



## **RESOLUTION OIV/OENO 365/2009**

### **REVISION OF THE MONOGRAPH ON ENZYMATIC PREPARATIONS (OENO 14/2003)**

THE GENERAL ASSEMBLY,

IN VIEW of article 2, paragraph 2 iv of the Agreement of 3 April 2001 establishing the International Organisation of Vine and Wine was founded,

TAKING NOTE of the works of the “Specification of Oenological Products” expert group,

CONSIDERING resolution Oeno 14/2003 adopted by the OIV

CONSIDERING the explanatory items mentioned in the introductory report in the annex

DECIDES on the proposal of Commission II “Oenology” to modify resolution Oeno 14/2003 in the International Oenological Codex by the following monograph:

### **ENZYMATIC PREPARATIONS**

The prescriptions described below concern all enzymatic preparations susceptible of being used during various operations that can be applied to grapes and their derivatives.

The prescriptions are based on the recommendations from the “General Specifications and Considerations for Enzymes used in Food Processing” drafted by the “Joint FAO/WHO Expert Committee on Food Additives (JECFA), 67th Session, Rome 20 -29 June 2006 published in 2006 in the FAO JECFA monographs.

#### **1. GENERAL CONSIDERATIONS**

Enzymatic preparations can be made from micro-organisms or plants.

When looking for synergies between various enzymatic activities including pectinase, cellulase and hemicellulase, mixtures of preparations made from different strains can be carried out. These preparations can contain one or more active compounds, in addition to supports, diluents, preservatives, antioxidants and other substances compatible with the good manufacturing practices and in accordance with local regulations. In certain cases, preparations can contain cells or cell fragments. Furthermore they can be in either liquid or solid form. The active substances can also

be immobilised on a support admitted for food use.

## **2. LABELLING**

The labelling of enzymatic preparations must at least specify the nature of the preparation (ex. pectinolytic enzyme), the activity (in units by g or ml), the batch number and the expiration date. Enzymatic preparations with multiple technological activities (cf. 4.1) must be mentioned as such on the label (for example: Enzymatic preparation with clarifying or aromatic enhancer properties).

If there is available space, it is desirable that the label has the following information: the main activity the preparation was standardised for, recommended dose and implementation conditions, storage condition for maintaining stability, the nature of additives and carriers used, the nature of enzymatic activities, batch number and expiration date. If there is not enough space, this information shall be indicated on the technical data sheet of the preparation.

The indication that enzymatic preparations were obtained by genetically modified organisms must be mentioned, where relevant.

## **3. ADMITTED ENZYMATIC PREPARATIONS**

All enzymatic preparations with activities presenting a technological interest duly proven in practice and meeting the conditions and criteria mentioned above, are accepted for the treatment of grapes and their by-products.

Enzymatic preparations used must not contain any substance, microorganism, nor enzymatic activity that:

- is harmful to health,
- is harmful to the quality of the products manufactured,
- can lead to the formation of undesirable products,
- or that will give rise or facilitate fraud.

## **4. ENZYMATIC ACTIVITIES**

### **4.1. General considerations**

Enzymatic preparations contain many enzymatic activities. Other than the main

enzymatic activities, (activities for which, respectively, the enzymatic preparation has been standardised) whose technological interest has been duly proven, secondary enzymatic activities are only tolerated if they are set within the technological constraint limits for manufacturing of enzymatic preparations. They must be as limited as possible. Activities which have a negative effect on wine (ex. Cinnamoyl esterase) should not be present in commercialised enzymatic products in oenology. Generally speaking, secondary activities must likewise comply with the requirements formulated above (point 3).

Generally speaking, the secondary activities present in a given preparation must not become the main reason to use the said preparation unless this preparation is declared as multiple technological effects. On a technological level, there is a distinction between the following preparations:

Maceration preparations: facilitate extraction of compounds such as colour, tannins,...

Clarification preparations: facilitate clarification and filtration of musts and wine

Aroma enhancers: reinforces and/or modifies aromatic profile of musts and wine

Stabilisation preparations: facilitates extraction of macromolecules or other substances with a stabilising effect on wine (yeast mannans).

When an enzymatic preparation generates multiple technological effects, duly noted in a practice, (ex. Clarification and aroma enhancer enzymes), whether they are the result of a main and/or secondary activity, they must be declared as such on the label. Different enzymatic activities responsible for these effects must be measured and indicated in the technical preparation data sheet.

Activities are expressed in nKat. (nkat = 1 nmole of transformed substrate or product formed per second by g or ml of the preparation).

## **4.2. Activity measurement**

The enzymatic activities presented are measured under the conditions corresponding to their biochemical characteristics. (pH, temperature) and if possible, the closest to activities encountered in the practice (grape juice, must or wine). The methods implemented must correspond to state of the art in analytical terms and, if possible, be validated in accordance with appropriate international standards (for example: ISO 78-20; ISO 5725).

Results are expressed in nanokatal/g or nanokatal/ml.

When the sought out technological effect results from the action of different enzymes within the same preparation, it is necessary to measure each enzymatic activity. Each of these activities will require special sheets, with the details of the analytical method.

## **5. SOURCES OF ENZYMES AND FERMENTATION ENVIRONMENT**

The microbial sources of enzymes must be non-pathogenic, non-toxic and genetically stable, and the fermentation broth should not leave harmful residues in enzymatic preparations. In the case of microorganisms, a safety study must be conducted in order to ensure that enzymatic preparation produced by a microorganism species (e.g. *Aspergillus niger*) does not present any health risk. This study can be based on principles brought forth on food enzyme guidelines published by the Scientific Committee for Food (SCF), or other equivalent organisations.

The techniques implemented must be compatible with good manufacturing practices and the prescriptions of the International Oenological Codex if yeast and/or lactic bacteria are used.

## **6. CARRIERS , DILUENTS, PRESERVATIVES AND OTHER ADDITIVES**

Substances used as carriers , diluents, preservatives or other additives must not, with a “carry over” effect, disseminate compounds in the grapes and derivative products, which are not compatible with regulations in force in different countries. Moreover, these compounds must not have a negative effect on the organoleptic properties of wine. In the case of immobilised enzymes, the carriers used must comply to standards on material in contact with foodstuffs. For this type of preparation, the content of compounds of the carriers used, susceptible to enter the musts and wine, should be determined and indicated on the label of the enzymatic preparation.

Preservatives such as KCl are added in the liquid enzyme concentrate during manufacturing. These substances prevent the development of micro-organisms during the different formulation operations of products. These substances can be found not only in liquid preparations but also in solid preparations. Given the inevitable “carry over” effect, only preservatives which are compatible to regulations in force in the different countries are authorised.

These substances must be clearly identified and indicated on the label or on the technical data sheet of the commercial product.

## **7. HYGIENE AND MAXIMAL LEVEL OF CONTAMINANTS**

Enzymatic preparations must be produced in accordance with good manufacturing practices:

### **7.1. Lead**

Proceed with the determination according to the method described in chapter II of the International Oenological Codex.

Content less than 5 mg/kg.

### **7.2. Mercury**

Proceed with the determination according to the method described in chapter II of the International Oenological Codex.

Content less than 0.5 mg/kg.

### **7.3. Arsenic**

Proceed with the determination according to the method described in chapter II of the International Oenological Codex.

Content less than 3 mg/kg.

### **7.4. Cadmium**

Proceed with the determination according to the method described in chapter II of the International Oenological Codex.

Content less than 0.5 mg/kg.

### **7.5. Salmonella sp**

Proceed with counting according to the method described in chapter II of the International Oenological Codex.

Absence checked on a 25 g sample.

### **7.6. Total coliforms**

Proceed with counting according to the method described in chapter II of the International Oenological Codex.

Content less than 30/per gram of preparation.

### **7.7. Escherichia coli**

Proceed with counting according to the method described in chapter II of the International Oenological Codex.

Absence checked on a 25 g sample.

## **7.8. ANTIMICROBIAL ACTIVITY**

Non-detectable

## **7.9. SPECIFIC MYCOTOXINS OF DIFFERENT PRODUCTION STRAINS**

Non-detectable

# **8. TECHNICAL DATA SHEET TO BE SUPPLIED BY MANUFACTURER**

Each type of enzymatic preparation must be defined using a technical data sheet. It must contain at least the following information:

- Name of enzyme and biological origin (e.g. pectolytic enzymes of *Aspergillus niger* or pectolytic enzyme of *A. oryzae* expressed as *A. niger*),
- Declared activity (in nKat/g or nKat/ml of preparation)
- Fields and application mode (technological effects and useful details for the implementation of the preparation) ,
- Stability of the preparation and expiration date period based on production date guaranteeing the maintaining of activity, under the given storage conditions (temperature),
- Types of reactions catalysed by the main enzymatic activities
- Main enzymatic activities with IUB number (for example Tannase 3.1.1.20),
- Secondary enzymatic activities with, if possible, the IUB number
- Types of carriers, diluents, preservatives and additives used and their respective contents,

If deemed useful, further information can be added to this technical data sheet.