An Industry-Sponsored Survey on
Genotoxicity and Oral Toxicity Study Data
From Enzymes Produced Using Protein Engineering

The Enzyme Technical Association (ETA), as a strong supporter of research in the enzyme industry, was recently asked to conduct a confidential survey of its membership concerning enzymes produced using protein engineering. Specifically, the survey requested information from the membership regarding the results from genotoxicity studies (Ames tests and chromosome aberration studies) and oral toxicity studies (acute oral and repeated dose studies) performed by industry on enzymes produced using protein engineering. A number of members of ETA participated in the survey.

Because much of this data has commercial value, the collection of data on the studies performed was done on a confidential basis, with all data provided by the members being de-identified and kept confidential by ETA.

The members participating in the survey provided information on a broad range of enzymes produced using protein engineering. These enzymes are used in numerous applications for human food, animal feed, and technical applications.

Reports on protein-engineered enzymes produced by 30 individual strains were submitted as part of the industry survey. The enzyme activities included acyltransferases, alpha-amylases, maltogenic alpha-amylases, pectate lyases, phospholipases, phytases, proteases, and xylanases. Based on the range of enzymes and number of tests performed, it is clear that the industry has done testing on a wide variety of enzymes with multiple applications. Of the 30 enzymes, all but 2 had been the subject of Ames tests, the results of which were all negative. For chromosome aberration testing, all but four had been tested and all the results showed that there were no adverse effects related to the treatment. Five of the strains were reported to have undergone additional testing such as micronucleus tests. In summary, for the 54 distinct Ames and chromosome aberration genotoxicity studies performed by the ETA membership there were no adverse effects related to the treatment in any of the studies.

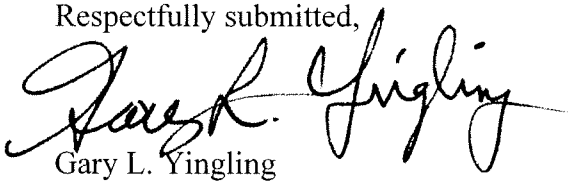
In addition to genotoxicity studies, the survey requested data regarding oral studies performed by industry, whether they were single, 14 day, or subchronic repeated dose oral studies ranging from 28 days to 90 days.

Of the 30 enzymes reported on, 15 had been the subject of an acute oral study. As to repeated dose oral studies, all but four had been the subject of one or more multi-day studies, with several strains having been the subject of several multi-day studies. Of the four that were not the subject of a multi-day study, an acute study had been performed. Therefore, of the 30 enzymes for which data was submitted, all have been the subject of at least one oral toxicity study and many of the enzymes have been the subject of more than one type of test. Again, no adverse effects were observed in any of a total of 47 distinct oral toxicity studies performed.

While the focus of the survey was genotoxicity and oral toxicity, an additional 18 studies of various types were reported on, including, for example, dermatologic irritation, ocular irritation and inhalation toxicity. Again, no data suggested an adverse event or problem.

The results of this survey of toxicology studies demonstrate that the general safety profile of enzymes produced using protein engineering is no different from that already established via extensive toxicological studies for non-protein-engineered enzymes as reviewed in Pariza and Johnson (2001), Olempska-Beer et al. (2006) and in many cases, reviewed by FDA in the GRAS Notice Program (accessible at http://www.cfsan.fda.gov/~rdb/opa-gras.html).

Respectfully submitted,



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