

# Great Britain and Northern Ireland Variety Lists / UK Plant Breeders' Rights Technical Protocol for Official Examination of Distinctness, Uniformity and Stability (DUS)

Barley

Hordeum vulgare L. sensu lato

November 2024

### Changes

- Updated title from United Kingdom to Great Britain and Northern Ireland
- Updated Plant Breeders' Rights to UK Plant Breeders' Rights
- Updated year to 2024
- Section A
  - Section 7.1
    - UPOV TGP/8 updated from 4 to 5 and updated date
- Section B
  - o Section 7.1
    - Updated language on seed submission
  - o Section 8.1
    - Updated Type of Seed language
- Section C
  - Section 5.6
    - Removed paragraph on selected seed sample. Updated paragraph numbers to match.
- Section D
  - o Section 5.3
    - Updated table to include new additional characteristic "Seed: Proanthocyanidin content"
- Section E
  - Section 4.1
    - Updated language

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# **Section A – General Information**

# 1 Purpose

1.1 This Protocol sets out the procedures for conducting tests and assessments in relation to official examinations of DUS, maintenance of reference stocks and verification of VCU submissions of varieties of winter and spring Barley entered for Variety Listing (VL) trials and/or Plant Breeders' Rights (PBR).

# 2 Scope

2.1 These procedures apply to all varieties of Barley (*Hordeum vulgare* L. *sensu lato*). Special procedures and responsibilities for Genetically Modified (GM) varieties are set out in Sections A5 and A6.

2.2 Except where specified in this protocol or authorised by the Plant Variety Rights Office for the UK, Animal and Plant Health Agency (APHA); only Variety List candidates, Plant Breeders' Rights candidates, candidates for Foreign Authorities and the reference varieties may be incorporated in the DUS tests.

# **3 Responsibilities**

3.1 The growing tests and assessments in this protocol are carried out under the responsibility of the Secretary of State for Environment, Food and Rural Affairs, Scottish Ministers, Welsh Ministers and the Minister for Agriculture, Environment and Rural Affairs in Northern Ireland (the National Authorities).

3.2 They are supervised, on behalf of the National Authorities, by officials of the Testing Authorities: APHA; the Scottish Government (SG); the Department of Agriculture, Environment and Rural Affairs for Northern Ireland (DAERA); and the Welsh Government (WG).

3.3 This protocol is authorised by the Plant Variety and Seeds Committee (PVSC). It cannot be amended without its approval. Requests and suggestions for amendment of the protocol should be put in writing to APHA or the Test Centre.

3.4 The procedures are administered by:

Plant Variety Rights Office for the UK Animal and Plant Health Agency Eastbrook Shaftesbury Road Cambridge CB2 8DR Email: <u>pvs.helpdesk@apha.gov.uk</u> The DUS growing tests and assessments in this protocol are co-ordinated and carried

out by: NIAB Barn 1 Park Farm Villa Road Impington Cambridge CB24 9NZ

Tel No: 01223 342200

3.6 The Test Centre is responsible for providing the appropriate facilities.

# **4 Non-Compliance with the Protocol**

4.1 Where the protocol uses the word "must" for any action then failure to carry out this action will result in non-compliance. Where non-compliance occurs or there are concerns regarding the validity of any data or tests this must be reported to APHA. Where this protocol uses the word "should" for any action this is the method to be followed unless there are clear technical reasons which can be justified by the Test Centre.

### **5** Responsibility for GM Releases

5.1 GM Release Consent Holders are responsible for GM releases. All parties involved in DUS work operating under a GM Release Consent must adhere to the instructions of the Release Consent where necessary, to comply with the relevant consent conditions. Where DUS protocol non-compliance occurs, this must be reported to the consent holder and the Test Centre who will notify APHA.

# **6** Procedures for GM Varieties

6.1 Applicants intending to enter GM candidates must consult APHA, well in advance of their application, about specific requirements under GM regulations.

6.2 The Test Centre must ensure that no test or trial sites are planted with GM candidates and/or varieties until APHA has given the specific clearances.

# 7 Associated Documents

7.1 The following documents are associated with this protocol

Reference	Title
Barley VCU Protocol	United Kingdom Variety List Trials: Protocol for Official Examination of Value for Cultivation and Use (VCU) of Cereals (wheat, barley, oats, triticale, rye and spelt wheat)
UPOV TG/1/3	General Introduction to the Examination of Distinctness, Uniformity and Stability and the Development of Harmonised Descriptions of New Varieties of Plants (19.04.2002).
UPOV TGP/8/5	Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability (28.10.2022).
UPOV TGP/9/2	Examining Distinctness (29.10.2015).
UPOV TGP/10/2	Examining Uniformity (01.11.2019).
UPOV TG/19/11	Guidelines for the Conduct of Tests for Distinctness, Uniformity and Stability, Barley (20.09.2018).
GB and NI Variety Lists	The Seeds (National Lists of Varieties) Regulations 2001 (as amended) and The Seeds (Variety Lists) Regulations (Northern Ireland) 2020
Plant Varieties Act 1997	Plant Breeders' Rights Regulations 1998 and Plant Varieties Act 1997
Plant Breeders' Rights 2019	The Plant Breeders' Rights (Amendment etc.) (EU Exit) Regulations 2019 as amended by The Animal Health, Invasive Alien Species, Plant Breeders' Rights and Seeds (Amendment etc.) (EU Exit) Regulations 2019 and The Plant Breeders' Rights (Amendment) (EU Exit) Regulations 2020

# **Section B – Application Requirements**

# 1 Purpose

1.1 The purpose of this section is to identify the specific requirements for Variety Listing and/or Plant Breeders' Rights applications, as appropriate.

# 2 Scope

2.1 These procedures apply to all applications.

# **3 Responsibilities**

3.1 The applicants are responsible for ensuring that these procedures are complied with.

# **4 Receipt of Applications**

4.1 The latest date for receipt of applications for Variety Listing and/or for Plant Breeders' Rights is stated on the GOV website (https://www.gov.uk/national-lists-of-agricultural-and-vegetable-crops).

4.2 The procedures for the submission of Variety Listing and Plant Breeders' Rights applications, Technical Questionnaires (TQs) and for payment of administration fees can be obtained from APHA PVS at the address shown in Section A or on the GOV website (<u>https://www.gov.uk/national-lists-of-agricultural-and-vegetable-crops</u>)

4.3 Applicants should note in the TQ, submitted with the application, any additional characteristics which may require examinations that are listed in the DUS characteristics section D, 5.2 or 5.3 (an additional fee may be required).

4.4 In the case of hybrid varieties, the TQ must include details of the nature of the hybrid and all parental lines.

# **5 Receipt of Seed**

5.1 The latest date for receipt of seed is stated in the Seed Gazette. In the absence of exceptional circumstances, seed submissions received after this date will be refused. Instructions for the delivery of seed will be made available to applicants by APHA (https://www.gov.uk/national-lists-of-agricultural-and- vegetable-crops).

### **6 Seed Quality Requirements**

6.1 The seed must satisfy the certification requirements for Basic Seed as laid down in the seed marketing legislation of the Devolved Administrations.

6.2 The seed must not be chemically treated. Seed treatment, if required, will be undertaken by the Test Centre. The chemicals applied and rates of application will be determined by the Test Centre.

# 7 Seed Quantity

7.1 Year 1

Туре	Further information	Seed quantity
Conventional Type	N/A	2 kg with 1000 seed weight given
Single Cross Hybrids	Hybrid Male sterile (female parent) Pollinator (male parent)	2 kg of each line with 1000 seed weight given
Single Cross Hybrids	Parental lines already under test or already on the GB and NI Variety List or with UK PBR need not be supplied	Parental lines already under test or already on the GB and NI Variety List or with UK PBR need not be supplied
Other Types	Contact APHA	Contact APHA

There are separate submissions of seed for VCU trials in Year 1. A sample of 200g of seed with be withdrawn from VCU submissions in Year 1 to authenticate the submission (see Section E5).

7.2 Year 2 and Further Year

Submissions None for DUS

A sample of 200g of seed will be withdrawn from VCU submissions in Year 2 and any further years to authenticate the submission (see Section E5). Applicants should refer to Trial Procedures for Official Examination of Value for Cultivation and Use (VCU) for Cereals for seed requirements and Section E4 dealing with replacement seed of a variety.

7.3 Shortfall in Seed Quantities

Where insufficient seed is available in the first instance a further stock must be supplied in the following year which will be authenticated against the original submission. An additional charge may be applied. This must be agreed in advance with APHA and the test centre.

7.4 Hybrid Barley

In the case of Hybrid Barley where insufficient seed stocks of parent lines are available, a minimum of 500g of each line should be supplied in the first instance. Further stocks should be supplied in the following year, and these will be authenticated against the original submission for which an additional charge may be applied. Where components of hybrids are on the GB and NI Variety Lists, or have UK PBR, seed need not be supplied unless specifically requested.

# 8 Labelling Requirements, Including Provisions for GM Varieties

8.1 Applicants must clearly label their seed, inside and outside the bag, with the following information:

- Applicant
- AFP number (if known)
- Breeder's Reference number or name
- Type of Seed (DUS)
- Quantity of seed and thousand seed weight
- In the case of hybrids, whether it is a parental line

8.2 All packages of GM material must be labelled clearly as "GMO" or "Genetically Modified Organism".

# **Section C – Growing Test Procedures**

# 1 Purpose

1.1 The purpose of this section is to provide details of the procedures used in the growing tests for DUS analysis.

# 2 Scope

2.1 These procedures apply to all varieties of Barley.

# **3 Responsibilities**

3.1 The Test Centre is responsible for conducting these procedures.

3.2 The Test Centre will be responsible for ensuring that no material supplied to it is used for any other purpose than the conduct of these procedures or the release of reference samples for authorised purposes (See Section E7).

# **4 Reference Varieties**

4.1 The principles governing the selection of reference varieties are set out in Appendix 1.

4.2 Seed of reference varieties will be supplied by the Test Centre.

# **5 Design of Tests**

5.1 The Test Centre is responsible for selecting a suitable site which should be on ground that has not normally had a Barley crop in the previous three years but may be less where it has been determined the risk of contamination is negligible.

5.2 Field husbandry should follow best practice for all operations and particularly as regards cultivation, drilling, fertiliser and spray application, use of irrigation and control of weeds, pests and diseases.

5.3 The minimum duration of tests should normally be two independent growing cycles. The National List and Seeds Committee (NLSC) must be informed on any proposed changes to the number of cycles.

5.4 The candidate variety will be grown in field-grown plots and compared with varieties which are in the same classification for the following grouping characters based on information given in the TQ; seasonal type; grain type (husked or non-husked); presence or absence of sterile spikelets in the ear (not applicable to six row varieties, where all the spikelets are fertile); presence or absence of lower leaf sheath hairs; number of rows in the ear; rachilla hair type of the grain; presence or absence of hairs in the ventral furrow of the grain; for male sterile parent lines as components of hybrid varieties: presence or absence of pollen production.

5.5 Plots are sown from the submitted seed in each year of test as follows:

Number of plots:	2
Total Number of Plants Examined/Va	riety: 2000
(approx.) No. of plants /m <sup>2</sup>	200-300
Time of sowing (winter Barley)	October - November
Time of sowing (spring Barley)	February - April

5.6 In the case of winter varieties, an additional plot is sown in late April during the first year of tests to examine the uniformity of the vernalisation response (characteristic 'Seasonal Type'):

Number of plots:	1
Total Number of Plants Examined/Variety:	minimum 500 plants
(approx.) No. of plants /m <sup>2</sup>	200-300
Time of sowing	Late April

5.7 At the end of the first recording year, reference varieties which are most similar to the candidate varieties are identified and may be sown in the second year of test.

5.8 At the end of the second year of tests, candidate varieties that are still not distinct may be grown in additional direct comparison plots. This requires approval from APHA, and an additional charge will be made to the applicant.

# 6 Records and Recording

6.1 All records and plot data should be in a form determined and validated by the Test Centre.

6.2 Characters, recording details and instructions are given in Section D. Any variant and abnormal plant or plants resulting from an adverse reaction to husbandry practice are recorded but excluded from analysis.

6.3 In the first test cycle, characters, as indicated in Section D, are recorded on all varieties in test and the data analysed to assess uniformity of the candidate varieties and to determine the most similar reference varieties for each candidate. (For details see Section G).

6.4 In the second test cycle, characters, as indicated in Section D, are assessed on all varieties in test and the data analysed and, together with those from the first year, used to assess distinctness and uniformity of the candidate varieties. (For details see Section G).

6.5 If the Test Centre notices unusual or novel characters in candidate varieties they must be noted and a photographic record taken.

# 7 Communications with the Applicant

7.1 The Test Centre will notify the applicant or the agent of any DUS problems at the earliest practical opportunity as they arise during the test. All such notifications must be copied to APHA.

7.2 In the case of distinctness problems, if confidentiality considerations allow, the applicant should be informed which variety is not distinct and be invited to submit any information which may help to distinguish them.

7.3 If DUS problems arise, applicants will be invited to visit the DUS tests by arrangement so that the material can be examined (if appropriate), and discussions held with the Test Centre.

7.4 After each test cycle the results are summarised and reported to the applicant and APHA by the Test Centre.

#### Section D

# Section D – Summary of DUS Characteristics to be Assessed, Method of Assessment and Standards Applied

# 1 Purpose

1.1 The purpose of this section is to summarise the characteristics to be assessed.

# 2 Scope

2.1 This section summarises characteristics, states of expression, method of observation and standards required for DUS assessment.

# **3 Responsibilities**

3.1 The Test Centre is responsible for co-ordinating the procedures in this summary of characteristics.

# **4** Organisation

4.1 The minimum duration of tests to assess characteristics is normally two independent growing cycles. Shorter durations may be applied for assessment of additional characteristics. Proposed changes to the number of growing cycles must be approved by the NLSC.

# **5 DUS Characteristics to be Assessed**

**Routine Characteristics** 

The following table summarises the DUS characteristics to be routinely examined.

Note:

\* a characteristic which must be examined according to UPOV

Guidelines. G a grouping characteristic

Type of observation of characteristics:

- MG Single measurement of a group of plants of parts of plants
- MS Measurement of a number of individual plants or parts of plants
- VG Visual assessment by a single observation of a group of plants or parts

of plants VS Visual assessment by observation of individual plants or parts of

plants

#### Section D

Number of plants or sample size for assessment

- A Sample size of 100 (See Section G 6.6)
- B Sample size of 2000

# **5.1 Barley Characteristics Routinely Recorded in DUS Tests**

UPOV TG/19/1 1	Character	Material examined	Number of plants or sample size for assessment	Method of assessment and recording	States of expression	D Method and Minimum Distance required	U Method: standard applied
1	Kernel: colour of aleurone layer	Submitted seed	A	VG	1 whitish 2 light grey blue 3 dark grey blue 4 purple 5 black	2 states	1% @95%
2*	Plant: growth habit	Field grown plot	В	VG	1 erect 3 semi-erect 5 intermediate 7 semi-prostrate 9 prostrate	2 - 3 states	0.1% @95%
3	Plant: intensity of green colour	Field grown plot	В	VG	1 light 2 medium 3 dark	2 states	0.1% @95%
4G*	Lowest leaves: hairiness of leaf sheaths	Sub sample in field grown plot	A	VG	1 absent 9 present	1 state	1% @95%
5*	Flag leaf: intensity of anthocyanin colouration of auricles	Field grown plot	В	VG	1 absent or very weak 3 weak 5 medium 7 strong 9 very strong	2 states	0.1% @95%
6	Flag leaf: attitude	Field grown plot	В	VG	1 erect 3 semi-erect 5 horizontal 7 semi-reflexed 9 reflexed	2 states	0.1% @95%
7*	Time of ear emergence: (first spikelet visible on 50% of ears)	Field grown plot	В	MG	1 very early 3 early 5 medium 7 late 9 very late	2 days	0.1% @95%

UPOV States of expression U Character D Method and Material examined Number of Method of TG/19/1 plants or assessment Minimum Method: Distance sample size for and recording standard assessment applied required Field grown plot VG 0.1% @95% 8 В 2 states Flag leaf: glaucosity of sheath 1 absent or very weak 3 weak 5 medium 7 strong 9 very strong 9\* Field grown plot VG 0.1% @95% В 1 absent or very Awns: anthocyanin colouration of tips 2 states weak 3 weak 5 medium 7 strong 9 very strong 10\* Field grown plot В VG 0.1% @95% Ear: glaucosity 1 absent or very 2 states weak 3 weak 5 medium 7 strong 9 very strong 11 Ear: attitude Field grown plot В VG 1 erect 2 states 0.1% @95% 3 semi-erect 5 horizontal 7 semi-drooping 9 drooping VG 0.1% @95% 25 Grain: anthocyanin colouration of nerves Field grown plot В 1 absent or very 2 states weak 3 weak of lemma 5 medium 7 strong 9 very strong 13\* MG 0.1% @95% Plant: length (stem, ear and awns) Field grown plot В 1 very short 2 states 3 short 5 medium 7 long 9 very long Field grown plot VG 0.1% @95% 14\* Ear: number of rows В 1 two 1 state 2 six 0.1% @95% 15\* Ear: development of sterile spikelets Field grown plot В VG 1 none or rudimentary 1 state ("deficiens") 2 full

Section D

Section D

16*	Sterile spikelet: attitude (in mid-third of	Samples from field	A for distinctness	VG	1 parallel	2 states	0.1% @95%
	ear)	grown plot	B for uniformity		2 parallel to divergent		_
					3 divergent		

UPOV TG/19/1 1	Character	Material examined	Number of plants or sample size for assessment	Method of assessment and recording	States of expression	D Method and Minimum Distance required	U Method: standard applied
17*	Ear: shape	Samples from field grown plot	A for distinctness B for uniformity	VG	1 strongly tapering 2 slightly tapering 3 parallel 4 fusiform	2 states	0.1% @95%
18*	Ear: density	Samples from field grown plot	A for distinctness B for uniformity	VG	3 sparse 5 medium 7 dense 9 very dense	2 states	0.1% @95%
19	Ear: length (excluding awns)	Samples from field grown plot	A for distinctness B for uniformity	VG	3 short 5 medium 7 long	2 states	0.1% @95%
20*	Awn: length (compared to ear)	Samples from field grown plot	A for distinctness B for uniformity	VG	1 very short 3 short 5 medium 7 long 9 very long	2 states	0.1% @95%
21	Rachis: length of first segment	Samples from field grown plot	A	VG	3 short 5 medium 7 long	2 states	1% @95%
22	Rachis: curvature of first segment	Samples from field grown plot	A	VG	1 absent or very weak 3 weak 5 medium 7 strong	2 states	1% @95%
23*	Median spikelet: length of glume and its awn relative to grain	Samples from field grown plot	A	VG	1 shorter 2 equal 3 slightly longer 4 longer	2 states	1% @95%
24*	Grain: rachilla hair type	Samples from field grown plot	A	VG	1 short 2 long	1 state	1% @95%
25	Grain: spiculation of inner lateral nerves of dorsal side of lemma	Samples from field grown plot	A	VG	1 absent or very weak 3 weak 5 medium 7 strong	2 states	1% @95%

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26*	Grain: type	Samples from field	A	VG	1 non-husked	1 state	1% @95%
		grown plot			9 husked		

							Section D
UPOV TG/19/1 1	Character	Material examined	Number of plants or sample size for assessment	Method of assessment and recording	States of expression	D Method and Minimum Distance required	U Method: standard applied
27*	Grain: hairiness of ventral furrow	Samples from field grown plot	A	VG	1 absent 9 present	1 state	1% @95%
28	Lemma: shape of base	Samples from field grown plot	A	VG	1 non bevelled 2 bevelled	1 state	1% @95%
29	Seasonal type	Field grown plot	Winter and alternative types: 750 plant plot test sown in late spring Spring types: TQ declaration	VG	1 winter type 2 alternative type 3 spring type	1 state	1% @95%

# **5.2 Special Category Characteristics**

These characters should only be used as a complement to confirm other morphological or physiological differences.

UPOV TG/19/1 1	Character	Material examine d	Number of plants or sample size for assessment	Method of assessment and recording	States of expression (Acid PAGE method)	D Method and Minimu m Distanc e require d	U Method: standard applied
31	C-Hordein composition: allele expression at locus Hor-1	Submitte d seed	20 grains for Distinctness 100 grains for Uniformity	Visual score	$\begin{array}{l} 1 - bands 27 + 30 + 32 + 37 + 39 \\ 2 - bands 27 + 30 + 32 + 34 + 37 + 39 \\ 3 - bands 27 + 30 + 32 + 37 \\ 4 - bands 32 + 37 + 41 \\ 5 - bands 27 + 30 + 32 + 37 + 39 + 41 \\ 6 - bands 32 + 37 + 38 \\ 7 - bands 35 + 38 \\ 8 - bands 32 + 37 + 39 + 41 \\ 9 - bands 38 + 41 + 42 \\ 10 - bands 30 + 32 + 37 \\ 11 - bands 30 + 32 + 37 \\ 11 - bands 34 + 37 \\ 12 - bands 31 + 34 + 37 + 38 + 41 \\ 14 - bands 32 + 37 + 41 + 43 \end{array}$	1 state	1% @95% see note below

						Section D		
UPOV TG/19/1	Character	Material examine d	Number of plants or sample size for assessment	Method of assessment and recording	States of expression (Acid PAGE method)	D Method and Minimu m Distance required	U Method: standard applied	
32	B-Hordein composition: allele expression at locus Hor-2	Submitte d seed	20 grains for Distinctness 100 grains for Uniformity	Visual score	1 - bands 71 +79 +83 +86 +94 +100 2 - bands 71 +82 +89 +100 3 - bands 76 +82 +83 +86 +100 4 - bands 66 +71 +76 +86 +93 +100 5 - bands 71 +78 +79 +90 +94 6 - bands 76 +81 +94 7 - bands 71 +72 +75 +82 +85 +86 +100 8 - bands 72 +76 +79 +90 +94 9 - bands 71 +76 +79 +86 10 - bands 71 +76 +79 +86 10 - bands 71 +78 +83 +86 +94 +100 11 - bands 71 +79 +83 +86 +90 12 - bands 71 +76 +79 13 - bands 71 +89 14 - bands 79 +83 +86 +90 15 - bands 67 +69 +71 +72 +78 +79 +85 +89 +94 16 - bands 71 +79 +83 +88 +94 17 - bands 69 +76 +79 +83 +93 18 - bands 71 +72 +79 +85 +86 +91 +100 19 - bands 72 +76 +100 20 - bands61 +71 +76 +79 +83 21 - bands 76 +81 +94 +100	1 state	1% @95% see note below	

Note – allowance is made for the presence of biotypes.

# **5.3 Approved Additional Characteristics**

The following table summarises the additional characteristics which have been approved by the NLSC for Barley.

Type of expression	Characteristic	Growth Stage	Method of observation	States of expression	Example varieties	Note
Qualitative	Production of pollen (male sterility)	60 – 65	VG	absent present	FM 99-18 MT 99-18	1 9
Qualitative	Seed: Proanthocyanidin content	Submitted seed	VG	absent present	Husky Origin	1 9

### **5.4 New Additional DUS Characteristics**

Applicants can suggest new additional characters on the Technical Questionnaire for testing DUS or after notification by the DUS Test Centre of distinctness problems. For procedures see Section F.

# Section E – Reference Seed Stock Maintenance and VCU Seed Stock Authentication Procedures

# 1 Purpose

1.1 This section sets out the procedures for reference seed stock maintenance and VCU seed stock authentication (if applicable).

# 2 Scope

2.1 These procedures apply to all reference collection varieties and VCU seed submissions where the VCU seed has not been taken from the same bulk as the seed used for the DUS test.

# **3 Responsibilities**

3.1 The Test Centre is responsible for conducting these procedures.

# **4 Procedures for Reference Seed Stock Maintenance**

4.1 The DUS seed sample submitted with the successful or pending application is considered to be the definitive seed of the variety. Subject to meeting the required certification standards (see Section B) the seed is dried and placed in storage under controlled and monitored refrigerated conditions as part of the official reference collection.

4.2 If during the normal tests there is any evidence that seed is deteriorating in storage, or that stocks are under 500 g, a request will be made to the maintainer asking for replacement seed (2 kg) of the variety. This replacement seed must be authenticated against the definitive seed.

4.3 Plots will be established from any replacement reference seed sample to be authenticated and compared visually with the definitive seed over a maximum of two recording seasons. Plots must be examined through all the growth stages from early growth habit to full harvest ripeness. If the new seed sample cannot be visually distinguished from the reference seed it will be accepted as representing the variety. It will then be considered as the definitive seed and substituted for the existing definitive seed in the reference collection.

4.4 In the event of the replacement sample not meeting the required acceptance standards, an additional replacement sample will be requested. If the additional replacement sample does not meet the acceptance criteria set out in 4.3, the variety will be deleted from the reference collection and the Variety Lists reviewed.

# **5 Procedures for VCU Seed Stock Authentication**

5.1 A representative sub-sample of seven grains from the VCU seed submission are compared to a representative sub-sample of seven grains from the DUS seed submission (definitive seed) by electrophoresis using the Acid PAGE method. If the VCU seed sample tested by the electrophoresis method does match the DUS seed, it will be considered to represent the variety.

5.2 If the VCU seed sample does not show the same banding pattern as the DUS seed sample a further electrophoresis test will be carried out. If the VCU seed sample still does not match the DUS seed,

side-by-side field plots of the two samples will be established and compared visually from early growth habit to full harvest ripeness.

5.3 If the VCU plot does not differ from the DUS plot in the comparison of field sown plots the VCU seed will be considered to represent the variety.

5.4 If the VCU plot can be visually distinguished from the definitive stock in the authentication plots then it will not be accepted as representing the variety.

# 6 Release of Reference Samples for Authorised Purposes

6.1 A maximum of 200 g of seed of reference samples can be supplied by the Test Centre, on request, to UK and UPOV DUS Testing Authorities and UK and OECD Seed Certification Agencies. The recipient will be notified in writing that this material, or any material derived from it, must not be supplied to a Third party or used for any other purpose than as a reference for official DUS testing or seed certification.

6.2 Provision of reference samples, other than in 6.1, to any other parties must be authorised by APHA.

# Section F – Procedures for Assessment of New Additional DUS Characters

# 1 Purpose

1.1 This section sets out the procedures for assessment of new additional DUS characters for varieties of Barley entered for Variety Listing and/or PBR trials.

# 2 Scope

2.1 These procedures apply to applications where additional DUS characteristics which have not been previously approved by the NLSC, are requested for use in the examination of DUS.

# **3 Responsibilities**

3.1 The Test Centre is responsible for liaising with the applicant to produce a proposed procedure for the conduct of new tests. This procedure must ensure that Distinctness, Uniformity and Stability requirements will be met.

3.2 All new additional characteristics must be authorised by the NLSC in consultation with the PVSC.

### **4 Reference Varieties**

4.1 The reference varieties must include varieties from which the candidate variety is not distinct, as well as other varieties for control purposes.

4.2 Seed of reference varieties will be supplied by the Test Centre.

# **5** Procedures

5.1 Details of the proposed special test or assessments will be submitted to the NLSC.

5.2 The NLSC may commission a test or trial to further investigate a proposal. The applicant will be advised by APHA of arrangements and costs.

5.3 Where the test for a character is approved by the NLSC it should be subsequently listed in Section D5.1, D5.2 or D5.3 as appropriate.

# **Section G – Procedures for DUS Decisions**

# 1 Purpose

1.1 This section sets out the standards used to assess distinctness, uniformity and stability of varieties of Barley.

# 2 Scope

2.1 These procedures apply to all varieties of Barley (*Hordeum vulgare* L. *sensu lato*) entered for Variety Listing and/or Plant Breeders' Rights tests and those being tested on behalf of Foreign Authorities.

# **3 Responsibilities**

3.1 The Test Centre is responsible for applying the criteria for DUS, set out in this procedure.

3.2 The Test Centre is responsible for producing the DUS report in accordance with these procedures and for ensuring that they are in accordance with UPOV guidelines.

# **4 Reference Varieties**

4.1 Appendix 1 sets out which varieties are considered as reference varieties for these procedures.

# **5** Distinctness

5.1 In accordance with associated document UPOV TG/1/3, varieties can be considered distinct where they have a different expression in a grouping character e.g. seasonal type; grain type (husked or non-husked); presence or absence of hairs on the lower leaf sheaths; presence or absence of sterile spikelets in the ear (not applicable to six row varieties); number of rows in the ear; rachilla hair type of the grain; presence or absence of hairs in the ventral furrow of the grain; for male sterile parent lines as components of hybrid varieties: presence or absence of pollen production.

5.2 The standard applied for distinctness over two years of test is a clear difference of one or two states in the expression of a characteristic in accordance with the table of characteristics given above in Section D.

#### Section F

5.3 Where varieties are grown in close proximity under the same conditions, and a direct comparison can be made, distinctness can be determined on the basis of visual observation. Characters are recorded using notes to represent states of expression (See Section D). In these circumstances, the basis for distinctness will be clearly recorded.

5.4 Hybrid

s Distinctness follows the principle of "hybrid first" i.e., if the final hybrid variety is not distinct then distinctness is examined by testing the parent lines – either the CMS (female parent) or the Restorer (pollen donor/male parent) must be clearly distinguishable from the respective male or female parent of the non- distinct hybrid variety. Hybridity may be used as a grouping character based on the TQ declaration made by the applicant. CMS female parent lines will only be compared to other CMS lines. Pollen production – absence or presence may also be used as a UK grouping character. It is tested by growing the male sterile parent line in isolated seed plots of approx. 2000 plants.

# **6 Uniformity**

6.1 Uniformity is assessed for all characteristics used to establish distinctness.

6.2 Uniformity is based on the assessment of off-types (variants) for visually observed characters.

6.3 The assessment of off-types is undertaken in both test cycles and the total should not exceed the number allowed using the population standards detailed below. Care is taken to ensure that the plants that are counted are not the result of any non-genetic factors such as environment, pest and disease.

6.4 In a sample size of 2,000 (characters marked as "B" in Section D), a population standard of 0.1% and an acceptance probability of at least 95% should be applied. For example, in a sample of 2000 plants, 5 off types are allowed.

6.5 In a sample size of 100 (characters marked as A in Section D), a population standard of 1% and an acceptance probability of at least 95% should be applied. For example, in a sample of 100 plants or parts of plants, 3 off-types are allowed.

6.6 For characters marked as "A" in Section D (with the exception of characteristic 1), the assessment of uniformity can be carried out in two stages. In the first stage, 20 plants or parts of plants are examined. If no off-types are observed the variety is declared uniform. If more than three off-types are found the variety is declared not to be uniform. If one to three off-types are observed, a further 80 plants or parts of plants should be examined.

6.7 The following uniformity standards apply to hybrids and their progenitor lines:

6.7.1 Hybrid B characters 10% @ 95% probability A characters 10% @ 95% probability

The maximum permissible number of off-types for B characters will be 225 plants or parts of plants in a sample of 2000. The sample size for assessment can be reduced to 200 plants. In this case, a maximum of 27 off-types are allowed. The maximum permissible number of off-types for A characters will be 15 plants or parts of plants in a sample of 100 plants or parts of plants.

6.7.2. Male sterile parent	B characters	0.2% @ 95% probability
	A characters	1% @ 95% probability

The maximum permissible number of off-types for B characters will be 8/2000 and the maximum permissible for A characters will be 3/100.

6.7.3. Male sterile single cross hybrids	
as parent in a 3-way hybrid) A characters	

B characters 0.5% @ 95% probability (used 1% @ 95% probability

The maximum permissible number of off-types for B characters will be 15/2000 and the maximum permissible for A characters will be 3/100.

6.7.4. The pollinator and restorer lines are usually inbred lines and will be tested as conventional self- pollinating varieties (see 6.4. and 6.5 above).

6.8 Resubmissions

For all varieties, except hybrid varieties, a resubmission of plant material (seed) may be allowed for the second growing cycle if in the first growing cycle the number of off-types did not exceed 15 plants in a sample size of 2000 plants (population standard of 0.5% with an acceptance probability of at least 95%) or 9 plants or parts of plants in a sample size of 100 (population standard of 5% with and acceptance probability of at least 95%).

# 7 Stability

7.1 A variety is considered sufficiently stable when there is no evidence to indicate that it lacks uniformity or fails to conform to the essential characteristics of its description in different submissions or in different tests.

7.2 Hybrids may be considered to lack stability if there is evidence that their progenitor lines lack uniformity or fail to conform to the essential characteristics of their description.

7.3For three-way hybrids with segregating progenitor lines, the production and maintenance schemes of all progenitor lines must indicate that the final hybrid (candidate) can, in terms of its genetic constitution, be consistently reproduced in each cycle of propagation.

# **8 DUS Report and Variety Description**

8.1 Upon completion of the DUS examination the DUS Summary report will be submitted to APHA and will be discussed at the relevant DUS Test Centre Meeting. This report will specify all non-routine characteristics for establishing distinctness.

8.2 The final DUS report, including the full variety description for positive reports, will be submitted to APHA. The characteristics to be used in the description are identified in Section D.

# **Appendix 1 – Reference Collection Varieties**

# **1 Variety Listing and Plant Breeders Rights**

1.1 The DUS reference collection, for any given category of plant variety comprises the following at the time when the application for the candidate is made:

1.1.1 All other candidate varieties already in DUS test in the UK or entering testing at the same time as the candidate.

1.1.2 All varieties with UK PBR.

1.1.3 All varieties on the OECD variety list that are listed by countries with comparable climatic conditions to the UK.

1.1.4 All varieties protected under National PBR (UPOV contracting parties) with comparable climatic conditions to the UK

1.1.5 Any varieties nominated by the applicant as being comparable i.e., known to be similar.

1.1.6 Any other varieties considered to be comparable i.e., known to be similar by the appropriate Test Centre or DUS Centre Group.

1.1.7 Other available comparable varieties in common knowledge.



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