

UPOV

TG/MUSHROOM (proj.1)

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## INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

GENEVA

DRAFT

## AGURICUS MUSHROOM \*

*Agaricus bisporus* L.  
*Agaricus bitorquis* L.  
*Agaricus arvensis* L.)

## GUIDELINES

## FOR THE CONDUCT OF TESTS

## FOR DISTINCTNESS, UNIFORMITY AND STABILITY

*to be considered by the  
 Technical Working Party for Vegetables at its thirty-eighth session,  
 to be held in Seoul, from June 7 to 11, 2004*

Alternative Names: \*

<i>Latin</i>	<i>English</i>	<i>French</i>	<i>German</i>	<i>Spanish</i>
<i>Agaricus bisporus</i> L. <i>Agaricus bitorquis</i> L. <i>Agaricus arvensis</i> L.	Agaricus Mushroom	Champignon de couche	Champignon	Champiñón

## ASSOCIATED DOCUMENTS

These guidelines should be read in conjunction with document TG/1/3, “General Introduction to the Examination of Distinctness, Uniformity and Stability and the Development of Harmonized Descriptions of New Varieties of Plants” (hereinafter referred to as the “General Introduction”) and its associated “TGP” documents.

\* 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](http://www.upov.int)), for the latest information.]

<u>TABLE OF CONTENTS</u>	<u>PAGE</u>
1. SUBJECT OF THESE TEST GUIDELINES.....	3
2. MATERIAL REQUIRED .....	3
3. METHOD OF EXAMINATION.....	3
3.1 Duration of Tests.....	3
3.2 Testing Place.....	3
3.3 Conditions for Conducting the Examination.....	3
3.4 Test Design .....	4
3.5 Number of Plants / Parts of Plants to be Examined.....	4
3.6 Additional Tests .....	4
4. ASSESSMENT OF DISTINCTNESS, UNIFORMITY AND STABILITY.....	4
4.1 Distinctness .....	4
4.2 Uniformity.....	5
4.3 Stability .....	5
5. GROUPING OF VARIETIES AND ORGANIZATION OF THE GROWING TRIAL.....	5
6. INTRODUCTION TO THE TABLE OF CHARACTERISTICS .....	6
6.1 Categories of Characteristics.....	6
6.2 States of Expression and Corresponding Notes.....	6
6.3 Types of Expression.....	6
6.4 Example Varieties .....	6
6.5 Legend.....	7
7. TABLE OF CHARACTERISTICS/TABLEAU DES CARACTÈRES/MERKMALSTABELLE/TABLA DE CARACTERES.....	8
8. EXPLANATIONS ON THE TABLE OF CHARACTERISTICS .....	13
8.1 Explanations covering several characteristics.....	13
8.2 Explanations for individual characteristics .....	13
9. LITERATURE.....	21
10. TECHNICAL QUESTIONNAIRE.....	22

## 1. Subject of these Test Guidelines

These Test Guidelines apply to all varieties of *Agaricus bisporus* L. , *Agaricus bitorquis* L. and *Agaricus arvensis* 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 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.

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

1 liter of spawn.

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.

## 3. Method of Examination

### 3.1 *Duration of Tests*

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

### 3.2 *Testing Place*

The tests should normally be conducted at one place. If any characteristics of the variety, which are relevant for the examination of DUS, cannot be observed at that place, the variety may be tested at an additional place.

### 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.

### 3.3.1 Type of observation – visual or measurement

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.

## 3.6 *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 minimum duration of tests recommended in section 3.1 reflects, in general, the need to ensure that any differences in a characteristic are sufficiently consistent.

#### 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.

#### 4.4 *Disease characteristics (not yet appropriate!!!)*

When disease resistance characteristics are used for assessing distinctness, uniformity and stability, records must be taken under conditions of controlled infection with a defined pathotype. If applicable, all resistances should be tested separately on each race and separately for each pathogen.

### 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 is 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 11)
- (c) Cap: color (characteristic 14)
- (d) Open Cap: central part of upper side (characteristic 19)

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*

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

6.4.2 Unless otherwise indicated, all example varieties mentioned in the Table of Characteristics represent the corresponding state of expression under standardised growing conditions. The variety descriptions preferably should, especially in case of this crop, state

the standardised growing conditions, such as temperature, CO<sub>2</sub>-level and relative humidity conditions, as well as the cultivation system.

#### 6.5 *Legend*

(\*) Asterisked characteristic – see Section 6.1.2

(QL) Qualitative characteristic – see Section 6.3

(QN) Quantitative characteristic – see Section 6.3

(PQ) Pseudo-qualitative characteristic – see Section 6.3

(a) – (b) See Explanations on the Table of Characteristics in Chapter 8, Section 8.1

(+) See Explanations on the Table of Characteristics in Chapter 8, Section 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
<b>1.</b> (*)	<b>Basidium: number of spores</b>					
(+)	<b>(b)</b>	two			Horronda, Horwitu	2
	or later ?	between 2 and 4				3
	<b>MG</b>	four			Horbita, Horvensis	4
<b>2.</b>	<b>Stipe: length</b>					
(+)	<b>(a)</b>	short			Horwitu	3
		medium			Le Lion B86, Somycel 76	5
	<b>MS</b>	long			Somycel 53	7
<b>3.</b>	<b>Stipe: diameter</b>					
(+)	<b>(a)</b>	small			Somycel 91	3
		medium			Somycel 76	5
	<b>MS</b>	large			Horronda, Horwitu	7
<b>4.</b>	<b>Stipe: ratio length/diameter</b>					
	<b>(a)</b>	small				3
		medium				5
	<b>MS</b>	large				7
<b>5.</b> (*)	<b>Stipe: shape in longitudinal section</b>					
(+)	<b>(a)</b>	rectangular			Horronda, Horvensis	1
	<b>VG</b>	narrow trapezoid			Horwitu	2



	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>6.</b>	<b>Stipe: swollen base in longitudinal section</b>					
(+)	(a)	absent			Horronda	1
	<b>VG</b>	present			Horbita	3
<b>7.</b>	<b>Stipe: distance from base to veil remnant ring</b>					
(+)	(a)	short			Commissaris Cremers	3
		medium			Horbita	5
	<b>VG</b>	long			Horvensis	7
<b>8.</b>	<b>Cap: height</b>					
(+)	(a)	short				3
		medium				5
	<b>MS</b>	tall				7
<b>9.</b>	<b>Cap: diameter</b>					
(+)	(a)	small			Commissaris Cremers	3
		medium			Somycel 76	5
	<b>MS</b>	large			Horronda	7
<b>10.</b>	<b>Cap: ratio height/diameter</b>					
	(a)	small				3
		medium				5
	<b>MS</b>	large				7
<b>11.</b>	<b>Cap: shape in longitudinal section</b>					
(*)						
(+)	(a)	obovate			Horvensis	1
		circular			Commissaris Cremers, Horronda	2
	<b>VG</b>	transverse elliptic			Horwitu	3

	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>12.</b>	<b>Cap: thickness in longitudinal section</b>					
(+)	(a)	thin			Le Lion B86, Somycel 76	3
		medium			Horronda	5
	<b>MS</b>	thick			Commissaris Cremers	7
<b>13.</b>	<b>Cap: amount of scales</b>					
(+)	(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
	<b>VG</b>	very high				9
<b>14.</b>	<b>Cap: color</b>	<b>xx: couleur</b>	<b>xx: Farbe</b>			
(*)	(a)	white	blanche	weiss	Royal 75, Somycel 91	1
		greyish white			Claron A3.01, Somycel 76	2
		pale yellowish			Horvensis	3
	<b>VG</b>	brown			Le Lion C9	4
<b>15.</b>	<b>Gills: color at time of breaking of the veil</b>					
(+)	(a)	pink			<i>Ex.var's</i>	1
	<b>or (a/b)?</b>	pale orange			Horvensis	2
		light brown			Horronda, Horwitu	3
	<b>VG</b>	dark brown			<i>Ex.var's</i>	4

	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>16.</b>	<b>Open Cap: diameter</b>	<i>Delete ?</i>				
(+)	(b)	small			Le Lion X13, Royal 75	3
		medium			Royal 20A	5
	<b>MS</b>	large			Somycel 76	7
<b>17.</b>	<b>Open Cap: thickness</b>	<i>Delete ?</i>				
(+)	(b)	Thin				3
		Medium			Horwitu, Le Lion X13	5
	<b>MS</b>	thick			Claron A5.1, Somycel 205	7
<b>18. (* )</b>	<b>Open Cap: margin</b>					
	(b)	not frayed			Claron A5.1, Royal 26A	3
		partly frayed			Horwitu, Somycel 205	5
	<b>VG</b>	frayed			Horronda	7
<b>19. (* )</b>	<b>Open Cap: central part of upper side</b>					
(+)	(b)	rounded			<i>Ex.Var's</i>	1
		flat				2
	<b>VG</b>	depressed				3
<b>20.</b>	<b>Discoloration of cutting surface</b>					
	(a)	weak			Commissaris Cremers	3
		medium			Horbita	5
	<b>VG</b>	strong				7

	English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
<b>21.</b> (*)	<b>Flushing pattern: earliness of first flush</b>					
	early				Le Lion X13, Horwitu	3
	medium				Claron A5.1, Royal 26A	5
	<b>MG</b> late				Le Lion X20, Somycel 205	7
<b>22.</b>	<b>Flushing pattern: duration of first flush</b>					
	short					3
	medium					5
	<b>MG</b> long					7
<b>23.</b> (*)	<b>Flushing pattern: earliness of second flush</b>					
	early					3
	medium				<i>Ex.Var's</i>	5
	<b>MG</b> late					7
<b>24.</b>	<b>Flushing pattern: duration of second flush</b>					
	short					3
	medium					5
	<b>MG</b> long					7

8. Explanations on the Table of Characteristics

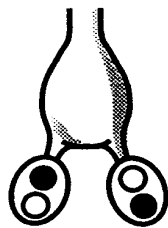
8.1 *Explanations covering several characteristics*

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

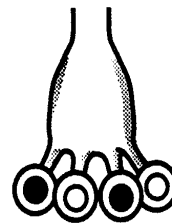
- (a) Stipe, cap and gills: Unless otherwise indicated, all characteristics of the fruit bodies, the cap, the stipe and the gills should be recorded at harvest maturity (button stage 1, 2 and 3 [see annex page] hand picked mushrooms; freshly harvested).
- (b) Open cap: The characteristics of the open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



3  
between 2 and 4

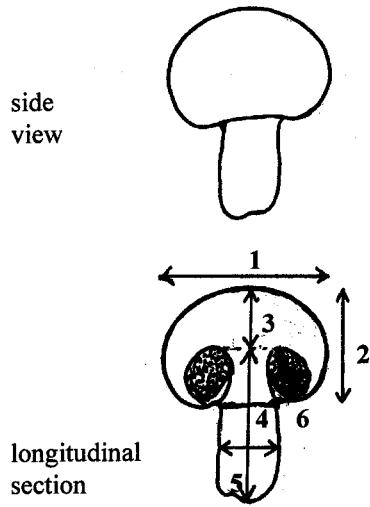
4  
four

Ad 2, 3, 7, 8, 9, 12, 16, 17 and 18: Mushroom: side view and longitudinal sections

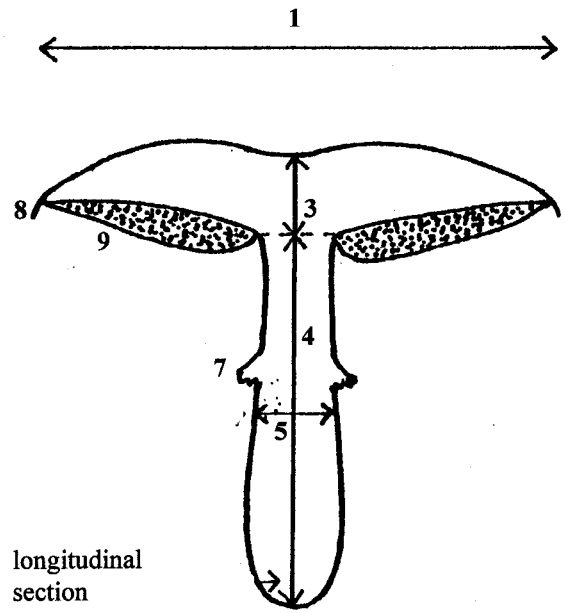
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Mushroom, 2003-05-23

- 15 -



**BUTTON**



**FLAT / FULLY SPREAD**

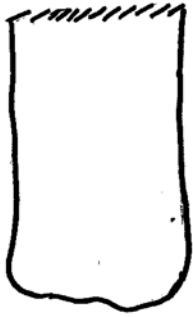
Explanation:

- 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 and 6: Stipe: shape in longitudinal section (5), swollen base (6)

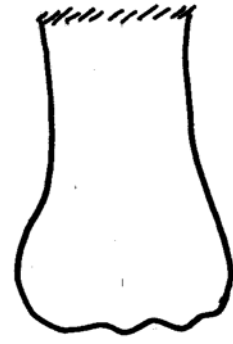




1  
rectangular

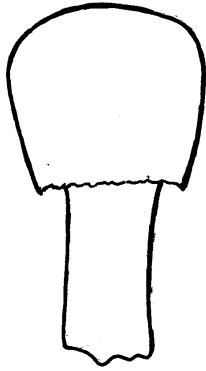


2  
trapezoid

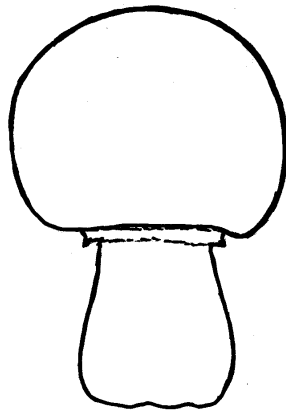


9  
swollen base  
present

Ad. 11: Cap: shape in longitudinal section



1  
obovate

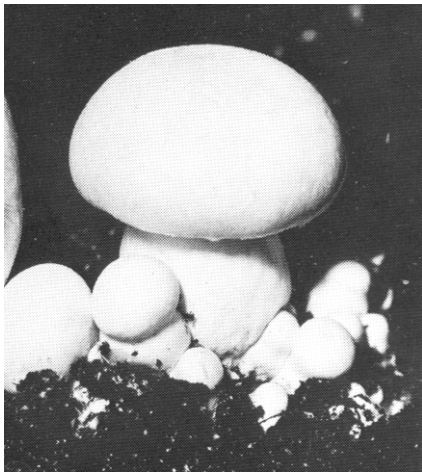


2  
circular



3  
transverse elliptic

Ad 13: Cap: amount of scales

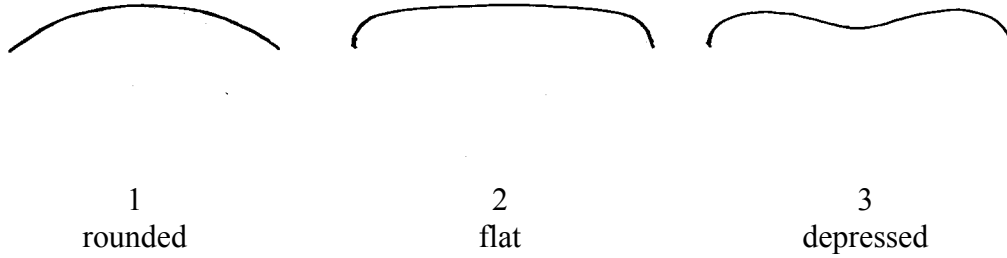


1  
absent of very low

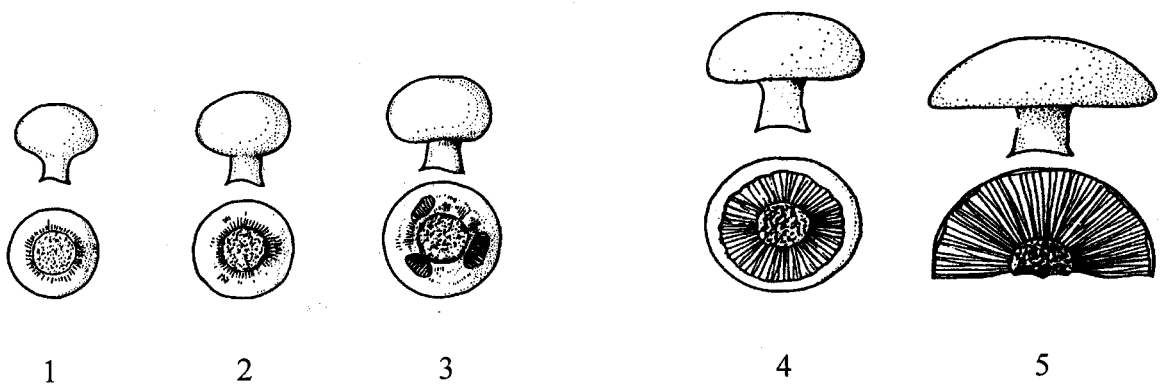


9  
very high

Ad. 19: Open Cap: central part of upper side



Ad. 15 and 18: Veil and Gills: (from below)

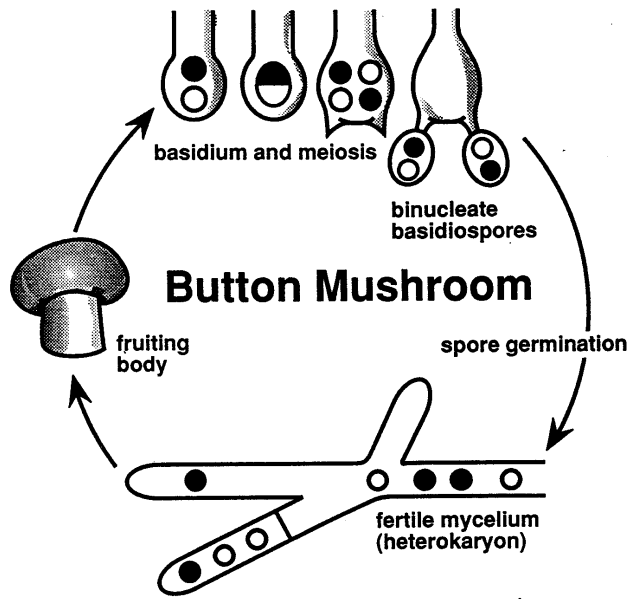


Explanation:

1, 2 and 3 - button stage  
1 and 2 - veil closed  
3 - veil breaking

4 - opening / gills visible  
5 - fully open / flat stage

Additional information: 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

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

Fritsche, G., 1979: "Breeding work with *Agaricus bitorquis*, Methods and Results of the Experimental Station in Horst," the Netherlands, Australian Mushroom Growers' Annual 2: 22-25.

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

Singer, R., 1986: "The *Agaricales* in Modern Taxonomy," Koeltz (Ger.), 981 pp and 80 pl.

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.

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 Latin Name	<input type="text" value="Agaricus bisporus L."/> <input type="text" value="Agaricus bitorquis L."/> <input type="text" value="Agaricus arvensis L."/>	
1.2 Common Name	<input type="text" value="Agaricus Mushroom"/>	
2. Applicant		
Name	<input type="text"/>	
Address	<input type="text"/>	
Telephone No.	<input type="text"/>	
Fax No.	<input type="text"/>	
E-mail address	<input type="text"/>	
Breeder (if different from applicant)	<input type="text"/>	
3. Proposed denomination and breeder's reference		
Proposed denomination (if available)	<input type="text"/>	
Breeder's reference	<input type="text"/>	

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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#### 4. Information on the breeding scheme and propagation of the variety

##### 4.1 Breeding scheme **ASW 15**

[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) totally unknown cross [ ]

##### 4.1.2 Mutation [ ] (please state parent variety)

##### 4.1.3 Discovery [ ] (please state where, when and how developed)

##### 4.1.4 Other [ ] (please provide details)]

##### 4.2 Method of propagating the variety

#### 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
<b>5.1 Basidium: number of spores</b> (1)		
two	Horronda, Horwitu	2[ ]
between 2 and 4		3[ ]
four	Horbita, Horvensis	4[ ]
<b>5.2 Stipe: shape in longitudinal section</b> (5)		
rectangular	Horronda, Horvensis	1[ ]
narrow trapezoid	Horwitu	2[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:	
<b>5.3 Cap: shape in longitudinal section</b>			
<b>(11)</b>			
obovate	Horvensis		1[ ]
circular	Commissaris Cremers, Horronda		2[ ]
transverse elliptic	Horwitu		3[ ]
<b>5.4 Cap: color</b>			
<b>(14)</b>			
white	Royal 75, Somycel 91		1[ ]
greyish white	Claron A3.01, Somycel 76		2[ ]
pale yellowish	Horvensis		3[ ]
brown	Le Lion C9		4[ ]
<b>5.5 Open Cap: central part of upper side</b>			
<b>(19)</b>			
rounded	<i>Ex.Var's</i>		1[ ]
flat			2[ ]
depressed			3[ ]
<b>5.6 Flushing pattern: earliness of first flush</b>			
<b>(21)</b>			
early	Le Lion X13, Horwitu		3[ ]
medium	Claron A5.1, Royal 26A		5[ ]
late	Le Lion X20, Somycel 205		7[ ]



TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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6. Similar varieties and differences from these varieties

*Please use the table, and space provided for comments, below 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 <b>similar</b> variety(ies)	Describe the expression of the characteristic(s) for <b>your</b> candidate variety
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*Example* *(example to be inserted)* *(example to be inserted)*

Comments:

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 Special conditions for the examination of the variety

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

Yes [ ]                      No [ ]

7.2.2 If yes, please give details:

7.3 Other information

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

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 or pesticide) | Yes [ ] | No [ ] |
| (c) Tissue culture  | Yes [ ] | No [ ] |
| (d) Other factors   | Yes [ ] | No [ ] |

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

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

Applicant's name

Signature

Date