Validation of the GHS Additivity Formula for Determining Acute Toxicity Classification of Mixtures: a Review of the Existing Research

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Introduction of GHS Additivity Formula

The most widely utilized method for determining acute toxicity classifications of mixtures in industry is the additivity formula, which is explained in the Globally Harmonized System of Classification and Labelling of Chemicals (GHS) "Purple Book." The GHS additivity formula works by taking the harmonic mean of the LD/LC50 values of the individual components that make up the mixture. The resulting value (the "acute toxicity estimate" or ATE) is then used for classification purposes as a substitute for mixture LD/LC50 test data.

The main principle of the GHS is to incorporate the best of existing methodologies (as opposed to creating new methodologies) for the purposes of harmonizing hazard communication. In the case of the GHS additivity formula, it was derived from the additivity formula that was already being used for classification of mixtures in the UN Recommendations on the Transport of Dangerous Goods ("Orange Book").

It is important to note that the additivity formula was not included in the Purple Book based on scientific validation of how accurately it calculates the LD/LC50 of a mixture – it is simply a regulatory construct that was chosen as the best alterative to estimating acute toxicity from all available methods (e.g. "simple threshold method" from OSHA 1994 HazCom Standard, or the "conventional method" from Dangerous Preparations Directive).

However, there has been an increased desire to validate the additivity formula for its accuracy in determining GHS classifications. Two recent studies were done (Dow Agrosciences, 2016 and BASF, 2018) and there was a pilot program launched last year by the EPA to look into the acceptability of the GHS additivity formula as an alternative to animal testing. The two aforementioned studies are discussed below, as well as a potential path going forward regarding the effort to validate the GHS additivity formula.

Dow Agrosciences Study (2016)

- **Selected 225 of their mixtures**
  - Composed entirely of ingredients of known acute toxicity
  - Contained at least one acutely toxic ingredient
  - Had some mixture-level *in vivo* acute toxicity test data – all studies were performed according to standard OECD (or equivalent) test guidelines, as described by the GHS

Accuracy of GHS Additivity Formula Compared to *in vivo* Test Data

Route	Number of mixtures	Correct Classification	Under-Classified	Over-Classified
Oral	199	67%	22%	11%
Dermal	179	93%	1%	6%
Inhalation	123	94%	3%	2%

Figure 1. This table shows the percentage of mixtures by route of exposure for which the GHS additivity method provided the correct/incorrect classification according to the *in vivo* test data in the Dow study. The reason for the different number of mixtures analyzed per route is due to the varying completeness of the acute toxicity test data on the 225 formulations.

Dow Agrosciences Study (2016), continued:

- **Notes on study:**
  - This study relies heavily or the dermal and inhalation routes, the vast majority of mixtures were "not classified." Given that the "not classified" range has no upper limit, it would be misleading to assume that the LD50 of the ATE from the additivity method and the LD/LC50 determined by the test data are similar simply because they both land in the "not classified" range.
  - The applicability of these results for other classes of mixtures is limited due to the fact that all mixtures in the study are agrochemicals
  - **Authors concluded** that the GHS additivity formula has a very high degree of accuracy for prediction of agrochemical mixture toxicity according to the GHS classification system.

BASF Study (2018)

- **Selected 210 of their mixtures**
  - All agrochemical formulations
  - Composed entirely of ingredients of known acute toxicity
  - Contained at least one of 8 orally-toxic ingredients
  - Some mixture-level *in vivo* acute toxicity test data available – all studies were performed according to standard OECD (or equivalent) test guidelines, as described by the GHS

Accuracy of GHS Additivity Formula Compared to *in vivo* Test Data

Route	Number of mixtures	Correct Classification	Under-Classified	Over-Classified
Oral	210	48%	39%	13%
Dermal	31	48%	6%	46%
Inhalation	128	61%	19%	20%

Figure 2. This table shows the percentage of mixtures by route of exposure for which the GHS additivity method provided the correct/incorrect classification according to the *in vivo* test data in the BASF study. The reason for the different number of mixtures analyzed per route is due to the fact that only 6 of the 8 orally-toxic ingredients are hazardous via inhalation, and only 1 of the 8 ingredients is hazardous via dermal exposure.

- **Notes on study:**
  - There is a fairly balanced breakdown of mixtures in terms of acute toxicity, which may have provided a more accurate reflection of the additivity formula's ability to predicted GHS classifications, in comparison the Dow study.
  - The small sample size for the dermal route analysis in this study makes it difficult to draw any significant conclusions.

BASF Study (2018), continued:

- The authors explain the lack of accuracy shown by the additivity formula in this study as potentially a result of chemical interaction of hazardous ingredients (it is presumed that the mixtures used in the Dow study did not contain as many hazardous ingredients). The main idea behind this theory is that each additional hazardous ingredient in a mixture increases the probability of synergistic or antagonist effects.

Combining the Data

The aforementioned EPA pilot program has not released any findings yet, and given that the two studies that were discussed are the only two publicly available studies of their kind, this is all the data there is to draw conclusions from at the moment. The below table summarizes some of the combined findings from these two studies:

Accuracy of GHS Additivity Formula Compared to *in vivo* Test Data (by hazard class)

Class. based on ATE	Route	Number of mixtures	Correct classification	Under-classified	Over-classified
NC	Oral	196	62%	38%	NA
5	Oral	69	32%	43%	25%
4	Oral	128	64%	16%	20%
NC	Dermal	179	98%	2%	NA
NC	Inhalation	171	88%	11%	NA
4	Inhalation	66	58%	9%	0%

Figure 3. This table combines the results from the BASF and Dow studies, identifying by route of exposure and hazard class, how accurate the GHS additivity formula was in matching the classification determined from the in vivo test data. Only hazard classes with a sample size of at least 50 are included in this table.

The Next Steps

If/when the EPA project is complete, it will provide the largest dataset of mixtures for the purpose of validating the GHS additivity method. However, because only data on agrochemicals is being collected, the findings may not be sufficient in determining the additivity method's accuracy in classifying other classes of mixtures.

If there is a large-scale study involving a variety of material types, with sufficient sample sizes for each level of acute toxicity, and sufficient sample sizes of mixtures with varying numbers of hazardous ingredients (e.g. mixtures with 3 hazardous ingredients, mixtures with 4 hazardous ingredients, etc.), then not only could the additivity formula be validated, but the data could be used to derive "adjustment factors" that would take certain factors (e.g. chemical interaction) into account that the standard additivity method is unable to account for.

References

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