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Generic dose response curves for predicting effects of herbicides on weeds or sensitive plant species

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Abstract

Computer-based decision support systems are an important tool for ensuring good practice in the aerial application of pesticides. By providing access to herbicide-plant dose-response curves through such systems, users are able to predict the effects of herbicides on sensitive plants outside of the spray area and weeds within the target area. However, difficulties arise when the operational scenario requires input of a herbicide/plant combination that is not found within the dose-response database. The purpose of the study described here was to evaluate whether the existing dose-response database could be used to derive generic dose-response curves for use in agriculture, horticulture or forestry.

Each dose-response curve was characterised using a term labelled "*s*", the ratio of herbicide doses required to give a 5% and a 95% yield reduction of the test plant species, and an index dose (in this case the dose giving a 50% yield reduction). The value of *s* differed considerably between trials with a range from 4.8 to 2800 (e.g. the upper extreme indicates that the dose required to give 5% yield reduction must be increased by a factor of 2800 for a 95% yield reduction).

Three generic dose-response curves were derived based on the mean value and the upper and lower extremes of *s*. With this information plus an estimate of the index dose (i.e. the dose required to reduce plant yield to 50% or some other selected value) an average, or extreme dose-response curve can be generated.

Keywords: AGDISP; dose response; herbicide; weed control

Introduction

Aerial application of herbicides is an important management tool in forestry and many other productive sectors (e.g. arable, pastoral, horticultural). Although herbicide application provides many benefits, the risk to sensitive environments from off-site spray drift is of ongoing concern (Bird et al., 1996), and, therefore, it is important for applicators to use best practice techniques.

Spray drift can be defined as the movement of pesticide droplets through the air at the time of application to locations outside of the target zone. Definition of best-practice spray application methods to minimise spray drift requires understanding of factors that influence spray droplet movement. The general principles of spray drift and deposition are well understood (Matthews, 1979; Yates et al., 1967). However, the large numbers of interacting factors that influence droplet movement (e.g. Teske & Barry, 1993; Teske et al., 1998) make accurate quantification of exposure of non-target vegetation to herbicides downwind of a target zone difficult. Nevertheless, this quantification is needed for accurate risk assessment. For this reason, over the last 20 years there has been substantial

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investment in the development and validation of aerial spray application simulation models.

The agricultural dispersal (AGDISP) model (Bilanin et al., 1989; Teske et al., 2003) is the most commonly used and well validated (Bird et al., 1996; 1999; Hewitt et al., 2002; Richardson et al., 1995) modelling system developed to calculate deposition of material from aerial pesticide application. The power of AGDISP is its ability to simulate the aircraft wing tip vortices that largely control near field movement of spray material (i.e. movement of spray material close to the aircraft). The strength of these vortices dissipates with time and distance from the aircraft. At distances beyond a few hundred metres downwind of the spray line, the vortices are weak enough that the prevailing meteorological conditions control further movement of the remaining airborne fraction of spray material.

The AGDISP model can be used to evaluate the effect of different operational (e.g. release height, nozzle type) and meteorological variables on spray drift. However, models that predict only spray deposition and drift are of limited value for risk assessment, because they only calculate exposure and provide no interpretation of the associated biological consequences. To overcome its current limitations, the AGDISP computational engine has been incorporated into a number of geographical information system (GIS)-based decision support systems such as SpraySafe Manager (SSM) (Ray et al., 1999; Schou et al., 2001) or Spray Advisor (Schou et al., 2009). In other words, the development of a more functional interface accessing supporting data can make the power of AGDISP more relevant to a range of end-users.

An innovation introduced into SSM about 15 years ago, and more recently into Spray Advisor, was the inclusion of a database that contained experimentally derived models relating the response of a number of plant species to specific herbicides. Combining herbicide exposure and biological response information enables users to undertake effective risk assessments and to minimise the likelihood of causing spray drift damage through choice of appropriate application equipment, operating conditions, or through definition of buffer zone (i.e. no-spray zone) width. However, there are significant practical limitations to this approach. For example, undertaking experiments to generate dose-response curves (Streibig, 1989), that describe the effects of different herbicide rates on plant species, is a time consuming and expensive exercise. Even for a single herbicide formulation and plant species, there are many factors that influence the dose-response relationship such as plant size, plant health, weather conditions, physiological status of the plant, adjuvants used in the spray mixture, and the response variable (Streibig & Kudsk, 1993).

Although the existing dose-response database covers over 50 combinations of herbicide type, formulation and plant species, it is still limited in terms of the range of scenarios actually encountered by users when they need to evaluate the likelihood of drift damage to non-target plant species. Individual plant dose-response curves are also not robust in terms of accounting for the range of factors that can influence plant sensitivity to herbicides (as described above).

The purpose of this study was to evaluate whether the existing dose-response database, initially developed for SSM, could be used to derive generalisations on the response of plants to various herbicide application rates that could be useful for risk assessments.

Methods

The dose-response data (for herbicide effectiveness) were derived from a series of trials each testing a different combination of herbicides and plant species (Tables 1 and 2) following methods described in Ray et al. (1996, 1999). In brief, test plants were generally raised in a glasshouse from seed or occasionally purchased as small seedlings from a nursery. In all cases, they were transplanted as small seedlings into square pots (180 x 180 x 180 mm) containing a commercial potting mix (Yates Bloom). Plants were either treated in the “young seedling” stage or “mature” stage. Young seedling plants were herbaceous species expected to be particularly herbicide-sensitive and therefore represented a worst-case scenario from a plant damage perspective. Conversely, field-grown, mature woody plants would be more typical of the growth stage of many plants actually targeted by herbicide treatments. Plants in the young seedling stage were kept well-watered in a glasshouse environment and treated at about 21 days after transplanting. Plants in the mature stage were kept in an outdoor environment, regularly watered, and treated about 4 – 6 months after transplanting. Control plants, which received no herbicide, were also grown. Generally, ten replicate plants were used for each dose, and forty replicate plants were used for the control. Control plants did not receive any treatment other than regular watering. At the time of treatment, 10 plants were randomly selected to estimate plant dry weight prior to treatment. Dry weights were obtained for each remaining plant at a fixed period (generally one month) after the herbicide application. In some cases, plant heights were also recorded before and after treatment but the analysis of these plants was handled in the same general way as plants where dry weight was the only response variable.

Herbicides were applied using a “track sprayer”. This sprayer consisted of an aspirated cabinet containing a variable speed conveyer belt with a TeeJet 80015E nozzle mounted above it. Plants were placed on the conveyor belt which transported them under the nozzle, operated at 200 kPa. Belt speed and nozzle emission were constantly monitored. The nozzle height was

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TABLE 1: Alphabetical summary of herbicides, additives and plant species used in the trials

Herbicide (Product)	**Additive (Manufacturer)**	**Common Name (Species)**
Glyphosate (Roundup)	Boost Penetrant (Dow AgroSciences)	Bean (*Vicia faba* L.)
Glyphosate (Roundup G2)	Pulse Penetrant (Nufarm Technologies, USA)	Browntop (*Agrostis capillaris* L.)
Hexazinone (Velpar 90)	Uptake (Dow AgroSciences NZ)	Coprosma (*Coprosma robusta* Raoul)
Hexazinone (Velpar DF)		Kanuka (*Kunzea ericoides* (A.Rich.) Joy Thomps.)
Metsulfuron (Escort)		Lettuce (*Lactuca sativa* L.)
Picloram (Picloram amine WSG)		Manuka (*Leptospermum scoparium* Forst.)
Quizalofop (Targa)		Radiata pine (*Pinus radiata* D. Don)
Terbuthylazine (Gardoprim)		Ryegrass (*Lolium perenne* L.)
Triclopyr (Grazon)		Sweetcorn (*Zea mays* L.)
		Tomato (*Solanum lycopersicum* L.)
		Wineberry (*Aristotelia serrata* (J.R.Forst. et G.Forst.) W.R.B.Oliv.)

adjusted to 50 cm above the average plant height. The swath width produced by the nozzle was measured at average plant height and the belt speed adjusted to give an application rate (total spray volume) of 200 L ha^{-1}.

In most cases, ten doses covering the range of interest were chosen, with the dose rates being increased using an exponential scale. In many cases a preliminary experiment was needed to identify this range (i.e. the doses needed to cover the range of plant response from no effect to complete growth suppression or mortality). Combinations of plants and their growth stages, and herbicides, additives, and their rates of application are shown in Table 2.

Analysis

The following response function has been found to perform well for experiments relating herbicide dose to plant response, such as dry weight, (Streibig et al. 1993):

$$y = c + d\,\exp(a + b\ln(x+k)) / (1 + \exp(a + b\ln(x+k))) \quad [1]$$

This is a logistic function using the log of the herbicide application rate, x, as the independent variable. The response variable, y, used in our study was generally the plant weight assessed at the end of the trial, (for one test of glyphosate on *Pinus radiata*, the response was the increase in plant height during the period of the trial). Since ln(0) is undefined, a small value for k, equal to half the minimum application rate, was included in the equation to allow the control treatment ($x = 0$) to be included in the analysis. The upper asymptote of the equation (the value of y when x is very large) is $c + d$, and lower asymptote (the value of y when x is very small) is d. The proportionate reduction in response between the upper and lower asymptotes for a given herbicide rate is:

$$r(x) = \exp(a + b\ln(x+k)) / (1 + \exp(a + b\ln(x+k))) \quad [2]$$

By rearranging Equation [2] to solve for x, the rate at which there is a p% reduction in response from the untreated control and the lower asymptote can be shown to be:

$$x_p = (p/100/(1/r(k) - p/100))^{1/b} \exp(-a/b) - k \quad [3]$$

The percentage reduction is controlled by the parameters a and b. The parameters c and d are not of general interest. To simplify comparisons between experiments, comparisons were made between the rate at which a 50% reduction in response occurred (x_{50}), and the ratio of the rates at which a 5% reduction and a 95% reduction occurred ($s = x_{95}/x_5$). The parameter x_{50} can be considered a location parameter, while s quantifies the slope of the response curve.

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TABLE 2: Summary of doses of herbicide and additives applied to a range of plant species

Herbicide	Additive rate (%)	Species	Plant type	Min. rate (kg ai ha^{-1})[1]	Max. rate (kg ai ha^{-1})[1]
Metsulfuron (Escort)	Pulse (0.2)	Bean	Herbaceous	0.0000006	0.018
Metsulfuron (Escort)	Pulse (0.2)	Coprosma	Woody	0.000000012	0.00018
Metsulfuron (Escort)	Pulse (0.2)	Kanuka	Woody	0.000000012	0.00018
Metsulfuron (Escort)	Pulse (0.2)	Manuka	Woody	0.000000012	0.00018
Metsulfuron (Escort)	Pulse (0.2)	Radiata	Woody	0.000006	0.18
Metsulfuron (Escort)	Pulse (0.2)	Ryegrass	Herbaceous	0.00012	0.036
Metsulfuron (Escort)	Pulse (0.2)	Sweetcorn	Herbaceous	0.000036	0.012
Metsulfuron (Escort)	Pulse (0.2)	Tomato	Herbaceous	0.0000006	0.018
Metsulfuron (Escort)	Pulse (0.2)	Wineberry	Woody	0.000000006	0.00018
Terbuthylazine (Gardoprim)	-	Browntop	Herbaceous	0.0005	15
Terbuthylazine (Gardoprim)	-	Tomato	Herbaceous	0.00015	5
Triclopyr (Grazon)	Boost (1)	Grapes	Woody	0.00006	0.6
Triclopyr (Grazon)	Boost (1)	Tomato	Herbaceous	0.00006	1.8
Picloram (Picloram amine WSG)	-	Tomato	Herbaceous	0.0000243	0.81
Glyphosate (Roundup)	Pulse (0.5)	Beans	Herbaceous	0.000036	1.08
Glyphosate (Roundup G2)	Pulse (0.5)	Browntop	Herbaceous	0.000036	1.08
Glyphosate (Roundup G2)	Pulse (0.5)	Coprosma	Woody	0.00036	3.6
Glyphosate (Roundup G2)	Pulse (0.5)	Kanuka	Woody	0.00036	3.6
Glyphosate (Roundup)	Pulse (0.5)	Lettuce	Herbaceous	0.00036	1.08
Glyphosate (Roundup G2)	Pulse (0.5)	Manuka	Woody	0.00036	3.6
Glyphosate (Roundup G2)	Pulse (0.5)	Radiata	Woody	0.000108	3.6
Glyphosate (Roundup)	Pulse (0.5)	Ryegrass	Herbaceous	0.00036	1.08
Glyphosate (Roundup G2)	Pulse (0.5)	Ryegrass	Herbaceous	0.000036	1.08
Glyphosate (Roundup)	Pulse (0.5)	Sweetcorn	Herbaceous	0.00036	1.08
Glyphosate (Roundup)	Pulse (0.5)	Tomato	Herbaceous	0.00002668	0.8004
Glyphosate (Roundup)	Pulse (0.5)	Tomato	Herbaceous	0.000036	1.08
Glyphosate (Roundup G2)	Pulse (0.5)	Wineberry	Woody	0.00036	3.6
Quizalofop (Targa)	Uptake (2)	Browntop	Herbaceous	0.000015	0.15
Quizalofop (Targa)	Uptake (2)	Ryegrass	Herbaceous	1.25E-09	0.00015
Hexazinone (Velpar 90)	-	Tomato	Herbaceous	0.00027	9
Hexazinone (Velpar DF)	-	Browntop	Herbaceous	0.000225	7.5

[1] ai = active ingredient.

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The percentage reduction in response can be written using x_{50} and s as:

$$p(x) = \frac{100\exp(2\ln(x - x_{50})(\ln(0.05) - \ln(0.95))/\ln(s))}{1 + \exp(2\ln(x - x_{50})(\ln(0.05) - \ln(0.95))/\ln(s))} \quad [4]$$

The objective of the study was to evaluate whether any generalisations could be made from the range of dose-response curves tested, using the parameters x_{50} and s. Analysis of variance was used to determine whether herbicide or plant type (categorised as either herbaceous or woody plants) influenced x_{50} and s. For variables that significantly influenced the response variable, Duncan's multiple comparison test was used to identify which means varied significantly at $p = 0.05$.

Modelling approach for evaluating dose-response relationships

The dose-response Equation [1] was fitted to data from each trial. From the estimated parameters a and b, estimates for x_{50} and s were calculated for each trial. Bootstrap standard errors were obtained for each parameter by resampling the residuals (Efron & Tibshirani, 1993). Subsequent analysis was undertaken to define the mean and variance of ln(s) which was assumed to be normally distributed. To determine its variance, it was necessary to take into account that estimates from each trial were, themselves, subject to experimental error. A simple calculation of the variance of the estimates of ln(s) would, therefore, tend to give an inflated value. A method proposed by Efron and Morris (1975) was used to correct for this, which requires known values for the standard errors of each estimate of ln(s). We used the bootstrap standard errors in our analysis.

Results and Discussion

Estimates of ln(x_{50}) and ln(s) were calculated for each trial (Table 3), along with bootstrap standard errors. The distribution of s across all trials was highly positively skewed. This suggested that a log transformation, or even a log-log transformation, should be applied to normalise s. The median value of ln(s) was 4.18, corresponding to s = 65. The mean of ln(s) corresponded to s = 115, while the mean of ln(ln(s)) corresponded to s = 38. This suggests that the log transformation did not fully compensate for the skewness in distribution, whereas the log-log transformation over-compensated.

The variance of ln(s) was 5.59, but after correcting for the within-study variance using the method of Efron and Morris (1975), this was reduced to 4.05. The uncorrected and corrected variances for ln(ln(s)) were respectively 0.412 and 0.216. Using the corrected variances, and assuming a normal distribution, the 5 and 95 percentiles of the distribution of s are 4.2 and 3100 for the log transformation, and 5.4 and 2500 for the log-log transformation. Using values midway between those obtained for the two transformations would result in a range for s of between 4.8 and 2800.

From this approach the following general conclusions could be made. A typical dose-response curve based on the median of ln(s) corresponds to s = 65. So, typically, the dose giving a 95% reduction in growth will be 65 times greater than the dose giving a 5% reduction in growth. However, the value of s differs considerably between trials. The likely range is from 4.8 to 2800, or, on the log scale, 1.57 to 7.94.

With picloram omitted from the analysis of variance, (as it is only represented in the dataset once), the only factor analysed that had a marginally significant effect on the slope parameter, s, was chemical type (p = 0.057). Slope was least for quizalofop (ln(s) = 2.38) and greatest for triclopyr (ln(s) = 8.05).

These results indicate that, without quantitative data, it is not realistic to make assumptions as to the nature of the dose-response relationship. However, from information such as that presented here, it is possible to create "average" general dose response curves that are representative of either extremes or typical situations. It is suggested that, for most purposes, a value for ln(s) of 4.18 will be appropriate, though in some cases, either of the extremes (1.57 or 7.94) may be more suitable. The typical and extreme curves are shown in Figure 1.

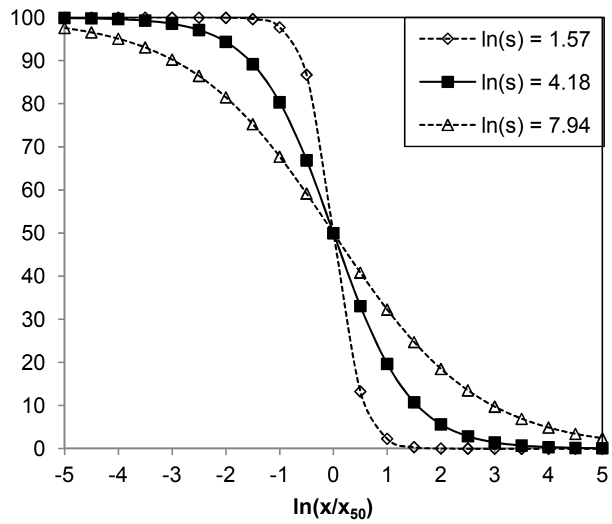

FIGURE 1: Typical and extreme dose-response curves.

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TABLE 3: Estimates of ln(x_{50}) and ln(s) and bootstrap standard errors for the function describing the relationship between herbicide dose and plant species.

Herbicide	Species	Date of application	ln(x_{50})	s.e.	ln(s)	s.e.
Metsulfuron	Beans	13 Oct 1995	-9.01	0.2	3.63	1.45
Metsulfuron	Coprosma	21 Nov 1997	-12.89	0.22	4.17	0.90
Metsulfuron	Kanuka	21 Nov 1997	-14.46	0.15	4.16	1.33
Metsulfuron	Manuka	21 Nov 1997	-13.90	0.47	7.29	2.25
Metsulfuron	Radiata pine	07 Jun 1998	-6.16	0.13	2.17	1.13
Metsulfuron	Ryegrass	22 Mar 1996	-5.73	0.24	3.49	1.00
Metsulfuron	Sweetcorn	16 Dec 1994	-7.21	0.41	5.51	2.12
Metsulfuron	Tomato	02 Nov 1995	-7.83	0.25	1.60	0.86
Metsulfuron	Wineberry	21 Nov 1997	-16.19	0.41	5.74	1.14
Terbuthylazine	Browntop	26 Feb 1997	-0.48	0.36	4.75	2.23
Terbuthylazine	Tomato	10 Feb 1997	-1.64	0.23	9.93	1.07
Triclopyr	Grapes	19 Jan 1996	-6.08	0.52	6.76	1.96
Triclopyr	Tomato	03 Nov 1995	-2.94	0.74	9.34	2.35
Picloram	Tomato	22 May 1996	-0.32	0.06	4.19	1.06
Glyphosate	Beans	13 Oct 1995	-5.07	0.32	5.97	2.16
Glyphosate	Browntop	01 Dec 1995	-2.11	0.11	3.83	0.54
Glyphosate	Coprosma	21 Nov 1997	-0.70	0.26	7.45	1.70
Glyphosate	Kanuka	21 Nov 1997	-2.66	0.21	6.71	0.63
Glyphosate	Lettuce	21 Nov 1994	-1.85	0.28	1.42	1.23
Glyphosate	Manuka	21 Nov 1997	-2.78	0.75	6.84	2.52
Glyphosate	Radiata	17 Mar 1999	-0.94	0.16	1.67	0.89
Glyphosate	Ryegrass	16 Dec 1994	-4.57	0.17	3.85	1.18
Glyphosate	Ryegrass	22 Mar 1996	-3.80	0.23	3.29	2.54
Glyphosate	Sweetcorn	16 Dec 1994	-4.00	0.57	2.00	2.46
Glyphosate	Tomato	02 Nov 1995	-2.51	0.14	4.02	0.90
Glyphosate	Tomato	23 Mar 1998	-2.32	0.13	5.51	0.75
Glyphosate	Wineberry	21 Nov 1997	-1.91	0.51	7.43	2.44
Quizalofop	Browntop	27 Jun 1997	-6.05	0.28	2.47	1.05
Quizalofop	Ryegrass	15 Apr 1997	-12.95	0.09	2.29	1.08
Hexazinone	Tomato	10 Feb 1997	-3.22	0.13	8.45	0.58
Hexazinone	Browntop	26 Feb 1997	-1.52	0.15	4.76	1.1

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The rate producing a 50% reduction in growth, ln (x_{50}), was strongly related to chemical type ($p < 0.0001$) but not plant type (Table 4). Values ranged from -1.06 for terbuthylazine to -10.38 for metsulfuron and values of ln(x_{50}) for metsulfuron and quizalofop, were significantly lower than those of all other chemicals.

TABLE 4: Influence of herbicide on the rate needed to produce a 50% reduction in growth (ln(x_{50})).

Chemical	**Mean[1]**
Metsulfuron	-10.38 (1.33) a
Quizalofop	-9.50 (3.45) a
Triclopyr	-4.51 (1.57) b
Glyphosate	-2.71 (0.37) b
Hexazinone	-2.37 (0.85) b
Terbuthylazine	-1.06 (0.58) b

[1] *The mean is shown with the standard error in brackets. Means followed by the same letter do not differ significantly at p = 0.05.*

For the practical application of these results it will be assumed that the user has enough information to provide a sensible value for x_{50}, the rate producing a 50% reduction in growth. A value of s based on the above analysis can then be chosen (e.g. ln(s) = 4.18). Equation [4] can then be used to generate a suitable dose-response curve. This procedure can be easily modified to use x_5, x_{95}, or indeed any other dose response rate in place of x_{50}.

Despite the large range in the value of s, in situations where there is an absence of appropriate data the described approach allows calculation of dose-response based on average, best-case, or worst-case scenarios. While further studies to try and explain the factors contributing to the size of s may be worthwhile, experience suggests that these factors will always be difficult to generalise.

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