PROTOCOL FOR TESTS ON DISTINCTNESS, UNIFORMITY AND STABILITY

X Triticosecale Witt.

TRITICALE

UPOV Code: TRITL

Adopted on 27/04/2022

Entry into force on 01/08/2022
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**CPVO-TP/121/3-Corr**

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1. SUBJECT OF THE PROTOCOL AND REPORTING

1.1 Scope of the technical protocol

This Technical Protocol applies to all varieties of *X Triticosecale Witt*..


1.2 Entry into Force

The present protocol enters into force on **01.08.2022**. Any ongoing DUS examination of candidate varieties started before the aforesaid date will not be affected by the approval of the Technical Protocol. Technical examinations of candidate varieties are carried out according to the TP in force when the DUS test starts. The starting date of a DUS examination is considered to be the due date for submitting of plant material for the first test period.

In cases where the Office requests to take-over a DUS report for which the technical examination has either been finalized or which is in the process to be carried out at the moment of this request, such report can only be accepted if the technical examination has been carried out according to the CPVO TP which was in force at the moment when the technical examination started.

1.3 Reporting between Examination Office and CPVO and Liaison with Applicant

1.3.1 Reporting between Examination Office and CPVO

The Examination Office shall deliver to the CPVO a preliminary report (“the preliminary report”) no later than two weeks after the date of the request for technical examination by the CPVO.

The Examination Office shall also deliver to the CPVO a report relating to each growing period (“the interim report”) and, when the Examination Office considers the results of the technical examination to be adequate to evaluate the variety or the CPVO so requests, a report relating to the examination (“the final report”).

The final report shall state the opinion of the Examination Office on the distinctness, uniformity and stability of the variety. Where it considers those criteria to be satisfied, or where the CPVO so requests, a description of the variety shall be added to the report.

If a report is negative the Examination Office shall set out the detailed reasons for its findings.

The interim and the final reports shall be delivered to the CPVO as soon as possible and no later than on the deadlines as laid down in the designation agreement.

1.3.2 Informing on problems in the DUS test

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 pertinent agreement, on matters of particular urgency, the applicant may be directly informed at the same time as the CPVO particularly if a visit to the trial is advisable.

1.3.3 Sample keeping in case of problems

If the technical examination has resulted in a negative report, the CPVO shall inform the Examination Office as soon as possible in case that a representative sample of any relevant testing material shall be kept.

2. MATERIAL REQUIRED

2.1 Plant material requirements

Information with respect to the agreed closing dates and submission requirements of plant material for the technical examination of varieties can be found on [https://public.plantvarieties.eu/publication](https://public.plantvarieties.eu/publication) in the special issue S2/S3 of the Official Gazette of the Office. General requirements on submission of samples are also to be found following the same link.
2.2 Informing the applicant of plant material requirements

The CPVO informs the applicant that

- he is responsible for ensuring compliance with any customs and plant health requirements.
- the plant material supplied should be visibly healthy, not lacking in vigor, nor affected by any important pest or disease.
- the plant material should not have undergone any treatment which would affect the expression of the characteristics of the variety, unless the competent authorities allow or request such treatment. If it has been treated, full details of the treatment must be given.

2.3 Informing about problems on the submission of material

The Examination Office shall report to the CPVO immediately in cases where the test material of the candidate variety has not arrived in time or in cases where the material submitted does not fulfil the conditions laid down in the request for material issued by the CPVO.

In cases where the examination office encounters difficulties to obtain plant material of reference varieties the CPVO should be informed.

3. METHOD OF EXAMINATION

3.1 Number of growing cycles

The minimum duration of tests should normally be two independent growing cycles.

The testing of a variety may be concluded when the Examination Office can determine with certainty the outcome of the test.

3.2 Testing Place

Tests are normally conducted at one place. In the case of tests conducted at more than one place, guidance is provided in TGP/9 "Examining Distinctness" http://www.upov.int/edocs/tgpdocs/en/tgp_9.pdf.

3.3 Conditions for Conducting the Examination

The tests should be carried out under conditions ensuring satisfactory growth for the expression of the relevant characteristics of the variety and for the conduct of the examination.

The optimum stage of development for the assessment of each characteristic is indicated by a number in the third column of the Table of Characteristics. The stages of development denoted by each number are described in Chapter 8.3.

3.4 Test design

Each test should be designed to result in a total of at least 2000 plants, which should be divided between at least two replicates.

The assessment of the characteristic “seasonal type” should be designed to result in a total of at least 300 plants.

If ear rows are used, the test should be conducted on at least 100 ear-rows.

In case of hybrids, the parent lines have to be included in the test and should be tested and assessed as any other variety. The observation on the hybrid variety itself should be made on at least 200 plants.

The design of the tests should be such that plants or parts of plants may be removed for measurement or counting without prejudice to the observations which must be made up to the end of the growing cycle.

3.5 Special tests for additional characteristics

In accordance with Article 23 of Implementing Rules Nº 874/2009 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.
3.6 Constitution and maintenance of a variety collection

The process for the constitution and the maintenance of a variety collection can be summarized as follows:

Step 1: Making an inventory of the varieties of common knowledge

Step 2: Establishing a collection ("variety collection") of varieties of common knowledge which are relevant for the examination of distinctness of candidate varieties

Step 3: Selecting the varieties from the variety collection which need to be included in the growing trial or other tests for the examination of distinctness of a particular candidate variety.

3.6.1 Forms of variety collection

The variety collection shall comprise variety descriptions and living plant material, thus a living reference collection. The variety description shall be produced by the EO unless special cooperation exists between EOs and the CPVO. The descriptive and pictorial information produced by the EO shall be held and maintained in a form of a database.

3.6.2 Living Plant Material

The EO shall collect and maintain living plant material of varieties of the species concerned in the variety collection.

3.6.3 Range of the variety collection

The living variety collection shall cover at least those varieties that are suitable to climatic conditions of a respective EO.

3.6.4 Making an inventory of varieties of common knowledge for inclusion in the variety collection

The inventory shall include varieties protected under National PBR (UPOV contracting parties) and Community PBR, varieties registered in the Common Catalogue, the OECD list, the Conservation variety list and varieties in trade or in commercial registers for those species not covered by a National or the Common Catalogue.

The variety collection shall include all varieties used as components (generally inbred lines) of all the hybrid varieties included in the variety collection, as well as varieties of common knowledge in their own right.

3.6.5 Maintenance and renewal/update of a living variety collection

The EO shall maintain seeds in conditions which will ensure germination and viability, periodical checks, and renewal as required. For the renewal of existing living material, the identity of replacement living plant material shall be verified by conducting side-by-side plot comparisons between the material in the collection and the new material.

4. ASSESSMENT OF DISTINCTNESS, UNIFORMITY AND STABILITY

The prescribed procedure is to assess distinctness, uniformity and stability in a growing trial.

4.1 Distinctness

4.1.1 General recommendations

It is of particular importance for users of this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 9 'Examining Distinctness' (http://www.upov.int/edocs/tgpdocs/en/tgp_9.pdf) prior to making decisions regarding distinctness. However, the following points are provided for elaboration or emphasis in this Technical Protocol.

To assess distinctness of hybrids, a pre-screening system on the basis of the parental lines and the formula may be established according to the following recommendations:

(i) description of parental lines according to the Technical Protocols;

(ii) check of the distinctness of the parental lines in comparison with the reference collection, based on the characteristics in the table of characteristics in order to screen the closest inbred lines;

(iii) check of the distinctness of the hybrid formula in comparison with those of the hybrids in common knowledge, taking into account the closest inbred lines;

(iv) assessment of the distinctness at the hybrid level of varieties with a similar formula.

Further guidance is provided in documents TGP/9 "Examining Distinctness" and TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability".
4.1.2 Consistent differences
The differences observed between varieties may be so clear that more than one growing cycle is not necessary. In addition, in some circumstances, the influence of the environment is not such that more than a single growing cycle is required to provide assurance that the differences observed between varieties are sufficiently consistent. One means of ensuring that a difference in a characteristic, observed in a growing trial, is sufficiently consistent is to examine the characteristic in at least two independent growing cycles.

4.1.3 Clear differences
Determining whether a difference between two varieties is clear depends on many factors, and should consider, in particular, the type of expression of the characteristic being examined, i.e. whether it is expressed in a qualitative, quantitative, or pseudo-qualitative manner. Therefore, it is important that users of these Technical Protocols are familiar with the recommendations contained in the UPOV-General Introduction to DUS prior to making decisions regarding distinctness.

If distinctness is assessed using the 2 x 1% criterion, the difference between two varieties is clear if the respective characteristics are significantly different in the same direction at the 1% level in at least two out of three years. The tests in each year are based on Student's two-tailed t-test of the differences between variety means with standard errors estimated using the residual mean square from the analysis of the variety x replicate plot means.

If distinctness is assessed by the combined over years distinctness analysis (COYD) the difference between two varieties is clear if the respective characteristics are different at the 1% significance level or less (p<0.01) in a test over either two or three years.

If the significance level or statistical methods proposed are not appropriate the method used should be clearly described.

4.1.4 Number of plants/parts of plants to be examined
Unless otherwise indicated, for the purposes of distinctness, all observations on single plants should be made on 10 plants or parts taken from each of 10 plants and any other observations made on all plants in the test, disregarding any off-type plants.

In the case of observations of parts taken from single plants, the number of parts to be taken from each of the plants should be 1.

4.1.5 Method of observation
The recommended method of observing the characteristic for the purposes of distinctness is indicated by the following key in the third column of the Table of Characteristics (see document TGP/9 “Examining Distinctness”, Section 4 “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

Type of observation: visual (V) or measurement (M)
"Visual" observation (V) is an observation made on the basis of the expert's judgment. For the purposes of this document, "visual" observation refers to the sensory observations of the experts and, therefore, also includes smell, taste and touch. Visual observation includes observations where the expert uses reference points (e.g. diagrams, example varieties, side-by-side comparison) or non-linear charts (e.g. colour charts). Measurement (M) is an objective observation against a calibrated, linear scale e.g. using a ruler, weighing scales, colorimeter, dates, counts, etc.

Type of record: for a group of plants (G) or for single, individual plants (S)
For the purposes of distinctness, observations may be recorded as a single record for a group of plants or parts of plants (G), or may be recorded as records for a number of single, individual plants or parts of plants (S). In most cases, "G" provides a single record per variety and it is not possible or necessary to apply statistical methods in a plant-by-plant analysis for the assessment of distinctness.

In cases where more than one method of observing the characteristic is indicated in the Table of Characteristics (e.g. VG/MG), guidance on selecting an appropriate method is provided in document TGP/9, Section 4.2.
4.2 Uniformity

4.2.1 It is of particular importance for users of this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 10 'Examining Uniformity' (http://www.upov.int/edocs/tgpdocs/en/tgp_10.pdf) prior to making decisions regarding uniformity. However, the following points are provided for elaboration or emphasis in this Technical Protocol:

4.2.2 The recommended sample size for the assessment of uniformity is indicated by the following key in the table of characteristics:

(A) sample size of 100 plants / parts of plants / ear rows
(B) sample size of 2000 plants

For the assessment of uniformity of mainly self-pollinated varieties, a population standard of 0.6% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 2000 plants, 18 off-types are allowed.

For the assessment of uniformity in a sample of 100 ear-rows, plants or parts of plants, a population standard of 6% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 100 ear-rows, plants or parts of plants, 10 off-types are allowed.

An ear-row is considered to be an off-type ear-row if there is more than 1 off-type plant within that ear-row.

For characteristics with the key "A" in the list of characteristics with the exception of characteristics 1 and 2 the assessment of uniformity can be done in 2 steps. In a first step, 20 plants or parts of plants are observed. If no off types are observed, the variety is declared to be uniform. If more than 6 off-types are observed, the variety is declared not to be uniform. If 1 to 6 off-types are observed, an additional sample of 80 plants or parts of plants must be observed.

For the assessment of uniformity of hybrid varieties, a population standard of 10% and an acceptance probability of at least 95% should be applied. In case of characteristics indicated by B, the sample size for the assessment of uniformity may be reduced to 200 plants. In case of a sample size of 200 plants, 27 off-types are allowed. In case of a sample size of 100 ear-rows, plants or parts of plants, 15 off-types are allowed.

Where the assessment of a hybrid variety involves the parent lines, the uniformity of the hybrid variety should, in addition to an examination of the hybrid variety itself, also be assessed by examination of the uniformity of its parent lines.

For all varieties except hybrid varieties, a re-submission of plant material may be allowed for the second growing cycle if in the first growing cycle the number of off-types did not exceed 28 plants in a sample size of 2000 plants (Population standard of 1 % with an acceptance probability of ≥ 95%) or 21 plants, parts of plants or ear rows in a sample size of 100 (Population standard of 15% with an acceptance probability of ≥ 95%).

4.3 Stability

4.3.1 It is of particular importance for users of this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 11 'Examining Stability' (http://www.upov.int/edocs/tgpdocs/en/tgp_11.pdf)

In practice, it is not usual to perform tests of stability that produce results as certain as those of the testing of distinctness and uniformity. However, experience has demonstrated that, for many types of variety, when a variety has been shown to be uniform, it can also be considered to be stable.

4.3.2 Where appropriate, or in cases of doubt, stability may be further examined by testing a new seed or plant stock to ensure that it exhibits the same characteristics as those shown by the initial material supplied.

5. GROUPING OF VARIETIES AND ORGANISATION OF THE GROWING TRIAL

5.1 The selection of varieties of common knowledge to be grown in the trial with the candidate varieties and the way in which these varieties are divided into groups to facilitate the assessment of distinctness are aided by the use of grouping characteristics.

5.2 Grouping characteristics are those in which the documented states of expression, even where produced at different locations, can be used, either individually or in combination with other such characteristics: (a) to select varieties of common knowledge that can be excluded from the growing trial used for examination of distinctness; and (b) to organise the growing trial so that similar varieties are grouped together.
5.3 The following have been agreed as useful grouping characteristics:
   a) Time of ear emergence (characteristic 6)
   b) Stem: density of hairs on neck (characteristic 10)
   c) Lower glume: hairs on external surface (characteristic 15)
   d) Seasonal type (characteristic 21)

5.4 If other characteristics than those from the Technical Protocol are used for the selection of varieties to be included into the growing trial, the EQ shall inform the CPVO and seek the prior consent of the CPVO before using these characteristics.

5.5 Guidance for the use of grouping characteristics, in the process of examining distinctness, is provided through the UPOV-General Introduction to DUS and document TGP/9 “Examining Distinctness”.

6. INTRODUCTION TO THE TABLE OF CHARACTERISTICS

6.1 Characteristics to be used

The characteristics to be used in DUS tests and preparation of descriptions shall be those referred to in the table of characteristics. 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 or by specific legislation on plant health. In the latter case, the CPVO should be informed.

The Administrative Council empowers the President, in accordance with Article 23 of Commission Regulation N°874/2009, to insert additional characteristics and their expressions in respect of a variety.

6.2 States of expression and corresponding notes

States of expression are given for each characteristic to define the characteristic and to harmonize descriptions. Each state of expression is allocated a corresponding numerical note for ease of recording of data and for the production and exchange of the description. All relevant states of expression are presented in the characteristic.

Further explanation of the presentation of states of expression and notes is provided in UPOV document TGP/7 “Development of Test Guidelines”.

6.3 Example Varieties

Where appropriate, example varieties are provided to clarify the states of expression of each characteristic.

6.4 Legend

For column ‘CPVO N°’:

G Grouping characteristic -see Chapter 5
QL Qualitative characteristic
QN Quantitative characteristic
PQ Pseudo-qualitative characteristic
(+ ) Explanations for individual characteristics -see Chapter 8.2

For column ‘UPOV N°’:

The numbering of the characteristics is provided as a reference to the UPOV guideline.

(*) UPOV Asterisked characteristic: Characteristics that are important for the international harmonization of variety descriptions.
For column ‘Stage, method’:
MG, MS, VG, VS - see Chapter 4.1.5

(A), (B) Recommended sample size for the assessment of uniformity - see Chapter 4.2.2

(a) Explanations covering several Characteristics - see Chapter 8.1
00-99 Explanations on growth stages - see Chapter 8.3

(w): winter type varieties
(s): spring type varieties
### 7. TABLE OF CHARACTERISTICS

<table>
<thead>
<tr>
<th>CPVO N°</th>
<th>UPOV N°</th>
<th>Stage, Method</th>
<th>Characteristics</th>
<th>Examples varieties</th>
<th>Note</th>
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<tbody>
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<td>1.</td>
<td>00</td>
<td>Seed: coloration with phenol</td>
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<td>(+)</td>
<td>VG/A</td>
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<td>QN</td>
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<td>very light to light</td>
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<td>UPOV N°</td>
<td>Stage, Method</td>
<td>Characteristics</td>
<td>Examples varieties</td>
<td>Note</td>
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<td>Plant: frequency of plants with recurved flag leaves</td>
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|         |         | very early to early | early to medium | Raptus (w)       | 5 |
|         |         | medium       | medium to late    | Adverdo (w)      | 7 |
| G       |         | very late    | late to very late |                    | 8 |
|         |         |              |                   | very late        | 9 |</p>
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<tr>
<td></td>
<td></td>
<td>long</td>
<td>Kolorit (w)</td>
<td></td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>long to very long</td>
<td></td>
<td></td>
<td>8</td>
</tr>
<tr>
<td></td>
<td></td>
<td>very long</td>
<td>Fronteira (w)</td>
<td></td>
<td>9</td>
</tr>
<tr>
<td>21.</td>
<td>24.</td>
<td></td>
<td>Seasonal type</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(+)</td>
<td>(*)</td>
<td>VG</td>
<td>winter type</td>
<td>Kasyno (w)</td>
<td>1</td>
</tr>
<tr>
<td>PQ</td>
<td></td>
<td>alternative type</td>
<td></td>
<td>RGT Bivouac (w)</td>
<td>2</td>
</tr>
<tr>
<td>G</td>
<td></td>
<td>spring type</td>
<td>Dublet (s)</td>
<td></td>
<td>3</td>
</tr>
</tbody>
</table>
EXPLANATIONS ON THE TABLE OF CHARACTERISTICS

8.1 Explanations covering several characteristics

Characteristics containing the following key in the third column of the Table of Characteristics should be examined as indicated below:

a) Observations should be made on spikelets in the mid-third of ear.

8.2 Explanations for individual characteristics

Ad. 1: Seed: coloration with phenol

Method for Determination of Phenol Reaction

- Number of seeds per test: 100 seeds. The grains should not have been treated chemically.
- Equipment: Petri dishes (approx. 9 cm diameter).
- Preparation of seeds: Soak in tap water for 16 to 20 hours, drain and remove surface water, place the grains with crease downwards, cover dish with lid.
- Concentration of solution: 1 per cent Phenol-solution (freshly made up).
- Amount of solution: The grains should be about 3/4 covered.
- Place: Laboratory
- Light: Daylight - out of direct sunshine.
- Temperature: 18 to 20°C.
- Time of recording: 4 hours (after adding solution).
- Scale of recording: See characteristic 1 in the Table of Characteristics.
- Note: At least two example varieties should be included as a control.

Any alternative method may be used if it gives the same results.

Ad 2: Coleoptile: anthocyanin coloration

Method for the Determination of Anthocyanin Coloration

- Number of seeds per test: 100 seeds
- Preparation of seeds: Set up non-dormant seeds on moistened filter paper covered with a Petri dish lid during germination.
- Place: Laboratory or glasshouse.
- Light: After the coleoptiles have reached a length of about 1 cm in darkness, they are placed in artificial light (daylight equivalent), 13000 to 15000 lux continuously for 3 - 4 days.
- Temperature: 15 to 20°C.
- Time of recording: Coleoptiles fully developed (about 1 week) at stage 09-11.
- Scale of recording: See characteristic 2 in the Table of Characteristics.
- Note: At least two example varieties should be included as a control when testing for distinctness.

Any alternative method may be used if it gives the same results.

Ad 3: Plant: growth habit

The growth habit should be assessed from the attitude of the leaves and tillers. The angle formed by the outer leaves and the tillers with an imaginary middle axis should be used.

1 erect 3 semi-erect 5 intermediate 7 semi-prostrate 9 prostrate
Ad. 4: Plant: frequency of plants with recurved flag leaves

1 (absent or very low): all or almost all flag leaves are rectilinear
3 (low): about 1/4 of the plants with recurved flag leaves
5 (medium): about 1/2 of the plants with recurved flag leaves
7 (high): about 3/4 of the plants with recurved flag leaves
9 (very high): almost all or all flag leaves are recurved

Ad. 6: Time of ear emergence

Time of ear emergence is reached when the first spikelet is visible on 50% of ears.

Ad. 10: Stem: density of hairs on neck

Ad. 13: Plant: length

The length of plant includes stem, ear, awns and scurs.

Ad. 14: Lower glume: length of first beak
Ad. 16: Straw: pith in cross section

Pith in cross section should be observed halfway between base of ear and uppermost node. All stems of the plant should be checked and the highest score per plant recorded.

Ad. 17: Ear: density
The density is the ratio of the number of spikelets per ear length.

Ad. 18: Ear: distribution of awns

Ad. 19: Ear: length of scurs or awns
Observations should be made at the tip of the ear.
Ad. 20: Ear: length

Length of ear should be observed excluding awns and scurs.

Ad. 21: Seasonal type

The seasonal type (need of vernalization) should be assessed on plots sown in springtime. Example varieties should always be included in the trial. When the example varieties behave according to its description, candidate varieties can be described. At the time when the latest spring type variety is fully mature (stage 91/92 of the Zadoks decimal code) growth stage reached by the respective variety should be assessed. The states of expression are defined as follows:

Winter type (high need of vernalization): the plants have reached stage 45 of the Zadoks decimal code (boots swollen) at maximum.

Alternative type (partial need of vernalization): the plants have exceeded stage 45 of the Zadoks decimal code (as a rule they have exceeded stage 75) and have reached stage 90 at maximum.

Spring type (no need or very weak need of vernalization): the plants have exceeded stage 90 of the Zadoks decimal code.

Seasonal type is not related to winter hardiness. Spring type varieties have no need for vernalization but may have winter hardiness.
### 8.3 Explanations on growth stages

The descriptions of the growth stages of the Zadoks decimal code for cereals (ZADOKS et al., 1974)

<table>
<thead>
<tr>
<th>Zadoks Decimal code</th>
<th>Description</th>
<th>Zadoks Decimal code</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Germination</strong></td>
<td></td>
<td><strong>Boot</strong></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>Dry seed</td>
<td>41</td>
<td>Flag leaf sheath extending</td>
</tr>
<tr>
<td>01</td>
<td>Start of imbibition</td>
<td>43</td>
<td>Boots just visibly swollen</td>
</tr>
<tr>
<td>03</td>
<td>Imbibition complete</td>
<td>45</td>
<td>Boots swollen</td>
</tr>
<tr>
<td>05</td>
<td>Radicle emerged from seed</td>
<td>47</td>
<td>Flag leaf sheath opening</td>
</tr>
<tr>
<td>07</td>
<td>Coleoptile emerged from seed</td>
<td>49</td>
<td>First awns visible</td>
</tr>
<tr>
<td>09</td>
<td>Leaf just at coleoptile tip</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| **Seedling growth** |             | **Inflorescence emergence** |             |
| 10                  | First leaf through coleoptile | 50                  | First spikelet of inflorescence visible |
| 11                  | First leaf unfolded | 53                  | 1/4 of inflorescence emerged |
| 12                  | 2 leaves unfolded | 55                  | 1/2 of inflorescence emerged |
| 13                  | 3 leaves unfolded | 57                  | 3/4 of inflorescence emerged |
| 14                  | 4 leaves unfolded | 59                  | Emergence of inflorescence completed |
| 15                  | 5 leaves unfolded |             |             |
| 16                  | 6 leaves unfolded | 60                  | Beginning on anthesis |
| 17                  | 7 leaves unfolded | 65                  | Anthesis half-way |
| 18                  | 8 leaves unfolded | 69                  | Anthesis completed |
| 19                  | 9 or more leaves unfolded |             |             |

| **Tillering**       |             | **Milk development** |             |
| 20                  | Main shoot only | 71                  | Caryopses watery ripe |
| 21                  | Main shoot and 1 tiller | 73                  | Early milk |
| 22                  | Main shoot and 2 tillers | 75                  | Medium milk |
| 23                  | Main shoot and 3 tillers | 77                  | Late milk |
| 24                  | Main shoot and 4 tillers |             |             |
| 25                  | Main shoot and 5 tillers | 83                  | Early dough |
| 26                  | Main shoot and 6 tillers | 85                  | Soft dough |
| 27                  | Main shoot and 7 tillers | 87                  | Hard dough |
| 28                  | Main shoot and 8 tillers |             |             |
| 29                  | Main shoot and 9 or more tillers | 91                  | Caryopses hard (difficult to divide with thumbnail) |

| **Stem elongation**  |             | **Dough development** |             |
| 30                  | Pseudo stem erection | 92                  | Caryopses hard (can no longer be dented with thumbnail) |
| 31                  | 1st node detectable | 93                  | Caryopses loosening in daytime |
| 32                  | 2nd node detectable | 94                  | Overripe, straw dead and collapsing |
| 33                  | 3rd node detectable | 95                  | Seed dormant |
| 34                  | 4th node detectable | 96                  | Viable seed giving 50% germination |
| 35                  | 5th node detectable | 97                  | Seed not dormant |
| 36                  | 6th node detectable | 98                  | Secondary dormancy induced |
| 37                  | Flag leaf just visible |             |             |
| 38                  | Flag leaf ligule/collar just visible | 99                  | Secondary dormancy lost |
9. LITERATURE

10. TECHNICAL QUESTIONNAIRE

The Technical Questionnaire is available on the CPVO website under the following reference: CPVO-TQ/121/3 - *X Triticosecale* Witt. - triticale