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# INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS GENEVA

# TECHNICAL WORKING PARTY ON AUTOMATION AND COMPUTER PROGRAMS

# Seventeenth Session Turku, Finland, June 29 to July 2, 1999

# REPORT

adopted by the Technical Working Party on Automation and Computer Programs

# Opening of the session

1. The Technical Working Party on Automation and Computer Programs (hereinafter referred to as "the Working Party") held its seventeenth session in Turku, Finland, from June 29 to July 2, 1999. The list of participants is reproduced in Annex I to this report.

2. The session was opened by Mr. John Law (United Kingdom) who welcomed the participants. He made the presentations around the table and encouraged those participants who were attending the TWC for the first time to ask questions if they had any doubts concerning the subjects under discussion.

# Adoption of the Agenda

3. The Working Party adopted the agenda as reproduced in document TWC/17/1, after having agreed to change its order, as proposed by the Chairman.

<u>Report on subjects of special interest to the Working Party raised during the thirty-fifth session of the Technical Committee</u>

4. Dr. Thiele-Wittig presented a brief report of the main items discussed during the previous sessions of the Technical Committee and apologized that, because of illness and an overload of work, the report had not been finished yet.

5. The Chairman referred to Circulars: U 2828 Revision of the General Introduction to the Test Guidelines; U 2829 Inclusion of technical information in the UPOV ROM; and U 2830 UPOV taxon code.

6. <u>Chairmanship of the Technical Working Party on Automation and Computer Programs</u> The Working Party noted that the Technical Committee proposed to the Council that it elect Mr. Wieslaw Pilarczyk, Poland, as Chairman of the TWC, as the candidate proposed by the Chairman of the TWC after consultation with members of the TWC.

7. The Working Party also noted that the following chairmen were proposed to the Council: TWF: Mr. Josef Harsanyi, Hungary; TWO: Ms. Elizabeth Scott, United Kingdom; TWV: Mrs. Julia Borys, Poland.

8. The Working Party was also informed that the Council had elected Mrs. Buitendag as Chairman of the Technical Committee and Mr. M. Camlin (United Kingdom) as Vice-Chairman. The Council had also elected Mr. M. Camlin as Chairman of the BMT and Ms. Françoise Blouet, France, as Chairman of the TWA.

9. <u>Application of COYD and COYU Analysis</u>: The Working Party was informed that, apart from TWA, other Technical Working Parties refused to use COYD and COYU, because the requirements that should be accomplished to use them were not fulfilled or there were too few cases where it could be applied. Experts also wondered if a method which had been originally created to solve problems in open pollinated varieties could be used on other crops.

10. The Technical Committee had asked about the minimum requirement of 20 varieties, and it was clarified that 20 varieties was only a rough indication which however could be lowered. The Technical Committee was informed that if all the requirements for its use were fulfilled, the COY approach could be used in self-fertilized and vegetatively propagated crops as well, but that experts were able to use another method if they considered that COY was unsuitable for the crop. It was pointed out that the main difficulty could be the lack of experience in the use of the COY approach of the experts involved in the DUS test. Delegates from France and Germany supported the idea that whenever the requirements of the COY approach were fulfilled, this was the best method to handle measured characteristics.

11. Some experts supported the idea that use of the COY approach was compulsory for every species proposed by the Technical Committee could be a misunderstanding, because it was clear that the use of the COY approach was a recommendation from the Technical Committee, and that sometimes this misunderstanding was the result of the status of the UPOV Test Guidelines and other UPOV recommendations in the different countries.

12. As a conclusion on the use of the COY method, the Technical Committee agreed that the COY approach was recommended for cross-fertilized species, but it could also be used in

self-fertilized species and vegetatively propagated materials as well, to handle measurements. When the requirements of the COY approach were not fulfilled, other methods could be used, for example the "t" test, or the LSD. The Technical Committee asked the TWC to consider other methods to be used in this second case, and the technical working parties could propose example situations.

13. <u>Concern over Too Small Differences</u>: The discussions at the Technical Committee meeting focused mainly on the possibility of having different levels of acceptance probability for different species or for different characteristics, and whether they should be included in the Test Guidelines. Some experts mentioned that this would enable them to take decisions without the necessity of a third year of testing. The Chairman of the TWC explained that the COY approach was designed to consider the two out of three testing years principle, and if that was going to be changed, other parameters should be changed. He also explained that if different levels of acceptance probability and population standard were used in different species or characteristics, they should be clearly stated in the Test Guidelines, if not this could lead to a misuse of the COY program with the consequent loss of reputation.

14. <u>Revision of the General Introduction to Test Guidelines, Harmonization of States of Expression and their Notes</u>: The second draft for a revised General Introduction was discussed briefly during two extra days by the Editorial Committee enlarged by the Chairmen of the Working Parties. A second meeting of an *ad hoc* group took place in Geneva, on May 10 and 11, 1999. As a result of that meeting, document TC/35/13 has been produced by the Office of UPOV for discussion during the Technical Working Parties (see item 7 of document TWC/17/1).

# UPOV ROM Plant Variety Database and Taxon Code

15. <u>UPOV ROM plant variety database:</u> The Working Party noted updated information provided by the Office of UPOV on the UPOV ROM Plant Variety Database. In 1998, a new version was issued every two months. It was mentioned that the initial objective of the UPOV ROM had been the use of the information in it for the examination of variety denominations. This meant that some member States had had problems getting data of varieties that were in their local market but were not included in the national list or protected. It was highlighted that it was necessary that member States and organizations which submit information for the UPOV ROM provide more than the minimum required. Concerning the possibility of including technical information in the UPOV ROM, the Office of UPOV explained that some member States have solved some of the problems. On the one hand, there were some technical problems related to the way in which the technical information is processed and stored and, on the other hand, there were limitations in making the information completely available from the legal point of view.

16. <u>Taxon code:</u> Mr. M.- H. Thiele-Wittig explained that the first objective of the taxon code was to solve the problems raised by the different species names used by different national offices.

17. The expert from Israel said that the code was very helpful and he was not in favour of combining the taxon code with agroeconomic groupings. The expert from France considered that the code was good and easy to use and that it was very important for people who worked

with more than one species, like administrative people or denomination examiners. The expert from UPOV was asked if the code would change when the species name changed. He replied that once the code was settled, it would not change whether the name of the species changed or not. He added that it was not a matter of nomenclature, it was a special code for specific purposes for UPOV use only.

18. Discussions were then focused on the technical information that could be included. The first question raised was whether this information should be included in the taxon code or in another code. A second question was on what technical information should be included (use only Technical Questionnaire information or full description).

19. The Chairman of the TWC had already asked the expert from UPOV about this discussion in the TWA meeting the week before. Mr. M.- H. Thiele-Wittig answered that there was a conflict between what experts wanted and the workload involved in achieving that aim. He explained that the TWA had finally agreed to have another code for the agroeconomic groupings, but there was no agreement on the extent of technical information to be included. Opinions varied from having only grouping characteristics or the full variety description.

The expert from Israel said that it would be useful to have information on the varieties, 20. even if it came from the breeder. The expert from France mentioned that to have technical information was an important need but that for France it would not be easy to submit it because it was stored in another way. He supported the idea that the agroeconomic groupings had to be separated in another field, with different options. Most experts agreed to have technical information in a field or code other than the taxon code. Some experts wondered about the possibility of including breeders' information and were of the opinion that only information from the national authority should be included. Other experts wondered what would happen if breeders' information was included and after the DUS trial the official description proved to be different. The UPOV expert explained that in the TWA it had been discussed whether breeders' information should be considered for pending applications while official information was recorded. The Chairman of the TWC suggested using temporary files for breeders' information or for information from the national authority but on provisional status (e.g. first year information). He finally said that when granting plant breeder's rights this information could be moved to final fields irrespective of whether it came from the breeder or from the office.

21. The UPOV expert wondered about the future delay in the UPOV ROM if more information was required from the member States, taking into account the present delay in the information submission from some offices. The Chairman was of the opinion that updates would come on time as the UPOV ROM became more and more involved in the work of the national offices.

22. The Working Party also discussed how to place the technical information. At first, it was proposed to include a first code for information on the use of the variety (e.g. winter type, spring type). Some experts considered that the word "use" was not suitable, because the use of a variety could change from country to country. It was proposed to use the word "type". A second level of information with more detailed data of the varieties was proposed. The Chairman asked what would be the ideal information. The UPOV expert replied that the full description was desirable. Once PBR had been granted the description was public and it

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should be made available, but it was well known that many offices sold the reports of the DUS tests and this would cause some difficulties from a practical point of view.

There were different opinions on the extent of the technical information that should be 23. included in this field. Some experts considered that the information of the Technical Questionnaire of the Test Guidelines would be enough. Other experts pointed out that there were many species without Test Guidelines, mainly in the ornamental group. Free text file would be useful for filing the information on these crops and for those offices that wanted to include further information to that required by the Technical Questionnaire. Other experts wondered if their national offices would agree to provide more information than that included in the Technical Questionnaire because that information was usually sold to other national authorities. The UPOV expert explained that the question of whether to pay or not was another matter, and that it was a mistake to mix both subjects. The experts from Israel and Germany also wondered about the proper use of detailed information included in the UPOV ROM. A compromise was suggested, i.e. to include in the UPOV ROM a few characteristics but to make the full description available to other offices upon request. The experts from Finland added that all the descriptors of the varieties in their country were public and there was no problem in submitting them, other than the workload. The expert from France said that the national offices would have to give up some income if UPOV wanted to include the full description of the variety, but added that experts wanted to have that information.

24. The expert from Korea mentioned that in his country two CD-ROMs with descriptive information on pepper and soybean varieties, including different descriptions of each variety in different places, had been developed. He explained that despite the limited use because of environmental influence national experts found it very useful to have the full descriptions.

25. The Working Party finally agreed to include the technical information in another code different to the taxon code. In respect of the extension of the information to be included in this code, there were different situations in different countries. Some countries supported the idea of including only the information from the Technical Questionnaire of the Test Guideline while others considered that the full description of the variety should be included. In any case, the workload to put the information into a proper format for submission was a problem.

26. A brief discussion took place on the possibility of having a reference number in addition to the taxon code and on whether spaces could cause any problem in short codes. The Working Party concluded that no reference number was necessary and dashes or spaces would cause no problems to the system.

### Image Analysis

27. <u>Flores, a pictorial database for ornamentals using JAVA</u>: The Working Party noted document TWC/17/5. The objective of this paper was to discuss a pictorial database for ornamental crops using JAVA, for pre-screening purposes, prepared by the experts from the Netherlands. The document explained that UPOV member States used a database system for descriptions. Pictorial information was stored in photos, in some cases was linked to other information related to the variety, but there was no use of the pictorial information. It mentioned that there were many attempts to retrieve information from pictorial databases (A

V Photo and Media Finder, QBIC and ARBIRS) but they were still not useful for variety testing.

The system consists of two main parts, the client-site and the server-site. It starts at the 28. client-site, when a user asks for the Flores.html document in his web-browser. The htmldocument, containing the applet (an applet is a small program that can run in a web-browser across the network) is sent to the client. The applet asks the user to load a local image (JPG or GIF). When the image has been loaded, the user should select the part of the picture (e.g. leafs, stem and flower/s), which is to be used for comparison. For this purpose an interactive segmentation of the image has to be performed. The applet has several tools to allow the user to indicate the part (object) of the image that is going to be used. The first tool is based on the thresholding of three-color histograms (for red, green and blue). The user can set upper and lower limits for the three colors. Another segmentation tool is region growing which is comparable to the magic wand in several image-processing programs like Adobe PhotoShop. There is also a standard segmentation tool, which directly selects the correct object, if the object was recorded under fixed conditions. The processed image contains a mask, which only allows the showing of the selected object. After segmentation the user can select different options for matching the image with the images in the database. The options include type of object (flower, plant, leaf, whole image) and type of matching (color histograms, combination of features). Then the image is sent to the server. After the server has processed the data, the applet receives the results of the matching. The results are a set of 25 records, in order of decreasing similarity. Each record contains the variety registration number of the object, the similarity, and a link to the URL-address of the image. The 25 images are downloaded in thumbnail format and shown in a new gallery window. A full size image can also be downloaded and shown in a new window, and subsequently be processed (segmented/matched) as described above.

29. Once the image has been received in the server-site, it is split into an RGB image (the full original image) and a binary mask image. Subsequently the image library of Scil-Image is called up through the JAVA Native Interface, using the color image, the binary image and the matching option as parameters. There are at the moment two options for matching: histogram-based and feature-based. The histogram-based option is a basic option, available for all objects, it is fast and easily calculated but results achieved are limited for accurate matching. It only uses basic color information. The feature-based option is object specific. Depending on the type of object indicated by the user, various features can be calculated, such as area, perimeter, length/width ratio, shape factor (perimeter<sup>2</sup>/(4. $\pi$ .area)), modus of red, green and blue (i.e. the value of the histogram bin with the highest count in the histogram) and the standard deviation of red, green and blue. Standardized recordings are necessary for optimal performance of the system.

30. The calculated features are weighted and used in linear combinations, where the weights and coefficient for the linear combinations are based on a linear discriminant analysis. Those which prove to be the most discriminating features are used for searching their nearest neighbors. The most similar objects in the feature space are found using the SR-Tree algorithm (SR =sphere/rectangle). Finding most similar images using this method is done only within the specified type of object.

31. By allowing user-driven segmentation in the applet, the system can combine the expertise of man in determining the kind of object by controlling the segmentation and the computer capacity for quantitative comparison. Nevertheless, as the system shows the resulting most similar images, the user is always in control of the final decision. Since multiple images of different flowers are available for each variety, the ranking of the most similar of the other images of the **same** variety can be used as a measure of retrieval accuracy. One limitation of the system is that it is not capable of obtaining an overall similarity for a variety. At the moment, the (prototype) database contains images of flowers of eighty rose varieties. At least three flowers per variety are available. The images are recorded under controlled conditions, so within the database all pictures can be compared. The system has been optimized for rose flowers. In the future it will be extended to other flower types and will provide a direct link to a relational database containing the variety information corresponding to the image.

32. The expert from the Netherlands was asked about the shading influence of the petals and the inner petals influence in the color of the photograph. He replied that it is up to the expert to extract the most similar variety. The Chairman said that it would be of interest to the Working Party to have a ring test to see the level of inconsistency and also the standardization required, two points that were of great interest for the group. Finally the expert from the Netherlands explained that one of the most important goals of this program was to have in the desktop computer the program that adapts the photograph to the requirements of the database.

33. Some experts commented on the work on image analysis in their countries. The expert from France mentioned the project that was being developed in his country. It worked for rose also, using color and physical appearance. There was another application on image analysis for seed recognition using the neuronal network approach. The expert from Germany said that in his country there were two projects, which worked with colors and shapes and the new approach enabled the experts from different field testing stations to work with the database in Hanover. The expert from the United Kingdom mentioned the VISOR project for carrots which would be extended to leek. Finally, at the request of the Chairman, the expert from France agreed to prepare a summary report on image analysis for the next meeting of the Working Party. The UPOV expert pointed out that the Working Party had to be clear in the intended use of image analysis, whether it could be used for distinctness, pre-screening or as supporting evidence only.

# Telecommunications, Exchangeable Software and Contacts

34. The expert from the United Kingdom introduced documents TWC/17/4 on developments in telecommunications within UPOV and TWC/17/7 on telecommunications, exchangeable software and contacts. The first document shows developments over the past two years. The links from UPOV web page, the e-mail list address, TWC documents and the JAVA version of the COYD program which allows an exploration of COYD on line. The interest of the TWF in developing its own web page was also mentioned. It was agreed that future web pages of other Technical Working Parties should be inside the UPOV Website. The Working Party regretted the lack of success of the e-mail bulletin board. Some experts considered that it was necessary to set up discussion subgroup to obtain better feedback while others said that this kind of bulletin should be managed by the UPOV Office and then forwarded to the relevant expert. Nevertheless, participants considered that they had limited

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time to answer or analyze problems outside their work. The availability of UPOV documents in electronic format was required by the participants. The UPOV expert explained that it was planned that whole documents would be available in electronic format and that to place incomplete documents in the UPOV web page was unacceptable to the Office.

35. The second document contained the information downloaded from the website http//www.bioss.sari.ac.uk/upov. It included an e-mail list of participants in the different UPOV Technical Working Parties, exchangeable software used by member States, database management systems in use, a COYD on line demonstration and index of TWC papers from 1986 to 1998, ordered by subjects. The Chairman encouraged the participants to look at the web page and said that anyone who wanted more details about the VISOR image analysis program could contact Mr. Mike Talbot (United Kingdom). He also added that it would be very useful if other member States were able to use the VISOR or FLORES image analysis systems with their own data. Some experts asked about the future developments of the UPOV web page. The expert from the UPOV Office replied that there were some options under consideration; one could be to have two parts for UPOV documents, one for free access and the other available by means of a password which would be provided to member States only. The participants were asked to send the web page address of their national offices and other institutions involved in the granting of plant breeders' rights to the UPOV Office in order to make the necessary links. Experts from Denmark, Finland, Netherlands, Republic of Korea and United Kingdom said that their offices had web pages. More countries were invited to supply information and to check the information they had given in the past. Changes and new information should also be sent by e-mail to Mr. Mike Talbot (United Kingdom) (e-mail: The information would also be available on Internet (http:// mike@bioss.sari.ac.uk). www.bioss.sari.ac.uk/links/upov/).

# Subgroup Meeting on Matters from the Working Group on Biochemical and Molecular Techniques and DNA Profiling in Particular (BMT) Related to the TWC

36. On the evening of July 29 experts from France, Germany, Mexico, Netherlands and United Kingdom held an informal subgroup meeting to discuss the request of the BMT to develop statistical methods to assess uniformity when using biochemical and molecular techniques.

37. The first point addressed by the subgroup was to consider on which technique the TWC was going to focus its work. The subgroup noted that in this field of technology, new techniques were launched every year and they had to make a compromise decision between a new technology that could last over time and one that was reliable for plant variety examination. Two techniques were finally selected to work on: microsatellites and AFLPs.

38. The second issue discussed by the subgroup was what species the TWC should test in the work. Considering the technical background available from the BMT and other Technical Working Parties three crops were selected: roses, oil-seed rape and ryegrass.

39. The subgroup also analyzed the present situation on the availability of information to work with. The conclusion was that national offices involved in plant breeders' rights or national listing did not have enough information. The subgroup considered that as

ASSINSEL was working with these techniques for essential derivation assessment it could be a good source of information.

40. The Working Party agreed on the proposal of the subgroup to contact ASSINSEL, the international working group on rye grass and the BMT. The specialist from Germany would contact the Canadian authorities to get information on oil-seed rape. Meanwhile the subgroup proposed that simulated data provided by the Netherlands and the United Kingdom could be used to develop the first generation of statistical tools required by the BMT.

41. The Working Party noted the oral report of this meeting and approved the actions proposed by the subgroup as mentioned in paragraph 40.

# Revision of the General Introduction to Test Guidelines

42. <u>Comments and discussion on the text of TC/35/13</u>: The Working Party noted document TC/35/13 presented by the Chairman and the expert from UPOV. They explained that during the last Technical Committee meeting this item had been forwarded to the Editorial Committee enlarged by the Chairmen of the Working Parties. During that meeting the Chairmen had been asked to obtain comments on the relevant items for their Technical Working Parties. An *ad hoc* meeting was held in Geneva on May 10 and 11, 1999, and as a result of that meeting document TC/35/13 was produced. It was explained that the aim was to have a document containing the general principles and rules for DUS testing that were not included in the individual Test Guidelines. It proposed to prepare a short main document with a complementary explanation in an enlarged document and a series of further documents in an exchangeable folder. Document TC/35/13 was the draft of the document with general principles, enlarged by complementary explanations.

43. The Working Party noted that in Annex II (list of complementary documents) it could be seen which Working Party was involved to a major degree and had to prepare the first drafts. The expert from France required more clarification on the last sentence of the remark on paragraph 36 and the rejection of different degrees of uniformity as a characteristic of distinctness. The discussions focused on paragraphs 43, 44 and 45 on quantitative and qualitative characteristics. Some experts wondered if it was necessary to keep the approach of characteristic type. They said that the proposal for paragraphs 43 to 45 did not help for statistical purposes and it would be better to relate characteristics as ordinal and nominal. Other experts considered the document proposal a good compromise between crop experts and statisticians and that further details could be included in document TGP/8.

44. In paragraph 51 the words "non-even distribution" should be replaced by "non-normal distribution". There were also comments on paragraph 58 and discussion focused on the necessity of considering whether combined characteristics should be "biologically meaningful" or not. The Working Party finally agreed to keep paragraph 58 unchanged but to redraft paragraph 71 as follows:

### "6.8 <u>Combined Characteristics</u>

"71 Cases can arise in which differences between two varieties may be observed in several separately assessed characteristics. If the combination of such data is used to establish distinctness (e.g. length/width ratio, but not any combination of characteristics

without biological meaning), it should be ensured that the degree of reliability is comparable with that provided for measured or normally visually observed characteristics and that uniformity of the combined characteristic is guaranteed."

45. At the request of the UPOV Office for more comments on uniformity assessment in bulk samples (paragraph 59), some experts mentioned the fact that in many cases more than one individual plant was necessary in order to get enough plant material to run the analysis. Other experts added that the high cost of some analyses rendered it impossible to perform repetitions. The expert from the United Kingdom explained that the concept of "conformity" was used in his country; four samples were taken, two used for the analysis and the other two stored and used for further analysis if the first two samples did not show coherent results. The expert from UPOV explained that the groups had to be careful with the concept of "conformity" instead of uniformity, and there was discussion on whether to require only conformity instead of uniformity. The expert from Denmark considered that it was a matter of the risks involved. He said that a paper could be prepared quantifying the risk of taking one plant individually or a plant group, for different options of plant numbers and grouping.

46. The experts made the following comments. They proposed that the assessment of distinctness and the descriptions should be split to avoid misunderstanding. They also considered that paragraphs 65 and 66 should be improved and clarified. In paragraph 81 "the acceptable level of variation" and "which is a further development....." should be deleted. In paragraph 82 "as those which are measured" should be deleted because it meant that a measurement had to be done. The Working Party agreed to keep the remark in paragraph 86 as part of the text of paragraph 85. Finally the last comments on the text were on paragraph 106, the replacement of "still another group" by "the third group"; and on paragraph 107 "third group" by "fourth group".

47. <u>Comments and discussion on the list of complementary documents in TC/35/13</u>: <u>TGP/8</u> <u>Definition of Good Statistical Practices</u>: The Working Party agreed that the document should contain more than definitions, but it should not be a statistical handbook. This document should include data type and handling and possible actions to be taken when ideal assumptions were not met. The Working Party considered that the most urgent matter was to define the chapters. Finally the following chapters were proposed by the TWC:

Chapter I: measured data, checking of the accomplishment of the assumptions, actions and methods when those assumptions were not met.

Chapter II: outliers, adequate randomization, one tail and two tail distributions, sufficient replications and numbers of plants for individual plant recording.

Chapter III: COY approach.

48. The Working Party agreed to have the chapter heading and structure for the proposed meeting on October 1, 1999, ready for further discussion on TC/35/13 and to provide a draft for January 2000 to the Technical Committee to be discussed during its next session. Experts from Germany would prepare the document.

49. <u>TGP/9 Handling of Measured Quantitative Characteristics</u>: The Working Party agreed that the United Kingdom would prepare the document and pointed out that subjects (d) and (e) were already on the TWC web page.

50. <u>TGP/10 Handling of Visually Assessed Characteristics</u>: The Working Party considered that, from the statistical point of view, there were only four types of characteristics, measured and observed, and inside each of these, two different types, ordinal and normal. The experts considered that, from their statistical point of view, these characteristics should be classified as visually assessed, ordinal type or normal type. The expert from Denmark proposed including something about true quantitative, non-true quantitative and qualitative characteristics in a special annex to the main document. The expert from UPOV explained that the task of the Working Party was to provide tools for the crop experts and the classification of the characteristics by the crop experts could not be changed. Finally, the Working Party agreed to have a set of four separate documents dealing on the one hand with distinctness and on the other hand with uniformity; both of them divided into quantitative and qualitative characteristics.

51. <u>TGP /11</u> Non-traditional, Non-morphological Characteristics and Methods for Variety <u>Testing</u>: The Working Party agreed that France would prepare a summary report on image analysis for the next meeting of the Working Party (see paragraph 33 of this document).

52. <u>TGP /18</u> Definition of Technical, Botanical and Statistical Terms Used in UPOV <u>Documents</u>: The Working Party agreed that this document had to be a glossary of terms and that it had to be closely linked to TGP/8.

53. <u>TGP/19 Further Statistical Methods</u>: The Working Party agreed that methods under development should not be included in this document. Subjects (a) and (b) had to represent the state of current knowledge. It was also agreed to change the title of the document to "Other Statistical Methods" and to delete subject (c), which was included in TGP/11.

# Incomplete Block Design

54. Efficiency of incomplete block designs in spring rape and mustard: The expert from Denmark introduced document TWC/17/8. The Working Party noted that these crops were some of the major crops for DUS testing in Denmark, with many candidate and reference varieties grown each year and, at the same time, some difficulties had been encountered in the establishment of distinctness of new candidates. Data from 2 trials (1997 and 1998) with Spring Rape and 2 trials (1997 and 1998) with Mustard have been used. All trials were laid out as  $\alpha$ -designs with 3 replicates. Eleven UPOV characteristics were recorded. The data from each trial was analysed by three models. A (complete block design), B (incomplete block design with fixed block effect) and C (incomplete block design with random block effect). For each of the 3 models (for individual trials) the LSD value for comparing varieties in the COY-D analysis were calculated. The expert concluded that incomplete block design proved to be a better method than complete block design, but not as great as expected and the impact it would make on the decision had not been analysed. He also mentioned that this method implied random distribution while DUS field trials were grouped. He finished by saying that blocks could be treated as different trials but no comparison between groups was allowed.

55. Efficiency of resolvable incomplete block design in DUS trial of French bean varieties. The expert from Poland introduced document TWC/17/2. He explained the experiment on French bean conducted in resolvable incomplete blocks in 1998 at the experimental station at Slupia Wielka. 40 varieties were tested, conducted in two replicates. The experiment was established as resolvable incomplete block design with 10 plots within every block. Both incomplete blocks and varieties within blocks were allocated at random. Five quantitative characteristics were used for the trial: c1 - plant height, c2 - pod length, c3 - pod median width, c4 - pod transverse width, and c5 was calculated as the ratio of pod transverse width by pod median width. He explained that the following analyses of variance were applied for the five characteristics:

(a) Analysis of incomplete blocks results according to field scheme applied,

(b) Analysis of randomized complete blocks (ignoring subdivision of replicates into incomplete blocks),

(c) Analysis of incomplete block results for artificial blocks received after subdivision of every incomplete block consisting of 10 plots into two smaller blocks consisting of 5 plots. It is a kind of post-blocking analysis (post-blocking).

56. The expert concluded that the effectiveness of incomplete block design depended on the characteristic involved, and it was higher for those which were more influenced by the environment.

57. The expert from UPOV mentioned that experts had to be aware that characteristics highly influenced by the environment were not suitable to assess distinctness. The expert from Poland said that there were some papers on alpha design for grouping purposes and that a paper could be prepared for the next TWC meeting. Most experts agreed that alpha design did not mean an automatic decrease in the cost of the trial, but sometimes it could give the same result with less repetitions, which was good for the allocation of big trials, and it was most useful for the space dependent characteristics.

58. The expert from the United Kingdom reported that in his country alpha design had been used for 10 years in sugar beet. The expert from Poland mentioned that pea varieties had been tested using incomplete block design and a paper could be prepared for the use of alpha design and grouping purposes. The expert from Germany reported that alpha design had been used for VCU trials for three years. The expert from UPOV reminded the experts that they should seek a solution for the crop experts. The Chairman of the TWC agreed on that and proposed to keep on studying the possibility of alpha design for DUS assessment, which he considered could be useful when the variety number under trial was very big, when decreasing the cost of the trials became a necessity and when there was not enough space to allocate the field trial. Experts agreed to inform the Technical Committee.

# Reference Collections

59. <u>Reduction of Herbage DUS trial sizes by cyclic planting of the reference collection and</u> <u>analysis by compensated data:</u> The expert from the United Kingdom introduced document TWC/17/11. She explained that the number of varieties included in the reference collections and the requirement of more space to place the field trials were increasing every year. Taking this into consideration, the expert gave different options for consideration. On the one hand, she mentioned the possibility of reducing the number of control varieties, but she considered it could lead to some candidate varieties not being compared with their most similar varieties and, on the other hand, she mentioned the possibility of reducing the number of plants or the numbers of replications, but both options would cause a weakening of the assessment of uniformity. Finally she explained the method of allocating the control varieties to three groups, which meant three groups of varieties which would not be grown in either year 1, or year 2 or year 3. Where data was missing for a reference variety, it was explained that it was compensated by the use of two years of data from before the test period. After PBR were granted those varieties were kept on trial for a fourth year after the three year testing period, they joined the reference collection and were allocated to a group in which growing would be omitted every third year.

60. The expert also explained that to minimize the risk of bias in the initial allocation of control varieties to groups they had been ranked by ear emergence and then allocated to a group. She said that there was likely to be a necessary balance in the numbers belonging to each group and in the future that should be done by transferring control varieties between the groups.

61. The expert explained that for cyclic planting, the matrix was incomplete for the control varieties and that COYU was applied to that matrix without data compensation because for COYU analysis only data from the control varieties within the test period were used to set the uniformity standard of the control varieties. She added that for distinctness purposes compensated data was used and the data had to be modified by a constant value following the joint regression analysis model originally proposed by Digby, P. (1979).

62. The expert mentioned that, from the resulting comparison of the method described above and the COYD and COYU on complete variety by test period year matrices, it was possible to conclude that for uniformity assessment, there were nearly identical results with 103 out of 105 decisions, while for the assessment of distinctness, from the 81 varieties passed by each approach, only 77 had been in common and 8 had been classified differently. She explained that the LSD tended to be smaller using cyclic controls with compensated data approach.

63. The expert concluded that a small reduction in stringency of distinctness and a slight increase in the stringency of uniformity were expected because of a slight overcompensation in using two years' past data for one year's missing from the test period in the distinctness testing, and the reduction in the information on the controls used to compile the uniformity standard in the uniformity testing. Finally she said that the national authorities of the United Kingdom had accepted the change to the new approach.

64. <u>Another method to reduce the reference collection</u>: The expert from the Netherlands made a presentation on a method to reduce the number of reference varieties under trial. He explained that in the Netherlands the reference collection for ryegrass had 190 varieties, it had one block, 20 plants per plot, 9 characteristics were evaluated and it took 3 years to list candidate varieties.

65. The Working Party was informed that the method consisted in measuring the variability of every characteristic in the collection Each one was then divided, according to a "yard stick" calculated on the bases of the visually assessed characteristics. As a result, the reference collection appeared divided into blocks with some varieties in each one of those blocks. The varieties were allocated to each block according to a randomization of the reference collection every three years. The expert said that the method assured that all variations in the reference collection would have been covered at the end of the trial period. The expert remarked that the method was focused on important characteristics, that it has potential use in a 1-year evaluation and that the reduction of the reference collection could range from one third to two thirds. He finally added that it was very important that the characteristics evaluated had to be independent in order to avoid measuring the same thing twice.

66. The Working Party asked the expert about the assessment of uniformity using that method. It was informed that uniformity was not assessed. The expert from UPOV stressed that uniformity had to be considered in every new method under evaluation. The Chairman mentioned that it was very useful to have some information from 1-year trials in order to inform the applicant how things were going. The expert from France said that other countries had different approaches to dealing with the problem of handling a big reference collection, in some cases previous information provided by the breeder was used for identifying the most similar varieties to be sown in the trial, but he regretted that it was difficult to lay down this approach in a paper.

# Application of a Threshold Model on a Number on UPOV Characteristics

67. The expert from the Netherlands introduced document TWC/14. He said that the objective of the document was that, by showing the results of the application of a threshold model to visually observed characteristics, it could provide a better understanding and lead to a reconsideration of the notes that had to be used.

68. Before discussing the core of the document, some comments about the types of the characteristics were given to the Working Party. It was mentioned that from the statistical point of view, two types of characteristics existed, quantitative and qualitative. Within quantitative characteristics there were continuous and discontinuous characteristics; and within qualitative there were ordinal and nominal characteristics. Some experts in the Working Party agreed that, on the one hand, in many cases it was necessary to count in round figures (e.g. number of leaves) for the assessment of a characteristic and, on the other hand, measured characteristics rather than round figures could be recorded. The expert from UPOV said that from the crop experts' point of view a quantitative discontinuous characteristic was a contradiction and that, by definition, quantitative characteristics had a continuous variation. He pointed out that crop experts wanted to treat those types in the same way (see also paragraph 50).

69. Discussions then focused on document TWC/17/14. The Working Party noted that the same information as for TWC/15/14 had been provided by France and Denmark. The data from France comprised the tendency to form inflorescence in the year of sowing in tall fescue and cocksfoot. The data from Denmark comprised the following characteristics:

Species	Characteristic	<u>UPOV N°</u>
Peas	Stipule: maximum density of flecking	34
Ryegrass	Plant: growth habit in autumn	2
	Leaf: color	5
Spring rape	Leaf: green color	4
	Leaf: lobes	5
	Leaf: dentation of margin	7
	Flower: color of petals	12
	Tendency to form inflorescences in year of sowing	22
Timothy	<b>Plant</b> : growth habit in 2 <sup>nd</sup> year before elongation	5

The document recalled that the threshold model assumed that there was an underlying 70. variable that determined in which category an individual plant lay and assumed that the variable followed a distribution (normal or logistic). For this paper, it was assumed that all the varieties had the same variation that followed a logistic distribution. Results for the characteristic "Tendency to form inflorescences in the year of sowing" in tall fescue showed that category 4 was very narrow and as a consequence the distance between 2 and 6 was much smaller than between 4 and 8. The same characteristic for cocksfoot gave a similar picture. It was concluded, for that characteristic, that it would be better to chose time-points closer to visiting the trials. The other characteristics had been studied in the same way. Finally the Working Party noted that the analysis had been done considering constant variances for all varieties. Further analysis could be done expanding the model with variances for each variety limited to analyzing not more than 20 varieties in a single run. In that case, results were comparable to those of analyses of measured characteristics. As a conclusion of the document, the Working Party noted that a characteristic had to be recorded on an ordinal scale and the variable had to be unimodal to be analyzed by threshold model. The results were analogous with those of measured characteristics and they could be used to consider the relocation of the notes.

71. The expert from UPOV pointed out that when Technical Working Parties do not set up the Test Guidelines in the right way it is due to errors by the national experts on dividing the characteristics. The Working Party concluded that threshold methods were good to establish whether the experimental division of a characteristic had to be reconsidered when revising the Test Guidelines of a given species and that it was necessary to keep on studying those methods.

# Handling Visually Assessed Characteristics

72. The expert from Germany introduced the document TWC/17/6. He explained that the document was a summary of the TWC documents dealing with different statistical methods for visually observed characteristics. The presentation focused on the type of characteristics and the method that could be used for its evaluation. He said that characteristics used for DUS assessment could be classified according the following table:

Type of scale	DESCRIPTION	Discrete/ Non-discrete	Example
Ratio scale (quantitative)	classes of fixed size and with exact zeropoint	Non-discrete	Absolute measurements (plant height)
		Discrete	Counts (number of plants)
Interval scale (quantitative)	classes of fixed size and without exact zeropoint	Non-discrete	Relative measurements (temperature in C)
		Discrete	Date
Ordinal (qualitative)	independent and non- exchangeable classes, different size of classes	Discrete	Attitude: erect=1, semi-erect=3, intermediate=5, semi-prostrate=7, prostrate=9
Nominal scale (qualitative)	independent and exchangeable classes, different size of classes	Discrete	Color of ear: white=1, colored=2

73. Then a report on suitable methods for the assessment of distinctness and uniformity for each type of characteristic followed, which can be resumed in the graphics as follows:

TYPE OF SCALE	DISCRETE OR NON DISCRETE EXPRESSION	CONDITION FOR DISTINCTNESS PROCEDURE			
		Degrees of freedom <20	Degrees of freedom >20	Unimodality	Other
Ratio scale (quantitative)	Non-discrete				
	Discrete	Long term LSD or 2*1 %	COY-D	????	????
Interval scale (quantitative)	Non-discrete				
	Discrete				
Ordinal (qualitative)	Discrete	Minimum distance >= 1		Threshold ??	???
Nominal scale (qualitative)	Discrete	Minimum distance = 1		???	???

# (i) <u>Methods for the assessment of distinctness</u>

# (ii) Methods for the assessment of uniformity

TYPE OF SCALE	DISCRETE OR NON DISCRETE EXPRESSION	CONDITION FOR UNIFORMITY PROCEDURE				
				Unimodality	Binary data	
Ratio scale (quantitative)	Non-disc	crete	2 from 3 years	COY-U		LSD for untransformed percentage of off-types
	Discrete		$1.6 \text{ s}^2$			
Interval scale (quantitative)	Non-discrete Discrete					
Ordinal (qualitative)	Discrete				Threshold ??	???
Nominal scale (qualitative)	Discrete c> 2				???	
		C< 2	Off-type procedure (binomial)			

74. The Working Party noted that it was necessary to agree on a denomination for the characteristics between statisticians and crop experts and they had to look for solutions still lacking for some situations. The expert from UPOV reminded the Working Party that it had to provide solutions for the different situations required by the crop experts and common denomination of the characteristics had to be agreed. Some experts considered that crop experts had to realise that, according to what was presented above, one group of characteristics would need more than one method. Other experts considered that there had to be a document in which both terminologies were explained. The Chairman considered that TWC/17/6 was a good document which explained the different types of characteristics and showed the solutions still pending. Some experts suggested to the Working Party that they follow up with some steps to deal with that matter. The first could be the comparative tables proposed in the present document, a second step to handle both definitions (crops experts and statisticians), a third step to prepare an explanatory document and the last one to place everything in the web in a way that enables to moving easily from one approach to another. The Working Party agreed to incorporate this section in the future document TP/8 of the General Introduction to the Test Guidelines and to circulate the participants at the meeting for their comments.

### Problems on Early Decision-making in DUS Testing

The expert from the United Kingdom introduced document TWC/17/10. The Working 75. Party noted that the main objective of the paper was to explore some of the statistical issues concerned with distinctness and uniformity testing when there was a wish to make a decision after only one sowing, mainly when handling measured characteristics. Two possibilities had been considered in the document: (1) decisions from a single sowing, and (2) decisions from sowings at two centers. For the first situation, it was explained that two main issues had to be considered when taking decisions on a one-year trial; one was that LSD had to be calculated from long term experience with the variability of the test and, the other, that when establishing a distinctness standard one would like to be confident that a distinctness decision would not change if information became available from later sowings. It was mentioned that one strategy had been proposed in document TWC/13/7. The expert presented an example using an LSD of 0.1 % instead of 1% to provide robustness against the variability that could occur in some trials. It was considered important to set high standards to ensure that a large difference in one year could not be reduced to non-significance after a further year. For the second situation, the Working Party noted the two options for combining the information proposed. One method could be to average variety means over the two centers and compare the average differences between varieties against a LSD based on the variety-by-center interaction, but that method would not satisfy the UPOV requirement for a variety to "be considered distinct if the difference has been determined in at least one testing place". Other disadvantages had been mentioned, such as defining how diverse centers must be in order to represent separate environments or the fact that averages over centers can dilute variety differences which exist in one center but are weak or absent in the other center. The other method for the decisions from sowings at two centers proposed was to make decisions separately for each center, but a difference could only be considered "clear and consistent" if it was observed in both centers, otherwise a second year of growing was necessary.

76. For the assessment of uniformity the expert explained that consideration had to be given as to how uniformity is to be assessed from one sowing. In order to achieve a uniformity

standard that was similar to that of a multi-sown test the probability level applied in the combined-over-years-uniformity (COYU) criterion might have to be adjusted. The principle of adjusting the COYU probability to allow decisions after 2 or 3 years had already been established, and for the case of two-center testing it seemed logical to seek uniformity at the center where distinctness had been established.

77. Some experts commented on the experiments that were being performed in their countries. The expert from Germany explained that they were trying to change from three-year trials to two-year trials in two locations for rye. The expert from France said that for many crops in his country there were two trials, one as principal and the second as backup. He added that differences between locations could cause more differences than between years. The Working Party agreed that there could be two different approaches. One could be the aim to take decisions after one year, and the other to make decisions in two years instead of three. Some experts said that it was not only a matter of reducing costs for the office, but for the breeder to get a quicker result also. The expert from UPOV explained that it took a long time to agree on one single testing place within UPOV member States and the TWC was now going in the other direction. The Working Party agreed that it was not its aim to change accepted UPOV rules, but to provide solutions for special situations and in that sense it was very useful to discuss new options.

### UPOV Questionnaire Concerning DUS and VCU Databases and Computer Systems

78. The expert from Poland introduced document TWC/17/3. The Working Party noted that answers from 16 countries had been collected concerning DUS testing and VCU testing. The information was considered surprising and the increase of the use of personal computer networks was highlighted. The Working Party agreed to repeat the questionnaire every two years. Some experts considered that harmonization was an important issue for the member States and it would be more relevant as the number of varieties grew and exchange of reports became more frequent.

# DUST for Windows (DUSTNT)

79. The Working Party noted the document TWC/17/9 and welcomed the new version of the DUST program for Windows. The expert from the United Kingdom explained that the new program had an easy-to-use interface and the possibility of converting Excel files to ASCII files as used by the program. She said that the minimum specification PC needed to run the software was a 486 DX processor with 36 Mbytes memory and that it would run under either Windows 95 or Windows NT. By contacting Dr. Sally Watson, Biometrics Division, Department of Agriculture for Northern Ireland, Newforge Lane, Belfast BT9 5PX, UK, Tel: (44) 1232 255292, Fax: (44) 1232 681216, e-mail: sally.watson@dani.gov.uk, the DUSTNT system could be obtained in three ways:

- through an FTP site from which the system may be downloaded via the Internet (www),
- by e-mail
- by conventional land mail using floppy disks

80. For the future, it was planned to develop an introductory tutorial and a user manual. The program is available only in English. The office offered to enquire about translation of the text files for the development of the three other language versions.

### Reports on New Developments in Member States

81. The Working Party received from some of its experts' short reports on recent developments in their countries.

82. The expert from Hungary presented a method on special applications of DUS variety descriptions as reproduced in document TWC/17/12. The aim of the method was to reduce the number of varieties included in DUS trials. He proposed forming groups of varieties where the members of each group were more similar to each other than to varieties belonging to other groups and to look after the most representative variety of each group. These representative varieties should be the ones to be included in DUS testing and in this way a reduction in the number of varieties under test could be achieved. He added that a program package had been developed at the National Institute for Agriculture Quality Control. He explained that for the calculation of similarities, every variety was compared to each other in every characteristic. According to the difference in the states of expression, different figures were given, ranging from 10 when there was no difference between the two varieties in a given characteristic, to 1 when the difference was greater than 0 (zero) but less than the DUS threshold. When the difference was greater than the DUS threshold, the figure given was 0 (zero). All these figures were added together. The maximum sum possible was 10 x Number of characteristics. Then the similarity was calculated by relating the sum of a given pair to the maximum possible (100 %). Finally the distance was assessed as follows:

Distance = 100 % - Similarity.

The expert said that the general formula could be modified by taking into account the distinctness thresholds and weight of the characteristics. He added that a given value of % distance should be taken (L= the distance threshold) and that the variety pairs having less distance value could be selected, and similarity groups of varieties could be built up with the varieties for a given distance threshold (L). Similarity groups were then represented by the variety that had the greater number of similarity connections in the group. The assessment of the threshold distance (L) was a very important issue. For this task, a method of comparative histograms between the real descriptive data set and a control variety description set created on random bases was planned. By relating the number of varieties involved in similarity groups to the total number of varieties under test, it was also possible to calculate the "similarity ratio of varieties" of a given crop.

83. The expert from Mexico reported that in his country there was a Technical Committee which is formed by ten members from the National Government, Research Institutes, Biomolecular Laboratories and Breeding Associations. He explained that the task of that committee was to establish criteria and norms for DUS assessment, the use of biomolecular techniques and database management. He also informed the Working Party about national developments on isozyme profile use for studying diversity, for characterization, for the assessment of similarities and for selection purposes in maize, dry bean, pepper, *Opuntia sp.* and squash. The expert from France informed the Working Party that COYD might be used for peas in the coming years. He reported that a new database structure had been set up that

enabled better handling of the information and that in his country they were moving towards a renewal of the technical part by developing new software. He also mentioned that the European Union had developed a new program to put the European Catalogue into electronic format. The expert from UPOV mentioned that the UPOV Office had received authorization to include the European Catalogue in the UPOV ROM in pdf format. The expert from the United Kingdom said that in the near future the COY approach would be used in more crops to get the benefits of reduced trials. The Chairman of the Working Party encouraged the participants to attend the next BMT meeting.

### Future Program, Date and Place of the Next Session

84. At the invitation of the expert from Ukraine, the Working Party agreed to hold its eighteenth session in Kiev, from June 12 (9 a.m.) to 15, 2000 (5 p.m.). The Working Party also considered the possibility of having a workshop on data handling at the same place, on June 9 and 10, previous to the next meeting for the countries of the region. During the eighteenth session, the TWC plans to discuss or rediscuss the following items:

(a) Report on subjects of special interest to the Working Party raised during the thirtyfifth session of the Technical Committee

- (b) Questions raised by other Technical Working Parties
- (c) Report on new developments in member States (oral reports)
- (d) UPOV ROM Plant Variety Database (oral report)
- (e) Revision of the General Introduction to Test Guidelines

(f) Summarized paper on image analysis (the expert from France to prepare a document)

(g) Spatial dependency and plant resources (the expert from the United Kingdom to prepare a document)

(h) Long-term alpha design trial on sugar beet (experts from the United Kingdom to prepare a document).

(i) Update paper on alpha design (the expert from Denmark to prepare a document)

(j) One year of alpha design compared to 2 and 3-years approach (the expert from the United Kingdom to prepare a document)

(k) Incomplete block design on peas (the expert from Poland to prepare a document)

(1) Alpha design considering variety grouping (the expert from Poland to present a document)

(m) New document on the result of the simulation study in ryegrass (experts from the Netherlands to prepare a document)

(n) Use of COY-D and COY-U approach in more than one location in forage crops (the expert from France to prepare a document)

(o) Items resulting from the fifth session of the Working Group on Biochemical and Molecular Techniques and DNA Profiling in Particular (BMT) (report by the Chairman)

(p) Telecommunications, exchangeable software and contacts (the expert from the United Kingdom to receive updated information and prepare updated versions of documents)

(q) Developments in the World Wide Web (the expert from the United Kingdom to report on the experience of the e-mail discussion group)

(r) List of statistical documents prepared by the TWC (the expert from the United Kingdom to prepare updated lists on the Internet to be downloaded four to six weeks before the session for printing and distribution on paper)

(s) List of statistical documents containing recommendations or methods of possible interest to the Technical Working Parties

(t) The paper of DUS Special Tests (the expert from the United Kingdom to prepare a document).

# Special Acknowledgement

85. The Working Party noted that Mr. Max-Heinrich Thiele-Wittig was going to retire and that this was the last meeting in which he would participate. The Working Party thanked Mr. Thiele-Wittig for his excellent work during the 26 years he has been working for UPOV and wished him a happy retirement.

# Visits, Demonstrations

86. In the afternoon of June 30, 1999, the Working Party visited the Boreal Plant Breeding Company, at Jokioinen (about 1 hour from Turku). It was explained that Boreal Plant Breeding was founded at the beginning of 1991 through the merger of the plant breeding of the Agricultural Research Center of Finland and Hakkija as well as Jokioinen seed center and that it was an entirely government-owned business enterprise. The Working Party visited the glasshouses and laboratories for plant breeding.

87. This report has been adopted by correspondence.

[Annex follows]

### TWC/17/13

# ANNEX

## LIST OF PARTICIPANTS

### I. <u>MEMBER STATES</u>

### **BULGARIA**

Mihaela Ivanova IORDANOVA (Mrs.), State Variety Testing Commission, 1a, Mednikarska str., Sofia 1040 (tel. +359 2 393 208, fax +359 2 393 208, e-mail: mihaelaio@hotmail.com)

Kristian Mirolubov MARKOV, State Variety Testing Commission, 1a, Mednikarska str., Sofia 1040 (tel. +359 2 393 208, fax +359 2 393 208)

### CZECH REPUBLIC

Lydie ČECHOVÁ (Mrs.), Central Institute for Supervising and Testing in Agriculture, ÚKZÚZ, 56901 Hradec Nad Svitavou (tel. +420-461 535 003, fax +420-461 22 748, e-mail: cechova@ooz.zeus.cz)

Tomáš ŠEVČÍK, Central Institute for Supervising and Testing in Agriculture, ÚKZÚZ, Hroznová 2, 656 06 Brno (tel. +420-5-435 48 359, fax +420-5-432 17 655, e-mail: sevcik@zeus.cz)

### **DENMARK**

Kristian KRISTENSEN, Danish Institute of Agricultural Sciences, Post Box 50, 8830 Tjele, (tel. +45-89 99 12 09, fax +45-89 99 1200, e-mail: kristian.kristensen@agrsci.dk)

### **FINLAND**

Kaarina PAAVILAINEN (Ms.), Plant Production Inspection Centre (KTTK), Seed Testing Department, P.O. Box 111, 32201 Loimaa (tel: +358-2-760 56 247, fax: +358-2-760 56 220, e-mail: kaarina.paavilainen@kttk.fi)

Leena PIETILÄ (Ms.), Plant Production Inspection Centre (KTTK), Seed Testing Department, P.O. Box 111, 32201 Loimaa (tel: +358 2 760 56216, fax: +358 2 760 56 222, e-mail: leena.pietila@kttk.fi)

# TWC/17/13 Annex, page 2

### FRANCE

Sylvain GRÉGOIRE, GEVES, INRA, La Minière, 78285 Guyancourt Cedex (tel. +33 -1-30 83 36 00, fax +33-1-30 57 01 47, e-mail: sylvain.gregoire@geves.fr)

## **GERMANY**

Uwe MEYER, Bundessortenamt, Osterfelddamm 80, 30627 Hannover (tel. +49-511-95 66 689, fax: +49-511-56 33 62, e-mail: uwe.meyer@bundessortenamt.de)

Beate RÜCKER (Ms.), Bundessortenamt, Osterfelddamm 80, 30627 Hannover (tel. +49-511-95 66 650, fax: +49-511-56 33 62, e-mail: beate.ruecker@bundessortenamt.de)

### **HUNGARY**

Zoltán VERESS, National Institute for Agricultural Qualification, Keleti K. u. 24, 1024 Budapest II (tel. +36-1-212-3127, fax +36-1-212-5800, e-mail: veressz@ommi.hu)

### **ISRAEL**

Baruch BAR-TEL, PBR Testing Unit, Volcani Center, POB 6, Bet Dagan, 50 250 (tel. +972-3-968 669, fax: idem, ilpbr\_tu@netvision.net.il)

# JAPAN

Kazuto HIGASHIMURA, Examiner of Variety Characteristics, National Center for Seeds and Seedlings, Ministry of Agriculture, Forestry and Fisheries, 2-2, Fujimoto, Tsukuba-shi, Ibaraki 305-0852 (tel. +81-298-38-6584, fax +81-298-38-6583, e-mail: niwayuji@ncss.go.jp and higashim@ness.go.jp)

### <u>MEXICO</u>

Porfirio RAMIREZ VALLEJO, Servicio Nacional de Inspección y Certificación de Semillas (SNICS), (National Service of Seed's Inspection and Certification), Lope de Vega 125-20. piso, Chapultopec Morales, 11570 México D.F. (tel. +52-5-203 9427, fax +52-5 250 6483, e-mail: eduardo.benitez@sagar.gob.mx and ramirez@calpos.calpos.mx)

### TWC/17/13 Annex, page 3

### **NETHERLANDS**

L.C.P. KEIZER, CPRO-DLO, Postbus 16, 6700 AA Wageningen (tel. +31-317-477 063, fax +31-317-418094, e-mail: l.c.p.keizer@cpro.dlo.nl)

Jac THISSEN, CPRO-DLO, Postbus 16, 6700 AA Wageningen (tel. +31-317-476 936, fax +31-317-418 094, e-mail: j.t.n.m.thissen@cpro.dlo.nl)

### POLAND

Wieslaw PILARCZYK, COBORU, Research Center for Cultivars Testing, 63-022 Słupia Wielka (tel. +48-61-285-2341 ext. 224, fax +48-61-285-3558, e-mail: wpilar@owl.au.poznan.pl)

### **RUSSIAN FEDERATION**

Svetlana MILOVANOVA (Ms.), State Commission of the Russian Federation for Selection Achievements Test and Protection, Orlicov per., 1/11, 107139 Moscow (tel.+ 95-204-49 26, fax ++95-207-86 26, e-mail: desel@agro.aris.ru

#### UKRAINE

Oleksandr ARISTOV, State Commission of Ukraine for Testing and Protection of Plant Varieties, 9, Suvorov street, Kyiv 252010 (tel. +38-044-290 3191, fax +38-044-290 3365, e-mail: aristov@vartest.kiev.ua)

Anatoliy BOCHKAROV, State Commission of Ukraine for Testing and Protection of Plant Varieties, 9, Suvorov street, Kyiv 252010 (tel. +38-044-290 3191, fax +38-044-290 3365, e-mail: anb@vartest.kiev.ua)

#### UNITED KINGDOM

John LAW, National Institute of Agricultural Botany (NIAB), Huntingdon Road, Cambridge CB3 OLE (tel. +44-1223-276 381, ext. 2254, fax +44-1223-277 602, e-mail: john.law@NIAB.com)

Sally WATSON (Ms.), Biometrics Division, Department of Agriculture for Northern Ireland (DANI), Newforge Lane, Belfast BT9 5PX (tel. 44-1232 255 292, fax +44-1232-681 216, e-mail: sally.watson@dani.gov.uk)

### TWC/17/13 Annex, page 4

### II OBSERVER STATES

#### **ESTONIA**

Pille ARDEL (Mrs.), Head, Variety Control Department, Plant Production Inspectorate, Viljandi Variety Testing Centre, 71024 Viljandi (tel. +372 43 34 650, fax: +372 43 34 650, e-mail: pille@matti.ee)

### GREECE

Miltiadis LAIDIS, Variety Research Institute, 57400 Sindos, Thessaloniki (tel. +30-31-799 684, fax: +30 31 799 392, e-mail: varinst@spark.net.gr)

### **REPUBLIC OF KOREA**

Chi Hong AHN, National Seed Management Office, 433 Anyang 6-dong, Anyang, Kyonggido 430 016 (tel. +82-343-446-2432, fax +82-343-448-1216, e-mail: chahn@seed.go.kr)

Jong-Soo SHIN, National Seed Management Office, 433 Anyang 6-Dong, Anyang, Kyonggi-Do 430 016 (tel. +82-331-240-3675, fax +82-331-240-3677, e-mail: mrshinjs@unitel.co.kr)

### III. OFFICER

John LAW, Chairman

### IV. OFFICE OF UPOV

Raimundo LAVIGNOLLE, Senior Program Officer, 34, chemin des Colombettes, 1211 Geneva 20, Switzerland (tel. +41-22-338 9565, fax +41-22-733 0336, e-mail: lavignolle.upov@wipo.int)

Max-Heinrich THIELE-WITTIG, Senior Counsellor, 34, chemin des Colombettes, 1211 Geneva 20, Switzerland (tel. +41-22-338 9152, fax +41-22-733 03 36, e-mail: upov.mail@wipo.int)

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