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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

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TECHNICAL WORKING PARTY FOR VEGETABLES

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REVISION OF DOCUMENT TGP/8:
PART II: TECHNIQUES USED IN DUS EXAMINATION
New Section 10 – Minimum Number of Comparable Varieties for the Relative Variance Method

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BACKGROUND

- 1. The Technical Working Party on Automation and Computer Programs (TWC), at its twenty-ninth session held in Geneva, from June 7 to June 10, 2011, received a presentation on "Uniformity Assessment on the Basis of the Relative Variance Method" from Mr. Nik Hulse (Australia). The TWC conditionally agreed with the proposal made by Australia. Doubts were expressed regarding some assumptions of the method and it was noted that further investigation would be made by Australia with respect to those assumptions and the F value used in the calculations (see document TWC/29/31 "Report" paragraphs 37 and 38).
- 2. The Technical Committee (TC), at its forty-eighth session, held in Geneva from March 26 to 28, 2012, considered the revision of document TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability" on the basis of document TC/48/19 Rev.. The TC noted that new drafts of relevant sections would need to be prepared by April 26, 2012, in order that the sections could be included in the draft to be considered by the TWPs at their sessions in 2012 (see document TC/48/22 "Report on Conclusions" paragraph 49).
- 3. The TC, at its forty-eighth session, noted the comments of the TWC with regard to certain of the assumptions of the method and noted that further investigations would be done by Australia with respect to these assumptions and the F value used in the calculations (see document TC/48/22 "Report on Conclusions" paragraph 65).
- 4. The new draft on "Uniformity Assessment on the Basis of the Relative Variance Method" for consideration by the TWPs at their sessions is provided in the Annex to this document.

[Annex follows]

ANNEX

TGP/8/1: PART II: 10: UNIFORMITY ASSESSMENT ON THE BASIS OF THE RELATIVE VARIANCE METHOD

10. UNIFORMITY ASSESSMENT ON THE BASIS OF THE RELATIVE VARIANCE METHOD

10.1 Use of the relative variance method

- 10.1.1 The relative variance for a particular characteristic refers to the variance of the candidate divided by the average of the variance of the comparable varieties (i.e. Relative variance = variance of the candidate/average variance of the comparable varieties). The data should be normally distributed. The relative variance method may be applied to any measured characteristic that is a continuous variable. Comparable varieties are varieties of the same type within the same or a closely related species that have been previously examined and considered to be sufficiently uniform (see document TGP/10, Section 5.2 "Determining acceptable level of variation").
- 10.1.2 Chapter 5 of the document "Examining Uniformity", TGP/10/1 explains that where it is not possible to visualize off-types then a comparison is made to comparable varieties as follows:
 - "5.1 The General Introduction, Chapter 6.4, explains that, in cases where there is a high level1 of variation in the expressions of characteristics for the plants within a variety, it is not possible to visualize which plants should be considered as off-types and the off-type approach for the assessment of uniformity is not appropriate. It clarifies that in such cases, uniformity can be assessed by considering the overall level1 of variation, observed across all the individual plants, to determine whether it is similar to comparable varieties. In this approach, relative tolerance limits for the level1 of variation are set by comparison with comparable varieties, or types, already known ("standard deviations approach"). The standard deviations approach means that a candidate variety should not be significantly less uniform than the comparable varieties."
- 10.1.3 In many situations relatively large scale trials are conducted with a large number of comparable varieties. In these cases an approach such as COYU may be considered appropriate. However, in trials where the number of available comparable varieties is typically low the Relative Variance method may be used.
- 10.1.4 For example, Chapter 7 of TGP/8/1 describes the Match approach and the varieties included in the trial as follows:
 - "7.2.3 The Match method typically involves relatively small scale trials where the number of varieties in the trials is limited to the candidate varieties and the most similar varieties of common knowledge."
- 10.1.5 Comparable varieties can be considered to be those that are similar in their relevant characteristics to the candidate variety and are sufficiently uniform. Consequently, the number of comparable varieties used for examining uniformity is determined by the number of similar varieties included in the trial for the purpose of examining distinctness.
- 10.1.6 Other varieties may be included in the trial for reasons other than that they are the most similar varieties to the candidate. For example, check or example varieties may be included to verify the expression of particular characteristics. The DUS examiner can exclude these as comparable varieties in the examination of uniformity.

10.2 Threshold limits for Relative Variance Method

- 10.2.1 In cross-pollinated varieties, a common recommendation in the UPOV Test Guidelines is to take 60 measurements per measured characteristic per variety. In essence, the variance ratio equates to the F statistic, and the tabulated value of F at P = 0.01 under df1 =60 (degrees of freedom of candidate) and df2 = ∞ (degrees of freedom of comparable variety(ies)) is 1.47. df2 = ∞ is chosen as a conservative estimate, as it is assumed that comparable varieties accurately represent the infinite number of possible comparable varieties for the species as a whole. Therefore, 1.47 is the threshold limit for cross-pollinated species with many comparable varieties.
- 10.2.2 However, when there are limited number of comparable varieties available for a species, it is not practical to use a conservative estimate of $df2 = \infty$. In those cases, it is recommend to use the actual sample

size of the comparable varieties to estimate the value of df2. For example, if the actual sample size of the comparable varieties is 60, and the number of comparable varieties is limited for that species, then the threshold limit is 1.84. (df1 =60, df2 =60).

10.3 The relative variance test in practice

10.3.1 When the calculated relative variance is lower than the tabulated value of F statistic, then it is reasonable to assume that the variances are equal and the candidate variety is uniform in that particular characteristic. If the calculated relative variance is higher than the tabulated value of F, then the null hypothesis, that the varieties have equal variances, is rejected. The candidate variety would then be deemed to have a higher variance than the comparable varieties for that particular characteristic and, therefore, would not meet the uniformity criteria.

10.4 Example of relative variance method

Example

- 10.4.1 In a DUS trial, a cross-pollinated candidate variety is grown together with a number of varieties representing the required level of uniformity for all relevant characteristics. In order to illustrate the calculation of the relative variance, an example with 4 comparable varieties is given. The variance data on plant height measurements for the five varieties are presented in Table 1. For each variety, 60 plants were measured for plant height:
- 10.4.2 The number of observations per variety is the same (n=60); therefore, we can take the average variance of the comparable varieties as their pooled variance.
- 10.4.3 The average variance for comparable varieties is (7.8 + 4.5 + 3.2 + 5.8)/4 = 5.32

Table 1: variances of candidate and comparable varieties for plant height data

Candidate	Comparable		Comparable		Comparable		Comparable	
		variety 1		variety 2		variety 3		variety 4
5.6	7.8		4.5		3.2		5.8	

If the variance of the candidate variety is lower than the average variance of the comparable varieties then no further test is required, it can be deemed that the candidate variety is sufficiently uniform in the relevant characteristic. However, if the variance of the candidate variety is higher than the average variance of the comparable varieties then the variances need to be compared using the relative variance method.

10.4.4 The relative variance for a particular characteristic refers to the variance of the candidate divided by the average of the variance of the comparable varieties.

Relative variance = variance of the candidate/average variance of the comparable varieties

$$= 5.6/5.32 = 1.05$$

- 10.4.5 For a sample size of 60, the threshold limit is 1.47; therefore, we can conclude that the candidate variety is sufficiently uniform for that characteristic.
- 10.4.6 This is conservative estimate of the relative variance method using $df2 = \infty$. If the variety is found to be non-uniform using this conservative approach then the competent authority may consider whether additional approaches, such as using the actual sample of the comparable varieties for the estimation of df2, are appropriate to provide a more precise estimate of uniformity.

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10.5 Relationship between relative variance and relative standard deviation

10.5.1 Sometimes in DUS trials, the uniformity data is presented in terms of standard deviations, not as variances. Mathematically there is a simple relationship between variance and standard deviation, as follows:

Standard deviation = square root of Variance

10.5.2 When making a decision on uniformity based on relative standard deviations, the same principle for acceptance or rejection applies for relative standard deviation; only the threshold limits are lower due to the square root of appropriate values. For example, for 60 samples the relative variance threshold is 1.47; however, for relative standard deviation the threshold is 1.21, which is the square root of 1.47.

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