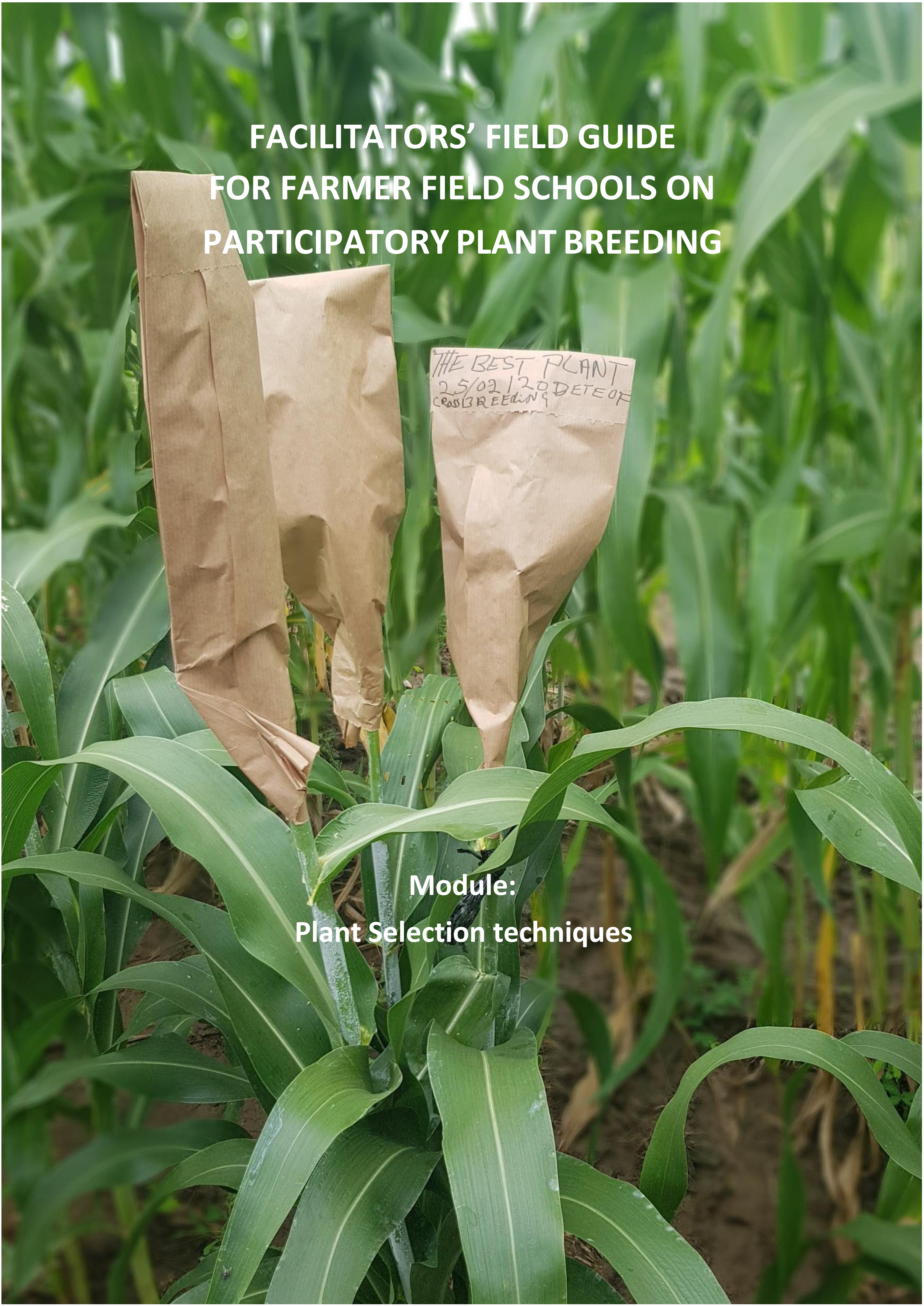
# FACILITATORS' FIELD GUIDE FOR FARMER FIELD SCHOOLS ON PARTICIPATORY PLANT BREEDING




## Module:

## Plant Selection techniques

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## Module: Selection techniques

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All illustrations by Irene Cécile (www.irenececile.com).

**Cover photo:** Zambia FFS selection of preferred sorghum plants

# Introduction

This module is part of the field guide for facilitators of Farmer Field Schools doing participatory plant breeding.

The module supplements other field guide modules that cover preparation and implementation of participatory plant breeding. It is more technical than the others and gives guidelines for one of the most important techniques available to a plant breeder: *the selection of plants from diverse plant populations*, whether existing landraces or varieties, or novel breeding populations. The focus is on using selection to enhance existing varieties or create new ones through 'Participatory Variety Enhancement' and 'Participatory Variety Development'. Additionally, it provides guidance on sourcing seeds for 'Participatory Variety Selection'. This poster explains the choice between these breeding methods.

It is impossible to give a simple 'cook-book' set of plant selection instructions applicable to all conditions, crops, reproductive systems, breeding objectives and capacities. This document gives possible trajectories--a description of options and possibilities, rather than a prescription writ in stone.

The techniques described are tailored to the conditions under which participatory plant breeding is carried out: with limited land and time, in farmers' fields and by farmers' hands.

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*Preparing the land for a PPB trial in a Zambian Farmer Field School*

# Introduction to Participatory Plant Breeding

Plant breeding is the science of changing the qualities, or *traits*, of crops to better suit the needs of the farmers growing them. It involves improving existing crop varieties, but also the creation of new ones, with new combinations of traits, by crossing different parent plants.

Farmers have done plant breeding in their way for approximately 10,000 years, always adapting to changing environments, tastes, cultures and markets. Scientists are newer to the field but have developed powerful techniques to make the breeding process quicker and more precise. Both approaches have their strengths and weaknesses, and in Participatory Plant Breeding (PPB) farmers and scientists combine forces to overcome these weaknesses and create crop varieties that are particularly well-suited to the preferences and growing conditions of smallholder farmers.

In the SD=HS programme, PPB is carried out by Farmer Field Schools (FFS), groups of approximately 25 farmers who get together every week or two to learn about plant breeding by doing it themselves, together with plant breeders. This FFS Starter Kit introduces how FFS work.

### From breeding objectives to selecting plants

PPB starts with a reflection on the changing world of the community: how have agroecosystems, culture, markets, infrastructure and climate changed, and what are the consequences for their crops? *How do crops need to change* to best respond to these external changes?

Defining the changes farmers wish to see in their crops is a collective decision-making process that needs to happen very carefully. After all, the decision will determine the work for years to come, and the participants of the FFS will remain motivated only if they feel the outcome of the work will benefit them – women, men, youngsters, poor and wealthier farmers alike.

The process of collectively reflecting on changes, identifying important traits and prioritizing them into breeding objectives is described in the Farmer Field School Diagnostic Stage module.

See also p. 34 for a discussion on how to address multiple traits in a breeding program.

In plant breeding, *changes are expressed in terms of traits*. Traits that can be observed, measured and selected. In the field, or, after harvest, in the kitchen: the size or colour of a seed, the number of panicles in a plant, cooking time, tolerance to a specific pest insect or disease. A community may have a wish list of many important traits they wish to see in a variety but will need to prioritize a maximum of three or four if they want their breeding efforts to be successful: there is no such thing as an 'ideal super variety'.

These prioritized traits are called *breeding objectives*. Breeding objectives are a balance between what farmers need and what is biologically possible within the science of plant breeding. Some traits are difficult to combine within the same plant, such as high yield and tolerance to environmental stresses, or better taste and extended shelf life.

Much of the progress towards the breeding objectives set by the FFS is achieved through *selection* among plants that vary for the traits of interest. Only the seed of those plants that match the interests of the FFS is collected and planted next year. The person selecting the plants can be seen as a *gatekeeper*: the stricter the gatekeeper, the stricter the selection of plants and traits that are allowed to pass to the next season. If the gatekeeper is not strict enough, too many plants are allowed to pass and there will be no change or improvement in the traits seen in the field the next season. However, if the gatekeeper is too strict and too few plants are allowed to pass, important traits may be lost, not enough diversity will be left in the field to select from, and the breeding efforts will fail. Finding this balance in selection intensity is an art as much as it is a science.

### Three methods of Participatory Plant Breeding

The SD=HS programme works with three methods of Participatory Plant Breeding. These are not the only methods that exist, but we have found them to work well within the social and pedagogical context of Farmer Field Schools. Which of these methods is chosen is decided by FFS and breeder together (see this poster with a decision-making tree) and depends on the farmers' breeding objectives and the traits already present in existing varieties. For each of the three methods, selection is approached in a different way.

The first, and simplest method is *Participatory Variety Selection* (PVS). PVS compares different varieties side by side, and in different locations, with the aim of choosing the ones best suited to local preferences and growing conditions. PVS does not require selection of individual plants, but because it must be repeated over several seasons, basic guidelines for the sourcing of seed for the next season are included in this guide for completeness.

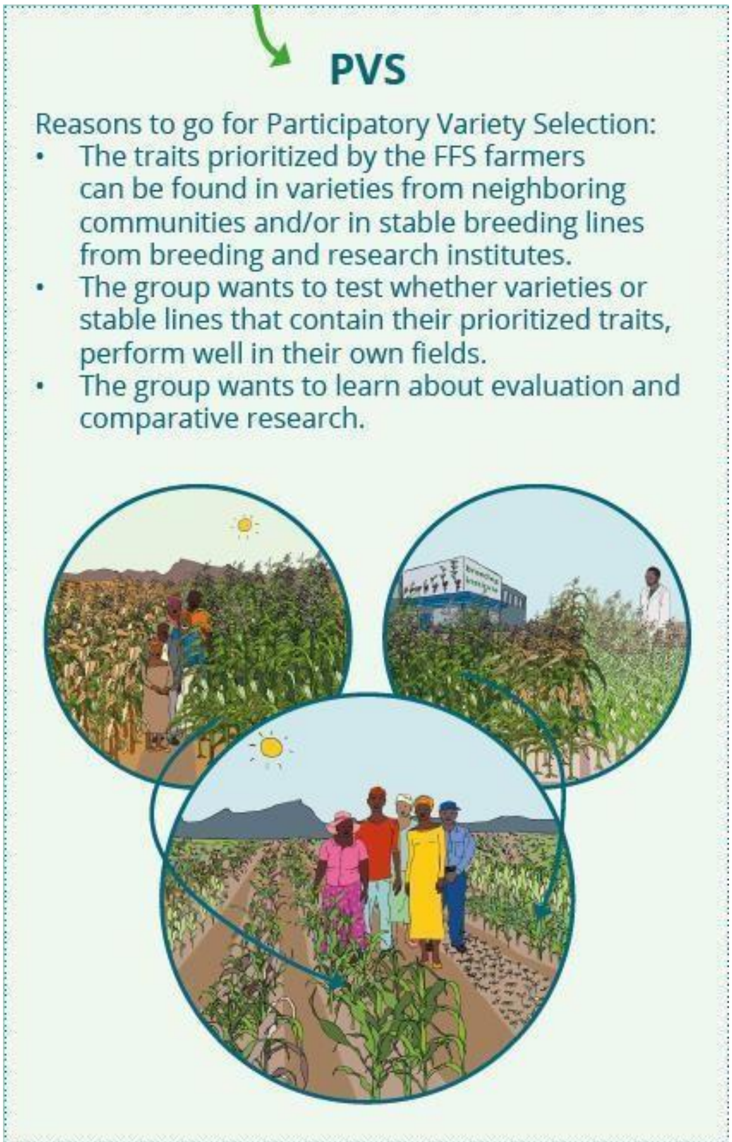
*Participatory Variety Enhancement* (PVE), the adaptation and improvement of existing varieties, and *Participatory Variety Development* (PVD), the creation of new varieties, are the two methods for which plant selection is crucially important. They are described in detail in this guide.

The instructions for selection in this guide are adapted to the context of smallholder farming communities, where land and time are limited. However, many of the methods used in FFS are the same as used by professional plant breeders when they carry out selection at their research stations.

All terms and definitions are explained in the section 'PPB concepts and techniques in more detail' (p. 22).

# I. Sourcing seeds in Participatory Variety Selection (PVS)

*For elaborate instructions on how to set up a PVS study see the [Module on Plot design for Participatory Varietal Selection]*




*Reasons to choose Participatory Variety Selection from poster about deciding PPB method.*

In a PVS study, farmers evaluate different varieties based on their breeding objectives. These can include modern varieties, traditional and farmers' varieties, as well as advanced (stable) breeding lines from breeding institutes. The aim is to observe the varieties and choose those that are desirable, but not to improve them further or change their traits. This means no selection of individual plants is needed in PVS; there is no need for a 'gatekeeper'.

However, since farmers and breeders wish to evaluate varieties under different growing conditions, most PVS studies require two or three seasons to complete. E.g., breeding objectives often include drought tolerance or resistance to a specific disease. If these conditions are absent in the FFS field one year, the study must be repeated a second time. In this case, new seed is required.

It is good practice for collaborating breeding institutes to provide the FFS with new seed of the same varieties each year the study is repeated, but if this does not happen, or if PVS varieties came from other sources (e.g., the market, or a neighbouring community) the FFS will need to secure seed for the next season themselves.

**How should an FFS secure seed to repeat a PVS study, if they are not provided with new seed by a breeding institute?**

The best option is to begin the first PVS year with more seed than is needed for planting and to keep the remaining seed as backup. This is also important in case drought or floods destroy the season's first crop, requiring replanting already in the first year. All remaining seed can be kept for the next season. Ask the collaborating breeder if they can provide more seed than is necessary for a single planting and make sure to store the seed well!

A second option, which is possible only for **self-pollinated crops** and not for cross-pollinated crops, is to save seed for the next season at harvest time. For self-pollinated crops, select seed according to normal farmer's practice, from the best performing plants in the centre of the plot. Avoid collecting seed from the borders. This minimizes the possibility of selecting off-types resulting from undesired cross-pollination: even self-pollinated crops have a 2 to 30% chance of cross-pollinating with other varieties grown nearby! Select as much seed as is required to plant next season's plot (400 seeds per PVS variety is more than enough) but collect at least double that amount to allow for replanting.

For **cross-pollinated crops**, selecting seed for the next season will not yield reliable results, as the harvested seed will be different from the original seed due to 'contamination' (or 'enrichment', depending on your perspective) between varieties grown in the PVS plot or even in nearby farms through cross-pollination. The best option is to go back to the reserve seed, if available, or to obtain seed from the original seed source.

### Completing a PVS study, moving on to seed production

Farmers must see with their own eyes how a variety performs before they will plant it in their fields. They are much too smart to adopt a variety based just on hearsay or the numbers written on a seed package, or even the small plots ('crop cuts') used by breeding institutes to assess a variety's performance.

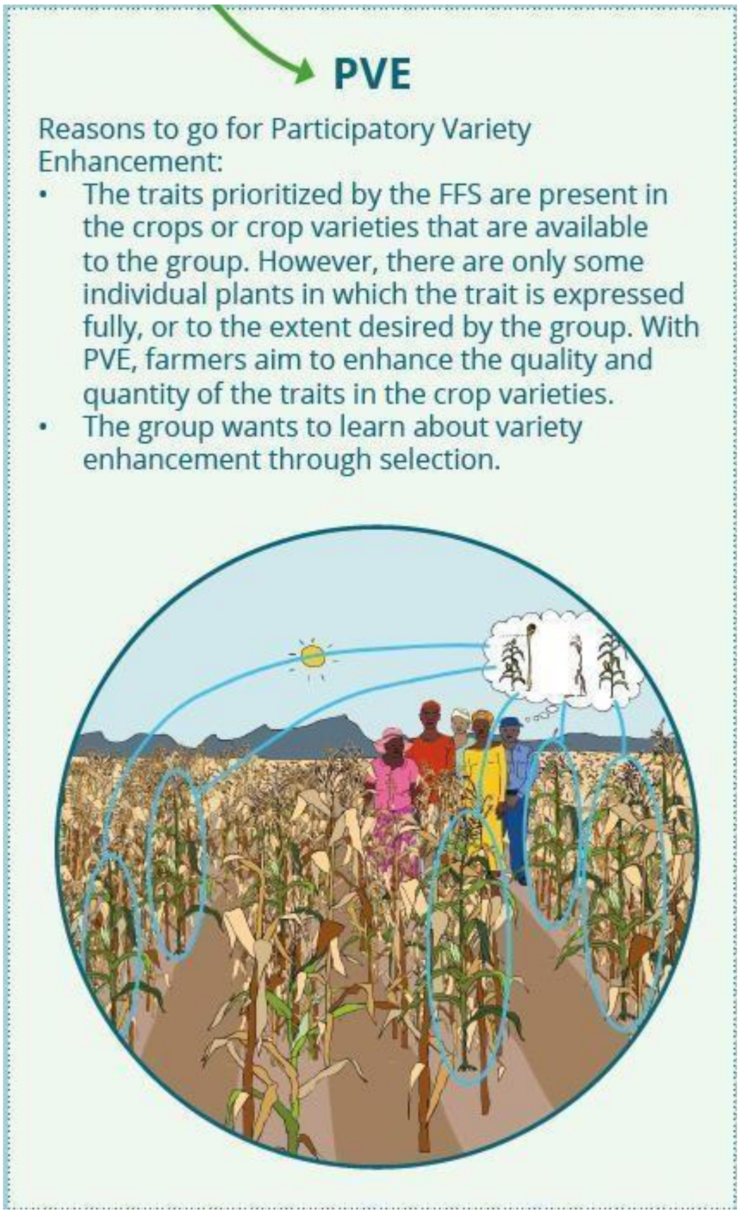
PVS plots are small and even though the FFS can agree that specific varieties are promising, most other farmers in the wider community won't appreciate the results until they can see and compare the variety and its yield in a production-sized plot with *growing conditions that are like* their own.

Therefore, complete a PVS study (or a PVE or PVD study, for that matter) by planting the most promising varieties in larger plots. The size very much depends on local availability of land, but half an acre (e.g. 20 x 100 meters) is good.

With the skills learned in the FFS, and if the participants are convinced with the results, the group may decide to continue the work by engaging in small-scale seed production of the most promising PVS varieties. The Module on Farmer Seed Production and Marketing gives guidance on how to do this.

# II. Selection of plants in Participatory Variety Enhancement (PVE)

*For elaborate instructions on how to do PVE see the [Module on Participatory Variety Enhancement]*




*Reasons to choose Participatory Variety Enhancement from poster about deciding PPB method.*

In PVE, the FFS works to maximize the potential of a valued local, 'traditional' or modern 'open pollinated variety', in accordance with their breeding objectives. This variety may have deteriorated over time or have become ill-adapted to changed conditions, but it has traits the FFS likes. The problem is that too few plants exhibit those traits: the variety shows variability (diversity).

> ***Open pollinated varieties** (OPVs) maintain their characteristics well over time, even if pollinated naturally, by wind or insects, in the field.*

The objective of PVE is to restore and/or improve the variety by **increasing the percentage of plants in the field that have the desired traits**. Or, put differently, by decreasing the percentage of plants that have undesirable traits. The aim is not necessarily to make the variety uniform, as the variability is often a valued characteristic of local varieties, helping them to perform in diverse and changing growing conditions.

While PVE is not about changing a variety, the selection of strong and elimination of weak plants can help a variety perform closer to its full potential, to what it is ideally capable of. For example, an increase in yield of 10 to 25% after three seasons is achievable.

There are **two big differences between PVE and the traditional 'mass selection'** practiced by farmers to maintain their varieties: in contrast to mass selection, i) plant selection in PVE is carried out through all growing stages and ii) poor-performing plants are physically eliminated from the field (only for cross-pollinated species).

> *PVE does not work with hybrids or with clonally propagated crops.* ***Why would this be?***
>
> PVE relies on the existence of (genetic) diversity within the crop population. Hybrids and clonally propagated tend to be genetically uniform. There are no differences between plants that farmers can use to guide their selection efforts.

PVE consists of a number of steps and techniques, which are described in detail in the *Module on Participatory Variety Enhancement*. Highlighted here are the steps that are relevant to the process of plant selection:

### Plot design

In the FFS, plots are always divided according to the number of FFS subgroups: if there are five subgroups, the plot is divided into five or ten subplots, giving each group the same amount of land to manage and the same number of plants to observe. Each subgroup also marks an equal number of plants to select for seed at harvest time.

If the crop is cross-pollinated, plot design should also consider **isolation**: to avoid pollen from other varieties of the same crop 'contaminating' the plants in the plot where PVE is being done, a 200-300 meters distance from plots with those other varieties, or an early or delayed planting time of approximately 15 days, should be maintained. Isolation is not necessary if the crop is self-pollinated.

> ***What if the terrain is hilly, or variable, and plants in one part of the plot look better than in another part?***
>
> *In smallholder farming communities, variability in land is more likely to be the rule than the exception. The advice is to use the following guidelines as standard practice.*
>
> In contrast to PVS, where each subplot should be comparable to other subplots to allow for a fair comparison between varieties (e.g. in hilly terrain, each subplot should include a lower-lying area and a higher part. Plants in the higher parts of the subplots are then compared with each other, as a separate set of plants, and plants in the lower parts as well), in PVE the plot should be subdivided as much as possible following its natural variability. This means that each subplot should cover a part of the larger plot where conditions are distinct and be separate from other subplots where conditions are different. E.g., one subplot high, the other low, one part with poor

soil, the other with good soil.

**Why is this?**

Even if plants in a subplot covering a poor part of the field seem to perform much worse than those in other subplots where conditions are better, it may be environmental variables that determine this performance, rather than the plants themselves. Soil nutrients, soil compaction, water availability, sun exposure, etc., often vary in different parts of a field. In the parts where the field is poor, the plants will perform worse than those in other parts of the field. But this does not mean the plants themselves are inferior – they may be very worthwhile. We simply cannot see the plants are worthwhile because the poor conditions prevent them from performing well.

To compensate for this variability, each subgroup still selects an equal number of best-performing plants from their subplot. For example, if 200 plants are selected in total and there are 5 subplots, 40 plants are selected from each subplot. The seed of all subplots is bulked for the next generation. Breeders call this method stratified mass selection.

### Planting

The **number of seeds planted should be between 2000-5000**. This corresponds to **a plot size of around 500-600 square meters**, depending on the crop and planting distance.

For example, with a planting distance of 25 cm between hills and 50 cm between rows (common for groundnut), this amounts to 4000 plants on a plot of 500 square meters. (See the *Module on Plot design for Participatory Varietal Selection* for examples for other crops).

### Selection

For **self-pollinated crops** (e.g., rice, beans, tomato)**:** only **positive selection** is needed, i.e. selection of well-performing plants with preferred traits. There is no need to eliminate poor-performing plants (negative selection). Plants should not be selected from the edges of the field because of the higher risk of cross-pollination with plants from neighboring fields.

***Positive selection****: selecting plants with desirable traits, by allowing them to reach maturity and selecting their seeds at the time of harvest*

**Why only positive selection?**

In self-pollinated crops, plants only or mainly pollinate themselves. The traits from inferior plants are not transferred to superior plants and do not end up in the seed selected for the next generation. Inferior plants can therefore be allowed to mature, they will simply not be considered for selection at the time of harvest.

Desired traits become visible at different growth stages: e.g., rapid root development (a trait related to drought tolerance) can be observed at the seedling stage, while grain size (a yield-related trait) can be evaluated only after harvest. Therefore, **positive selection must be conducted at all growth stages** and until the plants are mature. Plants with preferred traits are tagged using (plastic) ribbons.

**TIP**: It is useful to use differently coloured ribbons for different traits (breeding objectives) that are selected. E.g. yellow for height, blue for early flowering, red for disease resistance.

*Performing PVE to maximize yield of the local rice variety Jorayal Basmati from Nepal: the FFS selects plants that have a high number of grains per panicle. The best performing plants are tagged with green plastic ribbons. Photograph by Prashit Sthapit.*

For **cross-pollinated crops** (such as maize, pearl millet and gourds)**:** In addition to positive selection, **negative selection** of plants with undesirable traits must be carried out during the entire vegetative stage before **the plants are in flower**.

***Negative selection****: removing unwanted plants before they flower and cross-pollinate other plants, also called 'purifying selection'.*

**Why?** In cross-pollinated crops, pollen from plants with inferior traits can pollinate the flowers of superior plants. The resulting seed will carry the inferior traits, and this will frustrate breeding progress. For more on self- and cross-pollinated species, see also p. 23.

The most secure way of performing negative selection is by eliminating inferior plants. This minimizes the chance that seed from poor performing plants ends up in the next year's seed lot. "Practice selection with a hoe!" is what many plant breeders are taught in school: breeders must be merciless when it comes to eliminating plants that do not fit their breeding objectives. For farmers this can be difficult since negative selection effectively means destroying food.

Many mistakes are made in PVE when farmer-breeders fail to remove poor-performing plants from the field.

A (risky) alternative sometimes chosen in maize is to remove only the male flowers (tassels) of undesirable plants the moment they appear, instead of the entire plant. This way the plants and ears aren't allowed to mature. In addition to securing a harvest, the method has some merits if the traits the FFS selects become visible only in the later growth stages: time to maturity, or husk cover of the ear, for example.

The risk of allowing poor performing plants to mature is that their seed may end up with the seed of selected superior plants, which can set back the work by a season or more. To minimize this risk, properly mark all the plants that are positively selected and do not select seed from plants that are not marked.

*With PVE of cross-pollinating crops, negative selection starts as soon as the crop emerges from the ground. Two grandmothers were the only ones who still had seed of this Zambian community's local maize variety. After three years of strict selection, including negative selection, the variety is again reliable. The entire community is back to growing it, and others from outside have come to buy seed.*

### How many plants should be selected?

**In PVE, selection intensity is generally between 5% and 10%**. This means that *at most* 1 in 10 plants are selected for seed for the next year, i.e., 200 plants or fewer are selected if 2000 plants were initially planted. If more plants are selected, breeding progress would be too slow; if fewer plants are selected, important traits and diversity might be lost.

***Selection intensity****: the intensity with which we select good and remove poor plants; the strictness of the gatekeeper. The higher the selection intensity, the stricter we are in choosing plants, and the quicker the breeding progress.*

*A high selection intensity is indicated with a low percentage of plant selection (e.g. 5%); low selection intensity is indicated with a higher percentage (e.g. 20% of plants selected).*

Selection is strictly guided by the breeding objectives set by the FFS.

**If you want to go deeper: Some further thinking about selection intensity.**

While it is difficult to go wrong with the general rule of 5-10% selection intensity, there are many factors that can help decide whether to select fewer plants (towards 5% selection intensity) or more (towards 10% selection intensity). Some of these are listed below. The most important advice, however, remains: if in doubt, consult an experienced plant breeder!

→ If one selected trait is very important compared to other traits: select fewer plants

→ If several traits are equally important: select more

→ If desirable traits are found in different plants and not all in the same plant: select more

→ If desirable traits become visible prior to flowering (number of tillers, plant height, time to flowering): select fewer. If traits become visible after flowering or harvest (grain appearance, processing/cooking properties, taste): select more

*Explanation: Traits that appear later require the breeder to keep more plants in the field until maturity. As many plants as possible that might have the trait must be kept. Traits that show early can be selected with more precision.*

→ If the variety is in poor shape (many plants with unwanted traits): select fewer

→ If the selected trait is 'qualitative', i.e. can clearly be placed into a 'present' or 'absent' category (red or black seed colour, 8 or 12 seed rows on a cob, resistant to a disease): select fewer

→ If the selected trait is 'quantitative', i.e. can be measured along a range rather than put in a category, or is clearly influenced by the environment (plant height, number of productive tillers): select more

*Consult a breeder to know whether a trait can be easily selected or not!*

**Questions!**

**If 200 out of 2000 plants (10% selection intensity) of a cross-pollinated crop should be selected for seed by the end of the season, how many plants should be negatively selected/eliminated before flowering?**

Answer: There is no fixed percentage of plants that should be negatively selected. Only the better plants should be allowed to flower, but make sure that more than enough remain so that the FFS can select the 200 plants (10%) with the desired traits for seed for the next season.

**If ten plants of a cross-pollinated crop that were positively selected in the early vegetative stage, for example because they showed vigorous growth, end up producing small grains, should their seed still be selected for next season?**

Answer: Again, there is no clear yes or no; this is the 'art' part of plant breeding. The FFS may decide that vigorous growth is less of a priority than grain size (yield). In that case they will choose to discard the plants because of the risk they pose to reducing the yields of the other plants through cross-pollination. Especially if the remaining 190 plants also have reasonably good early growth, it is not worthwhile to keep the ten plants that were selected early.

If, on the other hand, vigorous growth is key, the plants should be kept!

### Harvesting and completion

At harvest time, superior panicles, heads, cobs or pods with positive traits are selected from the tagged plants. From these, good looking seeds that are healthy and disease-free are selected.

Selected **seed is *bulked*** (put together) into a single seed lot, or cobs and panicles might be combined in bunches and stored as such. Seed for the next season is taken from this lot.

**Selection must be repeated over several seasons** to achieve good results. **Three seasons** is the average, but it depends on several factors: the quality and variability of the initial seed lot, the breeding objectives set and how difficult they are to achieve, the weather conditions during the growing seasons, and how picky the FFS members are. 'Good enough' for own use in a community may not be 'good enough' for a seed enterprise or plant breeder who wishes to release seed to the market. Be clear about the final goal and remember: it is the FFS who collectively decide whether the work is complete.

Once a seed lot shows significant progress, even after one season, the FFS should conduct a comparative trial on a production-sized plot against the original (unimproved) seed lot to test the progress.

As with PVS above, and now that the FFS has produced a superior version of the variety, with good quality seeds, the group may decide to take up seed production and market the seed locally.

**If you want to go deeper: What if a local variety has more than one "type"?**

First remember that PVE selection is based on traits: the breeding objectives set by the farmers. Some basic traits must be uniform, for obvious reasons. E.g., the time to harvest or plant height in most crops.

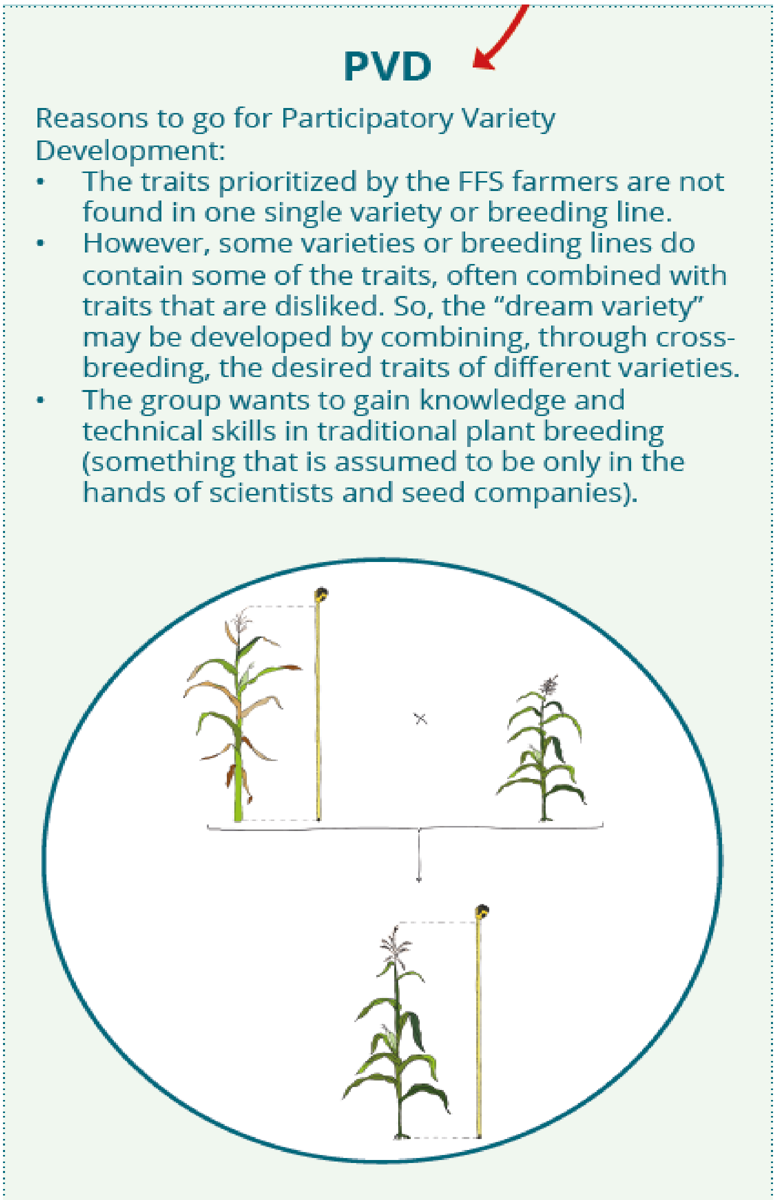
But traditional varieties can contain different 'types', with different traits, within a single population and there are reasons why such differences are sometimes desired. For example, the upland rice variety called "husband and wife" in Laos is composed of two main types; one is very high yielding but tends to lodge, the other is hardy and strong but less productive.

Planted together through broadcast seeding, the high yielding type is supported by the hardy type. If planted separately, both become useless. How do you carry out PVE with such a variety?

To ensure the types remain distinct, seed of different types is usually separated and planted in separate plots for easier selection. In this case of dependence however, the types are planted together and the best plants from each type are selected according to the distinct breeding objectives of that respective type. When the objectives are reached, the seed (if separated for selection purposes) is recombined to maintain the mixture that was useful to farmers and the growing condition.

The same approach applies to crop varieties that are grown together with varieties of completely different crops, e.g. beans and maize (and often gourds) in Latin America, or cowpea and sorghum intercropping in African countries. Their selection and improvement should not be done separately from their 'companion crops'.

# III. Selection of plants in Participatory Variety Development (PVD)




*Reasons to choose Participatory Variety Development from poster about choosing PPB method.*

### What is PVD, and when to do it?

'Participatory variety development' is *the creation of new varieties* with farmers taking active roles in the work, for example in creating new diversity by combining ('crossing') different parent varieties and by selecting promising plants from that diversity. PVD is done when individual existing varieties do not have the traits, or the combination of traits, that farmers need. The offspring resulting from a crossing will combine the traits of their parents, in many different combinations. This *new diversity* forms the raw material for PVD. Patient and skillful selection of these diverse plants until they become uniform and stable, requiring at least 7 seasons, or generations, is at the heart of plant breeding and can result in the creation of one or more new varieties.

*PVD is exciting, but requires a lot of work, determination, and patience. While there are many examples of farmers successfully creating their own varieties, we advise to do this with a committed FFS in close collaboration with a professional plant breeder.*

## Outline of a PVD trajectory

### What does a typical, multi-year PVD trajectory look like for an FFS and the plant breeder working with them?

The trajectory for PVD outlined here is practical and feasible within the context of a Farmer Field School. It combines, as much as possible, the complementary skills and resources of farmers and plant breeders, while minimizing the 'burden' of the work. There are many techniques to do plant breeding. Some are very advanced and difficult to practice in farmers' fields, while others can be used more easily by farmers, and all are variations of the basic methods explained in this document.

Before we go into more detail, a summary:

1. PVD, especially in the later stages of selection, should be undertaken by a committed FFS, who have gained skills with Participatory Variety Selection or Participatory Variety Enhancement. They must be ready and eager to commit to the work for a long period of time, often extending beyond the timeframe and support of individual projects. The plant breeder working with them must commit to supporting the FFS. It is not always necessary for the entire FFS to be involved all the time, as long as a few motivated FFS members continue to carry the work, engage with plant breeders, and share results with their peers.

2. There are two ways to begin PVD: i) to start with the crossing of parent varieties ('season 1' in the table below), or ii) to work with a *segregating population* derived from a crossing made earlier (e.g., 'Generation 4/season 5' in the table below). The second option takes less time and is preferable. However, *it only produces desirable results if the parental lines that were crossed to produce the segregating population were selected with the breeding objectives of the farmers in mind.*

> ***Segregating population**: the diverse plant population resulting from a cross. Genetic diversity can be seen in the different appearances of plants. These visible differences allow selection for breeding (see p. 23)*

3. When managed by an FFS, the first generations of a segregating population should be grown for several seasons until segregation is reduced, around Generation 4 or 5. Selection at this stage broadly follows the breeding objectives, but is not yet very stringent (i.e. plants with poor traits are removed, while the number of plants with preferred traits kept for the next round of selection is relatively large).

4. If the initial crossing is done in the FFS, the first seasons of growing the population, from Generation 1 to 4, may not require that much work. Unlike at later generations, weekly selection or monitoring is not yet needed. To make sure the FFS remains engaged throughout the season, meeting regularly and becoming stronger as a group, PVD can be done as a side study in parallel with PVS, PVE, or even seed production for economic purposes. After a few seasons, at Generation 4 or 5, when selection becomes more important and labour-intensive, the FFS should focus only on PVD.

5. After Generation 4 or 5, strict selection of plants, in the form of *pedigree selection* (for self-pollinated crops), or *recurrent selection* and negative selection (for cross-pollinated crops) is

applied to ‘shape’ the population into the desired variety.

> *In **pedigree selection** individual best-performing plants are selected from a segregating population and tested for several generations (see p. 36)*
>
> *In **recurrent selection**, plants are selected not just based on how they look, but also on how their offspring performs (see p. 36).*
>
> *Both techniques are used to increase the precision and speed of selection efforts.*

6. Around Generation 7, when the breeding population becomes more uniform and stable, begin performing preliminary yield trials and tests for taste, cooking quality, etc. A little bit more seed should be bulked after harvest to allow for these tests. Promising breeding lines can be disseminated to other FFS for multi-location testing and to share the burden on labour and land.

7. To increase the number of generations that can be grown in a single year and reduce the time needed to create a variety, seed can be ‘shuttled’ back and forth after every harvest between FFS and a breeding institute, where favourable growing conditions (e.g., irrigation) are available during the off-season. This is most important for locations with only one growing season per year.

The table and illustrations on the next pages give a step-by-step overview of the PVD trajectory.

Individual concepts and techniques are explained in detail in the section ‘PPB concepts and techniques in more detail’ (p. 22).

# Table: Year-by-year selection in participatory variety development

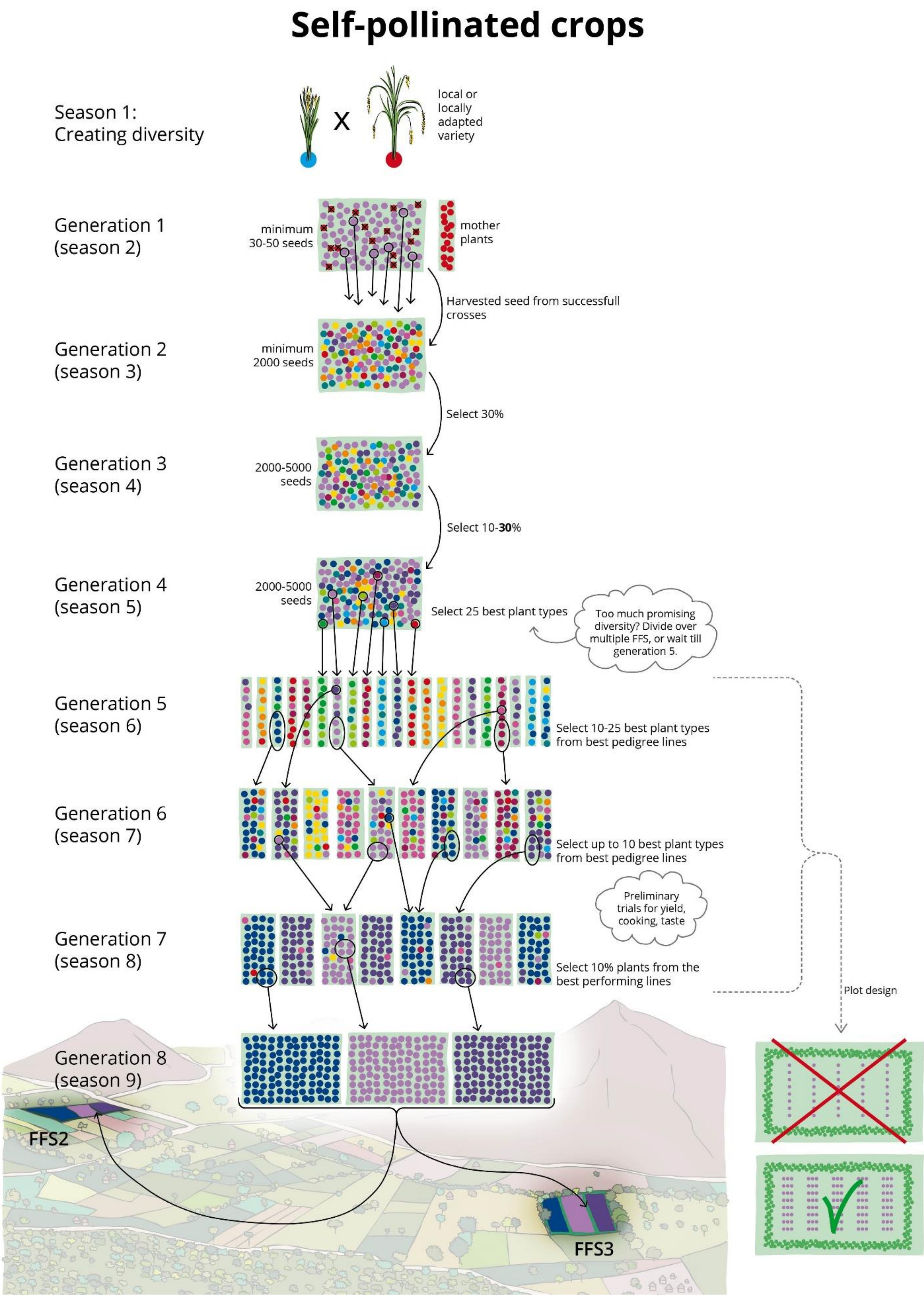
*Each term that requires explanation has a page number [23, 30, …] and is underlined. Clicking on the term or page number will take you to the relevant explanation in the document.*

**Note**: This table is an example of a possible PVD trajectory. It is by no means the only way and should not be seen as fixed. Depending on conditions, crop, breeding objectives, chosen method, (support) capacities and, alas, luck, numbers and timeframe may decrease or increase.

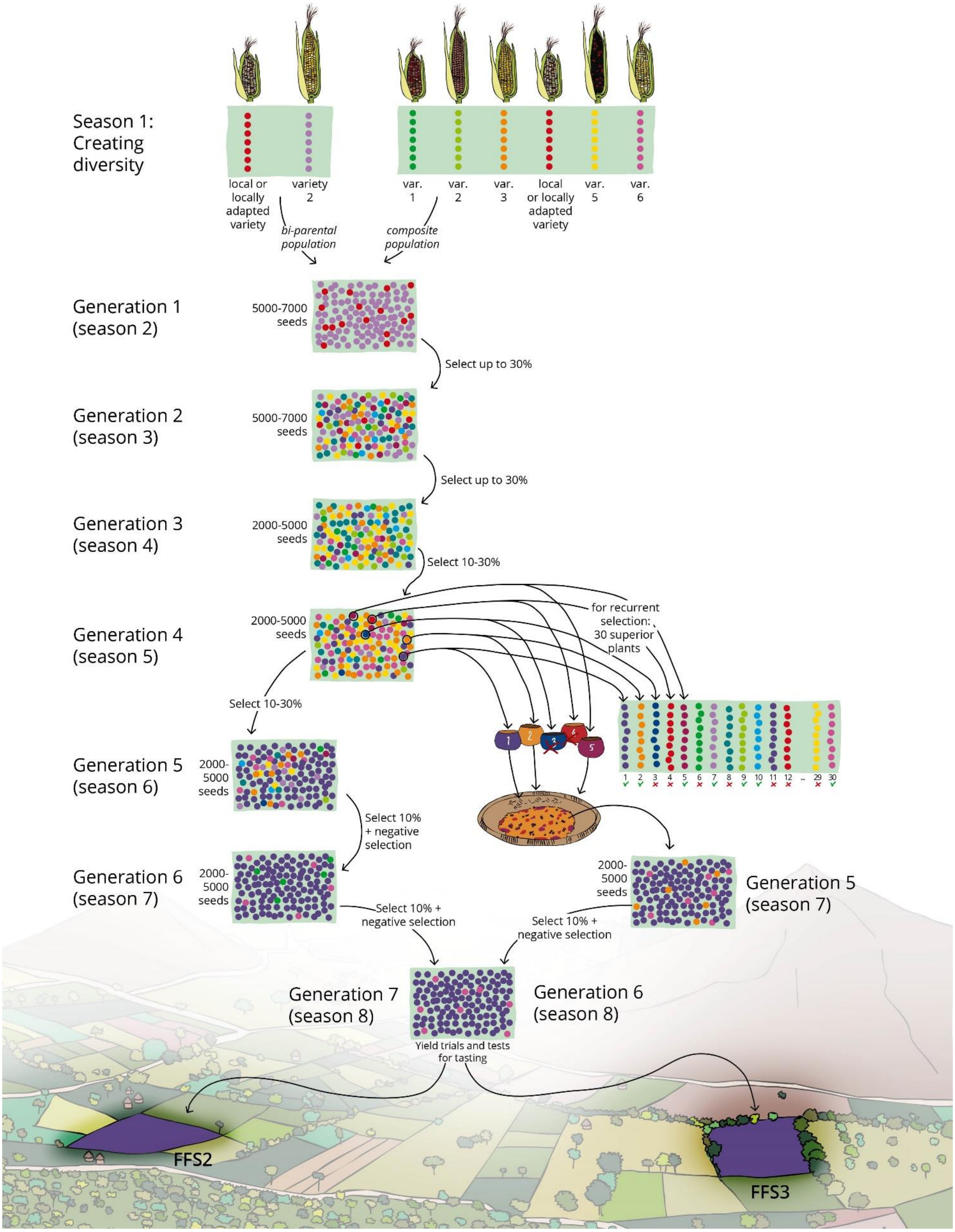
| Crop / Season, Generation | Stage | Self-pollinated[23] (e.g.: rice, beans, tomato) | Cross-pollinated[23] (e.g.: maize, pearl millet, cabbage) |
|---|---|---|---|
| **Season 0** | **Design** | Pre-selection of parent varieties, procuring seed | |
| **Season 1:** | **Creating new diversity** | Create a 'segregating population'[23] by crossing 2 parent varieties[27] Harvest all seed of successful crosses[4] and bulk the seed (minimum 30 – 50)<br>Optional: shuttle breeding[25] to speed up the breeding process | Create a 'bi-parental' (two-parent) population[27] by crossing 2 parent varieties, or create a 'composite population'[28] by crossing several (6 or more) diverse parent varieties<br>Harvest and bulk all seed<br>Optional: shuttle breeding[25] to speed up the breeding process |
| **Generation 1**<br>**Season 2** | | Plant all harvested seeds[30]<br>If it was not possible to check if crossing was successful in year 1 (e.g. for rice), plant Gen. 1 seeds alongside mother plants[28] ➔ compare and harvest and bulk seed from all successful crosses<br>Optional: shuttle breeding[25] (can be continued throughout) | Plant as many harvested seeds as possible, ideally 5000 – 7000[30]<br>Mass selection[32]: select seed from **30%** of the plants and bulk for the next season<br>Optional: shuttle breeding[25] (can be continued throughout) |
| **Generation 2**<br>**Season 3** | **Selection** | Plant 2000 seeds[30]<br>Bulk selection[9]: **30**% from all plants produced | Plant 5000 – 7000 seeds[30]<br>Mass selection[32]: **30**% from all plants produced |
| **Generation 3**<br>**Season 4** | | Plant 2000 – 5000 seeds[30]<br>Bulk selection[9]: 10 – **30**% selection intensity[31] | Plant 2000 – 5000 seeds[30]<br>Mass selection[32]: 10 – **30**% selection intensity[31] |
| **Generation 4**<br>**Season 5** | | Plant 2000 – 5000 seeds[30]<br>Bulk selection[9]: 10 – 30% selection intensity[31]<br>Positive selection[31] of superior plants during season<br>Now or at Gen. 5: Switch to pedigree selection[36] at the end of the growing season: select seed from the 25 most desirable plant types. | Plant 2000 – 5000 seeds[30]<br>Mass selection[32]: 10 – 30% selection intensity[31]<br>Optional: recurrent selection with progeny testing[36] to speed up selection process (requires 2 seasons) |

| | | | |
|---|---|---|---|
| | **Selection** | If necessary because of attractive variation, select more than 25 types. Possibly divide these over multiple FFS[37] | |
| **Generation 5**<br>**Season 6** | | Plant up to 25 lines per FFS (one line for each selected plant type).<br>Pedigree selection[36]: select best plants from best lines to continue with 10 – 25 lines | Plant 2000 – 5000 seeds[30]<br>Mass selection[32]: **10%** selection intensity[31]<br>Negative selection[32] of inferior plants |
| **Generation 6**<br>**Season 7** | | Plant 10 – 25 lines.<br>Pedigree selection[36]: select best plants from best lines to continue with maximum 10 lines<br>Harvest more seed to allow for tests at Gen. 7 | Plant 2000 – 5000 seeds[30]<br>Mass selection[32]: **10**% selection intensity[31]<br>Negative selection[32] of inferior plants<br>Harvest more seeds to allow for tests at Gen. 7 |
| **Generation 7**<br>**Season 8** | **Maintenance** | Plant up to 10 lines in slightly larger plots[39]<br>Preliminary yield trials, testing for taste, cooking, etc.<br>Select promising lines and disseminate to other FFS for multi-location testing[39] | Plant 2000 – 5000 seeds[30]<br>Preliminary yield trials, testing for taste, cooking, etc.<br>Continue to improve population through positive and negative selection. Disseminate seed to other FFS for multi-location testing[39] |
| **Generation 8**<br>**Season 9** | | Continue to improve breeding lines through positive selection and by removing outliers.<br>Stable self-pollinated variety[40] | Continue to improve population through positive and negative selection.<br>Selection intensity: select less than 10% of the plants for further seed production<br>Stable open-pollinated variety[40] |

# Illustrations: Year-by-year selection in participatory variety development



# Cross-pollinated crops



# PPB concepts and techniques in more detail

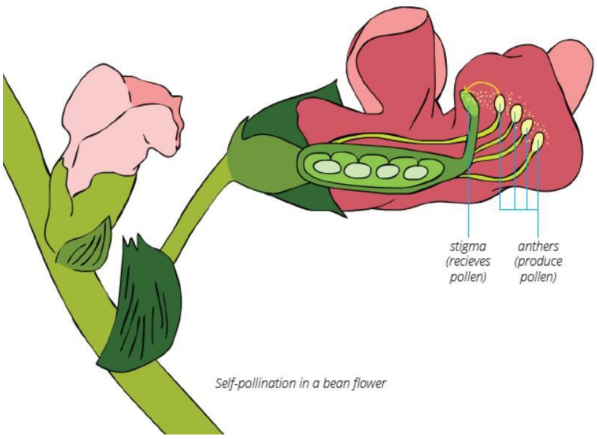
## I. General concepts

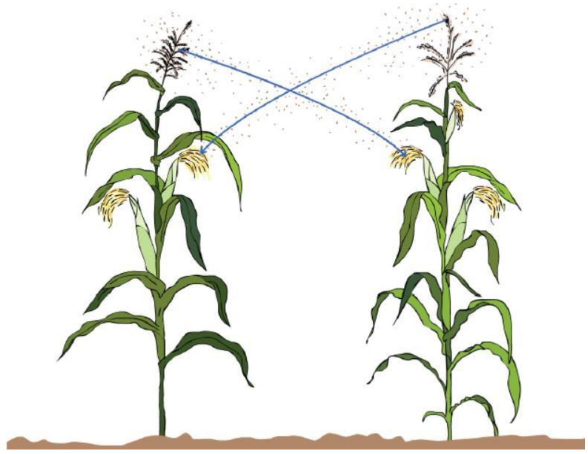
### The difference between self- and cross-pollinated plants

Self-pollinated crops such as rice, beans and tomato fertilize themselves; they are 'inbreeders'. The plant is both mother and father of the seed it produces and we do not usually have to worry about 'contamination' by genes from other plants of the same species growing in nearby fields or plots. In many self-pollinated crops, however, a percentage of plants can also be cross-pollinated. This ranges from as low as 1 – 2% (e.g., in barley and rice) to up to 30% in sorghum.

Cross-pollinated crops like maize, pearl millet and cabbage are fertilized by other plants; they are 'outcrossers'. The plant is mother of the seed it produces, but the seed's father is unknown, although it must grow nearby. This makes selection less predictable: every time a seed is selected, we know only half of its genes and only part of what the plant will look like.

This difference means selection in cross-pollinated crops happens very differently from selection in self-pollinated crops. For self-pollinators it is possible to focus selection on individual plants, and to follow their offspring (pedigree) generation after generation. For cross-pollinators such 'pure-line' selection is not possible. Instead, selection focuses on improving a group of plants (a population) by selecting many superior plants and allowing them to cross-pollinate to gain improved traits together, generation after generation. All or most plants in the population of cross-pollinated plants will contain some major traits for which selection takes place. Certain non-selected traits will also remain present, but to a much lesser extent, in fewer plants.



*The flowers of legume crops such as beans contain both male and female parts. They pollinate before the flower opens, making cross-pollination with other plants virtually impossible. Maize has separate male (on top) and female flowers (the ears) that flower at different times. This ensures cross-pollination with different plants.*

### What is a segregating population?

A segregating population is the offspring that results from the crossing of two or more distinctly different parents. Plants in the first generation after a crossing are referred to as F1, or the "first filial generation" (filial meaning "son" or "daughter)". They will be uniform, but starting with the second generation, the offspring will show high variation – our new diversity and the raw material for the selection process. From this moment and until the variability decreases and stabilises at a lower level, around the fifth or sixth generation, we refer to the plants as a segregating population.

Plants in a segregating population have the curious quality of being ‘unstable’: even for plants that self-pollinate, the seed from a desirable plant may not produce a similar-looking plant the next season. And seed harvested from an undesirable plant may produce a plant with desirable traits the next generation: the genes that carry the plant’s traits from parent to offspring are still being reshuffled inside the plant and inside the population; they are ‘segregating’. With each generation, segregation slows, stability increases and with it the probability that plants produce similar offspring as their parents. For this reason, *in the early generations of a segregating population, selection for visible traits is difficult and, for the risk of losing too much diversity, should not be done in too strict a manner. It becomes easier after arriving at Generation 4 or 5*.

This means that when an FFS receives a segregating population from a breeder, it is important to know the generation of that population: if it is at the 2$^{nd}$ or 3$^{rd}$ generation, it does not yet make sense to perform strict selection of individual plants. Instead, the advice is to grow the population over several seasons: eliminate off-types, allow natural selection to do its job and bulk seed of the healthy-looking plants after harvest. If the population is more advanced and segregation is reduced, Generation 4 or later, the plants are sufficiently stable to allow for strict selection (see ‘pedigree selection’, ‘recurrent selection’ and ‘negative selection’ below).

A distinct advantage of beginning PVD work with a segregating population rather than a crossing is that it gives the FFS a head start. They need less time (fewer seasons) to develop their own variety, while still having a vast range of diverse traits to select from. It also means that a wider range of techniques may be used to create the original population – techniques which are not feasible in an FFS as they require more expertise or advanced technology.

***Examples of more advanced techniques*** *are refined crossing schemes such as two-way, three-way and double crosses, back-crossing to ensure the transfer of a specific trait, the use of elite lines (breeding lines that have been bred to contain very desirable traits for e.g. yield, disease resistance, local adaptation), or mutant lines (seeds that have been exposed to chemicals or radiation to change their DNA and bring about new, ‘exotic’ traits), and using marker assisted selection to guarantee that selected plants carry the traits of interest. All these techniques can and have been used in PPB processes, but breeders must take time to discuss them with the farmers so that the techniques are understood, and farmers can decide whether they consent with their use. Without such agreement, distrust is likely to arise.*

*An FFS in Zimbabwe performing positive selection (marking) on a sorghum segregating population with clear variability in plant height, head size, seed colour and time to maturity.*

*This is a second attempt. The first time this FFS tried PVD, they performed very strict selection without knowing that what they had received from the breeding institute was a 2nd generation segregating population.*

*After two seasons it became clear that too few plants and too little diversity were left to continue the work and they had to start anew.*

## Shuttle breeding

PVD takes a long time, especially if the FFS chooses to start with a crossing rather than a more advanced Generation 4 or 5 segregating population. Whenever possible, we recommend speeding up the process as it strengthens the resolve of participating farmers. One way of doing so is to have more than one growing season in a single calendar year, thus 'advancing' the segregating population multiple generations. In warm climates this may be possible on-farm, often requiring irrigation during the dry season. In environments where this is not possible, the harvested seed may be brought back and forth ('shuttled') to a second location where growing conditions are favourable during the off-season: a crop breeding station, for example, where irrigation is available. Cold months must be avoided for crops that are not tolerant to cold. It is brought back to the community in time for the next growing season. Shuttle breeding is a great way to lessen the burden of the work and can help forge a strong collaboration between FFS and breeding institutes. In agroecosystems which normally only feature a single growing season, it can cut down the time needed to create a new variety from 9 to 5 years.

## Plot design

Plot design has a bearing on plant selection; in PVD plot design changes according to the stage and method of selection. Below are a few key considerations to keep in mind.

**Size**: the main concern is to plant sufficient seed to capture the necessary diversity. Plots for bulk- and mass selection should accommodate the number of seeds indicated in the table on p. 19: between 2000 and 7000, depending on the generation. This generally corresponds to **a maximum plot size of around 500-600 square meters**, depending on crop and planting distance.

***Bulk and mass selection*** *(p. 32) involve harvesting seeds from a population of plants collectively, without individual plant selection or regard to individual ancestry. The aim is to improve the entire population's traits.*

When moving to pedigree selection for self-pollinated crops, the number of seeds planted decreases, and so does the size of the plot: if, in Generation 5, we plant 25 pedigree lines in single rows of 30-50 seeds, a plot size of 150 – 200 square meters is enough. In subsequent generations the number of pedigree lines generally decreases, but since the number of seeds planted per line increases, the overall plot size remains the same.

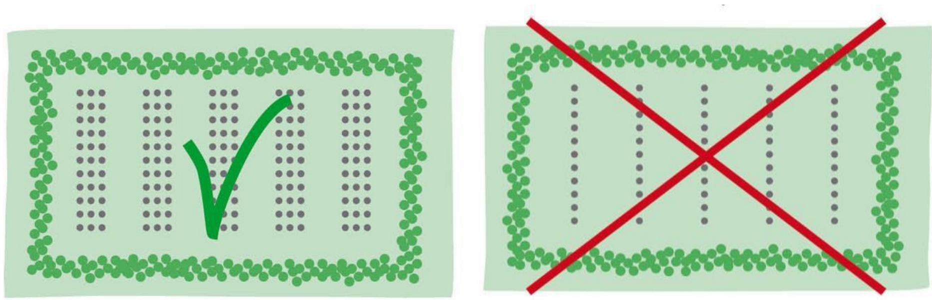
For recurrent selection in cross-pollinated crops, plot size is similar to pedigree selection, with one row of seeds planted to each of the 30 selected plants.

**Isolation**: for cross-pollinated crops, maintain the usual isolation distance or time to avoid contamination: 300 m from other plots with different varieties of the same crop, or 15 days early or delayed planting to avoid simultaneous flowering.

In self-pollinated crops isolation is not needed but avoid selecting plants from the plot borders to minimize the impacts of accidental outcrossing (remember that the percentage of outcrossing can be high, especially for crops like sorghum).

**Borders and variability in terrain:** for PVD, growing conditions should be similar throughout the plot. In the early generations of a segregating population, variability among plants is high and the predictability of selection efforts low. Variability in terrain would add a further unwelcome level of unpredictability and should be avoided. To respond to the need of creating varieties that respond well to different growing conditions, PVD can be conducted with the same population in different locations (FFSs) where those conditions exist.

Similarly, when comparing pedigree lines in later generations, variability in terrain should be avoided. Each line should be evaluated under the exact same growing conditions to allow for fair comparison. Pedigree lines planted near the borders of a plot tend to be over-selected as these parts of a plot have more nutrients, sunlight, and less competition. To reduce such bias, a border of the same crop can be planted around the PVD plot and pedigree lines should be planted in mini plots rather than single lines.



## II. Creating new diversity: making crosses

Without diversity, there can be no change! Participatory Variety Development creates new diversity where existing diversity is inadequate. We do this by crossing different parent varieties. Choosing the best parent varieties to make a cross is extremely important. To a large degree it will determine the success of the ensuing years of work. Make this choice together with a professional plant breeder, based on the breeding objectives set by the FFS. The breeder can also help assess beforehand *whether the traits the FFS wants to combine in a new variety are likely to be compatible*. For example, early maturity and big grain size often are not. Trying to achieve this for years in a row, without result, can be very frustrating!

### Self-pollinators: crossing two parent varieties

In the SD=HS programme, farmers often choose to cross a local variety with a modern or improved variety, including hybrids, with the aim of 'inserting' (adding) one or two traits from the improved variety into the local one, or vice versa (e.g. high yield, disease resistance, taste). See the *Meuong Phiang* 1 example below.

In selecting parent varieties for crossing, it is best to always choose varieties that are as different as possible from one another. Such crosses will produce segregating populations with more diversity and a higher potential for new promising varieties with strong traits.

### Cross-pollinators: crossing two parent varieties or creating a 'composite population'

For a farmer field school, there are two practical ways to create new diversity in cross-pollinated crops. Both methods create new diversity by combining the characteristics from the different parents. From this diversity a new 'open pollinated variety' (or varieties) can be developed. Both methods require selecting the parent varieties with care, making sure they have interesting traits that correspond with the breeding objectives identified by the FFS.

1. **Crossing two parent varieties (bi-parental cross)** can be done to improve an existing local variety in which an essential trait is missing or weakly expressed. That trait can be introduced or improved by crossing with another variety that is superior for that trait. Resistance to a new

**Note**: it is not feasible to only transfer one single trait or gene from parent to offspring. A combination of all the parents' genes in the offspring is inevitable – hence the need for repeated selection to weed out the unwanted variation.

It is also important that one of the parents is adapted to local conditions (such as the farmer variety mentioned above).

To make the cross, the two parent varieties are grown side by side in the FFS plot, *making sure flowering periods overlap*. Flowers are cross-pollinated manually and the flowers (or ears, panicles, heads) that have been pollinated are labelled.

## How to know if a crossing is successful?

Manual pollination requires skillful hands, as flowers are often small and delicate, and the time window for pollination may be limited to specific hours of the day, during only a few flowering days.

For some crops it will be apparent before harvest whether manual cross-pollination was successful. Groundnuts, for example, will grow pegs only from flowers that have been successfully pollinated. Flowers where manual cross-pollination failed wilt and die (when pollinating, the breeder removes the (male) anthers from the flowers so that the (female) stamen can only be pollinated by the foreign pollen inserted manually). All flowers where manual pollination succeeded must be labelled.

For other crops, like rice, successful pollination becomes visible only after planting the first generation (Gen. 1) seeds. For these crops, in the season following the one when the cross was made, Gen. 1 seeds should always be grown alongside a row of mother plants for comparison: seeds that produce plants that look like the mother plant have self-pollinated instead of cross-pollinated and should be discarded.

pest or disease, or a desired grain quality are examples of traits that may be improved by such crosses. One parent should be a well-known local variety that is lacking in a specific trait. The second parent should carry the trait to improve the local variety.

2. **A newly created composite population** is expected to have great diversity. Variation among plants in a composite population results from the traits (genes) coming together from many different parent varieties. This variation is expected for all traits for which the parents are different.
   *A composite breeding population requires 6 or more well-performing parent varieties*. At least one of them should be adapted to local growing conditions. The greater the difference, or 'genetic distance' between the parents, the higher the potential for new promising varieties. One way of ensuring this is by choosing parents that come from very different places.

To allow cross-pollination between parent varieties to take place, equal amounts of seed of each variety are planted together in a single plot. Cross- pollinated crops cross by themselves, without the help of human hands (most even have built-in mechanisms to prevent self-pollination). If varieties have different flowering times, plant them in rows at different time intervals to ensure synchronized flowering: early planting for late flowering varieties, and late planting for ones that flower early.

Make sure the plot is isolated, by flowering time or in distance, from other plots of the same crop.

**Note**: If storage facilities are available, it is very important to store part of the 2nd generation seeds (or the seeds of any generation, for that matter) for later. This saves a lot of work if at any time in the breeding process seeds are lost due to crop failure. Or if the FFS wishes to return to the original breeding population, for example to select for different traits or to repeat the selection.

**Meuang Nga x TDK1 → Meuong Phiang 1**

| | Meang Nga (local) | TDK1 (improved) | MP1 (new) |
|---|---|---|---|
| **Seed Color** | White | Dark Red | Light red |
| **Maturity (Days)** | Flowers in October (photosensitive) | 135-140 | 140-145 |
| **Crop season** | Wet | Wet and Dry | Wet and Dry |
| **Plant Height (cm)** | >130 | 90-100 | 130-150 |
| **Milling (%)** | 60% | 50-55% | >60 |
| **Grain Yield (t/ha)** | 2-2.5 | 4-6 | 4.5-6 |
| **Rice gall midge** | Resistant | Susceptible | Resistant |

*Mixing modern and traditional: an FFS in Meuong Phiang district in Laos crossed their local Meang Nga variety with TDK1, an improved variety developed by the Lao Rice Research Institute. The resulting Meuong Phiang 1 combines the yield, eating quality and non-photosensitivity of TDK1 with the resistance to gall midge and milling percentage (over 60%) of the traditional Meang Nga variety.*

*An interesting result of the continuous selection under real-life farming conditions is that the variety is adapted to and yields well in relatively poor, sandy soils. It grows quickly and covers competing weeds which makes it suitable for direct seeding (broadcasting). Adding too much fertilizer, on the other hand, will cause the variety to lodge and perform poorly.*

# III. How many seeds to plant at different generations?

## For self-pollinators:

At Generation 1:

The first season after making the cross, the seed from all successful crosses is planted. Depending on the crop and the effort required to manually cross-pollinate the flowers successfully, the number of Generation 1 seed may be limited, but a minimum of 30 to 50 seeds is advised. By Generation 2, there should be ample seed.

## For cross-pollinators:

At Generation 1:

After the parent plants have been allowed to cross-pollinate, we can assume that all harvested seed contains traits (genes) of several parent plants; there will be plenty of seed to harvest and plant. Plant as many as possible, ideally between 5000 – 7000.

At Generation 2: At this early stage, the breeding population is at its most diverse and very unstable. To capture all that diversity, as many seeds as possible should be planted, as this will be the basis from which selection will happen in the later generations:

*Plant no fewer than 2000 seeds for self-pollinated crops*. The maximum number of seeds planted is guided by the space available in the plot. It need not be higher than 5000 for self-pollinators.

*Plant no fewer than 5000 seeds for cross-pollinated crops*. The maximum number of seeds planted is guided by the space available in the plot. It need not be higher than 7000 for cross-pollinators.

Generation 3 and later:

In subsequent generations, the number of seeds planted can be reduced as homogeneity increases, staying within the range of 2000 – 5000 seeds for both cross- and self-pollinated crops.

**A general rule of thumb:** *the more segregation, or variability, in the breeding population, the higher the number of seeds needed to plant*. See also 'selection percentages' below.

# IV. Basic selection methods and selection intensity

## Selection intensity: 10% or 30%?

How many plants should be selected at each generation? There is no fixed rule, but remember the gatekeeper: select too many plants and breeding progress will be slow, select too few plants and there won't be enough plants with the desired traits to select from.

When a population is still unstable (meaning changing) and has more variation, usually during the first generations, it is wise to select a higher percentage of plants: up to 30%, and fewer if variability is lower. At this stage, our primary concern is to capture sufficient diversity for the next generation, including those traits, or combinations of traits, that may not yet be visible.

When, in later generations, the population becomes more stable and with less variation, our main concern is to shape the variety according to our breeding objectives and we become much stricter in the plants we select: at this stage we select only 10% of the plants for seed, or move to pedigree selection or recurrent selection to speed up the process.

Within this range of 10 – 30%, consider the list of factors on p. 13 ('*Some further thinking about selection intensity*') that help determine whether to select more or fewer plants.

### *Marking (positive selection) of superior plants*

Positive selection is used in all selection methods, for self- and cross-pollinated crops, to help the final selection of seeds at harvest time. Desired traits can become visible at different growth stages, and may disappear later in the plant's development, so marking well-performing plants is necessary throughout the entire growing season if we do not want to lose them from sight at the time of harvest. See p. 11 for more details.

Tagging of plants continues until the plants are mature. At harvest time, the best panicles, cobs, pods etc. with positive traits are selected from the tagged plants. After harvest, all good-looking seeds are harvested from these cobs or panicles.

## For self-pollinators: Bulk selection

Until Generation 4 or 5, our concern is mostly to 'advance' the breeding population without applying stringent selection by farmers: to grow several generations so that the diversity generated in the initial cross can become visible in the plants, and the major traits become more defined and distinct. This method is called 'bulk selection'. Bulk selection allows adverse environmental conditions to take out the weak plants through natural selection. The seed from plants that survive and look good is bulked into a single seed lot and used to produce the next generation.

*We do not perform strict selection in these early generations as this would result in a major loss of diversity* and endanger our selection work in later generations.

In bulk selection, up to 30% of the plants in the field are harvested for seed. Selection is not yet for traits, but rather by choosing not to harvest the weakest, shortest or tallest, earliest or latest plants. This results in progressively more homogeneity (uniformity) in each successive generation. If continued, bulk selection will eventually, after approximately 10 seasons, lead to a stable and relatively uniform variety. This seems very long, but *already after 5 or 6 seasons*, farmer-breeders can begin to cultivate the beginning variety (still referred to as a population) on production-sized plots, provided the farmer does not mind the variability and is keen to continue selection for desired traits.

The process can be sped up significantly, however, by using *pedigree selection* (see p. 36 below) from Generation 4 or 5 onwards.

## For cross-pollinators: Mass selection

The mass selection method in cross-pollinated crops is like the bulk selection method for self-pollinators, but with stronger selection intensity applied, from an earlier stage (Generation 2). Stronger selection is needed because the initial level of diversity is higher in populations of cross-pollinated crops. Focusing on key visible traits, well-performing plants are selected from the Generation 2 composite population, and their seed harvested and bulked to produce the next generation. We repeat the process until the population becomes more stable, around Generation 5, at which point we *increase selection intensity and begin to apply negative selection*.

Mass selection is simple and *can be very effective for traits that can be seen and selected before, or at the time of flowering* (e.g. flowering time, number of ears in maize, resistance to fall armyworm, plant and ear height, adaptation to changing conditions).

### *Negative selection of inferior plants*

After four seasons of mass selection, the population will begin to look more like a variety: still very diverse, but with a set of clearly present traits that correspond to the breeding objectives set by the FFS in most or all plants. At this point, we start to treat the population like a variety that should be improved and perform selection as we would when doing PVE. This *means we start actively eliminating undesirable traits from the population through **negative** selection*.

*Negative selection* in cross-pollinated crops should start as soon as the crop emerges from the ground. All weak, diseased plants and plants showing undesirable traits within a plot are removed. This process is also called 'roguing', as the 'rogue' plants are taken out. Roguing is done during the

*Bulk selection in a 4th generation sorghum population in Chirundu FFS in Zambia. The heads of plants selected to advance to the next generation are protected against bird predation before harvest.*

*One of the strengths of PPB is that **farmers and breeders look and select differently.** The breeder working with this community admitted that, afraid of losing critical diversity, he would always go through the field to secretly mark a few more plants after the community had finished their selection. The community, from their side, did the same: after the breeder advised the discontinuation of specific pedigree lines, they would nevertheless store the seeds in their community's seed bank as backup.*

entire vegetative growth stage and up to the point where flowers first appear. It must be done before any pollination can occur. All the while, **positive** selection is also carried out by marking well-performing plants.

To minimize the effects of environmental influences on selection efforts, remember to subdivide the plot into equally sized smaller subplots and select equal numbers of superior plants from each of them (see p. 10). Then bulk the seed.

Mass selection, coupled with negative selection, will result in a relatively stable and uniform 'open-pollinated variety' after approximately 8 seasons.

Recurrent selection (see p. 36, below) can be applied to speed up the process.

**How many traits can be selected in PVD?** An FFS usually comes up with 7 to 10 priority traits during the diagnostic stage. These can't all be realized, in any case not in equal measure, in a new variety.

**How do we choose?** As this example for sorghum in West Africa shows, arranging traits into priority categories adds focus and helps the FFS agree where selection should be strict and where it can be more flexible (adapted from E. Weltzien 2018).

| Trait | Preference demanded | Priority | Selection Target |
|---|---|---|---|
| **Grain color** | White | ***Must have*** | Reach threshold |
| **Plant height** | More than 2.0 m, less than 4.0 m | ***Must have*** | Reach threshold |
| **Resistance to a disease/pest** | *Striga* resistance | **Important** | Reach threshold |
| **Adaptation to drought** | Flowering at a fixed date | **Important** | Reach threshold |
| **Yield** | Better than local, under poor soil fertility conditions | ***Must have*** | Maximize |
| **Fodder quality** | Improved digestibility of dry fodder | Nice to have | Opportunistic |
| **Grain quality** | As efficient as local varieties for home processing | ***Must have*** | Reach threshold |

'Must have' traits with fixed thresholds form the strictest selection criteria. Having too many of these, more than 4 or 5, will make it difficult to find plants with all traits present. 'Important' traits, and traits where the aim is maximization rather than a hard threshold ('absent' or 'present'), are less critical. They can be treated as a guide rather than a rule: a breeding objective of 100-120 days to maturity, does not mean plants that mature at 125 days must be rejected. Especially if other 'must have' traits are present.

The likelihood of achieving the combination of chosen traits in a variety can be increased in the early stages of PVD by:

- Understanding whether breeding objectives are realistic and can be realistically combined in a variety (breeder and farmers discuss this together)
- Choosing the parent varieties so that they have the 'threshold traits' for the new variety
- Creating and selecting breeding populations that have all 'threshold traits' at expected levels, and a good diversity of 'maximize' traits (use this as a guideline for early (2nd and 3rd) generation bulk and mass selection)

Breeding objectives may also change or be discarded over the course of a PVD trajectory, as farmers become more familiar with the breeding population and repeatedly discuss and weigh their preferences.

Making invisible traits visible through **Marker assisted selection (MAS):** *With the breeding techniques described in this guide, it can take many generations before knowing whether 'invisible traits' such as taste, nutritional value or disease resistance are present in a breeding population. This can lead to disappointment.*

*Marker assisted selection helps find these traits by scouring a plant for parts of a gene ('genetic markers') that we know are associated with the trait of interest. This can greatly increase the speed and accuracy of breeding efforts. If the marker is found, we know the trait is present and will be expressed at some point in the future. If it isn't, we need to go back to the drawing table.*

*MAS requires a laboratory and is costly, but its use can be justified if the breeding efforts are likely to benefit a larger population. It is important to stress that MAS does not change a plant's genes. It should not be confused with genetic modification.*

**When should specific traits be selected?** In the early generations, selecting for all breeding objectives at the same time may not be feasible. There will not yet be any plants in which all desirable traits are combined (and if they do exist, they may change in the next generation), and some desirable traits are not yet visible.

In addition to the guidance on 'threshold' and 'maximize' traits above, it is advisable to begin selection efforts by looking for clearly visible morphological traits (shape or form-traits) such as plant height, panicle length, ear size, grain colour, degree of scattering. These traits are 'monogenic' or 'oligogenic' – their behaviour is determined by a single gene or a few genes – which makes them simpler, more predictable, and easier to select.

Starting at Generation 4 or 5, when the plants in the population look more similar, we can start selecting and testing for more complicated traits, such as taste or disease resistance. These traits are 'polygenic' – meaning that their behaviour is determined by multiple genes that add up to produce a certain trait. Where monogenic traits are usually either present or absent, polygenic traits are present to a degree, along a continuum. They are less obvious to see and select.

In some cases, one breeding objective is of paramount importance. Drought tolerance or disease resistance, for example. In such cases an exception to the above guidelines should be made. For even if such traits are polygenic and will only be expressed to their full potential in later generations, it can be useful if early (2nd or 3rd) generations are exposed to severe drought or disease stresses and to continue breeding efforts with a maximum number of the surviving plants.

# V. Speeding up selection using more advanced selection methods

## For self-pollinators:
## Pedigree selection

Pedigree selection is the most practiced selection method for self-pollinated crops. Breeding progress is much quicker than with bulk selection but requires more work. This is important to realize: in pedigree selection each line (seed stemming from a single or, in our case, multiple similar-looking plants) is planted separately, monitored separately, evaluated separately, harvested, processed and stored separately, and documented separately. While breeding institutes often start with pedigree selection already at Generation 2, this is not feasible in an FFS setting because of the high number of pedigree lines that need to be selected (more than 100) and the amount of work and land this implies. We start with pedigree selection after Generation 4 or 5.

(In manuals and scientific literature, this combination of bulk selection in early generations and pedigree selection in later generations is also referred to as 'modified bulk selection' or 'mass pedigree selection').

## For cross-pollinators:
## Recurrent selection/progeny testing

Recurrent selection is a method to improve the quality of the breeding population in cross-pollinated plants. It can be done to speed up mass selection early on or halfway in the breeding process. For farmers engaged in PVD, such a 'technical' step is an opportunity for learning, while speeding up the breeding progress strengthens their motivation to continue.

If a single breeding objective is of overriding importance, recurrent selection can be done as early as the 2$^{nd}$ generation. The risk of doing this so early is losing important genetic diversity in the breeding population.
If, on the other hand, multiple breeding objectives carry equal importance, recurrent selection should be postponed until later generations, around Generation 4 or 5.

***Why do populations become more stable over time****? Around generation 4, repeated inbreeding has led to a high level of '****homozygosity****'. In plants, traits are governed by genes, with most crops inhering two versions of each gene, one from the mother and one from the father. Homozygosity means that both versions of a gene in a seed are the same (the opposite is* ***heterozygosity****, where the two versions are different). Once homozygosity is reached for a trait, it becomes 'fixed' – all offspring inherit the same genes and maintain the trait consistently from one generation to the next. In the field, increased homozygosity means that seeds selected from a well-performing parent plant are likely to produce offspring with the same desired traits.*

At this point, as the plants in the population become more stable, it makes sense to begin selecting individual plant types to develop into breeding lines. Plant types are sets of multiple plants that look and behave the same. (While it is possible to base breeding lines on the seed of single plants, the risk of losing too much diversity is high and does not weigh up to the advantage of increased purity or precision).

**In choosing how many pedigree lines to form, we are again faced with the challenge:** select too few plants and we lose too much genetic diversity for further selection, select too many and there will be too much work, requiring too much land and potentially not enough progress. Working with 25 pedigree lines is manageable for an FFS, but this number is enough only if the population is sufficiently stable. If this is not yet the case and variability is still very high, the FFS may choose to select plants for up to 50 pedigree lines to capture sufficient diversity.

The FFS now has two options: to reduce workload, poor performing "junk" lines can be discarded very early in the season, leaving only the better lines to monitor and select from. Or they can partner with another FFS nearby, that can evaluate half of the selected plants.

Alternatively, continue with bulk selection for one more season.

Steps involved in pedigree selection (starting at Generation 4):

Generation 4:

- Shift from bulk to pedigree selection: Perform positive selection (marking) to identify and select seed from the 25 best performing plant types in the segregating population. If necessary because of attractive variation, select more than 25 plant types and (optional) divide these

Recurrent selection, in the simple form described here, addresses some important shortcomings of mass selection in cross-pollinated crops. In mass selection:

i) when selecting seed, we only know the plant it came from: its 'mother's half'. The 'father's half' of the seed can come from pollen from any other plant in the plot;
ii) we do not know whether the combination of known mother and unknown father will produce a plant that is any good;
iii) it is difficult to select for traits that become visible after flowering: negative selection must be done prior to flowering and plants that show negative traits only after they flower will have already cross-pollinated with superior plants.

The difference with mass selection is that in recurrent selection *plants are selected not solely based on how they look (before flowering), but also on how their offspring (progeny) performs*. This involves testing the offspring in so-called *progeny tests*. This way, even if we do not know the source of the pollen (the father), we are still able to evaluate whether that pollen helped to produce a good or a poor plant. In other words, do father and mother combine well? From the seed that was selected from well-performing plants in the original (e.g. Generation 4 or 5) population, only the ones that test well and result in good offspring are allowed back into the breeding population.

Recurrent selection with progeny testing involves *3 steps*, requiring *2 seasons*:

1. Select seed from the 30 best-performing (mother) plants in the breeding population (more, or as much as possible, if doing this

over multiple FFS.
Bulk the seed per plant type, store separately and mark well.

Generation 5:

- Plant the seed from selected plant types in separate 'progeny rows' or mini plots (e.g. 3 rows x 15 hills). If 25 plant types were selected, we will have 25 rows or mini plots.
- Evaluate each mini plot up to the point of maturity/harvest. Discard plots that do not yield good plants. Mark and select the most desirable plants in well-performing plots.
- Select plants for maximum 25 pedigree lines. Always bulk the seeds of plants that look similar, whether they come from the same mini plot or different ones, as they will constitute a single line in the following season. The seeds of selected plants that look different from the types already present are saved separately and planted as a new line in the next growing season. *It is common for pedigree lines to separate into multiple new lines. It is also common for multiple lines to come together again into one.*

Generation 6:

- Plant 10 to 25 pedigree lines. Evaluate lines and mark and select superior plants from superior lines. Discard pedigree lines with undesirable traits.
- Select plants for up to 10 lines. Harvest more seed to allow for testing in larger plots and/or dissemination to other FFS at Generation 7.
  Because of continuing, but much lower, segregation, selection within the pedigree lines is still needed to make sure the best plants compose the pedigree line at the end.

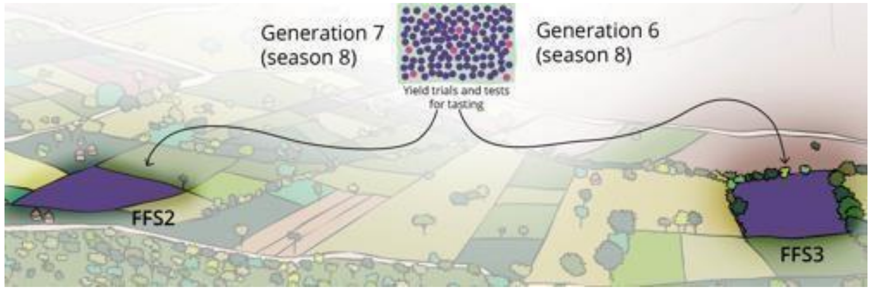
***Selection between and within pedigree lines:*** *pedigree selection is about choosing the lines that best fulfil our breeding objectives. But to bring them closest to those breeding objectives, selection within the lines must continue in generations 5 and 6, and until segregation stops entirely. Poor*

at a very early generation). Withhold half of the seed, keeping the seed of each selected plant separate. Mark and store these well.

2. Use the other half of the seed to plant: one row for the seed of each selected plant. Again, mark well, so that stored seed and planted seed of the same parent plant remain easily identifiable and can be matched.
   Evaluate each row up to the point of maturity/harvest and choose which rows should constitute the breeding population. (Since we won't be using the seeds produced in the evaluation plot for further breeding, there is no need to isolate the plot).
3. Go back to the seed that was withheld and *select only the seed lots that performed well in the progeny test* in the field. Bulk these seeds and grow them as the improved breeding population. Do NOT use the seed harvested from the evaluation plot, since these will have cross-pollinated!

Continue with mass selection in the new population.

Complete the process by conducting comparative trials for yield and breeding objectives (around Generation 7) and disseminating the population to other FFS for multi-location testing.



*plants in each line are removed, while only those plants are selected that show the traits that make that line (slightly) distinct from the other pedigree lines: plant height between 2 and 3 meters, 3-5 tillers, maturity in 100-115 days, etc.*

*This means that there* ***is no predetermined number of plants*** *to select per pedigree line, and* ***no fixed number of pedigree lines****. All lines that effectively meet (a subset of) the breeding objectives go to the next generation, with only those plants that look and behave alike.*

Generation 7:

- Grow up to 10 pedigree lines in slightly larger plots. The gradual reduction in the number of pedigree lines occurs because, with each generation, fewer lines excel in meeting the breeding objectives. Additionally, certain traits, such as aroma in rice, and disease vulnerability, emerge only from generation 5 onwards.
- Conduct preliminary yield trials and test for the other post-harvest breeding objectives, including cooking and eating qualities. Include a popular local control variety for comparison.
- Select only those lines that are promising enough to develop into new varieties and disseminate these ‘superior lines’ to FFS in other locations for testing in multiple locations.
- By this time, the plants in individual breeding lines will be near identical. Continue to improve the quality of the line through positive selection and by removing outliers.

Generation 8:

- If needed to achieve further uniformity, continue to improve the breeding lines by removing outliers.


*Pedigree selection in rice in Laos. The lines in the picture on the left are 4th generation, those on the right 5th generation. Notice the high uniformity within lines at generation 5, as well as the difference in earliness between the selected line and the other lines in the plot. Photo: SD=HS Whatsapp group.*

# VI. Completing the PVD process

With patience, skill, and some luck, participatory variety development yields a new variety.

Specifically, the steps outlined in this document produce 'open-pollinated varieties' (OPVs). OPVs have greater genetic diversity compared to the hybrids, 'pure lines', or 'pure varieties' often produced by commercial seed companies. For smallholder farmers, this has several advantages: by continuing to evolve, OPVs have the potential to adapt to changing growing conditions and increase resilience to pests and diseases. And even when pollinated naturally, by wind or insects, they maintain their characteristics well over time. This makes them very suitable for seed saving, helping farmers to break free from market dependence and secure a measure of sovereignty over their seeds.

***When is a PVD variety finished?*** In formal sector plant breeding, a variety is considered finished and new when it has been tested for "**DUS**" in multiple locations: it must be **D**istinct in its main characteristics from any existing varieties, exhibit **U**niformity across all plants within the variety, and be **S**table in the sense that traits do not change from one generation to the next.

It is similar in participatory plant breeding. Our selection work is done when:

i) the plants fulfil one or more of the breeding objectives (the trait(s) that make it *Distinct*);
ii) plants grown one season are similar to the plants from which the seed was selected in the previous season (*Stability*);
iii) while legislation differs from country to country, the *Uniformity* requirement for open-pollinated varieties developed by farmers often is less strict, for two reasons: farmer varieties are generally more diverse (heterogeneous) than formal varieties, which makes them more adaptable to real-life, variable growing conditions (in more mechanized market-oriented farming systems, farmers will require higher uniformity than in more subsistence-oriented farms). Secondly, it should be clear from the text in this document that uniformity requirements will be stricter for self-pollinated crops than for cross-pollinated crops, as the latter continue to cross-pollinate with nearby plants and, except for hybrids, show more heterogeneity between plants.

Once DUS tests are completed, a variety can be officially registered. In most countries, this is required before seed can be sold on the market. Some countries have simplified registration requirements for farmer varieties, which makes it easier for farmers to officially (legally) produce and sell seed of varieties created through PVD.