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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS
GENEVA

TECHNICAL WORKING PARTY FOR FRUIT CROPS

Twenty-Eighth Session
Wageningen, Netherlands, September 8 to 12, 1997

REPORT

adopted by the Technical Working Party for Fruit Crops

Opening of the Session

1. The twenty-eighth session of the Technical Working Party for Fruit Crops (hereinafter referred to as "the Working Party") was held at Wageningen, Netherlands, from September 8 to 12, 1997. The list of participants is presented in Annex I to this report.

2. Mr. Joost Barendrecht welcomed the participants to Wageningen. The session was opened by Mr. Chris Barnaby (New Zealand), Chairman of the Working Party.

Adoption of the Agenda

3. The Working Party adopted the agenda for its twenty-eighth session, which is reproduced in document TWF/28/1 Rev., after having agreed to slightly change the order of the items for discussion.

Short Reports on New Developments in Member States in Plant Variety Protection for Fruit Species (oral reports)

4. The Working Party received short reports from some of the experts on recent developments in their countries. The experts reported that in several countries preparations were under way to amend the laws to conform to the 1991 Act of the UPOV Convention. In some countries, new laws have already entered into force which conform with the 1991 Act. In several countries, applications for fruit varieties were only very few, in some European countries they had been further reduced as a result of the starting of the European Union (EU) protection system. The main species for which in the different countries applications had been received were apple, apricot, black and white current, cherry, citrus, grape, kiwifruit, nectarine, pear, peach, plum and strawberry. The expert from the EU reported that since the starting about two and a half years ago, about 5,500 applications had been received and about 2,200 titles granted. About 350 applications had been for fruit varieties. In Australia, a few applications for varieties of native fruit species had been received. In New Zealand, applications with resistance/tolerance against physiological disorders as the only distinguishing characteristic had been received.

Questions on the Testing of Varieties of Fruit Species

5. The Chairman explained that this item had been included on the agenda to offer the possibility to raise questions on certain problems which would, if possible, either be answered immediately or included as agenda item for the coming session.

6. The expert from South Africa raised the problem of preparing Test Guidelines for a genus with many groups (e.g. citrus) where especially for the size of fruits only the observation of a relative length, width or size compared to another length, width or size could be requested. Similar problems existed in *Prunus*. It might be necessary to prepare separate documents for the different groups but then the problem existed where to place hybrids between the groups. This problem apparently existed only in fruit species and not in ornamentals. Several experts agreed that separation into several documents might be preferable even if doubtful varieties would have to be tested according to two different documents.

7. The expert from Italy reported that for the digital imaging of the apple shape a problem was what to consider a representative fruit. It was difficult to find the right sampling scheme and the right number of fruits to be observed.

8. The expert from Slovakia raised the problem of finding the right example varieties when starting testing on a new species, especially the obtaining of plant material of older varieties no longer available on the market. The Chairman recalled that the example varieties in the UPOV Test Guidelines represented mainly the example varieties applicable in the country of the expert who drafted the Test Guidelines. In other more distant countries the experts would have to search for other example varieties of that region. The best starting point would be example varieties used in the neighboring countries.

Important Decisions Taken During the Previous Sessions of the Working Party and the Technical Committee

9. Mr. M.-H. Thiele-Wittig gave a brief report on the main items discussed during the previous session of the Technical Committee and referred participants needing further details to the full report reproduced in document TC/33/11.

10. The Working Party noted the latest stage of preparation of the UPOV Plant Variety Database on CD-ROM (UPOV-ROM) as set forth in Circular U 2554 dated July 16, 1997, distributing the third disc in 1997. The Office of UPOV aimed at issuing an updated disk every second month. Discussions were under way to include in the UPOV-ROM the OECD List, the European Union Catalogue and the List of Varieties from the Community Plant Variety Office of the European Union. Discussions with respect to the OECD List had been completed, and the UPOV-ROM to be distributed in November 1997 would contain the whole OECD List. It was expected that the UPOV-ROM would comprise several improvements before the end of the year and especially enable its use on a local network. It was also expected that in a few months it would be offered to the private sector at an annual subscription price of 750 CHF.

11. Several experts had had a chance to study the UPOV-ROM and expressed their satisfaction. The Working Party invited all the experts to contact their respective colleagues at national level for them to also see and assess the information on the disc and make any comments for further improvement. As several experts had not seen the UPOV-ROM, Mr. Thiele-Wittig gave a short demonstration of the content of the UPOV-ROM with its three parts, the combined database with the taxon information, the text part in pdf (portable document file) format with information from the member States on their data, all texts of the different Acts of the UPOV Convention, the Recommendations on Variety Denominations, the General Information Brochure, the lists of addresses of national PVR Offices, the list of UPOV publications and various other information and the part (password protected) containing the original data from the member States.

12. Trade names: The Working Party noted that the Technical Committee and the Administrative and Legal Committee, during its session on October 21, 1996, decided against the inclusion of the trade name in the Technical Questionnaire.

13. Definition of off-type, admixtures: The Working Party noted that the Technical Committee had considered that the definition of off-type was not clear. The previously prepared word "significant" had a statistical connotation and also, significance in leaves is different from that in fruits. The word "clear" was restricted more to what can be seen visually, while "significant" includes much more than seeing. It therefore seemed better to stay away from both "clear" and "significant" and search for a more general definition which, if need be, could differ depending on the genus or species under study. It was important to point out that the work aimed to distinguish a variety, so the word to be chosen should be considered in relation to distinctness.

14. The Working Party also noted that the Technical Committee noted the different positions on the concept of admixture in relation to off-type. It was mentioned that an admixture was a plant which did not belong to the variety and was clearly not an off-type. In other words, a barley seed within wheat was an admixture which might have been caused by mixing or in other ways, while an off-type belonged to and came from the variety through a

genetic difference expressed in the phenotype. The Working Party was reminded of the definition of admixture described in the General Introduction to Test Guidelines (TG/1/2), where admixture is included in off-types.

15. The Working Party further noted that the Technical Working Party for Ornamental Plants and Forest Trees (TWO) had discussed the question of off-types and admixtures and had come to the following new definition: "Any plant is to be considered an off-type if it differs in the expression of any characteristic, of the whole plant or of part of the plant, from that of the variety, taking into consideration the particular species. An admixture is considered to be an off-type."

16. The Working Party could agree to the first sentence of the definition from the TWO but two experts had difficulties with the last sentence. They agreed that admixtures should be treated the same way as other off-types and their number should be included in the number of off-types tolerated, but they had difficulty in calling them "off-types." The Working Party therefore proposed a rewording of the last sentence as follows: "An admixture has to be considered an off-type."

17. Transgenic/GM varieties: The Working Party noted that the Technical Committee reconfirmed its decision to include in the Technical Questionnaire of the Test Guidelines for Rape Seed and in future in other relevant Technical Questionnaires, a broad question whether the variety would "require authorization for release under legislation concerning especially the protection of the environment, human and animal health in the country in which the application is made" and whether such authorization had been obtained. The question was not intended to be limited to GM varieties but to elicit information where appropriate on other restrictions on release. The CAJ during its session held on October 21, 1996, decided to amend the text as follows:

"4.3(i) Does the variety require prior authorization for release under legislation concerning the protection of the environment, human and animal health?

Yes [ ] No [ ]

"Has such authorization been obtained?

Yes [ ] No [ ]

"If the answer to that question is yes, please attach a copy of such authorization."

18. Resistance characteristics: The Working Party noted the request from the Technical Committee and the preliminary answers received on a questionnaire as reproduced in document TWO/30/11. It also noted that for fruit species very little experience existed in the use of resistance characteristics. It therefore abstained from commenting on the document.

19. The Working Party noted the discussions on the screening of varieties in the Technical Committee and its request to study the subject and give a report of the discussions to its next session. The Working Party noted that at present in its field of competence there was no use made of electrophoresis or DNA marker for the screening of varieties and for the selection of varieties to be grown in the open or in the glasshouse. The Working Party was in principle against such use, but did not want to exclude it completely. The use had, however, to make sense. These methods should therefore only be admitted for screening if a strong correlation

existed between the characteristic in question (e.g. the band or bands in the case of electrophoresis) and morphological or physiological characteristics used in the Test Guidelines. If that was not the case and there was no connection to an expression in the plant, the screening by these means should not be admitted.

Updated Report From the Technical Working Party on Automation and Computer Programs (TWC) on Uniformity

20. List of statistical documents prepared by the TWC: The Working Party noted that the TWC had prepared document TWC/15/2 containing a list of documents produced by it, and document TWC/15/3 containing a topic index to those documents. The Working Party appreciated the updating of those lists and especially the topic index which made it easier to find a particular document on a given subject.

21. COYD and COYU analysis: The Working Party noted that the Technical Committee approved a revised version of the Combined-Over-Years Distinctness (COYD) criterion and the Combined-Over-Years Uniformity (COYU) criterion as contained in document TC/33/7 which replaces the version as contained in document TC/30/4 and that that version would become part of a revised General Introduction to Test Guidelines. It saw, however, little use in the field of fruit species as most tests were not randomized and contained few measurements, and some were only for one year.

22. Window version of DUSTW: The Working Party was also informed that as part of a pilot study into the production of a Windows version of DUSTX the general DUS data analysis package for the PC and thus applying among others the COYD and COYU criteria, a prototype program DUSTW had been produced. The prototype included the DUSTX programs: CHOSX, MERGX, ANALX, TESTX, TVRPX and UNSLX. It would run on 386, 486 and Pentium PC's under Windows 3.1 or Windows 95 (where an SX chip was used, a maths coprocessor is recommended). Whereas DUSTX was run from within MSDOS, the majority of today's software was run from within Windows. With DUSTW, or DUSTX for Windows the appearance of the program was more familiar to today's users and together with the greater interactive capabilities of Windows technology, the program was simpler to use and to learn. DUSTW was written with the DUSTX programs at its core, using the same control files to pass input and output file names and parameters to the programs. With DUSTW, instead of the user needing to edit the control files as necessary with DUSTX, the information was gathered by the program guiding the user to select filenames and options from windows displaying lists of filenames and options (including variety and character names where relevant). When the full version of DUSTW, or DUSTX for Windows was produced the user will be able to use data from Excel spreadsheets as well as from the carefully formatted ASCII files currently required by DUSTX. The program would also be capable of being run in languages other than English. More information can be found in document TWC/15/17.

23. Developments in the World Wide Web: The Working Party noted that in the TWC the importance of E-mail on the World Wide Web and the future trends had been discussed. With respect to UPOV, the situation was as follows: (a) the UPOV office in Geneva already had plans well advanced for the establishment of a Web server; the server would initially provide basic information about UPOV; its history, objectives, membership, structures, principal officers and in time, some of the formal documents, e.g. text of conventions, Test Guidelines,

would be placed on the server for access in electronic form; (b) an EU Fourth Framework FAIR Program proposal had recently been submitted by CPRO/NIAB/BioSS/GEVES to develop variety image database structures which might allow access from Web browsers and (c) the use of the Web for the provision of on-call training in science and technology was becoming increasingly important. An example of interest to crop specialists was the SMART system, a collaborative initiative aiming to provide user-friendly training in quantitative methods for scientists and technical specialists was available in six languages and which could be accessed at http://www.bioss.sari.ac.uk/smart/unix/smart. html.

24. The TWC had welcomed the offer made by the expert from the United Kingdom to set up an E-mail discussion group open to all TWC experts which would be used for discussion of certain subjects by the three special interest groups on visually-assessed characteristics, on BMT data, and on uniformity. It was also proposed whether it could be useful to have Internet structures which facilitated electronic communications and provided an information resource. These might include: (a) an E-mail discussion list where queries and news items might be posted; (b) one or more Web links on UPOV technical matters could be established; this could provide access to the Working Party documents as well as facilitating links between collaborating centers and individuals; (c) for short meetings involving small groups of individuals the possibility of using video conferencing facilities should be considered. The Working Party recommended that the Chairman should take part in the interest group on uniformity.

25. Testing of uniformity: finding the right population standard and decision rule for different sample sizes: The Working Party noted that the Technical Working Party on Automation and Computer Programs (TWC) had prepared document TWC/15/15 on balanced α and β risk tables (single sampling). Document TWC/11/16 was a help in finding the right sample size on the basis of the population standard. However, that document gave rise to some problems when trying to extend it to all species. Document TWC/15/15 listed the problems as (a) the population standard is often not known, (b) especially in new species it leads to small p errors but very large 2 p (consumer risk) errors, (c) the population standard for testing may be different to that required by other authorities, (d) self-fertilized species are treated differently to crop-fertilized species. It further questioned whether it was right that UPOV imposed a certain population standard for all varieties in a given species. If the population standard was necessary, UPOV needed to develop methods to estimate it from the acceptable number of off-types. It then proposed to calculate from the reference collection OC (Operating Characteristic) Curves.

26. UPOV documents in electronic form: The Working Party noted that the Technical Committee had considered the usefulness of documents in electronic form. It also noted that in the TWF a second distribution of technical reports had been made on discs. The Working Party again strongly supported making available the UPOV documents in electronic form. This should not be restricted to Test Guidelines but should cover various other documents, especially reports of meetings and other more important documents. Availability in electronic form would especially facilitate searches for certain subjects in existing documents or reproducing parts for new documents. It noted that the UPOV Test Guidelines may be available in electronic form by the end of the year. It asked that the report of the current session be made available via E-mail to those experts that had given their E-mail address in the list of participants. Once the UPOV home page was established, the UPOV-Test Guidelines should also be available on the WWW in parallel to a CD-ROM.

New Methods, Techniques And Equipment in the Examination of Varieties

27. Mr. Thiele-Wittig gave a short summary report of the fourth session of the Working Group on Biochemical and Molecular Techniques and DNA Profiling in Particular (BMT), referring for further details to document BMT/4/21 Prov.

28. During its session, the BMT had heard short presentations of research results on Azalea, Carnation, Maize, Oilseed Rape, Peach, Potato, Ryegrass, and Tomato; it had heard explanations on the usefulness and the limitations on statistical methods and especially on similarity, clustering and dendrograms, a review of methods for cluster analysis of marker data, on the use of the analysis of molecular variance (AMOVA) for distinctness studies and noted especially the frequent misuse of dendrograms as results of a study; it had heard reports on the correlation and causal linkage between DNA markers and morphological traits and on the relationship between genetic distance and morphological distance between varieties and that only in few cases there were correlations between morphological characteristics and DNA marker; it had noted the reconfirmation of the position of the breeders vis-à-vis DNA profiling and on the study on the use of DNA profiling methods by expert witnesses in disputes on essential derivation and on the effect of different plant breeding schemes in the evaluation of parentage between them and the judgment of essential derivation was not considered to be a task for the national authorities although the courts may approach national authorities for technical advice; it had very contradictory views on the possible use of DNA profiling for prescreening as a possible tool in DUS testing; it had noted that the biggest shortcoming remained in the checking and control of uniformity in characteristics obtained with biochemical or molecular markers, and had very lively and contradictory discussions on possibilities and consequences of the introduction of DNA profiling methods for DUS testing.

29. The next session of the BMT is scheduled to take place under the extended chairmanship of Mr. Joël Guiard, France, in Beltsville, United States of America, from September 22 to 24, 1998. During that session, discussions are planned on the following subjects: (a) Short presentation for research results or their follow-up on different species; (b) assessment of variability within varieties; (c) assessment of variability between varieties; (d) statistical methods: confidence intervals and accuracy of distance estimates; alternative to dendrograms; refinement of the analysis of molecular variance (AMOVA) for distinctness studies and tools to assess uniformity; combination of information from diverse data types (AFLP, SSR, morphological data, etc.); (e) position of the breeders vis-à-vis DNA profiling; (f) the use of DNA profiling methods by expert witnesses in disputes on essential derivation; (g) the use of DNA profiling for prescreening as a possible tool in DUS testing; (h) possibilities and consequences of the introduction of DNA profiling methods for DUS testing; (i) definition of variety; (j) future program of the BMT (date and place of the next session if any).

30. The Working Party noted that at the moment those methods still showed big discrepancies and offered no immediate help in the fruit sector. They seemed to be good for identification purposes but of limited use for distinctness. However, the Working Party had to keep its mind open for the future.

31. The expert from the United Kingdom gave a short report on the program on the use of image analysis for the measuring of starch amount in apples by measuring after an iondine test

the black area compared to the white area. She will prepare a written summary of that program for the next session.

32. The Working Party also noted the report of the Subgroup Meeting on Image Analysis of the TWO as reproduced in document TWO/29/17. The next meeting of that Subgroup will be held in Antibes, France, at the end of 1998. The Subgroup would not be limited to experts from the TWO. The main aim of the research was to arrive at comparable results in the measuring of existing characteristics, despite different hardware and software used. So far there was no aim to reach at new characteristics. The image analysis was therefore, different to electrophoresis or DNA techniques, mainly a different tool for what was already done by other tools.

Problems With the Testing of Earliness in Apples (New Zealand to Prepare a Document)

33. The Working Party noted document TWF/28/6 prepared by experts from New Zealand and the United Kingdom. The document raised the problem of applications for apple varieties with differences only in earliness of maturity. In the UPOV Test Guidelines maturity for consumption is foreseen while applicants claim maturity for harvesting/picking. The problem increases in the case of mutants where all other characteristics show no differences. As a possible solution the paper proposes the adoption of common test methods to assess maturity (e.g. starch tests); the methods could be added as an annex to TG/14/8; a general understanding within UPOV of what difference in earliness would be required (e.g. 3, 5, 7, 10 days apart from the nearest variety); a close liaison with the breeders to understand fully why they believe the variety is early; the need to understand very clearly how the seedling or mutant claims to be different from the parent(s), similar varieties and other type varieties around the claimed maturity date; the use of fruit color to determine maturity rather than eating qualities. Apple varieties bred to "ripen off the tree" could be harvested according to industry directed criteria, but actually tested after a defined period in storage when at maturity for consumption.

34. The Working Party agreed that there was obviously a discrepancy between what was observed and what applicants asked for. There were several post harvest tools available to partly solve the problem such as the starch test, the measuring of total soluble solids or measure the color. For these tests, however, more fruits were necessary than those available from the normal tests. In addition, it was difficult to observe the exact time for harvesting as although color seemed to be one main indicator, it was not the only one, some experts even considered color only secondary in importance for the judgment. Color was mainly important for the decision of the producer for picking as it was important for the attraction of the fruits for selling. The Working Party finally could only conclude that it needed (a) either more trees or longer testing periods (3 to 4 years more) to obtain more fruits for testing (b) to apply post harvest tests and (c) to try to obtain as much background information as possible from the applicant.

Instability in Vegetatively Propagated Crops

35. The expert from Belgium reported on his research on instability in vegetatively propagated species. He recalled that the term mutation originally only meant a change in the DNA sequence but that it was now often used to cover many more sources of variation. He also explained that by using new methods it was now possible to calculate the frequency of

mutations even though many mutations could not be separated by DNA methods. These mutations were not caused through changes of genes (gene mutations) but through other sources. In UPOV, experts would, however, examine changes expressed in the phenotype and hence look also at non-gene mutations.

36. To explain the higher frequency of non-gene mutations he recalled that in seed propagated species at each crossing, recombination and resetting of genes, repair would take place simultaneously and many abnormalities of the cell would be eliminated. In vegetative propagation these repairs would not take place and the cells would age and accumulate abnormalities as there were gene duplication, gene deletion, differences in the structure through transposable elements, which would finally have an effect on the phenotype. Although they also affected other characteristics they were most visible in the flower and fruit or the response to pathotypes or to stress as those characteristics were essential for the survival. As the cell had no possibility to repair itself and reset during a crossing, it had to search for other repair possibilities. Therefore some of the mutations resulting from those abnormalities in the cell were unstable and could change or remutate to the original version. Phenotypic changes could be caused by gene mutations, transposons, epigenetic effects and transgenetics. While only gene mutations would be detectable with DNA fingerprinting, new methods using RNA fingerprinting could also detect some non-gene mutations (epimutations). He finally gave some definitions of genotype, phenotype, epigenotype and epiphenotype, whereby the epigenotype covered all information on the genetic information of the totality of interactions of genes, between genes and the environment of the cell, much more than the DNA alone. As methods for obtaining transgenetic plants he mentioned the bombardment of the cell with a new gene or its introduction with *Agrobacterium tumefaciens*. He also mentioned gene silencing and the importance of gene-gene interactions which could lead to lower or higher expressions of a gene, or genes of opposite direction which would hybridize and thus suppress the expression of that gene.

37. He concluded that in view of the existing difficulties he was rather pessimistic with respect to possibilities to control uniformity and stability. Too many different mechanisms would influence the phenotype. Therefore vegetative propagation should be kept as short as possible, before going back to seed propagation. As certain phenotypic expressions were commercially very important and would be lost in a crossing, this was very contradictory. One basic fact that had to be kept in mind was that the rules for generative propagation would not apply for vegetative propagation. Genemarkers had very little use in vegetative propagation as, although they gave a certain picture of the genome, they only picked out certain points. Thus even with 450 markers the chances that one marker would find a changed gene were less than 1×10^5 and in epigenetic mutations no change of a gene existed and thus no genetic difference could be detected.

Testing of Rootstocks (TWF/28/3)

38. The Working Party noted document TWF/28/3 containing the results of a questionnaire issued. As the answers differed it was not easy to draw general conclusions. The Chairman made a summary of his findings as reproduced in Annex IV to this report. As the total number of applications per rootstock varieties was low, there was only limited need for Test Guidelines. For apple, grape, pear and walnut, a separate document on rootstock seemed to be favored by the majority. For *Prunus* the situation seemed to be difficult. The document did not touch the question of quince rootstocks in pear. It resulted from the discussions, that in

the end the question of whether to establish one single document including rootstock varieties or two separate documents would have to be decided species by species. If a separate rootstock document were prepared that document should not repeat flower and fruit characteristics from the fruit Test Guidelines but should merely make reference to those fruit Test Guidelines if those characteristics were necessary to establish distinctness of a rootstock variety.

39. In connection with these discussions the experts confirmed that for fruit varieties there existed at the national level no fixed list of routine characteristics tested each year. If the candidate variety was not sufficiently distinct from an existing variety the testing office would, on its own initiative, and without special request from the applicant do further tests and look for additional characteristics to establish distinctness and thus help the applicant who often does not know in which characteristic his variety was different from other similar varieties. Experts in the Working Party who also tested ornamental varieties confirmed that in the testing of ornamental varieties the office would also on its own initiative enlarge the test to find additional characteristics if otherwise the candidate variety would have to be declared not distinct. In fruits and ornamental species the expert would not check a list of characteristics fixed at the starting of the testing but look at the whole plant and observe any difference he may see irrespective of whether the characteristic was listed or not. That would apply in the same way for distinctness as it applied for uniformity.

Test Guidelines for Apple Rootstocks

40. After the general discussions on rootstocks a survey among the experts revealed that they were in favor of separate Test Guidelines for apple rootstocks. The Working Party thus noted the existing documents TG/14/5 and TWF/27/13 prepared by experts from the United Kingdom and made the following main changes in document TWF/27/13:

(i) Subject of these Guidelines: To have paragraph 3 from III moved here which should read: "Should characteristics of the flower, the fruit or the seed be necessary for distinction for those characteristics the Test Guidelines for Apple (fruit varieties), TG/14/8 should be used."

(ii) Material Required: The second sentence from the end of paragraph 1 to receive the addition: "Five plants are grown to produce trees."

(ii) Methods and Observations: To have paragraphs 1 and 2 replaced by the new standardized wording on observations determined by measurements, and on the required population standard (1% with one off-type permitted in 20 plants).

(iii) Grouping of Varieties: To have the first paragraph start with the words: "If necessary."

(iv) Table of Characteristics

Characteristics:

1, 2, 6, 12 To have the example variety M9 deleted

7, 26 To be deleted

10, 16 To have the asterisk deleted

5 To have "shine" replaced by "glossiness"

6 To have the bracketed content replaced by "at mid-length"

8 To be recorded "(as for 6)"

11 To have the word "narrow" deleted

12 To have state 3 read "medium brown" and to receive an asterisk

16 To receive new drawings

18 To read: "Expanding leaf: anthocyanin coloration" with the states "absent (1), present (9)"

19 To read: "Expanding leaf: hue of anthocyanin coloration" with the states "bronze (1), purple (2)" and to receive an asterisk

20 To have the states "semi-upwards (3), outwards (5), semi-downwards (7)"

21 to 24 To have the word "blade" added

22 To have a new characteristic 22(a) with an asterisk included reading: "Leaf blade: ratio length/width" with the states "small (3), medium (5), large (7)"

24 To read: "Leaf blade: length of tip" with the states "short (3), medium (5), long (7)"

25 To read: "Leaf blade: incisions of margin"

28 To have the additional states "absent or very weak (1)" and "very strong"

29 To have a new characteristic 29(a) with an asterisk included reading: "Petiole: ratio length of petiole/length of leaf blade" with the states "small (3), medium (5), large (7)"

31 To have the word "begin" added before "bud burst"

32, 33 To be deleted

(v) Technical Questionnaire: On the basis of the Technical Questionnaire of Loquat to request the indication of the species name and paragraph 5 to include characteristics 8, 12, 18 and 31.

Test Guidelines for *Pyrus* Rootstocks

41. The Working Party noted document TWF/27/10 prepared by experts from Germany and made the following main changes in that document:

(i) To have the new standardized wording of the Technical Notes copied from the amended wording for apple rootstocks (see paragraph 40 above).

(ii) Methods and Observations: Paragraph 4 to read: "Unless otherwise stated, all observations on the shoot should be made on one-year-old shoots."

(iii) Table of Characteristics

Characteristics

7, 8, 10 To be deleted

1 To read: "Plant: vigor"

2 To read: "Plant: number of basal shoots"

3 to 19 To have the words "one-year-old" deleted

14 To have the states: "elliptic, broad elliptic, round"

15 To have the second state read: "medium brown"

17 To have the words "apex of" added and the states to read "sharply pointed (1), bluntly pointed (2), rounded (3)"

19 To have "bulge" replaced by "support"

20, 21 To apply to the young shoot and to be placed after characteristic 2; characteristic 20 to have the words "before lignification" replaced by "during rapid growth"

22 To be changed as for apple

27 To read for state 4: "truncate"

28 To have "upper part" replaced by "apex"

31 To have "incisions of" deleted; the German expert to check whether there exists a state "serrate and crenate"

36 To have a new characteristic inserted after characteristic 36 reading: "Leaf: ratio length of blade/length of petiole"

38 To read: "Leaf: presence of stipules;" to be checked whether characteristics 38 to 40 apply to the "stipule" or "rudimentary leaflet"

40 To have the states read "short, medium, long"

(iv) Explanations

Characteristics

1 To be copied from apple

8 To be deleted

22 To be copied from apple

19, 27, 28, 31 To have new drawings to be prepared by experts from Germany

(v) Technical Questionnaire: To have 4.3 (ploidy) deleted, 4.5 reworded according to the standardized wording on GMO's

Standardization of Test Guidelines

42. The Working Party noted that the Technical Committee had taken note of document TC/33/8, Annex II to TC/33/3 and of the discussions held at the TWF, the TWO and the TWV on the harmonization of expression and Notes for different characteristics. On a proposal from the Editorial Committee, the Committee had agreed that the expert from South Africa would amend document TC/33/8. In connection with the above document, the General Introduction to Test Guidelines (TG/1/2) would also be revised and the first task for preparing a preliminary draft for a revised version would be carried out in a group consisting of members of the Editorial Committee, the Chairmen of all the Technical Working Parties and the Chairman and Vice-Chairman of the Technical Committee. The Office of UPOV will collect the information on which part of the General Introduction to Test Guidelines should be revised by the members of the above group. The Working Party noted the new document TWF/28/7 prepared by experts from South Africa and a collection of certain rules provisionally agreed upon by the Editorial Committee as reproduced in document TWF/28/9.

43. The expert from South Africa gave a short explanation of the basic principles of the document and explained the different cases appearing on the basis of a summary as reproduced in Annex II of this report and examples from document TWF/28/7. The Working Party praised the expert for that excellent document which for the first time clearly laid down the different cases. The Chairman invited all experts to study the documents TWF/28/7 and TWF/28/9 and apply the rules to new drafts they would prepare for the next session. If they encountered questions or had proposals for further improvements these should be sent to the expert from South Africa. The experts from Germany and Hungary asked to receive an electronic version for an easier search of certain cases.

Test Guidelines for Loquat

44. The Working Party noted document TG/159/1(proj.) and made the following main changes in that document:

(i) Methods and Observations: Paragraph 1 to refer to measurements; the bracketed contents of paragraph 6 to refer to 10% instead of 5-10%.

(ii) Table of Characteristics

Characteristics

18 To have "of peduncle" deleted

24 To have the order of states changed as follows: 4, 3, 2, 1, 5, 6 and to receive new drawings to be prepared by experts from Japan

25 To have state 3 read: "strongly angular"

32 To have the states "small, medium, large"

34 To have the bracketed content: "(pealing from stalk end)"

37, 43 To have the asterisk deleted

42 To have the order of the states changed into "elliptic, round, obovate"

47, 48, 49 To have the states "small, medium, large"

(iii) Literature: The expert from Japan to supply a new list.

(iv) Technical Questionnaire: To have the request for information on the situation concerning viruses and GMO included.

Test Guidelines for Mume (Japanese Apricot)

45. The Working Party noted document TG/160/1(proj.) and made the following main changes in that document:

(i) Conduct of Tests: The second sentence of paragraph 3 to read: "The fruits should be thinned to leave two or three fruits per spur."

(ii) Methods and Observations: Paragraph 1 to refer to measurements; in paragraph 5 the words "Unless otherwise stated" to be added.

(iii) Grouping of Varieties: To have characteristic 3 as additional grouping characteristic.

(iv) Table of Characteristics

Characteristics

4, 5 To be deleted

16 To have "blade" deleted

22 To have the method added, to be supplied by experts from Japan

26 To have the order of states changed as follows: "elliptic, round, oblate, ovate"

30 To have the state 2 before state 1

31 To read: "Fruit: anthocyanin coloration of skin (as for 30)"

34 To have the order of states changed as follows: "elliptic, broad elliptic, round, obovate"

35 To have state 2 read: "approximate right angled" and to receive a new drawing

36 To read: "Fruit: ratio stone diameter/fruit diameter

40 To be observed at 10% burst

(v) Technical Questionnaire: To have the request for information on the situation concerning viruses and GMO included.

Discussions on Working Papers on Test Guidelines

Test Guidelines for Citrus (Revision)

46. The Working Party noted documents TG/83/3 and TWF/27/14. Because of lack of time it had only a general discussion on document TWF/27/14. It noted the difficulty experienced by the expert from South Africa to cover in one single document all 18 different groups inside the citrus fruit trees. For certain characteristics it was not possible to know for each of the different groups whether it would be applicable and to indicate example varieties. Therefore no example variety existed for many characteristics and in many characteristics only a relative dimension could be requested (e.g. 80: Fruit rind: thickness in relation to diameter of fruit). This type of characteristic was not considered to be very useful and would in reality mean a different characteristic in each group as in practice not the relative length would be used but all varieties inside one group would be compared with each other. On the other hand the Working Party did not want to split the document and prepare separate Test Guidelines for different groups.

47. To find a solution to the problem, the Working Party decided to ask the expert from South Africa to prepare a list of characteristics without example varieties and to only mark each characteristic for the groups to which it could be applied. In a separate list for one or two main groups a list of example varieties could be prepared. The experts from Australia, Israel, Italy and Spain offered their help in that procedure and promised to send their comments to the expert from South Africa.

Test Guidelines for Grapevine

48. The Working Party noted documents TG/50/5, TWF/27/12 and TWF/28/4 and some oral comments by IPGRI and made the following main changes in document TWF/28/4:

(i) Material Required: As a minimum "15 plants on their own roots" should be required; the requirement on the use of rootstocks should be incorporated in the following paragraph and should read: "In the case of a variety with roots sensitive to *Phylloxera vastatrix* they should be grafted on a rootstock not being sensitive to that pest selected by the competent authority."

(ii) Methods and Observations: The first paragraph to read: "Unless otherwise indicated, all observations determined by measurement or counting should be made on 10 plants or parts of 10 plants." The second paragraph to apply to a population standard of 1% and to one off-type allowed in 15 plants. In paragraph 3 the first sentence to be deleted and the last sentence to read: "All observations on the mature leaf should be made on leaves on the middle third of the shoot just above the raceme." A fourth paragraph to be added to read: "4. All characteristics on the bunch and the berry should apply only to varieties for fruit production."

(iii) Technical of Characteristics:

Characteristics

1.1, 1.2 To have the code 7-9

3 To have the states "closed (1), slightly open (2), half open (3), wide open (4), fully open (5)"

42. To have the words "intensity of" deleted

6, 7, 26, 27, 13, 43(c), 48, 49, 51, 52 To have the word "none" in the first state replaced by "absent"

9, 10, 11, 12 To have the second state read: "green and red striped" and to receive a new drawing

17 To have state 3 read: "striate"

18 To be observed at stage "91-00"

22 To read: "Shoot: number of consecutive tendrils" with the states "up to two (1), three or more (2)" and to receive a drawing for explanations

23 To read: "Shoot: length of tendril"

30 to 53 To have the growth stage code replaced by "75-81"

31 To have the drawings improved by the addition of the OIV shapes

32 To have in the explanations a definition as follows: "A lobe is considered to be a lobe if there is a sinus depth of at least one third of the distance between the (imaginary) margin of a non-lobed leaf and the insertion of the petiole."

34 To have the following states "flat (1), V-shaped (2), involute (3), revolute (4), undulate (5)" and the new OIV drawings

39 To read: "Mature leaf: ratio length/width of teeth" with the states from "very small (1)" to "very large (9)"

40 To receive an additional state "one side straight, one side convex (5)" with another drawing

43(a) To read: "Mature leaf: tooth at petiole sinus"; the OIV Code to be "0-081.1"

43(b) To have the word "often" deleted, to have the OIV Code "0-081.2" and to receive a drawing

43(c) To have the OIV Code "0-605" and the IPGRI Code "7.1.34"

51, 52 To have the words "of black" added

56 To be placed before characteristic 30 and to have the following states: "pure male (1), predominantly male (2), male and female well developed (3), female dominant with straight stamens (4), female dominant with reflexed stamens (5)"

64 To have the following states: "oblong (1), narrow elliptic (2), elliptic (3), round (4), oblate (5), ovate (6), obtuse ovate (7), obovate (8), arched (9)"

70 To have the states "unclear (1), slightly clear (2), very clear (3)"

71 To read: "Berry: anthocyanin coloration of flesh" with the states "not colored (1), slightly colored (2), strongly colored (3)"

72 To have the states "soft (1), slightly firm (2), strongly firm (3)"

74 To have the word "other" inserted in state 4

76 To read: "Berry: ease of detachment of pedicel" with the states "difficult (1), slightly easy (2), very easy (3)"

77 To have the states "absent (1), rudimentary (2), well developed (3)"

(iv) Technical Questionnaire

Paragraph

1 To have the request for the indication of the species added

4.1 To be copied from the Test Guidelines for Loquat

4.3 To have a new 4.3 inserted on GMO varieties to be copied from Rape Seed; old 4.3 to become 4.4 and to be copied from apple (TG/14/8) with amendments to be proposed by the expert from Germany.

7.2 To have the additional group "Ornamental variety"

(v) Open Questions

The Working Party asked the expert from Germany to try solving the following open questions: (a) to include, if possible, more example varieties and to delete species names if example varieties were available; (b) to verify the asterisk for characteristic 5; (c) to improve the drawing for characteristic 9 to 12; (d) to check whether characteristic 43(a) should be deleted; (e) to check the states for characteristic 53; (f) to verify whether the characteristics from 57 onwards should only apply to varieties for fruit production only ; (g) to amend in the Technical Questionnaire the question on viruses. As IPGRI needed the data urgently, a preliminary draft resulting from the session should be sent to IPGRI. The expert from Germany would make an attempt to preliminarily solve as many of the above open questions as possible before the end of September or at the beginning of October. The document should, despite the open questions and their preliminary solution only, already be sent to the professional organizations for comments. Any possible discrepancy between the proposal from the German expert and the Working Party would be clarified during the next session of the Working Party.

Test Guidelines for Kiwifruit

49. The Working Party noted documents TG/98/3, TWF/27/17 and TWF/28/5. Lack of time allowed discussions only up to characteristic 25 of document TWF/28/5. The following main changes were made in that document:

(i) Subject of these Guidelines: To apply to ".... the genus *Actinidia* Lindl." instead of ".... any *Actinidia* species and interspecific hybrids."

(ii) Material Required: To have "meristem culture" replaced by "*in vitro* propagation."

(iii) Conduct of Tests: To have in paragraph 4: "8 vines" replaced by "8 plants."

(iv) Methods and Observations: To have the last sentence of paragraph 1 read: " For a sample size of 8 plants, the maximum number of off-types allowed in both cases would be 1." To have the last sentence of paragraph 6 read: "The most basal leaves of a shoot should be excluded." To have in the first sentence of paragraph 8 the words "or King" inserted after "terminal." To have an additional paragraph inserted reading: "All observations on the bud and the bud support should be made on dormant canes."

(v) Grouping of Varieties: To have the bracketed contents of paragraph 2 deleted and after "female" the words "and hermaphrodite" added

(vi) Table of Characteristics

Characteristics

1 To have the states "neuter (without flowers) or floral mutant (only petals, no stamen, no ovary) (1), female (2), hermaphrodite self fertile (3), male (4)"

2 To have the bracketed indications deleted

6 To read: "Young shoot: type of pubescence" with the first state "downy"

10 To have "upper side" replaced by "sunny side"

14 To read: "Stem type of pubescence" with the first state "downy"

15 To have state 2 read: "slightly conspicuous"

18 To read: "Stem: size of bud support (bud support diameter in relation to stem diameter)"

19 To read: "Stem: position of bud" with the states "sunken (1), slightly raised (2), strongly raised (3)"

21 To have the same states as characteristic 19

22 to 39 To apply to the leaf blade

23 To have the states checked

25 To have the states "far apart (1), slightly apart (2), touching (3), slightly overlapping (4), strongly overlapping (5)"

(vii) The expert from New Zealand will inquire about the copyright of the drawings used in the explanations.

Status of Test Guidelines

50. The Working Party agreed that the draft Test Guidelines for Mume (Japanese Apricot) and Loquat should be sent to the Technical Committee for final adoption. It agreed that the draft Test Guidelines for Apple Rootstocks, Grapevine and *Pyrus* should be sent to the professional organizations for comments and that the Working Papers on Test Guidelines for the other species mentioned on the agenda should be (re)discussed at its next session.

Future Program, Date and Place of Next Session

51. At the invitation of the expert from Australia, the Working Party agreed to hold its twenty-ninth session in Coolangatta, Australia, from November 9 to 14, 1998. It was planned that the following items would be discussed during the forthcoming session:

(a) Short reports on new developments in member States in plant variety protection for fruit species (oral reports);

(b) Questions on the testing of varieties of fruit species;

(c) Important decisions taken during the previous sessions of the Working Party and the Technical Committee (oral reports);

(d) Updated report from the Technical Working Party on Automation and Computer Programs (TWC) on uniformity;

(e) New methods, techniques and equipment in the examination of varieties;

(f) Testing of rootstocks

- Prunus Rootstocks (TWF/25/4, TWF/ 27/6)
- Walnut Rootstocks (TWF/26/8)

(g) Standardization of Test Guidelines (TWF/27/16, South Africa to prepare a new paper)

(h) Final discussion on draft Test Guidelines for

- Walnut (TG/125/4(proj.))
- Grapevine (TG/50/6(proj.))
- Apple Rootstocks
- *Pyrus* Rootstocks

(i) Discussions on working papers on Test Guidelines for

- Citrus (Revision) (TG/83/3, TWF/27/14, South Africa to prepare a new working paper)
- European Plum (Revision) (TG/41/4, TWF/25/6, TWF/27/8)
- Kiwifruit (TG/98/3, TWF/28/5)
- Pear (TG/15/1 and Corr., TWF/27/9, TWF/28/2)

52. In order to advance discussions on Test Guidelines, the Working Party agreed in a similar way to the Technical Working Party for Ornamental Plants and Forest Trees (TWO) to select for each of the species in the above planned list one leading expert and to ask the other countries whether they have a special interest in that species and would be willing to cooperate with the leading expert by correspondence in the preparation of a more advanced document. The document would then only be discussed in the full session of the Working

Party if it was in a fairly final stage and only a few changes might be required before its presentation to the professional organizations for comments. The leading expert would also check his draft against the documents TWF/28/7 and 9. It would be aimed at sending the final document to the Office of UPOV at least two months before the next session. The Office of UPOV was asked to prepare a Circular inviting experts from States which had not participated in the session to express their interest and send comments and remarks to the leading expert.

Visits

53. On September 9, 1997, the Working Party visited the CPRO-DLO at Wageningen/Elst. In the evening before it had received an introduction to the research going on in apple breeding, mainly on resistance to scab, mildew and cancer, on marker assisted breeding, on techniques to transform apple varieties with single genes and on durable disease resistance. A copy of the transparencies shown is reproduced in Annex III to this report.

54. In the afternoon of September 9, 1997, the Working Party visited the Dutch Inspection Service for Floriculture and Arboriculture (NAKB) a semi-governmental organization of growers at Horst where it received an inside view of the service on quality control, the control of administrative procedures and the promotion of healthy material. All of the some 4000 nurseries in the Netherlands have to be a member of that service which does obligatory inspection but also offers voluntary certification of plant material.

55. In the afternoon of September 9, 1997, the Working Party also visited the Fleuren Nursery at Baarlo which mainly propagated apple and pear trees where it visited the different presentation plots with different plant density (of up to 10,000 trees per hectare) and different cutting procedures.

56. *This report has been adopted by correspondence.*

[Four annexes follow]

TWF/28/10

ANNEX I

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[Annex II follows]

ANNEX II

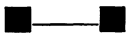
SUMMARY OF STATES OF EXPRESSION

1. QUALITATIVE

1.1 TRUE QUALITATIVE

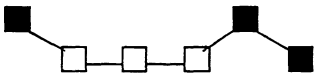
1.2. NON-TRUE QUALITATIVE

1.2.1 Only two states



1.2.2 More than two states

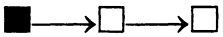
Non-linear: 

Non-linear + linear: 

2. QUALITATIVE EXPRESSED QUANTITATIVE

2.1. CONDENSED QUALITATIVE EXPRESSED TRUE QUANTITATIVE

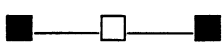
2.1.1 Only lower extreme fixed



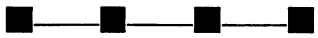
2.1.2 Medium state fixed

2.2. QUALITATIVE EXPRESSED NON-TRUE QUANTITATIVE

2.2.1 CONDENSED — Both extremes at limit



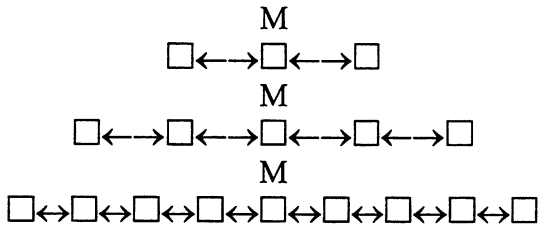
2.2.2 UNCONDENSED



3. QUANTITATIVE

3.1. TRUE QUANTITATIVE

3.1.1 No states fixed



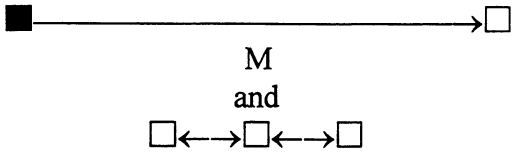
3, 5, 7

1, 3, 5, 7, 9

1, 2, 3, 4, 5, 6, 7, 8, 9

3.1.2 Only lower extremity fixed

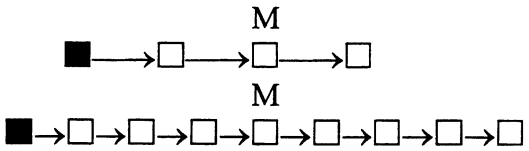
Clear absence



1, 9

3, 5, 7

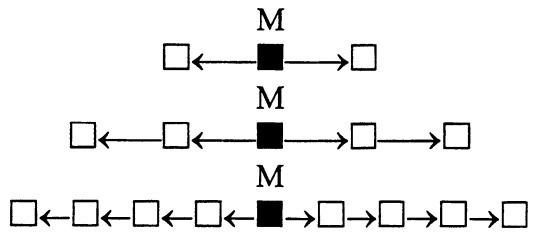
Unclear absence



1, 3, 5, 7

1, 2, 3, 4, 5, 6, 7, 8, 9

3.1.3 Only medium fixed



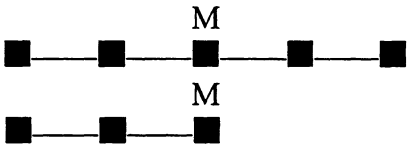
3, 5, 7

1, 3, 5, 7, 9

1, 2, 3, 4, 5, 6, 7, 8, 9

3.2. NON-TRUE QUANTITATIVE

3.2.1 Obvious limit definable for both extreme ends

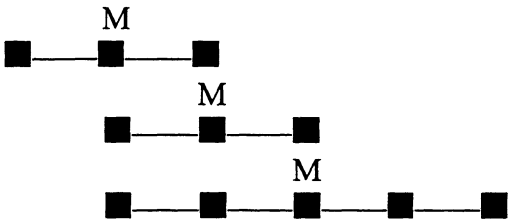


1, 3, 5, 7, 9

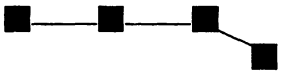
1, 3, 5 ?

Often qualitatively expressed

3.2.2 Without obvious limit to each extreme end



Symmetry easily distorted often qualitatively expressed, may become qualitative merely by addition of states not in the linear range



[Annex III follows]

ANNEX III

Apple breeding and apple research at CPRO-DLO

- Breeding new apple varieties
  (Bert Meulenbroek / Jac Verhaegh)
- Research for improving efficiency of breeding resistance to:
  * Scab (Joke Janse / Henk Schouten)
  * Mildew (Joke Janse)
  * Canker (Eric van de Weg / Karin Burger)
- Marker-assisted breeding in apple
  (Hans Jansen / Chris Maliepaard / Eric van de Weg)
- Development of techniques for transformation of apple with single genes
  (Klaas Puite / Jan Schaart)
- "Durable disease resistance in apple"
  (New EU-project coordinated by INRA; CPRO Henk Schouten and Joke Janse)

Choice of parents

- Combination of characteristics:
  - * resistance to scab
  - * resistance to mildew
  - * resistance to canker
  - * production
  - * growth type
  - * maturing time
  - * fruit size and fruit appearance
  - * taste
  - * storability
- Breeding value as known from earlier crosses

Resistance

- Develop testing methods for (partial) resistance
- Investigate genetic basis
- Investigate pathogene variability.

Resistance sources

- Scab:
  * *M. floribunda (Vf), M. micromalus (Vm)* and *M. pumila (Vr)* plus minor genes?
  * 'Antonovka'-types,
  * CPRO-selections ('Ecolette', 'Santana', etc),
  * 'Discovery', 'Lombart's Calville', etc).
  * D-numbers: high levels of resistance from crabapples.
- Mildew:
  * *M. zumi (Pl1) M. robusta (Pl2)* and 'White Angel' *(Plw)*: mono/digenic absolute resistance,
  * Cultivars with high level of (partial) resistance, like 'Prima', Ákane', etc.
  * D-numbers: high levels of resistance from crabapples.
- Canker:
  * Varieties with good resistance like: 'Clozeau', 'Golden Delicious', 'Jonathan' and 'Lombart's Calville',
  * Varieties with moderate resistance like: 'Elstar', 'Ecolette', 'Kloon 40' and.'Idared'

Fase	Task	Numbers
Crosses	* choosing parents	30
Seeds	* stratification * germination	18,000 17,000
Seedlings (2-3 weeks)	* scab resistance test	17,000
Seedlings (1 - 2 yrs)	* vigor * tree growth type * leaf size * mildew attach	6,000
Seedlings (3 - 7 yrs)	* production * fruit size * appearance * mildew attack <u>fruit sample</u> * taste * sugar/acid content * firmness * juiciness * storability	1,000 450
Selections (10 trees)	REPEAT	20
Variety testing	REPEAT	2 - 5

The development of 'Elstar'

Crossing ('Golden Delicious' x 'Ingrid Marie') 1955

Selection of the seedling (5544-240)..... 1965

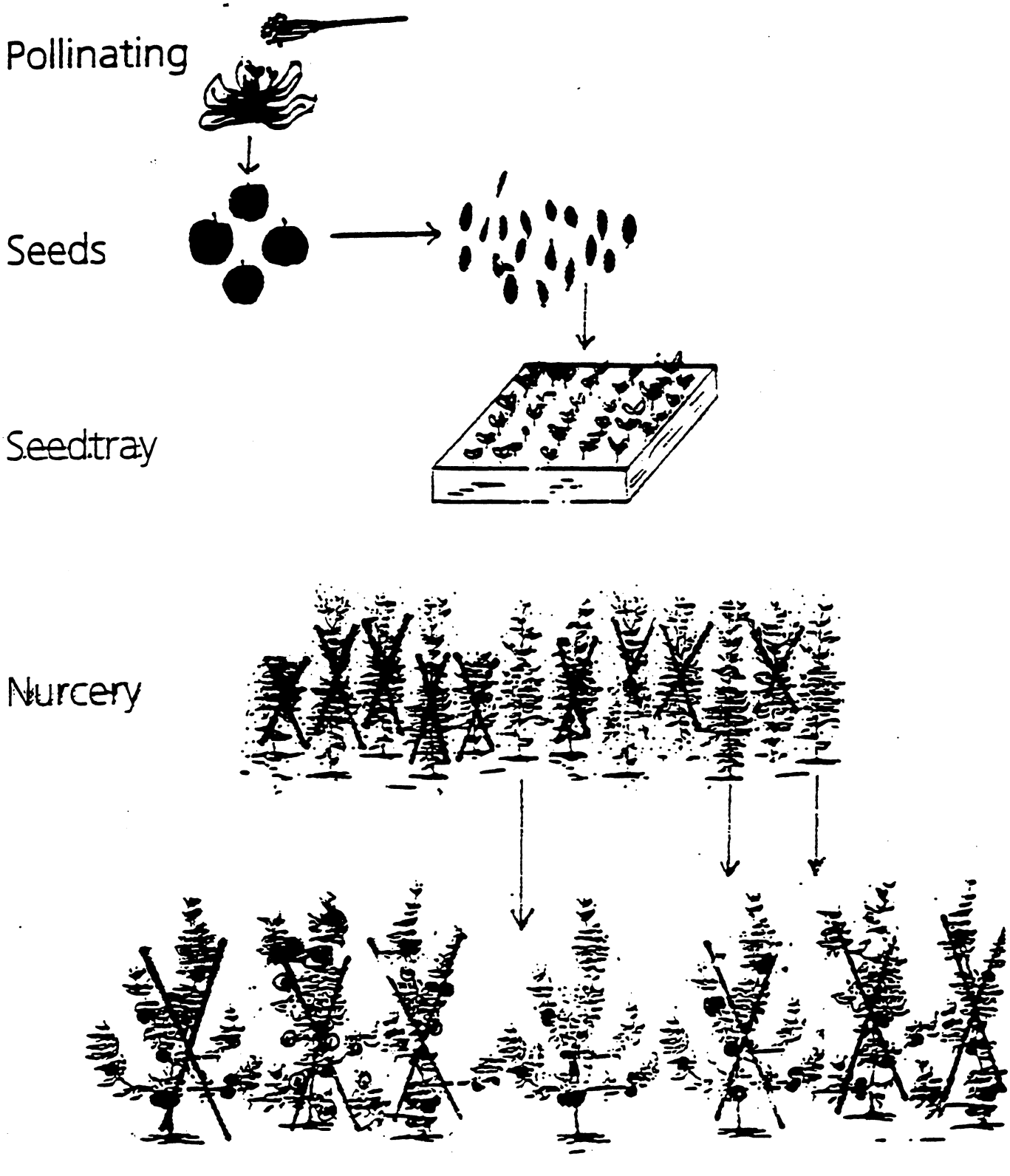
Start of evaluation of cultural value..... 1967

Decision for general testing for cultural value.... 1970

Possitive results from all Dutch trial stations 1975

Release of budwood 1975

Large quantities available in supermarkets 1982



[Annex IV follows]

ANNEX IV

ROOTSTOCK QUESTIONNAIRE TWF/28/3
A general summary of findings

A. The number of rootstock varieties under test is low. The main genus is *Malus*.

B. The broad view of member states who responded to the questionnaire is that when appropriate, separate lists of characteristics for rootstock varieties should be used and/or use of the lists of characteristics for fruit varieties with additional characteristics as necessary.

C. All of the possible guideline options would create problems of varying levels of difficulty. Options b, d, e and f were generally considered to be too complex, unnecessary and not entirely practical. The other three options could have the following problems:

i) the guideline could become difficult to use practically
ii) the guideline could have difficulty covering all characters and states in a genus
iii) the guideline may be unable to cover all variation within species
iv) some varieties do not fruit or flower
v) the fruit guideline would become too large
vi) appropriate example varieties

D. The testing of uniformity of vegetatively propagated rootstocks is not dependent on the guideline used. Most member states considered that there would be no problems whatever guideline option was used. The situation may be different for seed propagated rootstocks, however few countries report any experience.

E. Questions 6, 7 and 8 were not answered in detail. Most respondents stated that the guideline used does not really effect these questions providing that the guideline is clear about which species are included if the whole genus is not. There also appears to be little experience with sterile or late/non flowering rootstocks, interspecific hybrid rootstocks and seed propagated rootstocks.

F. There is a clear split in the respondents views on the significance of flower and fruit characters. Some considered it acceptable to ignore them and others did not. The significance of these characteristics does depend on the genera or species. It was suggested that flower and fruit characteristics could be a simple and very useful way to determine distinctness. There was also a mixture of views on whether non uniformity in flowers and fruit after a grant of rights would lead to withdrawal of the right. In practical terms, flowers and fruit are rarely, if ever, seen in commercial rootstock production.

G. The majority of respondents want a different guideline solution depending on the genera or species. The results suggest that the appropriate fruit guideline is a good starting point when preparing a rootstock guideline in the same genus or species.

For apple, pear, grape and walnut, option g, a or perhaps b are preferred.

For *Prunus* there is less consensus. There is general preference for options a, e or c. The *Prunus* rootstock group appears to be the most difficult to test and formulate guidelines for.

Consideration should also be given to the maturity of the test material. Should rootstock varieties be tested as trees, in a stool bed or both? The propagation method of the variety and the possible use of isoenzyme electrophoresis should also be considered.

Summarised by Chris Barnaby Chairman TWF.

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