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**FURTHER COMPARISON OF DECISIONS ON UNIFORMITY OF RYE VARIETIES
BASED ON COYU APPROACH AND ON BENNETT'S TEST**

Document prepared by experts from Poland

Summary

1. In document TWC/23/9 the year-by-year decisions concerning uniformity of rye varieties based on the UNIF approach and an approach based on the Bennett's test were compared. The decisions were, to a wide extent, consistent.

2. In this document the decisions concerning uniformity of rye varieties based on the COYU method and on a method that uses Bennett's test for coefficients of variations are compared. All considerations are illustrated with trial data concerning winter rye varieties. Both methods use the same results of a three-year series of DUS trials.

Introduction

3. In uniformity testing, the homogeneity of plants within varieties is tested. Differences between plants within a variety should not exceed a specified threshold. The methods used for checking uniformity depend both on the features of propagation of the variety and on the type of characteristic (i.e. quantitative, qualitative, or pseudo qualitative). In general, for self-pollinated and vegetatively propagated varieties, it is possible to count the number of off-type plants and compare with the maximum number of off-types allowed (threshold).

4. The threshold value depends on three parameters, namely on the population standard (the maximum number of off-type plants allowed), the sample size and the significance level of the statistical test.

5. A more complicated situation exists for cross-pollinated varieties to be clarified. Such varieties are usually less uniform than self-pollinated varieties. The decisions on uniformity of cross-pollinated varieties are based on the standard deviation calculated from pre-assumed sample and compared with standard deviations obtained within the same experiments for existing uniform varieties. Thus decisions are relative, depending on the set of varieties being compared within the DUS trial. A method and relevant statistical procedures has been developed within UPOV, and will be included in document TGP/8 "Comments on TGP documents". Because sample standard deviations depend on the levels of expression of the characteristics under consideration some additional procedures have been elaborated to remove these influences. The whole procedure is known as COYU (combined-over-years uniformity criterion). Other statistical procedures are not excluded if they address the mentioned problems in an appropriate way. For example, in document TWC/23/9, the Bennett's test for coefficients of variations was proposed as an alternative approach for testing the uniformity of varieties. In the mentioned paper these two approaches were compared. Only year-by-year comparisons were made and no meaningful differences between decisions based on those two methods were detected.

6. However in the case of rye varieties in Poland, the final decisions on uniformity are usually taken at the end of three-year period of DUS testing. Therefore in this paper, using the same data, the decisions concerning candidate varieties of rye based on COYU and a new method that uses the Bennett's test are compared. The following comparisons of these methods are made at different significance levels.

Trial data

7. The data used in document TWC/23/9 for year-by-year comparisons are used here to make over-years comparisons of the two approaches. The data concern twelve candidate varieties compared in a three-year series of trials conducted at the official variety testing experimental station at Słupia Wielka, Poland. A detailed description of these trials is given in document TWC/23/9. The characteristics observed and their codes were as follows:

8. C31- plant height, C32 - length between upper node and ear, C33 - length of ear, C10 - length of blade of leaf next to flag leaf, C11 - width of blade of leaf next to flag leaf, C51 - number of spikelets, C52 - length of rachis. The applied coding is in accordance with that used in official DUS testing in Poland. There were a total of 73, 83 and 75 varieties compared in years 1999, 2000 and 2001, respectively. Many other characteristics were also observed, but they were qualitative in nature and were not included in the statistical analysis.

The methods

9. The combined-over-years criterion (COYU) consists of calculation of the threshold value for all characteristics, in turn, using the formula:

$$UC = \bar{s}_d + t_p \sqrt{s^2 \left(\frac{1}{l} + \frac{1}{lw} \right)} \quad (1)$$

where

\bar{s}_d is the average of corrected standard deviations calculated over all varieties assigned to the reference collection (the set of varieties the new variety is compared with), s^2 is the sample variance among corrected standard deviations (of reference collection varieties) after removing the effects of years.

l stands for the number of years of trailing (usually 3 or 2),

w is the size of reference collection,

t_p means the critical value of one-side t-Student's distribution at probability p and degrees of freedom associated with s^2 (see Talbot [2000]). Usually the value of $p=0.001$ or 0.002 is accepted but other values are also admitted. If the (possibly adjusted) standard deviation of a particular variety is smaller than the UC value (threshold) for all considered characteristics, the variety is declared uniform. Therefore, if for just one characteristic, the standard deviation is larger than the threshold, the variety is considered to be non-uniform.

10. In this research the reference collection consists either of all registered varieties included in trials or of ten varieties with the closest mean values to the mean value of the variety being examined.

11. The details and justification of a Bennett's method are given in a paper by Bennet [1976]. The application of this method for a year-by-year approach is described in document TWC/23/9. For over-years situation, the method is quite similar and the only difference is that empirical coefficients of variation for varieties are averaged over years and the numbers of measurements from years added. So the value of test statistics for the hypothesis that there

are no differences among coefficients of variation against the relevant alternative hypothesis is calculated using formula

$$2Z = (n - v) \log \left(\frac{\sum_i \frac{n_i z_i^2}{1 + z_i^2}}{n - v} \right) - \sum_i (n_i - 1) \log \left(\frac{\frac{n_i z_i^2}{1 + z_i^2}}{n_i - 1} \right) \quad (2)$$

Where: n_i

is the number of independent observations of characteristic for i -th variety ($i = 1, \dots, v$);

$v=w+1$ means the size of reference collection plus one new variety,

$$n = \sum n_i ; z_i = \frac{\sum_{j=1}^l s_{ij} / \bar{x}_{ij}}{l},$$

s_{ij}^2 is the sample variance of all measurements of i -th variety for j -th year,

\bar{x}_{ij} is the adequate mean value.

All characteristics are analyzed one-by-one. The value of the test statistic is compared with χ^2 critical value at the appropriate significance level for w degrees of freedom, where w is the size of reference collection. The results obtained are presented below.

12. The number of varieties in the DUS trial is usually different in every year. This is because, every year new (candidate) varieties enter the system of testing and old candidates (accepted and rejected) are discarded. Furthermore in each DUS trial, only varieties to which new varieties are likely to be similar are grown. So both the number of candidate varieties and established (known) varieties can be different every year.

13. For the purpose of this research, from all varieties present in DUS trials in the period 1999-2000, only those present FOR the whole period were chosen. Therefore only results of 12 new (candidate) varieties and 19 varieties belonging to the reference collection were used.

14. The uniformity of varieties was examined using two methods of testing (COYU and Bennett's) at different levels of significance. The Bennett's test was applied in two versions. In the first version the uniformity of new varieties was tested against all established varieties, while in the second the uniformity of new varieties was tested against a subset of ten varieties with the most similar level of expression for the characteristics concerned (with similar mean values). The COYU analysis was performed with use of DUST package of Weatherup [1992]. The results of all calculations are presented in Tables 1, 2, 3 and 4.

Results

15. In Table 1, the numbers of candidate varieties declared as non-uniform by the methods under comparisons after testing at three different significance levels are given for all

characteristics. The two versions of the Bennett's method are distinguished by the capital letter W (whole set of reference collection) and S (subset of reference collection).

16. These calculations were done in order to identify the significance level and method of comparison which would give the most compatible results compared to those obtained by the COYU method. The chosen levels are close to the significance levels commonly used.

17. In Table 2, the numbers of discordant decisions (for all candidate varieties and all characteristics) are presented. Discordance means that one method (e.g. COYU) concludes that a variety is uniform while the other method (e.g. Bennett's method) concludes that the same variety not uniform or vice versa. The numbers of discordant results are given for both versions of the Bennett's test. In table 2, uniformity is denoted by "U" and lack of uniformity by "NU". For example, in the third column of table 2, the summary numbers of discordant results for COYU and Bennett's method (in its two versions), all at significance level 0.002, are given. In total, for significance level 0.002, there were four discordant results. When all tests were performed at level 0.0015, the number of discordant results was reduced to three.

18. In Table 3, COYU and the two versions of the Bennett's test are compared using side-by-side methods at different significance levels. The two versions of the Bennett's test gave exactly the same results on the uniformity of the candidate varieties. There were some discrepancies in the results concerning uniformity provided by COYU and Bennett's tests. Discrepancies between COYU and Bennett's tests were exactly the same for all applied significance levels of Bennett's tests. Nevertheless, these discrepancies are statistically not significant, which is illustrated in Table 4. Because the results of Bennett's test were the same for all significance levels, they are given together in Table 4. In Table 4, the results of the application of the exact Fisher test for side-by-side comparisons are shown. All empirical significance levels are meaningfully larger than standard $\alpha = 0.05$ level.

Observations and conclusions

19. The analysis of three-year series of DUS trials on rye varieties showed that:
- in the majority of cases, decisions on the uniformity of varieties were exactly the same for the COYU method and the Bennett's method (both versions),
 - the Bennett's methods seems to be slightly more tolerant (more varieties declared uniform) than COYU (when both applied at the same significance level),
 - further research is needed (for different species and series of trials) to make a more general conclusion on the behaviour of those methods.

References

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Table 1. Numbers of varieties declared as non-uniform

Characteristic	Significance level								
	0.002			0.0015			0.001		
	COYU	W	S	COYU	W	S	COYU	W	S
C31	1	1	1	1	1	1	1	1	1
C32	1	1	1	1	1	1	1	1	1
C33	0	0	0	0	0	0	0	0	0
C10	1	0	0	1	0	0	1	0	0
C11	2	0	0	2	0	0	2	0	0
C51	1	0	0	0	0	0	0	0	0
C52	0	0	0	0	0	0	0	0	0

Table 2. Summary numbers of discordant results for the methods under comparison for all characteristics

Bennett COYU		$\alpha=0.002$		$\alpha=0.0015$		$\alpha=0.001$	
		W & S		W & S		W & S	
		U	NU	U	NU	U	NU
$\alpha=0.002$	U	78	0	78	0	78	0
	NU	4	2	4	2	5	1
$\alpha=0.0015$	U	79	0	79	0	79	0
	NU	3	2	3	2	4	1
$\alpha=0.001$	U	79	0	79	0	79	0
	NU	3	2	3	2	4	1

Table 3. Two-by-two table of results concerning the uniformity of varieties

Bennett COYU		$\alpha=0.002$		$\alpha=0.0015$		$\alpha=0.001$	
		W & S		W & S		W & S	
		U	NU	U	NU	U	NU
$\alpha=0.002$	U	9	0	9	0	9	0
	NU	2	1	2	1	2	1
$\alpha=0.0015$	U	10	0	10	0	10	0
	NU	1	1	1	1	1	1
$\alpha=0.001$	U	10	0	10	0	10	0
	NU	1	1	1	1	1	1

Table 4. Comparison of results by exact Fisher test

Bennett COYU		$\alpha=0.002, 0.0015, 0.001$		
		W & S		α emp.
		U	NU	
$\alpha=0.002$	U	9	0	0.25
	NU	2	1	
$\alpha=0.0015$	U	10	0	0.17
	NU	1	1	
$\alpha=0.001$	U	10	0	0.17
	NU	1	1	

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