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# INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS GENEVA

## DRAFT

<u>Associated Document</u> <u>to the</u> <u>General Introduction to the Examination</u> <u>of Distinctness, Uniformity and Stability and the</u> Development of Harmonized Descriptions of New Varieties of Plants (document TG/1/3)

#### **DOCUMENT TGP/9**

#### **"EXAMINING DISTINCTNESS"**

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# **SECTION 1: INTRODUCTION**

From TGP/4

1.1 Article 7 of the 1991 Act of the UPOV Convention establishes that "a variety shall be deemed to be distinct if it is clearly distinguishable from any other variety whose existence is a matter of common knowledge at the time of filing the application."

1.2 The "General Introduction to the Examination of Distinctness, Uniformity and Stability and the Development of Harmonized Descriptions of New Varieties of Plants" (document TG/1/3), hereinafter referred to as "the General Introduction", states, with respect to common knowledge (see document TG/1/3, Section 5.2.2), that:

"Specific aspects which should be considered to establish common knowledge include, among others:

(a) commercialization of propagating or harvested material of the variety, or publishing a detailed description;

(b) the filing of an application for the grant of a breeder's right or for the entering of a variety in an official register of varieties, in any country, which is deemed to render that variety a matter of common knowledge from the date of the application, provided that the application leads to the grant of a breeder's right or to the entering of the variety in the official register of varieties, as the case may be;

(c) existence of living plant material in publicly accessible plant collections.

Common knowledge is not restricted to national or geographical borders."

Further information on varieties whose existence is a matter of common knowledge ("varieties of common knowledge") is provided in document TGP/3, "Varieties of Common Knowledge".

1.3 Although not exhaustive, and taking into account that these aspects have to be considered on a worldwide basis, it is clear that the list of varieties of common knowledge for a given species can be very large. Therefore, it may be useful to employ a process to reduce the number of varieties of common knowledge which need to be included in growing trials or other tests for direct comparison against a variety which is the subject of an application for plant breeders' rights ("candidate variety"). That process can be summarized in the following steps:

- 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 according to Section 2 "Constitution of Variety Collections" of document TGP/4 [cross ref.];
- Step 3: Selecting the varieties from the variety collection which should be included in the growing trial or other tests for the examination of distinctness of a particular candidate variety.

1.4 The identification of varieties of common knowledge which should be included in the variety collection (Step 1) and the establishment of a variety collection (Step 2) are addressed in

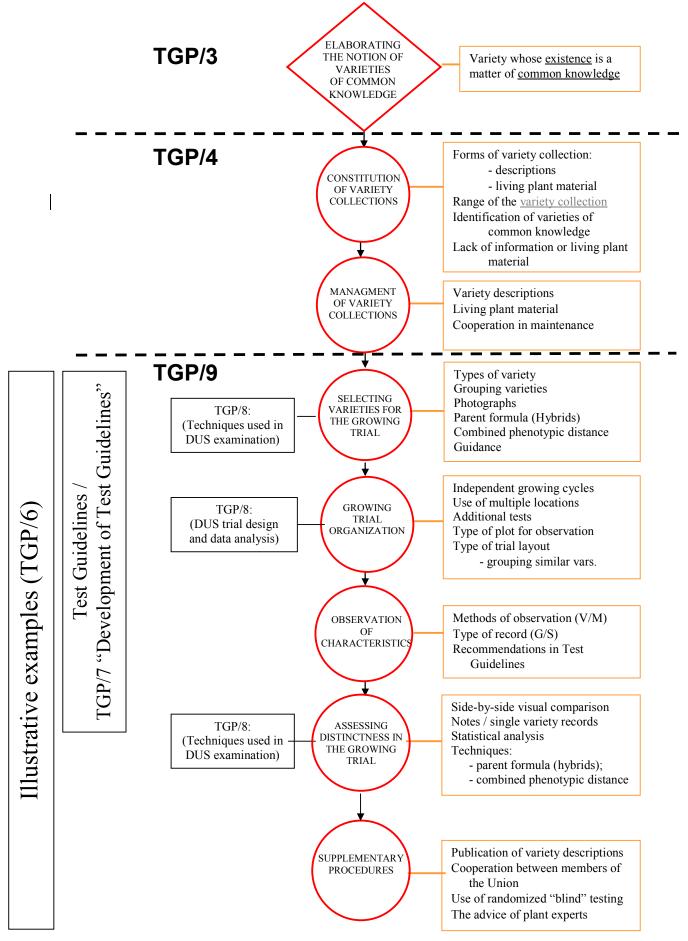
document TGP/4, "[Constitution and] Management of Variety Collections" [cross ref.]. The purpose of this document is to provide guidance on the use of the variety collection to select varieties for the growing trial (Section 2 of this document) and the examination of distinctness in

growing trials (Sections 3 to 5), with the aim of providing an effective examination of distinctness. The UPOV Convention has a means to address situations where a candidate variety is incorrectly considered to be distinct, in that it requires a breeder's right to be declared null and void if the distinctness requirement was not complied with at the time of the grant of the breeder's right (see Article 21(1)(i) of the 1991 Act and Article 10(1) of the 1978 Act). However, in order to maintain the effectiveness of protection, such cases should remain the exceptions and the procedures for assessing distinctness should be as robust as possible. In that regard, supplementary procedures may be used to reinforce the examination of distinctness. The use of supplementary procedures in the examination of distinctness is considered in Section 6 of this document.

1.5 For the purposes of this document, the term "growing trial" covers growing trials or other tests used in the examination of distinctness.

1.6 The following diagram presents a schematic overview of the process of examining distinctness as considered in documents TGP/3, TGP/4 and TGP/9:





## SECTION 2: SELECTING VARIETIES FOR THE GROWING TRIAL

## 2.1 Introduction

2.1.1 A key step (see Section 1.3 *[cross ref.]*) in the examination of distinctness is the selection of varieties of common knowledge, from within the <u>variety collection</u>, to be included in the growing trial.

2.1.2 This section (Section 2) explains how the variety description can be used for selecting varieties to be included in the growing trial. The General Introduction (Chapter 5.3.1.3) explains that "[...] where a candidate variety can be distinguished in a reliable way from varieties of common knowledge, by comparing documented descriptions, it is not necessary to include those varieties of common knowledge in a growing trial with the respective candidate variety. However, where there is no possibility of clearly distinguishing them from the candidate variety, the varieties should be compared with the candidate variety in a growing trial or other appropriate test. This emphasizes the importance of harmonization of variety descriptions in minimizing the workload of the DUS examiner." Thus, the description for a variety may not be sufficient to conclude that a variety is distinct from a candidate variety (see document TGP/4, Section 2.1.1 [cross ref.]). In such cases, the variety should be included in the growing trial unless supplementary procedures are used in a way which provides an effective examination of distinctness overall (see Section 6: Supplementary Procedures [cross ref.]).

A variety collection may be limited to certain types of variety or groups of varieties 2.1.3 within a species or subspecies (see TGP/4: Section 2.2.1.1 [cross ref.]). However, a variety collection may comprise more than one type or group of varieties. Therefore, the identification of types or groups of varieties within the variety collection can be a first step in the selection of varieties to be included in the growing trials. In this document, "type of variety" means that varieties of that type have a common trait, or traits, often physiological traits (e.g. long/short day varieties), by which they are recognized beyond the purposes of the examination of DUS. The term "group of varieties" means a grouping of varieties specifically for the purposes of the examination of distinctness (e.g. long/short rachilla hair type in barley). The General Introduction (Chapter 5.3.1.1) clarifies that it "is necessary to examine distinctness in relation to all varieties of common knowledge. However, a systematic individual comparison may not be required with all varieties of common knowledge. For example, where a candidate variety is sufficiently different, in the expression of its characteristics, to ensure that it is distinct from a particular group (or groups) of varieties of common knowledge, it would not be necessary for a systematic individual comparison with the varieties in that group (or those groups)." The same principle applies for types of varieties as for groups of varieties in this context.

2.1.4 The following paragraphs consider how suitable types or groups of varieties may be identified for the purpose of selecting varieties for the growing trial. Where it becomes apparent that information provided for a candidate variety or a variety in the variety collection was not sufficiently accurate, taking into account the anticipated level of environmental and observation variation, for example where it results in the wrong attribution of the type or group for the variety, it may be necessary to conduct a further growing trial containing the relevant varieties.

# 2.2 Types of varieties

Document TGP/4, "Constitution and management of variety collections", (see Section 2.1.1.2 *[cross ref.]*) identifies criteria with regard to types which might be used in the process of examining distinctness as follows:

From TGP/4: Sections 2.2.1.2

(i) recognition of different types of variety within the relevant UPOV Test Guidelines, or by the establishment of separate Test Guidelines for different variety types within, for example, the same species;

(ii) the variety collection might be limited by taking into account certain physiological traits of the varieties (e.g. earliness, day length susceptibility, frost resistance, etc.) according to the climatic conditions for which it is adapted.

# 2.3 Grouping varieties

2.3.1 Function

2.3.1.1 The selection of varieties to be grown in the trial with the candidate varieties is aided by the use of grouping characteristics to identify groups of varieties within the <u>variety collection</u>.

2.3.1.2 The General Introduction sets out the functions of grouping characteristics (see document TG/1/3, Section 4.8. Functional Categorization of Characteristics), as follows:

"1. Characteristics in which the documented states of expression, even where recorded at different locations, can be used to select, either individually or in combination with other such characteristics, varieties of common knowledge that can be excluded from the growing trial used for examination of distinctness.

"2. Characteristics in which the documented states of expression, even where recorded at different locations, can be used, either individually or in combination with other such characteristics, to organize the growing trial so that similar varieties are grouped together."

2.3.1.3 Function 1 above identifies the role of grouping characteristics in selecting varieties for the growing trial. Where UPOV has developed Test Guidelines (UPOV Test Guidelines), these will identify useful grouping characteristics. However, as indicated by Function 2, grouping characteristics are provided in the UPOV Test Guidelines for two reasons. Therefore, the use of each grouping characteristic for excluding varieties from the growing trial, as opposed to its use for organizing the growing trial so that similar varieties are grouped together (see S Section 3.6.2 *[cross ref.]*), should be considered carefully.

2.3.2 Criteria

2.3.2.1 The General Introduction sets out the criteria (document TG/1/3, Section 4.8, Functional Categorization of Characteristics) for the selection of grouping characteristics as follows:

"1. (a) Qualitative characteristics or

(b) Quantitative or pseudo-qualitative characteristics which provide useful discrimination between the varieties of common knowledge from documented states of expression recorded at different locations.

[ ... ]"

2.3.2.2 The states of expression of the grouping characteristics for the candidate varieties need to be known before the growing trial in order to be able to use that information in selecting varieties for the growing trial. For that reason, for the grouping characteristics identified in the UPOV Test Guidelines, information is requested in the Technical Questionnaire (TQ). Document TGP/7, "Development of Test Guidelines", (Guidance Notes 13.4) states that:

"(a) Grouping characteristics selected from the Table of Characteristics should, in general, receive an asterisk in the Table of Characteristics and be included in the Technical Questionnaire.

(b) TQ characteristics selected from the Table of Characteristics should, in general, receive an asterisk in the Table of Characteristics and be used as grouping characteristics. TQ characteristics are not restricted to those characteristics used as grouping characteristics;

(c) Asterisked characteristics are not restricted to those characteristics selected as grouping or TQ characteristics."

Whilst TQ characteristics are, in general, included in the Technical Questionnaire in order to act as grouping characteristics, it should be noted that, in certain cases, characteristics may be included in the Technical Questionnaire for reasons other than providing information on grouping. Therefore, TQ characteristics should not be assumed to always be appropriate for grouping.

2.3.2.3 The identification of characteristics as useful grouping characteristics in the UPOV Test Guidelines is based on the information which is likely to be available from other members of the Union and to be requested from the breeder in the Technical Questionnaire (Technical Questionnaire characteristics). However, further characteristics may also be useful for grouping where the information available to the DUS examiner provides useful discrimination between varieties from documented states of expression for those characteristics, e.g. where the variety descriptions are obtained from the same growing trial, such as can be the case from the first growing cycle where the DUS examination involves two growing cycles (see Section 2.3.5 *[cross ref.]*).

2.3.2.4 Where necessary, the criteria set out in Section 2.3.2.1 *[cross ref.]* can be used to identify suitable grouping characteristics from the Table of Characteristics in addition to those grouping characteristics listed in the Test Guidelines. Those criteria can also be used to identify suitable grouping characteristics where there are no UPOV Test Guidelines.

## 2.3.3 Use of grouping characteristics

2.3.3.1 The use of grouping characteristics to identify those varieties in the variety collection which can be excluded from the growing trial is influenced by the type of expression of the characteristics chosen. In that respect it is recalled that grouping characteristics should be qualitative characteristics or should be quantitative or pseudo-qualitative characteristics which provide useful discrimination between the varieties of common knowledge from documented states of expression recorded at different locations.

# Qualitative characteristics

2.3.3.2 The use of qualitative characteristics for grouping is relatively straightforward because, as a general rule, qualitative characteristics are  $[not] / [less likely to be^1]$  influenced by the environment (see document TG/1/3, Section 4.4.1) and for qualitative characteristics, the difference between two varieties may be considered clear if one or more characteristics have expressions that fall into two different states in the UPOV Test Guidelines (see document TG/1/3, Section 5.3.3.2.1). Therefore, in the case of qualitative characteristics, subject to the consistency of the observation on both the candidate variety and varieties in the variety collection, it is possible, in general, to exclude from the growing trial varieties which have a different state of expression to a candidate variety.

## Example (qualitative characteristic):

In the case of a qualitative characteristic: "Leaf: variegation", with the states absent (Note 1): present (Note 9), it would be possible to exclude from the growing trial varieties in the variety collection which have no variegation (Note 1), where the candidate variety is variegated (Note 9).

### *Quantitative and Pseudo-qualitative characteristics*

2.3.3.3 In the case of quantitative and pseudo-qualitative characteristics, it is not possible to specify a general rule for discriminating between varieties on the basis of documented states of expression recorded at different locations. However, such characteristics can be used for grouping where there is a sufficient difference in the states of expression of varieties in the variety collection and the candidate variety, subject to the consistency of the observation on both the candidate variety and varieties in the variety collection:

### *Example (quantitative characteristic):*

In the case of a quantitative characteristic, e.g. "Plant: height", represented on a 1 to 9 scale it might, for example, be possible to exclude from the growing trial varieties in the variety collection which are very short (Notes 1 and 2) or very tall (Notes 8 and 9), if the candidate variety is of medium height (Note 5).

## Example (pseudo-qualitative characteristic):

In the case of a pseudo-qualitative characteristic, e.g. "Petal: color", with the states: white (Note 1); yellow (2); green (3); pink (4); purple (5), it might, for

<sup>&</sup>lt;sup>1</sup> Change proposed by the TC. It has subsequently been observed that this change would cause divergence with the General Introduction.

example, be possible to exclude from the growing trial varieties in the variety collection which are yellow and green, if the candidate variety is pink.

In the case of both quantitative and pseudo-qualitative grouping characteristics, the range of varieties which can be excluded from the growing trial is determined by the influence of the environment on the states of expression, the difference in the environments where the varieties were observed and the consistency of the observation of the varieties in the variety collection.

2.3.3.4 The use of color characteristics for grouping is explored in document TGP/14, Section 2.3: Glossary of Technical, Botanical and Statistical Terms Used in UPOV Documents: Botanical Terms: Color: color characteristics *[cross ref.]*.

## 2.3.4 Combining grouping characteristics

As explained in Section 2.3.1.2 *[cross ref.]*, grouping characteristics "can be used to select, either individually or in combination with other such characteristics, varieties of common knowledge that can be excluded from the growing trial used for examination of distinctness". This clarifies that grouping characteristics can be used in combination to exclude varieties from the growing trial. Thus, for the examples in Section 2.3.3 *[cross ref.]*, it might be possible to use the characteristics in combination:

*Example (characteristics in combination):* 

## Candidate variety

Plant height:	medium (Note 5)
Leaf variegation:	present (Note 9)
Petal color:	pink (Note 4)

On the basis of the assumptions in Section 2.3.3, it could be possible to exclude from the growing trial concerning the candidate variety: any varieties with Plant height of Notes 1, 2, 8 or 9; any varieties with Leaf variegation: absent (Note 1); and any varieties with Petal color: yellow (Note 2) or green (Note 3).

2.3.5 Grouping using information from the same growing trial

2.3.5.1 Where information is obtained for all varieties from the same growing trial, e.g. from the first growing cycle where the DUS examination involves two growing cycles, it may be possible to obtain a higher level of discrimination from the grouping characteristics and, in particular, for quantitative and pseudo-qualitative characteristics. For example, in the case of the example for a quantitative characteristic in Section 2.3.3.3 *[cross ref.]*, "Plant: height", represented on a 1 to 9 scale, it was suggested that it might, for example, be possible to exclude from the growing trial varieties in the variety collection which are very short (Notes 1 and 2) and very tall (Notes 8 and 9), if the candidate variety was of medium height (Note 5) on the basis of information obtained from the first growing cycle of a DUS examination, it might be possible to exclude, for example, varieties in the variety collection with Note 3 or Note 7 from the second growing cycle.

2.3.5.2 In the case of some perennial crops, e.g. fruit trees, a second growing cycle may be conducted using the trial established for the first growing cycle. In such cases, the notion of

"excluding" varieties from the second growing cycle could mean that there would be no observation of the excluded varieties.

2.3.5.3 As noted in Section 2.3.2.3 *[cross ref.]*, the identification of characteristics as useful grouping characteristics in the UPOV Test Guidelines is based on the information which is likely to be available from other members of the Union and to be requested from the breeder in the Technical Questionnaire (Technical Questionnaire characteristics). However, further characteristics may also be useful for grouping where the information available to the DUS examiner provides useful discrimination between varieties from documented states of expression for those characteristics, e.g. where the variety descriptions are obtained from the same growing trial, such as from the first growing cycle where the DUS examination involves two growing cycles. This is particularly relevant for quantitative and pseudo-qualitative characteristics, for which the states of expression are particularly influenced by the environment.

# 2.3.6 Effectiveness of grouping

The use of grouping characteristics can be a very effective means of reducing the number of varieties which need to be included in the growing trial. In particular, in cases where there are a small number of candidate varieties and a good number of grouping characteristics with high levels of discrimination, the number of varieties in the variety collection which can be excluded from the growing trial can be high. However, in other situations, in particular where there are large numbers of candidate varieties and few grouping characteristics with high levels of discrimination, the possibilities to exclude varieties from the growing trial may be limited because there may be candidate varieties of many or all of the groups defined by the grouping characteristics. In such cases, the grouping characteristics may still play an important role in organizing the growing trial so that similar varieties are grouped together (see Section 3.6.2 *[cross ref.]*).

# 2.4 Photographs

2.4.1 Photographs can provide useful information for selecting varieties from within the variety collection to be included in the growing trial. In particular, photographs may provide information on characteristics not included in the TQ. This may, for example, concern shapes, plant structures or color patterns which are not easy for applicants to describe by means of Notes in the Table of Characteristics and, therefore, might not be included as characteristics in Section 5 of the TQ. In addition, the information provided in photographs on characteristics included in the TQ may be more discriminatory than that provided in Section 5 of the TQ and may allow more varieties to be excluded from the growing trial.

2.4.2 Document TGP/7, "Development of Test Guidelines", indicates that, where useful for the DUS examination, the UPOV Test Guidelines may require that a representative color photograph of the variety accompanies the information provided in the Technical Questionnaire. In such cases, it is recommended that guidance be provided by the authority to enhance the usefulness of the photograph (e.g. to include a metric scale and a color scale in the picture, to define what parts of the plant should be included, to specify the light conditions and the background color, etc). However, despite such guidance and the best endeavors of the breeder, photographs of the variety may not always accurately reflect its characteristics. Discrepancies between the characteristics of the variety and the appearance of the characteristics of the variety in a photograph, where those discrepancies arise solely from the photographic process, should not be a basis for rejecting an application for the variety.

## 2.5 Parent Formula of Hybrid Varieties

In the case of variety collections which contain hybrid varieties, it may be appropriate to use the parent formula as a basis to select varieties for inclusion in the growing trial. The use of the parent formula requires that the difference between parent lines is sufficient to ensure that the hybrid obtained from those parents is distinct. Details of the parent formula technique are provided in TGP/8 [cross ref.].

## 2.6 Combined Phenotypic Distance

## 2.6.1 Introduction

2.6.1.1 The General Introduction (document TG/1/3) explains in Section 5.3.3 that "A variety may be considered to be clearly distinguishable if the difference in characteristics is consistent and clear". Thus, the examination of distinctness can be based on a characteristic-by-characteristic approach, with a requirement that for two varieties to be considered distinct there should be a "minimum difference" for at least one characteristic. The "minimum difference" for a characteristic may either be set at a fixed level, determined by the DUS examiner or on the basis of statistics, or may be based on the judgement of the DUS examiner.

2.6.1.2 In the characteristic-by-characteristic / minimum difference approach, at least as a first step, differences between varieties which are less than the minimum difference for a characteristic are not considered: such differences may be considered in the growing trial where very similar varieties can be compared on the basis of the sum of a number of small differences.

2.6.1.3 When judging distinctness between varieties in the growing trial, DUS examiners consider the overall level of difference.

2.6.1.4 The "combined phenotypic distance" approach is intended to reflect the observation of the DUS examiner. As with distance measurement methods such as Mahalanbois and Gower, the combined phenotypic distance method combines the information for several characteristics for which there are differences. The result is a calculated value derived from all the differences observed. The distance measurement method can give more or less weighting to characteristics according to, for example, their genetic determination, their influence by the environment, the size of the difference and correlation between characteristics.

2.6.1.5 The appropriate parameters for the calculation of the combined phenotypic distance are determined by DUS examiners with the aim of reflecting the overall level of difference and of providing an objective basis for a decision on distinctness. Above a certain level, the combined phenotypic distance allows varieties which are very distinct to be excluded from the growing trial used for the examination of distinctness. The combined phenotypic distance can also be used to organize the growing trial so that the most similar varieties are grouped together.

2.6.1.6 The benefit of combined phenotypic distance is that it provides an overall assessment of the difference between varieties and optimizes the ability of DUS examiners to distinguish similar varieties.

2.6.1.7 The combined phenotypic distance is mainly applicable to self-pollinated varieties and is particularly efficient where there is a large number of varieties in the variety collection.

2.6.2 Methods

2.6.2.1 GAIA

### 2.6.2.1.1 Use of the GAIA methodology

The GAIA methodology is mainly used after a first growing cycle to identify those varieties of common knowledge which can be excluded from the subsequent growing cycle(s) because they have a "distinctness plus" GAIA distance (see Section 2.6.2.1.2 [cross ref.]) with respect to all the candidate varieties, thus demonstrating that all candidate varieties are distinct with respect to those particular varieties of common knowledge. GAIA can also identify similar varieties, on which the DUS examiner will need to focus attention in the subsequent growing cycle(s).

## 2.6.2.1.2 "Distinctness Plus"

2.6.2.1.2.1 The threshold for the combined phenotypic distance used to exclude varieties of common knowledge from the growing trial is called "Distinctness Plus" and is set by the DUS examiner at a level which is higher than the difference required to establish distinctness. This has the purpose of ensuring that all pairs of varieties having a distance equal or greater than the "Distinctness Plus" threshold would be shown to be distinct if grown in another trial.

2.6.2.1.2.1 It is important that the Distinctness Plus threshold is based on experience gained with the varieties of common knowledge and minimizes the risk of excluding varieties of common knowledge which should be compared to one or more candidate varieties in the subsequent growing trial(s).

2.6.2.1.3 Details of the GAIA method are provided in document TGP/8 [cross ref.].

2.6.2.2 Other Methods

[There are a range of other statistical methods in use in agricultural research that can be used in the examination of distinctness. Those include ANOVA and multiple range tests. Providing the underlying assumptions are met, those other statistical methods are as acceptable as the other methods mentioned in this section.]

# 2.7 Guidance and sources of information

2.7.1 The sections above identify factors which might be used for selecting varieties for inclusion in the growing trial. The following are useful sources of information in that respect:

- (a) DUS experts from other members of the Union;
- (b) breeders / applicants through:

(i) information provided in Section 5 of the Technical Questionnaire (Characteristics of the variety to be indicated by the applicant);

(ii) information provided in Section 6 of the Technical Questionnaire (Similar varieties and differences from these varieties);

(iii) information provided in Section 7 of the Technical Questionnaire (Additional information which may help in the examination of the variety, including e.g. photographs); and

- (iv) additional information;
- (c) other plant experts

2.7.2 The process of selecting varieties from within the variety collection, including decisions on which of the factors above are appropriate, requires appropriate knowledge of the variety collection and the requirements for distinctness and it is recommended that, where necessary, guidance is sought from relevant experts, particularly experienced DUS examiners.

[2.7.3 Some illustrations of the way in which the process is applied by members of the Union are provided in document TGP/6, "Arrangements for DUS Testing"]

# **SECTION 3: GROWING TRIAL ORGANIZATION<sup>2</sup>**

### 3.1 Introduction

General information concerning growing trial organization for DUS examination is provided in document TGP/8 *[cross ref.]*. The information in document TGP/8 includes guidance concerning replicated and randomized trial designs. The following section focuses on the situation where the growing trial is to be organized to facilitate side-by-side visual comparison.

## **3.2** Grouping of similar varieties

Section 2 *[cross ref.]* explains factors which might be used for selecting varieties for inclusion in the growing trial. Those factors can also be used to organize the growing trial such that similar varieties are grouped together in order to facilitate direct visual comparisons in the growing trial. The factors are:

(a) Grouping characteristics

As noted in Section 2.3.1.2 [cross ref.], grouping characteristics are:

"[...]

"2. Characteristics in which the documented states of expression, even where recorded at different locations, can be used, either individually or in combination with other such characteristics, to organize the growing trial so that similar varieties are grouped together."

(b) Photographs

See Section 2.4 [cross ref.]

(c) Parent Formula of Hybrid Varieties

See TGP/8 [cross ref.]

(d) Combined Phenotypic Distance

See TGP/8 [cross ref.]

<sup>&</sup>lt;sup>2</sup> The TC agreed that Section 3.1 to Section 3.6.1 should be repeated in document TGP/10, "Uniformity", or should be moved from TGP/9 to TGP/8, "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability". The TWC Chairperson agreed to the inclusion of those sections (Number of growing cycles; The notion of independent growing cycles; Use of multiple locations in the examination of distinctness; Additional Tests; Type of plot for observation; and Type of trial layout) in TGP/8 and they have been deleted from TGP/9.

# SECTION 4: OBSERVATION OF CHARACTERISTICS

### 4.1 Introduction

4.1.1 An important source of information on the observation of characteristics is the UPOV Test Guidelines, where those are available. In addition to the presentation of the characteristics and the states of expression in the Table of Characteristics (Chapter 7), further information is, where appropriate, provided in the Explanations on the Table of Characteristics (Chapter 8) in the form of explanations and/or illustrations. Document TGP/7, "Development of Test Guidelines", (see Guidance Note: GN 28) also explains that one of the purposes of including example varieties in the UPOV Test Guidelines is to illustrate the states of expression of a characteristic. However, the difficulty in selecting suitable example varieties which satisfy all the requirements for inclusion in the UPOV Test Guidelines means that a set of example varieties is not always provided for all characteristics.

4.1.2 In the absence of UPOV Test Guidelines, the principles set out in document TGP/7, "Development of Test Guidelines", provide suitable guidance with regard to the development and observation of characteristics for DUS testing.

Suitable training is required to ensure that observations by a DUS examiner for a 4.1.3 characteristic are consistent and that repeatability between observers can be achieved. Such consistency and repeatability are important for the use of variety descriptions in the process of examining distinctness (see Section 2 [cross ref.]), notwithstanding the fact that variation in variety descriptions will also occur as a result of the influence of the environment. Document TGP/7, "Development of Test Guidelines", (see Guidance Note: GN 28) explains that, in addition to illustrating the states of expression of a characteristic, the other purpose of example varieties in the UPOV Test Guidelines is to provide the basis for ascribing the appropriate state of expression to each variety and, thereby, to develop internationally harmonized variety descriptions. It is difficult to identify example varieties which can satisfy that universal requirement and that is reflected in the fact that, as mentioned above, a set of example varieties is not always provided for all characteristics. However, identification of a suitable set of example varieties for all characteristics at a DUS trial center does not present the same difficulties and is an important measure for ensuring consistency and repeatability of observations at a national or regional level.

# 4.2 Method of observation (Visual or measurement)

The expression of characteristics can be observed visually (V) or by measurement (M).

4.2.1 <u>Visual observation (V)</u>

4.2.1.1 Visual observation (V) is an observation made on the basis of the expert's judgement. 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. color charts).

4.2.1.2 Where they fulfill the requirements for the examination of DUS, visual observations can be used. They are generally quicker and cheaper than measurements but, because they are based on the expert's judgement, they have a particularly important requirement for training and

experience to ensure that observations by a DUS examiner for a characteristic are consistent and that repeatability between observers can be achieved.

## 4.2.2 <u>Measurement (M)</u>

Measurement (M) is an objective observation against a calibrated, linear scale e.g. using a ruler, weighing scales, colorimeter, dates, counts, etc.

### 4.2.3 <u>Selecting the Method of Observation</u>

The choice of the method for observations for the assessment of distinctness should take into account the following aspects:

(a) Type of expression of the characteristic:

*Qualitative (QL) characteristics:* qualitative characteristics are, in general, observed visually;

*Quantitative (QN) characteristics:* quantitative characteristics can be measured or visually observed. The General Introduction explains that:

"5.4.1 In cases where there is very little variation within varieties, the determination of distinctness is usually on the basis of a visual assessment, rather than by statistical methods."

[...]

"5.5.2.2.1 Quantitative characteristics are not necessarily assessed by measuring or counting and can be assessed visually. Where there is doubt regarding the use of a normally visually assessed quantitative characteristic as the distinguishing characteristic in relation to another variety, it should be measured, if that is possible with reasonable effort."

*Pseudo-qualitative characteristics:* Pseudo-qualitative characteristics are, in general, observed visually.

(b) Variability between and within varieties:

For the assessment of distinctness, visual observations are particularly suitable where there is sufficient variation between varieties, and a low level of variation within varieties. Measurements provide a higher level of information. The features of propagation determine the level of genetic variation within varieties. Vegetatively propagated, truly self-pollinated and mainly self-pollinated varieties normally have relatively little variation within varieties. Within cross-pollinated and synthetic varieties, variation is normally greater than for self-pollinated and vegetatively propagated varieties, especially in quantitative and some pseudo-qualitative characteristics.

(c) Number of varieties in the variety collection and in the growing trial: more precision may be necessary in order to examine distinctness where there is a large number of varieties included in the growing trial. Measurements provide more precise data.

(d) Resources (equipment, staff): visual observation is usually less time-consuming than measurements. However, measurements for some characteristics may be partly automated (e.g. imaging) and different characteristics may be measured simultaneously (e.g. thousand seed weight and kernel length; length and width of petals).

(e) Relation between workload and precision required.

# 4.3 Type of record(s)

# 4.3.1 <u>Introduction</u>

4.3.1.1 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).

4.3.1.2 The following sections consider the type of records which may be obtained and the way in which they may be used for the assessment of distinctness.

# 4.3.2 <u>Single record for a group of plants or parts of plants (G)</u>

4.3.2.1 If there is relatively little variation within varieties (excluding off-types), compared to the variation between varieties, the state of expression of a characteristic can be recorded as a single record for a group of plants or parts of plants (G), for the assessment of distinctness. These conditions are fulfilled in most characteristics in self-pollinated and vegetatively propagated varieties and for most qualitative and pseudo-qualitative characteristics in cross-pollinated varieties. In the case of some quantitative characteristics in self-pollinated and vegetatively propagated varieties, it may be appropriate to obtain records for individual plants (see Section 4.3.3.1 [cross ref.]).]

4.3.2.2 The record (G) may, for example, be in the form of: a Note (e.g. 1, 2, 3 etc.) corresponding to a state of expression in the UPOV Test Guidelines; a value (e.g. RHS Colour Chart reference number); a measurement (e.g. length (cm), weight (g), date (18-12-2005), count (3) etc.); an image etc.

4.3.2.3 The record (G) may result from an overall observation of a plot (e.g. leaf color, time of beginning of flowering) or it may result from an observation of parts of plants taken from a group of plants (e.g. color of lower side of leaf, hairiness of sheath of lowest leaf). The sample size of the group should be representative for the variety. Recommendations on an appropriate sample size are provided in the test guidelines.

# Example (VG)

Visual observation (VG): "Lowest leaf: hairiness of leaf sheaths" in barley (self-pollinated): leaves of several plants are observed and the appropriate state of expression recorded for the variety: Note 1 (absent), or Note 9 (present);

# Example (MG)

Measurement (MG): "Plant: height" in wheat (self-pollinated): an overall measurement in the plot.

4.3.2.4 In most cases, "G" provides a single record per variety and it is not possible or necessary to apply statistical methods for the assessment of distinctness. In some cases of "G", e.g. where there are several repetitions or plots, or more than one growing trial, more than one record per variety may be obtained, in which case statistical methods may be applied.

# 4.3.3 <u>Records for a number of single, individual plants or parts of plants (S)</u>

In cases where records for a number of single, individual plants are made (S), statistical analysis of those individual records may be used as the basis for the assessment of distinctness, or the records may be used to calculate a mean value for a variety or for a plot, which would be the basis for the assessment of distinctness.

# 4.3.3.1 Distinctness assessment based on variety mean value

Records for individual plants may be appropriate for some quantitative characteristics in self-pollinated and vegetatively propagated varieties. In particular, in the case of observations on certain parts of plants it might be necessary to measure a number of individual plants in order to determine the precise expression of the variety by calculating the mean value from individual measurements:

# Example (MS)

"Leaflet: length" in pea (self pollinated): a leaf from each of 20 plants is measured (MS). The value of each plant is used for calculation of the mean value, which can be considered in the same way as described in Section 4.3.2 [cross ref.].

# 4.3.3.2 Distinctness assessment based on individual plant records

If there is considerable variation within varieties, which is the normal situation for quantitative characteristics in cross-pollinated varieties, it is necessary to obtain records for individual plants in order to determine the mean expression and the variation within a variety. Distinctness is then assessed by comparing variety means calculated on the basis of the individual plant data, taking into account the random variation inherent in the variety means.

# Example (MS)

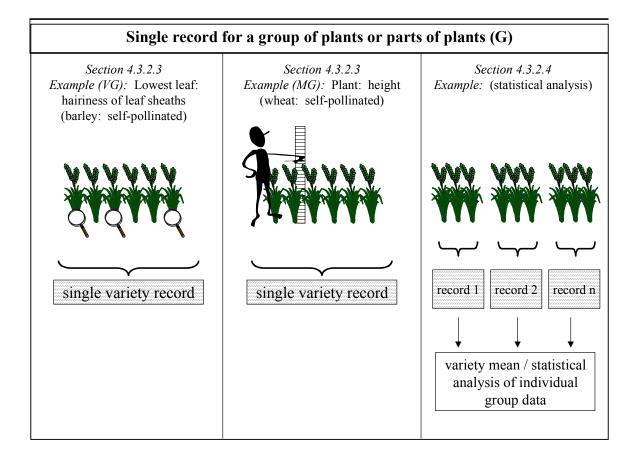
"Plant: natural height" in ryegrass (cross-pollinated): 60 plants are measured (MS). The value of each plant is used for calculation of the mean and for the variation within varieties in order to assess distinctness.

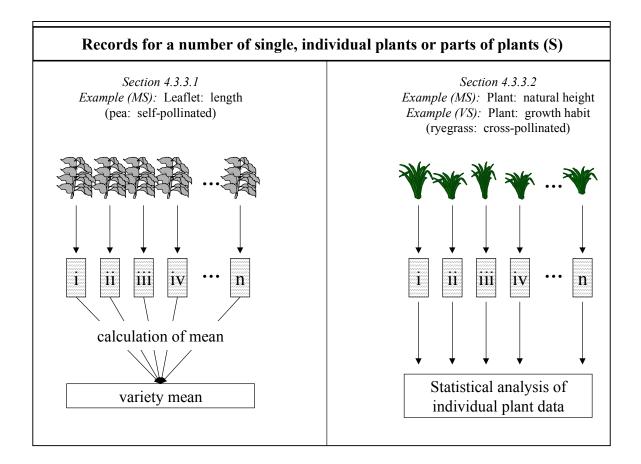
# Example (VS)

"Plant: growth habit" in ryegrass (cross pollinated): 60 plants are observed visually (VS). The value of each plant is used for calculation of the mean and for the variation within varieties in order to assess distinctness.

# 4.3.4 <u>Schematic summary</u>

The following diagram provides a schematic overview of a single record for a group of plants or parts of plants (G) and records for a number of single, individual plants or parts of plants (S):





## 4.4 **Recommendations in the UPOV Test Guidelines**

The indications used in UPOV Test Guidelines for the method of observation and the type of record are as follows:

Method of observation

- M: to be measured (an objective observation against a calibrated, linear scale e.g. using a ruler, weighing scales, colorimeter, dates, counts, etc.);
- V: to be observed visually (includes observations where the expert uses reference points (e.g. diagrams, example varieties, side-by-side comparison) or non-linear charts (e.g. color charts)

Type of record(s)

- G: single record for a variety, or a group of plants or parts of plants;
- S: records for a number of single, individual plants or parts of plants

### 4.5 Summary

The following table summarizes the common method of observation and type of record for the assessment of distinctness, although there may be exceptions:

	Type of expression of characteristic		
Method of propagation of the variety	QL	PQ	QN
Vegetatively propagated	VG	VG	VG/MG/MS
Self-pollinated	VG	VG	VG/MG/MS
Cross-pollinated	VG/(VS*)	VG/(VS*)	VS/VG/MS/MG
Hybrids	VG/(VS*)	VG/(VS*)	**

\* records of individual plants only necessary if segregation is to be recorded

\*\* to be considered according to the type of hybrid

# SECTION 5: ASSESSING DISTINCTNESS BASED ON THE GROWING TRIAL

## 5.1 Introduction

The process of determining which <u>varieties of common knowledge</u> to include in the variety collection and which varieties in the variety collection to include in the growing trial are considered in document TGP/4, "[Constitution and] Management of Variety Collections", *[cross ref.]* and Section 2 of this document *[cross ref.]*, respectively. This section considers the assessment of distinctness based on the growing trial and identifies certain techniques which can be used in the assessment of distinctness.

## 5.2 Approaches for assessing distinctness

## 5.2.1 Introduction

5.2.1.1 Approaches for assessment of distinctness based on the growing trial can be summarized as follows:

- (a) Side-by-side visual comparison in the growing trial;
- (b) Assessment by Notes / single variety records ("Notes"): the assessment of distinctness is based on the recorded state of expression of the variety for a characteristic [or a combination of characteristics]<sup>3</sup>;
- (c) Statistical analysis of growing trial data: the assessment of distinctness is based on a statistical analysis of the data obtained from the growing trial. This approach requires that, for a characteristic, there are a sufficient number of records for a variety.

5.2.1.2 The choice of approach or combination of approaches for the assessment of distinctness will depend on the method of observation and type of record (VG, MG, VS or MS), which is influenced by the features of propagation of the variety and the type of expression of the characteristic. The common situations are summarized by the table in Section 4.5 *[cross ref.]*. The purpose of the following sections is to consider how the assessment of distinctness is conducted for those different situations.

# 5.2.2 Side-by-side visual comparison ("Side-by-side")

5.2.2.1 Side-by-side visual comparison means that the assessment of distinctness is based on a direct visual comparison of varieties, side-by-side in the growing trial. This approach requires that the characteristics can be observed visually and indicates that the expression of the characteristic for a variety can be represented by a single record. It also requires that all similar varieties can be the subject of a direct side-by-side comparison in the growing trial. Such a requirement can be difficult to meet if the growing trial contains a large number of varieties and

<sup>&</sup>lt;sup>3</sup> The TC agreed that consideration should be given to adding "or a combination of characteristics" according to the outcome of discussions of Section 5.4.2.

there are limited possibilities for ensuring that all similar varieties are grouped together in the growing trial.

5.2.2.2 Side-by-side visual comparison is based on visual observation and, as explained in Section 4.2.1.2 *[cross ref.]*, because such observations are based on the expert's judgement, there is a particularly important requirement for training and experience.

5.2.2.3 In the case of vegetatively propagated and self-pollinated varieties, there is relatively little variation within varieties and visual assessment of distinctness is particularly suitable. However, where the range of variation within a variety is larger, because of the features of its propagation, and in particular for cross-pollinated and some types of hybrid varieties, determining distinctness on the basis of side-by-side visual comparison would require particular care.

5.2.2.4 In side-by-side comparisons of varieties, the following requirements for the assessment of distinctness should be considered:

# Qualitative characteristics

5.2.2.5 In general, side-by-side visual comparison is not necessary for qualitative characteristics, because varieties which have different states of expression for the same qualitative characteristic can be considered to be distinct (see Section 5.2.3 *[cross ref.]*).

## Pseudo-qualitative characteristics

5.2.2.6 The General Introduction explains with respect to pseudo-qualitative (PQ) characteristics that:

5.3.3.2.3 Pseudo-qualitative characteristics: "A different state in the Test Guidelines may not be sufficient to establish distinctness [...]. However, in certain circumstances, varieties described by the same state of expression may be clearly distinguishable."

5.2.2.7 The assessment of whether a pair of varieties are distinct, on the basis of a side-by-side visual comparison for a pseudo-qualitative characteristic, needs to take account of the variation within varieties.

### Quantitative characteristics

5.2.2.8 The General Introduction explains that, in the case of visually observed quantitative characteristics:

"5.5.2.2.2 A direct comparison between two similar varieties is always recommended, since direct pairwise comparisons are the most reliable. In each comparison, a difference between two varieties is acceptable as soon as it can be assessed visually and could be measured, although such measurement might be impractical or require unreasonable effort."

# 5.2.3 Assessment by Notes / Single variety records ("Notes")

5.2.3.1 Assessment by Notes / single variety records means that, for a particular characteristic, the assessment of distinctness is based on the recorded state of expression of a variety, obtained from the growing trial. The record may, for example, be in the form of: a Note corresponding

to a state of expression in the UPOV Test Guidelines (e.g. 1, 2, 3 etc.); a value (e.g. RHS Colour Chart reference number); a measurement (e.g. length (cm), weight (g), date (18-12-2005), count (3) etc.); an image etc.. The Notes / single variety records approach can be used for characteristics which are visually observed or measured, but requires that the expression of the characteristic for a variety can be represented by a single record for the purpose of the assessment of distinctness (VG, MG, mean of MS, mean of VS).

5.2.3.2 Where the requirements for distinctness assessment by Notes / single variety records are met it would usually also be possible to make a side-by-side visual comparison. However, in the case of assessment by Notes / single variety records, such proximity is not required, which is a particular advantage where the growing trial contains a large number of varieties and where there are limited possibilities for ensuring that all similar varieties are grouped together in the growing trial. On the other hand, because the varieties are not the subject of a side-by-side visual comparison, the difference required between varieties as a basis for distinctness is, with the exception of qualitative characteristics (see below), somewhat greater. The requirements for distinctness on the basis of Notes / single variety records are explained below:

Qualitative (QL) characteristics

5.2.3.3 The General Introduction provides guidance for qualitative characteristics as follows (see document TG/1/3):

5.3.3.2.1 Qualitative characteristics: "In qualitative characteristics, the difference between two varieties may be considered clear if one or more characteristics have expressions that fall into two different states in the Test Guidelines. Varieties should not be considered distinct for a qualitative characteristic if they have the same state of expression."

5.2.3.4 Thus, varieties which have different states of expression, i.e. different Notes, for the same qualitative characteristic can be considered to be distinct. Conversely, varieties which have the same Note for a qualitative characteristic should not be considered to be distinct for that characteristic.

Pseudo-qualitative (PQ) characteristics

5.2.3.5 The difference in Notes which may establish distinctness within a pseudo-qualitative characteristic is influenced by factors such as location, year and environmental variation within the trial. Also, as with quantitative characteristics, the range of the scale (number of Notes) varies. However, an important additional factor with pseudo-qualitative characteristics is that, whilst a part of the range is continuous, there is not an even distribution across the scale and the range varies in more than one dimension (e.g. shape: ovate (1), elliptic (2), circular (3), obovate (4): there is a variation in the length/width ratio and in the position of the widest point). This means that it is difficult to define a general rule on the difference in Notes to establish distinctness within a characteristic.

5.2.3.6 The difficulty in defining a general rule on the difference in Notes to establish distinctness within a pseudo-qualitative characteristic is reflected in the General Introduction (Chapter 5.3.3.2.3) which states that "A different state in the Test Guidelines may not be sufficient to establish distinctness [...]. However, in certain circumstances, varieties described by the same state of expression may be clearly distinguishable.". Nevertheless, it is the intention that the states and Notes in the UPOV Test Guidelines are useful for the assessment of distinctness. It is recalled that this section considers the assessment of distinctness based on the

information obtained from the growing trial and, therefore, refers to a situation where the states of expression and Notes are obtained for all varieties from the same growing trial in the same year. In such a situation, varieties with the same states of expression would not normally be considered to be clearly distinguishable.

5.2.3.7 The following examples illustrate why deciding on the number of Notes required to establish distinctness needs particular care:

Example 1:

Type of mottling: only diffuse (Note 1); diffuse and in patches (2); diffuse, in patches and linear bands (3); diffuse and in linear bands (4).

Example 2:

Shape: broad elliptic (Note 1), medium elliptic (2), narrow elliptic (3), ovate (4)

Example 3:

Color: green (Note 1), yellow green (2), green yellow (3), yellow (4), orange (5), red (6)

In the case of Examples 1 and 2, it is not appropriate to say that the "difference" between varieties with Notes 1 and 2 is less than between varieties with Notes 1 and 4, although they are respectively 1 and 3 Notes "different". In some cases, for example, the difference between Notes 2 and 3 may be greater than between Notes 1 and 4. However, Example 3 demonstrates that, in some parts of the range of some pseudo-qualitative characteristics, it might be possible to follow a similar approach to that used for quantitative characteristics e.g. varieties with states 2 and 3 (1 Note difference) have less difference than those with states 1 and 4 (3 Notes difference).

5.2.3.8 Color characteristics are, in general, pseudo-qualitative characteristics and are often recorded in the form of a color chart reference. Guidance on the use of color characteristics is provided in document TGP/14, Section 2.3: Glossary of Technical, Botanical and Statistical Terms Used in UPOV Documents: Botanical Terms: Color: color characteristics *[cross ref.]*.

Quantitative (QN) characteristics (vegetatively propagated and self-pollinated varieties)

5.2.3.9 The General Introduction states that:

"4.4.2 <u>Quantitative Characteristics</u>

"Quantitative characteristics" are those where the expression covers the full range of variation from one extreme to the other. The expression can be recorded on a one-dimensional, continuous or discrete, linear scale. The range of expression is divided into a number of states for the purpose of description (e.g. length of stem: very short (1), short (3), medium (5), long (7), very long (9)). The division seeks to provide, as far as is practical, an even distribution across the scale. The Test Guidelines do not specify the difference needed for distinctness. The states of expression should, however, be meaningful for DUS assessment."

5.2.3.10 Thus, it is the intention that the states and Notes in the UPOV Test Guidelines are useful for the assessment of distinctness. It is recalled that this section considers the assessment of distinctness based on the information obtained from the growing trial and, therefore, refers to

a situation where the states of expression and Notes are obtained for all varieties from the same growing trial in the same year. That situation is, in particular, reflected when the General Introduction states that:

"5.4.3 For quantitative characteristics, a difference of two Notes often represents a clear difference, but that is not an absolute standard for assessment of distinctness. Depending on factors, such as the testing place, the year, environmental variation or range of expression in the variety collection, a clear difference may be more or less than two Notes. Guidance is provided in document TGP/9, "Examining Distinctness."

5.2.3.11 A difference of two Notes is appropriate if the comparison between two varieties is performed at the level of Notes (VG, mean of VS). If the difference is only one Note, both varieties could be very close to the same border line (e.g. high end of Note 6 and low end of Note 7) and the difference might not be clear. When the comparison is performed at the level of measured values (MG, mean of MS) a difference smaller that two Notes might represent a clear difference.

5.2.3.12 Document TGP/7/1, "Development of Test Guidelines", (see Annex III: GN 20) explains that, in the case of quantitative characteristics, it is necessary to determine the appropriate range to describe the characteristic. In general, a standard "1-9" scale is used, but a "limited" range (Notes 1-5) and a "condensed" range (Notes 1-3) have also been accepted. Thus, when deciding on the number of Notes required to establish distinctness, the range of the scale needs to be taken into account.

5.2.3.13 In deciding, whether the "two-Note" standard is an appropriate basis for distinctness, it is also necessary to take into account the environmental variation within the growing trial.

5.2.3.14 It should also be recalled that a pair of varieties which are not distinct for a characteristic on the basis of Notes in the UPOV Test Guidelines may, for example in a subsequent growing trial, be the subject of a side-by-side visual comparison, where it may be possible to establish distinctness. However, in general, varieties with the same Note in the UPOV Test Guidelines would not normally be considered to be clearly distinguishable when based solely on the Notes.

5.2.3.15 In the case of single variety records other than Notes in the UPOV Test Guidelines, no general guidance can be made and the size of the difference required for distinctness will, as for Notes in the UPOV Test Guidelines, depend on factors such as the testing place, the year, environmental variation and the range of expression in the variety collection.

# 5.2.4 Statistical analysis of growing trial data

5.2.4.1 Where appropriate, the assessment of distinctness can be based on a statistical analysis of the data obtained from the growing trial. This approach requires that there is a sufficient number of records for a variety, e.g. records for a number of single, individual plants or parts of plants, whether obtained by measurement (MS) or by visual observation (VS). In most cases, when a single record is obtained by visual observation or measurement of a group of plants (VG / MG), this results in a single record per variety, in which case it is not possible or necessary to apply statistical methods for the assessment of distinctness. However, in some cases, e.g. where there are several repetitions or plots, or more than one growing trial, more than one record per variety may be obtained, in which case statistical methods can be applied,

although it is particularly relevant to check if the data obtained meets the assumptions required for a statistical procedure to be applied.

5.2.4.2 The assessment of distinctness by Notes / single variety records or side-by-side visual comparison is generally quicker and cheaper than the use of statistical analysis. However, as explained above, those approaches require that the expression of the characteristic for a variety can be represented by a single record. That requirement implies that there should be very little variation within varieties, which is usually met for all characteristics of vegetatively propagated varieties and self-pollinated varieties and for qualitative and pseudo-qualitative characteristics for cross-pollinated and hybrid varieties, except in cases of segregating characteristics. Thus, the most common use of statistical analysis of growing trial data is for quantitative characteristics of cross-pollinated and some hybrid varieties.

5.2.4.3 The General Introduction makes the following recommendations with regard to the use of statistical methods in the assessment of distinctness:

"5.5 Interpretation of Observations for the Assessment of Distinctness with the Application of Statistical Methods

5.5.1 <u>General</u>

5.5.1.1 For measured characteristics as well as for visually assessed<sup>[4]</sup> characteristics statistical methods can be applied. Appropriate methods have to be chosen for the interpretation of observations. The data structure and the type of scale from a statistical point of view (nominal, ordinal, interval or ratio) is decisive for the choice of appropriate methods. The data structure depends on the method of assessment<sup>[4]</sup> (visual assessment<sup>[4]</sup> or measurements, observation of plots or single plants) which is influenced by the type of characteristic, the features of propagation of the variety, the experimental design and other factors. DUS examiners should be aware of certain basic rules of statistics and especially the fact that their use is linked to mathematical assumptions and the use of experimental design practices, such as randomization. Therefore, those assumptions should be verified before applying statistical methods. Some statistical methods are quite robust, however, and can be used, with some caution, even if some assumptions are not fully met.

5.5.1.2 Document TGP/8, "Use of Statistical Procedures in DUS Testing", provides guidance on some appropriate statistical procedures for DUS assessment and includes keys for the choice of methods in relation to the data structure.

5.5.1.3 A combined characteristic should only be used for distinctness if the uniformity criteria for the combined characteristic itself, and not only its components, have been satisfied.

# 5.5.2 <u>Visually Assessed<sup>[4]</sup> Characteristics</u>

Non-parametric statistics may be used when visually assessed<sup>[4]</sup> characteristics have been recorded on a scale that does not fulfill the assumptions of the usual parametric statistics. The calculation of the mean value, for example, is only permitted if the Notes are taken on a graded scale which shows equal intervals throughout the scale. In the case of non-parametric procedures, the use of a scale that has been established on the basis of example varieties representative of the different states of the characteristics is

 $<sup>^4</sup>$  The term "assessed" would be more consistent with the use of the term "observed" in document TGP/9.

recommended. The same variety should then always receive about the same Note and thereby facilitate the interpretation of data. More details on the handling of visually assessed<sup>[4]</sup> characteristics are given in document TGP/9, "Examining Distinctness."

5.2.4.4 The suitability of statistical analysis and some requirements for their use are summarized below:

Qualitative (QL) characteristics

5.2.4.5 The General Introduction (Chapter 5.5.2.1) clarifies that "For visually assessed qualitative characteristics, different states of expression in direct comparisons are generally sufficient to assess distinctness. In most cases, therefore, no statistical methods are needed for the interpretation of the results."

Pseudo-qualitative (PQ) characteristics

5.2.4.6 The General Introduction (Chapter 5.5.2.3) explains that "The use of statistics for the assessment of pseudo-qualitative characteristics depends on the individual case, and no general recommendation can be made."

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Quantitative (QN) characteristics (vegetatively propagated and self-pollinated varieties)
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5.2.4.7 As indicated above, both side-by-side visual comparison and the assessment of distinctness on the basis of Notes / single variety records, are used for the assessment of distinctness for quantitative characteristics of vegetatively propagated and self-pollinated varieties. However, statistical methods can also be used for such situations where the necessary requirements are met.

5.2.4.8 The General Introduction clarifies the situation with regard to all visually observed, quantitative characteristics as follows:

- 5.5.2 <u>Visually Assessed<sup>[4]</sup> Characteristics</u>
- [...]
- 5.5.2.2 Quantitative Characteristics
- [...]

5.5.2.2.2 A direct comparison between two similar varieties is always recommended, since direct pairwise comparisons are the most reliable. In each comparison, a difference between two varieties is acceptable as soon as it can be assessed visually and could be measured, although such measurement might be impractical or require unreasonable effort.

5.5.2.2.3 The simplest case for establishing distinctness is when clear differences between varieties, in pair-wise comparisons, are of the same sign, provided these differences can be expected to recur in subsequent trials (e.g. variety A is consistently and sufficiently greater than B) and there are a sufficient number of comparisons. However, in most cases, establishing confidence that varieties are clearly distinguishable, is more complex. This is explained further in document TGP/9, "Examining Distinctness".

5.2.4.9 The situation referred to in the General Introduction that "However, in most cases, establishing confidence that varieties are clearly distinguishable, is more complex." does not, in general, apply to vegetatively propagated and self-pollinated varieties, but rather to the situation in cross-pollinated varieties and hybrid varieties. However, in most cases, quantitative characteristics are measured for such varieties and are handled as explained in Section 5.2.4.12 *[cross ref.]*.

5.2.4.10 The General Introduction clarifies the situation with regard to measured, quantitative characteristics for vegetatively propagated and self-pollinated varieties as follows:

### "5.5.3 <u>Measured Characteristics</u>

The following paragraphs provide guidance on the typical methods for examining distinctness according to the particular features of propagation of the variety:

[...]

### 5.5.3.1 Self-Pollinated and Vegetatively Propagated Varieties

UPOV has endorsed several statistical methods for the handling of measured quantitative characteristics. One method established for self-pollinated and vegetatively propagated varieties is that varieties can be considered clearly distinguishable if the difference between two varieties equals or exceeds the Least Significant Difference (LSD) at a specified probability level with the same sign over an appropriate period, even if they are described by the same state of expression. This is a relatively simple method but is considered appropriate for self-pollinated and vegetatively propagated varieties because the level of variation within such varieties is relatively low. Further details are provided in document TGP/9, "Examining Distinctness"."

5.2.4.11 Information on the Least Significant Difference (LSD) method is provided in document TGP/8 [cross ref.].

Quantitative (QN) characteristics (cross-pollinated varieties)

5.2.4.12 The General Introduction provides the following guidance with regard to the use of statistical methods for measured characteristics where individual plant data (MS) are available:

"5.5.3 <u>Measured Characteristics</u>

The following paragraphs provide guidance on the typical methods for examining distinctness according to the particular features of propagation of the variety:

[...]

5.5.3.2 Cross-Pollinated Varieties

5.5.3.2.1 COYD

UPOV has developed a method known as the Combined Over Years Distinctness (COYD) analysis, which takes into account variations between years. Its main use is for cross-pollinated, including synthetic, varieties but, if desired, it can also be used for self-pollinated and vegetatively propagated varieties in certain circumstances. This

method requires the size of the differences to be sufficiently consistent over the years and takes into account the variation between years. It is explained further in document TGP/9, "Examining Distinctness."

### 5.5.3.2.2 Refined COYD

A refinement to the COYD analysis, which is also provided, should be used to adjust the COYD analysis when environmental conditions cause a significant change in the spacing between variety means in a year, such as when a late spring causes the convergence of heading dates. It is supplemented by a further LSD method for cases where few varieties in the growing tests lead to less than about 20 degrees of freedom for the estimation of standard error.

### 5.5.3.2.3 Non-Parametric Procedures

Where COYD analysis cannot be used because the statistical criteria are not fulfilled, non-parametric procedures can be considered.

5.2.4.13 COYD can also be used where individual plant data are obtained by visual observation (VS), provided the data fulfill the necessary requirements (see document TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability" [cross ref.]). For example, visual observation of the attitude of the leaves of individual plants might be used for cross-pollinated varieties.

5.2.4.14 The following sections explain the COYD method and the  $2 \times 1\%$  criterion, which may be an appropriate method where the statistical criteria for COYD analysis are not fulfilled.

### Combined Over-Years Distinctness Criterion (COYD)

5.2.4.15 To assess distinctness for varieties on the basis of a quantitative characteristic it is possible to calculate a minimum distance between varieties such that, when the distance calculated between a pair of varieties is greater than this minimum distance, they may be considered as "distinct" in respect of that characteristic. Amongst the possible ways of establishing minimum distances is the method known as the Combined-Over-Years Distinctness (COYD).

- 5.2.4.16 The COYD method involves:
  - for each characteristic, taking the variety means from the two or three years of trials for candidates and established varieties and producing over-year means for the varieties;
  - calculating a least significant difference (LSD), based on variety-by-years variation, for comparing variety means;
  - if the over-years mean difference between two varieties is greater than or equal to the LSD then the varieties are said to be distinct in respect of that characteristic.
- 5.2.4.17 The main advantages of the COYD method are:
  - it combines information from several seasons into a single criterion (the "COYD criterion") in a simple and straightforward way;

- it ensures that judgements about distinctness will be reproducible in other seasons; in other words, the same genetic material should give similar results, within reasonable limits, from season to season;
- the risks of making a wrong judgement about distinctness are constant for all characteristics.

5.2.4.18 Details on the use of the Combined-Over-Years Distinctness (COYD) are provided in document TGP/8 [cross ref.].

# $2 \times 1\%$ criterion

5.2.4.19 For two varieties to be distinct using the  $2 \times 1\%$  criterion, the varieties need to be significantly different in the same direction at the 1% level in at least two out of three years in one or more measured characteristics. The tests in each year are based on Student's two-tailed t-test of the variety means with standard errors estimated using the plot residual mean square from the analysis of the variety × replicate plot means.

- 5.2.4.20 With respect to the  $2 \times 1\%$  criterion, compared to COYD, it is important to note that:
  - Information is lost because the criterion is based on the accumulated decisions arising from the results of t-tests made in each of the test years. Thus, a difference which is not quite significant at the 1% level contributes no more to the separation of a variety pair than a zero difference or a difference in the opposite direction. For example, three differences in the same direction, one of which is significant at the 1% level and the others at the 5% level would not be regarded as distinct.
  - Variety measurements on some characteristics are less consistent over years than on others. However, beyond requiring differences to be in the same direction in order to count towards distinctness, the 2×1% criterion takes no account of consistency in the size of the differences from year to year.

Other statistical methods

5.2.4.21 [There are a range of other statistical methods in use in agricultural research that can be used in the examination of distinctness. Those include ANOVA and multiple range tests. Providing the underlying assumptions are met, those other statistical methods are as acceptable as the other methods mentioned in this section.]

# 5.3 Summary of approaches for assessing distinctness based on the growing trial

The following table summarizes the common approaches to assessing distinctness based on the growing trial, taking into account the method of propagation, type of expression of the characteristic, method of observation and the type of record.

	Type of expression of characteristic		
Method of propagation of the variety	QL	PQ	QN
Vegetatively propagated,	Notes (VG)	Notes (VG)	Notes (VG/MG/MS)
self-pollinated		Side-by-side (VG)	Side-by-side (VG)
			Statistics (MG/MS)
Cross-pollinated	Notes (VG)	Notes (VG)	Statistics ([MG]/MS/VS)
	Statistics (VS*)	Side-by-side (VG)	Side-by-side (VG)
		Statistics (VS*)	Notes (VG/MG/MS)
Hybrids	Notes (VG)	Notes (VG)	**
	Statistics (VS*)	Side-by-side (VG)	
		Statistics (VS*)	

records of individual plants only necessary if segregation is to be recorded
to be considered according to the type of hybrid (see Section 4.3.3)

# 5.4 Techniques for assessing distinctness based on the growing trial

# 5.4.1 Parent Formula of Hybrid Varieties

In the case of variety collections which contain hybrid varieties, it may be appropriate to use the parent formula as a basis for assessing distinctness based on the growing trial. The use of the parental formula requires that the difference between parent lines is sufficient to ensure that the hybrid obtained from those parents is distinct. Details of the parent formula technique are provided in document TGP/8 [cross ref.].

# 5.4.2 Combined Phenotypic Distance<sup>5</sup>

# 5.4.2.1 Introduction

As explained in Section 2.6.1, when judging distinctness between varieties in the growing trial, DUS examiners may consider the overall level of difference. The following illustrations, presented on the basis of the model UPOV Variety Description (see document TGP/5 Section 6/1): Section 16 "Similar Varieties and Differences from These Varieties", are provided to indicate the use of a combination of characteristics for the determination of distinctness:

# Case 1: Field bean

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Leaflet: length	medium (Note 5)	medium to long (Note 6)
Pod: length	medium (Note 5)	medium to long (Note 6)

<sup>&</sup>lt;sup>5</sup> The TC agreed that consideration should be given to the use of a combination of characteristics including consideration of the stability of results. Examples of the use of a combination of characteristics to be provided by the European Community. In addition, to review whether Section 5.4.2 should be moved to Section 5.2, "Approaches for Assessing Distinctness".

## Case 2: Peas

	G G G G	G () ()
Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Stem: number of nodes up to and	medium (Note 5) [15.2]*	medium (Note 5) [16.3]
including first fertile node		
Pod: number of ovules	medium (Note 5) [8.7]**	medium to many (Note 6)
		[9.2]

# \* has a significantly lower first fertile node number (P=0.01) \*\* has a significantly lower number of ovules (P=0.02)

# Case 3: Rape Seed

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Time of flowering	medium to late (Note 6)	late (Note 7)
Flower: width of petals	broad (Note 7)	medium to broad (Note 6)

# Case 4: Rape Seed

Characteristic in which the similar variety is different	State of expression of similar variety	State of expression of candidate variety
Leaf: green color	dark (Note 7)	medium to dark (Note 6)
Leaf: dentation of margin	strong (Note 7)	medium to strong (Note 6)

# Case 5: Rape Seed

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Leaf: width	medium	medium to broad
Siliqua: length of beak	medium	medium to long
Seed cotyledon: length	medium	medium to long
Leaf: dentation of margin	weak to medium	medium

# Case 6: Rape Seed

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Flower: length of petals	medium to long	medium
Plant: total length including side	medium	short to medium
branches		
Seed cotyledon: length	medium to long	long

# Case 7: Red fescue

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Leaf: color	medium green (Note 5)	medium green to
		dark green (Note 6)
Stem: length	medium (Note 5)	short to medium (Note 4)

#### Case 8: Wheat

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Coleoptile: anthocyanin coloration	medium to strong (Note 6)	strong (Note 7)
Ear: glaucosity	medium (Note 5)	medium to strong (Note 6)

#### Case 9: Wheat

Characteristic in which the similar	State of expression of	State of expression of
variety is different	similar variety	candidate variety
Time of ear emergence	early to medium (Note 4)	medium (Note 5)
Ear: glaucosity	medium to strong (Note 6)	strong (Note 7)
Plant: length	medium to long (Note 6)	long (Note 6)

#### Case 10: Wheat

Characteristic in which the similar variety is different	State of expression of similar variety	State of expression of candidate variety
Plant: frequency of plants with	low	low to medium
recurved flag leaves		
Ear: glaucosity	strong	strong to very strong

## 5.4.2.2 Methods

5.4.2.2.1 GAIA

### Some reasons to sum and weight observed differences

5.4.2.2.1.1 When assessing distinctness, a DUS examiner first observes a variety characteristicby-characteristic. In the case of similar varieties, the DUS examiner also considers all observed differences as a whole. The GAIA software helps the DUS examiner to assess differences characteristic-by-characteristic and for all characteristics together.

5.4.2.2.1.2 A DUS examiner may see that two varieties are so distinct after the first growing cycle that it is not necessary to repeat the comparison. Those two varieties, which are "distinct plus" (see Section Section 2.6.2.1.2 [cross ref.]), are obviously distinct.

5.4.2.2.1.3 A DUS examiner may have a situation where two varieties receive a different note (e.g. Variety A is Note 3 for a given characteristic and Variety B is Note 4), but the two varieties are considered by the examiner to be similar. The difference could be due to the fact that the varieties were not grown very close each other (i.e. had different environmental conditions), or to variability of the observer when assessing the notes, etc.

5.4.2.2.1.4 Characteristics vary in their susceptibility to environmental conditions and the precision with which they are observed (i.e. visual observation/measurement). For characteristics which are susceptible to environmental conditions and which are not assessed very precisely, the examiner requires a large difference between Variety A and Variety B to be confident that the observed difference indicates distinctness.

5.4.2.2.1.5 For characteristics which are independent of environmental conditions and which are assessed precisely, the examiner can be confident in a smaller difference between Variety A and Variety B.

5.4.2.2.1.6 In the GAIA method, the examiner decides the appropriate weights for the observed differences for each observed characteristic. The software computes the sum of the weightings and indicates to the crop examiner which pairs of varieties are "distinct plus" and which are not. The examiner can then decide which of the varieties of common knowledge can be excluded from the subsequent growing cycle(s), because they are already obviously distinct from all candidate varieties.

# Computing GAIA phenotypic distance

5.4.2.2.1.7 The principle of the GAIA method is to compute a phenotypic distance between two varieties, being the total distance between a pair of varieties resulting from the addition of the weightings of all characteristics. Thus, the GAIA phenotypic distance is:

$$dist(i, j) = \sum_{k=1,nchar} W_k(i, j)$$

where:

dist(i, j) is the computed distance between variety i and variety j.

k is the  $k^{th}$  characteristic, from the *nchar* characteristics selected for computation.

 $W_k(i,j)$  is the weighting of characteristics k, which is a function of the difference observed between variety *i* and variety *j* for that characteristic k.

 $W_k(i,j) = f\left(OV_{ki} - OV_{kj}\right)$ 

where  $OV_{ki}$  is the observed value on characteristic k for variety i.

5.4.2.2.1.8 Details of the GAIA method are provided in document TGP/8 [cross ref.].

# 5.5 Illustrative scenarios

[Some illustrations of the process of assessing distinctness based on the growing trial as used by members of the Union are provided in document TGP/6, "Arrangements for DUS Testing".]

# **SECTION 6: SUPPLEMENTARY PROCEDURES**

### 6.1 Introduction

6.1.1 As explained in Section 1.4 *[cross ref.]*, supplementary procedures may be used to reinforce the examination of distinctness. The General Introduction explains that:

#### "5.3.1 Comparing Varieties

5.3.1.1 It is necessary to examine distinctness in relation to all varieties of common knowledge. However, a systematic individual comparison may not be required with all varieties of common knowledge. For example, where a candidate variety is sufficiently different, in the expression of its characteristics, to ensure that it is distinct from a particular group (or groups) of varieties of common knowledge, it would not be necessary for a systematic individual comparison with the varieties in that group (or those groups).

5.3.1.2 In addition, certain supplementary procedures may be developed to avoid the need for a systematic individual comparison. For example, the publication of variety descriptions, inviting comment from interested parties, or cooperation between members of the Union, in the form of an exchange of technical information, could be considered as supplementary procedures. However, such an approach would only be possible where the supplementary procedures, in conjunction with the other procedures, provide an effective examination of distinctness overall. Such procedures may also be appropriate for consideration of varieties of common knowledge, for which living plant material is known to exist (see Section 5.2.2) but where, for practical reasons, material is not readily accessible for examination. Any such procedures are set out in document TGP/9, "Examining Distinctness."

5.3.1.3 Further, where a candidate variety can be distinguished in a reliable way from varieties of common knowledge, by comparing documented descriptions, it is not necessary to include those varieties of common knowledge in a growing trial with the respective candidate variety. However, where there is no possibility of clearly distinguishing them from the candidate variety, the varieties should be compared with the candidate variety in a growing trial or other appropriate test. This emphasizes the importance of harmonization of variety descriptions in minimizing the workload of the DUS examiner."

6.1.2 In addition to the examples mentioned in the General Introduction, the making available of the list of varieties in the variety collection, the making available of lists against which candidate varieties have been examined and the use of panels of experts, are other examples of supplementary procedures. More information on some of these supplementary procedures is provided below. *[cross ref. document TGP/4, Section 2.3.2]* 

# 6.2 Publication of variety descriptions<sup>6</sup>

6.2.1 The General Introduction notes that the publication of variety descriptions inviting comment from interested parties may be considered as a supplementary procedure to avoid the need for a systematic individual comparison, for example to allow consideration of varieties of

<sup>&</sup>lt;sup>6</sup> See also document "Project to Consider the Publication of Variety Descriptions" (TWA/35/6, TWC/24/6, TWF/37/6, TWO/39/6, TWV/40/6)

common knowledge for which living plant material is known to exist but where, for practical reasons, material is not readily accessible for examination (see document TG/1/3, Section 5.3.1.2). In some such cases, varieties of common knowledge might be excluded from a growing trial based on the known expression of suitable grouping characteristics (see Section 2.3.2 *[cross ref.]*). However, in cases where there is insufficient information to exclude such varieties of common knowledge on the basis of grouping characteristics and they are not included in the growing trial, then publication of a detailed description of candidate varieties together with similar varieties included in the growing trial provide an opportunity for interested parties to comment. For example, where a candidate variety is found to be distinct from all varieties included in the growing trial, a supplementary step could be to publish a detailed variety description in the Plant Breeders' Rights Gazette together with a photograph showing the basis for distinctness from similar varieties included in the growing trial. The publication would then be followed by a time for comment on the distinctness of the candidate variety by interested parties, e.g. members of the public, breeders etc.. In the case of comments or objections, the authority would decide on the appropriate action, which could include a re-trial and a review of which varieties would need to be included in the growing trial. Where appropriate, the detailed description of the candidate variety and the basis for distinctness from similar varieties included in the new growing trial would then be re-published. An important consideration in the publication of variety descriptions is the influence of the environment and the source of the description. In particular, a review of the distinctness of varieties by descriptions produced in different environments or provided by different sources has many difficulties. In that respect, publication of variety descriptions with reference to similar varieties of common knowledge included in the same growing trial can aid review by other parties.

6.2.2 An example of the use of the publication of variety descriptions can be found in document TGP/6, Section 2.2, which explains the procedure used in Australia.

### 6.3 Cooperation between members of the Union

The General Introduction states that cooperation between members of the Union in the form of exchange of technical information could also be used as a supplementary procedure (see document TG/1/3, Section 5.3.1.2).

### 6.4 Use of randomized "blind" testing

6.4.1 After, or during, the examination, some doubts may exist over the distinctness of a variety on the basis of the growing trial. In such cases, the following situations are possible:

(a) with no differences observed, the application is rejected;

(b) with no conclusive difference observed and a claim from the breeder/applicant, the examining authority may decide to arrange additional tests.

6.4.2 In the case of visually observed characteristics, one possible arrangement for an additional test is "blind" testing.

6.4.3 The aim of "blind" testing is to assess distinctness between a pair of varieties avoiding any pre-judgement in the observation by making the samples in the trial anonymous (the expert is "blind" in respect to the identity of the variety in each plot). This kind of test plays a clarifying role when the differences between the candidate and (a) similar variety(ies) are not clearly definable. In such a case, another test during or after the examination of distinctness may provide evidence for a definitive decision by the authority.

6.4.4 The following are some examples of "blind" testing:

*Randomized variety plots*: duplicates samples of the same variety receive individual codes and are randomly distributed in the trial.

*Plots containing a mixture of varieties*: plots with a mixture of material from the varieties under examination are included in the trial. [This can be useful for seed-propagated varieties].

*Parts of plants of varieties*: randomized parts of plants from the varieties under examination (e.g. leaves or fruit).

6.4.5 Breeders/applicants may be part of the "blind" testing process. They may also be invited to visit the "blind" test and be requested to try to identify the plots of their variety.

6.4.6 At the end of the "blind" testing. a variety may be declared as distinct:

(a) if the expert and, where appropriate, the breeder/applicant always identify the variety; and

- (b) the difference can be considered as a clear difference for that characteristic.
- 6.4.7 In all cases, it is the authority which decides on distinctness.

# 6.5 The advice of [panels of] plant experts<sup>7</sup>

There may be cases where the assistance of a recognized plant expert or group of plant experts with extensive knowledge of varieties of common knowledge in a given genus, species or type of variety may be appropriate. In these cases, it is recommended that clear rules on the tasks and responsibilities of the plant expert or group of plant experts involved, as well as on the management of the information submitted for the purposes of examination, be established in order to maintain the transparency of the system.

[End of document]

<sup>&</sup>lt;sup>7</sup> The TC agreed to consider changing the title to "The advice of panels of experts".