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UPOV

INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

GENEVA

DRAFT

AGARICUS MUSHROOM[1] *

UPOV Code: AGARI

(*Agaricus bisporus* L.
Agaricus bitorquis L.
Agaricus arvensis L.
~~*Agaricus campestris* L.~~)

GUIDELINES

FOR THE CONDUCT OF TESTS

FOR DISTINCTNESS, UNIFORMITY AND STABILITY

prepared by an expert from the European Community

to be considered by the Technical Working Party for Vegetables
at its forty-first session, to be held in Nairobi, Kenya, from June 11 to 15, 2007

Alternative Names:*

Latin	*English*	*French*	*German*	*Spanish*
Agaricus bisporus L. *Agaricus bitorquis* L. *Agaricus arvensis* L. ~~*Agaricus campestris* L.~~	Agaricus Mushroom, Mushroom, Button 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.

[1] Proposal by HU to change terminology to BUTTON MUSHROOM

* These names were correct at the time of the introduction of these Test Guidelines but may be revised or updated. [Readers are advised to consult the UPOV Code, which can be found on the UPOV Website (www.upov.int), for the latest information.]

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1. Subject of these Test Guidelines

These Test Guidelines apply to all varieties of *Agaricus bisporus* L., *Agaricus bitorquis* L., and *Agaricus arvensis* L. ~~and *Agaricus campestris*~~ L. (Agaricaceae) (especially 'white and/or brown button mushroom')

2. Material Required

2.1 The competent authorities decide on the quantity and quality of the fungal ~~plant~~ 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 as a pure culture on a suitable medium.

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

1 litre[2] of spawn or 2 slant tubes containing a pure culture.

2.4 The quality of the material to be delivered should not be below the standards of commercial spawn for marketing in the country concerned, especially in regard to the quantity of hyphae. Mycelium on grain should be visible to the naked eye, the grain should not be colonized to such an extent that kernels stick together. The spawn should not be older than 6 months and having been stored under proper conditions (i.e. 2-4 °C).

2.5 The spawn 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.

2.6 The pure cultures must be shipped on slant agar tubes with appropriate medium such as PDA (peptose dextrose agar) or Malt extract agar. Tubes should be covered by cotton plugs or plastic caps allowing sterile air diffusion. Cultures should be fresh, i.e. not stored for longer than 2 weeks at low temperature.

3. Method of Examination

3.1 Duration of Tests

The minimum duration of tests should normally be two independent growing cycles (see also: Additional information: Life cycle of *Agaricus* on page …).

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".

[2] Proposal by HU to have quantity increased to 2-3 litres

3.3 Conditions for Conducting the Examination

3.3.1 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.

3.3.2 The recommended method of observing the characteristic is indicated by the following key in the second column of the Table of Characteristics:

MG: single measurement of a group of plants or 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.

3.4 Test Design

3.4.1 Each test should be designed to result in a total of at least 180 fruit bodies, which should preferably be divided between 6 replicates.

3.4.2 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.

3.5 Number of Plants / Parts of Plants to be Examined

Unless otherwise indicated, all observations should be made on 30 fruit bodies or parts taken from each of 30 fruit bodies per replicate.[3]

3.6 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

[3] Remark from HU as to whether observations should be made on the first or second flush, or both

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.

4.2 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, a population standard of 1% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 180 mushrooms, 4 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 tested, either by growing a further generation, or by testing a new spawn stock to ensure that it exhibits the same characteristics as those shown by the previous 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) Stipe: shape in longitudinal section (characteristic 5)

(b) Cap: shape in longitudinal section (characteristic 12)
(c) Cap: color (characteristic 15)
(d) Open Cap: central part of upper side (characteristic 21)

5.4 Guidance for the use of grouping characteristics, in the process of examining distinctness, is provided through the General Introduction.

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

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.

6.3 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

(*) Asterisked characteristic – see Chapter 6.1.2

QL: Qualitative characteristic – see Chapter 6.3
QN: Quantitative characteristic – see Chapter 6.3
PQ: Pseudo-qualitative characteristic – see Chapter 6.3

MG, MS, VG, VS: See Chapter 3.3.2

(a) – (b) See Explanations on the Table of Characteristics in Chapter 8.1

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

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

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
1. **(*)** **(+)**	**MG**	**Basidium: number of spores**					
	(b)	two				Broncoh, Horronda, Horwitu	2
QL		between 2 and 4					3
		four				Horbita, Horvensis	4
2.	**MS**	**Stipe: length**					
(+)							
QN	**(a)**	**short**				Horwitu, Le Lion C9	3
		medium				Broncoh, Le Lion B86, Somycel 76	5
		long				Somycel 53	7
3.	**MS**	**Stipe: diameter**					
(+)							
QN	**(a)**	small				Somycel 91	3
		medium				Broncoh, Somycel 76	5
		large				Horronda, Horwitu, Le Lion C9	7
4.	**MS**	**Stipe: ratio length/diameter**					
QN	**(a)**	small					3
		medium				Le Lion C9	5
		large				Broncoh	7
5. **(*)** **(+)**	**VG**	**Stipe: shape in longitudinal section**					
PQ	**(a)**	rectangular				Horronda, Horvensis	1
		~~narrow~~ trapezoid				Horwitu	2

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
6.	**VG**	**Stipe: swollen base in longitudinal section**					
(+)							
QL	**(a)**	absent				Horronda	1
		present				Horbita	9
7.	**VG**	**Stipe: intensity of swelling of base**					
QN	**(a)**	weak				Broncoh	3
		medium				Horbita	5
		strong					7
8.	**VG**	**Stipe: distance from base to veil remnant ring**					
(+)							
QN	**(a)**	short				Commissaris Cremers, Le Lion C9	3
		medium				Broncoh, Horbita	5
		long				Horvensis	7
9.	**MS**	**Cap: height**					
(+)							
QN	**(a)**	short					3
		medium				Broncoh	5
		tall					7
10.	**MS**	**Cap: diameter**					
(+)							
QN	**(a)**	small				Commissaris Cremers	3
		medium				Broncoh, Somycel 76	5
		large				Horronda	7
11.	**MS**	**Cap: ratio height/diameter**					
QN	**(a)**	small				Le Lion C9	3
		medium				Broncoh	5
		large					7

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
12. **(*)** **(+)**	**VG**	**Cap: shape in longitudinal section**					
PQ	**(a)**	obovate				Horvensis	1
		circular				Commissaris Cremers, Horronda	2
		transverse elliptic				Broncoh, Horwitu	3
13. **(+)**	**MS**	**Cap: thickness in longitudinal section**					
QN	**(a)**	thin				Le Lion B86, Somycel 76	3
		medium				Broncoh, Horronda	5
		thick				Commissaris Cremers	7
14. **(+)**	**VG**	**Cap: amount of scales**[4]					
QN	**(a)**	absent or very low				Somycel 91, Royal 70, Royal 75	1
		low				Horronda, Le Lion X13, Royal 24A	3
		medium				Horwitu	5
		high				Somycel 76	7
		very high					9
15. **(*)**	**VG**	**Cap: color**					
PQ	**(a)**	white				Royal 75, Somycel 91	1
		greyish white				Claron A3.01, Somycel 76	2
		yellowish white				Horvensis	3
		brown				Broncoh, Le Lion C9	4

[4] Proposal by HU to delete characteristics

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
16. (*)	**MG**	**Cap: firmness[5]**					
QN		soft					3
		medium				Le Lion C9	5
		firm					7
17. (+)	**VG**	**Gills: color at time of breaking of the veil**					
PQ	**(a)**	pink					1
		orange				Horvensis	2
		light brown				Horronda, Horwitu	3
		dark brown				Broncoh	4
18. (+)	**MS**	**Open Cap: diameter**					
QN	**(b)**	small				Le Lion X13, Royal 75	3
		medium				Royal 20A	5
		large				Broncoh, Somycel 76	7
19. (+)	**MS**	**Open Cap: thickness**					
QN	**(b)**	thin					3
		medium				Broncoh, Horwitu, Le Lion X13	5
		thick				Claron A5.1, Somycel 205	7

[5] Proposal by HU to delete characteristics

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
20. (*)	**VG**	**Open Cap: margin**					
QN	**(b)**	not frayed				Claron A5.1, Le Lion C9, Royal 26A	3
		partly frayed				Broncoh, Horwitu, Somycel 205	5
		frayed				Horronda	7
21. (*) (+)	**VG**	**Open Cap: central part of upper side**					
QL	**(b)**	rounded					1
		flat					2
		depressed				Broncoh	3
22.	**VG**	**Discoloration of cutting surface**					
QN	**(a)**	weak				Broncoh, Commissaris Cremers	3
		medium				Horbita	5
		strong					7
23. (*)	**MG**	**Flushing pattern: earliness of first flush**					
QN		early				Le Lion X13, Horwitu	3
		medium				Broncoh, Claron A5.1, Royal 26A	5
		late				Le Lion X20, Somycel 205	7
24.	**MG**	**Flushing pattern: duration of first flush**					
QN		short					3
		medium				Broncoh	5
		long					7

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
25. (*)	**MG**	**Flushing pattern: earliness of second flush**					
QN		early					3
		medium				Broncoh	5
		late					7
26.	**MG**	**Flushing pattern: duration of second flush**					
QN		short					3
		medium				Broncoh	5
		long					7
27. (*)	**MG**	**Fruit bodies: weight**[6]					
QN		low					3
		medium				Le Lion C9	5
		high					7

[6] remark by HU to delete characteristic

8. Explanations on the Table of Characteristics

8.1 Explanations covering several characteristics

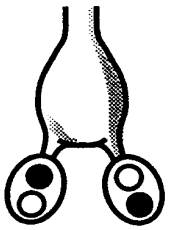
Characteristics containing the following key in the second column of the Table of Characteristics should be examined as indicated below:

(a) Stipe, cap and gills[7]: Unless otherwise indicated, all characteristics of the fruit bodies, the cap, the stipe and the gills should be recorded at harvest maturity (button stage 1, 2 and 3 [see annex page] hand picked mushrooms; freshly harvested).

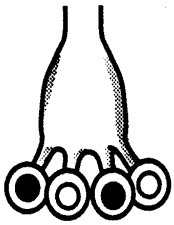
(b) Open cap: The characteristics of the open cap should be recorded as soon as the cap is fully spread (and not postponed until later date). Records should preferably be made from first and second flush; the third flush may give some additional information.

8.2 Explanations for individual characteristics

Ad 1: Basidium: number of spores



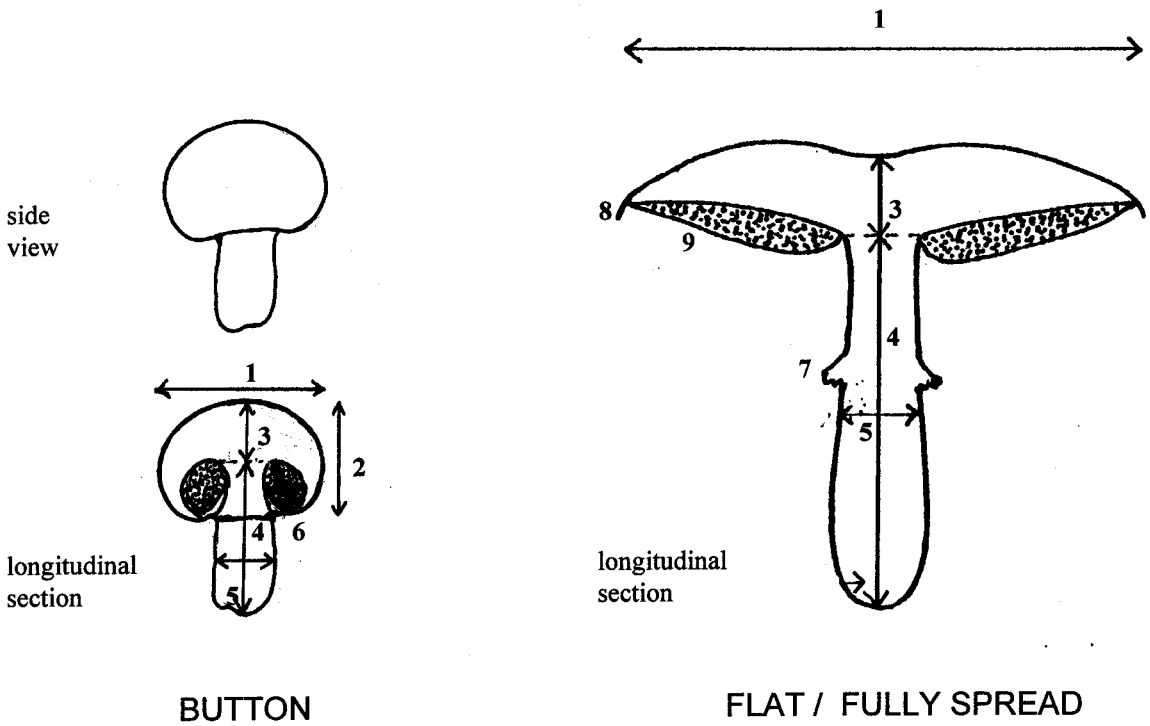
2
two



4
four

[7] Proposal by HU to replace by "Closed cap"

Ad 2, 3, 8, 9, 10, 13, 18, 19 and 20: Mushroom: side view and longitudinal sections



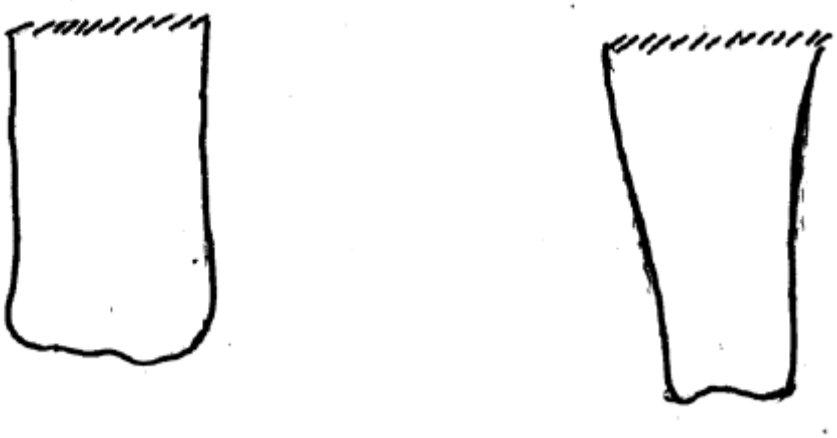
BUTTON

FLAT / FULLY SPREAD

Explanation:

1 - cap diameter
2 - cap height
3 - cap thickness
4 - stipe length
5 - stipe diameter
6 - veil
7 - veil remnant ring
8 - cap border
9 - gills

Ad. 5: Stipe: shape in longitudinal section



1
rectangular

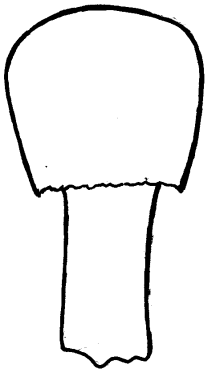
2
trapezoidal

Ad. 6: Stipe: swollen base in longitudinal section

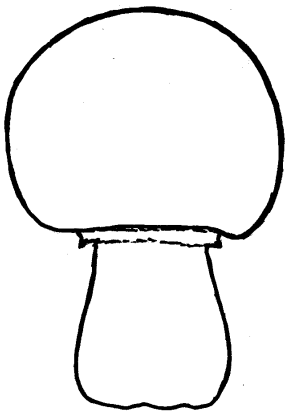
1
absent

9
present

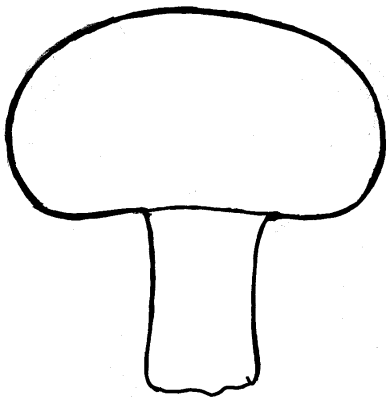
Ad. 12: Cap: shape in longitudinal section



1
obovate

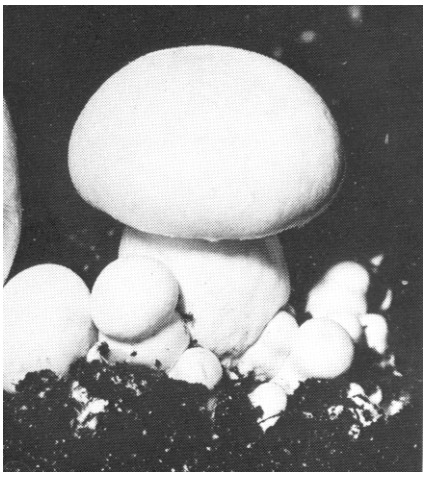


2
circular



3
transverse elliptic

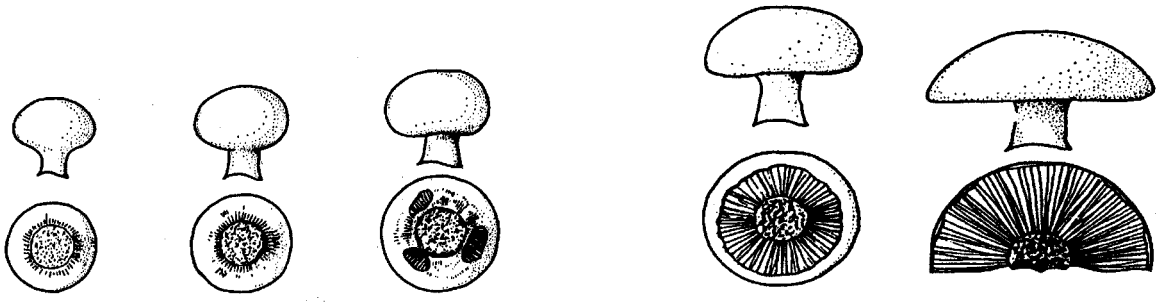
Ad 14: Cap: amount of scales



1
absent of very low

9
very high

Ad. 17 and 20: Veil and Gills: (from below)



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

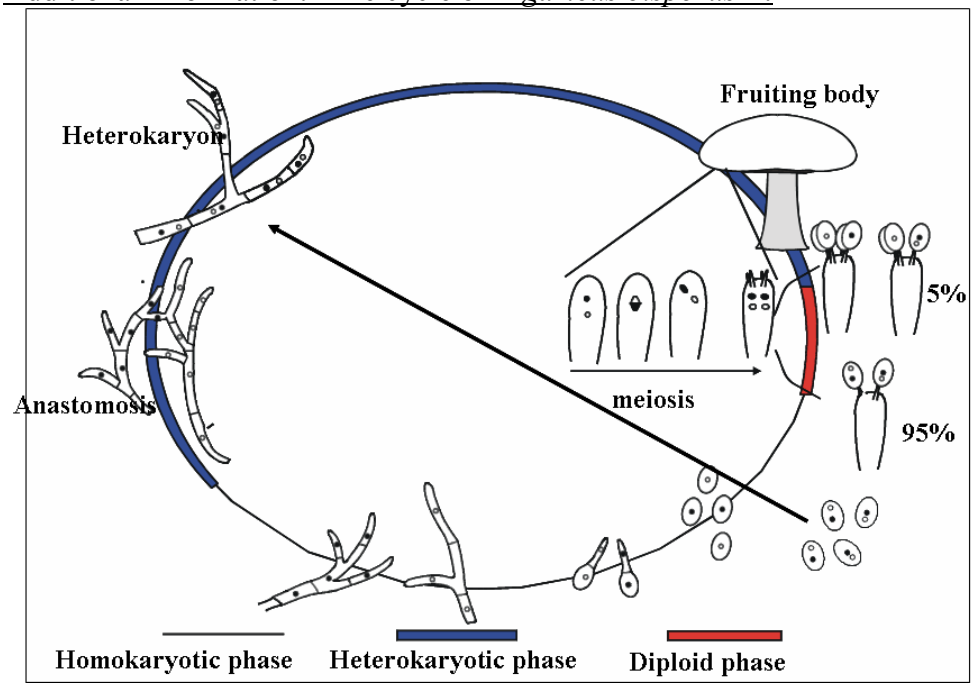
Ad. 21: Open Cap: central part of upper side

1
rounded

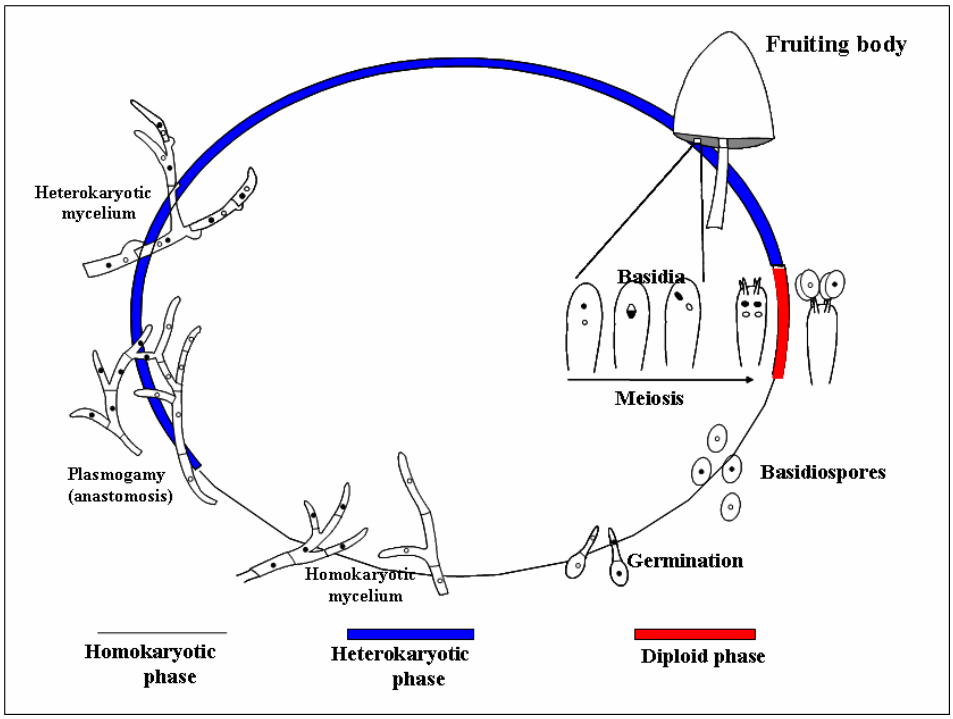
2
flat

3
depressed

Additional information: Life cycle of *Agaricus bisporus* L.



Life cycle of *Agaricus bisporus* var. *bisporus* L.



Life cycle of *Agaricus bitorquis* L.

9. Literature

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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* L. *Agaricus bitorquis* L. *Agaricus arvensis* L. ~~*Agaricus campestris*~~ L.
1.2 Common Name	Agaricus 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)

4.2 Method of propagating the variety

4.2.1 Vegetative propagation

(a) cuttings [ ]

(b) *in vitro* propagation [ ]

(c) other (state method) [ ]

4.2.2 Seed [ ]

4.2.3 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 (1)	**Basidium: number of spores**		
	two	Broncoh, Horronda, Horwitu	2[ ]
	between 2 and 4		3[ ]
	four	Horbita, Horvensis	4[ ]
5.2 (5)	**Stipe: shape in longitudinal section**		
	rectangular	Horronda, Horvensis	1[ ]
	narrow trapezoid	Horwitu	2[ ]
5.3 (12)	**Cap: shape in longitudinal section**		
	obovate	Horvensis	1[ ]
	circular	Commissaris Cremers, Horronda	2[ ]
	transverse elliptic	Broncoh, Horwitu	3[ ]
5.4 (15)	**Cap: color**		
	white	Royal 75, Somycel 91	1[ ]
	greyish white	Claron A3.01, Somycel 76	2[ ]
	pale yellowish	Horvensis	3[ ]
	brown	Broncoh, Le Lion C9	4[ ]
5.5 (21)	**Open Cap: central part of upper side**		
	rounded		1[ ]
	flat		2[ ]
	depressed	Broncoh	3[ ]

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

	Characteristics	Example Varieties	Note
5.6 (23)	**Flushing pattern: earliness of first flush**		
	early	Le Lion X13, Horwitu	3[ ]
	medium	Broncoh, Claron A5.1, Royal 26A	5[ ]
	late	Le Lion X20, Somycel 205	7[ ]

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			

[#]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.

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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 plant 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 plant 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 plant 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 plant 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]