking-speed calibration

When using the cart sprayer, the effective target zone is confined to a 45-cm-wide strip in the centre of the swath. Swath profiles (Fig. 1) of the Flak, Herbi, and Micromax show that this section receives the most uniform deposit. To calibrate the speed at which the cart sprayer was to move to achieve the desired application rates, deposit in the effective target zone was measured with a patternator. As there was a potential for error due to evaporation with this method, patterns were also checked using a more laborious spectrophotometric method. Both methods gave very similar results.

For the patternator method, a corrugated fibreglass sheet (1×3 m) was positioned 1.2 m beneath the spray atomiser, parallel to the swath line, and sloped slightly. Without moving

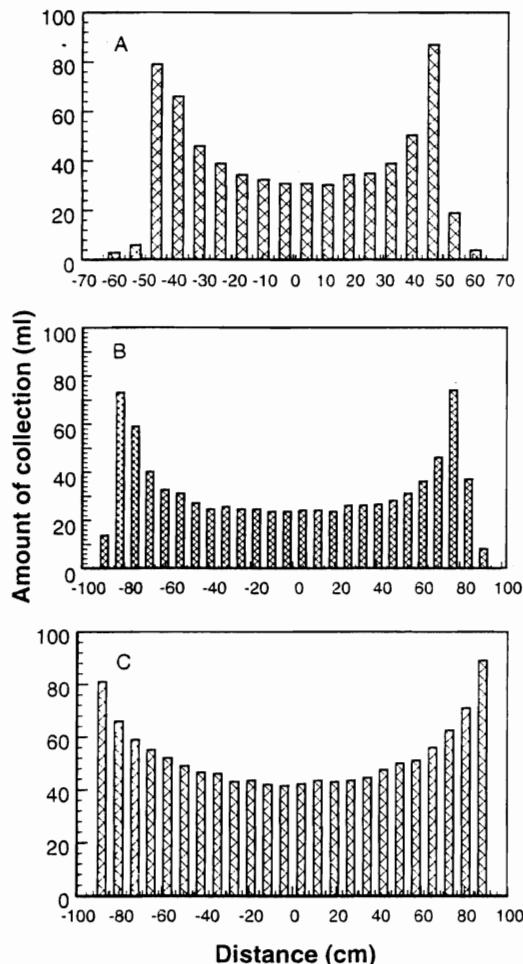


FIG. 1—Distribution pattern from Flak (A), Herbi (B), and Micromax (C) spinning disc atomisers. A patternator was used. Application parameters: spray mix = 0.1 kg a.e. glyphosate (as Vision®) in 30 l water; flow rate = 30 ml/min for Flak and 60 ml/min for Herbi and Micromax; rpm (revolutions per minute) = 4930, 2035, and 817 for Flak, Herbi, and Micromax, respectively; distance between patternator and nozzle = 1.2 m.

the cart, each spray solution was allowed to run through the sprayer at a given flow rate setting for 30 minutes. The furrows of the corrugated sheet channelled the spray from the effective target zone into beakers placed along the down-slope end of the sheet. In order to reduce the potential for errors due to evaporation, the distance that the liquid had to move on the surface of the fibreglass was minimised by positioning the atomiser head vertically over the lower edge of the sheet (i.e., only one-half of the emission was collected). The amount of spray solution collected in each beaker was measured and the swath profile and effective flow rate in the target zone were determined. Walking-speed could then be calculated to achieve the desired application rate.

In the spectrophotometric method, metal baking trays ($6 \times 22 \times 22$ cm; teflon-coated) were placed end to end down the centre of the spray swath and sprayed at a given walking speed and flow rate with a glyphosate solution containing 0.2% (w/v) tartrazine (Sigma Chemical Co. St. Louis, MO, USA). Immediately after spraying, the trays were rinsed with 50 ml distilled water. Aliquots of these rinsates were analysed for absorbance in a Spectronic 20 spectrophotometer at a wavelength of 429 nm. The corresponding dye concentrations were derived from standard curves and the spray deposit per unit area was calculated.

Phytotoxicity Experiments

Plant materials

Aspen plants were grown from seed collected in the vicinity of Sault Ste. Marie, Ontario, Canada. One month after sowing the seeds on to a soil-filled tray in the greenhouse, individual seedlings were transplanted to plastic pots (15×15 cm) filled with a mixture of potting soil (PRO-MIX BX, Premier Brands Inc., Stamford, CT, USA) and sand (3:1). Plants were irrigated daily and supplemented with fertiliser once every 2 weeks at the rate per pot of 250 ml fertiliser solution (Plant-Prod, all-purpose soluble fertiliser concentrate with chelated micronutrients, N-P-K = 20-20-20, Plant Products Company Limited, Brampton, Ontario, Canada) 1.5 g/litre. Greenhouse temperatures ranged between 18–28°C during the day and 15–22°C at night. Daylength in the greenhouse was maintained at a minimum of 14 h with supplemental lighting, while the humidity was ambient.

Aspen is a perennial species and can readily resprout if the roots are not killed. To control this weed effectively, root kill is therefore essential. Preliminary studies indicated that aspen should have an initial height of 1–1.2 m to ensure that stems will resprout after being cut to just above ground level. From the time of transplanting, this growth takes about 3 months to achieve under the greenhouse conditions described above. As the sample plants were grown they were trellised using metal rods to ensure an erect posture and thereby minimise contact with the walls of the cylinder shields during treatment.

Plant treatment and harvesting

Five droplet sizes of 177 (± 13), 394 (± 22), 667 (± 34), 1013 (± 105), and 1589 (± 130) μm in diameter and five spray volumes of 15, 30, 60, 90, and 120 ℓ/ha were evaluated. Sublethal a.i. rates of glyphosate, formulated as Vision®, were used. The rate of active ingredient was controlled by varying the concentration of herbicide in the spray solution. The spray volume rate, i.e., the volume deposited per unit area, was controlled by varying the cart speed or the number of passes with the cart and the flow rate. Tartrazine dye was added to all spray

solutions at a concentration of 0.2% (w/v). A Kromekote card and glass plate ($0.5 \times 20 \times 20$ cm) were placed at the upper mouth of an empty plant hood to verify droplet sizes and spray volumes for each treatment.

Three weeks after herbicide application, the stems of all plants from each treatment were cut to 10 cm above soil level. Four weeks after cutting, the fresh weight of regrowth was recorded and this value was used as a measure of root kill. The experiments employed a completely randomised block design, with 10 replicate plants sprayed in each treatment. Analyses of variance were carried out on all phytotoxicity data, using GLM procedures of Statistical Analysis System (SAS Institute Inc., Cary, NC, USA).

RESULTS AND DISCUSSION

Spray Calibration

Spread factor determination

With droplet sizes from 326 to 977 μm , there was a linear relationship between the droplet size and spread factor. The spread factor was greater with the larger droplets. With droplet sizes from 985 to 1684 μm , spread factor did not change with droplet diameter. There was no variation in spread factor due to the herbicide concentrations used in the phytotoxicity experiments of this study.

Spray droplet size calibration

Very narrow droplet spectra were obtained for all six different sizes of droplets produced by Flak, Herbi, and Micromax atomisers (Fig. 2). For example, 87% of the total number of droplets consisted of drops with diameters between 160 and 180 μm for the Flak atomiser (Fig. 3A) and 91% of total number of drops consisted of drops with diameters from 330 to 360 μm created with Herbi atomiser (Fig. 3B).

The generation of a large range of mono-sized droplets was always a limitation in previous studies on droplet size effects. These studies applied different droplet sizes mainly either by changing flat fan nozzles (Kudsk 1988), or by applying herbicide as individual drops (Merritt 1982; Prasad & Cadogan 1992). Flat fan nozzles produce wide droplet spectra. Droplet generators cannot simulate spray condition. In the present study, uni-sized droplets ranging from 177 to 1584 μm were sprayed.

Phytotoxicity Experiments

Effect of droplet size and spray volume

Change in droplet size did not alter glyphosate efficacy ($p = 0.67$) (Fig. 4). Phytotoxicity increased with decreasing spray volume rate ($p < 0.0001$). The interaction between these factors was not significant ($p = 0.99$). These results supported the findings of Kudsk (1988) and Merritt (1982).

At a fixed carrier volume, the total droplet coverage increased as droplet size decreased (Table 1). For example, if a spray volume of 30 ℓ/ha is used, an increase in droplet size from 117 to 1589 μm causes a four-fold decrease in droplet coverage. Since the herbicide concentration in the droplets does not change, the concentration gradient remains constant.

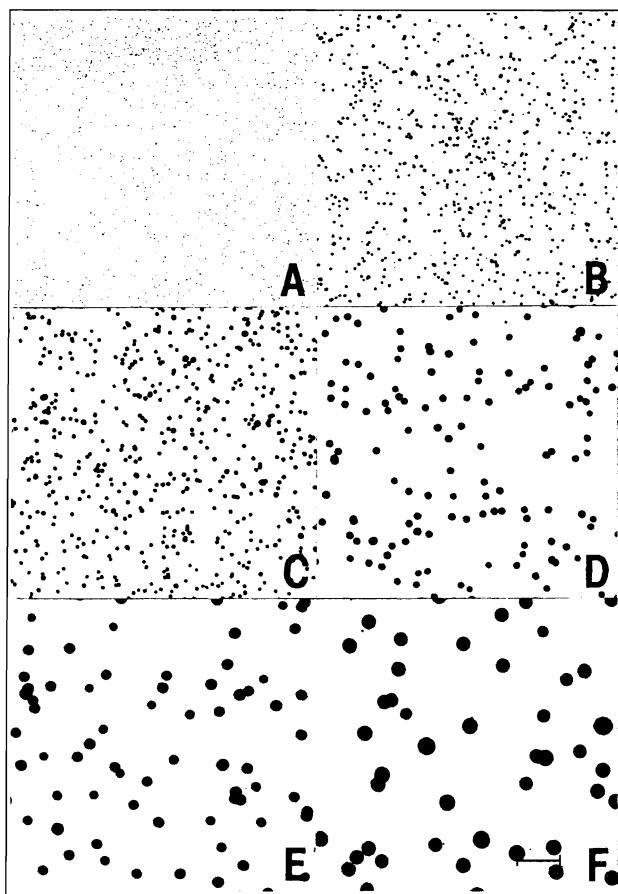


FIG. 2—Spray droplets collected on Kromekote cards. Flak (A), Herbi (B), and Micromax (C to F) atomisers were used. Spray mix = 0.1 kg a.e. glyphosate (as Vision®) in 30 l water with 0.2% (w/v) Erio Acid Red (EAR) dye (Ciba-Geigy Dyes Ltd, Dorval, Quebec, Canada); rpm = 4930 and 2035 for Flak ($177 \pm 13 \mu\text{m}$ [A]) and Herbi ($353 \pm 15 \mu\text{m}$ [B]), respectively. For Micromax, rpm = 1212, 817, 620, and 422 for droplets of $394 \mu\text{m}$ (C), $667 \mu\text{m}$ (D), $1013 \mu\text{m}$ (E), and $1589 \mu\text{m}$ (F), respectively. Scale bar = 1 cm.

TABLE 1—Relationship between droplet size, number of drops on unit area, and leaf area coverage. The spray volume was taken as 30 l/ha.

Droplet size (μm)	Number of droplets (No./ m^2)	Spread factor	Leaf area coverage (%)
177	1.034×10^6	1.77	7.94
394	9.372×10^4	2.03	4.70
667	1.932×10^4	2.36	3.76
1013	5.515×10^3	2.64	3.10
1589	1.429×10^3	2.64	1.97

These results suggest that the herbicide concentration gradient is more important than coverage in glyphosate phytotoxicity.

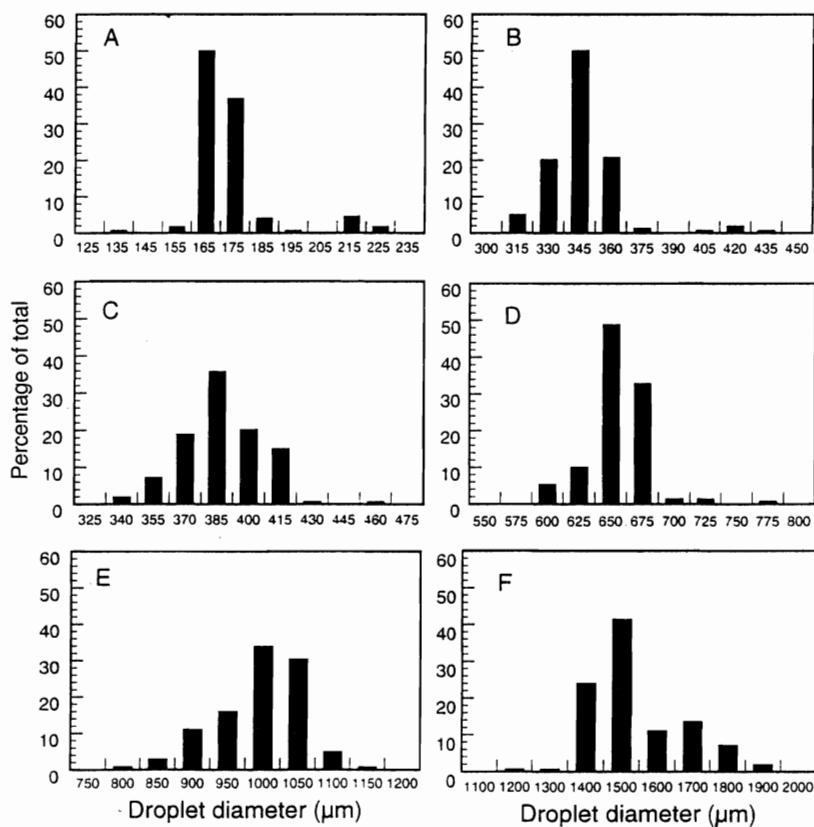


FIG. 3—Droplet spectra from the sprays corresponding to Fig. 2. More than 15 stains were measured for each spray application.

On the other hand, when droplet size is fixed and the carrier volume rate is increased, the droplet number and the droplet coverage also increase. However, at the same time, the glyphosate concentration gradient between the droplet and leaf decreases (in this trial as the spray volume increased from 15 to 120 l/ha, the glyphosate concentration decreased by a factor of eight).

Effect of droplet size and a.i. rate

Five droplet sizes ranging from 177 to 1589 μm in diameter and five a.i. rates ranging from 0.03 to 0.15 kg a.e./ha were tested. The spray volume rate (30 l/ha) was held constant. Droplet size had no effect on efficacy ($p = 0.18$) (Fig. 5). The interaction between the droplet size and a.i. rate was not significant ($p = 0.99$). Similar results were reported by Merritt (1982).

At a fixed a.i. rate, the number of droplets increases as droplet size decreases. More leaf surface area can be covered by a large number of small droplets than a small number of large droplets (Table 1). The lack of droplet size effect in this experiment suggests that this coverage is not important in glyphosate action.

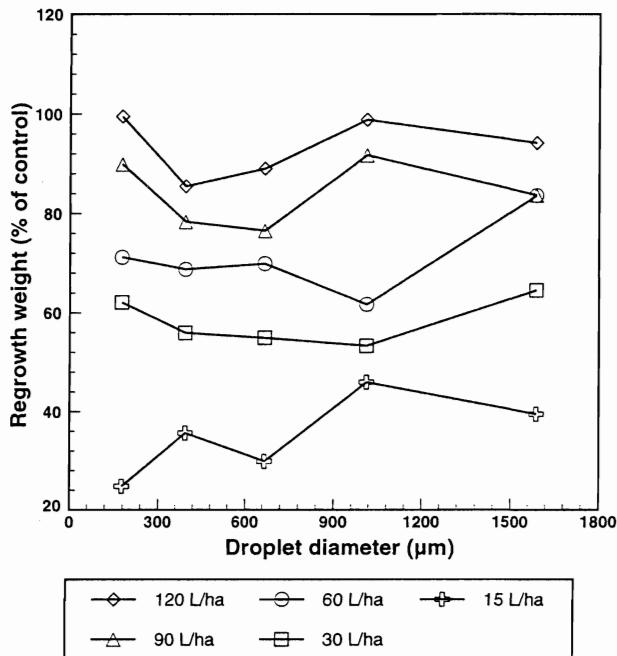


FIG. 4—Effect of droplet size and spray volume on efficacy of glyphosate on aspen. The a.i. rate was kept constant at 0.075 kg/ha. Each point represents mean of 10 measurements.

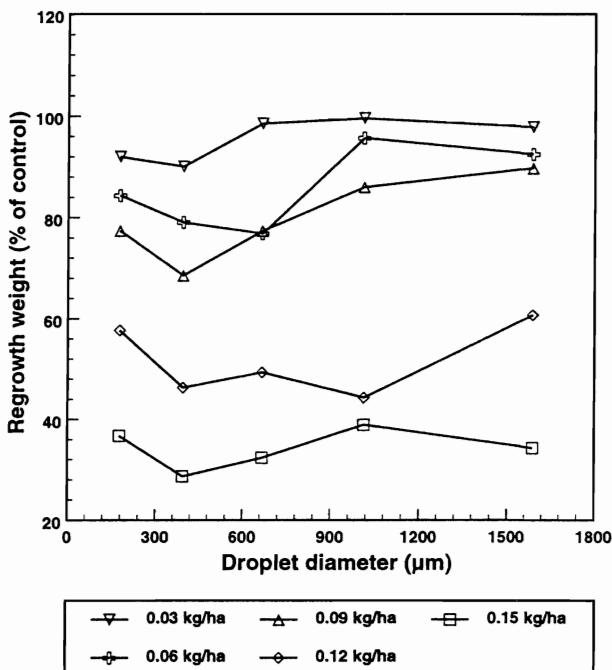


FIG. 5—Effect of droplet size and a.i. rate on the efficacy of glyphosate on aspen. Spray volume was held constant at 30 L/ha. Each point represents 10 measurements.

Effect of spray volume and a.i. rate

Droplet diameter was kept constant at 353 (± 15) μm . Five spray volumes ranging from 15 to 120 L/ha and five a.i. rates ranging from 0.04 to 0.12 kg a.e./ha were tested. Glyphosate efficacy increased with decreasing carrier volume rate ($p < 0.0001$) (Fig. 6). For example, the efficacy of 0.04 kg/ha in 15 L/ha is greater than that of 0.12 kg/ha in 120 L/ha and equivalent to that of 0.06 kg/ha in 30 L/ha (a commonly used operational spray volume for aerial forestry herbicide application in Canada). The interaction between the carrier volume rate and a.i. rate was not significant ($p = 0.61$).

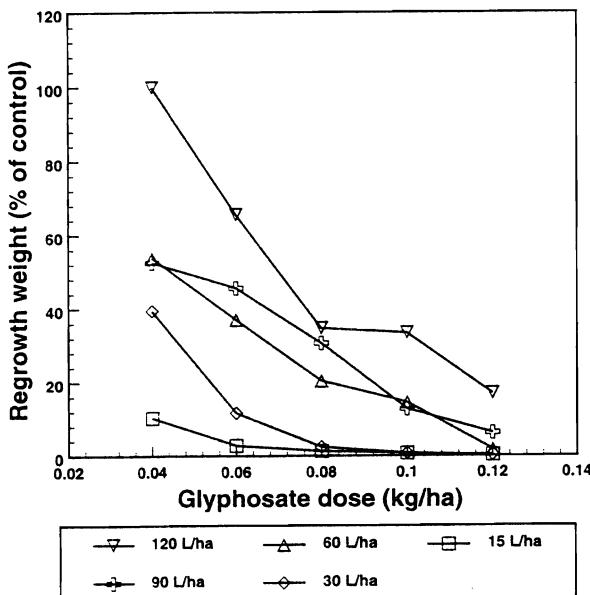


FIG. 6—Effect of spray volume and a.i. rate on efficacy of glyphosate on aspen. Droplet size was held constant at 353 μm . Each point represents 10 measurements.

As carrier volume rate decreases, coverage also decreases which means our data show an inverse relation between coverage and efficacy of glyphosate. Moreover, as the volume rate decreases, the concentration of glyphosate and surfactant in the spray droplets increases (by a factor of 8 in this experiment as the volume decreased from 120 to 15 L/ha). In Fig. 7 the results from this experiment are expressed in terms of efficacy v. glyphosate concentration in the spray mix. Over the range of a.i. rates tested, efficacy increased as glyphosate concentration increased, with maximum efficacy being obtained at 0.2–0.4 %.

Most studies on the effects of spray volume have been conducted using the same droplet size, changing herbicide concentration or spray solution concentration and droplet number (as in the present experiment) (Ambach & Ashford 1982; Buhler & Burnside 1983, 1987; Kudsk 1988; Merritt 1982; O'Sullivan *et al.* 1981; Sandberg *et al.* 1978; Stahlman & Philips 1979). It was suggested that glyphosate efficacy was enhanced by decreasing the spray volume and thereby increasing herbicide concentration. The present study supports this finding.

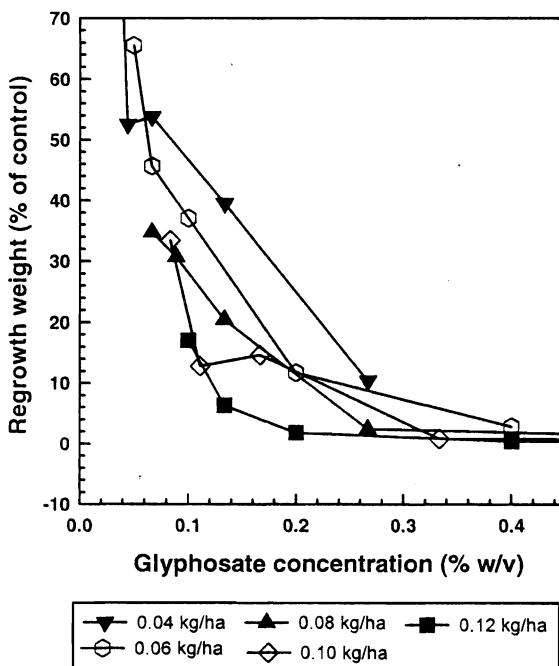


FIG. 7—Effect of glyphosate concentration and a.i. rate on efficacy of glyphosate on aspen. Conditions as for Fig. 6.

Increasing coverage by increasing spray volume has generally improved the efficacy of a number of systemic herbicides (Knoche 1994). However, glyphosate showed the opposite trend, which indicated that coverage is not the only factor affecting herbicide performance. Factors such as an increased concentration gradient of herbicide and/or surfactant between droplet deposit and the leaf surface, as indicated by the present study, also may be involved. As our study was designed to determine the effect of spray volume on efficacy of the only formulation of glyphosate available for forestry use in Canada, we were not able to distinguish between effects caused by increased herbicide concentration *v.* increased surfactant concentration. However, both factors would seem to be important as two studies which examined glyphosate and surfactant concentration independently found that increased concentration of either component enhanced efficacy (Jordan 1981) and glyphosate uptake (Gaskin & Holloway 1992) for particular rate and concentration ranges of the two components.

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