

TGP/13 Draft 2 ORIGINAL: English DATE: June 25, 2004

INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS GENEVA

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

DOCUMENT TGP/13

"GUIDANCE FOR NEW TYPES AND SPECIES"

General Guidance for Testing of New Species, Interspecific/Intergeneric Hybrids and New Types of Varieties

Document prepared by experts from the Community Plant Variety Office (CPVO) on the basis of a proposal prepared by experts from New Zealand and the United Kingdom

to be considered by the

Technical Working Party for Ornamental Plants and Forest Trees at its thirty-seventh session, to be held in Hanover, Germany, from July 12 to 16, 2004

and the

Technical Working Party for Fruit Crops at its thirty-fifth session, to be held in Marquardt (Potsdam), Germany, from July 19 to 23, 2004

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1. <u>Introduction</u>

1.1 The 1991 Act of the UPOV Convention requires that its provisions are, within a specified time, applied to all plant genera and species. Testing authorities, especially in the ornamental crops, are faced with an increasing number of applications for varieties which represent the first application for protection within a plant genus or species.

1.2 Sophisticated genetic techniques, together with new intercrossing and multiplication methods are continually becoming available to breeders, often transposed from more advanced breeding or multiplication systems used in other species. As a result, novel interspecific, or intergeneric, hybrids and new types of varieties are increasingly being introduced, where no experience exists in DUS testing.

1.3 This document seeks to provide general guidance for the examination of DUS (the "DUS Test") of varieties with the background described above. It considers new species, novel interspecific or intergeneric hybrids and new types of varieties, each being reviewed with respect to the points that need particular attention for organizing the DUS test and providing a variety description.

2. <u>New species</u>

2.1 Introduction to new species: what is considered as new species?

A new species is considered to be a species with which the organisation [authority?] carrying out the DUS test has no or very little previous experience.

2.2 Information provided in the technical questionnaire

2.2.1 The Technical Questionnaire (see TGP/7/1 Annex I: TG template) is a very valuable first source of information and it is of great importance that it is completed accurately and in full: in completion of the Technical Questionnaire, the breeder has the first important role to play, providing information on the origin, breeding background, maintenance and reproduction of the variety and a preliminary description of the most important characteristics. Information is often provided on the most similar varieties to the candidate variety which can be useful for confirming the descriptive information given for grouping purposes and can also be used to direct the authority towards the most appropriate varieties for inclusion in the DUS test. It is advisable to verify the information supplied by the breeder concerning the candidate variety and the most similar varieties. This can be done in a preliminary examination of test plant material, or when a growing trial has been completed and the essential descriptive characteristics of the new candidate variety have been determined.

2.2.2 For a candidate variety which is declared to have been "discovered and developed" from the wild species, details about this discovery and development should be provided. [While checking this element, the examiner should have in mind the Rio Convention on Biodiversity.] There should be a clear difference between the candidate and the wild species. This difference between the characteristics of the candidate and the wild botanical species should be clearly specified by the applicant.

2.2.3 Special attention should also been given that the applicant clearly specifies the growing conditions of the variety.

2.3 Preparation of the technical examination/Test Guidelines

2.3.1 Thus, an authority may receive an application for a variety of a species of which they have no previous experience. In this situation, the first step should be to determine whether or not UPOV Test Guidelines ("Test Guidelines") exist. If they do not, then a search should be made in TGP/5.10 or the "GENIE Database" to identify if other members of the Union have developed national guidelines and/or have experience of DUS testing in the species concerned. If this is unsuccessful, it will be necessary to establish an appropriate protocol for the DUS test. Test Guidelines of a similar species and close cooperation with the breeder can help in the development of an appropriate protocol.

2.3.2 For the preparation of new guidelines, guidance on the key issues to be addressed is provided in document TGP/7, "Development of Test Guidelines" and it could also be helpful to discuss testing with neighbouring countries. It should be noted that for the first applications of a new species, it might not be possible and necessary to develop guidelines with all details foreseen in document TGP/7/1. The DUS testing for a new species and for new variety types does not differ in principle from the testing of any variety. The difference is in the level of testing experience and the details known about that new species.

2.3.3 Before testing begins, it is important to learn as much about the new species as possible and to collect relevant information. Possible sources of information include botanical literature, trade and industry publications, trade catalogues, information available on the internet, national research institutes, amateur plant collectors, botanical gardens and the breeder. Knowledge of the growing conditions is certainly important information and sometimes, taking into account particular growing conditions, it might be more efficient to organize the testing at the premises of the breeder. Information concerning such an approach can be found in TGP/6 "Arrangements for DUS Testing".

2.3.4 Once an authority has acquired experience in testing a particular species, it should communicate this to the TC for updating of the document 'list of species in which practical knowledge has been acquired or for which national test guidelines have been established (document TGP/5.10).

2.4 Testing distinctness

2.4.1 It is necessary to examine if a new candidate variety is distinct from all other varieties whose existence is a matter of common knowledge (varieties of common knowledge). The selection of similar varieties for inclusion in the growing trial for comparison with a candidate variety is the usual first step in the DUS examination. In this context, it is important to ensure that the terms "variety" and "common knowledge" are clearly understood. These terms are clarified in document TG/1/3 (See Section 5.2 "Varieties of Common Knowledge"). Document TGP/3, "Varieties of Common Knowledge" may also be used to assist in understanding the meaning of the term common knowledge.

2.4.2 DUS testing of varieties of new species may differ from other variety testing in the determination of varieties of common knowledge as there may, or may not, be other similar varieties in existence.

2.4.3 It can be useful to examine the breeding origin of the candidate variety to increase information about the new species. The origin of a variety may be able to indicate the probability of varieties of common knowledge. It would therefore be useful to contact the authorities in the country of breeding origin to obtain information. The following are four scenarios:

- (i) Clonal propagation from a seedling or mutation, originating from a population in the wild, of a species not thought to be in cultivation. It is probable that there are no varieties of common knowledge. The candidate should nevertheless be clearly distinguishable from the wild species from which it is derived. This material might come from another part of the world, and might be difficult to obtain; however, the breeder might still possess material and might be requested to provide this material together with the candidate variety. Collections of wild species in botanical gardens could also be of help for the assessment of distinctness. The authority should prepare a description and assess uniformity and stability according to the principles for testing the first varieties of a new genus or species.
- (ii) Clonal propagation from a seedling in a population of a species which is in horticultural production. The species may be in commercial cultivation but possibly only exists as a heterogeneous group of seedlings. It is less probable that there are varieties of common knowledge because the species is known to exist only as a heterogeneous group of seedlings. The heterogeneous seedlings do not conform to a single description, are not uniform and cannot be considered as varieties. As a result this group of seedlings in commerce may not be relevant for the consideration of distinctness. If the seedlings have a level of uniformity in at least the main characteristics and have conformity to a single description, such that they satisfy the definition of a variety as set out in in the 1991 Act of the UPOV Convention, then these seedlings should be treated as varieties of common knowledge and, therefore, as relevant for DUS testing.
- (iii) Clonal selection for a desired attribute or form, possibly originating from mutation, from a species in horticulture. Other clonal forms may have been selected from within the species and exist in commerce as named or un-named varieties. The candidate variety should be compared with all other varieties existing in commerce and with any other appropriate varieties. The new variety should be distinct from such other varieties and should be uniform, not only for a characteristic which establishes distinctness, but for all relevant characteristics.
- (iv) A seed-propagated variety selected from within a population. Other seed-propagated varieties, whether named or un-named, could exist in commerce. The new seed-propagated variety should be compared with the other seed-propagated varieties, whether named or unnamed. The new variety must be distinct from the other seed-propagated varieties and uniformity assessed according to the reproductive system of the variety and the applicable UPOV uniformity method.

2.4.4 Each of the above scenarios will give some information about the species and provides an indication as to whether or not varieties of common knowledge might exist. For further information on the determination of distinctness in various crop types, reference

should be made to document TGP/9, "Examining Distinctness" and to individual Test Guidelines.

2.5 Testing Uniformity

2.5.1 Article 8 of the UPOV Convention states that a "variety shall be deemed to be uniform if, subject to the variation that may be expected from the particular features of its propagation, it is sufficiently uniform in its relevant characteristics" and so introduces the concept of an acceptable level of uniformity which is dependent upon the genetic make-up and system of propagation under consideration. General principles of testing Uniformity apply.

2.5.2 The General Introduction (document TG/1/3: Section 6.4) states that "where all the plants of a variety are very similar, and in particular for vegetatively propagated and self-pollinated varieties, it is possible to assess uniformity by the number of obviously different plants-"off-types"-that occur. However, where the range of variation within a variety is larger, because of the features of its propagation, and in particular for cross-pollinated, including synthetic, varieties, the plants are not all very similar and it is not possible to visualize which plants should be considered as atypical or "off-types". In this case the unformity can be assessed by considering the overall range of variation, observed across all the individual plants, to determine whether it is similar to comparable varieties".

2.5.3 The question of how uniformity can be assessed arises if the new variety is the first in a new species and, as a consequence, there are no other comparable varieties. In these circumstances, uniformity will need to be determined by another approach. The level of uniformity required of a candidate variety can sometimes be based upon past experience of what is known to be attainable by the breeding method used and has been shown to have been successful in the maintenance and multiplication of varieties of a similar type in other related species. Therefore, the following approaches may assist:

(a) Consideration of the levels of uniformity achieved in closely related species;

(b) Consideration of the level of natural uniformity in the family;

(c) Consulting the breeder in order to understand the breeding method;

(d) Considering the possibility of further improving the variety's uniformity, for example whether the breeder could easily remove more off-types;

(e) Based on available knowledge, considering how uniform a cross-pollinated variety from the new species could be.

2.5.4 For further information on the setting of uniformity standards in various crop types, reference should be made to document TGP/10, "Examining Uniformity" and to individual crop Test Guidelines.

2.6 Variety description

2.6.1 Chapter 4.3 of the General Introduction states that "to enable varieties to be tested and a variety description to be established, the range of expression of each characteristic in the Test Guidelines is divided into a number of states for the purpose of description, and the wording of each state is attributed a numerical note".

2.6.2 In the case of the examination of a candidate variety of a new species, varieties of common knowledge might not exist, or might exist but cannot be obtained, and it might not be possible to develop a range of expression for each characteristic. For this reason a description might be developed according to the Linnaean model. Descriptions of the wild species in a botanical dictionary might serve as guidance to prepare such a description. Only after applications for several varieties have been made for the same species, might it become possible to develop a list of characteristics with states of expression for that species.

3. Interspecific/Intergeneric hybrids

3.1 Introduction to Interspecific/Intergeneric Hybrids

An interspecific, or intergeneric, hybrid can be considered as a special form of a new species. Although there is a large overlap with the items which are handled in the new species paragraphs of chapter 2, this section considers the specific problems related to these hybrids.

3.2 Information provided by the Technical Questionnaire

Together with the species name of the parent varieties, the variety description and the species name of the most similar varieties are very useful information.

3.3 Preparation of the DUS examination/Test Guidelines

3.3.1 The parents may be well known but the resulting hybrid is new. An example is plumcots (*Prunus salicina* x *P. armeniaca*). In this case, Test Guidelines exist for both parent species and could be used separately or in combination. The first step, in determining whether existing Test Guidelines would be appropriate, is to assess the variety characteristics and determine if the variety is more like one parent than the other. If one parent has strongly influenced the variety characteristics, then the Test Guidelines for that parent might be used. If a small number of characteristics or states do not correspond to the Test Guidelines selected, then the Test Guidelines for the other parent are also available as a possible source of further appropriate characteristics to enable a full variety description. It may become necessary to prepare a new national guideline for the hybrid, and potentially new Test Guidelines if the interspecific or intergeneric hybrid varieties are to be grown in the territories of other members of the Union.

3.3.2 Some UPOV Test Guidelines are designed to cover the testing of all varieties in a genus. When Test Guidelines are prepared for a genus, they are usually most suited to a few species within the genus, where there is testing experience. This should not limit their use and genus based Test Guidelines are suitable for the testing of interspecific varieties.

3.3.3 Should no Test Guidelines exist for the parent species, it becomes necessary to prepare a new guideline and testing method for the hybrid using the principles in this document and in document TGP/7, "Development of Test Guidelines."

3.4 Testing Distinctness

The general principles should apply.

3.5 Testing Uniformity

The general principles should apply.

3.6 Variety description

As mentioned in paragraph 24, the parent guidelines should be used if possible.

4. <u>New types of varieties</u>

4.1 Introduction to new types of varieties

Techniques such as cell selection, protoplast fusion, *in vitro* fertilisation, embryo rescue and widecrossing, together with new intercrossing and multiplication methods are continually becoming available to breeders, often transposed from more advanced breeding or multiplication systems used in other species. As a result, novel variety types such as: complex hybrid variety systems with varying levels of plant-to-plant uniformity; highly uniform micropropagated clonal varieties derived from within sexually reproduced and more variable populations; and more variable seed-propagated varieties produced in hitherto highly uniform vegetatively propagated species, are increasingly being introduced. We now have the situation in several crop species where it is possible to have several different variety types being produced at the one time, each requiring a different standard of uniformity.

- 4.2 Information given in the Technical Questionnaire
- 4.3 Preparation of the technical examination
- 4.4 Testing distinctness
- 4.5 Testing uniformity
- 4.6 Variety description
- 5. <u>Literature</u>

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[Notes: Paragraphs of the original TGP 13, which have been omitted from this document are: 5 (partly), 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 21, 22, 23, 25, 26. Annex provides the text of those omitted paragraphs.]

[the Annex follows]

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ANNEX

OMITTED PARAGRAPHS FROM DOCUMENT TGP/13 Draft 1

GENERAL GUIDANCE FOR NEW SPECIES AND TYPES OF VARIETIES

1. INTRODUCTION

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4. For the granting of the breeder's right, the method of propagation of any variety has implications for the technical examination of its compliance with all the DUS criteria, but in particular for the consideration of uniformity. It is important to examine the naturally occurring reproductive system of the new candidate variety as this will influence the way in which it will have to be examined to determine whether it is distinct, sufficiently uniform and stable. It is therefore useful to briefly outline the four main types of plant reproductive systems and variety types which result from the different natural methods of reproduction adopted by plants, together with artificial hybridization systems, and to suggest the appropriate levels of uniformity which might reasonably be expected.

II. REPRODUCTIVE SYSTEMS AND VARIETY TYPES

Vegetatively Propagated Varieties - Clones

5. In vegetatively propagated varieties, the variety is produced by cloning a single plant. Once the initial cross or selection has been completed and the desirable phenotype identified, this can then be propagated vegetatively without further substantive genetic change. Vegetative propagation has the advantage for the plant breeding industry that plants propagated in this way, without recourse to the sexual cycle, are normally more uniform and more phenotypically stable than those reproduced sexually. Variety maintenance and multiplication is less complex than other forms of propagation except in certain species where deviations from description can easily result from natural somatic mutation. Indeed, in some crops many new protected varieties continue to arise in this way from spontaneous mutation. In some micropropagation systems there have also been problems with unacceptable levels of somaclonal variation and regeneration protocols have had to be carefully developed to try to avoid this.

6. A vegetatively propagated variety is derived from a single plant. Member plants of that variety should be phenotypically similar and varieties can be expected to exhibit reduced plant-to-plant variability, showing higher levels of uniformity and stability for all characteristics than other methods of propagation.

Self-Fertilized Varieties - Lines

7. For the plant breeding industry the main advantage of self-fertilization is that it confers a much greater degree of phenotypic uniformity in varieties than does cross-fertilization. Varieties of self-fertilized species tend towards homozygosity, at least in their main descriptor characteristics, which are positively selected for uniformity by the plant breeder. They can, as a result, more easily be maintained true to their description across repeated cycles of sexual

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multiplication as the male and female gametes are of increasingly similar (if not identical) genotypic makeup. Variety multiplication is therefore relatively straightforward provided initial selection and maintenance have been correctly carried out.

8. Self-fertilized varieties tend towards homozygosity and member plants will be phenotypically similar and, certainly similar for their main descriptor characteristics. Varieties should show low plant-to-plant variability and may be expected to have a high level of uniformity and stability.

Cross-fertilized Varieties - Populations

9. From the point of view of the natural environment, the cross-fertilized plant species are those which most easily can employ the variability brought about by sexual reproduction to adapt to environmental or cultural requirements. However, this plasticity and readiness to respond to the pressures of natural selection reflects a lack of uniformity and stability and means that multiplication and maintenance of cross-fertilized varieties true to their description is much more difficult.

10. Varieties are made up of populations of heterozygous and genetically different plants and are kept within certain descriptive ranges either by continuous re-selection or, more usually, by production of what is termed a synthetic variety. In the case of a synthetic variety, for example in forage grasses, a number of similar and desirable plants are selected and placed in a totally intercrossing environment (polycross). The aim is to produce a population of plants in the F1, which will be in genetic equilibrium and will be able to maintain the essential identity of the variety across a limited number of further generations in an open-pollinated system with a degree of isolation for the seed crop.

11. Cross-fertilized varieties are derived from populations of selected plants. In all subsequent generations the member plants, all of which are heterozygous, will show a range of expression for most characteristics that will be determined by the make-up of the initial founder plants. Varieties will therefore inherently show plant-to-plant variability and the permitted limits of this variation for any variety must be defined by statistical means in comparison with similar types or species. In maintenance and multiplication, such varieties are kept stable and true to description only with considerable care.

Hybrid Varieties

12. A hybrid variety results from the inducement of exclusive cross-fertilization between its component parental lines. This can be brought about by either manual (hand pollination, detasseling), chemical (gametocide) or genetic (self-incompatibility, male sterility) means. These may be used in various controlled crossing systems such as a two or three-way cross, inbred hybrid, top cross or a hybrid between two cross-fertilized varieties. The objective of the breeder is to utilize the heterosis or hybrid vigor which is expressed to most effect in the F1 generation to produce a favorable advantage in yield or some other desirable characteristic for the resultant progeny or hybrid variety. This heterosis is particularly well expressed in the case of crosses between inbred lines but can also be shown to advantage within other systems.

13. The hybrid variety produced from a single cross between two inbred lines is normally highly uniform because, in the F1 generation, all progeny are of the same predetermined,

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though heterozygous, genotype. The fact that heterozygosity is evident is of no consequence as there is normally no further multiplication beyond the F1 and the variety is maintained by repeatedly returning to a controlled cross of the parental lines. With heterozygous parents, e.g. multiple cross hybrids or hybrids between cross-fertilized varieties, the situation with respect to uniformity is as for cross-fertilized varieties except that, once again, as no further multiplication of the hybrid itself is intended, there is no need to consider any influence of genetic equilibrium upon stability across future generations.

14. Hybrid varieties are produced from a controlled cross between selected parents with the progeny representing the hybrid variety. While heterozygosity is present, phenotypic similarity and plant-to-plant uniformity is very high in the case of single-cross inbreds. In other hybrids, the level of uniformity has to be considered according to the uniformity of the parents. The need to achieve a satisfactory level of uniformity for distinctness purposes must be given careful consideration where a range of hybrid systems exist within the one crop. Acceptance of a very low level of uniformity in complex hybrids could greatly inhibit the scope for distinctness between new candidate varieties. For this reason, characterization of parent lines has been used in some situations in determining the distinctness of hybrid varieties. Where this approach is not used it will be necessary to give careful consideration to preserving the scope for development of new varieties. While uniformity remains a prerequisite of the hybrid variety it is not considered in respect of stability as multiplication is based upon maintenance of the parental lines and repeated enactment of the controlled crossing scheme to produce the hybrid.

IV. TESTING UNIFORMITY

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21. While there is a general acceptance that the first variety of a new type must, to some degree, be allowed to set the standard for uniformity, it is important that this is set at an appropriate level. If the standard is set too high then an unreasonably high target might be set for the future which could hinder breeding development. The higher the degree of uniformity for a characteristic or variety, the less scope there is for new variety selection. However, if the uniformity standard is set too high it may become almost unattainable thereby preventing the development of new varieties. The examination system must therefore always strive to find the optimal balance between these two goals.

22. Alternatively, if too low a standard is accepted this could also prevent further breeding progress as the first variety would have encompassed too large a proportion of the possible variability available within the type or species.

23. The main determinant of the plant breeding and multiplication systems utilized for variety production within any particular plant species is the natural mode of reproduction of that species and the ability of the plant breeder to control and modify this. At the early stages of development of the breeding history of most plant species the natural reproductive systems in place have therefore normally dictated what is considered to be sufficient uniformity. Therefore, in considering the standards to be required for uniformity of a new variety type in an already exploited crop or in a new plant species, it is important that full consideration is given to the influence of the inherent natural reproductive system of the crop species. It is important that efforts are made to understand the natural genetics and reproductive

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methodology of the type and species to which the new variety belongs to be able to have a realistic expectation of the level of uniformity that might reasonably be expected. This must be taken together with the modifying effects of the breeding and maintenance strategies employed by the breeder which must be studied to be sure that these will result in a true variety entity which will be able to be accurately described and either maintained or repeatedly produced true to its description over a period of years. Guidance can often be obtained from parallel breeding developments in other similar or related crop species.

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25. With vegetatively propagated, self-fertilized and single cross in-bred hybrid variety types, plant-to-plant uniformity of the phenotype can, within reason, be expected for most characteristics with some additional allowances for the occurrence of inbreds in the hybrids. The assessment of uniformity is normally carried out by the use of population standards. Firstly, the essential state of the variety should be defined either as a whole or across specific descriptor characteristics. Once the variety is clearly defined and described, any individual plants that do not conform to the variety description are identified as off-types. Depending upon the population standard appropriate for the species under examination, the sample size used for the examination of uniformity is determined together with the maximum allowable number of off-types within the sample to allow the variety to meet the uniformity criterion.

26. For the continuously expressed and measured characteristics normally used in cross-fertilized varieties or, when relevant, multiple cross hybrid populations, it is not appropriate to use population standards. Thus, it is not a fixed level of uniformity that is required of such types but a relative uniformity in comparison with other similar varieties. The spread of the normal distribution for each of the measured characteristics, as represented statistically by the standard deviation (SD), is normally employed to determine compliance with the uniformity criterion. For the candidate variety, the distribution of the individual plant data for each characteristic is examined alongside similar data for known and comparable reference varieties of similar type and must fall within tolerances based upon predetermined statistical probability levels.

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