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TC/28/3

# INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

#### GENEVA

# **TECHNICAL COMMITTEE**

Twenty - eighth Session Geneva, October 21 to 23, 1992

MATTERS ARISING FROM THE 1992 SESSIONS OF THE TECHNICAL WORKING PARTIES TO BE DEALT WITH BY THE TECHNICAL COMMITTEE

# Document prepared by the Office of the Union

This document summarizes, in its Annex, matters arising from the 1992 sessions of the Technical Working Parties, which have to be dealt with by the Technical Committee (hereinafter referred to as "the Committee"). They comprise: (i) Questions presented by the Technical Working Parties to the Committee; (ii) Important decisions taken by the Technical Working Parties and communicated to the Committee for information; (iii) Matters dealt with by the Technical Working Parties on the instructions of the Committee or in preparation for discussions planned in the Committee under separate agenda items. The headings of the different items are listed on pages 1 and 2 of the Annex.

To shorten references to the various Technical Working Parties in this document, use is made of the following codes that designate their documents:

[Annex follows]

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# MATTERS ARISING FROM THE 1992 SESSIONS OF THE TECHNICAL WORKING PARTIES TO BE DEALT WITH BY THE TECHNICAL COMMITTEE

#### Addressee of Test Guidelines

1. In connection with the discussionns on Test Guidelines for Soya Bean in the TWA, the question was raised as to who was the primary addressee of the UPOV Test Guidelines. As a result of those discussions, the TWA agreed that the addressees of the Test Guidelines were the national offices of the member States. Those national authorities would then either use the Test Guidelines in the given form or modify them to suit the national situation either for their own official testing or for testing done by applicants or breeders. They might, if applicable, add additional information addressed to the applicant or breeder to ensure thorough testing by him.

(see TWA/21/9 Prov., paragraph 39)

2. <u>The Committee is invited to take</u> the necessary decisions.

#### Separate Sets of Example Varieties for Different Regions

3. The TWA discussed, in connection with the Test Guidelines for Cereals, the need to also give other than European example varieties in the above documents. In this connection, the question arose whether sets of example varieties for different areas would have to be established separately or whether they could be combined in one single list. The TWA finally agreed to prepare proposals for separate sets of example varieties for different regions. The separate lists of example varieties should include for each example variety the expressions of the grouping characteristics. The whole question of example varieties should, however, be discussed by the Technical Committee.

(see TWA/21/9 Prov., paragraph 32)

4. <u>The Committee is invited to note</u> the <u>above information</u> and to <u>consider</u> possible steps to be taken.

#### Addition of States to Existing Qualitative Characteristics

5. The TWC dicussed at length the order of additional states of expression added to existing qualitative characteristics at the time of revision of a given Test Guidelines document. It proposed that the habit of adding additional states at the end of the list of existing states of qualitative characteristics should be discontinued. The new states should always be placed at their logical place, e.g. the place they would have occupied had they been included right from the beginning.

(see TWC/10/11 Prov., paragraph 4)

6. <u>The Committee is invited to take</u> the necessary decisions.

# One or Two Testing Centers

7. In the discussion on the Test Guidelines for Maize, the TWA discussed at length the differences between the member States as to the use of one or two testing places and their influence on the decision on distinctness. There were mainly two approaches, the first using only one location for the decisions on distinctness, but keeping a second testing place as a backup in case of unfavorable climatic or other conditions, and the second to use both testing places and obtain an average for the description of the variety. The TWA therefore asked the Subgroup to address the question again and come back with further remarks. At the same time, it asked the Technical Committee to discuss the problem for other species.

(see TWA/21/9 Prov., paragraph 34)

# 8. The <u>Committee</u> is <u>invited</u> to note the <u>above</u> information and to <u>consider</u> possible steps to be taken.

## Weighting of Characteristics

9. The TWA noted the weighting of characteristics in the screening of inbred lines of maize as explained by experts from France. It noted that the threshold for screening was higher than that applied for distinctness purposes. However, the TWA did not take any definite decision on the weighting of characteristics and left it to further discussion during its coming session or to the Technical Committee.

(see TWA/21/9 Prov., paragraph 37)

10. The Committee is invited to note the above information and to consider possible steps to be taken.

#### Test Guidelines for Rape

The TWA noted the discussions which had taken place in the Subgroup on 11. Rape on the different groups of varieties, inbred lines and narrow populations, hybrid varieties and synthetic varieties, and on the question whether the components of hybrids should be included in the test and whether unthreshed plants should be tested. At present it was premature to include electrophoretic characteristics in the Test Guidelines. There had been discussions on the different approaches with respect to off-types depending on whether one looked at distinctness (where some experts would admit a difference in several characteristics) or at the off-type in uniformity (where off-types in one characteristic would be sufficient to reject the variety). With respect to uniformity, standards would also still have to be established as in some countries not all characteristics were tested for uniformity. For hybrids no standards existed so far. The TWA finally agreed to submit the question whether the components of hybrids should be included in the test and whether unthreshed plants should be tested to the Technical Committee for discussion.

(see TWA/21/9 Prov., paragraph 43)

12. The <u>Committee</u> is <u>invited</u> to note the <u>above</u> <u>information</u> and to <u>consider</u> possible steps to be taken.

# Influence of the Method of Propagation on the Expression of Certain Characteristics

13. The TWV noted the difficulties in testing varieties of formerly seedpropagated species where vegetative propagation now led to differences which resulted only from the difference in propagation. Thus, in tomato obtained from tissue culture, epigenetic effects on the phenotype meant that changes in the physiological characteristics as, for example, earliness, but to a certain degree also fruit length, could be observed. In The Netherlands, through repetitive testing, the authorities will ensure that differences in propagation will not lead to a second grant of rights. Similar behavior was also found in cucumber, where more morphological characteristics changed. Here, it was tried by means of cuttings to remove the hormone effect of tissue culture on the expression of the characteristics of the variety. The TWV was aware that in the future more vegetative propagation may take place by tissue culture in laboratories and therefore it would have to watch developments very carefully.

(see TWV/26/11 Prov., paragraph 4)

14. The <u>Committee is invited to note</u> the <u>above information</u> and to <u>consider</u> possible steps to be taken.

#### Umbrella Varieties

The TWV noted that the Commission of the European Communities (EEC) had 15. completed its work on the separation of umbrella varieties into several different varieties. A 544-page document had been prepared by the Commission on the "EEC "Umbrella" Varieties Programme for Vegetables", on the "Renewal of the official acceptance of certain old vegetable varieties and derivation of varieties from them" as Council Directive 70/458/EEC on the marketing of vegetable seed. Copies of this voluminous document were handed out to the individual experts. Part I contains administrative and legal aspects in four chapters, namely (1) List of varieties classified as being "umbrella" varieties; (2) Splitting of the "umbrella" varieties and names to be borne by the varieties derived therefrom; (3) Implementation of the renewal of acceptance of the "umbrella" varieties in the National Catalogues of the Member States and in the Common Catalogue; (4) Persons responsible for the maintenance of the "umbrella" varieties officially registered at the time of the renewal; and Part II, technical aspects with three chapters, namely (1) Characteristics used to describe the varieties; (2) Descriptions of the varieties and (3) Maintenances of the varieties included in the investigation.

(see TWV/26/11 Prov., paragraph 15)

16. The Committee is invited to note the above information.

#### Application of the Combined Over-Years Distinctness (COYD) Analysis

After an enquiry made during the session, the TWC noted that in five of 17. the member States represented during the session COYD analysis would be applied to varieties of grasses and in some of them also to varieties of clover, sugar beet, Festuca, maize, rape seed, field bean, lucerne. It would be under study in some of the member States for varieties of flax, linseed, onion, shallot and leek. As agreed in the past, some countries would still apply COYD analysis at the 5% LSD level for a transitional period. They planned, however, to change as of 1994 to the 1% level. The TWC agreed that it was important to convince more member States to change to the COYD analysis. Therefore, the importance of the method should be stressed to others and for this purpose the expert from the United Kingdom was asked to rewrite the present document on COYD analysis, by foreseeing two parts: а first part in which the usefulness and the idea of the method would be explained in simple terms while, in the second part, the full method would be given including all information necessary to permit application of the method without the need to request the computer program in electronic form.

(see TWC/10/11 Prov., paragraph 8)

18. The <u>Committee</u> is <u>invited</u> to note the <u>above</u> <u>information</u> and to <u>consider</u> possible steps to be taken.

#### Long Term LSD Method

Having discussed at length the application of the long-term LSD method, 19. the TWC finally concluded that in all cases where at least 20 degrees of freedom were available the ordinary COYD analysis should apply. Only if less than 20 degrees of freedom were available due to the low number of varieties tested, should the long-term LSD method be applied. Before application, however, it was to be tested whether the long-term LSD was stable over the years for each crop and for each testing center. Should it prove not stable, could only be applied with great care. The TWC discussed the it recommendation to use the long-term LSD method provisionally. It would monitor the experience gained in the coming years before making a definite recommendation. The TWC agreed that there was always a certain natural grouping and it would therefore be left to the expert to decide within each group whether he would apply the long-term LSD method or not. Experts were invited to examine further whether it was possible to apply the closed group LSD method or not.

(see TWC/10/11 Prov., paragraph 7)

20. The Committee is invited to note the above information and to consider possible steps to be taken.

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#### Combined Over-Years Uniformity (COYU) Analysis

21. The TWC noted the introduction to document TWC/10/7 prepared by experts from Germany on the study of the proposed COYU analysis levels. The document concluded by stating that the probability levels agreed for experiments on grasses for 1991 and 1992 would allow smooth transition from the present uniformity criteria to the COYU criteria. After having heard the reports of the other experts on their experience with the application of COYU analysis, the TWC noted that none of the experts that had studied the application of that method had found anything which would prevent them from changing to the method with the required levels. The TWC thought, however, that some more experience and study would be necessary before a final decision on the level could be taken. It agreed therefore to continue trials next year with the levels as proposed during the last session of the TWC (rejection after 3 years: 0.2%, rejection after 2 years: 0.2% and acceptance after 2 years: 2%). The TWC agreed that the experts from Denmark would collect all information from the other member States for the preparation of a document to be circulated to all members of the Working Party.

### (see TWC/10/11 Prov., paragraph 9)

22. The TWC agreed furthermore that, as already decided for the COYD analysis, the expert from the United Kingdom would be asked to revise the present document on COYU analysis in a similar way, in two parts: a simple explanation and thereafter a full description of the method in a way that would enable experts to apply the method without the need to request the computer program in electronic form.

(see TWC/10/11 Prov., paragraph 10)

23. The <u>Committee</u> is <u>invited</u> to note the above information.

#### Multi-Variate Analysis

24. The TWC noted document TWC/10/4 on the Multi-variate Distinctness Criterion prepared by Dr. Weatherup (United Kingdom). The document concluded that Mahalanobis  $D^2$  statistics could be used to distinguish between variety pairs without much change in testing stringency as compared with COYD analysis. However, further work would be required to ensure the validity of the method in the DUS case where only two or three years, or replicates, of data are available. The statistics could also be used as an investigational tool to assist in the determination of the plant feature, such as a contrast of characteristic differences which could lead to distinctness being established by uni-variate procedures. The TWC agreed that this method should not be a method additional to COYD analysis. The method could, however, be used to confirm that COYD analysis worked well. It could also be used to identify those characteristics which were the distinguishing characteristics of the variety. It could be used as a safeguard towards making wrong decisions in using a number of uni-variate analyses. This method would give experts the possibility of detecting new relations between characteristics and arriving at newly derived characteristics for the use of distinctness. The experts from the United Kingdom would investigate the matter further and prepare a paper for the next session of the Working Party.

(see TWC/10/11 Prov., paragraph 13)

# 25. <u>The Committee is invited to note</u> the above information.

#### Testing of Uniformity, Off-types

26. The TWA noted document TWC/10/5, explaining the approach in selecting the right tables for the testing of uniformity of vegetatively propagated and truly self-pollinated varieties, as listed in document TC/XXV/8. The document stressed that it was imperative that the technical experts, when preparing Test Guidelines and discussing the maximum number of off-types admitted, should first fix the population standard, i.e. the maximum percentage of off-types allowed in varieties of that species if the whole population were to be examined.

(see TWA/21/9 Prov., paragraph 25)

27. After acceptance of document TC/XXV/8 by the Technical Committee, certain experts in the TWC asked what the present situation was with respect to the testing of uniformity for vegetatively propagated varieties and truly self-pollinated varieties and especially whether the table mentioned in paragraph 28 of the General Introduction to the Test Guidelines was completely replaced by document TC/XXV/8. The TWC therefore asked the Technical Committee to clarify the situation.

(see TWC/10/11 Prov., paragraph 12)

28. Having noted the decision of the Technical Committee, taken during its last session, that the table of maximum numbers of off-types, as reproduced in paragraph 28 of document TG/1/2, was to be replaced by the tables reproduced in document TC/XXV/8 and having noted the doubt expressed with respect to this replacement by the TWC and the latter's request for clarification by the Technical Committee, a TWA subgroup prepared a proposal for a text for replacement of paragraph 28 of document TG/1/2. After having amended the proposal in the meeting, the TWA proposed to the Technical Committee for final approval the following replacement for paragraph 28 of document TG/1/2 :

"For vegetatively propagated and self-fertilized species, the sample size and the maximum number of off-types will be given in the individual guidelines and be based on the tables in document TC/XXV/8. The crop experts choose the appropriate table when preparing the guidelines by first fixing the population standard, i.e. the maximum percentage of off-types allowed if the whole population were to be examined. The acceptance probability--i.e. the probability that a variety having P% of off-types is correctly considered uniform--and the sample size are then chosen. Small sample sizes increase the risk of accepting heterogeneous varieties.

#### Examples:

Population standard "P"	Acceptance probability	Sample size	Maximum number of off-types allowed	Risk of erroneously accepting a hetero- geneous variety with, for instance, 5% off-types
1%	95%	10	0	60%
18	95%	20	0	36%
1%	99%	100	3	16%
0.1%	99%	1000	3	3 8
0.1%	99%	2000	5	<b>?</b> &"

(see TWA/21/9 Prov., paragraph 27 and annex III)

# 29. <u>The Committee is invited to take</u> the necessary decisions.

#### Testing of Uniformity, Qualitative Characteristics

30. The TWA noted that the General Introduction to the Test Guidelines at present did not deal with clear off-types in qualitative characteristics of cross-fertilized plants. It agreed that the tables in document TC/XXV/8 could be used in the same way and that this fact should be clarified at the next revision of the General Introduction to the Test Guidelines.

(see TWA/21/9 Prov., paragraph 27)

# 31. The <u>Committee</u> is invited to take the necessary decisions.

#### Testing of Uniformity, Sample Size

32. The TWC had a lengthy discussion on sample size and the probability levels for the acceptance of a variety and on the effect of a low number of plants with respect to the risk of accepting varieties that were too heterogeneous. It stated that in certain cases, as for example in electrophoresis tests, the low number of seeds involved meant that it would be almost impossible to check uniformity if only 20 grains were used. This would raise the question what was a reasonable number to be tested compared with the workload involved. The TWC finally agreed to present the question to the other Technical Working Parties and ask for their comments. In addition, the experts from The Netherlands would check the ISTA rules with respect to uniformity and inform the TWC on their findings.

(see TWC/10/11 Prov., paragraph 11)

The TWA noted document TC/10/9, explaining certain consequences where 33. the sample size chosen was too small. The document stated that there should be equilibrium between the likelihood of erroneously accepting a heterogeneous variety as being uniform and the likelihood of rejecting a uniform variety as heterogeneous. The decrease in the probability of committing the one error would automatically mean an increase in the probability of committing the The TWA also noted the certification standards in The Netherlands. other. When used for testing as a prerequisite for plant variety protection, the population standard needed to be higher than for certification. In a small sample, the likelihood of admitting a heterogeneous variety was rather high. In this respect, the TWA also noted the remark that not all off-types could be observed with the same ease, some could be observed very easily, others with much more difficulty. The tables in document TC/XXV/8 were, however, based on the condition that all off-types were equally easily observed. The TWA further agreed that the tables listed in document TC/XXV/8 would lead to a low risk for the breeder, while leading to a higher risk for the consumer (meaning that a heterogeneous variety was still accepted as uniform). However, the tables were the best approximation to reality at present and the experts would always take into account certain exceptions. In order to further study the subject, the TWA proposed to the TWC that it discuss, during its next session, the way in which sequential sampling, that means taking subsamples out of the samples, could reduce the workload of the offices without unnecessarily increasing the risk of accepting varieties that were too heterogeneous.

(see TWA/21/9 Prov., paragraph 26)

34. The TWA also discussed problems connected with the low sample size in the case of electrophoretic characteristics. It noted that at present the sample size of the gel varied between 10 and 80 from one country to another. To justify a low sample number in electrophoretic characteristics, it was stated that electrophoresis was a very precise tool which was not influenced by environment, and which used the same standard as, for example, used in ear rows. It was necessary, however, to study the risk involved via sequential analysis and especially to give a better description of the risk when reducing the sample size. The experts in favor of a low sample size were ready to accept a higher consumer risk because of the very precise method. The TWA agreed that it would discuss the sample size and the study of subsampling on the basis of the paper on sequential analysis which the experts from The Netherlands had offered to prepare.

(see TWA/21/9 Prov., paragraph 29)

35. The <u>Committee is invited to note</u> the <u>above information and to consider</u> possible steps to be taken.

#### Number of Statistical Documents

36. The TWA also noted certain criticisms of the high number of documents produced by the TWC, many of which seemed too theoretical and were difficult for the crop experts to understand and apply. It noted at the same time that, without the help of the TWC and the establishing of the COYD analysis, national offices would be at a loss with respect to the testing of, for example, varieties of grasses and would have to reject many varieties to the detriment of the breeder. It also noted that, as concerns the testing of TWV and TWO varieties, the methods developed by the TWC were hardly used as the

TWO very seldom applied statistics and statistics were at any rate not applicable to most of the vegetable species. As to the criticism concerning the difficulty of understanding the TWC documents, the TWA had been informed that the TWC had issued a document (TWC/10/3) in which the different methods were explained and that a revised version of that document containing a much simpler introduction to each of the methods was under preparation for the coming session of the TWC.

(see TWA/21/9 Prov., paragraph 28)

# 37. The <u>Committee</u> is <u>invited</u> to note the above information.

### Computer Format for Exchange of Descriptions of Varieties.

38. The Working Party agreed that it would need a standardized computer format for the exchange of descriptions of varieties in electronic form. It set up a small subgroup with experts from Germany, France and the United Kingdom to prepare a draft format for the exchange of lists of varieties under test in electronic form including their grouping characteristics. The expert from the United Kingdom would prepare a paper for circulation via the Office of UPOV on that format. Comments on the paper were to be sent to the expert of the United Kingdom for compilation into a document for the next session of the TWC.

(see TWC/10/11 Prov., paragraph 15)

39. <u>The Committee is invited to note</u> the above information.

#### Access to International Data, Computing Centre Electronic Communications.

40. The TWC noted a table on computing centre electronic communications, circulated during the meeting. The experts were asked to inform Mr. Talbot (United Kingdom) on any updating of that table. The updated version is contained in document TWC/10/12.

(see TWC/10/11 Prov., paragraph 17)

41. The <u>Committee</u> is <u>invited</u> to note the above information.

## Programs Which Can Be Readily Assimilated in Other Plant Variety Computer Systems

42. The TWC noted that no request for changes to the information as included in Annex VIII of document TWC/VI/13 had been received by the experts from the United Kingdom. The TWC asked its experts to send any further information and especially additional information on commercial packages used by the member States as well as any relevant macros to Mr. Talbot (United Kingdom) for the preparation of an updated list.

(see TWC/10/11 Prov., paragraph 18)

43. The Committee is invited to note the above information.

# Review of Documents on Statistical Methods Discussed During Past Sessions

The TWC noted document TWC/10/3 containing a review of statistical 44. documents. After a general discussion on the way the papers should be presented in such a document, the TWC agreed that each paper should contain a brief description of the method, followed by a short history of its development, with a review of the most important papers prepared in that connection. In addition to the short list of most important papers, to be annexed to each part, the document should also contain, at the very end, a complete list of all the documents issued, including circulars of importance and also other documents which did not have the code TWC, but which were important and had been prepared during the process of reaching the final Thus, for example, the document TC/XXV/8, prepared for the conclusion. Technical Committee, should be part of that document. The papers on COYD and COYU should be worded in such a way that they could form part of the annexes to the revised General Introduction to the Test Guidelines, after inclusion of a few sentences in the final text of that revised General Introduction. Both papers should comprise a first part in which the method would be described in simple terms to be understood by a non-statistician and a second part containing the full description of the method, including the program for the calculation of COYU and COYD, to enable experts to do the final calculation on the basis of the information on paper without having to request the computer program in electronic form. The TWC agreed that all those involved in the preparation of document TWC/10/3 would re-write their parts of that document accordingly. Henceforth, all documents should also contain key words to ease reference. The revised document TWC/10/3 should not only explain methods that have already been finalized, but also methods during their development. The TWC would update that document every year either by additions to the existing document or, after several editions, by a completely revised new document.

(see TWC/10/11 Prov., paragraph 21)

# 45. The <u>Committee</u> is <u>invited</u> to note the above information.

#### Handling of Visually Assessed Characteristics

The TWC noted document TWC/10/8 in which a first step had been made to 46. start work on the data of visually observed characteristics which were difficult to handle and therefore often avoided by statisticians. The document explained the different scales included, the nominal scale, the ordinal or rank scale, interval and ratio scale and counts. As a result of this study, it was concluded that descriptive statistics gave the expert an intuitive understanding of the data additional to his experience. Attention should be paid to strongly correlated characteristics and it should be checked whether some of them could be eliminated to save time and cost. The analysis of variance could be applied for an equatory use if the data had a 1 to 9 scale with a nondegenerated sampling distribution. Fy values for years allowed the identification of characteristics which were mostly influenced by year effects. Fv values made it possible to determine whether characteristics had a high discriminative power or not. A comparison of LSD values derived by the analysis of variance with the minimum distance derived by expert knowledge provided the expert with a helpful check of his work. As the study was only a first step in identifying and handling visually observed characteristics, more experience would be necessary and further studies would have to be made,

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especially by extending the studies to other species. The techniques were very helpful in establishing or revising Test Guidelines and should be used by the experts when drafting new or revising existing Test Guidelines.

(see TWC/10/11 Prov., paragraph 22)

47. The TWC also noted a description given by the expert from The Netherlands on an analysis of ordinal data studied by him. He concluded that, for data recorded on an ordinal scale, the assumptions underlying the analysis of variance may be violated. A threshold model was therefore proposed which could be used as a basis for an alternative method of analysis. The types of results of the analysis were similar to those obtained from the analysis of variance and could be used to indicate pair-wise the differences between varieties. Further investigations into the properties of the analysis would be carried out by him to get an insight into the applicability of the method proposed. He offered to prepare a document for discussion during the coming session of the Working Party. He invited the other experts to send him data for checking for such a paper.

(see TWC/10/11 Prov., paragraph 23)

48. The TWC agreed that it was necessary to encourage more work on the analysis of visually assessed characteristics in order to understand those characteristics better. It therefore proposed that everybody should check the application of the analysis to visually assessed characteristics at home and present results for discussion during the coming session of the TWC.

(see TWC/10/11 Prov., paragraph 24)

49. The Committee is invited to note the above information.

<u>Consequences of Newly Introduced Characteristics on Protected Varieties and</u> Their Description

50. The TWA discussed the question of the legal situation if, with the introduction of new characteristics, the description of existing varieties which had not been tested for those characteristics had to be extended. This led to the question whether the protected plant material would define the description, or whether the description was only defined by the description established on paper at the time of granting protection. Would a breeder also have to keep his variety uniform in those characteristics that had not been observed at the time of granting the breeder's right, but which had been uniform, in order to keep his variety material in conformity with the deposited material?

(see TWA/21/9 Prov., paragraph 22)

51. <u>The Committee is invited to take</u> the necessary decisions.

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# New Methods, Techniques and Equipment in the Examination of Varieties (Item 7 on the Draft Agenda)

#### New Methods in the Examination of Vegetable Varieties

52. The expert from France in the TWV reported that so far new methods such as electrophoresis or DNA methods would not be used for taking a decision for the DUS, in order not to reduce the minimum distance between varieties, but that research was planned or already on-going for several species. Thus, for pea and field pea, electrophoresis was applied as a routine matter for supplementing the variety description. Electrophoresis was applied to asparagus and work would begin with respect to carrot and garlic. In garlic so far there had been no success as only three groups could be separated. Therefore, DNA probes would be tried out in France for this species. It was also planned to apply DNA probes to sweet pepper. In onions and carrots also research with image analysis had started.

#### (see TWV/26/11 Prov., paragraph 9)

53. The expert from Germany in the TWV reported on the trials of electrophoresis in asparagus, making use of the EEC trial held in conjunction with France, Italy and The Netherlands. In asparagus, three different isoenzymes would be checked on phyloclades of 80 plants. Preliminary studies were being undertaken with respect to beans and peas with the intention of using electrophoresis in the post-control. A feasibility study was underway with respect to the use of image analysis, but results could only be expected at the end of this year. It was planned to study shape characteristics in particular. Color measurements had been undertaken so far only with respect to ornamental species.

(see TWV/26/11 Prov., paragraph 10)

The expert from The Netherlands in the TWV reported on studies on image 54. analysis, measuring the pod shape of peas and beans and the bulb shape of onions and shallot and blistering in lettuce. The pod shape of peas and beans would be used this year as routine for all varieties. When comparing measurements and observations with the help of image analysis, it became apparent that the measurements by hand showed a bigger variation than those by image analysis. With respect to DNA probes, a study was underway on tomato, especially in the area of the nonexpressed genome. A high degree of polymorphism was observed. This year all varieties would be checked with this method. Color measurements would only be taken with respect to ornamental species. So far electrophoresis had been applied in The Netherlands only to agricultural species, but this year a study had also started with respect to various vegetable species, as for example shallot and asparagus. The Dutch Board of Plant Breeders' Rights had decided that the characteristics obtained with the help of electrophoresis, RFLPs or PCR may be added to the description of the variety if related to the sample. This would be especially important for mushroom varieties.

(see TWV/26/11 Prov., paragraph 11)

55. The expert from the EEC in the TWV reported that the EEC catalogue did not mention new techniques, but the approach to seed control made clear reference to new techniques. There were provisions intended to develop alternative seed control and to supplement official seed inspection with these methods. It was also intended to use electrophoresis in EEC comparative trials.

#### (see TWV/26/11 Prov., paragraph 12)

56. The TWV agreed that it was also necessary to listen to the breeder and to encourage the use of new methods. However, that use had to be made cautiously and the method could never replace field work. Therefore, it was necessary to continue to develop the field trials further.

(see TWV/26/11 Prov., paragraph 13)

57. The <u>Committee</u> is <u>invited</u> to note the above information.

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## <u>General Discussion on the Use of Electrophoresis in the Examination of</u> Varieties

In a survey, taken during the meeting of the TWA, on which of the member 58. States already used electrophoresis in the testing of distinctness and which member States planned to use it, it appeared that most member States were very cautious with respect to characteristics obtained with the help of electrophoresis especially as most breeders would be afraid of the obligation to make all the varieties uniform with respect to characteristics obtained with the help of electrophoresis. However, several member States reported that where the variety could not be distinguished by other traditional characteristics, and if the breeder agreed, they would accept a characteristic obtained with the help of electrophoresis as the final resort for taking a decision and declaring the variety distinct. In that case, however, a prerequisite was that the old variety from which the new variety was otherwise not distinct had also to be uniform in that characteristic and that the breeder maintained his variety uniform in that characteristic. As some States therefore accept characteristics obtained with the help of would electrophoresis, UPOV had to advance in its discussions to prevent it being overtaken by events.

(see TWA/21/9 Prov., paragraph 9)

59. The TWA agreed to make a survey on how far member States already used electrophoresis in other species. Dr. Camlin (GB) would prepare a questionnaire to be circulated via UPOV for preparation of a document for the next session of the TWA.

(see TWA/21/9 Prov., paragraph 10)

60. The TWA therefore reconfirmed what it had already agreed during its last session, namely that

(i) electrophoretic characteristics should be included in the Table of Characteristics and not in an annex to the Test Guidelines;

(ii) the characteristics should not have an asterisk;

(iii) it had to be studied further whether the characteristics could be used alone or only in combination with a traditional characteristic and whether a difference in one of the characteristics alone would be sufficient to establish distinctness.

(see TWA/21/9 Prov., paragraph 11)

61. The TWA noted that at present the discussions had centred on the methods to be used only, but not on the interpretation of the results or on the definition of the characteristics. It agreed that it should not be left to the individual member States to decide how to interpret the results.

(see TWA/21/9 Prov., paragraph 8)

62. With respect to the definition of the characteristics obtained with the help of electrophoresis, the TWA proposed that the characteristic should be defined as the absence or presence of a given allele.

(see TWA/21/9 Prov., paragraph 12)

63. <u>The Committee is invited to take</u> the necessary decisions.

#### Consequences of the Introduction of Electrophoresis in the Test Guidelines

64. With respect to the consequences of the introduction of electrophoresis in the Test Guidelines, the TWA agreed that Mr. Guiard (France) would prepare a document which would restrict itself not only to electrophoresis, but also enlarge to new sophisticated methods used for identification and for distinctness purposes. That document would include three chapters, namely:

- (i) Current developments in the field of description of varieties
- (ii) Consequences of the distinctness of varieties and breeding activities
- (iii) Solutions which should be used to maintain a good protection system.

(see TWA/21/9 Prov., paragraph 15)

65. With respect to the current developments (i), the document would discuss the tendency to search increasingly for smaller and smaller differences, to use ever more precise methods to describe varieties and research into new characteristics independent of environment. It would mention the pressure on breeders to go further in developments and the fact that breeders would always think that their variety was new. It would take into account the possibility of reading viability, which was not available before, and the rapid evolution of the methods and tools. As there was a wish for an exchange of descriptions, there was a need to have characteristics independent of environment which would lead to the introduction of characteristics closer to the expression of the gene. The offices would have no choice and indeed no reason to reject

characteristics of this kind. With respect to the consequences of distinctness (ii), the document would mention the existence of increasingly closer varieties, the fact that if no limit was placed there would always be a possibility of finding differences, that there was no reason to reject a new characteristic, the risk of moving towards ever more uniform varieties, the decrease in protection for the breeder which would result from that fact and the transfer of the exercise of protection to the dependency stage. With respect to the solutions for maintaining good protection (iii), the document would start with the correct interpretation of the new version of Article 1(iv) and Article 7 of the Convention of 1991, the study of the question species by species, the use of the genetic background of the characteristics and would promote a multi-variate approach to distinctness, as there was a need to introduce the notion of weighting of characteristics.

(see TWA/21/9 Prov., paragraph 16)

Having noted the outline of the document, some experts in the TWA fully 66. agreed with the intended document, while others warned that not just any new characteristic should be acceptable as one should be aware that the introduction of a new characteristic would be an act to the detriment of the breeder of the existing variety. Sophisticated characteristics should only be used once general agreement had been reached within UPOV. Others stated that, in general, when testing distinctness a set of several small differences was observed and it should be discussed whether that set of differences should be other sophisticated methods which would help to find considered or distinctness in the presence of the set of small differences. When looking for small differences it should always be kept in mind that when accepting such small differences for distinctness, the breeder should also be able to keep his variety stable within this small range. On the other hand, it should always be kept in mind that true breeding should be rewarded. When accepting certain methods, it should be kept in mind that it should be possible to define the method with accuracy and the effort should remain reasonable.

(see TWA/21/9 Prov., paragraph 17)

67. The breeder present as an observer in the meeting of the TWA stated that, in his personal opinion, many breeders were not in favor of too many varieties on the market as that would reduce the value of protection for each variety. The commercial advantage of each variety would be larger if fewer varieties were admitted. Therefore not just any variety should be admitted to the market and the introduction of the system of essential derivation would work in that direction. On the other hand, however, authorities should be less reluctant to admit new, modern methods in the testing of varieties. If these methods were used in the breeding of varieties, they should also be admitted in testing by the authorities. Their application could be an important cost-saving factor.

(see TWA/21/9 Prov., paragraph 18)

68. The Committee is invited to take the necessary decisions.

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69. The TWA furthermore agreed that, in future, the question of electrophoresis would not be discussed in separate subgroups on electrophoresis, but by the subgroup for the species concerned when it discussed revision or establishment of a new Test Guidelines document.

(see TWA/21/9 Prov., paragraph 21)

70. <u>The Committee is invited to note</u> the above information.

#### Electrophoresis in Cereals

71. Having taken a decision of principle on the use and definition of the characteristics obtained with the help of electrophoresis, the TWA tried to apply that decision to the testing of wheat, barley and oats. It noted that genetic knowledge differed for different species, for example there was good knowledge with respect to hordeins in barley and glutenins in wheat, but there was less knowledge with respect to gliadins in wheat and with respect to avenins in oats.

(see TWA/21/9 Prov., paragraph 13)

72. With respect to the different methods recommended for barley, the TWA finally recommended that the SDS-PAGE method be used but would state in the Test Guidelines that where B-hordeins and C-hordeins only, and no D-hordeins, were concerned, the acid PAGE method may be used as well. Therefore, the TWA proposed that the Subgroup on Cereals should meet again to prepare the following proposals:

(a)	For hordeins:	characteristics on the absence or presence of alleles
(b)	For glutenins:	characteristics on the absence or presence of alleles
(C)	For gliadins:	if possible characteristics on the absence or presence of
		alleles (otherwise study the question further)
(d)	For <b>avenins:</b>	characteristics with band patterns and an agreed method (to be proposed for an intermediate period until more genetic information was available)

In the Test Guidelines, the method would have to be included and, in the case of barley, the full SDS-PAGE method together with a table establishing correspondence with the Acid-PAGE method.

(see TWA/21/9 Prov., paragraph 14)

73. <u>The Committee is invited to note</u> <u>the above information and to consider</u> <u>possible steps to be taken</u>.

### Electrophoresis in Maize

74. The TWA also noted the results of the testing of electrophoresis in maize and the proposal to start with the testing of isozymes. It invited all other member States to join in the planned ring test as laid out in paragraph 21 of document TWA/21/3. Any member State interested in joining the test, should contact the French experts.

(see TWA/21/9 Prov., paragraph 36)

# 75. The <u>Committee</u> is invited to note the above information.

#### Color Measurements

The TWA noted document TWO/25/2, prepared by the Technical Working Party 76. for Ornamental Plants and Forest Trees, on the measuring of color and document TWA/21/7 on the measurement of leaf color in ryegrass, prepared by TWA experts in the United Kingdom. Document TWA/21/7 concluded that the results of the comparison of visual rating with measured color characteristics were very encouraging, since color measurements were likely to be utilized in cases where color differences were observed via recorders in the field trials. It was, however, wrong to use the machine data to establish distinctness and, in that respect, the document supported the proposal, mentioned in document TWO/25/2, to use the measurements as support only, as otherwise there was a risk that one would use machine data to create differences that did not really exist. In visual observations, the observer would view each plant and would form a final impression of the color of that plant which would include the actual leaf color of all leaves including diseased and senescent leaves. The observation would be dependent on the environment and, in general, would be a relative observation rather than an absolute one. Measurements by the machine should only be used to confirm observed differences and be of additional help in establishing the conformity of the variety. For registration purposes, however, there was still a need for the adoption of a standardized method before it could be used for the observation of uniformity. The document had been prepared on the basis of data from one year and therefore the results needed to be checked again.

(see TWA/21/9 Prov., paragraph 23)

77. The TWA, having discussed the possibilities of measuring color for agricultural species, confirmed the position taken by the document that it would only use differences seen by the eye. It could not see much interest in using the measurement of color for agricultural species. However, it would keep an eye on the progress made in the Technical Working Party for Ornamental Plants and Forest Trees and it asked the Office of UPOV to submit document TWA/21/7 to the TWO for information.

(see TWA/21/9 Prov., paragraph 24)

78. The Committee is invited to note the above information.

# Central Computerized Data Base (Item 8 on the Draft Agenda)

The TWPs noted the history of the discussions in the different Technical 79. Working Parties which finally led to the establishment of document TWC/10/2 as a result of a questionnaire issued by the Office of UPOV. They partly also noted the discussions held on the same subject by other TWPs and their proposals to set up a small subgroup which could comprise experts from the different TWPs, as well as an expert with administrative and legal knowledge, to develop a minimum proposal for the checking of variety denominations and another proposal with further items which could be usefully included in a future UPOV data base. Many experts were worried about the cost of setting up and running such a data base. The TWPs held lengthy discussions on the usefulness of such a data base, on its possibilities and on what should be stored in it. It was important to create an efficient system for all member Too much parallel work should be avoided between UPOV and the EC States. which had simultaneously planned to develop a data base for the future EC Plant Variety Protection Office.

80. The TWPs agreed to support the proposal to set up a small subgroup to discuss the minimum content of such a data base. They proposed that, at the beginning, the data base should be limited to administrative information and to the checking of variety denominations. At present, no technical information should be included. The TWPs finally proposed to develop two proposals, one limited to the checking of variety denominations (the minimum information) and another with about 20 further items, and to ask for the cost of these two proposals. The list of items to be included in the UPOV data base as minimum information reads as follows:

# "List of Minimum Information to be Included in the UPOV Central Computerized Data Base

_	Latin Name
_	
-	Application Number
-	Registration Number
-	Country of Application for PBR
-	Country of Application for National Listing
-	Place of Publication (nonprotected, nonlisted varieties)
-	Applicant
-	Breeder
-	Maintainer
-	Breeder's Reference
-	Approved Variety Denomination
-	Proposed Variety Denomination
	Date of Application for PBR
-	Date of Application for National Listing
-	Date of Proposed Denomination
-	Date of Approval of Denomination
	Date of Rejection of Denomination
-	Date of Grant of Protection
-	Date of Inclusion in National List
-	Source of Information
-	Remarks"

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81. Should a subgroup be convened to discuss final items together with experts from WIPO, the TWA proposed that Mr. Bar-Tel (Israel) be made a member of that subgroup and, should he be prevented from attending, Mr. Ghijsen (The Netherlands). The TWC proposed Mr. Grégoire (France) and Dr. Laidig (Germany) and the TWV, Mr. Breuils (France) for election as a member of that subgroup. In addition, it could also be useful to invite an expert from the EC to participate in those discussions.

82. With respect to the maximum delay in the distribution of information, it was proposed that it should be three months at the beginning with the possibility of reducing it to one month at a later time. With respect to the kind of information to be extracted from the data base, there should not be any limitation and it should be possible to combine all information in the data base.

83. The TWPs stressed that, in addition to the discussions on the setting up of a data base, parallel discussions should take place to standardize the national gazettes, especially the information contained in the different sections, to facilitate a future exchange of information or, once a UPOV data base had been established, the inclusion of that information directly in the UPOV Data Base.

(see TWA/21/9 Prov., paragraphs 5 and 6, TWC/10/11 Prov., paragraph 16, TWV/26/11 Prov., paragraph 7)

84. The Committee is invited to take the necessary decisions.

# Cooperation with Breeders in the Testing of Varieties (Item 9 on the Draft Agenda)

The TWA noted the report of the last session of the TWA and the 85. excellent discussion on cooperation with breeders in the testing of varieties which had taken place last year and on the different ways of having the breeder participate in the testing of varieties selected in the different member States. The TWA further reviewed the participation of breeders in the discussion in the Technical Working Parties and Subgroups which had taken place in past years and the need for help in testing by the breeder because of the opening up of plant variety protection to the whole plant kingdom. As the technical experts from the breeders had been nominated to attend the session of the TWA rather late this year, the experts looked for possibilities of increasing the participation of the breeders in their discussion. In this respect, it also pointed to the decision of the Council and the Technical Committee to invite breeders to selected items to be discussed in a given Working Party or Subgroup meeting. It therefore agreed that at the time of planning the coming session, it would take more time to select the items to which breeders would be especially invited to send experts.

(see TWA/21/9 Prov., paragraph 30)

86. The TWA noted that the system and principles for the testing of vegetables and the participation of breeders in the testing was under review at present in the Netherlands. The expert from The Netherlands promised to prepare a document for discussion during the coming session of the TWA.

(see TWA/21/9 Prov., paragraph 31)

87. The TWV had a rather short exchange of views on cooperation with breeders in the testing of varieties, without coming to any conclusions. The subject was obviously of less importance to the testing of vegetable species and the TWV therefore agreed to discontinue discussions on the subject.

(see TWV/26/11 Prov., paragraph 14)

88. The <u>Committee is invited to note</u> the <u>above information</u> and to <u>consider</u> possible steps to be taken.

#### Definition and Examination of Hybrid Varieties (Item 10 on the Draft Agenda)

89. The TWA noted the report on the testing of distinctness of hybrids in France on the basis of the inbred lines and the formula. It noted that this testing was only considered to be a preliminary grouping of the varieties to find out those lines and hybrid varieties which would have to undergo a more precise test. Thus, the testing of the inbred lines would not replace the testing of the hybrid itself.

(see TWA/21/9 Prov., paragraph 35)

90. The Committee is invited to note the above information and to consider possible steps to be taken.

#### Minimum Distances Between Varieties (Item 11 on the Draft Agenda)

The TWC noted document TWC/10/6 on differences between varieties. 91. The document explained the zero hypothesis which meant that when two varieties "a" and "b" were compared, the normal assumption was to consider the two varieties identical. Because of the use of a limited number of plants, a statistical test had to be applied to analize whether the difference between the two random samples was significant (>LSD). It further explained the two types of error that could be made in this respect. The first one was to decide that two varieties were different on the basis of the results of the experiment whereas in reality they were not (the error was called probability level and given the symbol alpha). The second kind of error was to decide that the two varieties were not different, whereas in reality they were (the error was called discriminating power and given the symbol beta). It furthermore gave various alternatives which were used to prevent acceptance of unusually small differences. Those alternatives were:

(i) Standard procedure, use of alpha = 0.01 whether the LSD was small or large;

(ii) Set the minimum value (threshold) for LSD. Normally alpha was set at 0.01, but if that gave an LSD, for instance, smaller than 1.5, then 1.5 was used as LSD;

(iii) Decrease alpha (for example, divide by 2) for the characteristics in which unusually small variances sometimes occured;

(iv) Add a certain fixed value, also called minumum difference, to the LSD. That method was described in paper TWC/VIII/14.

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92. The TWC followed the conclusion of the paper that the alternatives (iii) and (iv) were not acceptable and that alternative (i) was the best and alternative (ii) second best if it was not possible to apply alternative (i). The TWC stated that restricting the approach characteristic by characteristic was wrong. Statistics were only a tool to help in reaching a decision and the decision would have to be taken per variety and not per characteristic. From the statistical point of view, alternative (ii) was the best for making corrections to an LSD value. Statistics could only help in solving the problem of minimum distance. The TWC agreed to ask Mr. Ghijsen (The Netherlands) to prepare a further paper on minimum distance for discussion during its coming session.

(see TWC/10/11 Prov., paragraph 20)

93. <u>The Committee is invited to note</u> <u>the above information and to consider</u> possible steps to be taken.

[End of Annex and of Document]