**APPENDIX 2-6. Atrazine Species Sensitivity Distribution Analysis for Terrestrial Plants**

SSDs were fit to toxicity data for terrestrial plants exposed to atrazine. Five distributions were tested and a variety of methods were used to determine whether different subsets of data should be modeled independently. Ultimately, the results from the vegetative vigor and seedling emergence SSDs were used.  **Table 1** provides a summary of the results.

**Table 1. Summary statistics for SSDs fit to atrazine test results.**

|  |  |  |
| --- | --- | --- |
| **Statistic** | **Vegetative Vigor** | **Seedling Emergence** |
| Best Distribution (by AICc) | Normal | Gumbel |
| Goodness of fit P-value | 0.813 | 0.7602 |
| CV of the HC05 | 0.2717 | 0.4707 |
| HC05 | 0.0242 | 0.0037 |
| HC10 | 0.0347 | 0.0047 |
| HC50 | 0.1233 | 0.0146 |
| HC90 | 0.4376 | 0.0855 |
| HC95 | 0.6958 | 0.169 |

1. Data

Data used in this analysis are detailed in **Tables 9** and **10** (at the end of the document) and were from registrant submitted studies as well as an ECOTOX query (**APPENDIX 2-2**). **Table 2** provides the distribution of the test results for atrazine including the number of species represented. Distributions were created for terrestrial plants based on IC25 values for various metrics of growth from 14- and 28-d seedling emergence studies and 14-, 28-, and 42-d vegetative vigor studies.

**Table 2. Distribution of test results available for atrazine.**

|  |  |  |
| --- | --- | --- |
| **Data Subset** | **Test results** | **Species** |
| Vegetative Vigor | 93 | 36 |
| Seedling Emergence | 16 | 9 |

**Figures 1** and **2** show the distribution of test results among species from the vegetative vigor and seedling emergence studies, respectively, indicating that several species have been repeatedly tested (seven species have been tested at least 5 times each), but the majority of species have been tested fewer than three times.

 

**Figure 1. Distribution of the number of test results per species in atrazine vegetative vigor data.**



**Figure 2. Distribution of the number of test results per species in atrazine seedling emergence data.**

Five potential distributions for the atrazine data were considered, including log-normal, log-logistic, log-triangular, log-gumbel, and Burr. To fit each of the first four distributions, the toxicity values were first common log (log10) transformed. Finally, effect thresholds and five quantiles from the fitted SSDs (HC05, HC10, HC50, HC90, HC95) were calculated and reported.

1. Comparison of distributions using AICc

Akaike’s Information Criterion corrected for sample size (AICc) was used to compare the five distributions for the aquatic plant dataset. For these comparisons all SSDs were fit using maximum likelihood. The AICc suggested that the normal distribution provided the best fit for the vegetative vigor data and the gubmel distribution provided the best fit for the seedling emergence data (**Tables 3 and 4**).

**Table 3. Comparison of distributions for vegetative vigor toxicity data for atrazine.**

| **Distribution** | **AICc** | **∆AICc** | **Weight** | **HC05** |
| --- | --- | --- | --- | --- |
| normal | -45.0266 | 0 | 0.3265 | 0.0242 |
| logistic | -44.9734 | 0.0532 | 0.3179 | 0.0234 |
| gumbel | -43.1765 | 1.8501 | 0.1295 | 0.0282 |
| triangular | -42.9126 | 2.1140 | 0.1135 | 0.0235 |
| burr | -42.8993 | 2.1273 | 0.1127 | 0.0264 |

**Table 4. Comparison of distributions for seedling emergence toxicity data for atrazine.**

| **Distribution** | **AICc** | **∆AICc** | **Weight** | **HC05** |
| --- | --- | --- | --- | --- |
| gumbel | -39.6212 | 0 | 0.2971 | 0.0037 |
| triangular | -39.4532 | 0.1680 | 0.2732 | 0.0028 |
| logistic | -38.8917 | 0.7295 | 0.2063 | 0.0025 |
| normal | -38.7859 | 0.8353 | 0.1957 | 0.0027 |
| burr | -34.8808 | 4.7404 | 0.0278 | 0.0036 |

1. Goodness of fit

The plot of the cumulative distribution functions for the best-fit distributions (as determined by AICc) suggest little evidence of lack-of-fit (**Figures 3** and **4**). Bootstrap goodness-of-fit tests did not show evidence for lack-of-fit (P-values > 0.05, **Tables 5** and **6**) for any of the distributions. The coefficient of variation for the HC05 was below 1 for all distributions.



**Figure 3. Log-normal SSD for atrazine toxicity values for terrestrial plants at the vegetative vigor life stage.** Black points indicate single toxicity values. Red points indicate average of multiple toxicity values for a single species. Blue line indicates full range of toxicity values for a given taxon.



**Figure 4. Log-gumbel SSD for atrazine toxicity values for terrestrial plants at the seedling emergence.** Black points indicate single toxicity values. Red points indicate average of multiple toxicity values for a single species. Blue line indicates full range of toxicity values for a given taxon.

**Table 5. Range of HC05 values for atrazine SSDs for terrestrial plants at the vegetative vigor life stage.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Distribution** | **Method** | **HC05** | **SE** | **CV** | **Lower CI** | **Upper CI** | **P** |
| Normal | ML | 0.0242 | 0.0066 | 0.2717 | 0.015 | 0.0407 | 0.8132 |
| Normal | MO | 0.0237 | 0.0063 | 0.2657 | 0.0143 | 0.0386 | 0.7802 |
| Normal | GR | 0.0213 | 0.0061 | 0.2844 | 0.0108 | 0.0346 | 0.6424 |
| Logistic | ML | 0.0234 | 0.0071 | 0.3052 | 0.0132 | 0.0415 | 0.6683 |
| Logistic | MO | 0.0242 | 0.007 | 0.2893 | 0.0131 | 0.0394 | 0.8002 |
| Logistic | GR | 0.0203 | 0.0058 | 0.287 | 0.0089 | 0.0318 | 0.8002 |
| Triangular | ML | 0.0235 | 0.0063 | 0.2672 | 0.0177 | 0.0414 | 0.0629 |
| Triangular | MO | 0.023 | 0.0058 | 0.2536 | 0.0142 | 0.0373 | 0.5784 |
| Triangular | GR | 0.022 | 0.0057 | 0.2583 | 0.0129 | 0.0354 | 0.4745 |
| Gumbel | ML | 0.0282 | 0.0051 | 0.1828 | 0.0209 | 0.0409 | 0.2607 |
| Gumbel | MO | 0.0333 | 0.0066 | 0.199 | 0.0227 | 0.0489 | 0.2787 |
| Gumbel | GR | 0.0302 | 0.0061 | 0.2022 | 0.0177 | 0.0408 | 0.3077 |
| Burr | ML | 0.0264 | 0.0082 | 0.3117 | 0.0146 | 0.0463 | 0.5764 |

ML=maximum likelihood, MO= moment estimators, and GR=graphical methods

LCp and UCp=projections of the confidence limits of the HC05 (LCx and UCx) onto the cumulative distribution function of the fitted distribution.

**Table 6. Range of HC05 values for atrazine SSDs for terrestrial plants at the seedling emergence life stage.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Distribution** | **Method** | **HC05** | **SE** | **CV** | **Lower CI** | **Upper CI** | **P** |
| Normal | ML | 0.0027 | 0.0022 | 0.8378 | 0.001 | 0.0098 | 0.3616 |
| Normal | MO | 0.0024 | 0.0021 | 0.881 | 0.000704 | 0.0081 | 0.2887 |
| Normal | GR | 0.0017 | 0.0015 | 0.8828 | 0.000252 | 0.0059 | 0.1998 |
| Logistic | ML | 0.0025 | 0.0021 | 0.8472 | 0.00077 | 0.009 | 0.4595 |
| Logistic | MO | 0.0025 | 0.0023 | 0.9467 | 0.000561 | 0.0094 | 0.4046 |
| Logistic | GR | 0.0015 | 0.0014 | 0.9479 | 8.47E-05 | 0.0053 | 0.2408 |
| Triangular | ML | 0.0028 | 0.0025 | 0.9063 | 0.0015 | 0.0108 | 0.5295 |
| Triangular | MO | 0.0023 | 0.002 | 0.8497 | 0.000793 | 0.0081 | 0.2488 |
| Triangular | GR | 0.0019 | 0.0016 | 0.8306 | 0.000435 | 0.0064 | 0.1688 |
| Gumbel | ML | 0.0037 | 0.0017 | 0.4707 | 0.0021 | 0.009 | 0.7602 |
| Gumbel | MO | 0.0036 | 0.002 | 0.5517 | 0.0016 | 0.0094 | 0.7113 |
| Gumbel | GR | 0.0026 | 0.0015 | 0.5823 | 0.000579 | 0.0067 | 0.3976 |
| Burr | ML | 0.0036 | 0.0022 | 0.606 | 0 | 0.0091 | 0.6454 |

ML=maximum likelihood, MO= moment estimators, and GR=graphical methods

LCp and UCp=projections of the confidence limits of the HC05 (LCx and UCx) onto the cumulative distribution function of the fitted distribution.

1. Test for the need to model results separately by vegetative vigor and seedling emergence life stage

Examination of the cumulative distribution functions plotted on similar axes for vegetative vigor and seedling emergence studies does not support combining the datasets into one distribution. The 95% bootstrap confidence intervals for the separate distributions do not overlap substantially (**Figure 5**). Given these differences, the vegetative vigor and seedling emergence distributions will be used separately.

**Figure 5. SSDs for vegetative vibor IC25s (normal) and seedling emergence IC25s (gumbel) for atrazine**. Blue lines show the distribution and upper and lower confidence interval for species from vegetative vigor studies. Red lines show the distribution and upper and lower confidence interval for species from seedling emergence studies.

1. Calculation of other quantiles

**Tables 7 - 9** provides estimates of the HC05 as well as other quantiles of the fitted SSDs.

**Table 7. Estimated quantiles of the fitted SSDs for atrazine IC25s for terrestrial plants at the vegetative vigor life stage.**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Distribution** | **Method** | **HC05** | **HC10** | **HC50** | **HC90** | **HC95** |
| Normal | ML | 0.0242 | 0.0347 | 0.1233 | 4.38E-01 | 6.27E-01 |
| Normal | MO | 0.0237 | 0.0341 | 0.1233 | 4.46E-01 | 6.41E-01 |
| Normal | GR | 0.0213 | 0.0314 | 0.1233 | 4.83E-01 | 7.12E-01 |
| Logistic | ML | 0.0234 | 0.0355 | 0.1207 | 4.10E-01 | 6.22E-01 |
| Logistic | MO | 0.0242 | 0.0366 | 0.1233 | 4.15E-01 | 6.28E-01 |
| Logistic | GR | 0.0203 | 0.032 | 0.1233 | 4.74E-01 | 7.50E-01 |
| Triangular | ML | 0.0235 | 0.0335 | 0.1502 | 6.73E-01 | 9.59E-01 |
| Triangular | MO | 0.023 | 0.0317 | 0.1233 | 4.79E-01 | 6.61E-01 |
| Triangular | GR | 0.022 | 0.0306 | 0.1233 | 4.96E-01 | 6.90E-01 |
| Gumbel | ML | 0.0282 | 0.0357 | 0.1062 | 5.86E-01 | 1.13E+00 |
| Gumbel | MO | 0.0333 | 0.0409 | 0.1045 | 4.56E-01 | 8.00E-01 |
| Gumbel | GR | 0.0302 | 0.0378 | 0.1061 | 5.35E-01 | 9.93E-01 |
| Burr | ML | 0.0264 | 0.0374 | 0.1173 | 4.39E-01 | 7.01E-01 |

**Table 8. Estimated quantiles of the fitted SSDs for atrazine IC25s for terrestrial plants at the seedling emergence life stage.**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Distribution** | **Method** | **HC05** | **HC10** | **HC50** | **HC90** | **HC95** |
| Normal | ML | 0.0027 | 0.0041 | 0.0176 | 7.61E-02 | 1.15E-01 |
| Normal | MO | 0.0024 | 0.0037 | 0.0176 | 8.32E-02 | 1.29E-01 |
| Normal | GR | 0.0017 | 0.0029 | 0.0176 | 1.07E-01 | 1.78E-01 |
| Logistic | ML | 0.0025 | 0.004 | 0.0157 | 6.25E-02 | 9.99E-02 |
| Logistic | MO | 0.0025 | 0.004 | 0.0176 | 7.64E-02 | 1.26E-01 |
| Logistic | GR | 0.0015 | 0.0028 | 0.0176 | 1.11E-01 | 2.09E-01 |
| Triangular | ML | 0.0028 | 0.004 | 0.0185 | 8.61E-02 | 1.24E-01 |
| Triangular | MO | 0.0023 | 0.0034 | 0.0176 | 9.08E-02 | 1.34E-01 |
| Triangular | GR | 0.0019 | 0.0029 | 0.0176 | 1.07E-01 | 1.63E-01 |
| Gumbel | ML | 0.0037 | 0.0047 | 0.0146 | 8.68E-02 | 1.71E-01 |
| Gumbel | MO | 0.0036 | 0.0046 | 0.0144 | 8.55E-02 | 1.69E-01 |
| Gumbel | GR | 0.0026 | 0.0036 | 0.0152 | 1.44E-01 | 3.41E-01 |
| Burr | ML | 0.0036 | 0.0048 | 0.015 | 7.80E-02 | 1.44E-01 |

**Table 9** provides all of the available IC25 values for vegetative vigor and seedling emergence studies. These data sets are the same as reported in the effects characterization and provide additional data for the formulated products. Values that were included in the SSD (*i.e.,* TGAI) are marked with an \*.

Table 9. Nontarget Terrestrial Plant Seedling Emergence Toxicity (Tier II). All definitive endpoints are used quantitatively in the SSD

|  |  |  |
| --- | --- | --- |
| **Genus** | **Species** | **IC25** |
| Avena | sativa | 0.004 |
| Allium | cepa | 0.009 |
| Lolium | perenne | 0.007 |
| Brassica | oleracea | 0.014 |
| Daucus | carota | 0.003 |
| Cucumis | sativus | 0.013 |
| Lactuca | sativa | 0.0025 |
| Glycine | max | 0.19 |
| Solanum | lycopersicum | 0.034 |
| Avena | sativa | 0.0403 |
| Brassica | oleracea | 0.03 |
| Solanum | lycopersicum | 0.053 |
| Allium | cepa | 0.034 |
| Brassica | oleracea | 0.018 |
| Lactuca | sativa | 0.048 |
| Solanum | lycopersicum | 0.15 |

Table 10. Nontarget Terrestrial Plant Vegetative Vigor Toxicity (Tier II). All definitive endpoints are used quantitatively in the SSD

|  |  |  |
| --- | --- | --- |
| **Genus** | **Species** | **IC25** |
| Lactuca | sativa | 0.003577 |
| Solanum | lycopersicum | 0.00465 |
| Rudbeckia | hirta | 0.00473 |
| Leucanthemum | vulgare | 0.010059 |
| Geum | canadense | 0.01073 |
| Fragaria | virginiana | 0.017883 |
| Bellis | perennis | 0.029123 |
| Lactuca | sativa | 0.029776 |
| Lycopus | americanus | 0.032637 |
| Rumex | crispus | 0.032941 |
| Lactuca | sativa | 0.035767 |
| Rumex | crispus | 0.046578 |
| Solanum | lycopersicum | 0.04918 |
| Lycopus | americanus | 0.051504 |
| Solanum | nigrum | 0.05991 |
| Lycopus | americanus | 0.061698 |
| Helianthus | annuus | 0.064381 |
| Geum | canadense | 0.065275 |
| Eutrochium | maculatum | 0.069746 |
| Prunella | vulgaris | 0.07469 |
| Eutrochium | maculatum | 0.076899 |
| Lycopus | americanus | 0.077793 |
| Geum | canadense | 0.078956 |
| Lactuca | canadensis | 0.086735 |
| Alliaria | petiolata | 0.088523 |
| Lactuca | sativa | 0.089328 |
| Helianthus | strumosus | 0.089417 |
| Solanum | lycopersicum | 0.089775 |
| Poa | compressa | 0.109983 |
| Lycopus | americanus | 0.121965 |
| Phalaris | arundinacea | 0.123396 |
| Centaurea | cyanus | 0.127348 |
| Triticum | aestivum | 0.132338 |
| Bellis | perennis | 0.134126 |
| Bellis | perennis | 0.134448 |
| Digitalis | purpurea | 0.138561 |
| Symphyotrichum | lateriflorum var. lateriflorum | 0.143962 |
| Fragaria | x ananassa | 0.146645 |
| Glycine | max | 0.147539 |
| Rudbeckia | hirta | 0.148531 |
| Raphanus | sativus | 0.158269 |
| Digitalis | purpurea | 0.165226 |
| Phalaris | arundinacea | 0.168999 |
| Alliaria | petiolata | 0.177047 |
| Geum | canadense | 0.188134 |
| Elymus | lanceolatus | 0.194107 |
| Centaurea | cyanus | 0.203004 |
| Symphyotrichum | lateriflorum var. lateriflorum | 0.217284 |
| Digitalis | purpurea | 0.240327 |
| Leucanthemum | vulgare | 0.244253 |
| Symphyotrichum | lateriflorum var. lateriflorum | 0.249475 |
| Geum | canadense | 0.256628 |
| Rudbeckia | hirta | 0.279921 |
| Inula | helenium | 0.34778 |
| Leucanthemum | vulgare | 0.355613 |
| Lactuca | sativa | 0.355613 |
| Solidago | canadensis | 0.369294 |
| Rudbeckia | hirta | 0.37186 |
| Centaurea | cyanus | 0.407824 |
| Rudbeckia | hirta | 0.413511 |
| Solidago | canadensis | 0.432038 |
| Rudbeckia | hirta | 0.434041 |
| Triticum | aestivum | 0.456655 |
| Vicia | americana | 0.469442 |
| Alliaria | petiolata | 0.473913 |
| Leucanthemum | vulgare | 0.558144 |
| Solidago | canadensis | 0.756677 |
| Inula | helenium | 0.878983 |
| Andropogon | gerardii | 1.933205 |
| Avena | sativa | 2.4 |
| Allium | cepa | 0.61 |
| Brassica | oleracea | 0.014 |
| Daucus | carota | 1.7 |
| Cucumis | sativus | 0.008 |
| Lactuca | sativa | 0.33 |
| Glycine | max | 0.026 |
| Solanum | lycopersicum | 0.72 |
| Avena | sativa | 0.35 |
| Allium | cepa | 0.038 |
| Lolium | perenne | 0.24 |
| Brassica | oleracea | 0.059 |
| Daucus | carota | 0.054 |
| Cucumis | sativus | 0.015 |
| Lactuca | sativa | 0.022 |
| Glycine | max | 0.018 |
| Solanum | lycopersicum | 0.029 |
| Avena | sativa | 0.2 |
| Allium | cepa | 0.1 |
| Daucus | carota | 0.31 |
| Cucumis | sativus | 0.13 |
| Lactuca | sativa | 0.061 |
| Glycine | max | 0.004 |
| Solanum | lycopersicum | 0.12 |

\* Indicates study was conducted with TGAI and value used to derive SSD.

 NA = Studies are acceptable for ECOTOX but have not been formally reviewed by EFED scientists.

1If a species was not represented with a 96 hour study, and other values were available from shorter/longer duration studies (up to 10 days) then the data were included and the duration was listed next to the LC50 value as an indicator. If there were multiple other durations, the value from the study closer to a 96h duration was selected and if there were multiple values for the same duration, the range is indicated.