

PUBLIC CONSULTATION ON PROPOSED AMENDMENTS TO THE FOOD REGULATIONS REGARDING PERMITTED ENZYMES FOR FOOD USE

Aim

The Agri-Food and Veterinary Authority (AVA) is seeking feedback from the food industry (local food enzyme manufacturers and importers), on the proposed amendments to the Food Regulations concerning permitted enzymes for food use.

Background

The Eighth Schedule lists enzymes in alphabetical order among other permitted general purpose food additives, with information on the enzyme name, enzyme production strain, and indication of whether the enzyme is produced from a genetically modified source.

Further characterisation of the permitted enzymes such as the production source (from animal, plant, microbial etc), the Enzyme Commission number (EC number), as well as the donor organism and donor gene (for enzymes produced from genetically modified microbial sources) are not provided. In addition, certain enzymes that have a long history of usage by the food industry (such as bromelain) are not listed under the Eighth Schedule.

Such information is important to the industry so that they can clearly identify the permitted enzymes and their sources.

Proposed Amendments

Revised presentation of the permitted food enzymes under the Eighth Schedule

AVA proposes to amend the Eighth Schedule of the Food Regulations to group all permitted enzymes together under a new section titled “Permitted Enzymes”. In this new section, permitted enzymes will be further classified based on their sources (animal, plant, and microbial). In addition, the EC number, as well as information on the donor organism and donor gene (for enzymes produced from genetically modified microbial sources) will be specified.

In addition, 158 more new enzymes, which have been evaluated to be safe for use in foods will be incorporated into the new section on enzymes.

The proposed format for the new section on permitted enzymes under the Eighth Schedule can be found in the **ANNEX**.

Benefits to industry

The proposed amendment is a trade facilitating measure. With the proposed inclusion of 158 more enzymes in the Eighth Schedule, the industry can look forward to making use of a greater variety of enzymes in the manufacture of their food products.

The revised format for listing of the permitted enzymes is similar to that of major developed countries such as Australia, Canada and New Zealand. The revised format aims to provide clarity to the industry on the identity of permitted enzymes for food use in Singapore. With the availability of such clear and unambiguous information, it is envisaged that the industry will be better equipped to comply with the requirements of the Food Regulations.

Request for comments

AVA invites views and comments on:

- i. Whether the proposed format of presentation provides clarity on the identity of permitted enzymes for food use.
- ii. Whether additional information on the characterisation of enzymes should be included in the proposed format, and the rationale for inclusion.
- iii. Whether any additional enzymes should be included in the Eighth Schedule, and the justification for inclusion.

Procedure and timeframe for submitting views and comments

AVA welcomes views and comments on the above proposal. All submissions should be clearly and concisely written, and should provide a reasoned explanation for any proposed revisions.

Submissions should reach AVA no later than 12:00 p.m., 8 June 2015, through mail, or email, to the following addresses:

Mail:

52 Jurong Gateway Road #14-01 Singapore 608550
(Attention: Ms Leong Ai Ling)

Email:

leong_ai_ling@ava.gov.sg

We regret that we will not be able to address or acknowledge every feedback or comment received. However, the feedback or comments will be consolidated and a summary of the key comments received will be published, together with AVA's responses, on the AVA website after the close of the consultation exercise. The summary will not disclose the identity of person(s) or organisation(s) providing the feedback or comments.

ANNEX

PERMITTED ENZYMES

(A) Enzymes derived from animal sources

| Enzyme | EC Number | Source |
|--------------------------------------|-----------|--|
| Catalase | 1.11.1.6 | Bovine liver |
| Lactoperoxidase | 1.11.1.7 | Bovine milk |
| Lipase, triacylglycerol | 3.1.1.3 | Bovine stomach; salivary glands or forestomach of calf, kid or lamb; porcine or bovine pancreas |
| Lysozyme | 3.2.1.17 | Egg whites |
| Pancreatin or (pancreatic elastase) | 3.4.21.36 | Pancreas of the hog or ox |
| Pepsin | 3.4.23.1 | Bovine or porcine stomach |
| Phospholipase A2 | 3.1.1.4 | Porcine pancreas |
| Rennet | 3.4.23.4 | Aqueous extracts from the fourth stomach of calves, kids, lambs, and adult bovine animals, sheep and goats |
| Thrombin | 3.4.21.5 | Bovine or porcine blood |
| Trypsin | 3.4.21.4 | Porcine or bovine pancreas |

(B) Enzymes derived from plant sources

| Enzyme | EC Number | Source |
|---------------|------------|--|
| Alpha-amylase | 3.2.1.1 | Malted cereals |
| Actinidin | 3.4.22.14 | Kiwifruit (<i>Actinidia deliciosa</i>) |
| Beta-Amylase | 3.2.1.2 | Malted cereals Sweet potato (<i>Ipomoea batatas</i>) |
| Bromelain | 3.4.22.4 | Pineapple fruit/stem (<i>Ananas comosus</i> and <i>Ananas bracteatus</i> (L)) |
| Ficin | 3.4.22.3 | <i>Ficus</i> spp. |
| Lipoxidase | 1.13.11.12 | Soyabean whey or meal |
| Papain | 3.4.22.2 | <i>Carica papaya</i> (L) (Fam. <i>Caricaceae</i>) |

(C) Enzymes derived from microbial sources

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|----------------------------------|-----------|-----------------------------------|------------------------|--------------------|
| Alpha-acetolactate decarboxylase | 4.1.1.5 | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Bacillus subtilis</i> | <i>Bacillus brevis</i> | Alpha-acetolactate |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|--------------------------------|-----------|---------------------------------------|---|---------------|
| | | | | decarboxylase |
| Alpha-amylase | 3.2.1.1 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> | <i>Aspergillus niger</i> | Alpha-amylase |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | <i>Bacillus amyloliquefaciens</i> | Alpha-amylase |
| | | <i>Bacillus licheniformis</i> | | |
| | | <i>Bacillus licheniformis</i> | <i>Bacillus licheniformis</i> | Alpha-amylase |
| | | <i>Bacillus licheniformis</i> | <i>Bacillus licheniformis</i> and <i>Bacillus amyloliquefaciens</i> | Alpha-amylase |
| | | <i>Bacillus licheniformis</i> | <i>Geobacillus stearothermophilus</i> | Alpha-amylase |
| | | <i>Bacillus licheniformis</i> | <i>Pseudomonas stutzeri</i> | Alpha-amylase |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Bacillus subtilis</i> | <i>Bacillus megaterium</i> | Alpha-amylase |
| | | <i>Bacillus subtilis</i> | <i>Geobacillus stearothermophilus</i> | Alpha-amylase |
| | | <i>Geobacillus stearothermophilus</i> | | |
| | | <i>Geobacillus stearothermophilus</i> | <i>Geobacillus stearothermophilus</i> | Alpha-amylase |
| | | <i>Rhizopus oryzae</i> | | |
| Alpha-arabinofuranosidase | 3.2.1.55 | <i>Aspergillus niger</i> | | |
| Alpha-galactosidase | 3.2.1.22 | <i>Aspergillus niger</i> | | |
| Alpha-glucosidase (or maltase) | 3.2.1.20 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | | |
| Aminopeptidase | 3.4.11.1 | <i>Aspergillus oryzae</i> | | |
| | | <i>Lactococcus</i> | | |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|---|-----------|--------------------------------------|-----------------------------|--------------|
| | | <i>lactis</i> | | |
| Amylomaltase | 2.4.1.25 | <i>Bacillus amyloliquefaciens</i> | <i>Thermus thermophilus</i> | Amylomaltase |
| Asparaginase | 3.5.1.1 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> | <i>Aspergillus niger</i> | Asparaginase |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Aspergillus oryzae</i> | Asparaginase |
| Aspergillopepsin I | 3.4.23.18 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | | |
| Aspergillopepsin II | 3.4.23.19 | <i>Aspergillus niger</i> | | |
| Beta-amylase | 3.2.1.2 | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| Beta-fructofuranosidase (invertase or saccharase) | 3.2.1.26 | <i>Aspergillus japonicus</i> | | |
| | | <i>Aspergillus niger</i> | | |
| | | <i>Saccharomyces cerevisiae</i> | | |
| Beta-galactosidase (or lactase) | 3.2.1.23 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Bacillus circulans</i> ATCC 31382 | | |
| | | <i>Kluyveromyces lactis</i> | | |
| | | <i>Kluyveromyces marxianus</i> | | |
| | | <i>Saccharomyces sp.</i> | | |
| Beta-glucanase (endo-beta glucanase or endo-1,3-beta-glucanase) | 3.2.1.6 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Disporotrichum dimorphosporum</i> | | |
| | | <i>Humicola insolens</i> | | |
| | | <i>Rasamsonia emersonii</i> | | |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|---------------------------------|-----------|--|------------------------------|------------------------|
| | | <i>Trichoderma longibrachiatum</i> | | |
| Beta-glucosidase | 3.2.1.21 | <i>Aspergillus niger</i> | | |
| Carboxylesterase | 3.1.1.1 | <i>Rhizomucor miehei</i> | | |
| Catalase | 1.11.1.6 | <i>Aspergillus niger</i> | | |
| | | <i>Micrococcus luteus</i> | | |
| Cellulase | 3.2.1.4 | <i>Aspergillus niger</i> | | |
| | | <i>Penicillium funiculosum</i> | | |
| | | <i>Rasamsonia emersonii</i> | | |
| | | <i>Trichoderma longibrachiatum</i> | | |
| | | <i>Trichoderma viride</i> | | |
| Chymosin | 3.4.23.4 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> var. <i>awamori</i> | <i>Camelus dromedarius</i> | Chymosin |
| | | <i>Escherichia coli</i> K-12 strain GE81 | | |
| | | <i>Kluyveromyces lactis</i> | | |
| Cyclodextrin glucanotransferase | 2.4.1.19 | <i>Paenibacillus macerans</i> | | |
| Dextranase | 3.2.1.11 | <i>Chaetomium gracile</i> | | |
| | | <i>Penicillium lilacinum</i> | | |
| Endo-arabinase | 3.2.1.99 | <i>Aspergillus niger</i> | | |
| Endo-protease | 3.4.21.26 | <i>Aspergillus niger</i> | | |
| Glucan 1,3-beta-glucosidase | 3.2.1.58 | <i>Trichoderma harzianum</i> | | |
| Endo-1,3-beta-xylanase | 3.2.1.32 | <i>Humicola insolens</i> | | |
| Endo-1,4-beta-xylanase | 3.2.1.8 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> | <i>Aspergillus niger</i> | Endo-1,4-beta-xylanase |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Aspergillus aculeatus</i> | Endo-1,4-beta-xylanase |
| | | <i>Aspergillus oryzae</i> | <i>Humicola lanuginosus</i> | Endo-1,4-beta-xylanase |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|---|-----------|--------------------------------------|---|---|
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus licheniformis</i> | <i>Bacillus licheniformis</i> | Endo-1,4-beta-xylanase |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Bacillus subtilis</i> | <i>Bacillus subtilis</i> | Endo-1,4-beta-xylanase |
| | | <i>Humicola insolens</i> | | |
| | | <i>Trichoderma longibrachiatum</i> | | |
| Glucoamylase (or amyloglucosidase) | 3.2.1.3 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> | <i>Aspergillus niger</i> | Glucoamylase (or amyloglucosidase) |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Rhizopus delemar</i> | | |
| | | <i>Rhizopus niveus</i> | | |
| | | <i>Rhizopus oryzae</i> | | |
| Glucose isomerase (or xylose isomerase) | 5.3.1.5 | <i>Actinoplanes missouriensis</i> | | |
| | | <i>Bacillus coagulans</i> | | |
| | | <i>Microbacterium arborescens</i> | | |
| | | <i>Streptomyces olivaceus</i> | | |
| | | <i>Streptomyces olivochromogenes</i> | | |
| | | <i>Streptomyces murinus</i> | | |
| | | <i>Streptomyces rubiginosus</i> | | |
| | | <i>Streptomyces rubiginosus</i> | <i>Streptomyces rubiginosus</i> | Glucose isomerase (or xylose isomerase) |
| Glucose oxidase | 1.1.3.4 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Aspergillus niger</i> | Glucose oxidase |
| Glycerophospholipid cholesterol acyltransferase | 2.3.1.43 | <i>Bacillus licheniformis</i> | <i>Aeromonas salmonicida subsp. salmonicida</i> | Glycerophospholipid cholesterol acyltransferase |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|----------------------------------|-----------|------------------------------------|--|--------------------------|
| Hexose oxidase | 1.1.3.5 | <i>Hansenula polymorpha</i> | <i>Chondrus crispus</i> | Hexose oxidase |
| Inulinase | 3.2.1.7 | <i>Aspergillus niger</i> | | |
| Lipase, monoacylglycerol | 3.1.1.23 | <i>Penicillium camembertii</i> | | |
| Lipase, triacylglycerol | 3.1.1.3 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus niger</i> | <i>Candida antarctica</i> | Lipase, triacylglycerol |
| | | <i>Aspergillus niger</i> | <i>Fusarium culmorum</i> | Lipase, triacylglycerol |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Fusarium oxysporum</i> | Lipase, triacylglycerol |
| | | <i>Aspergillus oryzae</i> | <i>Humicola lanuginosa</i> | Lipase, triacylglycerol |
| | | <i>Aspergillus oryzae</i> | <i>Rhizomucor miehei</i> | Lipase, triacylglycerol |
| | | <i>Aspergillus oryzae</i> | <i>Humicola lanuginosa</i> and <i>Fusarium oxysporum</i> | Lipase, triacylglycerol |
| | | <i>Candida rugosa</i> | | |
| | | <i>Hansenula polymorpha</i> | <i>Fusarium heterosporum</i> | Lipase, triacylglycerol |
| | | <i>Mucor javanicus</i> | | |
| | | <i>Penicillium roquefortii</i> | | |
| | | <i>Rhizomucor miehei</i> | | |
| | | <i>Rhizopus arrhizus</i> | | |
| <i>Rhizopus niveus</i> | | | | |
| <i>Rhizopus oryzae</i> | | | | |
| Lysophospholipase | 3.1.1.5 | <i>Aspergillus niger</i> | | |
| Maltogenic alpha-amylase | 3.2.1.133 | <i>Bacillus subtilis</i> | <i>Geobacillus stearothermophilus</i> | Maltogenic alpha-amylase |
| Maltotetrahydrolyase | 3.2.1.60 | <i>Bacillus licheniformis</i> | <i>Pseudomonas stutzeri</i> | Maltotetrahydrolyase |
| Mannan endo-1,4-beta-mannosidase | 3.2.1.78 | <i>Aspergillus niger</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Trichoderma longibrachiatum</i> | | |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|--------------------------------------|-----------|------------------------------------|-----------------------------------|-------------------------------|
| Metalloproteinase | 3.4.24.4 | <i>Aspergillus oryzae</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus coagulans</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| Mucorpepsin (or aspartic proteinase) | 3.4.23.23 | <i>Aspergillus oryzae</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Rhizomucor miehei</i> | Aspartic proteinase |
| | | <i>Cryphonectria parasitica</i> | | |
| | | <i>Rhizomucor miehei</i> | | |
| Pectin esterase | 3.1.1.11 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Aspergillus aculeatus</i> | Pectin esterase |
| Pectin lyase | 4.2.2.10 | <i>Aspergillus niger</i> | | |
| Phospholipase A1 | 3.1.1.32 | <i>Aspergillus oryzae</i> | <i>Fusarium venenatum</i> | Phospholipase A1 |
| Phospholipase A2 | 3.1.1.4 | <i>Aspergillus niger</i> | Porcine pancreas | Phospholipase A2 |
| | | <i>Streptomyces violaceoruber</i> | | |
| 3-Phytase | 3.1.3.8 | <i>Aspergillus niger</i> | | |
| 4-Phytase | 3.1.3.26 | <i>Aspergillus oryzae</i> | <i>Peniophora lycii</i> | 4-Phytase |
| Polygalacturonase (pectinase) | 3.2.1.15 | <i>Aspergillus niger</i> | | |
| | | <i>Aspergillus oryzae</i> | | |
| | | <i>Aspergillus oryzae</i> | <i>Aspergillus aculeatus</i> | Polygalacturonase (pectinase) |
| | | <i>Rhizopus oryzae</i> | | |
| | | <i>Trichoderma longibrachiatum</i> | | |
| Pullulanase | 3.2.1.41 | <i>Bacillus acidopullulyticus</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus licheniformis</i> | | |
| | | <i>Bacillus licheniformis</i> | <i>Bacillus deramificans</i> | Pullulanase |
| | | <i>Bacillus subtilis</i> | | |
| | | <i>Bacillus subtilis</i> | <i>Bacillus acidopullulyticus</i> | Pullulanase |

| Enzyme | EC Number | Production strain | Donor organism | Donor gene |
|--|-----------|-----------------------------------|------------------------------|--|
| | | <i>Bacillus subtilis</i> | <i>Bacillus deramificans</i> | Pullulanase |
| | | <i>Bacillus subtilis</i> | <i>Bacillus naganoensis</i> | Pullulanase |
| | | <i>Klebsiella pneumoniae</i> | | |
| Serine proteinase | 3.4.21.14 | <i>Aspergillus oryzae</i> | | |
| | | <i>Bacillus amyloliquefaciens</i> | | |
| | | <i>Bacillus halodurans</i> | | |
| | | <i>Bacillus licheniformis</i> | | |
| | | <i>Bacillus subtilis</i> | | |
| Serine protease with trypsin specificity | 3.4.21.4 | <i>Fusarium venenatum</i> | <i>Fusarium oxysporum</i> | Serine protease with trypsin specificity |
| Transglucosidase | 2.4.1.24 | <i>Aspergillus niger</i> | | |
| Transglutaminase | 2.3.2.13 | <i>Streptomyces mobaraensis</i> | | |
| Urease | 3.5.1.5 | <i>Lactobacillus fermentum</i> | | |

Notes:

Trichoderma longibrachiatum – also known as *Trichoderma reesei*

Kluyveromyces marxianus – former names *Saccharomyces fragilis* and *Kluyveromyces fragilis*

Kluyveromyces lactis – former name *Saccharomyces lactis*

Rhizomucor miehei – former name *Mucor miehei*

Micrococcus luteus – former name *Micrococcus lysodeikticus*

Paenibacillus macerans – former name *Bacillus macerans*

Rasamsonia emersonii – former name *Talaromyces emersonii*

Klebsiella pneumoniae – former name *Klebsiella aerogenes*

Streptomyces mobaraensis – former name *Streptoverticillium mobaraensis*

Humicola lanuginosa – also known as *Thermomyces lanuginosus*

Mucor javanicus – also known as *Mucor circinelloides* f. *circinelloides*

Penicillium roquefortii – also known as *Penicillium roqueforti*

Hansenula polymorpha – also known as *Pichia angusta*

Geobacillus stearothermophilus – former name *Bacillus stearothermophilus*