



INTERNATIONAL PROGRAMME ON CHEMICAL SAFETY

WORLD HEALTH ORGANIZATION

**TOXICOLOGICAL EVALUATION OF CERTAIN  
FOOD ADDITIVES**

**WHO FOOD ADDITIVES SERIES 22**

Prepared by:

The 31st meeting of the Joint FAO/WHO Expert  
Committee on Food Additives (JECFA)

World Health Organization, Geneva 1988

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First published 1988

Printed in Great Britain at the University Press, Cambridge

Library of Congress cataloging in publication data available.

British Library cataloguing in publication data

Joint FAO/WHO Expert Committee on Food Additives,  
Meeting (31st: 1987: Geneva).

Toxicological evaluation of certain food additives and  
contaminants.

1. Food additives -- Toxicology
2. Food contamination
3. Food poisoning

I. Title II. World Health Organization

ISBN 0 521 36928 2

The preparation of this document was supported by the International  
Programme on Chemical Safety (IPCS), Geneva, Switzerland.

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## PREFACE

The monographs contained in this volume were prepared by the thirty-first Joint FAO/WHO Expert Committee on Food Additives (JECFA), which met in Geneva, Switzerland, 16-25 February 1987. These monographs summarize the safety data on selected food additives by the Committee. Generally, the compounds on which monographs were prepared are those on which substantial safety data exist. The data reviewed in these monographs form the basis for acceptable daily intakes (ADIs) established by the Committee.

The thirty-first report of JECFA has been published by the World Health Organization in the WHO Technical Report Series (No. 759). The participants in the meeting are listed in Annex 3 of the present publication and a summary of the conclusions of the Committee is included as Annex 4.

Specifications established by the thirty-first meeting of JECFA have been issued separately by FAO under the title *Specifications for the identity and purity of certain food additives*, FAO Food and Nutrition Paper, No. 38. These toxicology monographs should be read in conjunction with the specifications and the report.

Reports and other documents resulting from previous meetings of the Joint FAO/WHO Expert Committee on Food Additives are listed in

## Annex 1.

JECFA serves as a scientific advisory body to FAO, WHO, their Member States, and the Codex Alimentarius Commission, primarily through the Codex Committee on Food Additives, regarding the safety of food additives and contaminants in food. Committees accomplish this task by preparing reports of their meetings and publishing specifications and toxicological monographs, such as those contained in this volume, on substances that they have considered.

The toxicological monographs contained in this volume are based upon working papers that were prepared by temporary advisers in advance of the thirty-first JECFA meeting. A special acknowledgement is given to those who prepared these working papers: Dr C.L. Galli, Professor of Experimental Toxicology, University of Milan, Milan, Italy; Dr S.I. Shibko, Associate Director for Regulatory Evaluation, Center for Food Safety and Applied Nutrition, Food and Drug Administration, Washington, DC, USA; and Dr Ronald Walker, Professor of Biochemistry, University of Surrey, Guildford, Surrey, England.

Many proprietary unpublished reports are referenced. These were voluntarily submitted to the Committee by various producers of the food additives under review and in many cases these reports represent the only safety data available on these substances. The temporary advisers based the working papers they developed on all the data that were submitted, and all these studies were available to the Committee when it made its evaluations.

From 1972 to 1975 the toxicology monographs prepared by Joint FAO/WHO Expert Committees on Food Additives were published by WHO in the WHO Food Additives Series; after 1975 this series became available only in the form of unpublished WHO documents provided on request by the Organization. Beginning with the 1985 monographs, they are published by Cambridge University Press, which should ensure that these monographs are more widely known and available.

The preparation and editing of the monographs included in this volume have been made possible through the technical and financial contributions of the Participating Institutions of the International Programme on Chemical Safety (IPCS), which support the activities of JECFA. IPCS is a joint venture of the United Nations Environment Programme, the International Labour Organisation, and the World Health Organization, which is the executing agency. One of the main objectives of IPCS is to carry out and disseminate evaluations of the effects of chemicals on human health and the quality of the environment.

The designations employed and the presentation of the material in this publication do not imply the expression of any opinion whatsoever on the part of the organizations participating in the IPCS concerning the legal status of any country, territory, city, or area or its authorities, or concerning the delimitation of its frontiers or boundaries. The mention of specific companies or of certain manufacturers' products does not imply that they are endorsed or recommended by those organizations in preference to others of a similar nature that are not mentioned.

Any comments or new information on the biological or toxicological data on the compounds reported in this document should be addressed to: Joint WHO Secretary of the Joint FAO/WHO Expert Committee on Food Additives, International Programme on Chemical Safety, World Health Organization, Avenue Appia, 1211 Geneva 27, Switzerland.

See Also:

Toxicological Abbreviations



## ENZYME PREPARATIONS

Problems in evaluating the safety of enzymes in food processing were discussed at the fifteenth, eighteenth and twenty-ninth meetings of the Expert Committee, when principles relating to their evaluation were elaborated (Annex 1, references 26, 35, and 70). At its present meeting, the Committee reaffirmed those principles, which have been consolidated in Annex III of "Principles for the Safety Assessment of Food Additives and Contaminants in Food" (Annex 1, reference 76).

For the purpose of toxicological evaluation, the enzyme preparations under present consideration were grouped into the following classes:

- Class III - Enzymes derived from Aspergillus oryzae;
- Class IV - Enzymes derived from Aspergillus niger; and
- Class V - Enzymes derived from Trichoderma reesei, Trichoderma harzianum, Penicillium funiculosum, Aspergillus alliaceus.

The guidelines established by JECFA for these classes of enzymes provide a basis for the toxicological studies required for their evaluation.

At the twenty-ninth meeting the Committee concluded that, when enzyme preparations from either class IV or class V are added directly to food but not subsequently removed, an acceptable daily intake should be established to ensure that levels of the enzyme preparations in food are safe. In order to evaluate the information received on the estimate of the amount of enzyme preparations used in the toxicological studies and levels of consumption resulting from their use in food, the Committee adopted the concept of enzyme total organic solids (TOS), which is defined as follows;  $\% \text{ TOS} = 100 - (A + W + D)$ , where A = % ash, W = % water, and D = % diluent and carrier (Ad hoc Enzyme Technical Committee, 1981; Pariza & Foster, 1983). This concept overcomes the problem that enzyme preparations of different activities and forms were used in the toxicological studies. It also takes into account that most of the organic solids in this fraction are not the enzyme per se.

In establishing acceptable daily intakes for the enzymes in classes IV and V, the Committee noted that the animal feeding studies were primarily of short-term duration. It, therefore, concluded that it would be appropriate to use a safety factor greater than the usual 100.

## REFERENCES

Ad hoc Enzyme Technical Committee (1981). The 1978 enzyme survey, summarized data, National Academy of Sciences/National Research Council/Food and Nutrition Board, Committee on GRAS List Survey, Phase III, National Academy Press, Washington, D.C.

Pariza, M.W. & Foster, E.M. (1983). Determining the Safety of Enzymes used in Food Processing, J. Food Protection, 46: 453-468.

See Also:

Toxicological Abbreviations





## ENZYMES DERIVED FROM ASPERGILLIS NIGER

### **EXPLANATION**

A. niger is a contaminant of food and was not considered in the same light as those organisms regarded as normal constituents of food. It is necessary to show that the strains used in enzyme preparations do not produce mycotoxins.

Microbial carbohydrases prepared from some varieties of A. niger were evaluated at the fifteenth meeting of the Committee, at which time a temporary ADI "not limited" was established (Annex 1, reference 26). A toxicological monograph was prepared (Annex 1, reference 27). An adequate 90-day study in rats was requested. Since the previous evaluation, additional data have become available on a number of carbohydrases, which are summarized and discussed in the following monograph. These enzymes were considered by the Committee to encompass the carbohydrases previously considered. The previously published monograph has been expanded and reproduced in its entirety below.

### AMYLOGLUCOSIDASES (E.C. 3.2.1.3)

#### BIOLOGICAL DATA

##### Biochemical aspects

No information available.

##### Toxicological studies

#### **Special studies on aflatoxin-related effects**

##### Ducklings

Four groups of 5 ducklings received in their diet 0, 1, 5, or 10% enzyme preparation for 29 days. Growth, feed consumption, survival, behaviour, and mean liver weights were comparable, in all groups. No gross or histopathological lesions of the liver were seen (FDRL, 1963a).

Four groups of 5 ducklings received in their diet 0, 1, 5, or 10% enzyme preparation for 29 days. Growth, feed consumption, survival, behaviour, and development were comparable in all groups. No gross liver lesions were seen at autopsy and mean liver weights of treated animals were similar to those of controls. Histopathology of the livers was normal. No toxic elements were noted (FDRL, 1963b).

#### **Acute toxicity<sup>1</sup>**

Species	Route	LD <sub>50</sub> (mg/kg b.w.)	Reference
Mouse	oral	> 3,200	Hunt & Garvin, 1963
		> 4,000	Hunt & Garvin, 1971
		> 3,200	Willard & Garvin, 1968
		> 4,000	Garvin et al., 1966

Rat	oral	10,000	Gray, 1960
		31,600	Kay & Calendra, 1962
		> 3,200	Willard & Garvin, 1968
		> 4,000	Garvin et al., 1966
		12,500 - 20,000	Kapiszka & Hartnage, 1978
Rabbit	oral	> 4,000	Garvin et al., 1966
Dog	oral	> 4,000	Garvin et al., 1966

- <sup>1</sup> These data were obtained with several different commercial enzyme preparations.

### Short-term studies

#### Rats

Three groups of 10 male rats received 0, 0.5, or 5% enzyme preparation in their diets for 30 days. No adverse effects related to treatment were observed regarding growth, appearance, behaviour, survival, food consumption, haematology, organ weights, or gross pathology (Garvin *et al.*, 1966).

Two groups of 10 male and 10 female rats received either 0 or 5% enzyme preparation in their diets daily for 91 days. No differences from controls were observed regarding appearance, behaviour, survival, weight gain, haematology, organ weights, or gross pathology (Garvin & Merubia, 1959).

Two groups of 10 male and 10 female ARS Sprague-Dawley rats were fed diets containing 5 or 10% of the test enzyme preparation (equivalent to 3.5 or 7 g enzyme preparation/kg b.w./day) for 90 to 94 days. A control group of 20 male and 20 female rats were maintained on the diet alone. No signs of toxicity were observed during the test period. Body-weight gain and food consumption were similar between test and control groups. Differential blood counts were within the normal range at weeks 4 and 8 of the study in both test and control animals. At the end of the study serum clinical chemistry parameters, organ weight analyses, and gross and microscopic pathology showed no compound-related effects (Garvin *et al.*, 1972).

### Long-term studies

No information available.

#### Observations in man

No information available.

### COMMENTS

Several short-term feeding studies in rats on amyloglucosidase preparations from *A. niger* have been performed. One study, in which the preparation was fed at up to 10% of the diet, was considered to be acceptable by current standards. No compound-related effects were observed in this study or in duckling tests that were performed to investigate potential aflatoxin-related effects.

The evaluations by the Committee of the carbohydrates and the protease from *A. niger* are summarized at the end of this section.

### REFERENCES

FDRL (1963a). Unpublished report No. 84600e. Submitted to WHO by



Laboratories, Inc., Elkhart, IN, USA.

FDRL (1963b). Unpublished report No. 84600f. Submitted to WHO by Laboratories, Inc., Elkhart, IN, USA.

Garvin, P.J. & Merubia, J. (1959). Unpublished report. Submitted to WHO by Baxter Laboratories, Inc.

Garvin, P.J., Willard, R., Merubia, J., Huszar, B., Chin, E., & Gilbert, C. (1966). Unpublished report. Submitted to WHO by Baxter Laboratories, Inc.

Garvin, P.J., Ganote, C.E., Merubia, J., Delahany, E., Bowers, S., Varnado, A., Jordan, L., Harley, G., DeSmet, C., & Porth, J. (1972). Unpublished report from Travenol Laboratories, Inc., Morton Grove, IL, USA. Submitted to WHO by Gist-brocades NV, Delft, Holland.

Gray, E.H. (1960). Unpublished report. Submitted to WHO by Miles Laboratories, Inc., Elkhart, IN, USA.

Hunt, R.F. & Garvin, P.J. (1963). Unpublished report. Submitted to WHO by Baxter Laboratories, Inc.

Hunt, R.F. & Garvin, P.J. (1971). Unpublished report. Submitted to WHO by Travenol Laboratories, Inc., Morton Grove, IL, USA.

Kapiszka, E.L. & Hartnage, R.E. (1978). The acute oral toxicity of Diazyme concentrate and Diazyme 325 in the rat. Unpublished report No. 16 from Miles Laboratories, Inc., Elkhart, IN, USA. Submitted to WHO by Miles Laboratories, Inc., Elkhart, IN, USA.

Kay, J.H. & Calendra, J.C. (1962). Unpublished report. Submitted to WHO by Miles Laboratories, Inc., Elkhart, IN, USA.

Willard, R. & Garvin, P.J. (1968). Unpublished report. Submitted to WHO by Travenol Laboratories, Inc., Morton Grove, IL, USA.

$\beta$ -GLUCANASE (E.C. 3.2.1.6)

#### BIOLOGICAL DATA

##### Biochemical aspects

No information available.

##### Toxicological studies

(The TOS of the enzyme preparation used for toxicity studies was 49%).

#### Special Studies on mutagenicity

The enzyme preparation was tested for mutagenic activity using 5 strains of *Salmonella typhimurium* (TA98, TA100, TA1535, TA1537, and TA1538 both with and without metabolic activation (S-9 fraction). The preparation was not mutagenic or toxic at concentrations up to 40 mg/ml (McConville, 1980).

A cytogenic bone marrow study was performed using adult male Chinese hamsters. Groups of adult male hamsters received up to 5000 mg/kg b.w./day of the enzyme preparation for 5 consecutive days. Treatment did not result in an increased frequency of chromosomal aberrations in bone marrow (McGregor & Willins, 1981).

#### Acute toxicity

Species	Route	Sex	LD <sub>50</sub> (ml/kg b.w.)	Reference
Mouse (NMRI)	oral	M & F	30	Novo, 1978a
Rat (Wistar)	oral	-	28.1	Novo, 1978b

### Short-term studies

#### Rats

Three groups, each containing 5 male and 5 female Wistar/Mol SPF rats, were dosed orally by gavage once a day for 14 days with enzyme preparation at dose levels equivalent to 2.5, 5.0, or 10 ml/kg b.w. No clinical changes were observed. Body-weight gains of test and control animals were similar. At termination of the study, measurements of organ weights showed no compound-related effects (Novo, 1978c).

In another study, 4 groups, each containing 15 male and 15 female Wistar/Mol SPF rats, were dosed by gavage once a day for 90 days with enzyme preparation at dose levels equivalent to 0, 2.5, 5.0, or 10 ml/kg b.w. Deaths, primarily in the high-dose group, appeared to be related to injury during dosing. No clinical signs were observed in the other test animals. Male rats in the high-dose group showed decreased weight gain and marked decrease in food intake. Haematology studies showed increased platelet counts and decreased clotting times in the high-dose group at week 6, but this effect was not apparent at week 12. No other effects were reported. Clinical chemistry and urinalysis values at weeks 6 and 12 were within the normal range. At termination of the study, organ weight analysis showed a marked increase in relative weights of the spleen and testes of the males in the high-dose group. Gross and histopathological examination of the principal organs and tissues showed no compound-related effects (Perry *et al.*, 1979).

#### Dogs

Three groups, each containing one male and one female beagle dog, received single doses of 5, 10, or 15 ml/kg b.w. of the enzyme preparation over a 4-day period. Following a 7-day observation period the dogs were sacrificed and subjected to macroscopic post-mortem examination. No compound-related effects were observed, with the exception of vomiting during the first 4 days of the study. In another study, dogs were administered consecutive doses of 15 ml/kg b.w./day for 9 days, and 10 ml/kg b.w./day for 5 days. No deaths occurred during the course of the study. The only clinical sign noted was excessive salivation and emesis shortly after dosing. Body weights, electrocardiograms, haematological parameters, blood serum chemistry, organ weights, gross pathology, and histopathology showed no compound-related effects (Osborne *et al.*, 1978).

In another study, three groups, each containing 3 male and 3 female beagle dogs, were dosed with the enzyme preparation by gavage once a day, seven days a week, for 13 weeks, at dose levels equivalent to 2, 5, or 10 ml/kg b.w./day. Two dogs in the high-dose group died during the course of the study, which the authors concluded was due to respiratory distress as a result of foreign material in the lungs. Vomiting was reported after dosing in the high-dose group. Haematological parameters at weeks 6 and 12 were within normal limits, with the exception of a significant increase in WBC count, specifically in the group mean neutrophil counts, in the high-dose group. Clinical chemistry values were within the normal range at weeks 8 and 12, with the exception of slight increases in blood glucose and

cholesterol in the high-dose group. Urinalysis showed no compound-related effects. At termination of the study, organ-weight analyses and gross and histopathological examination of the principal organs and tissues showed no compound-related effects (Greenough *et al.*, 1980).

#### Long-term studies

No information available.

Observations in man

No information available.

#### COMMENTS

This enzyme preparation was not genotoxic in microbial or in mammalian test systems. Short-term studies in rats and dogs resulted in no observed compound-related effects at levels up to 5 ml/kg b.w./day of enzyme preparation.

The evaluations by the Committee of the carbohydrases and the protease from *A. niger* are summarized at the end of this section.

#### REFERENCES

Greenough, R.J., Brown, J.C., Brown, M.G., Cowie, J.R., Maule, W.J., & Atken, R. (1980).  $\beta$ -Glucanase 13 week oral toxicity study in dogs. Unpublished report No. 1630 from Inveresk Research International, Musselburgh, Scotland. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

McConville, M. (1980). Testing for mutagenic activity with *S. typhimurium* strain TA98, TA100, TA1535, TA1537, and TA1538 of fungal  $\beta$ -glucanase. Unpublished report No. 1751 from Inveresk Research International, Musselburgh, Scotland. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

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Novo (1978a). Acute oral toxicity of  $\beta$ -glucanase given to mice. Unpublished report No. 1978-06-30 RKH/PNi from Novo Industri A/S, Bagsvaerd, Denmark. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

Novo (1978b). Acute oral toxicity of  $\beta$ -glucanase given to rats. Unpublished report No. 1978-07-17 RICH/PNi from Novo Industri A/S, Bagsvaerd, Denmark. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

Novo (1978c). Oral toxicity of  $\beta$ -glucanase given daily to rats for 14 days. Unpublished report No. 1978-08-21 RKH/PNi from Novo Industri A/S, Bagsvaerd, Denmark. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

Osborne, B.E., Cockrill, J.B., Cowie, J.R., Maule, W., & Whitney, J.C. (1978). Beta-glucanase, dog acute and maximum tolerated dose study. Unpublished report No. 1208 from Inveresk Research International, Musselburgh, Scotland. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

Perry, C.J., Everett, D.J., Cowie, J.R., Maule, W.J. & Spencer, A. (1979).  $\beta$ -glucanase toxicity study in rats (oral administration by

gavage for 90 days). Unpublished report No. 1310 from Inveresk Research International, Musselburgh, Scotland. Submitted to WHO by Novo Industri A/S, Bagsvaerd, Denmark.

#### HEMI-CELLULASE

#### BIOLOGICAL DATA

##### Biochemical aspects

No information available.

##### Toxicological studies

##### Special studies on mutagenicity

The enzyme preparation was tested for mutagenic activity using *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 both with and without metabolic activation (S-9 fraction). The test substance was not mutagenic or toxic at concentrations up to 5 mg/plate (Clausen & Kaufman, 1983).

In an *in vitro* cytogenetic test using CHO-K1 cells, both with and without metabolic activation (S-9 fraction), the enzyme preparation at test levels up to 2.5 mg (dry wt)/ml did not induce chromosomal aberrations (Skovbro, 1984).

##### Acute toxicity

No information available.

##### Short-term studies

###### Rats

Four groups, each containing 5 male and 5 female Wistar MOL/W rats, were dosed by gavage once a day for 90 days with the enzyme preparation at doses equivalent to 0, 100, 333, or 1000 mg/kg b.w./day. No significant clinical changes were observed. Body-weight gain and food intake were similar among test and control animals. Haematologic and clinical chemistry measurements at termination of the study were within normal ranges. Post-mortem examinations, measurements of organ weights, and histopathology showed no compound-related effects. Slight increases in kidney and adrenal weights in the mid-dose group were not associated with histopathological effects, and did not show a dose response (Kallesen, 1982).

##### Long-term studies

No information available.

##### Observations in man

No information available.

#### COMMENTS

This enzyme preparation was not genotoxic in microbial or in mammalian test systems. In a limited 90-day study in rats, no effects were observed at the highest dose administered (1 g/kg b.w./day). This enzyme preparation contained high levels of pectinase. The pectinase enzyme preparation summarized below may be identical to this hemi-cellulase preparation, which provides added assurance of the safety of this preparation.

The evaluations by the Committee of the carbohydrases and the protease from *A. niger* are summarized at the end of this section.

#### REFERENCES

Clausen, B. & Kaufman, U. (1983). Unpublished report from Obmutat Laboratiet. Submitted to WHO by Grinsted Products A/S, Brabrand, Denmark.

Kallesen, T. (1982). A 90-day toxicity study. Unpublished report No. 10023 from Scantox Biological Laboratory Ltd., Denmark. Submitted to WHO by Grinsted Products A/S, Brabrand, Denmark.

Skovbro, A. (1984). *In vitro* mammalian cytogenetic test (according to OECD Guideline No. 473). Unpublished report No. 10398 from Scantox Biological Laboratory Ltd., Denmark. Submitted to WHO by Grinsted Products A/S, Brabrand, Denmark.

PECTINASE (E.C. 3.1.1.11; 3.2.1.15; 4.2.2.10)

#### BIOLOGICAL DATA

Biochemical aspects

No information available.

Toxicological studies (The TOS of the commercial preparation is approximately 5%).

#### Acute toxicity

Species	Route	LD <sub>50</sub> (ml/kg b.w.)	Reference
Rat	oral	18.8-22.1	Porter & Hartnagel, 1979

#### Short-term studies

##### Rats

Two groups of 10 male and 10 female ARS Sprague-Dawley rats were fed diets containing 5 or 10% of the test enzyme preparation (equivalent to 3.5 or 7 g of the enzyme preparation/kg b.w./day), for 90 to 94 days. A control group of 20 male and 20 female rats was maintained on the diet alone. No signs of toxicity were observed during the test period. Body-weight gain and food consumption were similar among test and control groups. Differential blood counts at weeks 4 and 8 of the study were within the normal range in test and control animals. At the end of the study serum clinical chemistry analyses, organ weight analyses, and gross and microscopic pathology showed no compound-related effects (Garvin *et al.*, 1972).

#### Long-term studies

No information available.

Observations in man

No information available.

#### COMMENTS

In a short-term study in rats, no adverse effects were observed

at dietary levels of the enzyme preparation up to the equivalent of 7 mg/kg b.w./day. This enzyme preparation may be identical to the hemi-cellulase preparation summarized above. The hemi-cellulase enzyme preparation summarized above also contained high levels of pectinase, which provides added assurance of the safety of this preparation.

#### REFERENCES

Garvin, P.J., Ganore, C.E., Merubia, J., Delahany, E., Bowers, S., Varnado, A., Jordan, L., Harley, G., DeSmet, C., & Porth, J. (1972). Carbohydrase from *Aspergillus niger* (pectinase, cellulase and lactase). Unpublished report from Travenol Laboratories, Inc., Horton Grove, IL, USA. Submitted to WHO by Gist-brocades NV, Delft, Holland.

Porter, M.C. & Hartnagel R.E. (1979). The acute oral toxicity of a new pectinase product in the rat. Unpublished report No. 11 from Miles Laboratories, Inc., Elkhart, IN, USA. Submitted to WHO by Enzyme Technical Association, Washington, DC, USA.

#### PROTEASE

No information available.

#### GENERAL COMMENTS ON ENZYMES FROM *A. NIGER*

*Aspergillus niger* is a contaminant of food. Although there may be possible strain differences in *A. niger*, and different cultural conditions might be used to prepare the various enzymes, the available toxicity data, which consist primarily of short-term feeding studies in rats and some studies in dogs, show that all the enzyme preparations tested were of a very low order of toxicity. The enzyme preparations tested were non-mutagenic in bacterial and mammalian cell systems. Studies on some strains of *A. niger* used to prepare carbohydrases showed no aflatoxin or related substance production. These studies provide the basis for evaluating the safety of enzyme preparations derived from *A. niger*. It was also noted that the enzyme preparations tested exhibit a number of enzyme activities, in addition to the major enzyme activity. Thus, there may be considerable overlap of the enzyme activities of the different enzyme preparations so that safety data from each preparation provides additional assurance of safety for the whole group of enzymes.

Since the enzyme preparations tested were of different activities and forms, and most of the organic materials in the preparations are not the enzyme *per se*, the numerical ADI is expressed in terms of total organic solids (TOS) (see introduction to enzyme preparations section).

#### EVALUATION

##### Level causing no toxicological effect

All enzyme preparations tested showed no-observed-effect levels greater than 100 mg TOS/kg b.w./day in 90-day studies in rats.

##### Estimate of acceptable daily intake

0-1 mg TOS/kg b.w. for each of the enzyme preparations.

See Also:

Toxicological Abbreviations