

**Part 158 Nontarget Plant Protection Data Requirements:
Guidance for Calculating Percent Survival in Seedling
Emergence Studies**

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**Office of Pesticide Programs
Environmental Protection Agency**



I. Purpose

The purpose of this document is to clarify EFED's recommendations for calculating percent survival in seedling emergence studies.

II. Issue

The Seedling Emergence guideline (OCSPP 850.4100, January 2012, EPA 712-C-012) specifies that the percent survival endpoint should be calculated as the number of surviving plants divided by the number of planted seeds that emerged. Accurately capturing seedling survival, as defined in OCSPP 850.4100 (*i.e.*, number of surviving plants divided by the number of planted seeds that emerged), would require additional tracking of the fate of individual seeds and seedlings because daily observations cease after 50% of the control seeds emerge. The 850.4100 guideline states:

“Controls are observed daily for number of seedlings emerged until 50% of seedlings have emerged. Number of seedlings emerged, number of surviving and dead seedlings, and visual symptoms are observed and recorded for treatment and controls at day 0, 7 and 14 post-emergence (and day 21 post-emergence if test is extended (see paragraph (e)(2) of this guideline)) of 50% of control plants.”

In practice, several seeds are typically planted in a single pot and are not individually tracked or observed daily; therefore, there is a possibility of a seedling or seedlings emerging and then dying during the same period that another seedling or seedlings emerge in the shared pot. In such cases, mortality of a seedling or seedlings would likely be missed (because the dead seedling would be ‘replaced’ by a seedling emerging later).

Therefore, it has been standard practice in the Environmental Fate and Effects Division for several years to calculate survival based on the number of surviving plants at the end of the study as a percentage of the number of seeds planted (**Equation 1**). This method more accurately predicts potential impacts to non-target plants from the use of pesticides.

Calculating % survival based on **Equation 1** captures effects to both seeds and seedlings. For example, if 10 seeds were planted and only three emerged, all of which survived, the survival rate would be considered 100% if based on the survival calculation as defined in the OCSPP 850.4100 guideline (**Equation 2**), even though only 30% of the original planted seeds survived. In contrast, the same data would result in a survival rate of 30% based on **Equation 1**, a more meaningful ecological endpoint to evaluate impacts on non-target plants. The calculation using **Equation 1** is consistent with documentation associated with the Comprehensive Environmental Toxicity Information System, the statistical package currently being used in the Environmental Fate and Effects Division.

III. Resolution

For all seedling emergence studies, survival should continue to be calculated based on number of surviving plants at the end of the study as a percentage of the number of seeds planted (**Equation 1**).

Equation 1:

$$\% \text{Survival} = (\# \text{ surviving plants at end of study} / \# \text{ planted seeds}) * 100$$

If the study report includes sufficient information (*i.e.*, emergence status of each individual seed was tracked throughout the study), then an additional calculation of the percent survival of emerged plants (**Equation 2**) may be compared to the control and reported in the Data Evaluation Record (DER) along with results from **Equation 1**.

Equation 2:

$$\% \text{Survival of Emerged Plants} = (\# \text{ surviving plants at end of study} / \# \text{ plants emerged}) * 100$$