E

TG/259/2(proj.3)
ORIGINAL: English
DATE: 2016-11-02

INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

Geneva

DRAFT

AGARICUS

UPOV Code(s):

AGARI_BIS

Agaricus bisporus (Lange.) Sing.

GUIDELINES

FOR THE CONDUCT OF TESTS

FOR DISTINCTNESS, UNIFORMITY AND STABILITY

prepared by experts from European Union
to be considered by the
Enlarged Editorial Committee
at its meeting, to be held in Geneva,
from 2017-01-11 to 2017-01-12

Disclaimer: this document does not represent UPOV policies or guidance

Alternative names:*

Botanical name	*English*	*French*	*German*	*Spanish*
Agaricus bisporus (Lange.) Sing.	Mushroom	Champignon de couche	Champignon	Champiñón

The purpose of these guidelines ("Test Guidelines") is to elaborate the principles contained in the General Introduction (document TG/1/3), and its associated TGP documents, into detailed practical guidance for the harmonized examination of distinctness, uniformity and stability (DUS) and, in particular, to identify appropriate characteristics for the examination of DUS and production of harmonized variety descriptions.

ASSOCIATED DOCUMENTS

These Test Guidelines should be read in conjunction with the General Introduction and its associated TGP documents.

TABLE OF CONTENTS PAGE

1. Subject of these Test Guidelines

These Test Guidelines apply to all varieties of *Agaricus bisporus* (Lange.) Sing..

2. Material Required

2.1 The competent authorities decide on the quantity and quality of the material required for testing the variety and when and where it is to be delivered. Applicants submitting material from a State other than that in which the testing takes place must ensure that all customs formalities and phytosanitary requirements are complied with.

2.2 The material is to be supplied in the form of spawn or pure culture on a suitable medium.

2.3 The minimum quantity of material, to be supplied by the applicant, should be:

(a) 15 litres of spawn
or
(b) 2 slant tubes or agar plate (petri dish), containing a pure culture.

2.4 The material supplied should be visibly healthy, not lacking in vigor, nor affected by any important pest or disease.

2.5 The material should not have undergone any treatment which would affect the expression of the characteristics of the variety, unless the competent authorities allow or request such treatment. If it has been treated, full details of the treatment must be given.

3. Method of Examination

3.1 Number of Growing Cycles

3.1.1 The minimum duration of tests should normally be two independent growing cycles.

3.1.2 The two independent growing cycles should be in the form of two separate cultivations.

3.1.3 The growing cycle is considered to be from spawn inoculation until the end of the first flush. Extension of the cultivation period can be requested by the applicant if the distinctness can only be demonstrated in the second and/or third flush.

3.2 Testing Place

Tests are normally conducted at one place. In the case of tests conducted at more than one place, guidance is provided in TGP/9 “Examining Distinctness”.

3.3 Conditions for Conducting the Examination

The tests should be carried out under conditions ensuring satisfactory growth for the expression of the relevant characteristics of the variety and for the conduct of the examination.

3.4 Test Design

3.4.1 The design of the tests should be such that fruit bodies or parts of fruit bodies may be removed for measurement or counting without prejudice to the observations which must be made up to the end of the growing cycle.

3.4.2 Each test should be designed to result in a total of at least 90 fruit bodies in the first flush, which should be divided equally over 3 replicates. 45 fruit bodies should be collected at stage 2 and 45 fruit bodies should be collected at stage 5 (see chapter 8.3).

3.4.3 A minimum growing surface per strain of 1m² is advised in order to obtain sufficient fruiting bodies in both stages.

3.5 Additional Tests

Additional tests, for examining relevant characteristics, may be established.

4. Assessment of Distinctness, Uniformity and Stability

4.1 Distinctness

4.1.1 General Recommendations

It is of particular importance for users of these Test Guidelines to consult the General Introduction prior to making decisions regarding distinctness. However, the following points are provided for elaboration or emphasis in these Test Guidelines.

4.1.2 Consistent Differences

The differences observed between varieties may be so clear that more than one growing cycle is not necessary. In addition, in some circumstances, the influence of the environment is not such that more than a single growing cycle is required to provide assurance that the differences observed between varieties are sufficiently consistent. One means of ensuring that a difference in a characteristic, observed in a growing trial, is sufficiently consistent is to examine the characteristic in at least two independent growing cycles.

4.1.3 Clear Differences

Determining whether a difference between two varieties is clear depends on many factors, and should consider, in particular, the type of expression of the characteristic being examined, i.e. whether it is expressed in a qualitative, quantitative, or pseudo-qualitative manner. Therefore, it is important that users of these Test Guidelines are familiar with the recommendations contained in the General Introduction prior to making decisions regarding distinctness.

4.1.4 Number of fruit bodies or parts of fruit bodies to be Examined

Unless otherwise indicated, for the purposes of distinctness, all observations on single fruit bodies should be made on 90 fruit bodies or parts of fruit bodies taken from each of 90 fruit bodies and any other observations made on all fruit bodies in the test, disregarding any off-type fruit bodies.

4.1.5 Method of Observation

The recommended method of observing the characteristic for the purposes of distinctness is indicated by the following key in the second column of the Table of Characteristics (see document TGP/9 “Examining Distinctness”, Section 4 “Observation of characteristics”):

MG: single measurement of a group of fruit bodies or parts of fruit bodies
MS: measurement of a number of individual fruit bodies or parts of fruit bodies
VG: visual assessment by a single observation of a group of fruit bodies or parts of fruit bodies
VS: visual assessment by observation of individual fruit bodies or parts of fruit bodies

Type of observation: visual (V) or measurement (M)

“Visual” observation (V) is an observation made on the basis of the expert’s judgment. For the purposes of this document, “visual” observation refers to the sensory observations of the experts and, therefore, also includes smell, taste and touch. Visual observation includes observations where the expert uses reference points (e.g. diagrams, example varieties, side-by-side comparison) or non-linear charts (e.g. color charts). Measurement (M) is an objective observation against a calibrated, linear scale e.g. using a ruler, weighing scales, colorimeter, dates, counts, etc.

Type of record: for a group of fruit bodies (G) or for single, individual fruit bodies (S)

For the purposes of distinctness, observations may be recorded as a single record for a group of fruit bodies or parts of fruit bodies (G), or may be recorded as records for a number of single, individual fruit bodies or parts of fruit bodies (S). In most cases, “G” provides a single record per variety and it is not possible or necessary to apply statistical methods in a fruit body-by-fruit body analysis for the assessment of distinctness.

In cases where more than one method of observing the characteristic is indicated in the Table of Characteristics (e.g. VG/MG), guidance on selecting an appropriate method is provided in document TGP/9, Section 4.2.

4.2 Uniformity

4.2.1 It is of particular importance for users of these Test Guidelines to consult the General Introduction prior to making decisions regarding uniformity. However, the following points are provided for elaboration or emphasis in these Test Guidelines:

4.2.2 For the assessment of uniformity of cross-pollinated varieties, a population standard of 1% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 90 fruit bodies, 3 off-types are allowed.

4.3 Stability

4.3.1 In practice, it is not usual to perform tests of stability that produce results as certain as those of the testing of distinctness and uniformity. However, experience has demonstrated that, for many types of variety, when a variety has been shown to be uniform, it can also be considered to be stable.

4.3.2 Where appropriate, or in cases of doubt, stability may be further examined by testing a new stock to ensure that it exhibits the same characteristics as those shown by the initial material supplied.

5. Grouping of Varieties and Organization of the Growing Trial

5.1 The selection of varieties of common knowledge to be grown in the trial with the candidate varieties and the way in which these varieties are divided into groups to facilitate the assessment of distinctness are aided by the use of grouping characteristics.

5.2 Grouping characteristics are those in which the documented states of expression, even where produced at different locations, can be used, either individually or in combination with other such characteristics: (a) to select varieties of common knowledge that can be excluded from the growing trial used for examination of distinctness; and (b) to organize the growing trial so that similar varieties are grouped together.

5.3 The following have been agreed as useful grouping characteristics:

(a) Cap: color (characteristic 8)
(b) Gills: color (characteristic 19)
(c) Basidium: spores (characteristic 21)

5.4 Guidance for the use of grouping characteristics, in the process of examining distinctness, is provided through the General Introduction and document TGP/9 “Examining Distinctness”.

6. Introduction to the Table of Characteristics

6.1 Categories of Characteristics

6.1.1 Standard Test Guidelines Characteristics

Standard Test Guidelines characteristics are those which are approved by UPOV for examination of DUS and from which members of the Union can select those suitable for their particular circumstances.

6.1.2 Asterisked Characteristics

Asterisked characteristics (denoted by *) are those included in the Test Guidelines which are important for the international harmonization of variety descriptions and should always be examined for DUS and included in the variety description by all members of the Union, except when the state of expression of a preceding characteristic or regional environmental conditions render this inappropriate.

6.2 States of Expression and Corresponding Notes

6.2.1 States of expression are given for each characteristic to define the characteristic and to harmonize descriptions. Each state of expression is allocated a corresponding numerical note for ease of recording of data and for the production and exchange of the description.

6.2.2 In the case of qualitative and pseudo-qualitative characteristics (see Chapter 6.3), all relevant states of expression are presented in the characteristic. However, in the case of quantitative characteristics with 5 or more states, an abbreviated scale may be used to minimize the size of the Table of Characteristics. For example, in the case of a quantitative characteristic with 9 states, the presentation of states of expression in the Test Guidelines may be abbreviated as follows:

State	*Note*
small	3
medium	5
large	7

However, it should be noted that all of the following 9 states of expression exist to describe varieties and should be used as appropriate:

State	*Note*
very small	1
very small to small	2
small	3
small to medium	4
medium	5
medium to large	6
large	7
large to very large	8
very large	9

6.2.3 Further explanation of the presentation of states of expression and notes is provided in document TGP/7 “Development of Test Guidelines”.

6.3 Types of Expression

An explanation of the types of expression of characteristics (qualitative, quantitative and pseudo-qualitative) is provided in the General Introduction.

6.4 Example Varieties

Where appropriate, example varieties are provided to clarify the states of expression of each characteristic.

6.5 Legend

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
1 2	**3 4**	**5 6**	**7**			
	Name of characteristics in English	**Nom du caractère en français**	**Name des Merkmals auf Deutsch**	**Nombre del carácter en español**		
	states of expression	types d'expression	Ausprägungsstufen	tipos de expresión		

1 Characteristic number

2 (*) Asterisked characteristic – see Chapter 6.1.2

3 Type of expression
QL Qualitative characteristic – see Chapter 6.3
QN Quantitative characteristic – see Chapter 6.3
PQ Pseudo-qualitative characteristic – see Chapter 6.3

4 Method of observation (and type of plot, if applicable)
MG, MS, VG, VS – see Chapter 4.1.5

5 (+) See Explanations on the Table of Characteristics in Chapter 8.2

6 (a)-(f) See Explanations on the Table of Characteristics in Chapter 8.1

7 Growth stage key See Explanations on the Table of Characteristics in Chapter 8

7. Table of Characteristics/Tableau des caractères/Merkmalstabelle/Tabla de caracteres

		English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
1.		**QN VG**	**(+)**				
		Mycelium: intensity					
		weak				J10263	1
		medium				Horronda, Sylvan A15	2
		strong				Brawn, Heirloom	3
2.		**QN VG**	**(+)**				
		Number of pins					
		few				Horronda	3
		medium				Amycel 2400	5
		many				Horwitu, Sylvan A15	7
3.	**(*)**	**QN MG**	**(+)**	**2**			
		Time beginning of harvest					
		early				Brawn, Euromycel 30	3
		medium				Amycel 2400, Sylvan A15	5
		late				Euromycel 58	7
4.	**(*)**	**QN MG**	**(+)**	**2**			
		Time of peak of first flush					
		early				Heirloom	3
		medium				Amycel 2400, Sylvan A15	5
		late				Brawn, Euromycel 58	7
5.	**(*)**	**QN MS/VG**		**(a), (b), (f)**	**2**		
		Stipe: length					
		short				Brawn	3
		medium				Broncoh, Sylvan A15	5
		long				Amycel 2400, Horwitu	7
6.	**(*)**	**QN MS/VG**	**(+)**	**(a), (b)**	**2**		
		Stipe: diameter					
		narrow				Somycel 53	3
		medium				Brawn, Broncoh	5
		broad				Horronda	7

		English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
7.	**(*)**	**QN MG/MS/VG**	**(+) (a)**	**2**			
		Stipe: ratio length/diameter					
		low				Brawn	3
		medium				Sylvan A15	5
		high				Somycel 53	7
8.	**(*)**	**PQ VG**	**(a)**	**2**			
		Cap: color					
		white				Sylvan A15	1
		greyish white				Somycel 76	2
		brown				Amycel 2400	3
9.	**(*)**	**QN VG**	**(a)**	**2**			
		Only varieties with brown cap: Cap: intensity of color					
		very light				Broncoh, J10263	1
		light				Amycel 2400	3
		medium				Heirloom	5
		dark				Brawn	7
		very dark				BP-1	9
10.		**QL VG**	**(+) (a)**	**2**			
		Only varieties with brown cap: Stipe: color					
		white				Brawn, Heirloom	1
		greyish white				Amycel 2400	2
11.		**QL VG**	**(+) (a)**	**2**			
		Stipe: oxidation at cutting edge					
		absent				Sylvan A15	1
		present				Heirloom, Somycel 53	9

		English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
12.	**(*)**	**QN MS/VG**	**(a), (b), (f)**	**2**			
		Cap: height					
		short				J10263	3
		medium				Brawn, Sylvan A15	5
		tall				Euromycel 58	7
13.	**(*)**	**QN MS/VG**	**(a), (b), (f)**	**2**			
		Cap: diameter					
		small				Horwitu	3
		medium				Broncoh	5
		large				Heirloom, Sylvan A15	7
14.	**(*)**	**QN MS/VG**	**(+) (a)**	**2**			
		Cap: ratio height/diameter					
		low				Somycel 76	3
		medium				Broncoh, Sylvan A15	5
		high				Heirloom	7
15.		**QL VG**	**(+) (a)**	**2**			
		<u>Only varieties with brown cap</u>: Cap: Shade of scales compared to surface					
		lighter				Amycel 2400, Heirloom	1
		darker					9
16.	**(*)**	**QN MS/VG**	**(a), (b), (f)**	**2**			
		Cap: thickness in longitudinal section					
		thin				J10263	3
		medium				Broncoh, Horronda	5
		thick				Sylvan A15	7

		English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
17.	**(*)**	**QN**	**VG**	**(+)**	**(a)**	**2**			
		Cap: scaling							
		absent or very weak						Somycel 53	1
		weak						Horwitu	3
		medium						Heirloom, Horronda	5
		strong						Somycel 76	7
		very strong						Broncoh	9
18.		**QN**	**VG**	**(+)**	**(a)**	**2**			
		Cap: thickness of veil							
		thin						J10263	1
		medium							2
		thick						Horronda, Sylvan A15	3
19.	**(*)**	**PQ**	**VG**		**(d)**	**3**			
		Gills: color							
		pink						BP-1	1
		light brown						Horronda, Horwitu	2
		dark brown						Broncoh	3
20.		**QL**	**VG**	**(+)**	**(d)**	**3**			
		<u>Only varieties with brown cap</u>: Veil: annulus color							
		white						Amycel 2400, Sylvan 800	1
		brown						Brawn, Heirloom	2
21.	**(*)**	**QL**	**VG**	**(+)**	**(d)**				
		Basidium: spores							
		absent						J10263	1
		present						Sylvan A15	9
22.		**QN**	**MG**			**4**			
		Time of opening of cap							
		early						Horwitu	3
		medium						Amycel 2400, Sylvan A15	5
		late						Brawn, Heirloom	7

		English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
23.	**(*)**	**QN**	**VG**		**(c), (e)**	**5**			
		Open cap: stipe distance from base to annulus							
		short						Amycel 2400	3
		medium						Broncoh	5
		long						Horwitu	7
24.	**(*)**	**QN**	**MS/VG**		**(c), (e)**	**5**			
		Open cap: diameter							
		small						Horwitu	3
		medium						Broncoh, Sylvan A15	5
		large						Amycel 2400, Heirloom	7
25.	**(*)**	**QN**	**MS/VG**		**(c), (e)**	**5**			
		Open cap: thickness					**Sombrero abierto: espesor**		
		thin						J10263	3
		medium						Horwitu, Sylvan A15	5
		thick						Brawn, Heirloom	7
26.	**(*)**	**QN**	**VG**		**(c)**	**5**			
		Open cap: fraying of margin							
		absent or weak						Amycel 2400, J10263	1
		moderate						Broncoh, Horwitu	2
		strong						ML0406	3
27.	**(*)**	**PQ**	**VG**		**(c)**	**5**			
		Open cap: shape of central part of upper side							
		rounded						Euromycel 58, ML1496	1
		plane						Heirloom	2
		depressed						Broncoh	3

8. Explanations on the Table of Characteristics

8.1 Explanations covering several characteristics

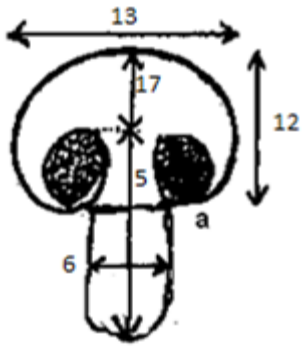
Characteristics containing the following key in the second column of the Table of Characteristics should be examined as indicated below:

(a) Stipe, cap: Observations on the stipe and the cap should be made at growth stage 2, when the fruit body appears as a button mushroom with the veil closed

(b)

side view

longitudinal section



a: veil

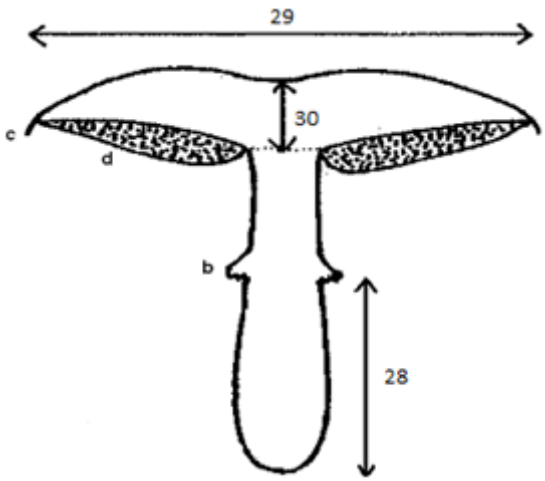
5: Stipe: length
6: Stipe: diameter
12: Cap: height
13: Cap: diameter
16: Cap: thickness in longitudinal section

BUTTON

(c) Open cap: Observations on the open cap should be made at growth stage 5, when the cap of the fruit body is fully open and flat

(d) Gills: Observations on the gills should be made at growth stage 3, when the fruit body appears as a button mushroom with the veil breaking.

(e)



longitudinal section

b: annulus
c: cap border
d: gills

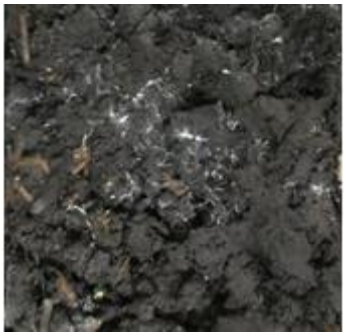
23: Stipe: distance from stipe to annulus
24: Open cap: diameter
25: Open cap: thickness

FLAT/FULLY SPREAD

(f) The fruit bodies observed at growth stage 2 should be cut longitudinally.

8.2 Explanations for individual characteristics

Ad. 1: Mycelium: intensity



1
weak

2
medium

3
strong

Ad. 2: Number of pins

A pin is a young primordial fruit body. The number of pins larger than 3 mm is visually observed 4 days after aeration.

Ad. 3: Time beginning of harvest

The time of the first day of harvest is recorded when more than 5 fruiting bodies in the first flush have reached growth stage 2.

Ad. 4: Time of peak of first flush

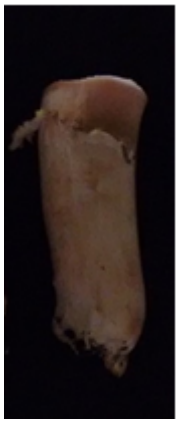
The dates of harvest of fruit bodies at growth stage 2 are recorded. The time of the peak of the first flush is the time at which the largest number of fruit bodies is harvested.

Ad. 6: Stipe: diameter

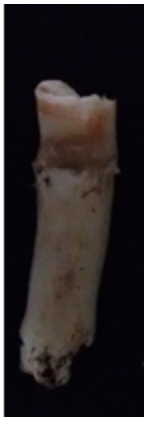
To be observed in the middle of the stipe.

Ad. 7: Stipe: ratio length/diameter

3
low



5
medium



7
high

Ad. 10: Only varieties with brown cap: Stipe: color

The stipe color is observed at harvest.

Ad. 11: Stipe: oxidation at cutting edge

The stipes are cut transversally in the middle. Oxidation of the cutting edge should be observed 2 to 10 minutes after cutting.

Ad. 14: Cap: ratio height/diameter

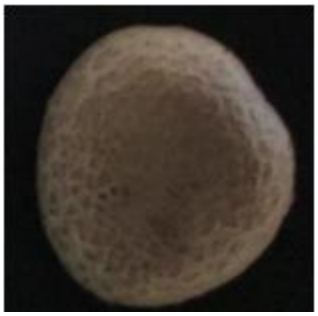
3
low

5
medium

7
high

Ad. 15: Only varieties with brown cap: Cap: Shade of scales compared to surface

1
lighter



9
darker

Ad. 17: Cap: scaling

1
absent or very weak

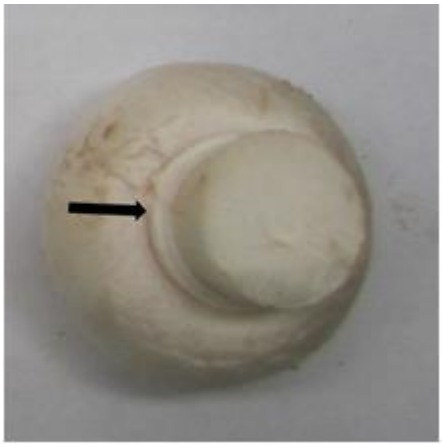
3
weak

5
medium

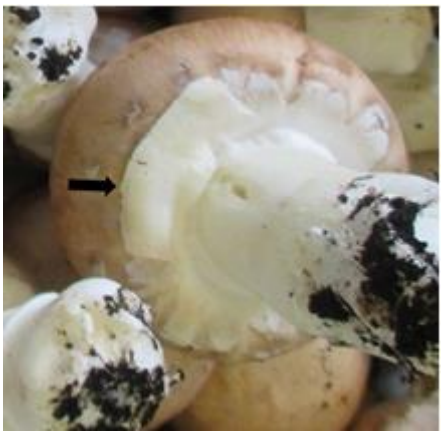
7
strong

9
very strong

Ad. 18: Cap: thickness of veil



1
thin



3
thick

Ad. 20: Only varieties with brown cap: Veil: annulus color

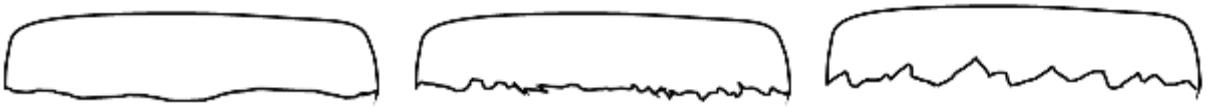
1
white

2
brown

Ad. 21: Basidium: spores

To be observed by making a sporeprint according to the methodology described by Singer (1986). If spores are formed, a sporeprint can be obtained by allowing a stage 3 fruiting body to ripen at room temperature above a sheet of white paper, which is placed below the gills. Spores of a fungal body fall onto the surface of the paper underneath. Presence of spores is reveled after two days, when a clear black-brown print on the paper has been obtained.

Ad. 26: Open cap: fraying of margin



1
absent or weak

2
moderate

3
strong

Ad. 27: Open cap: shape of central part of upper side

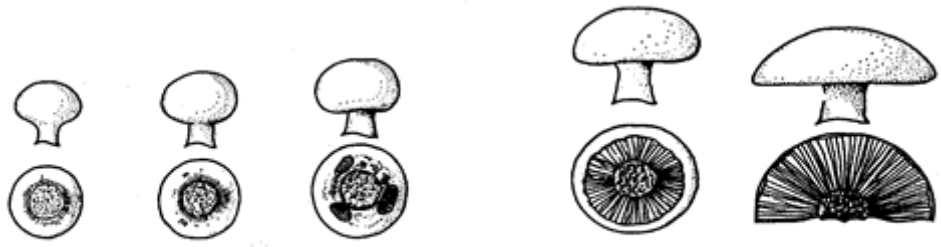
1
rounded

2
plane

3
depressed

8.3 Growth Stages and life cycle of *Agaricus bisporus*

Growth Stages

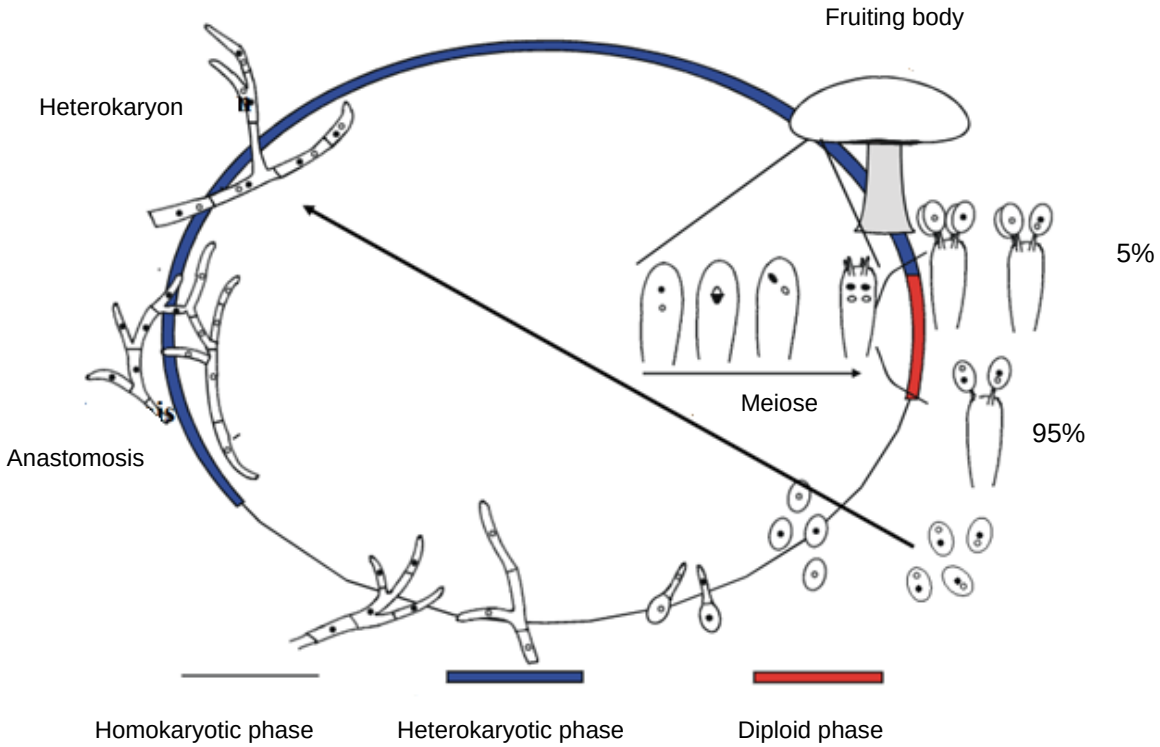


1 2 3 4 5

Explanation:

1, 2 and 3	-	button stage
1 and 2	-	veil closed
3	-	veil breaking
4	-	opening/gills visible
5	-	fully open/flat stage

Life cycle of *Agaricus bisporus*



9. Literature

Flegg, P.B., Spencer, D.M. and Wood, D.A., 1985: The biology and technology of the cultivated mushroom. J. Wiley & Son, 347 pp.

Fletcher, J.T. & Gaze R.H., 2007: Mushroom growing. In: Mushroom pest and disease control: a colour handbook, Manson Publishing Ltd, pp. 7-21.

Foulongne-Oriol., M, Rodier, A., Caumont, P., Spataro, C., Savoie, J.M., 2011: Agaricus bisporus cultivars: hidden diversity beyond apparent uniformity? In: Proceedings of the 7th international conference on mushroom biology and mushroom products, vol 2. pp 9–16.

Fritsche, G., 1964: Versuche zur Frage der Merkmalsübertragung beim Kulturchampignon Agaricus (Psalliota) bisporus (Lge.) Sing. Der Züchter 34-2: 76-93.

Fritsche, G., 1988: Spawn: properties and preparation, In: The Cultivation of Mushrooms, Darlington Mushroom Laboratories, pp. 91-99.

Neut, A. van der, 1991: The development of a set of characteristics for DUS tests of cultivated mushroom varieties. In: Genetics and breeding of Agaricus, Pudoc Wageningen, pp. 153-160.

Nichols, 1985. Post-harvest physiology and storage. Pp 195-210. In: Flegg P.B., Spencer D.M., Wood D.A. 1985: The biology and technology of the cultivated mushroom. J. Wiley & Son, 347 pp.

Parra Sánchez L.A. 2008: Fungi Europaei. Agaricus L. – Allopsalliota vol 1. Candusso Edizioni, 824 pp.

Parra Sánchez L.A., 2013: Fungi Europaei. Agaricus L. – Allopsalliota vol 2, Candusso Edizioni, 1168 pp.
Singer, R. 1986. The Agaricales in modern taxonomy, 4th edition. Koelts, Koenigstein, DE.

Vooren, J.G. van de, Polder, G. & Heijden, G.W.A.M. van der, 1991: Application of image analysis for variety testing of mushroom. Euphytica 57: 245-250.

Vooren, J.G. van de, Polder, G. & Heijden, G.W.A.M. van der, 1992: Identification of mushroom cultivars using image analysis. Transactions of the ASAE 35-1: 347-350.

10. Technical Questionnaire

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

	Application date: (not to be filled in by the applicant)

TECHNICAL QUESTIONNAIRE
to be completed in connection with an application for plant breeders' rights

1. Subject of the Technical Questionnaire

1.1 Botanical name	*Agaricus bisporus* (Lange.) Sing.
1.2 Common name	Mushroom

2. Applicant

Name	
Address	
Telephone No.	
Fax No.	
E-mail address	
Breeder (if different from applicant)	

3. Proposed denomination and breeder's reference

Proposed denomination (if available)	
Breeder's reference	

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

#4. Information on the breeding scheme and propagation of the variety

4.1 Breeding scheme

Variety resulting from:

4.1.1 Crossing

(a) controlled cross [ ]

(please state parent varieties)

(b) partially known cross [ ]

(please state known parent variety(ies))

(c) unknown cross [ ]

4.1.2 Mutation [ ]

(please state parent variety)

4.1.3 Discovery and development [ ]

(please state where and when discovered and how developed)

4.1.4 Other [ ]

(please provide details)

Authorities may allow certain of this information to be provided in a confidential section of the Technical Questionnaire.

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

4.2 Method of propagating the variety

4.2.1 Vegetative propagation

(a) *In vitro* propagation [ ]
(b) Other (state method) [ ]

4.2.2 Other [ ]
(Please provide details)

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

5. Characteristics of the variety to be indicated (the number in brackets refers to the corresponding characteristic in Test Guidelines; please mark the note which best corresponds).

	Characteristics	Example Varieties	Note
5.1 (4)	**Time of peak of first flush**		
	very early		1 [ ]
	very early to early		2 [ ]
	early	Heirloom	3 [ ]
	early to medium		4 [ ]
	medium	Amycel 2400, Sylvan A15	5 [ ]
	medium to lated		6 [ ]
	late	Brawn, Euromycel 58	7 [ ]
	late to very late		8 [ ]
	very late		9 [ ]
5.2 (8)	**Cap: color**		
	white	Sylvan A15	1 [ ]
	greyish white	Somycel 76	2 [ ]
	brown	Amycel 2400	3 [ ]
5.3 (13)	**Cap: diameter**		
	very small		1 [ ]
	very small to small		2 [ ]
	small	Horwitu	3 [ ]
	small to medium		4 [ ]
	medium	Broncoh	5 [ ]
	medium to large		6 [ ]
	large	Heirloom, Sylvan A15	7 [ ]
	large to very large		8 [ ]
	very large		9 [ ]
5.4 (19)	**Gills: color**		
	pink	BP-1	1 [ ]
	light brown	Horronda, Horwitu	2 [ ]
	dark brown	Broncoh	3 [ ]
5.5 (21)	**Basidium: spores**		
	absent	J10263	1 [ ]
	present	Sylvan A15	9 [ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

6. Similar varieties and differences from these varieties

Please use the following table and box for comments to provide information on how your candidate variety differs from the variety (or varieties) which, to the best of your knowledge, is (or are) most similar. This information may help the examination authority to conduct its examination of distinctness in a more efficient way.

Denomination(s) of variety(ies) similar to your candidate variety	Characteristic(s) in which your candidate variety differs from the similar variety(ies)	Describe the expression of the characteristic(s) for the **similar** variety(ies)	Describe the expression of the characteristic(s) for **your** candidate variety
Example	*Cap: color*	*greyish white*	*brown*

Comments:

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

#7. Additional information which may help in the examination of the variety

7.1 In addition to the information provided in sections 5 and 6, are there any additional characteristics which may help to distinguish the variety?

Yes [ ] No [ ]

(If yes, please provide details)

7.2 Are there any special conditions for growing the variety or conducting the examination?

Yes [ ] No [ ]

(If yes, please provide details)

7.3 Other information

Authorities may allow certain of this information to be provided in a confidential section of the Technical Questionnaire.

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:

8. Authorization for release

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

Yes [ ] No [ ]

(b) Has such authorization been obtained?

Yes [ ] No [ ]

If the answer to (b) is yes, please attach a copy of the authorization.

9. Information on material to be examined or submitted for examination

9.1 The expression of a characteristic or several characteristics of a variety may be affected by factors, such as pests and disease, chemical treatment (e.g. growth retardants or pesticides), effects of tissue culture, different rootstocks, scions taken from different growth phases of a tree, etc.

9.2 The material should not have undergone any treatment which would affect the expression of the characteristics of the variety, unless the competent authorities allow or request such treatment. If the material has undergone such treatment, full details of the treatment must be given. In this respect, please indicate below, to the best of your knowledge, if the material to be examined has been subjected to:

(a)	Microorganisms (e.g. virus, bacteria, phytoplasma)	Yes [ ]	No [ ]
(b)	Chemical treatment (e.g. growth retardant, pesticide)	Yes [ ]	No [ ]
(c)	Tissue culture	Yes [ ]	No [ ]
(d)	Other factors	Yes [ ]	No [ ]

Please provide details for where you have indicated “yes”.

.....

10. I hereby declare that, to the best of my knowledge, the information provided in this form is correct:

Applicant’s name

Signature Date

[End of document]