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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

GENEVA

DRAFT

HEMP

UPOV Code: CANNB_SAT

Cannabis sativa L.

GUIDELINES

FOR THE CONDUCT OF TESTS

FOR DISTINCTNESS, UNIFORMITY AND STABILITY

prepared by an expert from the Netherlands

to be considered by the Technical Working Party for Agricultural Crops at its thirty-eighth session, to be held in Seoul, Republic of Korea, from August 31 to September 4, 2009

Alternative Names:*

Botanical name	English	French	German	Spanish
Cannabis sativa L.	Hemp	Chanvre	Hanf	Cáñamo

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.

^{*} 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. <u>Subject of these Test Guidelines</u>

These Test Guidelines apply to all varieties of Cannabis sativa L.

2. <u>Material Required</u>

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 seed or young plants of sufficient size and with sufficient development to express all the characteristics of the variety in the first growing cycle.

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

Vegetatively propagated varieties: 50 young plants (potted, non-flowering).

Seed-propagated varieties: 500 grams of seed.

In the case of hybrid varieties an additional 200 grams of seed of each parental component should be submitted.

2.4 In the case of seed, the seed should meet the minimum requirements for germination, species and analytical purity, health and moisture content, specified by the competent authority.

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

2.6 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 it has been treated, full details of the treatment must be given.

3. <u>Method of Examination</u>

3.1 Number of Growing Cycles

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

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

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 Type of observation

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

3.3.3 Type of plot for observation

The recommended type of plot in which to observe the characteristic is indicated by the following key in the second column of the Table of Characteristics:

- A: field test (see 3.4)
- B: seedling test (seed-propagated varieties only)

3.4 Test Design

3.4.1 In case of open pollinated varieties, hybrids or inbred lines each test should be designed to result in a total of at least 200 plants, which should be divided between 2 replicates.

3.4.2 In case of vegetatively propagated varieties each test should be designed to result in a total of at least 40 plants.

3.4.3 The design of the tests should be such that plants or parts of plants may be removed for measurement or counting without prejudice to the observations which must be made up to the end of the growing cycle. As Hemp is very sensitive to environmental conditions seed propagated varieties are recommended to be grown at a plant density of about 40 plants per m^2 and observations on border plants should be avoided.

3.5 Number of Plants / Parts of Plants to be Examined

Unless otherwise indicated, all observations on single plants should be made on 40 plants or parts taken from each of 40 plants and any other observations made on all plants in the test.

3.6 Additional Tests

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

4. <u>Assessment of Distinctness, Uniformity and Stability</u>

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.

4.2 Uniformity

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:

Male plants may occur in seed-propagated monoecious varieties. For seed propagated, monoecious varieties a maximum of 5% male plants is accepted.

(a) Cross-pollinated varieties

The assessment of uniformity of open-pollinated varieties should be according to the recommendations for cross-pollinated varieties in the General Introduction.

(b) Inbred lines and hybrid varieties

For the assessment of uniformity of inbred lines and single hybrids, a population standard of 2% with an acceptance probability of 95% should be applied (male plants excluded). In the case of a sample size of 40 plants, 2 off-types are allowed. In case of a sample size of 200 plants, 7 off-types are allowed.

(c) Vegetatively propagated varieties

For the assessment of uniformity of vegetatively propagated 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 40 plants, 2 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 seed or plant stock to ensure that it exhibits the same characteristics as those shown by the previous material supplied.

5. <u>Grouping of Varieties and Organization of the Growing Trial</u>

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.

The following have been agreed as useful grouping characteristics:

- (a) Time of beginning of female flowering (characteristic 13)
- (b) Plant: sex expression (characteristic 16)
- (c) Plant: natural height (characteristic 18)

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

6. <u>Introduction to the Table of Characteristics</u>

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: field test

- B: seedling test
- (a)-(c) See Explanations on the Table of Characteristics in Chapter 8.1
- (+) See Explanations on the Table of Characteristics in Chapter 8.2

0003, etc. stage of observation: 4-digit code for growth stage – see Chapter 8.3

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

		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
1.	VG B	Cotyledon: shape					
QN	0003	narrow elliptic	Example variety Yuso	31 more commonly k	nown as Uso 31 ?	Carmen, Yuso 31	1
		medium elliptic					2
		broad elliptic				Ruby, Tegege	3
2.	VG B	Cotyledon: intensity of green color					
QN	0003	light					1
		medium					2
		dark				BundvGem	3
3.	VG B	Hypocotyl: intensity of anthocyanin coloration					
QN	0003	absent or very weak				Carmen	1
		weak				Hlukhivs'ki 18	3
		medium				Hlera	5
		strong				Zolotonos'ki 15	7
		very strong					9
4.	VG	Plant: coloration of	f				
(+)		the crown					
PQ	1006	yellow				Vellow Apex	1
		light green				Hlukhivs'ki 33	2
		green				Ermakivs'ki	3
		violet	(red) purple instead of	violet ?		Dniprovs'ki 14, BundyGem	4

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
5.	VG	Leaf: anthocyanin coloration	DE : independent f	rom 4 ?			
QN	10xx	absent or very weak				Anka, BundyGem, Carmen, Calavos, FibreGem, Kepnock	1
		weak					3
		medium					5
		strong					7
		very strong					9
6.	VG	Leaf: intensity of					
(+)		green color					
QN	(c)	light				Anka	1
	2101 2201 2301	medium				Kinai egylaki	2
		dark				Carmen, Tiborszállási	3
7. (*)	VG	Leaf: size					
QN	(a)	small				Finola	3
	2101 2201 2301	medium				Carmen	5
		large				BundyGem	7
8. (*) (+)	VG	Leaf: number of leaflets					
QN	(a)	few					1
	2101 2201 2301	medium				Finola	2
		many				BundyGem	3

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
9.	MS	Central leaflet: length					
QN	(a)	short				Fasamo	3
	2101 2201 2301	medium				Dniprovs'ki 11	5
		long				Hlukhivs'ki 10	7
10.	MS	Central leaflet: width	1				
QN	(a)	narrow				Fasamo	3
	2101 2201 2301	medium				Dniprovs'ki 18	5
		broad				Hlukhivs'ki	7
11.	MS	Petiole: length					
QN	(a)	short				Anka, Ermakivs'ki, Finola	1
	2101 2201 2301	medium				Hlukhivs'ki 57, Tegege	2
		long				Bundygem,Fibregem, Hlukhivs'ki 33	3
12. (*)	VG	Petiole: anthocyanin coloration					
QN	(a)	absent or very weak	Example varieties nee	eded (asterisked char.))		1
	2101 2201 2301	weak				Ruby	2
		medium					3
		strong					4
		very strong					5

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
13. (*) (+)	VG	Time of beginning of female flowering	Remarks from severa difficult to observe. F female flowers with s	l experts : beginnin R prefers : Time of tyle).	ng of female flowering is f flowering (i.e. 50% of		
QN	2201 2301	very early					1
		early				Finola	3
		medium				Tiborszállási	5
		late				Kompolti	7
		very late					9
14.	VG	Inflorescence: anthocyanin coloration of male flowers					
QN	2102 2304	absent or very weak				Kompolti	1
		weak				Carmen	3
		medium				Lovrin 110	5
		strong					7
		very strong					9

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
15. (+)	MG	Inflorescence: THC content	Proposed by seve characteristic as i Despite substanti hemp varieties, d or two (DUS) tria significant (smal	eral experts. NL, CZ ar it is very sensitive to en al differences between lifferences among fibre als are not enough to de l) differences.	nd DE are not in favour of this nvironmental conditions. fibre hemp and medicinal hemp varieties are small. One etermine consistent and	9	
QN	(b)	absent or very low				Hlera, Hlukhivs'ki 33, Santhica 23	1
		very low to low				Anka, BundyGem, Epsilon 68, FibreGem.	2
	2202 2203 2302 2305	low				Carmen, Férimon, Yuso 31	3
		low to medium				Calavos, Fibrimon 56, Kepnock, Ruby	4
		medium				Ermakivs'ki, Tegege	5
		high				Yus 9	7
		very high				Krasnodars'	9
16. (*) (+)	VG	Plant: sex expression					
QL	2102 2202 2302 2304	monoecious				Anka, Ermakivs'ki, Fibrimon 21	1
		dioecious				Kompolti, Yuso 31	2
		gynoecious				Uniko B	3
17.	VG	Plant: number of primary lateral branches	NL : to be deleted	d, as this characteristic	is sensitive to plant density		
QN	(b)(c)	none or very few				Carmen, BundyGem, FibreGem	1
	2202 2302	medium					2
		many					3

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
18. (*) (+)	VG/ MG	Plant: natural height					
QN	(b)	short				Carmen, Finola, Yuso 31,	3
	2202 2302	medium				Hlukhivs'ki 33	5
		long				Dniprovs'ki 11	7
19. (*)	VG	Main stem: color					
PQ	(b)(c)	yellow				Chameleon, Hlukhovs'ki 10	1
	2202 2302	yellow green				Zhovtosteblyovi	2
		light green	NL and FR : light g	reen = yellow greer	1?	BundyGem, FibreGem, Yuso 31	3
		medium green				Hlera, Tiborszállási	4
		dark green				Kompolti, Zolotonos'ki 11	5
		violet	Purple instead of vic	olet ?		Fibranova	6
20.	MS	Main stem: length of internodes					
QN	(b)(c)	short				Finola, Fasamo	3
	2202 2302	medium				Ruby, Sinelnikivs'ki 3	5
		long				Dniprovs'ki 11	7
21.	MS/ VG	Main stem: thickness					
QN	(b)(c)	thin				Carmen	1
	2202 2302	medium				Dniprovs'ki 11	2
		thick				Carmagnola, Deni	3

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
22.	VG	Main stem: number of grooves					
(+)		of grooves					
QN	(b)(c)	few					1
	2202 2302	medium				Fedora 17	2
		many				Yuso 31	3
23.	VG	Main stem: pith in	FR and DE : to be dele	eted			
(+)		cross-section					
QN	(b)	absent or very narrow					1
	2204 2306	narrow				Carmen, Bundygem, Fibregem,	2
		medium					3
		broad				Deni	4
24.	MG	Seed: weight					
QN	2205 2307	very low				Deni	1
		low				Fasamo	2
		medium				Yuso 31, Kompolti	3
		high				Hlukhivs'ki 10	4
		very high					5
25.	VG	Seed: color of testa					
PQ	2205 2307	light grey				Hlukhivs'ki 10	1
		grey				Fibriko TC, Hlukhivs'ki 58, Lipko	2
		grey brown					3
		yellowish brown					4
		brown				Carmen, Secuieni 1	5
		purplish					6

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		English	français	deutsch	español	Example Varieties/ Exemples/ Beispielssorten/ Variedades ejemplo	Note/ Nota
26.	VG	Seed: reticulation					
(+)							
QN	2205 2307	absent or very weak				FibreGem, Hlukhivs'ki 10	1
		medium				Calavos, Kompolti, Odnodomni 9CHS	2
		strong				BundyGem, Hlukhivs'ki 58	3
27.	VG	Seed: shape in lateral view					
PQ	2205 2307	narrow elliptic	FR and UA: 3 express CZ: rounded, elliptic, o	ions only: rounded, elor ovate. NL will provide l	ngated, oblong; better explanation.	Carmen	1
		ovate					2
		broad ovate				BundyGem, FibreGem, Ruby, Tegege	3
		semi broad elliptic				Calavos	4
		semi oblate					5
28.	MG	Stem: bast fiber	See remark at 15.				
(+)		content					
QN	(b)	very low					1
	3003	low				Ermakivs'ki, Fedora 19	3
		medium				BundyGem, FibreGem, Ruby	5
		high				Carmen, Kompolti	7
_		very high				Beniko, Yuso 31	9
Add.		Main stem: technical length	Proposed by BG. Not point of stem and inflo	recommended by NL, as prescence) and influence	s characteristic is hard to ed by the environment.	define (separation	
Add.		Inflorescence: length	See previous remark				
Add.		Inflorescence: density	See previous remark				

8. <u>Explanations on the Table of Characteristics</u>

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) Observations should be done on the last opposite, fully expanded leaves.
- (b) Male plants should be excluded from the observation.
- (c) Observations should be done on the middle third part of the plant.

8.2 Explanations for individual characteristics

Ad. 4: Plant: coloration of the crown



plants above: red purple crown plants below: light green crown

Ad. 6: Leaf: intensity of green color



from left to right: medium, dark and light green

Ad. 8: Leaf: number of leaflets

Medium number of leaflets is 7 (average number of leaflets). Few is less than 7 leaflets. Many is more than 7 leaflets.

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Ad.13: Time of beginning of female flowering

50% of plants with styles on first female flower visible

Ad. 15: Inflorescence: THC content

The method to determine the THC content is based on a quantitative determination of Δ^9 -tetrahydrocannabinol by gas chromatography after extraction with a suitable solvent.

Sampling

The sample should be taken from the upper 30 cm of the main stem, containing the female inflorescence. Sampling should be carried out in the period from 20 days after the beginning of female flowering up to the end of flowering. The sample should be dried as soon as possible (within 48 hours) at a temperature below 60° C. Samples should be dried to a constant weight and to a moisture content of 8 - 13 %. After drying samples can be stored (without crushing) at 25° C in a dark place.

Determination of THC content (see also Cole, 2003).

1. Preparation of the test sample

Remove stems and seeds over 2 mm in size from the dried samples.

Grind the dried samples to obtain a semi-fine powder (passing through a 1 mm mesh sieve). The powder may be stored for 10 weeks at below 25° C in a dark dry place.

2. Reagents and extraction solution

Reagents

- Δ^9 -tetrahydrocannabinol, pure for chromatographic purposes.

- squalane, pure for chromatographic purposes, as an internal standard.

Extraction solution

- 35 mg of squalane per 100 ml hexane.

3. Extraction of Δ^9 *-tetrahydrocannabinol*

Weigh 100 mg of the powdered test sample, place in a centrifuge tube and add 5 ml of extraction solution containing the internal standard.

Place in an ultrasound bath and leave for 20 minutes. Centrifuge for 5 minutes at 3000 r.p.m. and then remove the supernatant THC solution. Inject the solution into the chromatograph and carry out a quantitative analysis.

4. Gas chromatography

a). Apparatus

- gas chromatograph with a flame ionization detector and a split/splitless injector

- column allowing good separation of cannabinoids, for example a glass capillary column 25 m long and 0,22 mm in diameter impregnated with a 5% non-polar phenyl-methyl-siloxane phase.

b). Calibration ranges

At least three points including points 0,04 and 0,50 mg/ml Δ^9 -THC in extraction solution.

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c). Experimental conditions The following conditions are given as an example for the column referred to in a). oven temperature 260° C injector temperature 300° C detector temperature 300° C d). Injection volume: 1 µl

Results

THC should be determined to two decimals in grams of Δ^9 -THC per 100 grams of analytical sample dried to constant weight. A tolerance of 0,03 g per 100 grams applies. The results are expressed in % dry weight.

State of expression	Range of THC (% dry weight)	
1	< 0,001	absent to very low
2	0,001 - 0,049	very low to low
3	0,050 - 0,099	low
4	0,100 - 0,199	low to medium
5	0,200 - 0,299	medium
6	0,300 - 0,499	medium to high
7	0,500 - 0,999	high
8	1,000 - 2,000	high to very high
9	>2,000	very high

States of expression for range of THC content:

Ad. 16: Plant: sex expression

Cannabis sativa L. is normally dioecious. Monoecious plants occasionally occur naturally but are specially created by breeding activity (Bócsa, 1998). Genetics of sex expression is complex and sex expression can be modified by environmental factors. Varieties are rarely 100% monoecious. Male plants may occur for several generations segregating from breeders' seed. Monoecious varieties are varieties which do not contain more than 5% male plants. Dioecious varieties consisting of hermaphrodite and male plants should have a stable proportion of more than 5% male plants. Gynoecious varieties are 100 % female. They are usually vegetatively propagated.

Ad 18: Plant: natural height

Natural height of flowering plant including inflorescence.

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Ad. 22: Main stem: number of grooves



many grooves (observation to be done on the middle third part of the plant).

Ad. 23: Main stem: pith in cross section

Observations to be taken on node below the last opposite leaves



Ad. 26: Seed reticulation

Black mosaic patterns: marbling of testa



Anka: black mosaic pattern absent to very slight





Kepnock: black mosaic pattern medium to strong

FibreGem: black mosacic pattern medium

Ad. 28: Stem: bast fiber content

The extraction of bast fibers from straw by alkaline extraction is an old timeconsuming method (Bredemann, 1922) to determine the gravimetric fiber yield of bast fiber plants. This method has been modified, without changing the principles of the analysis.

Modified Bredemann method.

A single straw sample of 20 plants per replicate (whole stems, representative for thickness, etc) is cut into pieces of 50-100 cm length and bound in bundles of approximately 100 grams. After the determination of the dry mass (drying balance) the bundled samples are boiled for 1 hr. in a solution of 1.5% caustic soda. After the treatment the straw is placed on a fine screen and sprayed with cold water. Thus the bastfibers are easily removed from the core. The fibers are separated (stripped off completely) and collected. If necessary a second scouring process can be performed e.g. in a laboratory dying machine (Ahuba Turbocolor), with 1% caustic soda + 0,1% detergent, which will remove remaining adherent bast parts and by-products. To avoid losses the fiber sample is wrapped into a fine meshed nylon stocking. After neutral rinsing the fibers are dried and the dry mass is analyzed by a moisture analyzer. This way fiber losses (shortest fragments and trash) can be reduced to 1%.

Fiber content can also be determined by mechanical decortication.

8.3 Growth stages for Hemp

All characteristics should be recorded at the appropriate time for the plant concerned. Growth stages of hemp are recorded by a four-digit code describing the principal growth stages, depending on the sex of the plant followed by detailed developmental stages (Mediavilla, Vito *et al.*, 1998):

Principal growth stages

Four principal stages describe the life cycle of a plant and are coded by their first digit of the four-digit code.

First-digit of code	Definition
0	Germination and emergence
1	Vegetative stage
2	Flowering and seed formation
3	Senescence

Secondary growth stages

The secondary growth stages are described by the second digit, which indicates the sex of the plant, the third and fourth digits indicating the developmental stage of the plant.

Code	Definition	Remarks			
Germinatio	on and emergence				
0000	Dry seed				
0003	Cotylodons unfolded				
Vegetative	stage refers to main stem. Leaves are co	onsidered unfolded when leaflets are at least one cm long			
1002	1 st leaf pair	1 leaflet			
1004	2 nd leaf pair	3 leaflets			
1006	3 rd leaf pair	5 leaflets			
10xx	Last opposite leaf pair	$xx = 2(n^{th} leaf pair)$			
Flowering	and seed formation refers to the ma	in stem including branches			
2000	GV point (i.e. induction of	Change of phyllotaxis on the main stem from			
	flowering)	opposite to alternate. Distance between			
		petioles of alternate leaves at least 0.5 cm			
2001	Flower primordia	Sex nearly indistinguishable			
	Dioecious plant				
Male					
2100	Flower formation	First closed staminate flowers			
2101	Beginning of flowering	First opened staminate flowers			
2102	Flowering	50% opened staminate flowers			
2103	End of flowering	95% of staminate flowers opened or withered			
	Female				
2200	Flower formation	First pistillate flowers			
		Bract with no styles			
2201	Beginning of flowering	Styles on first female flowers			
2202	Flowering	50% of bracts formed			
2203	Beginning of seed maturity	First seeds hard			
2204	Seed maturity	50% of seeds hard			
2205	End of seed maturity	95% of seeds hard or shattered			
	Monoec	ious plant			
2300	Female flower formation	First pistillate flowers			

		Perigonal bracts with no styles
2301	Beginning of female flowering	First styles visible
2302	Female flowering	50% of bracts formed
2303	Male flower formation	First closed staminate flowers
2304	Male flowering	50% opened staminate flowers
2305	Beginning of seed maturity	First seeds hard
2306	Seed maturity	50% of seeds hard
2307	End of seed maturity	95% of seeds hard or shattered
Senescence		
3001	Leaf dessication	Leaves dry
3002	Stem dessication	Leaves dropped
3003	Stem decomposition	Bast fibers free

9. <u>Literature</u>

Bócsa, I., 1998: Genetic Improvement : Conventional Approaches. In: Advances in Hemp Research. Paolo Ranalli (Ed.). Haworth Food Products Press, New York. 272 pp.

Bredemann, G., 1922: Die Bestimmung des Fasergehaltes in Bastfaserpflanzen bei züchterischen Untersuchungen. Faserforschung 2. Leipzig : Hirzel Verlag. S. 239-258.

Clarke, R. C., 1998: Botany of the Genus *Cannabis*. In: Advances in Hemp Research. Paolo Ranalli (Ed.). Haworth Food Products Press, New York. 272 pp.

Cole, M.D., 2003: The analysis of controlled substances – a systematic approach. John Wiley and Sons Ltd., Chichester, UK. ISBN 0-471-49252-3.

Mediavilla, Vito, Manuel Jonquera.\, Ingrid Schmid-Slembrouck and Alberto Soldati, 1998: cimal code for growth stages of hemp (*Cannabis sativa* L.). Journal of the International Hemp Association 5(2): 67-72

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10. <u>Technical Questionnaire</u>

TEC	CHNICAL QUESTIONNAIR	E	Page {x} of {y}	Reference Number:
				Application date: (not to be filled in by the applicant)
	TE to be completed in com	CH nect	NICAL QUESTIONN tion with an applicatio	VAIRE n for plant breeders' rights
1.	Subject of the Technical Qu	esti	onnaire	
	1.1 Botanical name	Car	nnabis sativa L.	
	1.2 Common name	Hei	mp	
2.	Applicant			
	Name			
	Address			
	Telephone No.			
	Fax No.			
	E-mail address			
	Breeder (if different from a	opli	cant)	
	L			
3.	Proposed denomination and	bre	eeder's reference	
	Proposed denomination (if available)			
	Breeder's reference			

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TE	CHNI	CAL QI	UEST	IONNAIRE	Page $\{x\}$ of $\{y\}$	Ret	ference Number:
[#] 4.	⁴ 4. Information on the breeding scheme and propagation of the						e variety
	4.1 Breeding scheme						
		Variet	ty resu	ılting from:			
		4.1.1	Cros	ssing			
			(a)	controlled cr (please state	ross parent varieties)		[]
			(b)	partially kno (please state	own cross known parent variet	y(ies))	[]
			(c)	unknown cro	DSS		[]
		4.1.2	Muta (plea	ation ase state paren	t variety)		[]
		4.1.3	Disc (plea and	overy and dev ase state where how develope	velopment e and when discovere d)	d	[]
		4.1.4	Othe (plea	er ase provide de	tails)		[]

 $^{^{\#}}$ Authorities may allow certain of this information to be provided in a confidential section of the Technical Questionnaire.

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TECHNICAL Q	UESTIONNAIRE	Page $\{x\}$ of $\{y\}$	Reference Number:
4.2 Method of			
4.2.1	Seed-propagated var	rieties	
	(a) Self-pollinatio	on	[]
	(b) Cross-pollinat (i) population (ii) synthetic	ion 1 variety	[]
	(c) Hybrid		[]
	(d) Other (please provid	e details)	[]
4.2.2	Vegetatively propag	ated varieties	
	(a) cuttings		[]
	(b) <i>in vitro</i> propag	gation	[]
	(c) other [state me	ethod]	
4.2.3	Other (please provide detai	ils)	[]

In the case of hybrid varieties the production scheme for the hybrid should be provided on a separate sheet. This should provide details of all the parent lines required for propagating the hybrid e.g.

Single Hybrid

(... female parent ...) x (... male parent ...)

Three-Way Hybrid

(... female line ...) x (... male line ...)

=> single hybrid used as female parent x (... male parent ...)

and should identify in particular:

(a) any male sterile lines

(b) maintenance system of male sterile lines.

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TEC	HNICAL QUESTIONNAIRE Page {x} of {y} Reference	e Number:	
5. correc	Characteristics of the variety to be indicated (the number esponding characteristic in Test Guidelines; please mark esponds).	in brackets refers t the note which	to the best
	Characteristics	Example Varieties	Note
5.1 (13)	Time of beginning of female flowering		
	very early		
	early	Finola	3[]
	medium	Tiborszállási	5[]
	late	Kompolti	7[]
	very late		
5.2 (16)	Plant: sex expression		
	monoecious	Anka, Ermakivs'ki, Fibrimon 21	1[]
	dioecious	Kompolti, Yuso 31	2[]
	gynoecious	Uniko B	3[]
5.3 (18)	Plant: natural height (flowering plant including inflorescence)		
	short	Carmen, Finola, Yuso 31,	3[]
	medium	Hlukhivs'ki 33	5[]
	long	Dniprovs'ki 11	7[]

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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	Characteristic(s) in	Describe the expression	Describe the
variety(ies) similar to	which your candidate	of the characteristic(s)	expression of the
your candidate variety	variety differs from the	for the similar	characteristic(s) for
	similar variety(ies)	variety(ies)	your candidate variety
Example	Plant: height	short	tall

Comments:

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TEC	HNICAL Q	UESTIONNAIRE	Page {x}	of {y}	Reference Number:
[#] 7.	Additional	information which	may help in	the exami	nation of the variety
7.1	In addition characteris	n to the information tics which may help	n provided to distingu	in section ish the var	is 5 and 6, are there any additional iety?
	Yes []	No []	
	(If yes, plea	ase provide details)			
7.2	Are there a	my special condition	ns for growi	ing the vari	ety or conducting the examination?
	Yes []	No []	
	(If yes, plea	ase provide details)			
7.3	Other infor	rmation			
	Main use				
	(a) (b) (c) (d)	bast fibre and wo oil seed pharmaceuticals other (please provide de	ody core etails)		[] [] []
8.	Authorizat	ion for release			
	(a) Does the protecti	the variety require on of the environme	prior autho ent, human	rization for and animal	r release under legislation concerning health?
	Yes	[]	No	[]	
	(b) Has s	such authorization b	een obtaine	d?	
	Yes	[]	No	[]	
	If the answ	ver to (b) is yes, plea	se attach a	copy of the	e authorization.

 $^{^{\#}}$ Authorities may allow certain of this information to be provided in a confidential section of the Technical Questionnaire.

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

.....

9.3 Has the plant material to be examined been tested for the presence of virus or other pathogens?

Yes []

(please provide details as specified by the Authority)

No

[]

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]