Phenotypic characterization kit II

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Contents and type of traits:

General data about the phenotyping assay

- Number of plants per accession
- Field name
- Country
- Cultivation type (Open field/Greenhouse/Mesh tunnel)
- Temperature °C (mean, maximum, minimum values over the cropping period)
- Relative humidity (mean, maximum, minimum values over the cropping period)
- Watering (flooding, local irrigation)
- Surface (m2)
- Number of days of culture (from planting to the end of the crop)
- Type of soil (clay, fine silt, silt, total sand)
- Fertilization (indicate the overall splitting of macronutrients: kg ha^{-1} of N, P_2O_5 and K_2O)
- Mode of cultivation (brief description)

- Plant pruning (yes/no)
- Number of stem (by plant)
- Total irrigation volume (m3/ha)
- Rainfall (m3/ha)
- Daily global solar radiation
- Reference crop evapotranspiration (Mean Eto)
- Reference crop evapotranspiration (Total Eto)
- UTM coordinates
- Take note of the accession with low or lacking viability

A Morphological characterization. **Qualitative traits**

1. Growth habit (1, determinate/ 2, semideterminate/ 3, indeterminate).

2. Foliage density (1, very sparse/ 3, sparse/ 5, intermediate/ 7, dense).

3. Inflorescence (1, uniparous/ 2, fishbone/ 3, forked/ / 4, compound/ 5, irregular).

4. Leafy inflorescence (1, not leafy/ 2, leafy/ 3, leafy with shoots).

5. Style position (1. inserted/ 2. same level as stamen/ 3. slightly exserted/ 4. highly exserted). Take it three times (early, medium, late trusses).

6. Green shoulder (1, uniform/ 2, light green/ 3, medium green/ 4, dark green).

7. External fruit color (1, yellow/ 2, orange/ 3, pink/ 4, red/ 5, purple/ 6, brown/ 7, green). Check that pink coloured fruits have colourless skin.

8. Fruit predominant shape (1, flat/ 2, rectangular/ 3, elipsoid/ 4, obovoid/ 5, round/ 6, oxheart/ 7, long/ 8, heart/9, bell pepper).

9. Fruit set sequence (1, very poor/ 3, poor/ 5, intermediate/ 7, good/ 9, very good).

10. Fruit size homogeneity (1, extremely variable/ 3, low/ 5, intermediate/ 7, high).

11. Irregular transversal section (1, round/ 2, angular/ 3, irregular/ 4, highly irregular).

12. Easiness of fruit to detach from the pedicel (1, very easy/ 3, easy/ 5, intermediate/7, difficult).

13. Blossom end scar condition (1, open/ 2, closed/ 3, both).

14. Ribbing at calyx end(1, very weak/ 2, weak/3, intermediate/4, strong).

15. Macrocalix (1, absence/ 2, presence).

A Morphological characterization. **Quantitative traits**

1. Traits for fruit characterization:
    1. Fruit firmness (measured with a penetrometer (Type used in Valencia: Fruit pressure tester mod. FT 0.11 Lbs.) (Descriptor optional). Measured in 6 fruits. Two opposite measures per fruit in the equatorial part of the fruits.
    2. Colour (L,a,b parameters) Measured in Valencia with Chroma Meter CR-400/410. Konica Minolta (Descriptor optional). Measured, at the green shoulder (breakers stage) and outside the green shoulder (at the midpoint of the fruit length) at the red ripe stage, in 6 fruits per accession (3 measurements per fruit).

B Agronomic characterization. **Qualitative traits**

1. Puffiness appearance (1, absent/ 3, slight/ 5, intermediate/ 7, severe).

2. Presence and incidence of radial cracking (1, absent/ 3, scarced; less than 5% of fruits affected/ 5, intermediate; between 5% and 20%/ 7, abundant; more than 20%).

3. Presence and incidence of concentric cracking (1, absent/ 3, scarced; less than 5% of fruits affected/ 5, intermediate; between 5% and 20%/ 7, abundant; more than 20%).

4. Fruit fasciation (1, absent/ 3, slight; less than 5% of fruits affected/ 5, intermediate; between 5% and 20%/ 7, severe; more than 20%).

5. Blossom-end rot (1, absent/ 3, slight; less than 5% of fruits affected/ 5, intermediate; between 5% and 20%/ 7, severe; more than 20%).

B Agronomic characterization. **Quantitative traits**

1. Flowering earliness. Number of days to from sowing until 50% of plants have at least one open flower in a uniform growing environment.

2. Ripening earliness. Number of days from sowing until 50% of plants have at least one ripe fruit.

3. Ripening uniformity of the whole plot. For this descriptor two measures have to be taken:

- Number of days from sowing until all plants have at list one ripe fruit (M1).

- Number of days from sowing until the first plant has a ripe fruit (M2).

The descriptor will be calculated as M1 – M2.

4. Yield. Measured in three plants per accession, chosen randomly within the plot. Yield will be estimated from quantitative data obtained on trusses 1 to 4. In these trusses count and weight all the fruits in a per plant basis (separate commercial to non-commercial fruits). On the remaining trusses (>5th) count the number of fruits and estimate the yield using the mean fruit weight calculated on trusses 1 to 4. For plants conducted on one stem and pruned, the main stem should be cutted at approximately 2 m height. In Long Shelf Life accessions total yield (all trusses) will be evaluated.

- Commercial yield on trusses 1st to 4th (kg/plant) (fruits without cracking, blossom-end rot, small size and fasciated fruits).

- Non-commercial yield on trusses 1st to 4th (fruits with cracking, blossom-end rot or small size) (kg/plant). Taking in account that in some traditional varieties fruit shape could include a certain degree of variability, fasciated fruits could be admitted as commercial (variety 'Valenciana'). Besides, some traditional varieties have predominantly this type of fruits (accession TRVA3030 'Fuenterrobles' (Phenotyping kit 2 images). So these fruits must be counted as commercial yield.

- Number of commercial and non-commercial fruits on trusses 1st to 4th.

- Total number of fruits per plant.

- Mean fruit weight (g) = commercial + non-commercial yield/total number of fruits on trusses 1st to 4th.

- Total yield (commercial + non-commercial, kg/plant): calculate using the following formulae: mean fruit weight*total number of fruits.

Samples for metabolic profiling

Froze 2 biological replicates per accession (each replicate coming from different groups of plants). Harvest the fruits at the Red Ripe stage (RR). Each replicate should be formed by at least 6 fruits. For big fruits take a slice of 1/8 of the fruit; for small fruits take half of the fruit. Discard locular content and froze, using nitrogen liquid, solely the pericarp. Store the samples at -80°C. Sample all the accessions during the same period to minimize environmental variation (during one week if possible).

Pictures

Take pictures of plant, inflorescences, fruits and unusual characteristics of each accession as indicated in pictures (Annex images). Add always a ruler for size reference.

Heterogeneous accessions

If the accession is clearly a mixture of two or more types separate the seeds from selfing and characterize the two or more types. Label the segregants using the last number of the new TRADITOM codification.

Observations.

Write down unusual characteristics no considered in the descriptors used.

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