



# PBRO

## Plant Breeders' Rights Office

<b>TEST GUIDELINE</b>
<b>APPLE (Fruit Varieties)</b>
<i>Malus domestica</i> Borkh.
<b>Version Date: 2019/09</b>

Completed forms are to be returned to:

Plant Breeders' Rights Office  
59 Camelot Drive  
Ottawa, Ontario, Canada  
K1A 0Y9

## TABLE OF CONTENTS

A.	INTRODUCTION.....	3
B.	REFERENCE VARIETIES.....	3
C.	COMPARATIVE PHOTOGRAPHS .....	3
D.	CHARACTERISTICS.....	4
E.	CONDUCTING COMPARATIVE TESTS & TRIALS.....	5
	General Principles .....	5
	Specific Protocols for Apple fruit varieties.....	6
F.	DESCRIPTION FORM.....	7
	Varieties in trial.....	7
	Description of the comparative tests and trials .....	8
	Table of characteristics .....	9
H.	EXPLANATIONS AND ILLUSTRATIONS.....	17

## A. INTRODUCTION

To complete the examination process, the following requirements are to be submitted to the Plant Breeders' Rights (PBR) Office within 6 months of completion of the site examination conducted by a PBR examiner:

- a completed description form (Section F); this information is important for providing a thorough description of the candidate variety in the event that the breeders' right on this variety is challenged,
- a description of the trials; this information is important to ensure that as much as possible, the trial could be duplicated in the event a re-trial is required, and
- 1 or more comparative photographs.

More detailed information on PBR trials can be found in the document *Guidelines for Conducting PBR Comparative Tests and Trials* which is available at the PBR web-site or from the PBR Office.

## B. REFERENCE VARIETIES

- 1) Candidate varieties must be compared to the most similar variety(ies) available in Canada at the time of application. If there are other similar foreign varieties, these should also be used for comparison.
- 2) The selection of the reference variety(ies) to be grown in the trial with the candidate variety(ies) is aided by the use of grouping characteristics which are known from experience not to vary in the documented states of expression, or to vary only slightly, even when the variety is produced at different locations. For the list of grouping characteristics for this crop kind, see Specific Protocol in Section E.

## C. COMPARATIVE PHOTOGRAPHS

- 1) Comparative photographs of characteristics which clearly demonstrate that the candidate variety is distinct from the reference variety(ies) must be submitted.
- 2) Both the candidate and reference variety(ies) should be in the same frame of the photograph. If this is not possible, a common reference measure (e.g. ruler) should be included in each photograph.
- 3) The photographs should be based on plant material from the comparative tests and trials.
- 4) The comparative photographs should clearly show how the candidate variety is distinct from the reference varieties. For example, if the candidate variety is different from the reference variety in plant height, growth habit, etc., then photographs of the whole plants may be necessary. However, if the distinguishing characteristics are more specific or related to smaller plant parts, such as flower colour, leaf size, etc., then it would be more appropriate to remove these parts from the plants and take close-up photographs. For the latter examples, use of a macro lens is recommended. Also, the subject of the photographs should occupy as much of the frame of the photograph as possible.

- 5) The candidate and reference variety(ies) should be clearly labelled in the comparative photographs. The candidate variety should be labelled with the proposed denomination. The reference variety(ies) should be labelled with the name by which they were protected (if such is the case) or by the name by which they are known (commercial name).
- 6) Where suitable, photographs taken indoors should be done against a grey-neutral background while photographs taken outdoors should be taken on an overcast day or in light shade, not in full sun.
- 7) The comparative photographs must be submitted in jpeg format if available. The jpeg files can either be submitted on a disc or as an attachment to an e-mail addressed to the PBR Office. As much as possible, submit each jpeg file in a 4x6" format (10.15 x 15.24 cm).

## **D. CHARACTERISTICS**

- 1) To assess distinctness, uniformity and stability, the characteristics listed in this document should be used. If additional characteristics are needed, a description of these characteristics, including the rating of the observation for all varieties, must be submitted.
- 2) Characteristics used to describe the candidate variety must also be described for all reference varieties.
- 3) The asterisked (\*) characteristics are compulsory. It is recommended that the other characteristics also be completed to ensure a full description of the variety is on record.
- 4) Many characteristics have a rating system which falls within a 1-9 note scale. Characteristics may be rated with intermediate note values where the observation of the characteristic falls between one state of expression and another. For example, where the states of expression for a characteristic are: small (3), medium (5), large (7) and your observation rates between small (3) and medium (5), a note of (4) may be assigned. Also, extremes of the scale (e.g. 1 or 9) may be used.

## E. CONDUCTING COMPARATIVE TESTS & TRIALS

### General Principles

- 1) The trials used for the purpose of demonstrating distinctness, uniformity and stability (DUS) of the candidate variety must be conducted in Canada.
- 2) The trials 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) The design of the trial 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.
- 4) Additional tests for examining relevant characteristics may also be used (e.g. tests for disease resistance). The protocols of these additional tests must be described in full.
- 5) All observations and measurements must be based on plant material from the PBR trials conducted in Canada.
- 6) Observations must be taken for both the candidate and reference varieties from plants grown at the same location under the same conditions.
- 7) Where more than one growing cycle of trials is conducted, the trials must be grown at the same location.
- 8) Where more than one growing cycle of trials is conducted, the characteristic ratings and measurements (including the mean, number of observations, standard deviation and range) from each growing cycle should be submitted as two separate sets of data.
- 9) When measured characteristics are used to demonstrate distinctness, you may be required to submit additional statistical support (e.g. LSD, ANOVA, p-value, etc.).
- 10) It is recommended that the raw data (e.g. leaf measurements) be retained in the event that the data are required by the PBRO to verify test results. If raw data are not available, further trials may be required.

## Specific Protocols for Apple, Fruit Varieties

- 1) Data must be collected from trials conducted for the duration of 1 growing cycle. However, the plants must be of a suitable maturity to be exhibiting characteristics representative of the variety. Otherwise, further growing cycles may be required.
- 2) As a minimum, the trial must include a total of:
  - a. 5 trees for varieties resulting from crossing
  - b. 10 trees for varieties resulting from mutation.
- 3) **(a) Varieties resulting from crossing:**
  - Unless otherwise indicated, all observations should be made on 5 trees or parts of 5 trees.
  - In the case of parts of the trees, the number to be taken from each of the trees should be 2.**(b) Varieties resulting from mutation:**
  - Unless otherwise indicated, all observations should be made on 10 trees or parts of 10 trees.
  - In the case of parts of the trees, the number to be taken from each of the trees should be 1.
- 4) For the assessment of uniformity, a population standard of 1% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 5 trees, no off-types are allowed. In the case of a sample size of 10 trees, 1 off-type is allowed.
- 5) Grouping Characteristics

To select appropriate reference varieties for Apple, fruit varieties ensure that when they are compared to the candidate variety, the reference variety shows the same state of expression for as many of the following grouping characteristics as possible:

- i) Tree: type (characteristic 2)
- ii) Only varieties with ramified tree type: Tree: habit (characteristic 3)
- iii) Fruit: general shape (characteristic 28)
- iv) Fruit: relative area of over colour (characteristic 36)
- v) Fruit : hue of over colour – with bloom removed (characteristic 37)
- vi) Fruit: pattern of over colour of skin (characteristic 39)
- vii) Time of beginning of flowering (characteristic 55)
- viii) Time of eating maturity (characteristic 57)

## F. DESCRIPTION FORM

Information reported in this document may be accessible or protected as required under the provisions of the Access to Information Act. Information that could cause you or your organization harm if released is protected from disclosure as defined in Section 20 of the Access to Information Act.

**Varieties in trial:** List the denominations of the candidate variety (CV) and the reference varieties (RV) which have been selected for use in these comparative tests and trials.

**PBR APPLICATION NUMBER OF CV:** \_\_\_\_\_

	<b>Denomination</b>
<b>CV</b>	
<b>RV1</b>	
<b>RV2</b>	
<b>RV3</b>	
<b>RV4</b>	

**Description of the comparative tests and trials:** Fill in the following table with the requested information, if applicable. The information requested in this section should be completed to ensure that as much as possible, the trial could be duplicated in the event a retrial was required.

<b>Location of trial:</b>
<b>Year(s) when comparative data was collected:</b>
<b>Only fill out the trial parameters listed below which are applicable to this trial.</b>
<b>Number of trees per variety:</b>
<b>Spacing between trees (m) or planting density:</b>
<b>Spacing between rows (m):</b>
<b>Rootstock variety:</b>
<b>Year trees planted:</b>
<b>Other information (describe):</b>



**Table of characteristics:** Fill in the following table as much as possible to ensure a full description of the variety is available. This could be necessary in the event that the breeder's right on this variety is challenged.

Characteristic		CV	RV1	RV2	RV3	RV4
<b>For characteristic 1, tree vigour should be observed in summer when the tree is in peak vegetative growth and should be considered as the overall abundance of vegetative growth.</b>						
1		<b>Tree: vigour</b> (1-very weak, 3-weak, 5-medium, 7-strong)				
<b>For characteristics 2 and 3, tree type and habit, observe bare trees in winter.</b>						
2	(*) (+)	<b>Tree: type</b> (1-columnar, 2-ramified)				
3	(*) (+)	<b><u>Only varieties with ramified tree type: Tree: habit</u></b> (1-upright, 2-spreading, 3-drooping, 4-weeping)				
<b>For characteristic 4, type of bearing, observations should be made (forty) 40 days after flowering.</b>						
4	(+)	<b>Tree: type of bearing</b> (1-on spurs only, 2-on spurs and long shoots, 3-on long shoots only)				
<b>For characteristics 5 to 9, one year old shoots, observations should be made on lateral dormant shoots in winter, on trees that have completed at least one growing cycle at the testing location.</b>						
<b>For characteristic 5, the thickness of the one-year-old shoot should be observed in the centre of the middle internode. Measurements can be made using a Vernier calliper gauge.</b>						
5		<b>One-year-old shoot: thickness</b> (3-thin, 5-medium, 7-thick, 9-very thick)				
<b>For characteristic 6, the length of the internode should be observed in the middle third of the shoot. Measurements can be made using a Vernier calliper gauge.</b>						
6	(*)	<b>One-year-old shoot: length of internode (cm)</b>				
		mean				
		range				
		standard deviation				
		number measured				

LEGEND: (\*) compulsory characteristic which must always be completed  
 (+) see illustration or clarification in section G for additional information

Characteristic			CV	RV1	RV2	RV3	RV4
7		<b>One-year-old shoot: colour on sunny side</b> (1-greenish brown, 2-reddish brown, 3-light brown, 4-medium brown, 5-dark brown)					
8		<b>One-year-old shoot: pubescence on distal half of shoot</b> (1-absent or very sparse, 3-sparse, 5-medium, 7-dense, 9-very dense)					
9	(*)	<b>One-year-old shoot: number of lenticels</b> (3-few, 5-medium, 7-many)					
<p><b>For characteristics 10 to 18, observations should be made on fully developed leaves from the middle third of vigorous current season shoots from the outside of the tree.</b></p>							
10	(*) (+)	<b>Leaf blade: attitude in relation to shoot</b> (1-upwards, 2-outwards, 3-downwards)					
11	(*)	<b>Leaf blade: length (cm)</b>					
		mean					
		range					
		standard deviation					
12	(*)	<b>Leaf blade: width (cm)</b>					
		mean					
		range					
		standard deviation					
13	(*)	<b>Leaf blade: ratio length/width</b> (3-small, 5-medium, 7-large)					
14		<b>Leaf blade: intensity of green colour</b> (3-light, 5-medium, 7-dark)					
15	(+)	<b>Leaf blade: incisions of margin (upper half)</b> (1-crenate, 2-bicrenate, 3-serrate type 1, 4-serrate type 2, 5-biserrate)					
16		<b>Leaf blade: pubescence on lower side</b> (1-absent or sparse, 2-medium, 3-dense)					

Characteristic			CV	RV1	RV2	RV3	RV4
17	(*)	<b>Petiole: length (mm)</b>					
		mean					
		range					
		standard deviation					
		number measured					
18		<b>Petiole: extent of anthocyanin colouration from base</b> (3-small, 5-medium, 7-large)					
<p>For characteristics 19 to 22, observations should be made on the second or subsequent flowers, at the start of anther dehiscence.</p>							
<p>For characteristic 19, the “balloon stage” is the phenological stage in the course of flower development when the calyx is fully expanded and the petals are recognizable, having partially expanded and inflated but are closed, covering the internal flower organs. The balloon stage is usually 1-2 days before the petals unfold.</p>							
19	(*)	<b>Flower: predominant colour at balloon stage</b> (1-white, 2-yellowish pink, 3-light pink, 4-dark pink, 5-medium red, 6-dark red, 7-purple)					
20	(*)	<b>Flower: diameter with petals pressed into horizontal position (mm)</b>					
		mean					
		range					
		standard deviation					
		number measured					
21	(*) (+)	<b>Flower: arrangement of petals</b> (1-free, 2-intermediate, 3-overlapping)					
22	(+)	<b>Flower: position of stigmas relative to anthers</b> (1-below, 2-same level, 3-above)					
<p>For characteristic 23, observations should be made on young fruit forty (40) days after flowering.</p>							
23		<b>Young fruit: extent of anthocyanin over colour</b> (1-absent or very small, 3-small, 5-medium, 7-large, 9-very large)					

Characteristic		CV	RV1	RV2	RV3	RV4
<p>Unless otherwise indicated, observations on the fruit should be made on ten (10) typical fruits taken from a minimum sample of twenty (20) fruits, at the time of ripeness for eating. The terminal (king) fruit should be excluded from the sample.</p>						
24	(*)	<b>Fruit: size</b> (1-very small, 2-very small to small, 3-small, 4-small to medium, 5-medium, 6-medium to large, 7-large, 8-large to very large, 9-very large)				
25	(*)	<b>Fruit: height (cm)</b>				
	(+)	mean				
		range				
		standard deviation				
26	(*)	<b>Fruit: diameter (cm)</b>				
	(+)	mean				
		range				
		standard deviation				
26	(+)	number measured				
		number measured				
		number measured				
		number measured				
27	(*)	<b>Fruit: ratio height/diameter</b> (1-very small, 3-small, 5-medium, 7-large, 9-very large)				
28	(*)	<b>Fruit: general shape</b> (1-cylindrical waisted, 2-conic, 3-ovoid, 4-cylindrical, 5-ellipsoid, 6-globose, 7-obloid)				
29		<b>Fruit: ribbing</b> (1-absent or weak, 2-moderate, 3-strong)				
30		<b>Fruit: crowning at calyx end</b> (1-absent or weak, 2-moderate, 3-strong)				
31	(*)	<b>Fruit: size of eye</b> (3-small, 5-medium, 7-large)				
32		<b>Fruit: length of sepal</b> (3-short, 5-medium, 7-long)				
33	(*)	<b>Fruit: bloom of skin</b> (1-absent or weak, 2-moderate, 3-strong)				
34		<b>Fruit: greasiness of skin</b> (1-absent or weak, 2-moderate, 3-strong)				
35	(*)	<b>Fruit: ground colour</b> (1-not visible, 2-whitish yellow, 3-yellow, 4-whitish green, 5-yellow green, 6-green)				

LEGEND: (\*) compulsory characteristic which must always be completed  
 (+) see illustration or clarification in section G for additional information

Characteristic			CV	RV1	RV2	RV3	RV4
36	(*)	<b>Fruit: relative area of over colour</b> (1-absent or very small, 3-small, 5-medium, 7-large, 9-very large)					
37	(*)	<b>Fruit: hue of over colour (with bloom removed)</b> (1-orange red, 2-pink red, 3-red, 4-purple red, 5-brown red)					
38	(*)	<b>Fruit: intensity of over colour</b> (3-light, 5-medium, 7-dark)					
39	(*)	<b>Fruit: pattern of over colour</b> (1-only solid flush, 2-solid flush with weakly defined stripes, 3-solid flush with strongly defined stripes, 4-weakly defined flush with strongly defined stripes, 5-only stripes (no flush), 6-flushed and mottled, 7-flushed, striped and mottled)					
40	(*)	<b>Fruit: width of stripes</b> (3-narrow, 5-medium, 7-broad)					
41	(*)	<b>Fruit: area of russet around stalk attachment</b> (1-absent or small, 2-medium, 3-large)					
42		<b>Fruit: area of russet on cheeks</b> (1-absent or small, 2-medium, 3-large)					
43	(*)	<b>Fruit: area of russet around eye basin</b> (1-absent or small, 2-medium, 3-large)					
44		<b>Fruit: number of lenticels</b> (3-few, 5-medium, 7-many)					
45		<b>Fruit: size of lenticels</b> (3-small, 5-medium, 7-large)					
46	(*)	<b>Fruit: length of stalk (cm)</b>					
		mean					
		range					
		standard deviation					
		number measured					
47	(*)	<b>Fruit: thickness of stalk</b> (3-thin, 5-medium, 7-thick)					
48	(*) (+)	<b>Fruit: depth of stalk cavity</b> (3-shallow, 5-medium, 7-deep)					
49	(*) (+)	<b>Fruit: width of stalk cavity</b> (3-narrow, 5-medium, 7-broad)					
50	(*) (+)	<b>Fruit: depth of eye basin</b> (3-shallow, 5-medium, 7-deep)					

LEGEND: (\*) compulsory characteristic which must always be completed  
(+) see illustration or clarification in section G for additional information

Characteristic			CV	RV1	RV2	RV3	RV4
51	(*) (+)	<b>Fruit: width of eye basin</b> (3-narrow, 5-medium, 7-broad)					
<p><b>For characteristic 52, the firmness of the flesh may be measured using a penetrometer.</b></p>							
52	(*)	<b>Fruit: firmness of flesh</b> (1-very soft, 3-soft, 5-medium, 7-firm, 9-very firm)					
53	(*)	<b>Fruit: colour of flesh</b> (1-white, 2-cream, 3-yellowish, 4-greenish, 5-pinkish, 6-reddish)					
54	(*) (+)	<b>Fruit: aperture of locules (in transverse section)</b> (1-closed or slightly open, 2-moderately open, 3-fully open)					
<p><b>For characteristic 55, time of beginning of flowering should be observed when 10% of the flowers are fully open.</b></p>							
55	(*)	<b>Time of beginning of flowering</b> (1-very early, 3-early, 5-mid-season, 7-late, 9-very late)					
<p><b>For characteristic 56, time of harvest is the optimum time of picking to achieve fruit in peak condition for eating.</b></p>							
56		<b>Time for harvest</b> (1-very early, 3-early, 5-mid-season, 7-late, 9-very late)					
<p><b>For characteristic 57, time of eating maturity is the period when a fruit has reached optimum colour, firmness, texture, aroma and flavour for consumption. Depending on the type of fruit, this period can occur directly after removal from the tree (e.g. early varieties) or after a period of storage or conditioning (e.g. later varieties).</b></p>							
57	(*)	<b>Time of eating maturity</b> (1-very early, 2-very early to early, 3-early, 4-early to mid-season, 5-mid-season, 6-mid-season to late, 7-late, 9-late to very late, 9-very late)					

<b>Other Information</b>		
<b>58</b>	(*)	<p><b>Describe any additional characteristics which distinguish the candidate variety from the reference variety (ies) (e.g. other morphological differences, resistance to pests, resistance to disease, etc.)</b></p>
<b>59</b>	(*)	<p><b>Describe any off-types and/or variants observed during the multiplication of this variety. The frequency of occurrence of off-types and/or variants should be noted.</b></p>

<b>Other Information</b>		
<b>60</b>	(*)	<p><b>Indicate which characteristics (including the characteristic number) from this form are most useful in distinguishing the candidate variety from the reference variety (ies).</b></p>
<b>61</b>		<p><b>List any additional comments.</b></p>



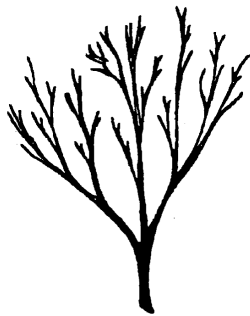
## H. EXPLANATIONS AND ILLUSTRATIONS

**Characteristic 2: Tree: type**

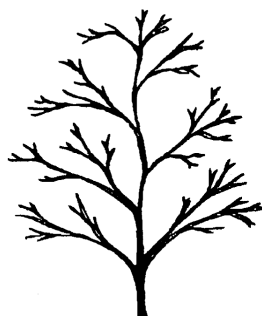
**Columnar:** A compact spur-type tree form with virtually no side branches. Closely spaced, short fruiting spurs are produced along the main stem.

**Ramified:** A form where trees have well developed branches.

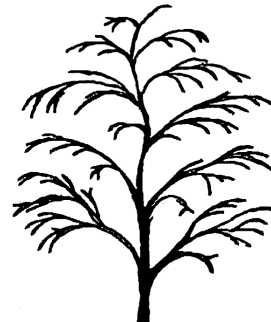
**Characteristic 3: Only varieties with ramified tree type: Tree: habit**



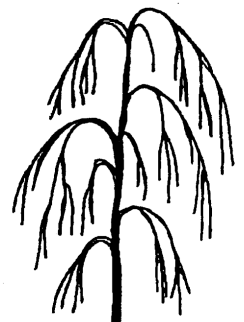
1  
upright



2  
spreading

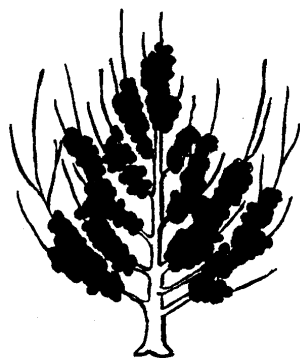


3  
drooping

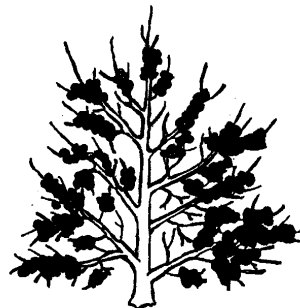


4  
weeping

**Characteristic 4: Tree: type of bearing**



1  
on spurs only

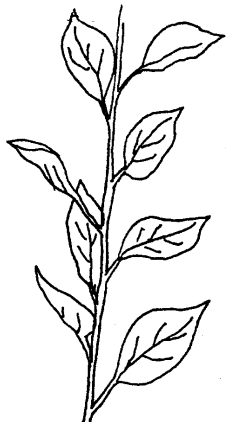


2  
on spurs and long shoots



3  
on long shoots only

**Characteristic 10: Leaf blade: attitude in relation to shoot**



1  
upwards



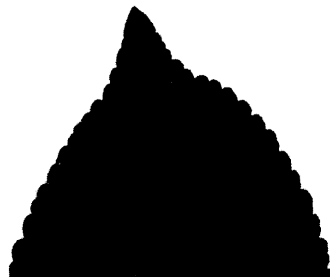
2  
outwards



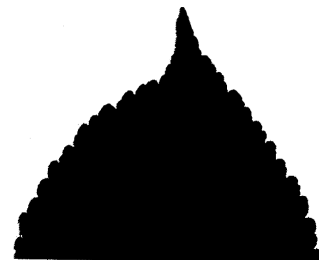
3  
downwards

**Characteristic 15: Leaf blade: incisions of margin (upper half)**

The predominant type of incision should be observed.



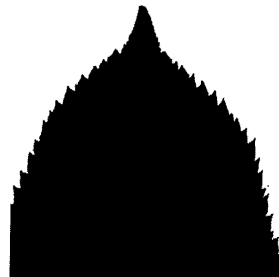
1  
crenate



2  
bicrenate



3  
serrate type one

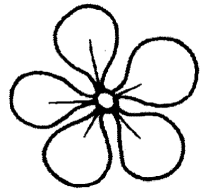


4  
serrate type two

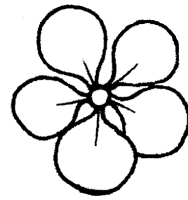


5  
biserrate

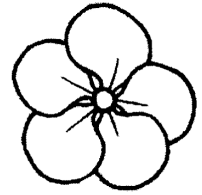
**Characteristic 21: Flower: arrangement of petals**



1  
free



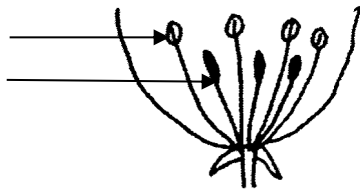
2  
intermediate



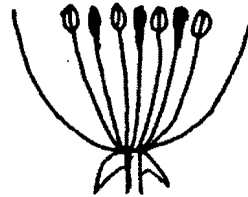
3  
overlapping

**Characteristic 22: Flower: position of stigmas relative to anthers**

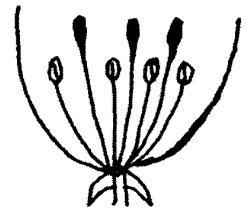
anther  
stigma



1  
below



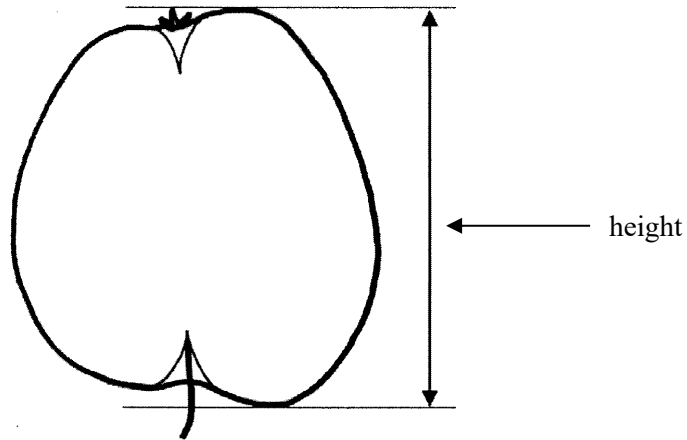
2  
same level



3  
above

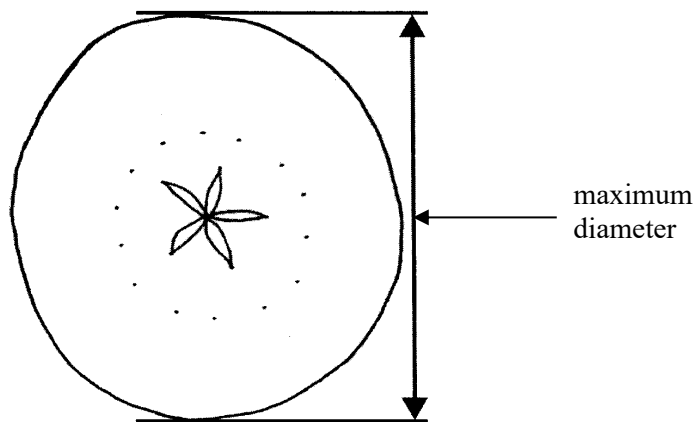
**Characteristic 25: Fruit: height**

The maximum height should be observed.

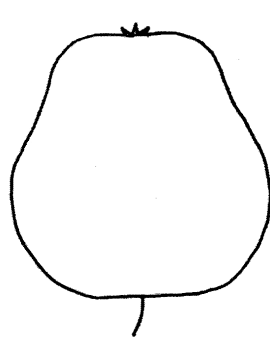


**Characteristic 26: Fruit: diameter**

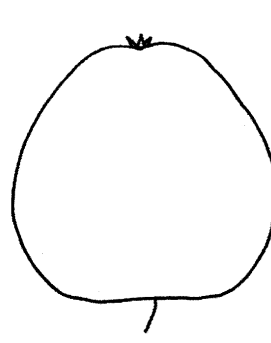
The maximum diameter should be observed.



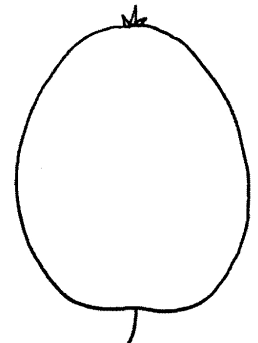
Characteristic 28: Fruit: general shape



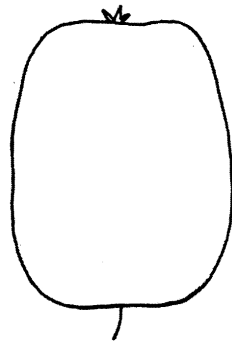
1  
cylindrical waisted



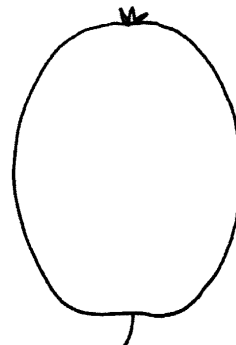
2  
conic



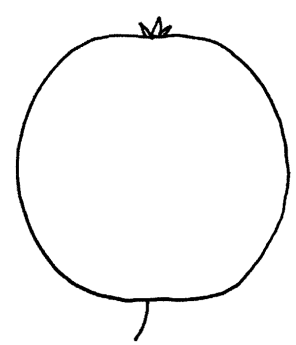
3  
ovoid



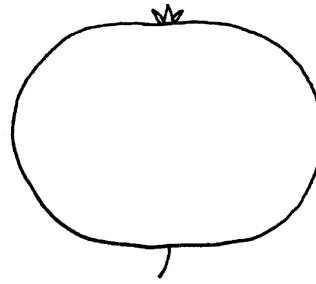
4  
cylindrical



5  
ellipsoid



6  
globose



7  
obloid

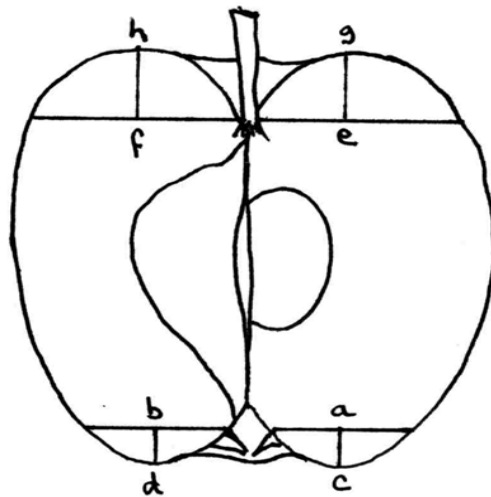
**Characteristic 48-51: Fruit: depth and width of stalk cavity; depth and width of eye basin**

Fruits should be cut through the central axis as accurately as possible. Stalk cavity and eye basin depth and width should be measured from the sectioned fruits. The following diagram indicates the position of lines scored, using a knife or scalpel, on the fruit prior to measuring these characteristics.

- X The lines a-b and e-f must be at right angles to the axis of the fruit.
- X The line a-b is marked at the base of the sepals.
- X The line e-f is marked at the insertion of the stalk.
- X The lines a-c and b-d indicate eye basin depth. They are drawn at right angles to the line a-b to the point where the basin curve levels out.
- X The lines e-f and f-h indicate stalk cavity depth. They are drawn at right angles to the line e-f to the point where the stalk cavity levels out.
- X In the case of asymmetric or irregular sections, the larger side should be considered.

f-h = depth of stalk cavity  
(characteristic 48)

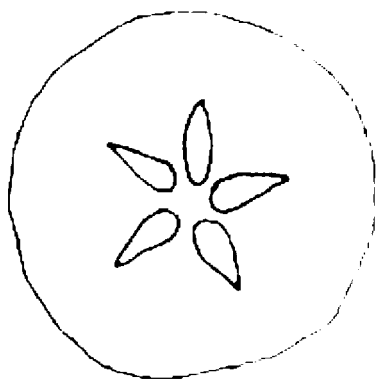
e-f = width of stalk cavity  
(characteristic 49)



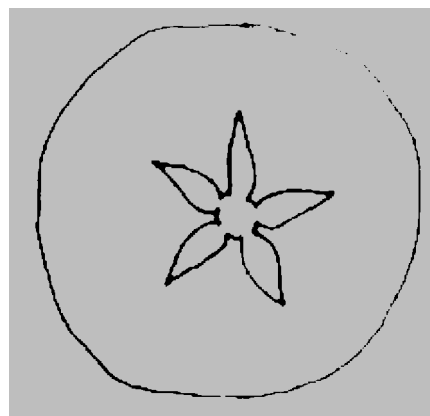
a-b = width of eye basin  
(characteristic 51)

a-c = depth of eye basin  
(characteristic 50)

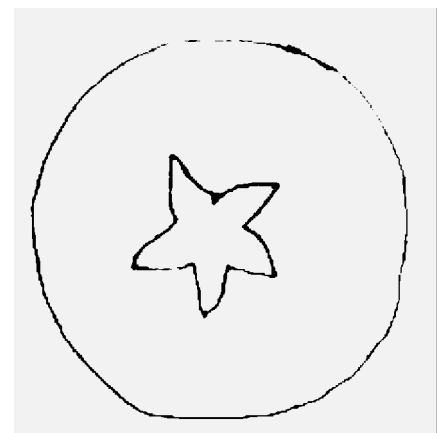
**Characteristic 54: Fruit: aperture of locules (in transverse section)**



1  
closed or slightly open



2  
moderately open



3  
fully open