



**EUROPEAN UNION**

**COMMUNITY PLANT VARIETY OFFICE**

**PROTOCOL FOR DISTINCTNESS, UNIFORMITY AND STABILITY TESTS**

*Cichorium intybus* L. partim

**INDUSTRIAL CHICORY**

UPOV Species Code: CICHO\_INT\_SAT

**Adopted on 01/12/2005**

## **I SUBJECT OF THE PROTOCOL**

The protocol describes the technical procedures to be followed in order to meet the Council Regulation 2100/94 on Community Plant Variety Rights. The technical procedures have been agreed by the Administrative Council and are based on general UPOV Document TG/1/3 and UPOV Guideline TG/172/4 dated 06/04/2005 for the conduct of tests for Distinctness, Uniformity and Stability. This protocol applies to all varieties of *Cichorium intybus* L. **partim** of the family *Compositae*, excluding Witloof chicory (CPVO-TP/173/1) and leaf chicory (UPOV-TG/154/3).

## **II SUBMISSION OF SEED AND OTHER PLANT MATERIAL**

1. The Community Plant Variety Office (CPVO) is responsible for informing the applicant of

- the closing date for the receipt of plant material;
- the minimum amount and quality of plant material required;
- the examination office to which material is to be sent.

A sub-sample of the material submitted for test will be held in the variety collection as the definitive sample of the candidate variety.

The applicant is responsible for ensuring compliance with any customs and plant health requirements.

2. Final dates for receipt of documentation and material by the Examination Office

The final dates for receipt of requests, technical questionnaires and the final date or submission period for plant material will be decided by the CPVO and each Examination Office chosen.

The Examination Office is responsible for immediately acknowledging the receipt of requests for testing, and technical questionnaires. Immediately after the closing date for the receipt of plant material the Examination Office should inform the CPVO whether acceptable plant material has been received or not. However if unsatisfactory plant material is submitted the CPVO should be informed as soon as possible.

3. Plant material requirements

The final dates for request for technical examination and sending of Technical Questionnaire by the CPVO as well as submission date of plant material by the applicant can be found in the S2 supplement of the CPVO Official Gazette and the CPVO website ([www.cpvo.europa.eu](http://www.cpvo.europa.eu)).

|                       |                                                                                                                                                                                                                                                                                                                                                            |
|-----------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Quality of seed:      | Should not be less than the standards laid down for certified seed in Annex 2 of Council Directive 2002/55/EC.                                                                                                                                                                                                                                             |
| Quality of plants:    | Should not be less than the standards laid down for plants in EC Directive 92/33 and implementing measures.                                                                                                                                                                                                                                                |
| Seed Treatment:       | The plant material must not have undergone any treatment unless the CPVO and the examination office allow or request such treatment. If it has been treated, full details of the treatment must be given.                                                                                                                                                  |
| Special requirements: | -                                                                                                                                                                                                                                                                                                                                                          |
| Labelling of sample:  | <ul style="list-style-type: none"><li>- Species</li><li>- File number of the application allocated by the CPVO</li><li>- Breeder's reference</li><li>- Examination reference (if known)</li><li>- Name of applicant</li><li>- The phrase "On request of the CPVO"</li><li>- In the case of a split sample, the quantity of seed being submitted.</li></ul> |

### **III CONDUCT OF TESTS**

#### 1. Variety collection

A variety collection will be maintained for the purpose of establishing distinctness of the candidate varieties in test. A variety collection may contain both living material and descriptive information. A variety will be included in a variety collection only if plant material is available to make a technical examination.

Pursuant to Article 7 of Council Regulation No. 2100/94, the basis for a collection should be the following:

- varieties listed or protected at the EU level or at least in one of the EEA Member States;
- varieties protected in other UPOV Member States;
- any other variety in common knowledge.

The composition of the variety collection in each Examination Office depends on the environmental conditions in which the Examination Office is located.

Variety collections will be held under conditions which ensure the long term maintenance of each accession. It is the responsibility of Examination Offices to replace reference material which has deteriorated or become depleted. Replacement material can only be introduced if appropriate tests confirm conformity with the existing reference material. If any difficulties arise for the replacement of reference material Examination Offices must

inform the CPVO. If authentic plant material of a variety cannot be supplied to an Examination Office the variety will be removed from the variety collection.

2. Material to be examined

Candidate varieties will be directly compared with other candidates for Community plant variety rights tested at the same Examination Office, and with appropriate varieties in the variety collection. When necessary an Examination Office may also include other candidates and varieties. Examination Offices should therefore make efforts to co-ordinate the work with other Offices involved in DUS testing of industrial chicory. There should be at least an exchange of technical questionnaires for each candidate variety, and during the test period, Examination Offices should notify each other and the CPVO of candidate varieties which are likely to present problems in establishing distinctness. In order to solve particular problems Examination Offices may exchange plant material.

3. Characteristics to be used

The characteristics to be used in DUS tests and preparation of descriptions shall be those referred to in the Annex 2. All the characteristics shall be used, providing that observation of a characteristic is not rendered impossible by the expression of any other characteristic, or the expression of a characteristic is prevented by the environmental conditions under which the test is conducted. In the latter case, the CPVO should be informed. In addition the existence of some other regulation e.g. plant health, may make the observation of the characteristic impossible.

The Administrative Council empowers the President, in accordance with Article 23 of Commission Regulation N° 1239/95, to insert additional characteristics and their expressions in respect of a variety.

4. Grouping of varieties

The varieties and candidates to be compared will be divided into groups to facilitate the assessment of distinctness. Characteristics which are suitable for grouping purposes are those which are known from experience not to vary, or to vary only slightly, within a variety and which in their various states of expression are fairly evenly distributed throughout the collection. In the case of continuous grouping characteristics overlapping states of expression between adjacent groups is required to reduce the risks of incorrect allocation of candidates to groups. The characteristics which may be used for grouping are the following:

- (a) Ploidy (characteristic 1)
- (b) Leaf: length (characteristic 4)
- (c) Leaf: intensity of green colour (characteristic 6)
- (d) Root: length (characteristic 14).
- (e) Male sterility (22)

5. Trial designs and growing conditions

The minimum duration of tests will normally be two independent growing cycles. Tests will be carried out under conditions ensuring normal growth. The size of the plots will be such that plants or parts of plants may be removed for measuring and counting without prejudice to the observations which must be made up to the end of the growing period.

The test design is as follows

As a minimum, each test should include a total of 100 plants which should be divided between two or more replicates.

All observations determined by measurements or counting should be made on 40 plants or parts of 40 plants.

All observations on the leaf should be made on the full-grown leaf before deterioration, this means 2 to 3 weeks before harvesting the roots.

All observations on the root should be made immediately after harvesting; assessment of total sugar content within a week from harvesting the roots.

6. Special tests

In accordance with Article 83(3) of Council Regulation No. 2100/94 an applicant may claim either in the Technical Questionnaire or during the test that a candidate has a characteristic which would be helpful in establishing distinctness. If such a claim is made and is supported by reliable technical data, a special test may be undertaken providing that a technically acceptable test procedure can be devised.

Special tests will be undertaken, with the agreement of the President of CPVO, where distinctness is unlikely to be shown using the characters listed in the protocol.

7. Standards for decisions

a) **Distinctness**

A candidate variety will be considered to be distinct if it meets the requirements of Article 7 of Council Regulation No. 2100/94.

b) **Uniformity**

For the assessment of uniformity of open-pollinated varieties, relative uniformity standards should be used.

For the assessment of uniformity of hybrid varieties relative uniformity standards should be applied, excluding clearly recognisable inbred plants. For these clearly recognisable inbred plants a population standard of 3% with an acceptance probability of at least 95% should be applied.

Table of maximum numbers of clearly recognisable inbred plants.

| Number of plants | off-types allowed |
|------------------|-------------------|
| 28-46            | 3                 |
| 47-66            | 4                 |
| 67-88            | 5                 |
| 89-110           | 6                 |
| 111-134          | 7                 |

c) **Stability**

A candidate will be considered to be sufficiently stable when there is no evidence to indicate that it lacks uniformity.

#### **IV REPORTING OF RESULTS**

After each recording season the results will be summarised and reported to the CPVO in the form of a UPOV model interim report in which any problems will be indicated under the headings distinctness, uniformity and stability. Candidates may meet the DUS standards after two growing periods but in some cases three growing periods may be required. When tests are completed the results will be sent by the Examination Office to the CPVO in the form of a UPOV model final report.

If it is considered that the candidate complies with the DUS standards, the final report will be accompanied by a variety description in the format recommended by UPOV. If not the reasons for failure and a summary of the test results will be included with the final report.

The CPVO must receive interim reports and final reports by the date agreed between the CPVO and the examination office.

Interim reports and final examination reports shall be signed by the responsible member of the staff of the Examination Office and shall expressly acknowledge the exclusive rights of disposal of CPVO.

## V **LIAISON WITH THE APPLICANT**

If problems arise during the course of the test the CPVO should be informed immediately so that the information can be passed on to the applicant. Subject to prior agreement, the applicant may be directly informed at the same time as the CPVO particularly if a visit to the trial is advisable.

The interim report as well as the final report shall be sent by the Examination Office to the CPVO.

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## ANNEXES TO FOLLOW

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## ANNEX II

### Technical Questionnaire

#### Types of expression of characteristics:

- QL – Qualitative characteristic
- QN – Quantitative characteristic
- PQ – Pseudo-qualitative characteristic

#### Type of observation of characteristics:

- MG – Single measurement of a group of plants or parts of plants
- MS – Measurement of a number of individual plants or parts of plants
- VG – Visual assessment by a single observation of a group of plants or parts of plants
- VS – Visual assessment by observation of individual plants or parts of plants

When a method of observation is attributed to a certain characteristic, the first differentiation is made depending if the action taken is a visual observation (V) or a measurement (M).

The second differentiation deals with the number of observations the expert attributes to each variety, thus the attribution of either G or S.

If a single observation of a group consisting of an undefined number of individual plants is appropriate to assess the expression of a variety, we talk about a visual observation or a measurement made on a group of plants, thus we attribute the letter G (either VG or MG). If the expert makes more than one observation on that group of plants, the decisive part is that we have at the end only one data entry per variety which means that we have to deal with G (e.g. measurement of plant length on a plot – MG, visual observation of green colour of leaves on a plot – VG).

If it is necessary to observe a number of individual plants to assess the expression of a variety, we should attribute the letter S (thus either VS or MS). Single plant data entries are kept per variety for further calculations like the variety mean (e.g. measurement of length of ears – MS, visual observation of growth habit of single plants in grasses – VS). The number of individual plants to be observed in such cases is stated in section III.5.



## ANNEX I

### TABLE OF CHARACTERISTICS TO BE USED IN DUS-TEST AND PREPARATION OF DESCRIPTIONS

| CPVO<br>N° | UPOV<br>N° | Characteristics                                       | Examples             | Note |
|------------|------------|-------------------------------------------------------|----------------------|------|
| <b>1.</b>  | <b>1.</b>  | <b>Ploidy</b>                                         |                      |      |
| QL         | QL         | diploid                                               | Turquoise            | 2    |
| VS         | VS         | triploid                                              | Perle                | 3    |
|            |            | tetraploid                                            |                      | 4    |
| G          |            | polyploid                                             | Ivoire               | 5    |
| <b>2.</b>  | <b>2.</b>  | <b>Plant: height (at end of first growing season)</b> |                      |      |
| (+)        | (+)        | short                                                 | Perle                | 3    |
| QN         | QN         | medium                                                | Orchies              | 5    |
| VG         | VG         | tall                                                  | Katrien, Luxor       | 7    |
| <b>3.</b>  | <b>3.</b>  | <b>Foliage: attitude</b>                              |                      |      |
| QN         | QN         | erect                                                 | Luxor, Madona, Rubis | 1    |
| VG         | VG         | semi-erect                                            | Fruitosa, Orchies    | 3    |
|            |            | horizontal                                            | Faste                | 5    |
| <b>4.</b>  | <b>4.</b>  | <b>Leaf: length</b>                                   |                      |      |
| (+)        | (+)        |                                                       |                      |      |
| QN         | QN         | short                                                 | Perle                | 3    |
| MS/VG      | VG         | medium                                                | Orchies              | 5    |
| G          |            | long                                                  | Jade, Luxor          | 7    |
| <b>5.</b>  | <b>5.</b>  | <b>Leaf: width</b>                                    |                      |      |
| (+)        | (+)        | narrow                                                | Eva, Luxor, Vanessa  | 3    |
| QN         | QN         | medium                                                | Rubis                | 5    |
| MS/VG      | VG         | broad                                                 | Jade                 | 7    |

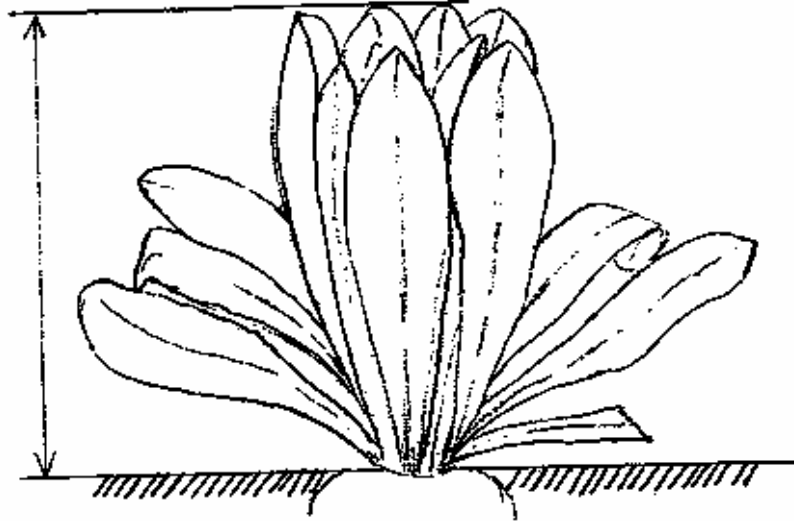
| CPVO<br>N° | UPOV<br>N° | Characteristics                               | Examples      | Note |
|------------|------------|-----------------------------------------------|---------------|------|
| <b>6.</b>  | <b>6.</b>  | <b>Leaf: intensity of green colour</b>        |               |      |
| QN         | QN         | light                                         | Eva           | 3    |
| VG         | VG         | medium                                        | Katrien       | 5    |
| G          |            | dark                                          | Madona, Rubis | 7    |
| <b>7.</b>  | <b>7.</b>  | <b>Leaf: glossiness</b>                       |               |      |
| QN         | QN         | absent or very weak                           |               | 1    |
| VG         | VG         | weak                                          | Luxor         | 3    |
|            |            | medium                                        | Rubis         | 5    |
|            |            | strong                                        |               | 7    |
| <b>8.</b>  | <b>8.</b>  | <b>Leaf: shape in cross section</b>           |               |      |
| QN         | QN         | concave                                       |               | 1    |
| VG         | VG         | flat                                          | Luxor, Madona | 2    |
|            |            | convex                                        |               | 3    |
| <b>9.</b>  | <b>9.</b>  | <b>Leaf: blistering</b>                       |               |      |
| QN         | QN         | absent or very weak                           | Jade          | 1    |
| VG         | VG         | weak                                          | Luxor         | 3    |
|            |            | medium                                        | Bergues       | 5    |
|            |            | strong                                        | Cassel        | 7    |
| <b>10.</b> | <b>10.</b> | <b>Leaf: anthocyanin coloration of midrib</b> |               |      |
| QN         | QN         | absent or very weak                           | Bergues       | 1    |
| VG         | VG         | weak                                          | Luxor, Rubis  | 3    |
|            |            | medium                                        |               | 5    |
|            |            | strong                                        |               | 7    |
| <b>11.</b> | <b>11.</b> | <b>Leaf: undulation of margin</b>             |               |      |
| QN         | QN         | weak                                          | Madona, Rubis | 3    |
| VG         | VG         | medium                                        | Marlene       | 5    |
|            |            | strong                                        |               | 7    |

| CPVO<br>N° | UPOV<br>N° | Characteristics                            | Examples                             | Note |
|------------|------------|--------------------------------------------|--------------------------------------|------|
| <b>12.</b> | <b>12.</b> | <b>Leaf: number of incisions of margin</b> |                                      |      |
| QN         | QN         | absent or very few                         | Luxor                                | 1    |
| VG         | VG         | few                                        | Marlene, Rubis                       | 3    |
|            |            | medium                                     | Katrien                              | 5    |
|            |            | many                                       |                                      | 7    |
| <b>13.</b> | <b>13.</b> | <b>Leaf: depth of incisions of margin</b>  |                                      |      |
| QN         | QN         | shallow                                    | Bergues                              | 3    |
| VG         | VG         | medium                                     |                                      | 5    |
|            |            | deep                                       | Capucijnerbaard                      | 7    |
| <b>14.</b> | <b>14.</b> | <b>Root: length</b>                        |                                      |      |
| QN         | QN         | short                                      |                                      | 3    |
| MS/VG      | MS         | medium                                     | Madona, Marlene                      | 5    |
| G          |            | long                                       | Magdeburger Spitzkopf                | 7    |
| <b>15.</b> | <b>15.</b> | <b>Root: maximum width</b>                 |                                      |      |
| QN         | QN         | narrow                                     | Magdeburger Spitzkopf                | 3    |
| MS/VG      | MS         | medium                                     | Luxor, Rubis                         | 5    |
|            |            | broad                                      | Bergues                              | 7    |
| <b>16.</b> | <b>16.</b> | <b>Root: shape of shoulder</b>             |                                      |      |
| (+)        | (+)        | flat                                       | Luxor                                | 1    |
| PQ         | PQ         | slightly rounded                           | Madona, Rubis                        | 2    |
| VG         | VG         | clearly rounded                            |                                      | 3    |
|            |            | conical                                    | Magdeburger Spitzkopf                | 4    |
| <b>17.</b> | <b>17.</b> | <b>Root: total sugar content</b>           |                                      |      |
| (+)        | (+)        | very low                                   | Sabau 3                              | 1    |
| QN         | QN         | low                                        | Luxor, Markise                       | 3    |
| MG         | MG         | medium                                     | Brinco, Orchies, Vanessa             | 5    |
|            |            | high                                       | Dageraad, Fredonia, Katrien, Marlene | 7    |
|            |            | very high                                  | Eva                                  | 9    |

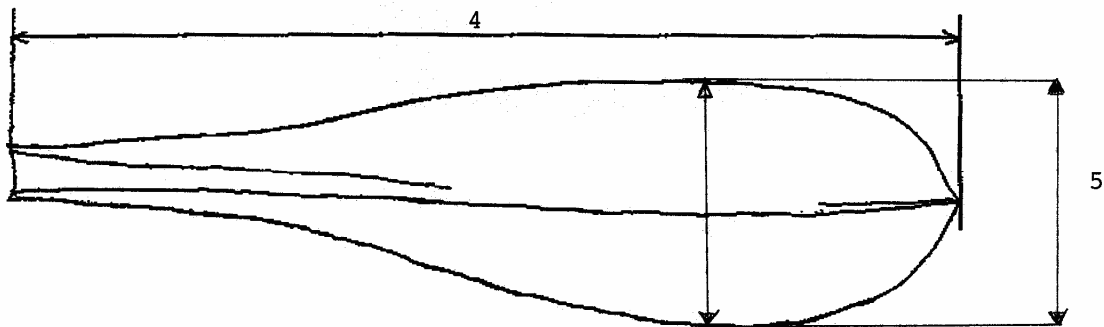
| CPVO<br>N° | UPOV<br>N° | Characteristics                                | Examples         | Note |
|------------|------------|------------------------------------------------|------------------|------|
| <b>18.</b> | <b>18.</b> | <b>Bolting tendency (from an early sowing)</b> |                  |      |
| QN         | QN         | absent or very weak                            | Katrien, Orchies | 1    |
| MS/VG      | VG         | weak                                           | Bergues, Marlene | 3    |
|            |            | medium                                         | Madona           | 5    |
|            |            | strong                                         | Vanessa          | 7    |
|            |            | very strong                                    | Inula            | 9    |
| <b>19.</b> | <b>19.</b> | <b>Flowering stem: height</b>                  |                  |      |
| QN         | QN         | short                                          |                  | 3    |
| VG         | VG         | medium                                         |                  | 5    |
|            |            | tall                                           |                  | 7    |
| <b>20.</b> | <b>20.</b> | <b>Flowering stem: branching</b>               |                  |      |
| QN         | QN         | weak                                           |                  | 3    |
| VG         | VG         | medium                                         |                  | 5    |
|            |            | strong                                         |                  | 7    |
| <b>21.</b> | <b>21.</b> | <b>Flower: colour</b>                          |                  |      |
| PQ         | PQ         | white                                          |                  | 1    |
| VG         | VG         | pink                                           |                  | 2    |
|            |            | blue                                           | Luxor            | 3    |
| <b>22.</b> | <b>22.</b> | <b>Male sterility</b>                          |                  |      |
| QL         | QL         |                                                |                  |      |
| VS         | VS         | absent                                         | Luxor            | 1    |
| G          |            | present                                        | Turquoise        | 9    |

## EXPLANATIONS AND METHODS

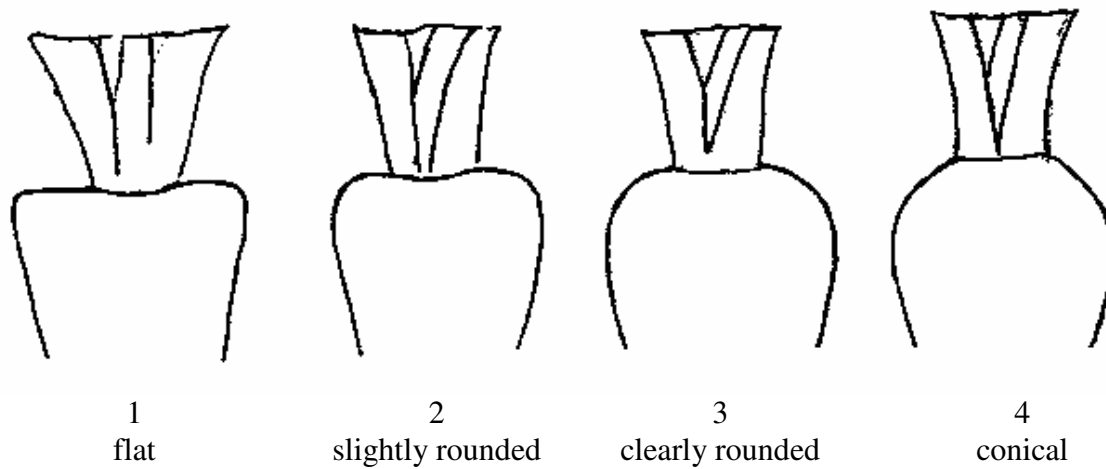
Ad 2: Plant: height (at end of first growing season)



Ad 4 and 5: Leaf: length (4) and width (5)



Ad 16: Root: shape of shoulder



Ad 17: Root: total sugar content

The total sugar content should be measured on the basis of bulk samples.

A sample of 25 roots should be taken randomly from each plot. The roots should be thoroughly washed and all impurities should be removed.

A representative sub-sample of pulp is produced by taking small quantities of material from throughout each of the roots i.e. from the top to the base, at equal distances, and from the outer to the central part of the root. This can be achieved, for example, by making incisions to the centre of the root at 2-3 cm intervals along the length of each root.

The sub-sample of pulp is homogenized and the resultant juice is then filtered under pressure. Readings for the juice are then taken from a refractometer. Three separate readings should be taken to obtain a representative result.

## LITERATURE

Frese, L., Dambroth, M. and Bramm, A., 1991: Breeding Potential of Root Chicory (*Cichorium intybus* L. var. *sativum*) Plant Breeding 106, 107-113.

## **ANNEX II**

The Technical Questionnaire is available on the CPVO website under the following reference:  
CPVO-TQ/172/2