Potential and limitations of standardized descriptors for the genus *Abelmoschus* (okra)

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Summary

The limitations of a set of standardized descriptors are becoming apparent during trials for the following reasons:

- there is monomorphism within a specific species
- there are descriptors which are not planned but interesting
- descriptor states are lacking
- the absence of guidelines for selecting a subsample of descriptors

In this paper we make comments and proposals (on the basis of the existing set of descriptors) that should allow a set of descriptors to be elaborated in the future that is less constraining and better suited to the purpose of describing existing diversity and its organization.

Introduction

The management of crop genetic resources is not limited to the storage of seeds only. In such cases, collections become quickly unusable, thus it is essential to describe, as best as possible, samples to implement a reliable information basis for their effective use. We will try to issue comments on a general level, but many remarks will of course be linked with observations from West Africa.

Okra is an annual, herbaceous plant. The monoic flowers are self-compatible. The rate of allogamy differs according to varieties and ecological conditions (Martin, 1983; Hamon et Koechlin, submitted a,b).

Seeds are kept in fruits for cultivated forms with the exception of those from very wet, forest zones. These fruits, when collected, are sorted individually into groups of similar fruits or put together in a basket (Hamon and Charrier, 1983). There is no information available on the parents, or even on the mother plant. In principle, one sampling per fruit should be done. The starting set would then include between 50 and 150 half-sister seeds.

Often many fruits are put together in order to reduce the number of samples and to increase the number of seeds. This practice induces irreversible mistakes such as the mixture of species or varieties.

In this paper we will, on the basis of our experience (Hamon, 1988; Hamon *et al*, in press) analyze critically the list of descriptors which was proposed by Charrier (1984) and used as a reference by IBPGR. We will underline the reliability of some of those descriptors, the limits of others and the unusefulness of some of them in specific conditions.

Comments and proposals (descriptors)

1. *The key steps of characterization*

The characterization of samples or the evaluation of their diversity includes many key steps; these are mainly:

1.1 "Passport data". Data linked to the origin of the germplasm

In the past these data were often missing (Peeters and Williams, 1984). Data about the country of origin, collector's number, geographical coordinates and sometimes ecological data of the collecting sites are now available for recently collected accessions, but information on their role in local agricultural practices is often lacking. But excess of information at different levels (store, backyard primitive cultivar, advanced cultivar soils type) is often difficult to interpret.

1.2 The description of the sample at the collecting stage

This is the most important step. Inaccurate or incomplete information will have irreversible consequences.

The description of an okra specialist may be quite different from that of a generalist who does not know the diversity of the genus. At a simple level, confusing *A. esculentus* with *A. caillei* is common for at least three reasons: 1) *A. caillei* is not yet well known; 2) African farmers consider *A. caillei* as a late type of *A. esculentus*; 3) there are no traits of classical botany which can be applied to the pods (Stevels, 1988).

2. *Identification of the accession*

For easier reading, we have not respected the order of descriptors in Charrier (1984); however, we will systematically refer to its numbering.

2.1 (D 1.1.1) Accession number

The accession number is unique and should also be employed during evaluation. Other passport data do not need to be referred to, but the corresponding file will be consulted, if necessary. However, the geographical coordinates should be included, as they will serve as a variable for analysis.

2.2 Coordinates of collection sites

- (D 2.2.4.) Country of origin

  The international code proposed by IBPGR (1976) is used

- Geographical coordinates of the collecting site

  (D 2.2.7.) Latitude, (D 2.2.8.) Longitude

2.3 (D 1.5) Scientific name

The name of the species as provided by the collector and included in the passport data should be distinguished from the identification of the evaluator. Experience shows that the co-existence of two cultivated species in Africa is an important source of mistakes; similarly, the taxonomy within *A. moschatus* and *A. manihot* is not yet very clear.

Proposal

1) *A. esculentus*, 2) *A. caillei*, 3) Natural hybrid between *A. esculentus* and *A. caillei*, 4) Mixture of *A. esculentus* and *A. caillei* in the same sample, 5) *H. sabdariffa* (often included as samples), 6) *H. cannabinus* (often included as samples), 7) *A. moschatus*, 8) *A. manihot*, 9) Unknown, 10) *A. ficulneus*, 11) and 12) and following numbers: not yet designated.

N.B. Fruit shape, seed striation and zymograms are very reliable identifiers.

2.4. The homogeneity of the accession

The homogeneity of the sample is commonly accepted as a basic starting point. This is often not the case, notably in Africa, and this should be noted as soon as the evaluation allows. The level of heterogeneity can be linked with the status of the sample, but it is quite often the case that many characters segregate. It is up to the evaluator to fix limits in a global manner (see below) or for some characters (refer further for each character).

1) Yes (variety of fixed type), 2) No (heterogeneity but same species), 3) No (heterogeneity with two different species), 4) No (two different species plus hybrids).

3. *Quantitative characters linked with growth*

3.1 General remarks

A certain number of rules should be respected for the characterization to be useful. These are:

- describe accurately the site, including data on sowing period and pluviometry;
- use well-known standards, at least two (or three) for each species;
- carry out evaluation separately for each species but include in each evaluation the standards from other species in order to create a reference basis;
- indicate on how many plants the observations were made.

Around 30 plants should be enough, in the first evaluation phase, to provide a good picture. If the sample is obviously not a fixed form or an interfecondation, there are no real solutions except to create subsamples and purify them later on.

3.2 Variables for vigour and growth

Three variables are retained. These are:

D 6.1.2. Maximum plant height (cm)
D 6.1.9. Maximum numbers of internodes
D 6.1.3. Stem diameter at the base (mm)

These three descriptors will cause problems if the conditions of observation are not precisely defined. Indeed:

1) the diversity of growth cycles within the same species is important. Okra growth is continuous until death. Thus, maximal value should be estimated on dead plants. This implies numerous visits and risks of errors;

2) it is more difficult to compare *A. esculentus* with *A. caillei* in West Africa because the former ends it cycle when the latter starts to flower; wild species have cycles of undetermined duration and are more or less perennials.

In conclusion, there is therefore a need to identify the species of the sample quickly and to proceed accordingly with separate trials.

The best time to observe *A. esculentus* is around 80 to 90 days after sowing. At this time, most *A. caillei* plants will still be growing. The optimal time for this species, which has slower growth, is around 100-110 days. Finally, the speed of growth of wild species is much slower. There is therefore a need to be extremely careful when making these observations.

3.3 Variables for habit and plant structure

Plant structure and habit are characters which are taking different modalities. With *A. esculentus* there is no real problem, because there are only a few branches, which can be clearly seen at the base. They are easy to count and their length can be measured. There are more difficulties with *A. caillei* whose structure is much more branched with eventually secondary branches. The same occurs with most of *A. manihot;* it becomes nearly impossible to observe number and length of branches of *A. moschatus*, which is bushy.

These characters are nevertheless interesting because, beyond a simple description, they serve as reference parameters for the yield. Indeed, the fruit yield on the stem is little influenced by the production on branches, which for *A. caillei* and wild species is often of greater importance.

Proposals

- Plant habit

  1) Unique orthotrop axis, 2) Dense branching at the base followed by an orthotrop axis without branches, 3) Base without branching but densely branched apex, 4) Densely branched all over the plant

- Length of the branches

  The number of branches can be counted but the length should be estimated, because of the time taken for measurement, in the case of many accessions

  0) No branch, 1) Rarely more than 10 cm, 2) Frequently more than 10 cm.

4. *Inflorescence and fruit*

4.1 General remarks

The flowering of okra starts with the emergence of a first flower which may be followed, depending on the species, by a new flower every day, or a minimum of one new flower per week, continuing until the end of the cycle.

A flower opens during the morning, closes in the afternoon and the corolla will fall the following morning. The young fruit will grow very quickly and can reach, for *A. esculentus*, several centimeters in a few days.

The observation of flowering characters requires a lot of work if results are to be precise (in terms of days). Observations of terms of weeks after flowering will reduce the workload.

The observation of pods, which, unless exceptions, are homogeneous, is important. Observations should be done on fruits which are representative and not on the ones which are obviously misshaped.

The actual codification is too restrictive. The diversity observed in Africa shows that more importance should be given to traits which are used in breeding or at least for variety identification. Measurements should be done on completely mature and dried fruits.

4.2 Flowering and fruit earliness

Four descriptors are retained to assess flowering and fruit earliness; these are:

D 6.2.1. First flowering
D 6.2.2. First flowering node
D 6.2.3. First fruit-producing node
D 6.2.X. Flowering span

N.B. 999 is noted if no flowering occurs

The flowering span is a new descriptor which could be interesting in that it provides an index of grouped flowering. This can be difficult to assess, because it requires daily visits, but its observation can be simplified by a weekly registration of the production of young fruits. The observation of the first 10-15 plants per accession is possible.

4.3 Fruit shape

Five descriptors are selected as follows:

D 4.2.8. Fruit length at maturity
D 4.2.X. Fruit width
D 4.2.11 Number of ridges per fruit
D 4.2.6. Position of fruits on the main stem
D 6.3.1. Weight of 1000 seeds

N.B. The number of ridges per fruit and their position on the main stem are useful only for cultivated forms and in this case require the selection of descriptor states. The number of ridges is always five for wild forms. This descriptor is an exception to the quantitative continuity, as the number of ridges is either zero if these are not well marked, or it varies between 5 and 12. There are nearly always variations on the same plant, thus assessment is difficult and the formation of classes should be preferred.

Proposed descriptors

D 4.2.11. Number of ridges per fruit (simple genetic character with intermediate heterozygotes)

1) Smooth fruits or ridges unmarked until the base, 2) 5 ridges, 3) Frequently more than 5 but less than 9, 4) Frequently more than 9

D 4.2.6. Position of fruit on main stem

Homogeneity: 1) Erect, 2) Intermediate, 3) Horizontal, 4) Slightly falling, 5) Totally falling. *Heterogeneity*: 6) Presence of 1 and 2, 7) Presence of 2 and 3, 8) Presence of 3 and 4, 9) Presence of 4 and 5

5. *Colouring of diverse plant organs*

5.1 General remarks

Colour traits are not easy to assess due to uncertainties with colour variations, even in a homogeneous accession. This applies not only to green and red variations, but also to all other colour characters, except for the internal flower spot. The superimposition of one colour upon another is common with okra, the green or red background being influenced by other colours. The ideal would be to print a scale of colour references on paper in order to avoid observations being influenced by the previous accessions. There is also a need for staff with a very reliable sense of colour and its gradations. Thus, except for clear cases, there is a need to be prudent and to do two evaluations. It should not be forgotten that, due to the complexity of the genetic base of the pigmentation (Mehetre *et al.*, 1980), it is rare to find a totally homogeneous accession. Accessions should be noted as heterogeneous when differences are marked beyond the level of gradations.

5.2 Colouring of vegetative organs

4 characters are selected. These are:

D 4.1.4. Stem colour
D 4.1.6. Lamina colour
D 4.1.X Leafrib colour
D 4.1.Y Petiole colour

- D 4.1.4. Stem colour

  Homogeneity: 1) Green (unless special cases, gradations will not be noted), 2) Green with clearly noticeable red spurs, 3) Anthocyanic (red or purple) even if there are some green spurs. *Heterogeneity*: 4) Rather red, 5) Rather green, 6) Complete segregation

- Leaf colour

  Leaf colour is assessed from that of lamina and ribs. Lamina is of green colour, but eventually, in some cases (plants with slightly anthocyanic stems) red spots may appear on aged leaves. It is better not to note this, as the stage of appearance of this phenomenon is difficult to assess. Only important red spots, which are rare, should be noted.

  For the ribs we can distinguish the following sections:

  - the point of convergence of the ribs (foliar spots); the basal half of the ribs; the distal half of the ribs

  Quantification is very difficult. It is preferable to limit ourselves to the presence of anthocyanic pigmentation on basal and/or the distal half.

- D 4.1.6. Lamina colour

  1) Green, 2) Green with some red veins, 3) Green with important red spots

- D 4.1.X. Rib colour

  Homogeneity: 1) Totally green, 2) Red at the ribs' joining point, thereafter totally green, 3) Green with some red spots widespread on the lower half of the ribs, 4) Green but with plenty of red spots all along the rib, 5) Ribs nearly totally red. *Heterogeneity*: 6) Heterogeneity (1+2), 7) Heterogeneity (2+3), 8) Heterogeneity (2+4 or 5), 9) Complete segregation

- D 4.1.Y. Petiole colour

  1) Green, 2) Red above but green below, 3) Red on both sides

5.3 Flower and pod colour

Three descriptors are selected. These are:

D 4.2.5. Petal blotch
D 4.2.7. Fruit colour
D 4.2.X. Colour of the darkest ridges

- D 4.2.5. Petal blotch

  Okra flowers all have a red petal blotch in the centre of the flower. Nevertheless, the blotch on the external face may be absent. This character is simple monogenic; we will therefore note the three following cases:

  Homogeneity: 1) Internal, 2) External. *Heterogeneity*: 3) Segregating

- D 4.2.7. Fruit colour

  A large scale of colours and gradations is involved in addition to the usual difficulties associated with the observation of colour markers. This is particularly true when red and green are together or when diverse nuances of green must be described. The diversity of fruit colour reaches its maximum in West Africa.

  It is important, before starting the notations, to become acquainted with the descriptor states. It would certainly be most useful to build a file of colour references.

  N.B. The numeration follows its gradual implementation, hence its apparent disorder. Reordering of the descriptor states could be useful.

  1) Whitish green to white, 2) Common green, 3) Green background plus more or less red spots, 4) White background plus more or less red spots, 5) Red, 6) Green towards black, 7) Light green but not white (1+2), 8) Mixture of (2+3), 9) Violet to purple, 10) Mixture of (2+6), 11) Dark green but not black, 12) Mixture of (3+5), 13) Mixture of (6+5), 14) Water green (characteristic of Sudan), 15) Pink, 16) Mixture of (3+6)

- D 4.2.X. Darker colour of fruit ridges and spines on fruits and seed hairiness

  1) No hairs, 2) Slightly hairy, 3) Very hairy, 4) Slightly spiny, 5) Numerous spines, 6) Spines on fruits and seed hairiness, 7) Seed hairiness but no spiny fruits

6. *Electrophoretic markers*

The use of enzymatic diversity for varietal identification has been used for many crops. With regard to okra, the low level of polymorphism (Hamon, 1988) shows that this technique, using classical systems, only allows (with a few exceptions) the two cultivated species *A. esculentus* and *A. caillei* to be distinguished.

For wild species we only have data for *A. moschatus* and *A. manihot* (Hamon, 1989). These results show, however, that the level of diversity for these species is clearly superior and that these markers could be useful.

7. *The descriptors at a species level*

We have not examined, in this paper, descriptors that are used at the species level, i.e. those which do not vary within a given species.

The example of zymograms, as outlined above, is a particular case. These observations will be made at the stage of species identification (refer to para 2.3) and could eventually be included within passport data.

8. *Non-independent descriptors*

We cannot develop the problems linked with non-independent descriptors, some aspects of which have been treated in other papers (Hamon, 1988); nevertheless, knowledge of these may allow substantial time-savings when making observations.

Descriptors are often presented as autonomous elements and this is inaccurate. Indeed, numerous quantitative descriptors are correlated with each other because their purpose is to:

- assess, in a different way, the same biological phenomenon;
- assess different aspects of the same organ.

Similarly for the qualitative variables, information on one may allow, with good probability, the state of another descriptor to be guessed at; this applies for many colour descriptors.

Discussion

We have presented in this paper the potential and limits of a set of standardized descriptors for the genus *Abelmoschus*. Our work is based on the initial list proposed by Charrier (1984), which was not considered exhaustively, but the most used and most easily assessable descriptors in that list were picked out.

We have proceeded by regrouping descriptors which can be measured or observed simultaneously. Their number is reduced and in some cases this number could be further reduced when some quantitative descriptors are correlated or when qualitative ones are not independent.

The most important recommendations we would like to make are the following:

- make sure species are correctly identified before being characterized (this can be done at the time of collecting or performed on the seeds with the help of zymograms);
- compare only what can be compared, i.e. distinguish in time and in the field the characterization of each species. Be careful not to plan so many observations that they become unmanageable;
- select in respect of the evaluation site some standards (two very different per species) which will be systematically included in all trials. This will allow more reliable comparisons;
- check the genetic homogeneity of the sample: autogamy and varietal fixation are far from being the rule. About ten artificial selfings should be performed in each sample: this means about 1000 seeds.

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