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LETTUCE

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Lactuca sativa L.

GUIDELINES

FOR THE CONDUCT OF TESTS

FOR DISTINCTNESS, UNIFORMITY AND STABILITY

*prepared by an expert from France**to be considered by the**Technical Committee at its sixty-first session,
to be held Geneva from 2025-10-20 to 2025-10-21**Disclaimer: this document does not represent UPOV policies or guidance*

This document contains the following changes proposed by the Technical Working Party for Vegetables (TWV), at its fifty-ninth session¹, presented in **grey highlight**:

- (a) Addition of new characteristics 44 to 46 “Resistance to *Bremia lactucae* (BI) - Isolates BI: 38EU, BI: 39EU, BI: 40EU” after “Resistance to *Bremia lactucae* (BI) - Isolate BI: 36EU” to the Table of Characteristics;
- (b) Revision of explanation Ad. 38 to 43 “Resistance to *Bremia lactucae* (BI), several isolates”,
- (c) Addition of the new characteristics 44 to 46 “Resistance to *Bremia lactucae* (BI) - Isolates BI: 38EU, BI: 39EU, BI: 40EU” to the Technical Questionnaire, section 5.

Alternative names:*

Botanical name	English	French	German	Spanish
<i>Lactuca sativa</i> L.	Lettuce	Laitue	Salat	Lechuga

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.]

¹ held by electronic means, from May 5 to 8, 2025.

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1. Subject of these Test Guidelines

These Test Guidelines apply to all varieties of *Lactuca sativa* L.

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 seed.

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

15,000 seeds

The seed should meet the minimum requirements for germination, species and analytical purity, health and moisture content, specified by the competent authority. In cases where the seed is to be stored, the germination capacity should be as high as possible and should, be stated by the applicant.

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

2.5 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. Method of Examination

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*

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 Each test should be designed to result in a total of at least 60 plants, which should be divided between at least 2 replicates.

3.4.2 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.

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 plants or parts of plants to be Examined

Unless otherwise indicated, for the purposes of distinctness, all observations on single plants should be made on 20 plants or parts of plants taken from each of 20 plants and any other observations made on all plants in the test, disregarding any off-type plants.

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 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

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 plants (G) or for single, individual plants (S)

For the purposes of distinctness, observations may be recorded as a single record for a group of plants or parts of plants (G), or may be recorded as records for a number of single, individual plants or parts of plants (S). In most cases, "G" provides a single record per variety and it is not possible or necessary to apply statistical methods in a plant-by-plant 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 These Test Guidelines have been developed for the examination of seed-propagated varieties. For varieties with other types of propagation the recommendations in the General Introduction and document TGP/13 “Guidance for new types and species”, Section 4.5 “Testing Uniformity” should be followed.
- 4.2.3 For the assessment of uniformity of seed-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 60 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 further examined by testing a new seed 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) Seed: color (characteristic 1)
 - (b) Leaf: anthocyanin coloration (characteristic 11)
 - (c) Time of beginning of bolting (characteristic 35)
 - (d) Resistance to *Bremia lactucae* (BI) Isolate BI: 29EU (characteristic 38)

In a first step, the collection should be divided according to types as described in the Table 1. In cases of doubt to which type a variety belongs to, it should be tested under consideration of all relevant types. The different types of Lettuce are explained in Chapter 8.3

- 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”.

Table 1

Type	Example varieties	Plant: degree of overlapping of upper part of leaves (Char. 3)	Leaf: number of divisions (Char. 6)	Leaf: thickness (Char. 17)	Leaf: undulation of margin (Char. 20)	Leaf: venation (Char. 25)	Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Head: shape in longitudinal section (Char. 27)
Butterhead type	Clarion, Maikönig, Sartre	medium to strong	absent or very few	thin to thick	absent to weak	not flabellate	circular or narrow oblate
Novita type	Norvick	absent or weak	absent or very few	thin to medium	very weak to medium	flabellate	-
Iceberg type	Great Lakes 659, Roxette, Saladin, Vanguard 75	strong	absent or very few	thick	absent to medium	flabellate	circular or narrow oblate
Batavia type	Aquarel, Curtis, Funnice, Felucca, Grand Rapids, Masaida, Visyon	absent or weak to strong	absent or very few	medium to thick	weak to very strong	flabellate	broad elliptic, circular or narrow oblate
Frisée d'Amérique type	Bijou, Blonde à couper améliorée	absent or weak	absent or very few	thin	absent to strong	flabellate or not flabellate or semi	-
Lollo type	Lollo rossa, Revolution	absent or weak	absent or very few	thin	strong to very strong	flabellate	-
Oakleaf type	Catalogna, Kipling, Muraï, Salad Bowl	absent or weak	few to many	thin	absent to weak	flabellate or not flabellate or semi	-
Multi-divided type	Curletta, Duplex, Jadigon, Rodagio	absent or weak	medium to very many	thin	weak to very strong	flabellate	-
Frillice type	Frilett	absent or weak	absent or very few	thick	weak to strong	flabellate	-
Cos type	Actarus, Blonde maraîchère, Pinokkio	absent or weak to medium	absent or very few	medium to thick	absent to weak	not flabellate	narrow elliptic
Gem type	Craquerelle du Midi, Sucrine, Xanadu	absent or weak to medium	absent or very few	medium to thick	absent to weak	not flabellate	broad elliptic, circular or narrow oblate
Stem type	Celtuce, Guasihong	absent or weak	absent or very few	thin to medium	absent to weak	not flabellate	-

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:

<i>State</i>	<i>Note</i>
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:

<i>State</i>	<i>Note</i>
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)-(b) See Explanations on the Table of Characteristics in Chapter 8.1
- 7 Not applicable

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

	English		français		deutsch		español		Example Varieties Exemples Beispielssorten Variedades ejemplo		Note/ Nota
1.	(*)	PQ	VG								
		Seed: color		Semence : couleur		Samen: Farbe		Semilla: color			
		white		blanche		weiß		blanco		Verpia	1
		yellow		jaune		gelb		amarillo		Durango	2
		brown		marron		braun		marrón		Oaklin	3
		black		noire		schwarz		negro		Kagraner Sommer 2	4
2.	(*)	QN	MS/VG		(a)						
		Plant: diameter		Plante : diamètre		Pflanze: Durchmesser		Planta: diámetro			
		very small		très petit		sehr klein		muy pequeña		Tom Thumb	1
		small		petit		klein		pequeña		Gotte à graine blanche	3
		medium		moyen		mittel		media		Clarion, Verpia	5
		large		grand		groß		grande		Great Lakes 659	7
		very large		très grand		sehr groß		muy grande		El Toro	9
3.	(*)	QN	VG	(+)	(a)						
		Plant: degree of overlapping of upper part of leaves		Plante : degré du chevauchement de la partie supérieure des feuilles		Pflanze: Stärke des Überlappens des oberen Teils der Blätter		Planta: grado de solapamiento de la parte superior de las hojas			
		absent or weak		nul ou faible		fehlend oder gering		ausente o débil		Actarus, Aquarel, Blonde à couper améliorée, Curtis, Lollo rossa	1
		medium		moyen		mittel		medio		Augusta, Clarion, Fiorella	2
		strong		fort		stark		fuerte		Roxette, Vanguard 75	3
4.		QN	MS/VG	(+)	(a)						
		<u>Only varieties with Plant: degree of overlapping of upper part of leaves: absent or weak: Plant: number of leaves</u>		<u>Seulement les variétés avec Plante : degré de chevauchement de la partie supérieure des feuilles : nul ou faible : Plante : nombre de feuilles</u>		<u>Nur Sorten mit Pflanze: Stärke des Überlappens des oberen Teils der Blätter: fehlend oder gering: Pflanze: Anzahl Blätter</u>		<u>Solo variedades con Planta: grado de solapamiento de la parte superior de las hojas: ausente o débil: Planta: número de hojas</u>			
		few		petit		wenige		bajo		Lollo rossa	3
		medium		moyen		mittel		medio		Muraï	5
		many		grand		viele		alto		Felucca, Sartre, Xandra	7
5.		QN	VG	(+)	(b)						
		Leaf: attitude		Feuille : port		Blatt: Stellung		Hoja: porte			
		erect		dressé		aufrecht		erecto		Feria, Pinokkio	1
		semi-erect		demi-dressé		halbaufrecht		semierecto		Expedition, Sartre	3
		horizontal		horizontal		horizontal		horizontal		Divina	5

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
6.	(*)	QN	VG	(+)	(b)			
		Leaf: number of divisions	Feuille : nombre de divisions		Blatt: Anzahl Teilungen	Hoja: número de divisiones		
		absent or very few	nul ou très petit		fehlend oder sehr wenige	ausentes o muy bajo	Fiorella, Lollo rossa	1
		few	petit		wenige	bajo	Curletta, Rodagio	3
		medium	moyen		mittel	medio	Ezabel, Jadigon	5
		many	grand		viele	alto	Expedition, Multired 54	7
		very many	très grand		sehr viele	muy alto	Excite, Ezfrill, Telex	9
7.		PQ	VG	(+)	(b)			
		<u>Only varieties with Leaf: number of divisions: absent or very few: Leaf: shape</u>	<u>Seulement les variétés avec Feuille : nombre de divisions : nul ou très petit : Feuille : forme</u>		<u>Nur Sorten mit Blatt: Anzahl Teilungen: fehlend oder sehr wenige: Blatt: Form</u>	<u>Solo variedades con Hoja: número de divisiones: ausentes o muy bajo: Hoja: forma</u>		
		triangular	triangulaire		dreieckig	triangular		1
		lanceolate	lancéolée		lanzettlich	lanceolada	Qingyuanewoju	2
		medium oblate	arrondie aplatie moyenne		mittel breitrund	achatada media	Stylist	3
		narrow oblate	arrondie aplatie étroite		schmal breitrund	achatada estrecha	Commodore, Fiorella	4
		circular	circulaire		kreisförmig	circular	Verpia	5
		broad elliptic	elliptique large		breit elliptisch	elíptica ancha	Amadeus	6
		medium elliptic	elliptique moyenne		mittel elliptisch	elíptica media	Xanadu	7
		narrow elliptic	elliptique étroite		schmal elliptisch	elíptica estrecha	Verte maraîchère	8
		linear	linéaire		linear	lineal	Hongwoju	9
		broad obtrullate	losangique transverse large		breit verkehrt rautenförmig	rómbica ancha		10
		obovate	obovale		verkehrt eiförmig	oboval	Raisa	11
		oblanceolate	oblancéolée		verkehrt lanzettlich	oblanceolada	Xiangshengcai	12
8.		PQ	VG	(+)	(b)			
		<u>Only varieties with Leaf: number of divisions: absent or very few: Leaf: shape of apex</u>	<u>Seulement les variétés avec Feuille : nombre de divisions : nul ou très petit : Feuille : forme de l'extrémité</u>		<u>Nur Sorten mit Blatt: Anzahl Teilungen: fehlend oder sehr wenige: Blatt: Form der Spitze</u>	<u>Solo variedades con Hoja: número de divisiones: ausentes o muy bajo: Hoja: forma del ápice</u>		
		acute	aiguë		spitz	agudo	Celtuce	1
		obtuse	obtuse		stumpf	obtuso	Actarus	2
		rounded	arrondie		abgerundet	redondeado	Blonde maraîchère, Maserati	3
		obcordate	obcordiforme		verkehrt herzförmig	obcordiforme	PS 6545691	4

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
9.	QN	VG	(+)	(b)				
	<u>Only varieties with Leaf: number of divisions: absent or very few: Leaf: longitudinal section</u>		<u>Seulement les variétés avec Feuille : nombre de divisions : nul ou très petit : Feuille : section longitudinale</u>		<u>Nur Sorten mit Blatt: Anzahl Teilungen: fehlend oder sehr wenige: Blatt: Längsschnitt</u>	<u>Solo variedades con Hoja: número de divisiones: ausentes o muy bajo: Hoja: sección longitudinal</u>		
	concave		concave		konkav	cóncava	Sunstar	1
	flat		plate		flach	plana	Clarion, Lollo rossa	3
	convex		convexe		konvex	convexa	Tiago	5
10.	QN	VG	(+)	(b)				
	<u>Only Oakleaf type varieties: Leaf: width of lobes</u>		<u>Seulement les variétés de type Feuille de chêne : Feuille : largeur des lobes</u>		<u>Nur Sorten des Typs Eichblatt: Blatt: Breite der Lappen</u>	<u>Solo variedades de tipo Oakleaf: Hoja: anchura de los lóbulos</u>		
	narrow		étroits		schmal	estrecha	Kibrille, Rougini	3
	medium		moyens		mittel	media	Bandolin, Ribaï	5
	broad		larges		breit	ancha	Horix, Starix, Vizir	7
11. (*)	QN	VG	(+)	(b)				
	Leaf: anthocyanin coloration		Feuille : pigmentation anthocyanique		Blatt: Anthocyanfärbung	Hoja: pigmentación antociánica		
	absent or very weak		nulle ou très faible		fehlend oder sehr gering	ausente o muy débil	Clarion	1
	weak		faible		gering	débil	Du bon jardinier	3
	medium		moyenne		mittel	media	Lollo rossa, Luana	5
	strong		forte		stark	fuerte	Merveille des quatre saisons	7
	very strong		très forte		sehr stark	muy fuerte	Iride, Revolution	9
12. (*)	PQ	VG		(b)				
	Leaf: hue of anthocyanin coloration		Feuille : teinte de la pigmentation anthocyanique		Blatt: Ton der Anthocyanfärbung	Hoja: tonalidad de la pigmentación antociánica		
	reddish		rougeâtre		rötlich	rojiza	Lollo rossa	1
	purplish		pourpre		purpurn	purpúrea	Iride	2
	brownish		brunâtre		bräunlich	amarronada	Luana, Maravilla de Verano	3
13.	QN	VG	(+)	(b)				
	Leaf: area covered by anthocyanin coloration		Feuille : surface couverte par la pigmentation anthocyanique		Blatt: Fläche der Anthocyanfärbung	Hoja: superficie cubierta por la pigmentación antociánica		
	very small		très petite		sehr klein	muy pequeña	Steirer Krauthauptel	1
	small		petite		klein	pequeña	Diablo	3
	medium		moyenne		mittel	media	Luana	5
	large		grande		groß	grande	Merveille des quatre saisons	7
	very large		très grande		sehr groß	muy grande	Bijou, Revolution	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
14. (*)	PQ	VG	(+)	(b)				
	Leaf: color		Feuille : couleur		Blatt: Farbe	Hoja: color		
	green		vert		grün	verde	Verpia	1
	yellowish green		vert jaunâtre		gelblichgrün	verde amarillento	Dorée de printemps	2
	greyish green		vert grisâtre		gräulichgrün	verde grisáceo	Celtuce, Du bon jardinier	3
15. (*)	QN	VG		(b)				
	Leaf: intensity of green color		Feuille : intensité de la couleur verte		Blatt: Intensität der Grünfärbung	Hoja: intensidad del color verde		
	very light		très claire		sehr hell	muy claro		1
	light		claire		hell	claro	Blonde maraîchère, Lollo Bionda	3
	medium		moyenne		mittel	medio	Aquarel, Clarion	5
	dark		foncée		dunkel	oscuro	Expedition, Verpia	7
	very dark		très foncée		sehr dunkel	muy oscuro	Pascal, Verdatrix	9
16.	QN	VG		(b)				
	Leaf: glossiness of upper side		Feuille : brillance de la face supérieure		Blatt: Glanz der Oberseite	Hoja: brillo del haz		
	absent or very weak		nulle ou très faible		fehlend oder sehr gering	ausente o muy débil	Divina, Du bon jardinier	1
	weak		faible		gering	débil	Duplex, Fiorella, Sartre	3
	medium		moyenne		mittel	medio	Funnice	5
	strong		forte		stark	fuerte	Noisette, Redair	7
	very strong		très forte		sehr stark	muy fuerte	Bijou	9
17. (*)	QN	VG		(b)				
	Leaf: thickness		Feuille : épaisseur		Blatt: Dicke	Hoja: grosor		
	very thin		très mince		sehr dünn	muy delgada	Stefano	1
	thin		mince		dünn	delgada	Bijou, Lollo rossa, Raisa	2
	medium		moyenne		mittel	media	Curtis, Expedition	3
	thick		épaisse		dick	gruesa	Frilett, Roxette	4
	very thick		très épaisse		sehr dick	muy gruesa		5
18. (*)	QN	VG		(b)				
	Leaf: blistering		Feuille : cloûre		Blatt: Blasigkeit	Hoja: abullonado		
	absent or very weak		nulle ou très faible		fehlend oder sehr gering	ausente o muy débil	Duplex, Sartre	1
	weak		faible		gering	débil	Fiorella	3
	medium		moyenne		mittel	medio	Commodore	5
	strong		forte		stark	fuerte	Blonde de Paris, Xanadu	7
	very strong		très forte		sehr stark	muy fuerte	Blonde de Doulon, Iride, Karioka	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
19.	QN	VG/VS	(+)	(b)				
	Leaf: size of blisters		Feuille : taille des cloques		Blatt: Größe der Blasen	Hoja: tamaño del abullonado		
	small		petites		klein	pequeño	Dorée de printemps, Rodagio	3
	medium		moyennes		mittel	medio	Clarion	5
	large		grandes		groß	grande	Fiorella	7
20. (*)	QN	VG/VS	(+)	(b)				
	Leaf: undulation of margin		Feuille : ondulation du bord		Blatt: Wellung des Randes	Hoja: ondulación del borde		
	absent or very weak		nulle ou très faible		fehlend oder sehr gering	ausente o muy débil	Tiago	1
	weak		faible		gering	débil	Commodore	3
	medium		moyenne		mittel	media	Noisette, Pentared	5
	strong		forte		stark	fuerte	Calmar, Invicta	7
	very strong		très forte		sehr stark	muy fuerte	Lollo rossa	9
21.	PQ	VG	(+)	(b)				
	Leaf: type of incisions of margin		Feuille : type de découpures du bord		Blatt: Typ der Randeinschnitte	Hoja: tipo de incisiones del borde		
	crenate		crénelé		gekerbt	crenada	Gloire du Dauphiné	1
	regularly dentate		régulièrement denté		regelmäßig gezähnt	dentada regularmente	Soliflore	2
	irregularly dentate		irrégulièrement denté		unregelmäßig gezähnt	dentada irregularmente	Rodagio	3
	bidentate		bidenté		doppelt gezähnt	bidentada	Great Lakes 118	4
	tridentate		tridenté		dreifach gezähnt	tridentada	Expedition	5
22.	QN	VG	(+)	(b)				
	Leaf: depth of incisions of margin		Feuille : profondeur des découpures du bord		Blatt: Tiefe der Randeinschnitte	Hoja: profundidad de las incisiones del borde		
	absent or very shallow		absentes ou très peu profondes		fehlend oder sehr flach	ausentes o muy poco profundas	Actarus, Clarion, Tiago	1
	shallow		peu profondes		flach	poco profundas	Pentared, Unicum	3
	medium		moyennes		mittel	medias	Santarinas	5
	deep		profondes		tief	profundas	Expedition	7
	very deep		très profondes		sehr tief	muy profundas		9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
23.	QN	VG	(+)	(b)				
	Only varieties with Leaf: type of incisions of margin: irregularly dentate, bi- or tridentate: Leaf: depth of secondary incisions of margin		Seulement les variétés avec Feuille : type de découpures du bord : irrégulièrement denté, bidenté ou tridenté : Feuille : profondeur des découpures secondaires du bord		Nur Sorten mit Blatt: Typ der Randeinschnitte: unregelmäßig gezähnt, doppelt oder dreifach gezähnt: Blatt: Tiefe der sekundären Randeinschnitte	Solo variedades con Hoja: tipo de incisiones del borde: dentadas irregularmente, bidentadas o tridentadas: Hoja: profundidad de las incisiones secundarias del borde		
	shallow		peu profondes		flach	poco profundas	Great Lakes 659	3
	medium		moyennes		mittel	medias	Expedition	5
	deep		profondes		tief	profundas		7
24.	QN	VG	(+)	(b)				
	Leaf: density of incisions of margin		Feuille : densité des découpures du bord		Blatt: Dichte der Randeinschnitte	Hoja: densidad de las incisiones del borde		
	very sparse		très lâches		sehr locker	muy laxa		1
	sparse		lâches		locker	laxa	Maravilla de Verano	3
	medium		moyennes		mittel	media	Calmar	5
	dense		denses		dicht	densa	Grand Rapids	7
	very dense		très denses		sehr dicht	muy densa	Locarno	9
25. (*)	QN	VG	(+)	(b)				
	Leaf: venation		Feuille : nervation		Blatt: Aderung	Hoja: nervadura		
	not flabellate		non flabelliforme		nicht fächerförmig	no flabeliforme	Verpia, Xanadu	1
	semi-flabellate		semi-flabelliforme		halb fächerförmig	semiflabiliforme	Kibrille, Muraï	2
	flabellate		flabelliforme		fächerförmig	flabeliforme	Locarno, Roxette	3
26.	QN	MS/VG		(a)				
	Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Head: size		Seulement les variétés avec Plante : degré du chevauchement de la partie supérieure des feuilles : moyen ou fort : Pomme : taille		Nur Sorten mit Pflanze: Stärke des Überlappens des oberen Teils der Blätter: mittel oder stark: Kopf: Größe	Solo variedades con Planta: grado de solapamiento de la parte superior de las hojas: medio o fuerte: Cogollo: tamaño		
	very small		très petite		sehr klein	muy pequeño	Tom Thumb	1
	small		petite		klein	pequeño	Xanadu	3
	medium		moyenne		mittel	medio	Fiorella, Soraya	5
	large		grande		groß	grande	Great Lakes 659	7
	very large		très grande		sehr groß	muy grande	Blonde maraîchère, El Toro	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
27. (*)	QN	MS/VG	(+)	(a)				
	Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Head: shape in longitudinal section		Seulement les variétés avec Plante : degré du chevauchement de la partie supérieure des feuilles : moyen ou fort : Pomme : forme en section longitudinale		Nur Sorten mit Pflanze: Stärke des Überlappens des oberen Teils der Blätter: mittel oder stark: Kopf: Form im Längsschnitt	Solo variedades con Planta: grado de solapamiento de la parte superior de las hojas: medio o fuerte: Cogollo: forma en sección longitudinal		
	narrow elliptic		elliptique étroite		schmal elliptisch	elíptica estrecha	Verte maraîchère	1
	broad elliptic		elliptique large		breit elliptisch	elíptica ancha	Amadeus, Sucrine	2
	circular		circulaire		kreisförmig	circular	Verpia	3
	narrow oblate		aplatie arrondie étroite		schmal breitrund	achatada estrecha	Ametist	4
28.	QN	VG		(a)				
	Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Head: density		Seulement les variétés avec Plante : degré du chevauchement de la partie supérieure des feuilles : moyen ou fort : Pomme : densité		Nur Sorten mit Pflanze: Stärke des Überlappens des oberen Teils der Blätter: mittel oder stark: Kopf: Dichte	Solo variedades con Planta: grado de solapamiento de la parte superior de las hojas: medio o fuerte: Cogollo: densidad		
	loose		lâche		locker	laxa	Nanda	3
	medium		moyenne		mittel	media	Delice, Daguan	5
	dense		dense		dicht	densa	Atella, Islandia	7
	very dense		très dense		sehr dicht	muy densa	Rubette	9
29.	QN	MS/VG	(+)	(a)				
	Only Stem type varieties: Stem: length		Seulement les variétés de type Tige : Tige : longueur		Nur Sorten des Typs Stengelsalat: Stengel: Länge	Solo variedades de tipo tallo: Tallo: longitud		
	short		courte		kurz	corto	Wuweijianye	3
	medium		moyenne		mittel	medio	Zipixiang	5
	long		longue		lang	largo	Guasihong	7
30.	QN	MS/VG	(+)	(a)				
	Only Stem type varieties: Stem: width		Seulement les variétés de type Tige : Tige : largeur		Nur Sorten des Typs Stengelsalat: Stengel: Breite	Solo variedades de tipo tallo: Tallo: anchura		
	narrow		étroite		schmal	estrecho	Ailaowoju	1
	medium		moyenne		mittel	medio	Guasihong, Zipixiang	2
	broad		large		mittel	ancho	Guasihong	3
31.	PQ	VG	(+)	(a)				
	Only Stem type varieties: Stem: shape in longitudinal section		Seulement les variétés de type Tige : Tige : forme en section longitudinale		Nur Sorten des Typs Stengelsalat: Stengel: Form im Längsschnitt	Solo variedades de tipo tallo: Tallo: forma en sección longitudinal		
	cylindrical		cylindrique		zylindrisch	cilíndrico	Chiwoju	1
	conical		conique		kegelförmig	cónico	Guasihong	2
	fusiform		fusiforme		spindelförmig	fusiforme	Zipixiang	3

	English		français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
32.	PQ	VG	(a)				
	Only Stem type varieties: Stem: color		Seulement les variétés de type Tige : Tige : couleur	Nur Sorten des Typs Stengelsalat: Stengel: Farbe	Solo variedades de tipo tallo: Tallo: color		
	whitish green		vert blanchâtre	weißlichgrün	verde blanquecino	Wuweijianye	1
	light green		vert clair	hellgrün	verde claro	Chiwoju	2
	medium green		vert moyen	mittelgrün	verde medio	Yangwoju	3
	greenish purple		pourpre verdâtre	grünlichpurpurn	púrpura verdoso	Guasihong	4
	purplish red		rouge pourpre	purpurrot	rojo purpúreo	Hongwosun	5
33.	PQ	VG	(a)				
	Only Stem type varieties: Stem: color of flesh		Seulement les variétés de type Tige : Tige : couleur de la chair	Nur Sorten des Typs Stengelsalat: Stengel: Farbe des Fleisches	Solo variedades de tipo tallo: Tallo: color de la médula		
	yellowish white		blanc jaunâtre	gelblichweiß	blanco amarillento	Wuweijianye	1
	whitish green		vert blanchâtre	weißlichgrün	verde blanquecino	Chiwoju	2
	light green		vert clair	hellgrün	verde claro	Yangwoju	3
	medium green		vert moyen	mittelgrün	verde medio	Guasihong	4
	dark green		vert foncé	dunkelgrün	verde oscuro	Chiwosun	5
34.	QN	MG/VG					
	Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Time of harvest maturity		Seulement les variétés avec Plante : degré du chevauchement de la partie supérieure des feuilles : moyen ou fort : Époque de maturité de récolte	Nur Sorten mit Pflanze: Stärke des Überlappens des oberen Teils der Blätter: mittel oder stark: Zeitpunkt der Erntereife	Solo variedades con Planta: grado de solapamiento de la parte superior de las hojas: medio o fuerte: Época de madurez para cosecha		
	very early		très précoce	sehr früh	muy temprana	Gotte jaune d'or	1
	early		précoce	früh	temprana	Pantlika, Sucrine	3
	medium		moyenne	mittel	media	Clarion	5
	late		tardive	spät	tardía	Blonde maraîchère, Calmar	7
	very late		très tardive	sehr spät	muy tardía	El Toro, Pinokkio	9
35. (*)	QN	MG/VG	(+)				
	Time of beginning of bolting		Époque de début de montaison	Zeitpunkt des Schoßbeginns	Época del comienzo de la subida de la flor		
	very early		très précoce	sehr früh	muy temprana	Blonde à couper améliorée	1
	early		précoce	früh	temprana	Gotte à graine blanche	3
	medium		moyenne	mittel	media	Pantlika	5
	late		tardive	spät	tardía	Hilde II	7
	very late		très tardive	sehr spät	muy tardía	Erika, Roxette	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
36.	QN	VG	(+)					
	Axillary sprouting		Développement des bourgeons axillaires		Seitentriebbildung	Brotación axilar		
	absent or weak		absent ou faible		fehlend oder gering	ausente o débil	Claridia, Shotter, Valmaine, Xanadu	1
	medium		moyen		mittel	media	Actarus	2
	strong		fort		stark	fuerte	Amible, Bassoon	3
37.	QN	VG	(+)					
	Bolting stem: fasciation		Hampe florale : fasciation		Schoßender Stengel: Verbänderung	Tallo floral: fasciación		
	absent or very weak		absente ou très faible		fehlend oder sehr gering	ausente o muy débil	Aquarel, Gotte à graine blanche	1
	weak		faible		gering	débil	Verte maraîchère	3
	medium		moyenne		mittel	media	Amadeus	5
	strong		forte		stark	fuerte	Rugini	7
	very strong		très forte		sehr stark	muy fuerte	Sartre, Verdetrix	9
38.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 29EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 29EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 29EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 29EU		
	absent		absente		fehlend	ausente	Argelès	1
	present		présente		vorhanden	presente	Balesta	9
39.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 30EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 30EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 30EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 30EU		
	absent		absente		fehlend	ausente	Argelès, Colorado	1
	present		présente		vorhanden	presente	Balesta	9
40.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 31EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 31EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 31EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 31EU		
	absent		absente		fehlend	ausente	Colorado, RYZ910457	1
	present		présente		vorhanden	presente	Argelès, Balesta	9
41.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 33EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 33EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 33EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 33EU		
	absent		absente		fehlend	ausente	Kibrille, RYZ2164	1
	present		présente		vorhanden	presente	RYZ910457	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
42.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 35EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 35EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 35EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 35EU		
	absent		absente		fehlend	ausente	Design, Kibrille	1
	present		présente		vorhanden	presente	Bartoli	9
43.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 36EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 36EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 36EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 36EU		
	absent		absente		fehlend	ausente	Bartoli, RYZ2164	1
	present		présente		vorhanden	presente	Design, Kibrille	9
44.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 38EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 38EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 38EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 38EU		
	absent		absente		fehlend	ausente	Design, Kibrille	1
	present		présente		vorhanden	presente	Bartoli	9
45.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 39EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 39EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 39EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 39EU		
	absent		absente		fehlend	ausente	Bartoli, Dandie, RYZ2164	1
	present		présente		vorhanden	presente	Design, Kibrille	9
46.	QL	VG	(+)					
	Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 40EU		Résistance à <i>Bremia lactucae</i> (BI) Isolat BI: 40EU		Resistenz gegen <i>Bremia lactucae</i> (BI) Isolat BI: 40EU	Resistencia a <i>Bremia lactucae</i> (BI) Aislado BI: 40EU		
	absent		absente		fehlend	ausente	Bartoli, RYZ2164	1
	present		présente		vorhanden	presente	Kibrille	9
47.	QL	VG	(+)					
	Resistance to <i>Lettuce mosaic virus</i> (LMV) Pathotype II		Résistance au <i>Lettuce mosaic virus</i> (LMV) Pathotype II		Resistenz gegen <i>Lettuce mosaic virus</i> (LMV) Pathotyp II	Resistencia al <i>Lettuce mosaic virus</i> (LMV), Patotipo II		
	absent		absente		fehlend	ausente	Bijou, Hilde II, Sprinter, Sucrine	1
	present		présente		vorhanden	presente	Capitan, Corsica	9
48.	QL	MS/VG	(+)					
	Resistance to <i>Nasonovia ribisnigri</i> (Nr) Biotype Nr: 0		Résistance à <i>Nasonovia ribisnigri</i> (Nr) Biotype Nr: 0		Resistenz gegen <i>Nasonovia ribisnigri</i> (Nr) Biotyp Nr: 0	Resistencia a <i>Nasonovia ribisnigri</i> (Nr) Biotipo N° 0		
	absent		absente		fehlend	ausente	Abel, Green Towers, Nadine	1
	present		présente		vorhanden	presente	Barcelona, Bedford, Dynamite, Silvinas	9

	English		français		deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
49.	QN	MS/VG	(+)					
	Resistance to <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Race 1		Résistance à <i>Fusarium</i> <i>oxysporum</i> f.sp. <i>lactuca</i> (Fol) Race 1		Resistenz gegen <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Pathotyp 1	Resistencia a <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Raza 1		
	absent or low		absent ou faible		fehlend oder gering	ausente o baja	Cobham Green, Patriot	1
	medium		moyenne		mittel	media	Affic, Fuzila, Natexis	2
	high		élevée		hoch	alta	Costa Rica No. 4, Romasol	3
50.	QN	MS/VG	(+)					
	Resistance to <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Race 4		Résistance à <i>Fusarium</i> <i>oxysporum</i> f.sp. <i>lactuca</i> (Fol) Race 4		Resistenz gegen <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Pathotyp 4	Resistencia a <i>Fusarium oxysporum</i> f.sp. <i>lactuca</i> (Fol) Raza 4		
	absent or low		absent ou faible		fehlend oder gering	ausente o baja	Costa Rica No. 4, Gisela	1
	medium		moyenne		mittel	media	Ballerina, Patriot	2
	high		élevée		hoch	alta	Lomeria, Palmos	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) Plant, head and stem: Observations should be made at harvest maturity. For varieties with degree of overlapping of upper part of leaves absent or weak observations should be made just before deterioration and before bolting.
- (b) Leaf: For varieties with degree of overlapping of upper part of leaves medium or strong observations should be made on the largest outer leaves, at harvest maturity. For varieties with degree of overlapping of upper part of leaves absent or weak observations should be made on the largest leaves, just before deterioration and before bolting. For Stem type varieties observations should be made on leaves at the middle third of the stem, just before deterioration and before bolting.

8.2 *Explanations for individual characteristics*

Ad. 3: Plant: degree of overlapping of upper part of leaves

Observations should be made on leaves at the heart of the plant to form a head.



1
absent or weak



2
medium



3
strong

Ad. 4: Only varieties with Plant: degree of overlapping of upper part of leaves: absent or weak: Plant: number of leaves

In case of doubt, observations can be made by cutting the plant in half.



3
few

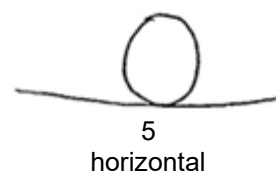
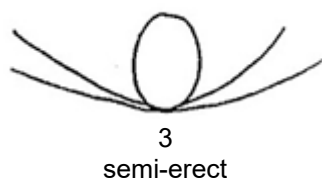
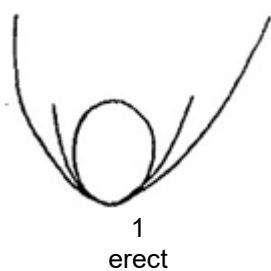


5
medium



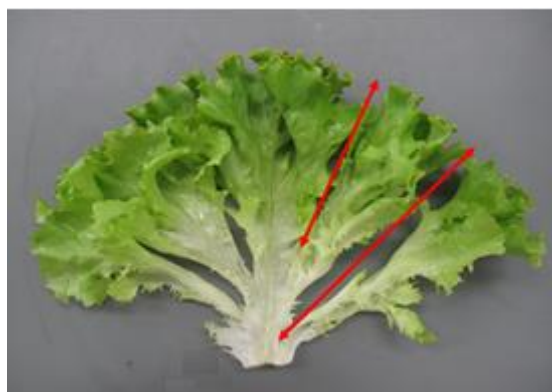
7
many

Ad. 5: Leaf: attitude



Ad. 6: Leaf: number of divisions

Observations should be made only on the incisions more than halfway to the midrib of the whole leaf.



1
absent or very few



3
few



5
medium

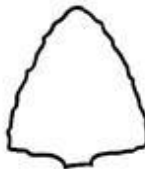
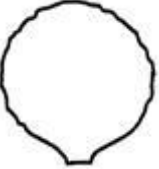
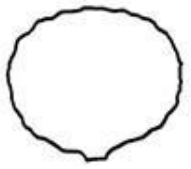
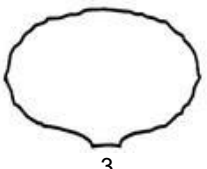


7
many



9
very many

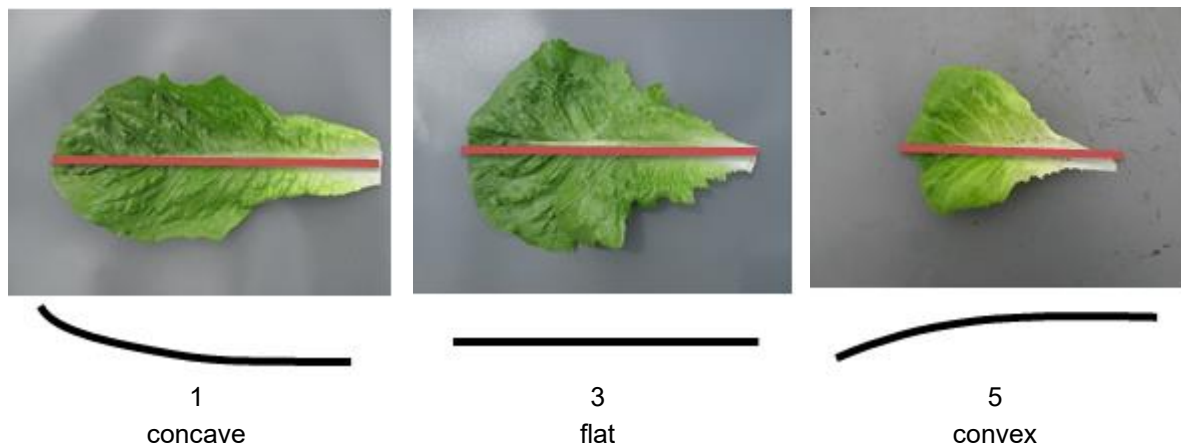
Ad. 7: Only varieties with Leaf: number of divisions: absent or very few: Leaf: shape

width (ratio length/width)	< broadest part >		
	below middle	at middle	above middle
narrow (high)		 9 linear	
	 2 lanceolate	 8 narrow elliptic	 12 oblanceolate
		 7 medium elliptic	
medium (medium)	 1 triangular	 6 broad elliptic	 11 obovate
		 5 circular	
		 4 narrow oblate	 10 broad obtrullate
broad (low)		 3 medium oblate	

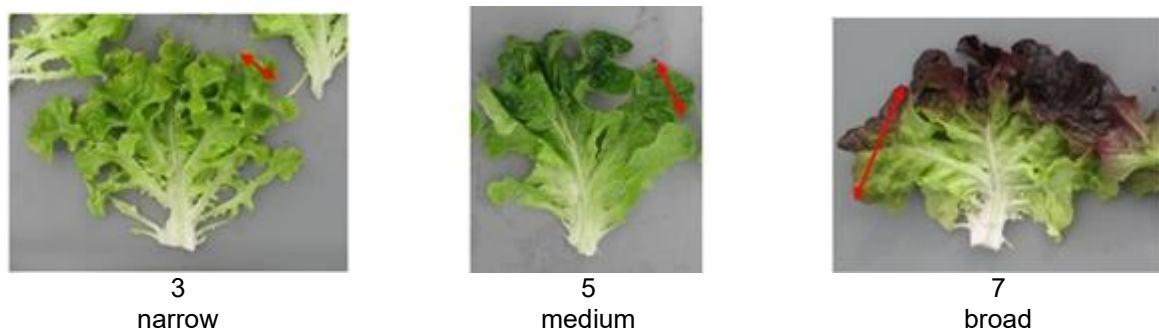
Ad. 8: Only varieties with Leaf: number of divisions: absent or very few: Leaf: shape of apex



Ad. 9: Only varieties with Leaf: number of divisions: absent or very few: Leaf: longitudinal section



Ad. 10: Only Oakleaf type varieties: Leaf: width of lobes



Ad. 11: Leaf: anthocyanin coloration

Ad. 12: Leaf: hue of anthocyanin coloration

Anthocyanin coloration (Char. 11)	Hue of anthocyanin coloration (Char. 12)		
	1 reddish	2 purplish	3 brownish
1 absent or very weak	Clarion		
3 weak	Du bon jardinier, Steirer Krauthauptel		Brauner Trotzkopf, Diablo, Maravilla de Verano
5 medium	Lollo rossa		Frisée d'Amérique, Luana, New Red Fire, Salad bowl rossa
7 strong	Jadigon		Duplex, Merveille des quatre saisons
9 very strong	Revolution	Iride	Multired 54

Ad. 13: Leaf: area covered by anthocyanin coloration

Observations should be made on the total area of diffused and/or localised anthocyanin coloration.



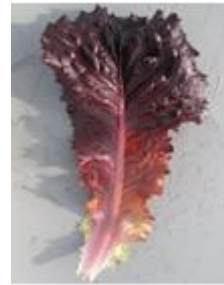
3
small



5
medium



7
large



9
very large

Ad. 14: Leaf: color

Ad. 15: Leaf: intensity of green color

Only to observe for green varieties and for two-colored varieties with 'Leaf: area covered by anthocyanin coloration' less than large (less than note 7 to 9), so the green color of the leaf can be observed without picking a leaf from the plant.

Intensity of green color (Char. 15)	Color (Char. 14)		
	1 green	2 yellowish green	3 greyish green
1 very light			
3 light	Blonde maraîchère, New Red Fire	Lollo Bionda, Steirer Krauthauptel	Celtuce
5 medium	Ballerina	Aquarel, Australische Gele, Dorée de printemps	Clarion, Du bon jardinier, Durango
7 dark	Actarus, Baby Star, Expedition, Verpia		Webbs Wonderful
9 very dark	Pascal, Verdetrix		

Ad. 19: Leaf: size of blisters

Observations should be made on the whole leaf.



3
small



5
medium



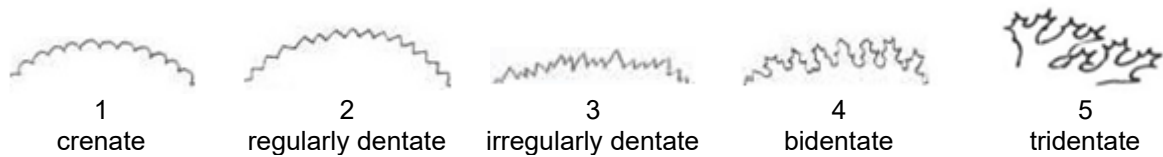
7
large

Ad. 20: Leaf: undulation of margin

Observations should be made on undulation of margin of apical part; also apical part in case of divided leaves.

Ad. 21: Leaf: type of incisions of margin

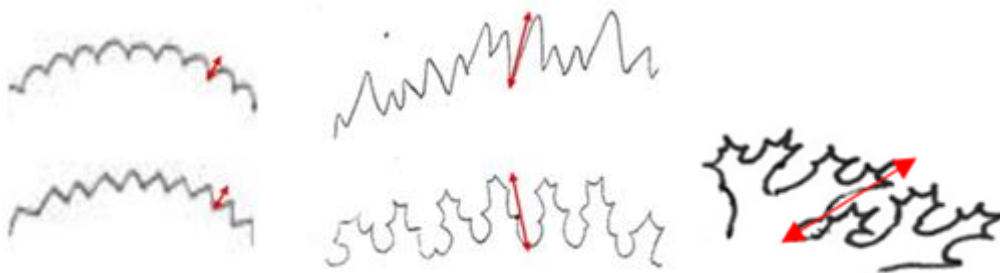
Observations should be made on incisions of the margin at the distal half of the leaf.



Ad. 22: Leaf: depth of incisions of margin

Observations should be made on incisions of the margin at the distal half of the leaf. For varieties with irregularly dentate, bidentate or tridentate incisions describe the deepest incisions and use Char. 23 for the secondary incisions.

The following drawings illustrate how to observe this characteristic for the different types of incisions.



Ad. 23: Only varieties with Leaf: type of incisions of margin: irregularly dentate, bi- or tridentate: Leaf: depth of secondary incisions of margin

Observations should be made on secondary incisions of the margin at the distal half of the leaf. In case of tridentate incisions observations should not be made on tertiary incisions of the margin (the most shallow ones).

Ad. 24: Leaf: density of incisions of margin

Observations should be made on all incisions of the margin at the distal half of the leaf, so in case of irregularly dentate or bidentate both primary and secondary incisions, in case of tridentate also tertiary incisions.

Ad. 25: Leaf: venation



1
not flabellate

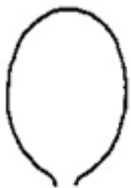


2
semi-flabellate

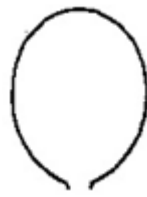


3
flabellate

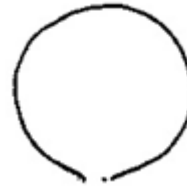
Ad. 27: Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Head: shape in longitudinal section



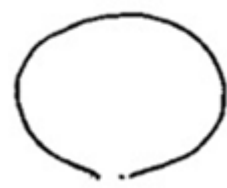
1
narrow elliptic



2
broad elliptic

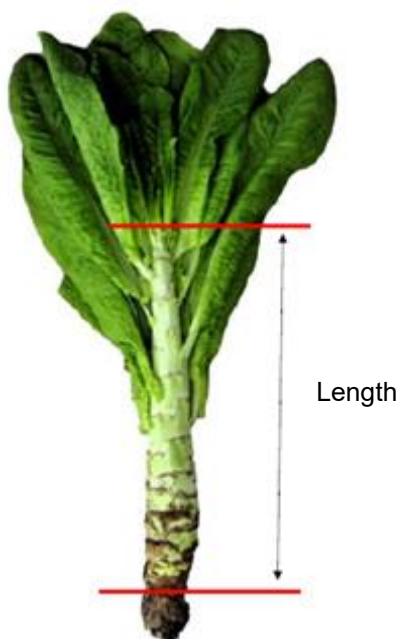


3
circular



4
narrow oblate

Ad. 29: Only Stem type varieties: Stem: length



Ad. 30: Only Stem type varieties: Stem: width

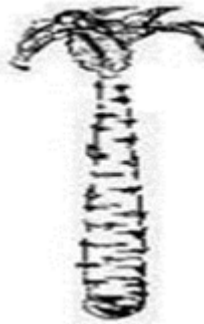
Observations should be made on the broadest part of the stem.



Ad. 31: Only Stem type varieties: Stem: shape in longitudinal section



1
cylindrical



2
conical



3
fusiform

Ad. 35: Time of beginning of bolting

Observations should be made in a trial with more than 12 hours of day light as lettuce varieties need a long photo period to induce bolting.

Observations should be made when 50% of the plants start to bolt. The top of the bolting stem can be seen or felt at the top of the plant.

Ad. 36: Axillary sprouting

Formation of secondary sprouts beside the main head. Arrow points at one of the secondary sprouts. Observations should be made in overripe stage, just before bolting.



Ad. 37: Bolting stem: fasciation

Observations should be made on the stem of bolted plants after the first flowers are open. For varieties with very late time of beginning of bolting and with strong degree of overlapping of leaves, the cover leaves of the head may be incised just before deterioration in order to be able to observe fasciation.



1
absent or
very weak



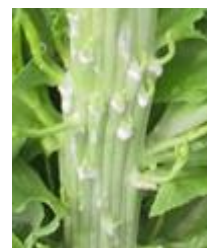
3
weak



5
medium



7
strong



9
very strong

Ad. 38 to 46: Resistance to *Bremia lactucae* (BI), several isolates

1.	Pathogen	<i>Bremia lactucae</i>
2.	Quarantine status	no
3.	Host species	lettuce - <i>Lactuca sativa</i> L.
4.	Source of inoculum	GEVES ² (FR) or Naktuinbouw ³ (NL)
5.	Isolate	BI: 29-31EU, 33EU, 35EU, 36EU, 38-40EU
6.	Establishment isolate identity	test on differentials (see table below)
7.	Establishment pathogenicity	test on susceptible varieties

² matref@geves.fr

³ resistentie@naktuinbouw.nl

8.	Multiplication inoculum	
8.2	Multiplication variety	susceptible variety, for example Green Towers. For isolates with a higher number than BI: 16EU, a variety with defeated resistance may be preferable to keep the isolate fit.
8.8	Shelflife/viability inoculum	2 hours at room temperature; 2 days in refrigerator
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 plants
9.2	Number of replicates	-
9.3	Control varieties	(informative) differentials (see table below)
9.4	Test design	-
9.5	Test facility	climate room
9.6	Temperature	15°C-18°C
9.7	Light	adequate for good plant growth; seedlings should not be etiolated. option: reduced light 24 hours after inoculation
9.8	Season	-
9.9	Special measures	plants may grow on wet blotting paper with or without a nutrient solution, on sand or on potting soil (see point 13). high humidity (>90%) is essential for infection and sporulation.
10.	Inoculation	
10.1	Preparation inoculum	washing off from leaves by vigorous shaking in a closed container
10.2	Quantification inoculum	counting spores; spore density should be 3×10^4 - 1×10^5
10.3	Plant stage at inoculation	cotyledon stage
10.4	Inoculation method	spraying till run-off. option: reduced light 24 hours after inoculation
10.5	First observation	beginning of sporulation on susceptible varieties (around 7 days after inoculation)
10.6	Second observation	3-4 days after first observation (around 10 days after inoculation)
10.7	Final observations	14 days after inoculation two of these three observations may be sufficient, the third notation is optional for observation of evolution of symptoms in case of doubt. the day of maximum sporulation should occur in this period.
11.	Observations	
11.1	Method	visual observation of sporulation and necrotic reaction to infection
11.2	Observation scale	resistant: class 0 no sporulation, no necrosis class 1 no sporulation, necrosis present class 2 weak sporulation (much less than susceptible control) with necrosis class 3 weak sporulation (less than susceptible control and not evolving between second and third observation) with necrosis class 4 very sparse sporulation (not evolving between second and third observation) without necrosis susceptible: class 5 reduced sporulation (compared to susceptible control) without necrosis class 6 normal sporulation without necrosis
11.3	Validation of test	Validation on controls If varieties show the same level of sporulation as the susceptible control but with necrosis, another test on bigger plants or another substrate must be undertaken
12.	Interpretation of data in terms of UPOV characteristic states	class 0, 1, 2, 3 and 4: resistant class 5 and 6: susceptible

13.	Critical control points	reaction of standards (the infection pressure may vary between experiments, leading to slight differences in sporulation intensity); when the reactions are not clear the experiment should be repeated. the sowing on soil can be used to see necrosis, but weak sporulation (much less than susceptible control) can appear; when testing on sand, spores can be confused with grains of sand. in case of use of nutritive solution on blotting paper, a fungicide can be added to avoid contamination by saprophytes.
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For reference: The International Bremia Evaluation Board (IBEB) produces regular updates of the host differential reaction table. The most recent table is available through ISF at <http://www.worldseed.org/our-work/plant-health/other-initiatives/ibeb/>. Pictures for the observation scale are also provided.

Isolates	Differentials	Green Towers	Dandie	R4T57D	UC Dm14	NunDm15	CGDm16	Colorado	FrRsal-1	Argelès	RYZ 2164	RYZ910457	Bedford	Balesta	Bartoli	Design	Kibrille	Fenston	Bataille	RYZ20007
BI: 29EU		+	-	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-	-
BI: 30EU		+	-	+	+	+	-	+	-	+	+	-	-	-	-	+	-	-	-	-
BI: 31EU		+	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	-	-	-
BI: 32EU		+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	+	-	-	-
BI: 33EU		+	-	+	+	+	+	+	+	+	+	-	-	-	-	+	+	-	-	-
BI: 34EU		+	-	+	+	-	+	+	+	+	+	+	-	-	+	(-)	-	-	-	-
BI: 35EU		+	-	+	+	+	+	+	+	+	+	+	-	-	-	+	+	-	-	-
BI: 36EU		+	+	+	+	-	+	+	+	+	+	+	-	-	+	-	-	-	-	-
BI: 38EU		+	-	+	+	+	-	+	+	+	+	+	-	-	-	+	+	-	-	+
BI: 39EU		+	+	+	+	-	+	+	+	+	+	+	-	-	+	-	-	-	-	+
BI: 40EU		+	-	+	+	+	+	+	+	+	+	+	+	-	+	(-)	-	-	-	-

Legend: (+) indicates slightly reduced sporulation, (-) indicates no sporulation with necrosis or very weak sporulation, as defined in the harmonized scale given in Fig. 1. on <https://worldseed.org/our-work/disease-resistance/other-initiatives/ibeb/>

Ad. 47: Resistance to *Lettuce mosaic virus* (LMV) Pathotype II

Resistance to pathotype II to be tested in a bio-assay (method i) and/or in a DNA marker test (method ii).

(i) Bio-assay

1.	Pathogen	<i>Lettuce mosaic virus</i>
2.	Quarantine status	no
3.	Host species	lettuce - <i>Lactuca sativa</i> L.
4.	Source of inoculum	GEVES ⁴ (FR) or Naktuinbouw ⁵ (NL)
5.	Isolate	pathotype II (isolates LMV-0 and Ls1 belong to the same pathotype)
6.	Establishment isolate identity	resistant and susceptible controls
7.	Establishment pathogenicity	susceptible control inoculation
8.	Multiplication inoculum	
8.2	Multiplication variety	susceptible control
8.3	Plant stage at inoculation	2-3 leaves
8.4	Inoculation medium	0,05 M PBS, 0,25% (w/v) Na ₂ SO ₃ 0,5% C ₅ H ₁₀ NNaS ₂ .3H ₂ O, 4% carborundum and 5% active charcoal
8.5	Inoculation method	rubbing; optionally repeat after 4 d; 1-2 h high humidity after inoculation
8.6	Harvest of inoculum	homogenized fresh leaf in buffer (50% w/v); freeze-dried leaves can be kept less than 1 year in storage, long term storage at -80°C
8.7	Check of harvested inoculum	compare with mock inoculation with LMV buffer + carborundum + charcoal
8.8	Shelflife/viability inoculum	2 h at 4°C or on ice
9.	Format of the test	
9.1	Number of plants per genotype	at least 20
9.2	Number of replicates	1
9.3	Control varieties	susceptible: Bijou (red), Hilde II (green), Sprinter (green), Sucrine (green) resistant: Capitan (green), Corsica (green), Multired 80 (red)
9.4	Test design	several mock-inoculated plants in the same tray
9.5	Test facility	climate chamber
9.6	Temperature	after inoculation 15-22°C
9.7	Light	12-16 h light ca. 5000 lux
10.	Inoculation	
10.1	Preparation inoculum	fresh leaf ground in fresh LMV buffer incl. carborundum and active charcoal
10.3	Plant stage at inoculation	1st leaf well-developed at 1st inoculation, optionally 4 days later 2nd inoculation
10.4	Inoculation method	rubbing, rinse carborundum off
10.7	Final observations	21 days post inoculation
11.	Observations	
11.1	Method	visual estimate of mosaic severity; compare with standards, preferably with standards of same growth type.
11.2	Observation scale	resistant = no symptoms susceptible = growth retardation, young leaves with mosaic, leaf curling

⁴ matref@geves.fr

⁵ resistentie@naktuinbouw.nl

11.3	Validation of test	standards should conform to description
12.	Interpretation of data in terms of UPOV characteristic states	classify resistant or susceptible per plant, see 11.2.
13.	Critical control points	Sprinter is less susceptible than many other susceptible varieties, this variety can be used to detect low inoculation pressure in a specific experiment. anthocyanin coloration in leaves may mask mosaic symptoms and an earlier observation date for green varieties may be possible, depending on the reaction of the standard varieties in the test.

(ii) DNA marker test

The gene *mo1* (with its recessive alleles *mo1¹* or *mo1²*) gives resistance to LMV pathotype II. Alleles for resistance *mo1¹* and *mo1²* and the dominant allele for susceptibility *mo1⁰* can be detected by the co-dominant marker as described by V. Nicaise *et al.* (2003). Specific aspects:

1.	Pathogen	<i>Lettuce mosaic virus</i> pathotype II
2.	Functional gene	<i>mo1</i> (with two recessive alleles for resistance <i>mo1¹</i> and <i>mo1²</i> and one dominant allele for susceptibility <i>mo1⁰</i>)
3.	Probes and primers for Taqman PCR	
3.1.	Assay 1	to distinguish <i>mo1¹</i> genotypes from <i>mo1⁰</i> and <i>mo1²</i> genotypes (6 base deletion at nucleotide position 344-349):

Probe	DNA sequence '5-'3	Fluorophore color (optional)
Pr-del-mo1	GGCTCAAGGAGCTGACTTCTATTG	Texas Red (Susceptible)
Pr-del-mo1 ¹	GGCTCATGACTTCTATTG	6FAM-MGB (Resistant <i>mo1¹</i>)

Primers	DNA sequence '5-'3
Fw-del-mo1	CAACAACATACATCGACCAA
Rev-del-mo1	CTTCCCACTTAGGCTCGAT

Sequence amplicon: '5-'3

The amplicon sequence of the *mo1⁰* and *mo1²* allele:

TTACAACAACATACATCGACCAAGCAAGTTGGCTCAAGGAGCTGACTTCTATTGTTTCAAGAA
TAAAATCGAGCCTAAGTGGGAAGACC

The amplicon sequence of the allele for resistance *mo1¹*:

TTACAACAACATACATCGACCAAGCAAGTTGGCTCATGACTTCTATTGTTTCAAGAATAAAATC
GAGCCTAAGTGGGAAGACC

3.2.	Assay 2	to distinguish <i>mo1²</i> genotypes from <i>mo1⁰</i> and <i>mo1¹</i> genotypes (SNP at nucleotide position 228):																	
<table><tr><td>Probe</td><td>DNA sequence '5-'3</td><td>Fluorophore color (optional)</td></tr><tr><td>Pr-SNP228-<i>mo1</i></td><td>CTCCCTCTGCTAAGTC</td><td>6FAM-MGB (Susceptible)</td></tr><tr><td>Pr-SNP228-<i>mo1²</i></td><td>ACTCCCTCTCTAAGT</td><td>VIC-MGB (Resistant <i>mo1²</i>)</td></tr></table> <table><tr><td>Primers</td><td>DNA sequence '5-'3</td></tr><tr><td>Fw-SNP228-<i>mo1</i></td><td>GCATCCGCTCGAGCATTC</td></tr><tr><td>Rev-SNP228-<i>mo1</i></td><td>CTACCCCAAGCGACTTGCTT</td></tr></table> Sequence amplicon: '5-'3 The amplicon sequence of the <i>mo1⁰</i> and the <i>mo1¹</i> allele: TCAGCATCCGCTCGAGCATTCTTGGACTTTCTGGTTCGATACTCCCTCTGCTAAGTCCAAGCA AGTCGCTTGGGGTAGTTCCATGCGCC The amplicon sequence of the allele for resistance <i>mo1²</i>: TCAGCATCCGCTCGAGCATTCTTGGACTTTCTGGTTCGATACTCCCTCTCCTAAGTCCAAGCA AGTCGCTTGGGGTAGTTCCATGCGCC				Probe	DNA sequence '5-'3	Fluorophore color (optional)	Pr-SNP228- <i>mo1</i>	CTCCCTCT G CTAAGTC	6FAM-MGB (Susceptible)	Pr-SNP228- <i>mo1²</i>	ACTCCCTCT C TAAGT	VIC-MGB (Resistant <i>mo1²</i>)	Primers	DNA sequence '5-'3	Fw-SNP228- <i>mo1</i>	GCATCCGCTCGAGCATTC	Rev-SNP228- <i>mo1</i>	CTACCCCAAGCGACTTGCTT	
Probe	DNA sequence '5-'3	Fluorophore color (optional)																	
Pr-SNP228- <i>mo1</i>	CTCCCTCT G CTAAGTC	6FAM-MGB (Susceptible)																	
Pr-SNP228- <i>mo1²</i>	ACTCCCTCT C TAAGT	VIC-MGB (Resistant <i>mo1²</i>)																	
Primers	DNA sequence '5-'3																		
Fw-SNP228- <i>mo1</i>	GCATCCGCTCGAGCATTC																		
Rev-SNP228- <i>mo1</i>	CTACCCCAAGCGACTTGCTT																		
4.	Format of the test																		
4.1	Number of plants per genotype	at least 20 plants																	
4.2	Control varieties	Homozygous allele for susceptibility <i>mo1⁰</i> present: Sprinter, Sucrine Homozygous allele for resistance <i>mo1¹</i> present: Capitan, Kanaryole Homozygous allele for resistance <i>mo1²</i> present: Corianas Mix DNA to have heterozygous controls																	
5.	Preparation																		
5.1	Preparation DNA	Harvest per individual plant a part of a young leaf. Isolate total DNA with a standard DNA isolation protocol.																	
5.2	Preparation PCR	Pipette each DNA sample and a commercial real-time PCR mastermix into individual wells for assay 1 and for assay 2. Analyse the samples in a real-time PCR machine capable of reading the fluorophores of all the probes, with reaction conditions suitable for the mastermix used.																	
6.	PCR conditions	(detailed test protocol available through Naktuinbouw ⁶ (NL))																	
Assay 1:																			
<table><tr><td></td><td>Temperature</td><td>Time</td><td>Ramping speed</td></tr><tr><td>Initial activation of enzyme</td><td>95°C</td><td>2' 00"</td><td></td></tr><tr><td>40 cycles</td><td>95°C</td><td>0' 15"</td><td>5°C/sec</td></tr><tr><td></td><td>65°C</td><td>0' 48"</td><td>5°C/sec</td></tr></table>					Temperature	Time	Ramping speed	Initial activation of enzyme	95°C	2' 00"		40 cycles	95°C	0' 15"	5°C/sec		65°C	0' 48"	5°C/sec
	Temperature	Time	Ramping speed																
Initial activation of enzyme	95°C	2' 00"																	
40 cycles	95°C	0' 15"	5°C/sec																
	65°C	0' 48"	5°C/sec																
Assay 2:																			
<table><tr><td></td><td>Temperature</td><td>Time</td><td>Ramping speed</td></tr><tr><td></td><td>95°C</td><td>2' 00"</td><td></td></tr><tr><td>40 cycles</td><td>95°C</td><td>0' 15"</td><td>5°C/sec</td></tr><tr><td></td><td>60°C</td><td>0' 48"</td><td>5°C/sec</td></tr></table>					Temperature	Time	Ramping speed		95°C	2' 00"		40 cycles	95°C	0' 15"	5°C/sec		60°C	0' 48"	5°C/sec
	Temperature	Time	Ramping speed																
	95°C	2' 00"																	
40 cycles	95°C	0' 15"	5°C/sec																
	60°C	0' 48"	5°C/sec																
Analysis at end point RFU.																			

⁶ Naktuinbouw: resistentie@naktuinbouw.nl

7.	Observations																								
7.1	Observations scale																								
Assay 1:																									
<table><tr><td>Signal giving Fluorophore</td><td></td><td></td></tr><tr><td>FAM (<i>mo1¹</i>)</td><td>Texas Red (<i>mo1⁰</i> or <i>mo1²</i>)</td><td></td></tr><tr><td>-</td><td>x</td><td>Homozygous <i>mo1⁰</i> or <i>mo1²</i>, or heterozygous <i>mo1⁰mo1²</i></td></tr><tr><td>x</td><td>-</td><td>Homozygous <i>mo1¹</i></td></tr><tr><td>x</td><td>x</td><td>Heterozygous <i>mo1⁰mo1¹</i> or <i>mo1¹mo1²</i></td></tr><tr><td>-</td><td>-</td><td>No result, repeat test</td></tr></table>			Signal giving Fluorophore			FAM (<i>mo1¹</i>)	Texas Red (<i>mo1⁰</i> or <i>mo1²</i>)		-	x	Homozygous <i>mo1⁰</i> or <i>mo1²</i> , or heterozygous <i>mo1⁰mo1²</i>	x	-	Homozygous <i>mo1¹</i>	x	x	Heterozygous <i>mo1⁰mo1¹</i> or <i>mo1¹mo1²</i>	-	-	No result, repeat test					
Signal giving Fluorophore																									
FAM (<i>mo1¹</i>)	Texas Red (<i>mo1⁰</i> or <i>mo1²</i>)																								
-	x	Homozygous <i>mo1⁰</i> or <i>mo1²</i> , or heterozygous <i>mo1⁰mo1²</i>																							
x	-	Homozygous <i>mo1¹</i>																							
x	x	Heterozygous <i>mo1⁰mo1¹</i> or <i>mo1¹mo1²</i>																							
-	-	No result, repeat test																							
Assay 2:																									
<table><tr><td>Signal giving Fluorophore</td><td></td><td></td></tr><tr><td>FAM (<i>mo1⁰</i> or <i>mo1¹</i>)</td><td>VIC (<i>mo1²</i>)</td><td></td></tr><tr><td>(x) (FAM RFU << VIC RFU)</td><td>x</td><td>Homozygous <i>mo1²</i></td></tr><tr><td>x</td><td>-</td><td>Homozygous <i>mo1⁰</i> or <i>mo1¹</i>, or heterozygous <i>mo1⁰mo1¹</i></td></tr><tr><td>x</td><td>(x) (FAM RFU >> VIC RFU)</td><td>Heterozygous <i>mo1⁰mo1²</i> or <i>mo1¹mo1²</i></td></tr><tr><td>-</td><td>-</td><td>No result, repeat test</td></tr></table>			Signal giving Fluorophore			FAM (<i>mo1⁰</i> or <i>mo1¹</i>)	VIC (<i>mo1²</i>)		(x) (FAM RFU << VIC RFU)	x	Homozygous <i>mo1²</i>	x	-	Homozygous <i>mo1⁰</i> or <i>mo1¹</i> , or heterozygous <i>mo1⁰mo1¹</i>	x	(x) (FAM RFU >> VIC RFU)	Heterozygous <i>mo1⁰mo1²</i> or <i>mo1¹mo1²</i>	-	-	No result, repeat test					
Signal giving Fluorophore																									
FAM (<i>mo1⁰</i> or <i>mo1¹</i>)	VIC (<i>mo1²</i>)																								
(x) (FAM RFU << VIC RFU)	x	Homozygous <i>mo1²</i>																							
x	-	Homozygous <i>mo1⁰</i> or <i>mo1¹</i> , or heterozygous <i>mo1⁰mo1¹</i>																							
x	(x) (FAM RFU >> VIC RFU)	Heterozygous <i>mo1⁰mo1²</i> or <i>mo1¹mo1²</i>																							
-	-	No result, repeat test																							
7.2	Validation of the test	Control varieties should give the expected results.																							
8.	Interpretation of data in terms of UPOV characteristic states	The combination of the two PCR assays leads to the following predicted result in a bio-assay with LMV pathotype II:																							
<table><tr><td></td><td></td><td colspan="3">Assay 2 (<i>mo1²</i>)</td></tr><tr><td></td><td></td><td>absent</td><td>present homozygous</td><td>heterozygous</td></tr><tr><td rowspan="3">Assay 1 (<i>mo1¹</i>)</td><td>absent</td><td>susceptible (<i>mo1⁰</i>)</td><td>resistant (<i>mo1²</i>)</td><td>susceptible (<i>mo1⁰/mo1²</i>)</td></tr><tr><td>present homozygous</td><td>resistant (<i>mo1¹</i>)</td><td>-</td><td>-</td></tr><tr><td>heterozygous</td><td>susceptible (<i>mo1⁰/ mo1¹</i>)</td><td>-</td><td>not yet validated</td></tr></table>					Assay 2 (<i>mo1²</i>)					absent	present homozygous	heterozygous	Assay 1 (<i>mo1¹</i>)	absent	susceptible (<i>mo1⁰</i>)	resistant (<i>mo1²</i>)	susceptible (<i>mo1⁰/mo1²</i>)	present homozygous	resistant (<i>mo1¹</i>)	-	-	heterozygous	susceptible (<i>mo1⁰/ mo1¹</i>)	-	not yet validated
		Assay 2 (<i>mo1²</i>)																							
		absent	present homozygous	heterozygous																					
Assay 1 (<i>mo1¹</i>)	absent	susceptible (<i>mo1⁰</i>)	resistant (<i>mo1²</i>)	susceptible (<i>mo1⁰/mo1²</i>)																					
	present homozygous	resistant (<i>mo1¹</i>)	-	-																					
	heterozygous	susceptible (<i>mo1⁰/ mo1¹</i>)	-	not yet validated																					
		Heterozygous plants (<i>mo1⁰mo1¹</i> or <i>mo1⁰mo1²</i>) are predicted to be susceptible in the bio-assay, as <i>mo1¹</i> and <i>mo1²</i> are recessive alleles. Heterozygous plants <i>mo1¹mo1²</i> need a conclusion from a bio-assay. Varieties showing a mixture of genotypes (heterozygous plants <i>mo1⁰mo1¹</i> , <i>mo1⁰mo1²</i> or homozygous <i>mo1⁰</i> plants (susceptible predicted phenotype) and homozygous <i>mo1¹</i> or <i>mo1²</i> plants (resistant predicted phenotype)) are predicted to be non-uniform in the bio-assay. In case the DNA marker test result does not confirm the declaration in the TQ, a bio-assay should be performed to observe whether the variety is resistant due to on another mechanism.																							

Ad. 48: Resistance to *Nasonovia ribisnigri* (Nr) Biotype Nr: 0

1. Pathogen	<i>Nasonovia ribisnigri</i>
2. Quarantine status	no
3. Host species	lettuce - <i>Lactuca sativa</i> L.
4. Source of inoculum	Naktuinbouw ⁷ (NL)
5. Isolate	Nr: 0, preferably red colored biotype
6. Establishment isolate identity	the ends of the legs are black, size 1.5-2.5 mm
7. Establishment pathogenicity	with susceptible control Abel or Green Towers
8. Multiplication inoculum	
8.2 Multiplication variety	Abel or Green Towers
8.3 Plant stage at inoculation	4 to 6 leaves
8.5 Inoculation method	transfer ~5 aphids per plant
8.6 Harvest of inoculum	transfer to Petri-dish; shake off when aphids are numerous carefully remove aphids using a fine painting brush when only few are available
8.7 Check of harvested inoculum	check the black ends of the aphids legs
8.8 Shelf life/viability inoculum	a few hours in shadow
9. Format of the test	
9.1 number of plants per genotype	at least 20
9.2 number of replicates	no
9.3 Control varieties	susceptible: Abel, Green Towers, Nadine resistant: Barcelona, Bedford, Dynamite, Silvinas
9.4 Test design	
9.5 Test facility	glasshouse
9.6 Temperature	after inoculation: 20-22°C, keep below 26°C
9.7 Light	daylight
9.9 Special measures	containment of winged aphids needs special attention
10. Inoculation	
10.1 Preparation inoculum	transfer by shake-off or with brush into Petri-dish
10.3 Plant stage at inoculation	2 to 3 week old seedlings
10.4 Inoculation method	transfer 5 small or medium sized aphids to each plant
10.7 Final observations	15 to 20 days post inoculation
11. Observations	
11.1 Method	count red aphids per plant; if many aphids are present, strong growth reduction can be observed; for this observation, a separate aphid free tent is necessary for blanks
11.2 Observation scale	0 no aphids 1 1-5 aphids 2 6-10 aphids 3 >10 aphids
11.3 Validation of test	controls should be >95% ok; if >5% plants are in class 2 or off-type, the experiment should be repeated
12. Interpretation of data in terms of UPOV characteristic states	0 or 1 resistant 3 susceptible
13. Critical control points	allow sufficient time for the aphids born after inoculation to mature and turn red; as soon as this is the case, the test must be concluded; this may be before 15 days post inoculation. only adult, red aphids are counted; young aphids are transparent and do not count

⁷ resistentie@naktuinbouw.nl

Ad. 49: Resistance to *Fusarium oxysporum* f.sp. *lactucae* (Fol) Race 1

1. Pathogen	<i>Fusarium oxysporum</i> f.sp. <i>lactucae</i>								
2. Quarantine status	EPPO alert list								
3. Host species	lettuce - <i>Lactuca sativa</i> L.								
4. Source of inoculum	NIAS Genebank ⁸ (JP), CREA-SCS ⁹ (IT), Naktuinbouw ¹⁰ (NL), GEVES ¹¹ (FR)								
5. Isolate	Fol: 1								
6. Establishment isolate identity	use microscope and inoculation to lettuce susceptible standard								
7. Establishment pathogenicity	use lettuce susceptible standard								
8. Multiplication inoculum									
8.1 Multiplication medium	inoculation by sowing on contaminated soil: Wheat bran-soil medium inoculation by soaking seedlings: on synthetic liquid medium (e.g. Potatoes Dextrose Broth)								
8.6 Harvest of inoculum	inoculation by sowing on contaminated soil: 7-10 day-old culture inoculation by soaking seedlings: 15 days								
9. Format of the test									
9.1 Number of plants per genotype	at least 30, in case of doubt 60								
9.2 Number of replicates	at least 2								
9.3 Control varieties	absent or low resistance: Cobham Green, Patriot (Cobham Green is slightly less susceptible than Patriot) medium resistance: Affic, Fuzila, Natexis (Natexis is the lower level of medium resistance) high resistance: Costa Rica No.4, Romasol								
9.4 Test design	include control varieties								
9.5 Test facility	greenhouse or climate room								
9.6 Temperature	25-28 °C (day) / 20 °C (night)								
9.7 Light	under natural day length								
10. Inoculation	two methods can be used for inoculation:								
	<table border="1"> <thead> <tr> <th>sowing seeds on contaminated soil</th><th>soaking seedlings</th></tr> </thead> <tbody> <tr> <td>wheat bran-soil medium culture mixed with sterilized soil</td><td>soaking of roots and of hypocotyl axis for 5 to 15 min in the inoculum suspension</td></tr> <tr> <td>soil: culture = 20 : 1</td><td>spores are harvested and adjusted to 10⁶ to 10⁷ sp/ml</td></tr> <tr> <td>seeds stimulated to emerge (remark: avoid seeds rotted by factors other than pathogen)</td><td>cotyledons to 2 or 3 leaves appearing</td></tr> </tbody> </table>	sowing seeds on contaminated soil	soaking seedlings	wheat bran-soil medium culture mixed with sterilized soil	soaking of roots and of hypocotyl axis for 5 to 15 min in the inoculum suspension	soil: culture = 20 : 1	spores are harvested and adjusted to 10 ⁶ to 10 ⁷ sp/ml	seeds stimulated to emerge (remark: avoid seeds rotted by factors other than pathogen)	cotyledons to 2 or 3 leaves appearing
sowing seeds on contaminated soil	soaking seedlings								
wheat bran-soil medium culture mixed with sterilized soil	soaking of roots and of hypocotyl axis for 5 to 15 min in the inoculum suspension								
soil: culture = 20 : 1	spores are harvested and adjusted to 10 ⁶ to 10 ⁷ sp/ml								
seeds stimulated to emerge (remark: avoid seeds rotted by factors other than pathogen)	cotyledons to 2 or 3 leaves appearing								
10.1 Preparation inoculum									
10.2 Quantification inoculum									
10.3 Plant stage at inoculation									
10.4 Inoculation method	two methods can be used, as described above								
10.5 First observation	7- 10 days post inoculation								
10.6 Second observation	14 days post inoculation								
10.7 Final observations	20-25 days post inoculation (sowing or soaking). One or two of these 3 observations may be sufficient. The observation for inoculation by soaking is destructive since stems are cut for the observation of vessels.								

⁸ genebank@nias.affrc.go.jp

⁹ scs.sa@crea.gov.it

¹⁰ resistentie@naktuinbouw.nl

¹¹ matref@geves.fr

11. Observations

11.1 Method

visual and/or counting number of plants with symptom; as information calculate a disease index.

11.2 Observation scale

inoculation by sowing seeds on contaminated soil	inoculation by soaking seedlings
Class 0: healthy	Class 0: plant without symptoms and healthy vessels 
Class 1: slightly stunting, growing reduction	Class 1: plant with brown vessels only below the cotyledon without yellowing and wilting 
Class 2: severely stunting	Class 2: plant with brown vessels above the cotyledon, without yellowing and wilting 
Class 3: dead plant	Class 3: plant yellowing and wilting, brown vessels 
	Class 4: dead plant 

11.3 Validation of test	Validation based on control varieties. Expected behavior of control varieties: Absent or low resistance: most plants in classes 3 and 4, few plants observed in classes 2 and 1 Medium resistance: most plants in classes 1, 2 and 3 High resistance: most plants in classes 0 and 1, few plants in class 2
12. Interpretation of data in terms of UPOV characteristic states	Results should be compared with the results of the control varieties based on distribution of plants over the classes. For varieties close to medium resistance borders, an additional statistical analysis can be used to finalize the assessment of uniformity, and relative position regarding the results of control varieties.

Ad. 50: Resistance to *Fusarium oxysporum* f.sp. *lactucae* (Fol) Race 4

1. Pathogen	<i>Fusarium oxysporum</i> f.sp. <i>lactucae</i>
2. Quarantine status	NO : EPPO alert list
3. Host species	lettuce - <i>Lactuca sativa</i> L.
4. Source of inoculum	e.g. GEVES ¹² (FR)
5. Isolate	Fol: 4, e.g. isolate 04750888 reference strain validated in an interlaboratory test ¹³ Other validated isolates may be used, as long as producing the same results on the differential set.
6. Establishment isolate identity	The most recent table of differential set is available through ISF at https://worldseed.org/our-work/disease-resistance/differential-hosts/
7. Establishment pathogenicity	use lettuce susceptible standard
8. Multiplication inoculum	
8.1 Multiplication medium	on liquid medium (e.g. Potatoes Dextrose Broth)
8.6 Harvest of inoculum	15 days
9. Format of the test	
9.1 Number of plants per genotype	at least 30 plants, in case of doubt 60 plants
9.2 Number of replicates	at least 2 replicates
9.3 Control varieties	absent or low resistance: Gisela medium resistance: Ballerina and Patriot (Ballerina is the lowest level of medium resistance) high resistance: Lomeria, Palmos
9.4 Test design	include control varieties
9.5 Test facility	greenhouse or climate room
9.6 Temperature	23-26°C
9.7 Light	under natural day length
10. Inoculation	
10.1 Preparation inoculum	spores are harvested
10.2 Quantification inoculum	adjust to 10 ⁶ sp/ml
10.3 Plant stage at inoculation	cotyledons to 2 or 3 leaves appearing
10.4 Inoculation method	soaking roots and hypocotyl for 5 to 15 min in the inoculum suspension
10.5 First observation	-
10.6 Second observation	-
10.7 Final observations	20-25 days post inoculation
11. Observations	
11.1 Method	visual and/or counting number of plants with symptom.
11.2 Observation scale	

¹² matref@geves.fr

¹³ See ISF EG DRT Fol: 4 resistance in lettuce <https://worldseed.org/document/isf-final-report-fol-race-4-lettuce/>

inoculation by soaking seedlings	
Class 0: plant without symptoms and healthy vessels	
Class 1: plant with brown vessels only below the cotyledon without yellowing and wilting	
Class 2: plant with brown vessels above the cotyledon, without yellowing and wilting	
Class 3: plant yellowing and wilting, brown vessels	
Class 4: dead plant	

11.3 Validation of test

Validation based on control varieties.

Expected behavior of control varieties:

Absent or low resistance: most plants in classes 3 and 4, few plants observed in classes 2 and 1

Medium resistance: most plants in classes 1, 2 and 3

High resistance: most plants in classes 0 and 1, few plants in class 2

12. Interpretation of data in terms of UPOV characteristic states

Results should be compared with the results of the control varieties based on distribution of plants over the classes.

For varieties close to medium resistance borders, an additional statistical analysis can be used to finalize the assessment of uniformity, and relative position regarding the results of control varieties.

8.3 Lettuce types

See also 5.3 for a table to determine the type using several characteristics.



Butterhead type

Heading; thin to rather thick, tender leaves with a clear midrib; leaf shape circular to transverse broad elliptic; in general no incised margin; head shape ranging from broad elliptic to transvers elliptic.



Novita type

Cross between Butterhead and Iceberg type for glasshouse growing. Open heading; leaf structure like Butterhead, incisions of the margin as Iceberg.



Iceberg type

Heading with strong or very strong overlapping of upper part of leaves; thick and crispy leaves, predominantly green and greyish green, leaf margin hardly to rather strongly incised, no clear midrib but with flabellate venation.



Batavia type

Open to strong heading; generally medium thick, rather strongly blistered leaves, predominantly yellowish or medium green; leaf margin with weak to strong undulation.



Frisée d'Amérique type

Non-heading, loose, generally quite large plant; thin leaves. Compared to Lollo type in general less undulating margin and showing more leaf blade. Compared to Batavia type, leaves are thinner. Mainly used for babyleaf production.



Lollo type

Non-heading; thin leaves with strongly undulated leaf margin. The plant as a whole shows mainly the undulating leaf margins. In general strongly blistered leaves, blisters are rather small.



Oakleaf type

Thin, divided leaves; divisions have an oakleaf or lobed shape with in general a rounded tip. Radichetta or Catalogna with acute tip of the division. Heart can be loose to dense.



Multi-divided type

Non-heading; thin, medium to very strong divided leaves. Tip of divisions can be undulated and incised. Plant may look as a Lollo type, but leaves are always divided.



Frillice type

Non-heading; thick, crispy leaves, sometimes weakly divided. Clearly incised leaf margin.



Cos type

Elongated and rather tough leaves with a clear midrib, head shape in longitudinal section elliptic, length of head $>1.5 \times$ diameter; heading can be very late.



Gem type

Tough leaves with clear midrib, head shape short elliptic to slightly obovate. Some types only have a tightly filled heart, others are more similar to a short Cos type. Suitable for semi-arid conditions.



Stem type

Forms a fleshy stem before bolting, at least under (semi-)short day conditions; leaves are mainly tough and have a clear midrib. Leaves and/or stem are consumed.

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10. Technical Questionnaire

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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	Application date: (not to be filled in by the applicant)
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TECHNICAL QUESTIONNAIRE to be completed in connection with an application for plant breeders' rights	
1.	Subject of the Technical Questionnaire
1.1	Botanical name Lactuca sativa L.
1.2	Common name Lettuce
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:
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#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 []

(b) partially known cross []

(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)

--

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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4.2 Method of propagating the variety

4.2.1 Seed-propagated varieties

(a) Self-pollination

[]

(b) Other (please provide details)

[]

4.2.2 Other
(Please provide details)

[]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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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 Seed: color (1)			
white	Verpia	1 []	
yellow	Durango	2 []	
brown	Oaklin	3 []	
black	Kagraner Sommer 2	4 []	
5.2 Leaf: number of divisions (6)			
absent or very few	Fiorella, Lollo rossa	1 []	
very few to few		2 []	
few	Curletta, Rodagio	3 []	
few to medium		4 []	
medium	Ezabel, Jadigon	5 []	
medium to many		6 []	
many	Expedition, Multired 54	7 []	
many to very many		8 []	
very many	Excite, Ezfrill, Telex	9 []	
5.3 Leaf: anthocyanin coloration (11)			
absent or very weak	Clarion	1 []	
very weak to weak		2 []	
weak	Du bon jardinier	3 []	
weak to medium		4 []	
medium	Lollo rossa, Luana	5 []	
medium to strong		6 []	
strong	Merveille des quatre saisons	7 []	
strong to very strong		8 []	
very strong	Iride, Revolution	9 []	
5.4 Leaf: hue of anthocyanin coloration (12)			
reddish	Lollo rossa	1 []	
purplish	Iride	2 []	
brownish	Luana, Maravilla de Verano	3 []	

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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Characteristics	Example Varieties	Note
5.5 Leaf: area covered by anthocyanin coloration (13)		
very small	Steirer Krauthauptel	1 []
very small to small		2 []
small	Diablo	3 []
small to medium		4 []
medium	Luana	5 []
medium to large		6 []
large	Merveille des quatre saisons	7 []
large to very large		8 []
very large	Bijou, Revolution	9 []
5.6 Leaf: color (14)		
green	Verpia	1 []
yellowish green	Dorée de printemps	2 []
greyish green	Celtuce, Du bon jardinier	3 []
5.7 Leaf: intensity of green color (15)		
very light		1 []
very light to light		2 []
light	Blonde maraîchère, Lollo Bionda	3 []
light to medium		4 []
medium	Aquarel, Clarion	5 []
medium to dark		6 []
dark	Expedition, Verpia	7 []
dark to very dark		8 []
very dark	Pascal, Verdatrix	9 []
5.8 Only varieties with Plant: degree of overlapping of upper part of leaves: medium or strong: Time of harvest maturity (34)		
very early	Gotte jaune d'or	1 []
very early to early		2 []
early	Pantlika, Sucrine	3 []
early to medium		4 []
medium	Clarion	5 []
medium to late		6 []
late	Blonde maraîchère, Calmar	7 []
late to very late		8 []
very late	El Toro, Pinokkio	9 []

TECHNICAL QUESTIONNAIRE		Page {x} of {y}	Reference Number:
Characteristics	Example Varieties	Note	
5.9 Time of beginning of bolting (35)			
very early	Blonde à couper améliorée	1 []	
very early to early		2 []	
early	Gotte à graine blanche	3 []	
early to medium		4 []	
medium	Pantlika	5 []	
medium to late		6 []	
late	Hilde II	7 []	
late to very late		8 []	
very late	Erika, Roxette	9 []	
5.10 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 29EU (38)			
absent	Argelès	1 []	
present	Balesta	9 []	
5.11 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 30EU (39)			
absent	Argelès, Colorado	1 []	
present	Balesta	9 []	
not tested		[]	
5.12 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 31EU (40)			
absent	Colorado, RYZ910457	1 []	
present	Argelès, Balesta	9 []	
not tested		[]	
5.13 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 33EU (41)			
absent	Kibrille, RYZ2164	1 []	
present	RYZ910457	9 []	
not tested		[]	
5.14 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 35EU (42)			
absent	Design, Kibrille	1 []	
present	Bartoli	9 []	
not tested		[]	
5.15 Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 36EU (43)			
absent	Bartoli, RYZ2164	1 []	
present	Design, Kibrille	9 []	
not tested		[]	

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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Characteristics	Example Varieties	Note
5.16 (44) Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 38EU		
absent	Design, Kibrille	1 []
present	Bartoli	9 []
not tested		[]
5.17 (45) Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 39EU		
absent	Bartoli, Dandie, RYZ2164	1 []
present	Design, Kibrille	9 []
not tested		[]
5.18 (46) Resistance to <i>Bremia lactucae</i> (BI) Isolate BI: 40EU		
absent	Bartoli, RYZ2164	1 []
present	Kibrille	9 []
not tested		[]
5.19 (47) Resistance to <i>Lettuce mosaic virus</i> (LMV) Pathotype II		
absent	Bijou, Hilde II, Sprinter, Sucrine	1 []
present	Capitan, Corsica	9 []
not tested		[]
5.20 (48) Resistance to <i>Nasonovia ribisnigri</i> (Nr) Biotype Nr: 0		
absent	Abel, Green Towers, Nadine	1 []
present	Barcelona, Bedford, Dynamite, Silvinas	9 []
not tested		[]
5.21 (49) Resistance to <i>Fusarium oxysporum</i> f.sp. <i>lactucae</i> (Fol) Race 1		
absent or low	Cobham Green, Patriot	1 []
medium	Affic, Fuzila, Natexis	2 []
high	Costa Rica No. 4, Romasol	3 []
not tested		[]
5.22 (50) Resistance to <i>Fusarium oxysporum</i> f.sp. <i>lactucae</i> (Fol) Race 4		
absent or low	Costa Rica No. 4, Gisela	1 []
medium	Ballerina, Patriot	2 []
high	Lomeria, Palmos	3 []
not tested		[]

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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
<i>Example</i>	<i>Plant: diameter</i>	<i>medium</i>	<i>medium to large</i>
Comments:			

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#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

Type (see 5.3 and 8.3 in the Test Guidelines for Lettuce (document TG/13/11 Rev.) for explanations):

Type	Example varieties	
Butterhead type	Clarion, Maikönig, Sartre	☐
Novita type	Norvick	☐
Iceberg type	Great Lakes 659, Roxette, Saladin, Vanguard 75	☐
Batavia type	Aquarel, Curtis, Funnice, Felucca, Grand Rapids, Masaida, Visyon	☐
Frisée d'Amérique type	Bijou, Blonde à couper améliorée	☐
Lollo type	Lollo rossa, Revolution	☐
Oakleaf type	Catalogna, Kipling, Murai, Salad Bowl	☐
Multi-divided type	Curletta, Duplex, Jadigon, Rodagio	☐
Frillice type	Frilett	☐
Cos type	Actarus, Blonde maraîchère, Pinokkio	☐
Gem type	Craquerelle du Midi, Sucrine, Xanadu	☐
Stem type	Celtuce, Guasihong	☐

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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".

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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

[End of document]