

VII D. Enzymes

Commercial enzyme products may be obtained from edible plants and animals and from non-toxicogenic, non-pathogenic microorganisms. Questions about the microbial source of the enzyme (see **Chapter VII E**) and the nature and level of enzyme preparation in the food are of concern in evaluating the safety of commercial enzyme products because they influence the type and level of contaminating impurities in the food.

In general, enzyme preparations from organisms with a history of safe use do not require the same level of toxicological testing as enzymes from sources without a history of safe use in food. The safety of commercial enzyme products from sources without a history of safe use in food usually is evaluated on a case-by-case basis, but some generalizations about toxicology tests for these food additives can be made.

Because of the protein nature of enzymes and their susceptibility to digestion when consumed, residues of pure enzymes in processed food would be expected to have only limited toxic potential. If highly purified preparations of microbial enzymes are used in food processing, exposure to the enzymes is usually reduced to the parts-per-billion range. Such a level of exposure would ordinarily be too low to pose a safety concern, and toxicological testing may not be required. An exception to this generalization may occur if review by the Center's chemists results in concern for the presence in the enzyme preparation of a toxic material used in the purification process; however, this is unlikely because of the requirement that food grade chemicals be used in purification.

In most cases, however, commercial enzyme products from microbial sources are only partially purified. A variety of uncharacterized extraneous substances ("impurities") of biological origin may be present in the enzyme preparation at levels comparable to the active ingredient. These substances have no technical effect in food processing, but are allowed to remain in the enzyme products because the impurities do not interfere with enzyme function. In addition, the enzyme preparation may contain multiple enzyme activities that serve a variety of useful functions in processing food. When the types and levels of impurities in commercial enzyme products from microbial sources are considered to be significant, the Agency may recommend that safety be established by appropriate toxicity testing. Such a requirement usually can be met by 90-day toxicity studies in the rat and the dog. However, if review of the safety of the enzyme preparation raises questions about chemical contaminants, stability of the microbial strain, production of toxic products, *etc.*, additional studies may be needed.

Enzyme products may be added directly to the food to be processed (*e.g.*, rennet) or they may be immobilized on an insoluble matrix for use in processing liquid foods. Enzymes are immobilized by secure bonding (usually by means of a chemical reaction) to an insoluble matrix. Liquid food products (*e.g.*, corn syrup) may be processed by passage over a column of the immobilized enzyme. Only negligible amounts of the immobilized enzyme are expected to enter the processed food. Depending on the nature of the immobilization matrix, however, some potential exists for contamination of the processed food by chemicals used in the immobilization process. If the Agency decides that information about the nature of the fixing agent and its potential migration to food raise questions of safety for foods processed by passage over an immobilized microbial enzyme, the Agency will recommend that the immobilized enzyme be subjected to 90-day toxicity studies in the rat and the dog or other appropriate study.

As described in the preceding paragraphs, a variety of factors will be taken into account by the Agency in deciding what information is needed to assess the safety of additives that are enzymes. Before conducting toxicity studies to assess the safety of such additives, petitioners should consult with Agency scientists. A comprehensive review of the safety concerns relating to additives that are enzymes will be issued in a separate publication.