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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

Geneva

ENLARGED EDITORIAL COMMITTEE

Geneva, January 9 and 10, 2013

REVISION OF DOCUMENT TGP/8*:* PART II: TECHNIQUES USED IN DUS EXAMINATION, NEW SECTION 10: MINIMUM NUMBER OF COMPARABLE VARIETIES FOR RELATIVE VARIANCE METHOD

Document prepared by the Office of the Union

BACKGROUND

1. The Technical Committee (TC), at its forty-eighth session, held in Geneva from March 26 to 28, 2012, considered the proposal for a revision of Section 10: Uniformity Assessment on the Basis of the Relative Variance Method on the basis of document TC/48/19 Rev. "Revision of document TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability", Annex XIV. The TC noted the comments of the Technical Working Party on Automation and Computer Programs (TWC) with regard to some of the assumptions of the method and noted that further investigations would be done by Australia with respect to those assumptions and the F value used in the calculations (see document TC/48/22 "Report on Conclusions" paragraph 65).

2. The TC agreed with the workplan for the development of TGP/8 presented in Annex XV to document TC/48/19 Rev. which indicated that Section 10: Uniformity Assessment on the Basis of the Relative Variance Method would be considered by the TWPS in 2012. 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 Technical Working Parties (TWPs) at their sessions in 2012 (see document TC/48/22 "Report on Conclusions" paragraphs 49 and 78).

3. The following abbreviations are used in this document:

TC:	Technical Committee
TC-EDC:	Enlarged Editorial Committee
TWA:	Technical Working Party for Agricultural Crops
TWC:	Technical Working Party on Automation and Computer Programs
TWF:	Technical Working Party for Fruit Crops
TWO:	Technical Working Party for Ornamental Plants and Forest Trees
TWPs:	Technical Working Parties
TWV:	Technical Working Party for Vegetables

COMMENTS BY THE TECHNICAL PARTIES IN 2012

The TWA, TWV, TWC, TWF and TWO considered documents TWA/41/26, TWV/46/26, TWC/30/26, TWF/43/26, TWO/45/26, respectively, and commented as follows:

General	The TWA agreed that Chapter 10.2 “Threshold limits for Relative Variance Method” of the Annex to document TWA/41/26 should be considered by the TWC for incorporation in document TGP/8/1 Section 10. The TWA agreed that the remaining paragraphs were already covered by TGP/8/1 Section 10 (see document TWA/41/34 “Report”, paragraph 38).	TWA
	The TWV agreed with the proposal of the TWA that Chapter 10.2 “Threshold limits for Relative Variance Method” of the Annex to document TWV/46/26 should be considered by the TWC for incorporation into document TGP/8/1 Section 10 (see document TWV/46/41 “Report”, paragraph 37).	TWV
	The TWC noted the comments made by the TWA and TWV and agreed that Section 10.2 should be incorporated in document TGP/8. It requested the drafter to prepare a new draft after checking whether the remaining sections were already covered under section 10 of the TGP/8 (see document TWC/30/41 “Report”, paragraph 36).	TWC

4. Annex I to this document contains the text proposed by the drafter (Mr. Nik Hulse, Australia) for replacement of the text of Section 10: “Uniformity Assessment on the Basis of the Relative Variance Method”, as considered by the TWPs, at their sessions in 2012.

5. Annex II to this document contains the proposed text by drafter (Mr. Nik Hulse, Australia) for revision of Section 10: “Uniformity Assessment on the Basis of the Relative Variance Method” of the current text of Section 10 of document TGP/8/1, on the basis of comments by the TWPs, at their sessions in 2012. The amendments to the text of Section 10 of document TGP/8/1 are indicated by highlighting and strikethrough for deletions and highlighting and underlining for additions.

[Annexes follow]

ANNEX I

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 document 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 is a limited number of comparable varieties available for a species, it is not practical to use a conservative estimate of df2 = ∞. In those cases, it is recommended 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 a conservative estimate of the relative variance method using df2 = ∞. 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.

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.

[Annex II follows]

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~~,~~ irrespective of the method of propagation of the variety. 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 In cross-pollinated varieties, a common recommendation in the UPOV Test Guidelines is to take 60 measurements per characteristic per variety. In essence, the variance ratio equates to the F statistic, and the tabulated value of F at P = 0.01 under df_1 =60 (degrees of freedom of candidate) and $df_2 = \infty$ (degrees of freedom of comparable variety(ies)) is ~~1.60.~~1.47, $df_2 = \infty$ 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, ~~1.6~~. 1.47 is the threshold limit for cross-pollinated species with ~~60~~ measurements per characteristics per variety. For different sample sizes, a different F statistic should be used for the df_1, although the df_2 should remain at ∞.

10.2 Threshold limit for different sample sizes

10.2.1 ~~Different threshold limits of F (at P = 0.01) should be applied for different sample sizes of the candidate variety. The df_1 will vary according to different sample sizes of the candidate variety.~~ However, ~~in all cases the df_2 will be considered to be ∞, to cover the whole range of possible~~ when there is a limited number of comparable varieties ~~within~~ available for a species ~~thus providing~~, it is not practical to use a conservative estimate of ~~the threshold. Under these conditions and taking the relevant values from the F table, Table 1 shows the threshold limits that would apply for different sample sizes of the candidate varieties. In the case of different sample sizes than those included in Table 1, the correct threshold limit should be used for the exact~~ df2 = ∞. In those cases, it is recommended 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).

~~*Table 1: Threshold limit for relative variance for some different sample sizes*~~

~~SAMPLE SIZE OF CANDIDATE~~	~~THRESHOLD LIMIT FOR RELATIVE VARIANCE~~
~~30~~	~~2.03~~
~~40~~	~~1.81~~
~~50~~	~~1.68~~
~~60~~	~~1.60~~
~~80~~	~~1.49~~
~~100~~	~~1.43~~
~~150~~	~~1.33~~
~~200~~	~~1.28~~

~~Source: Table of F published in 'Tables for Statisticians' Barnes &Noble, Inc. New York~~

~~10.2.2For a given sample size, if the relative variance exceeds the threshold limit, the candidate variety will be deemed to be non-uniform for that characteristic.~~

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 ~~presented in Table 1, for the relevant sample size~~, 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 ~~2~~1. For each variety, 60 plants were measured for plant height:

~~*Table 2 variances of candidate and comparable varieties for plant height data*~~

~~Candidate~~	~~Comparable Variety 1~~	~~Comparable Variety 2~~	~~Comparable Variety 3~~	~~Comparable Variety 4~~
~~5.6~~	~~7.8~~	~~4.5~~	~~3.2~~	~~5.8~~

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

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.

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	

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 ~~Now, in Table 1,~~ for a sample size of 60, the threshold limit is ~~1.60~~1.47; therefore, we can conclude that the candidate variety is sufficiently uniform for that characteristic.

10.4.6 This is a conservative estimate of the relative variance method using df2 = ∞. 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.

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 Therefore, when dealing with relative standard deviations, Table 1 needs to be modified to include the square roots of the threshold limits, which is presented in Table 4.~~

~~*Table 4: Threshold limit for relative standard deviations for some different sample sizes*~~

~~SAMPLE SIZE OF CANDIDATE~~	~~THRESHOLD LIMIT FOR RELATIVE STANDARD DEVIATIONS~~
~~30~~	~~1.42~~
~~40~~	~~1.35~~
~~50~~	~~1.30~~
~~60~~	~~1.26~~
~~80~~	~~1.22~~
~~100~~	~~1.20~~
~~150~~	~~1.15~~
~~200~~	~~1.13~~

~~10.5.3~~ 10.5.2 When making a decision on uniformity based on relative standard deviations, ~~the examiner needs to use Table 4, instead of Table 1, to get the appropriate threshold limits.~~ ~~T~~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 ~~1.60~~1.47; however, for relative standard deviation the threshold is ~~1.26~~ 1.21, which is the square root of ~~1.60~~1.47.

[End of Annex II and of document]