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TESTING UNIFORMITY OF APPLE VARIETIES ARISING FROM MUTATION

Document prepared by the Office of the Union

1. The Technical Committee (TC), at its forty-ninth session held in Geneva from March 18 to 20, 2013, noted that an expert from New Zealand would make a presentation on testing of uniformity of Apple varieties arising from mutation at the TWF session in 2013 (see document TC/49/41 "Report on the Conclusions", paragraph 117).
2. Subsequent to the TC meeting, the expert from New Zealand agreed to prepare a document to be presented at all the TWP sessions in 2013, which is reproduced in the following Annex to this document.
3. *The TWV is invited to consider the information on the testing of uniformity of apple varieties arising from mutation in New Zealand, as presented in the Annex to this document.*

[Annex follows]

ANNEX

THE ASSESSMENT OF UNIFORMITY AND STABILITY BY OFF –TYPES ON THE BASIS OF TWO SAMPLES FOR APPLES VARIETIES ORIGINATING AS MUTATIONS IN NEW ZEALAND

Background

Apple varieties originating as mutations have the potential to exhibit greater variability of expression within a variety than varieties originating as seedlings. For this reason an assessment of uniformity using a larger sample size is considered necessary. A larger sample size provides more rigor in the test, but does require additional space for trees which did create a problem due to space limitations in the main testing area at the Cultivar Centre of the New Zealand Institute for Plant and Food Research (Cultivar Centre), the entrusted authority for apple DUS examination on behalf of the Plant Variety Rights Office. A solution was to site additional trees at another location agreed to by the breeder or agent. The second set of trees is often part of commercial or other trial plantings managed by the breeder or agent and in most cases this requirement is easily complied with. The second set of trees is only used for the assessment of uniformity and stability, removing any other requirements for the second test location to have trees of similar or reference varieties.

Testing practice and supply of trees

Information regarding the testing requirements is supplied to the applicant after application. The trees supplied to the Cultivar Centre are used for the determination of DUS, with the second set of trees at the second location used for uniformity and stability assessment only.

Five (5) trees are required to be supplied on MM106 rootstock to the Cultivar Centre. The trees should be second generation trees and recommended to have no more than 20% of the trees coming from any single stick of budwood. Currently all other varieties originating from mutations are on MM106 rootstock and variety to variety comparison would not be possible using other rootstock varieties. All trees must be individually labeled clearly identifying each tree. Trees are supplied between 1 June and 30 September.

In addition to the five trees supplied to the Cultivar Centre, separate test trees will be required for the assessment of uniformity and stability. These trees can be located on a site selected by the breeder or agent and approved by the Plant Variety Rights Office. This trial should be established or existing trees designated at the same time as trees are supplied to the Cultivar Centre, and the Plant Variety Rights Office notified, otherwise the examination towards grant of PVR may be delayed or declined. The location of these additional trees is to be supplied to the Cultivar Centre, with contact information, when Cultivar Centre trees are supplied. The minimum number of trees required is 20 trees on MM106 or 30 trees on M9. The trees for the assessment of uniformity and stability should be second generation trees recommended to have no more than 20% of the trees (e.g. 6 out of 25 or 30) coming from any single stick of budwood.

A recent development has been the decision to complete the move of all varieties in the collection at the Cultivar Centre on to M9 rootstock. This has been underway for several years for varieties originating as seedlings and is now being progressively carried out for mutation varieties. From 2015, trees of apple varieties at the Cultivar Centre will be on M9 rootstock, the number of trees required for testing being ten for all candidate varieties.

Assessment of trees*Purpose*

The purpose of the assessment is to detect evidence of atypical expression, mutation or other genetic instability. Such a problem might be expected to manifest itself in one of two ways:

- A part of a tree producing fruit with skin colouration or patterning outside the expected range for the candidate variety, e.g. block colouring instead of striping, reversion to parent variety colouring.
- A tree producing fruit with chimeral striping.

The assessment is not to measure the variation normally found within a variety such as that caused by the position of fruit on the tree.

Procedure

The assessment is to be carried out at the normal harvest time. The assessment requires that a minimum of 20 trees should each produce at least 40 fruit. If the number is less the assessment should be postponed until the following harvest. The assessment procedure is carried out over two harvests or growing seasons

Inspect each tree, checking for a part of the tree that may be carrying a number of fruit with skin coloration or patterning clearly different from typical expression for the variety. Such a tree part should be described and photographed. The tree should be recorded as an off-type and marked in case the expert or examiner should wish to inspect it.

With each remaining individual tree (i.e. excluding any that may have been already recorded as a possible off-type), harvest all atypical fruit and place in a separate container per tree. Then:

- Inspect the fruit in each container recording signs of genetic instability, in particular chimeral striping.
- Compare the fruit in each container against that in all others. This is to check whether any one tree shows differing types of atypical expression.

All atypical fruit in a container are counted and the type of atypical expression recorded for each fruit. As a guide, a tree may be identified as an off-type tree if the number of atypical fruit harvested is a third or greater in relation to fruit still on the tree. For a sample size of 40 fruit, a minimum of 13 off-type fruit could lead to the tree being recorded as an off-type. Any atypical expression could result in the tree being determined as an off type and fruit number is not the only factor.

If there are problems identifying fruit characteristics and there is uncertainty as to whether or not the expression is atypical, record a description of the nature of the problem and the numbers involved.

The harvested atypical fruit and samples of fruit with typical expression from both locations are later examined and compared side by side in the laboratory by the examiner and any expert advisors.

No statistical analysis is applied to any data or observations recorded from either set of trees.

With 20 trees the maximum number of off-type trees is 1. The table below gives the off-types permitted with tree sample sizes greater than 20. (It is based upon an acceptance probability of 95% and population standard 1% - these bases obtained from document TGP /8/1)

Trees sampled	Maximum off-types
6-35	1
36-82	2
83-137	3

Examination of results

The results of the assessment on trees at the Cultivar Centre and on those from the additional trees at the second location are not combined and are treated as two separate samples. If both samples are determined to meet the uniformity standard then the variety will be considered to be uniform. The same approach applies if both samples are determined to exceed the standard, the variety lacks sufficient uniformity. At the end of testing, after two growing seasons, the variety has been assessed for uniformity and stability on four occasions.

It is possible that the two assessments in a single season are not consistent. If a variety has questionable uniformity it is more likely to be evident in the larger number of trees at the additional location, than in the smaller tree numbers at the Cultivar Centre. This situation has occurred for very few varieties and no conclusion regarding uniformity is arrived at until two seasons or growing cycles have been completed. Should the inconsistency of results at the end of testing prevent a final decision regarding uniformity and stability a further testing season can be undertaken. A variety could be determined to lack uniformity if off-type trees were recorded in the additional trial for both seasons but no off-type trees were recorded at

the Cultivar Centre. The reverse situation could also apply. Consistency over the two seasons in the same trial location is considered more important than the consistency between the two trial locations in the same year.

In the future, the number of trees available for assessment at the Cultivar Centre will increase from five to ten due to the change from trees on MM106 to M9 rootstock. It is not expected that there will be any change to the existing practice and a larger trial size on another site will continue to be required.

Further information about a candidate variety

It is often the case that DUS testers have relatively little information about the candidate variety beyond that provided on the technical questionnaire and a photo. When trees are supplied or made available for testing at the second location, it can be difficult to check that trial trees are true to type and fit the information available. For varieties originating as mutations the typical expression of the variety may not be clear on young trees. The requirement for a breeder or agent to make available additional trees at a second location provides the opportunity to compare the material at the Cultivar Centre with the additional trees. This increases the information available about a variety at an earlier stage and assists to confirm that the trees supplied to the Cultivar Centre, which are used for the determination of distinctness, are true to type. Should the expression of key characteristics on trees in both trials differ, this can be recorded at an early stage in testing and acted upon.

[End Annex and of document]