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Seed sources of agroforestry trees in a farmland context

- a guide to tree seed source establishment in Nepal

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Titel

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Cover photo

Breeding seedling seed orchard of *Dalbergia sissoo*. Photo: Erik Kjær 1998

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Abbreviations

TISC	Tree Improvement and Silviculture Component of NARMSAP
DFSC	(former) Danida Forest Seed Centre, now *Forest & Landscape Denmark*,The Royal Veterinary and Agricultural University, Denmark
NARMSAP	Danida/HMG Natural Resource Management Sector Assistance Programme, Kathmandu, Nepal
OECD	Organisation for Economic Co-operation and Development
EU	European Union
FAO	Food and Agriculture Organization of the United Nations
MPBS	Multiple population breeding strategy
BSO	Breeding Seed Orchard

1 Introduction

This document originates from an on-going cooperation between the authors on the appropriateness of different seed sources for procurement of trees to small holders. The document has been developed as a part of the partnership between NARMSAP (Danida/HMG Natural Resource Management Sector Assistance Programme) in Nepal, Danida Forest Seed Centre (since 2004: *Forest & Landscape Denmark*), and World Agroforestry Centre (ICRAF).

Definitions of tree seed sources have been developed and applied in many countries, and standardised (slightly differently) by OECD, EU, FAO, DFSC and others. However, the experience from 10 years of work with development and provision of better tree seed to farmers in Nepal has been that none of these definitions are fully suitable for tree seed sources in Nepal. We have therefore developed the present publication with the objective to

1. Develop and adopt definitions and classification suitable for the Nepalese farmland context
2. Provide a practical guideline for establishment and management of such types of tree seed sources in Nepal, and
3. Discuss advantages and disadvantages for the different seed source type.

The guideline consists of four sections: an introduction that introduces the TISC context, section two that introduces the Nepalese framework, in section three we define and discuss different options in terms of types of seed sources, and in section four we discuss some of the more specific details related to the establishment of planted seed sources.

The publication is based on our joint experiences gained in Nepal, on which we have formulated guidelines for the future work in Nepal. Although specific to Nepal we believe that some of the recommendations can be of general interest.

1.1 The TISC context

The Tree Improvement and Silviculture Component (TISC) under Natural Resource Management Sector Assistance Programme (NARMSAP) supports domestication of woody and multipurpose trees in Nepal by establishing genetically diverse seed sources. The seed sources are designed to produce offspring with properties desired by the farmer (vigour, fodder, fruiting or other, depending on species). Nepal has an advanced community forestry programme. In TISC's domestication strategies, farmers are identified as the major agents of domestication, because it is assumed that only involvement by community groups and farmers can ensure successful development and deployment of good seed sources of important tree species. One of the main functions of TISC is to support the development of a decentralised network of producers and customers of tree seed (Dhakal *et al.* 2001). The owners of the network are farmer associations/seed cooperatives with relatively little technical knowledge and financial resources.

Collection of wood and branches for household consumption and fodder in the tropical Terrai landscape. Chitwan, Sauraha. Photo: Erik D. Kjær.

The design and function of the seed sources should therefore be as simple as possible without compromising the possibilities for a continuous improvement of the genetic material delivered to the farmers. TISC's focus is on seed sources of multipurpose tree species. However, the domestication of valuable timber species is also considered to be important.

Definitions of seed source types have been presented by many authors (see e.g. Willan & Barner, 1989). However, these mainly apply to industrial plantation forestry and are not directly applicable to the situation in Nepal. In this document, we therefore develop and use definitions that in general should accommodate to the domestication processes in Nepal. Definitions that reflect a continuum of seed sources with increasing possibilities for improvement (and a correspondingly increased investment), ranging from seed sources in farmland and natural forest to seed orchards with control of the heredity of every single tree. However, it is not only a question of increasing the expected gain from improvement. Increasing the security of healthy plantings (i.e. reducing the risk of failure) is also a major objective. Given the extreme climatic variation within Nepal, this requires considerations on *which* seed sources can be used *where* (deployment guidelines). We therefore discuss the seed source types in *a planting zone context*, an aspect elaborated in the sections below.

2 A framework for working with seed sources in Nepal

Many different tree species (especially fodder trees) are planted by farmers and for many species it is likely that the demand will increase further in the future. Given the extraordinarily large ecological variation in Nepal, special attention should be given to ecological considerations. Different species and seed sources will be best at different planting sites and therefore the challenge is to *match species and seed sources to planting site*. Well-adapted seed sources may ensure a reliable yield for farmers, while maladapted seed sources may result in loss - or even total failure (Lillesø *et al.* 2001b).

Farmland tree species are used mainly for fodder and harvesting of fruits and are relatively fast growing, however, their genetic potential is underutilised. The genetic potential can be released to the benefit of farmers if a proper species-site and seed sources-site matching is performed, and if the species are brought into a careful but unsophisticated domestication process.

In the tree planting zones system mapped by TISC (Lillesø *et al.* 2001b), planting sites with similar environmental conditions are grouped together into zones (in some cases the same tree planting zone might appear physically separate) for which specific seed sources can be developed and thereby increase farmers planting success. The 'tree planting zones' can be recognised in the field by farmers and will be utilised where there is the greatest potential for seed demand. The system of 'Tree Planting Zones' will be part of a tree seed distribution system that will contribute to the improvement of the living conditions for the poor farmers (smallholders) in Nepal.

The farmers demand for fodder and fruit tree species in Nepal is of many species in small quantities, and the demand is therefore best fulfilled through a decentralised distribution carried out by farmer associations, seed cooperatives and private suppliers. In the short term the demand will have to be fulfilled by collection from trees occurring on the farmers' fields, while in the longer term the genetic potential of the species can be released through establishment of simple and intermediate breeding seed orchards (Dhakal *et al.* 2001; Lillesø *et al.* 2001b).

Simple level BSO of endangered Dalbergia latifolia at Teelkane, Chitwan in the Central Nepal. Photo: Lokendra Dhakal

2.1 Identifying species for domestication

Domestication occurs when humans plant trees and the planting material is not harvested in natural populations. Improved growth, quality and adaptation can be achieved by carefully selecting the best seed sources when raising seedlings for a given planting (Kjær, 1999).

Many of the species that are of potential interest to the farmers for planting have become rare in the natural forest and are only to an extremely limited extent available on the market (Lillesø *et al.* 2001a). The limited distribution of species and their non-availability on the market appear to have created a situation where farmers in a given area can only select among a relatively small number of the species that have potential for that area (Lillesø *et al.* 2001a). Because farmers in different areas use different species, the total number of species used by farmers is large.

There is no straightforward way of selecting high priority species for farmers for domestication when the market for seed is imperfect. The identification of species for domestication must be based on farmers' preferences and on information concerning selection of most preferred species from a wide range of available choices. A very important part of decentralised seed distribution is therefore to make a range of good planting material easily available on the market along with information on the species, i.e. to make the market more perfect and widen the available choices.

TISC will therefore use a combination of market surveys and preference studies, and let the market itself determine which species have a potential demand. In particular, TISC will support introduction of new seed sources and exchange of information between farmers (Dhakal *et al.* 2001). The farmers use a large number of species, and the strategy at TISC is therefore to develop seed sources for a correspondingly large number of species. It will be up to the market to decide whether these species in the long run will attract sufficient interest from the farmers.

3 Defining seed sources

The main purpose for TISC in classifying seed sources is to be able to provide users and seed source managers with information on the genetic quality of seed from different types of seed sources.

Ideally, the classifications should correspond as closely as possible to international standards in order to facilitate exchange of seed and information. The *'OECD Scheme for control of forest reproductive material moving in international trade'* (OECD Scheme) gives some definitions. The OECD scheme has the advantage of being generally recognised and implemented in many – mainly West European – countries. However, the OECD definitions are not well suited for species-diverse natural forests, and they are of little use for seed sources of trees that are scattered on farmland. Furthermore, TISC supports low input domestication programmes for many species rather than for a few traditional industrial tree improvement programmes. Therefore, TISC has developed a modified terminology that fits the Nepalese situation better (Table 1), as discussed below.

3.1 Selecting seed source types for the domestication programme

A fundamental part of the TISC Seed Programme is to support the creation of decentralised seed supply organisations and to introduce species into the seed supply systems, thereby letting the species prove themselves before supporting investments into BSOs.

All seed sources of indigenous species will start either as sources in the natural forest or as sources in the farmland. The most demanded species will be collected and planted as seed sources (for breeding and production) in collaboration with farmer associations/seed cooperatives. The effort put into each species will vary and different strategies are therefore applied depending on the importance of the species.

The challenge will be to develop an extensive programme that still can provide fast growing, healthy, genetically diverse and stable planting material for a large number of species. The workload for TISC should be realistic – even after a large number of seed sources have been established – and much of the responsibility for maintaining the seed sources should be in the hands of seed cooperatives or the Forest User Groups[1].

The types of seed sources that TISC has adapted within the framework of tree planting zones (Table 1) are described in more details below:

[1] Nepal has one of the most advanced programmes for participatory forest management in the world and a large proportion of the national forest has been handed over to Forest User Groups.

Table 1: TISC modified seed sources scheme.

Type	Location of seed trees	Normal ownership of seed trees	Genetic content	Timing of seed production	Criteria for classification as 'selected'
Farmland seed source - natural	In agricultural landscape Scattered trees remaining from natural forests, or natural regeneration from such	Farmers Public land	Local origin representing the original population in the area	Immediate production Seed production has a trade off with branch and leaf production	Number of trees Distance between trees All seed trees should be of acceptable quality
Farmland seed source - planted	In agricultural landscape Scattered trees Borderline tree around/ within farms Trees permanently intercropped Roadside trees	Farmers Public land	Local or non-local origin	Immediate production Production of seed has a trade off with branch and leaf production, and/or shade of agricultural crop	Number of trees Distance between trees Number of involved farms All seed trees should be of acceptable quality
Natural forest	Trees growing naturally in high forest or woodlands (pristine or subject to different degrees of human influence)	FUG Government Public land	Local origin Often influenced by human activity	Immediate production Seed production may be limited in some forests due to limited fruit set and difficult collection	Number of trees Size of forest Not degraded through over-exploitation All seed trees should be of acceptable quality
Plantation	Trees planted in a plantation or woodlot	Farmers FUG Government Public land	Trees with unknown origin	Immediate production	Number of trees Optional thinning Trees of good quality compared to plantings in the region
Provenance plantation	Trees planted in a plantation or woodlot	Farmers FUG Seed coop Government Public land	Trees with known origin (planted with seedlings from documented seedlot)	Production only after some years, quality can be improved further by selective thinning	Origin and quality of founding seed source Design and thinning.
Bulked BSO (simple level Breeding Seed Orchard)	Trees planted in a plantation, woodlot or maybe farmland with the purpose of seed collection	FUG Seed coop Government Public land	Planted exclusively with offspring from carefully selected trees Genetically diverse Documented	Production only after some years, quality improves further by thinning	Quality of founding seed trees and their origin Number of progenies Design and thinning
Family BSO (intermed. level Breeding Seed Orchard)	Trees planted in a plantation, woodlot or maybe farmland with the purpose of seed collection	FUG Seed coop Government Public land	Planted exclusively with offspring from carefully selected trees Progenies are kept separate in the planting Genetically diverse Documented	Production only after some years, quality improves further by thinning	Quality of founding seed trees and their origin Number of progenies Design and thinning Genetic quality based on quantitative genetic analysis

The purpose of the establishment of BSOs is to release the potential gain of the target species to the benefit of the farmers. The cost (TISC manpower and expertise as well as farmers' input) of releasing this gain should be minimal to enable inclusion of as many species as possible. This will inevitably involve a trade-off whereby only some of the total potential gain will be released. However, the realised gain, together with a transparent market, will be of immense value to the farmers.

3.2 Seed sources for immediate production

The seed programme should have an arsenal of seed sources that can on the one hand provide documented seed for the users in the short term, and on the other hand leave room for increasing gain by increasing investment in the seed sources for the species of highest priority. For the Nepalese tree seed programme we have defined three types of seed sources: farmland seed sources, selected stands in natural forests and selected plantations. All three types consist of trees that already are growing in the forests or open landscape, and can therefore be used for seed production right away. The 'establishment phase' thus merely requires development of appropriate documentation. An important objective of this documentation is to provide a basic '*source identification*' that is crucial for successful application of Nepalese Tree Planting Zones which shall protect against seed sources being mismatched to planting sites in the environmentally heterogeneous Nepal.

The seed sources must meet some criteria in order to be classified as *selected.* These criteria shall ensure that seed lots have good genetic variation and originate from good trees. These criteria are therefore discussed in more detail in the following sections.

Farmland seed sources

Many of the best fodder and fruit tree species in Nepal have over time become very rare in the natural forest and now mainly survive in the farmland within the agroforestry system.

Many species of trees are planted or grown on the same farm. One farm may therefore have seed trees from more than one species. We define a farmland seed source as a number of trees growing in the farmland, confined to an elevation belt in a geographical area region, all trees being in the same planting zone.

Farmland seed sources can consist of scattered trees remaining from natural forests (natural regeneration from such trees) or from planted trees. In the first case, the origin is known (= local), but the conversion of natural forest to agricultural land may have involved heavy selection: directly (e.g. cutting of trees with particular good stem form) and/or indirectly (natural selection following altered microclimate and/or effects on pollinating animals).

In the case of planted trees, the origin may sometimes be known, but most often this is not the case. For less planted, indigenous species (excluding *Dalbergia sissoo,* and *Pinus roxburgii*) the origin is probably mostly local, because indigenous species (other than *D. sissoo,* and *P. roxburgii*) are only found in very small quantities in nurseries supported by NGOs and other organisations (cf. Lillesø *et al.*, 2001a, based on a nursery survey in 15 districts from 1998). The genetic base may still be narrow for many planted farmland seed sources, because the trees can easily origin from seed collected on a few trees.

Many tree species in Nepal have often been planted by the farmer from cuttings/seeds/wildlings from an unknown (but most often small) number of mother trees, and in some cases the farmer therefore will be able to provide such information. However, it will often be difficult to infer on the genetic basis of specific trees, and often the information is lacking altogether. This is a serious complication when we want to ensure that the seed from the farmland seed source includes an appropriate level of genetic diversity, because strong relatedness between the farmland trees increases the risk of collecting inbred seed. Even if the trees are self-incompatible (cannot self-pollinate), it means that one may underestimate the required number of seed trees. The effective number of father trees (pollen donors, N_{ep}) is also likely to be smaller than would be anticipated from the actual number of trees in the area, and again this could lead to less genetic diversity in the seed crop than expected from the number of seed trees.

There is no simple and quick solution to the risk of high relatedness among trees on farmland. When planting new improved seed sources (see the sections on BSOs below), the problem can be handled though the design and breeding strategy, but for the source that must serve for immediate production one must accept that the seed may be somewhat inbred because trees may be pollinated by close relatives. Still, part of the problem can be addressed by including trees from a number of different farms. The idea behind this approach is that seed trees on different farms are likely to be less related than seed trees on the same farm, because it is assumed that farmers often have propagated trees by grafting (or seed collection) from older trees on their own or neighbouring farms. Collection of seed from a number of different farms therefore reduces the risk of procuring seed with a very low level of genetic diversity. It will not reduce the average level of inbreeding 'here and now', but will increase the level of diversity and thereby reduce the risks involved in using trees over a large area that represent a genetically narrow base. Also, the tree planting farmers may use their new trees for future seed collection. At that time they will benefit from being able to collect non-inbred seed because they planted trees from a genetically diverse seed source.

The problem with unknown relatedness of farmland seed trees is less problematic if these are of known origin. This will be the case if the farmland trees are remaining trees from the natural vegetation at the site, or if the planted seed trees are of known origin. The 'Number of farms' criterion does therefore not apply to such cases.

In order to be classified as *selected* seed source, the farmland seed source should meet a number of general criteria (Table 1 and 2). These include:

- Basic documentation
- A minimum number of healthy and vigorous trees that have been identified and marked
- Minimum requirements on the distribution of these seed collection trees - in order to ensure that seed is collected on trees that have other trees of the same species growing within pollination distance in order to reduce risk of collecting selfed seed
- In the case of planted trees: the seed trees in the farmland seed source should be 'good trees' as evaluated by local farmers using the species. In

practice, this criterion can be handled by defining ideotypes[2], based on farmers' description of 'good trees' of a given species in a given region.

- In case of unknown origin: seed trees should be distributed on several farms (*cf.* above) depending on normal propagation method.

Not all individual trees will flower every year, and some years some farmers may prioritise sale of seed less than fodder production (from fodder trees). As a matter of general rule 50 trees should initially be identified for each seed source in each zone in order to assure collection from at least 20-30 trees *(N_f)* which is required to maintain adequate genetic variability, (see Table 2).

The collected seed or planting material (from at least 10-30 farmers) should be mixed thoroughly in fairly equal proportions from each farmer to reduce the risk that customers get a mix of seeds from related mother trees (see table 2).

The above criteria shall ensure an acceptable genetic diversity in the seed lots. Genetic diversity in the seed lots will depend on a number of additional factors that are normally out of control of the seed collector including: N_{ep} (average effective number of pollen fathers per seed tree), variation between seed trees in their fertility (how much seed they produce), and the degree of relatedness between seed trees on the same farms (and between different farms). Based on some assumptions we have chosen the criteria in a way that we expect the seed lots to include an effective population size of 20 or more in most years[3]. The criteria are rather conservative in the sense that we expect higher levels of diversity in most years. Especially the number of pollen fathers is for safety reasons assumed to be low ($N_{ep} = 2$), cf. footnote 3 below. Most studies on pollen flow in trees would indicate higher N_{ep} (Smouse and Sork, 2004), but they are based on studies in forests, and we really do not have any studies that would reflect the situation of the Nepalese farmland. The reduction factor due to unequal seed production is set to 50%, based on experience from a number of other species (Bila, 2000, and Kjær unpublished).

Information and/or agreements between the owner of the seed trees, and the seed collector(s) regarding the future management of the trees in a seed source is important. This shall be in support of management that improves the quality of the seed source (e.g. letting only the best trees flower, or removing of inferior trees by cutting), and protect against management that deteriorates the quality. TISC will assist the seed cooperatives and farmers in preparing a seed source description and outline 'good management practice'.

At this stage, we have identified the following species for which classified farmland seed sources seems to be an appropriate strategy for procurement of good quality seed to farmers in Nepal: *Artocarpus lakoocha, Bauhinia purpurea, Bauhinia variegata, Boehmeria rugulosa, Choerospondias axillaris, Ficus auriculata, Ficus glaberrima, Ficus glomerata, Ficus hispida, Ficus lacor, Ficus neriifolia, Ficus semicordata, Ficus subincisa, Grewia optiva, Litsea cubeba, Litsea monopetala, Saurauia napaulensis.*

[2] Ideotype: Donald (1968) suggested the importance of having a model (ideotype) as a goal for breeding programs. This concept has had an impact on breeding programs and ideal plant types of many crops have been described (in Zaheer *et al.*, 1999).

[3] For this purpose we first estimate the effective population number as $N_e = 4*N_{ep}*N_f/(1+N_{ep})$, this estimate is then corrected with the (i)effect of unequal seed production from different seed trees (by reducing with a fixed factor of 0.5) and (ii) relatedness between trees on the same farm (by reducing with a factor between 0.2 to 0.5, depending on propagation technique). N_{ep} is the average, effective number of pollen fathers (here assumed to be 2), whereas N_f is number of seed trees collected from.

Table 2. General criteria for classification of farmland seed sources as selected

Species characteristics	Minimum number of selected, superior trees to be included in an 'improved' farmland seed source	Minimum number of trees that must have contributed to a given seed lot (N_f)	Minimum number of farms on which seed trees grow (N_{farms}) (only when origin is unknown)
Trees that farmers normally propagate by cuttings or grafting (e.g. Ficus) [4]	50	30	15
Trees that farmers propagate from either seed or cuttings/grating[5]	50	25	10
Trees that farmers normally propagate from seed from multiple sources[6]	50	20	5
Predominant pollinator	**Maximum distance from the seed trees to other trees of the species**[7]		
Beetles	100 meters		
Insects	200 meters		
Wind	200 meters		
Bats, birds, large bees	200 meters		
Fig wasps	1.000 meters		

Note: The criteria are designed in order to obtain a high level of diversity in the seed lot. They should therefore be seen as a 'minimum' - more trees will be an improvement. The guidelines are general recommendation that may need modification for each specific species as these vary in distribution, fertility and floral biology.

Stands in natural forest

From a seed source manager's point of view, natural forests differ from plantations in four important ways (Kjær, 1999), and it is therefore best to provide separate descriptions to selected stands in natural forests and in plantations.

(i) Neighbouring trees in natural forests may be siblings, which have germinated and grown next to each other in a gap formed by logging or natural disturbance – creating a family structure such that trees close to each other may tend to be more closely related than more distant trees. This is contrary to a plantation where family structure is less likely to occur (seeds have been mixed after collection), but where the planting on the other hand may originate from seed collected on few trees (depending on species).

(ii) The genetic origin of old plantations will often be unknown and it can not be assumed *a priori* that trees in a plantation will show long term adaptation to the local ecological conditions. Species in a natural forest are expected to have grown on the site for many generations, and are therefore adapted to local conditions.

(iii) Plantations consist of relative uniform stands with trees of equal age. Density is typically high, and the seed source managers will therefore be able to select the best trees out of a number of neighbouring trees. This selection is to some extent expected to reflect genetic differences (depending on sites, species and characters). Selection in natural forests, on the contrary, is expected to yield very moderate – if any – genetic gains (still depending on trait and species).

[4] Strong relatedness can be anticipated, and we therefore assume a reduction factor on N_e, $r = N_{farm}/N_f = 0.5$

[5] Fairly strong relatedness can be anticipated, we therefore assume a reduction factor on N_e, $r = N_{farm}/N_f = 0.4$

[6] Some relatedness can be anticipated, and N_e we therefore assume a reduction factor on N_e, $r = N_{farm}/N_f = 0.25$

[7] Based on Nason *et al* (1998)

(iv) Seed production and the ease of collection may be very different in plantations and natural forest. In some cases it may be more difficult to collect seed from the natural forests, whereas pollination may be critical in plantations.

In order to be classified as *selected* seed source, the natural stand should meet a number of general criteria (Table 1). A stand selected in natural forest should contain many, sexually mature trees with good form and health of the target species. Seed sources will preferentially be identified in Forest User Group (FUG) forests, and seed production should become part of the multipurpose production from the forest (seed production will rarely be the main use of the stand). The seed source may consist of several FUG forest stands within the same planting zone. The total number of fertile seed trees in the seed source should be at least 50 healthy and vigorous trees. Collection of trees of good quality (based on individual species preference, ideotypes) will be an advantage, but most important is that the trees on average look good. In the case of limited seed demand, seed should preferentially be collected from trees spread throughout the forest. Not all identified seed trees may flower every year but collection should be made from at least 30 trees in any given year.

In natural forest – where the natural history is not known – it is difficult to infer on the expected superiority of individual trees for important characters. For timber species, average form should be assessed to ensure that the stand has not been subject to severe negative selection.

At this stage, we have identified the following species for which classified natural forest stands seems to be an appropriate strategy for procurement of good quality seed to farmers in Nepal: *Acacia catechu, Albizia lebbeck, Albizia procera, Alnus nepalensis, Bombax ceiba, Cinnamomum tamala, Dalbergia latifolia, Fraxinus floribunda, Garuga pinnata, Michelia champaca, Phyllanthus emblica, Pinus roxburghii, Pinus wallichiana, Prunus cerasoides, Taxus baccata, Pterocarpus marsupium, Toona ciliata.*

TISC should collaborate with FUGs in carrying out inventory of the FUG forest area and to incorporate seed production into the operational plan of the FUG forests. The inventory will be part of the TISC seed source description. Seed production should be part of an integrated management of the stands generating multiple forest products (seed and silviculture).

At the time of selection an agreement should be made between the FUGs and the seed cooperative on future management of the trees. FUG-FUG networking for marketing and information sharing will be facilitated by TISC. TISC will also assist seed cooperatives and FUGs in preparing seed source descriptions and management.

Plantations – unknown origin

Trees planted in plantations may often serve as high producing seed sources where seed collection is easy. The origin of such plantations is often unknown.

Seed sources for exotic species can by their very nature not be established in natural forests, and species already established in plantations in Nepal (*e.g.*

Pinus patula) can serve as seed source for immediate production. Seed sources will preferentially be identified in Forest User Group (FUG) plantations.

In order to be classified as *selected* seed source, the plantation seed source should meet a number of general criteria (Table 1). Selected stands in plantations should be identified as healthy stands with trees of at least average quality as compared to other stands in the area. Upon approval of the stand for seed production, selective thinning may be applied.

A minimum area of one hectare where seed can be collected may be sufficient - provided it is known that the plantation was established from well-mixed seed of a good, representative collection. This size of area will ensure possibilities of collecting from 50-100 seed trees at a sufficient spacing (14-10 meters) even after thinning.

For small plantation stands from unknown seed source, it should be considered mixing seed collected from several stands within the same Planting Zone. This is parallel to the case with farmland seed sources. However, the problem is probably less pronounced for the plantations, as each consists of many trees. The likelihood that a plantation originated from seed collected from one tree (or few) will be lower for plantations with many trees, and for species with small seed.

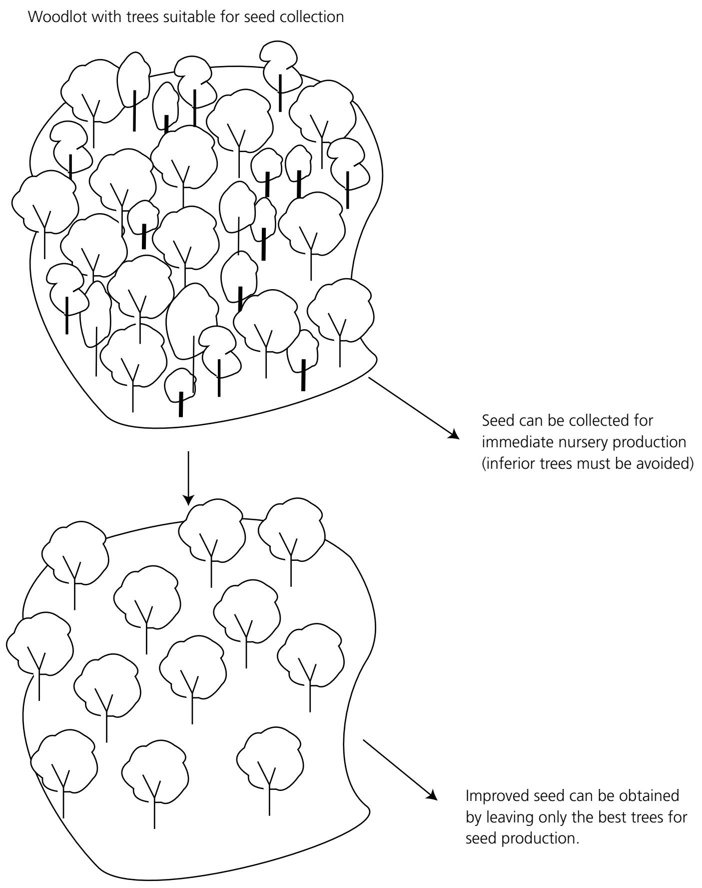

Figure 1. Seed sources for immediate production

Provenance plantations – known origin
Plantations with known origin can be a good seed source, if the origin is suitable and the plantation has been managed appropriately. If isolated from other trees of the same species, it can be an effective way to get access to large quantities of seed of exotic species from a smaller seedlot of superior origin. Also, as the tree stand has grown in Nepal for some time, natural selection may have caused some local adaptation of the seed source. If so, the seed stand can be superior to new introductions. If applied appropriately simple selective thinning regimes can further improve the quality for economic important traits.

Several exotic species have become popular among farmers in Nepal, but the origin of the available material is often unknown or may come from introductions with narrow genetic base (from few mother trees) (Granhof 1994). For many exotic multipurpose species it may furthermore be difficult to import seed from a specific (and large) number of mother trees - instead it may be possible to obtain bulked seed from specific provenances. Existing populations with known, well suited origin can be applied for seed collection right away, but most often such stands must first be established, and seed production will then only initiate after some years.

At this stage, we have identified the following species for which plantation seed sources seem to be an appropriate strategy for procurement of good quality seed to farmers in Nepal: *p.t.*: *Azadirachta indica (naturalised), Cryptomeria japonica (naturalised), Eucalyptus camaldulensis, Flemingia macrophylla, Leucaena diversifolia, Melia azedarach (naturalised), Morus alba, Pinus patula, Tectona grandis*.

For species where original introduction(s) is suspected to have limited genetic variation – but a landrace may have developed over some generations – collections should be carried out following the planting zone system. If possible, introduction of suitable material from abroad from reliable seed dealers could provide new genetic material.

3.3 Seed sources for breeding and production: Multiple Population Breeding Programme and BSOs

BSOs (breeding seed orchards) are plantings established with seed collected from a number of selected seed trees and mixed in a specific planting with the purpose of producing improved seed (see figure 2, page 22).

Seed orchards will only produce seed after some years, but will provide seed of significantly higher genetic quality (usually higher potential for growth).

Seed orchards should also be matched with suitable planting areas by using the Planting Zones map. However, development and application of BSOs can be more specific because it involves specific testing in so-called 'Multiple populations'. The idea behind the multiple population breeding strategy is to divide the breeding population into sub-populations that are kept separate throughout future generations of tree improvement in order to produce propagation materials adapted to different environments (MPBS, Namkoong *et al.* 1980).

The term Breeding Seed Orchard (BSO) was developed by Barnes (1984). It is a special type of seed orchard where seed production and breeding (testing) is combined. It can be a relatively simple and fast way of domesticating tree species, and adapted to different levels of investment depending on the type of BSO used, intensity of selection, and whether only within family variation is utilised or whether also between-family-variation is utilised.

The Multiple Population Breeding Programme in Nepal was initiated for *D. sissoo* in 1994 following a strategy agreed upon among government forestry institutions in 1993. The strategy was outlined by Gibson (1994) based on the so-called BSO concept and presented in Thomson (1994). In the following years TISC started to establish BSOs for multipurpose high priority species using two types[8] of BSOs as defined by Barnes (1984).

The unique feature of the BSOs compared to the other source types is that they consist of seed from selected trees, include selection of superior trees in the seed source, and include a level of diversity that will make them suitable for future breeding (the reason for calling them *Breeding* Seed Orchards). For the Nepalese situation TISC defines two types of BSOs:

- Bulked BSO (often referred to as simple level) with no family control. All families are bulked before planting in this type BSO
- Family BSO (often referred to as intermediate level BSO). The information about origin of each seedling is maintained through seed processing, nursery production and planting, and is documented in the establishment report. In the BSO, seedlings from a given mother tree are planted next to each other in identified plots. These plots are replicated several times thoughout the BSO (cf. section 4.3-4.5 below).

The genetic content of the two types of BSOs is in principle the same, and the activities in the mobilisation phase (identification of superior trees in forest/landscape, seed collection and documentation of seed collected) are the same. The differences between Bulked BSO and Family BSO refer to the nursery, establishment, and subsequent management and analysis phases, where the Family BSOs are much more complicated to handle as discussed in more details below.

In Nepal, focus is on development of decentralised programmes for decentralised production and distribution of tree seed to improve the livelihood of smallholders in Nepal. The key concept in this programme is that production of seed should mainly be carried out by farmer organisations, and the simplicity of the Bulked BSO is therefore an import asset. This is especially important considering the anticipated relatively large increase in the number of BSOs to be established, and also because of an increasing collaboration with farmer associations in the breeding programme.

[8] Barnes (1984) also described a third type: Intensive or advanced level BSO with controlled pollination. This type was not deemed cost effective for Nepal.

The Family level BSO has three advantages over the Bulked BSO:

1. Information on genetic parameters such as potential gain, genetic correlation between characters, expected effect of different selection intensity and methods, and genotype-by-environment interaction can be inferred by measuring the trees in the BSO and comparing the progenies' performance relative to each other (see e.g. Kjær *et al.* 2001).
2. The level of genetic diversity can be monitored and controlled because the relative contribution of each family remains known throughout the lifetime of the BSO (cf.section 4.2 below). This will be especially important as breeding proceeds over generations.
3. The gains from selection can be higher in most cases (especially low heritability traits).

The ability to gain information on genetic parameters (1) is an important advantage of the Family BSO over the Bulked BSO, because it makes it possible to estimate the genetic response on selection for different traits. One can compare gains from different selection scenarios and based on these estimates decide which trait to select for. The correlated response (changes in correlated characters due to selection for a given trait) can also be important to know.

Duer and Maarschalkerweerd (2004), for example analysed the Nepalese Family BSO of *Bauhinia purpurea* at Teel Kane (Terrai region of Nepal, established by TISC) in order to estimate and compare genetic parameters for growth rate and fodder values. Based on this study, they concluded that simple selection for fast growth is not likely to influence the nutritious value of the fodder, because fast growth and nutrient value had low genetic correlation. A further result was that breeding for nutrient value was not an interesting option: the genetic variation for this trait was low compared to the relatively high levels of genetic variation in biomass.

Hansen *et al.* (2005) analysed two Family BSOs of *Dalbergia sissoo* (in the Terai region of Nepal, established by TISC) in order to study the differences between natural seed sources from different parts of Nepal, and to quantify the genotype-by-interaction between the two BSO sites. They found that seed collected from different parts of Nepal performed differently in terms of growth, and that substantial genotype-by-environment interaction could be observed. Based on these results they could conclude that the applied multiple population strategy is appropriate and recommend maintaining separate breeding populations in future breeding generations.

Dhakal *et al.* (2005) studied mortality in the Nepalese Family BSO of *Dalbergia sissoo* at Sunari (Eastern Nepal) following heavy infection by *Aristobia horridula* beetles. They found that substantial genetic variation exists in resistance against infection, and that breeding could play an important role in combating the pest.

The three above investigations are examples on practical answers that could only be found because the BSOs have family control. However, this does not mean that all BSOs should be of the family type. This kind of information can be obtained as long as single (or a few) Family BSOs are available for each species. Based on genetic analysis of these few Family BSOs established and maintained by TISC, recommendations can be given on how to manage and select in Bulked BSOs of the same species established elsewhere, as part of a decentralised approach.

The decrease in genetic diversity due to phenotypic selection in Bulked BSOs cannot be monitored directly because there is no family control. However, the question can be addressed based on genetic estimates from a single (or a few) Family BSOs of the same species, and recommendations on selection can be issued based on such results.

Based on the above discussion, we find it valuable to combine the two approaches, mainly building on Bulked BSOs, but establishing and analysing some Family BSOs in order to gain knowledge and thereby be able to guide the future activities. The Family BSOs will also serve the important function of maintaining a core breeding population for breeding and gene conservation.

Bulked BSOs (= Simple level BSOs)
This type of (simple level) BSO is utilised for important and farmer planted species or species with large potential for future planting**.**

Seed for multiplication is collected from a number of selected (unrelated) trees. Bulked BSOs are established from the bulked seedlot (*i.e.* no family control). Seed handling and propagation of the seedlings is easier, and the establishment of the seed source is also less demanding. No complicated layout is required (layout in a square is convenient to minimise cost of fencing and protection), but it is important to plant the BSO at a homogeneous site in order to ease comparision between neighbouring trees and thereby increase the gains from the subsequent selections (selective thinnings). These thinning operations should be applied as careful phenotypic selection.

Seed should always be kept in separate bags during collection and seed processing, but will often be mixed prior to nursery production in order to ease the seedling production and avoid the cumbersome random mixing of seedling afterwards. However, in the case of low germination percent, one might consider growing the seedlings separated by family in the nursery, and only mix them afterwards before planting. This will allow one to compensate, if some families germinate much poorer than others.

At this stage, we have identified the following species for which simple level BSOs seems to be an appropriate strategy for procurement of good quality seed to farmers in Nepal:
Azadirachta indica, Acacia catechu, Bombax ceiba. Toona ciliata, Cinnamomum tamala, Albizia lebbeck, Albizia procera, Fraxinus floribunda, Dalbergia latifolia, Pterocarpus marsupium.

A large number of additional species are candidates for establishment in this kind of BSO in the future.

Registered Seed stand of Dalbergia sissoo at Lalbandi, Central Nepal. Photo: Lokendra Dhakal

Family BSOs (Intermediate level BSO)

Family BSOs are established with family control through the whole process of seed handling, nursery propagation, and overall selection combines within family and between-family-selections. The seed orchards can therefore be assessed for a number of traits prior to the genetic thinning, and the thinning operations are performed carefully, based on analysis of these data. This type of seed source demands relatively careful input for assessment and will not be standard for more than a handful of species (cf. discussion above).

Design, establishment and thinning of such seed sources requires specific genetic insight and computer skills. Also, the plots should be measured and analysed at regular intervals to determine which families to remove or retain and support additional information that can feed into breeding decisions. Therefore, this type of BSO is not suited for a completely decentralised approach. They can still be established in cooperation with FUG or cooperatives, but will require the know-how of TISC staff.

Inclusion of a number of families from other planting zones in a selected number of BSOs will be beneficial, because it allows estimation of the genotype by environment interaction, see Hansen *et al.* (2005). The planting zone system can thus be evaluated for the species involved allowing 'adaptive genetic management' as genetic insight increases over time. These extra families will be removed before seed harvest, especially if they show significantly poorer growth.

Furthermore, *between-family-selection* also requires a larger number of families to be included in the orchard. As a consequence, less expert-intensive options do not include *between-family selection*.

In most cases preferences will be given to designs that only include selection--between-trees within-families only (cf. below section on BSO design). The choice of whether or not to use *between-family-selection* in the BSO is a trade-off between expected gain from tree improvement versus availability of experts within the tree domestication programme. The main factor increasing the demand for expert input in *between-family-selection* in the seed orchards is

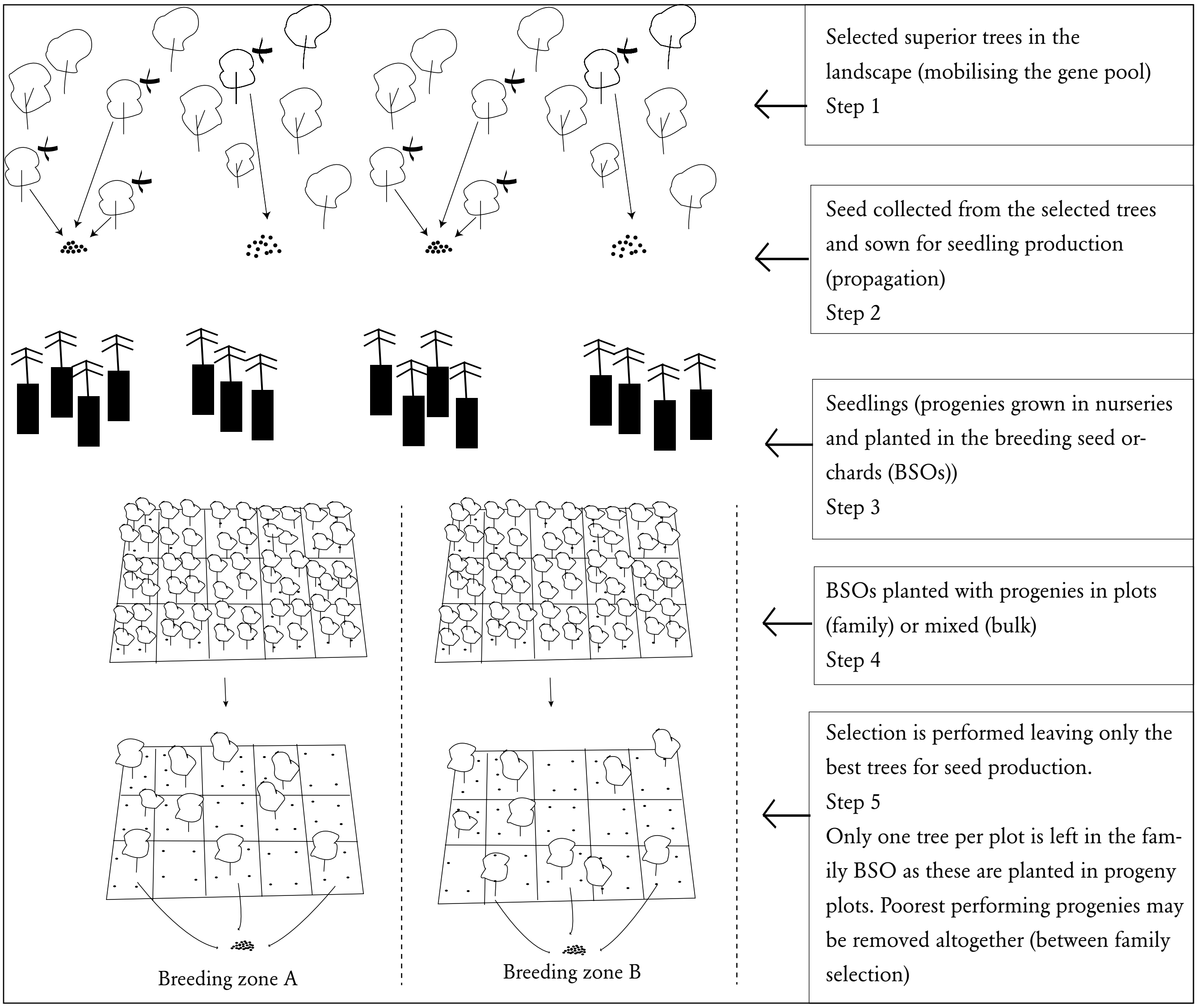

Figure 2. Breeding seed orchards

that because it requires careful measurement and expert analysis. Family BSO designed not to include *between-family-selection* (cf. the section below) *may* be assessed and analysed, but it is not required prior to the genetic thinnings, which makes things a lot simpler. Thinning operations can simply be performed based on careful phenotypic selection between half sibs in each plot.

Trained staff (TISC) will still have to design the layout of all Family BSOs, while measurement and analysis will not be a strict prerequisite when no families are removed.

At this stage, we have identified the following species for which Family BSOs (intermediate level BSOs) seem to be an appropriate strategy for procurement of good quality seed to farmers in Nepal:

Dalbergia sissoo, Bauhinia purpurea, Bauhinia variegata, Artocarpus lakoocha, Choerospondias axillaris[9]*, Ficus*[10] *auriculata, Ficus neriifolia, Ficus semicordata, Ficus subincisa, Ficus hispida, Ficus glaberrima, Ficus glomerata, Ficus lacor, Flemengia macrophylla, Guazuma ulmifolia, Grewia optiva, Litsea cubeba, Garuga pinnata, Saurauia napaulensis, Litsea monopetala, Boehmeria rugulosa, Michelia champaca.*

[9] *Choerospondias axillaris* is dioecious (having the male and female reproductive organs in separate individuals). Special attention to this special biology must be taken into consideration when designing BSOs for this species

[10] Specialised wasps (one wasp species for every *Ficus* species) pollinate *Ficus* species and some species are dioecious). Special attention to this special biology must be taken into consideration when designing BSOs for species in this *genus*.

4 Selection, design and management of BSOs

4.1 Selection of seed trees for initial mass propagation and/or future breeding

The number of trees used for initial seed collection should be of a certain size. Many future activities may depend on the genetic material sampled at this stage, and the initial collection should be considered a one-off investment that to a large extent determines the future quality. It is an important objective to maintain genetically diverse seed sources and therefore the number of families should in general not be less than 30-40 (Figure 2, Step 1).

The required number of seed trees is especially large, if it is anticipated that many families should be removed (combined *within- and between-family-selections*, cf. Figure 2, Step 5), because 30 families should remain after thinning. An initial number of at least 80-100 families is therefore required if family selection is to be applied.

30-40 seed trees may be sufficient in each breeding zone for Family BSOs with only *within-family-selection* because all families will be maintained for seed production and future breeding. Still, a larger number of families is desirable, if more intense domestication activities for some reason will be initiated in the future, *e.g.* if the species is given higher priority.

Seed collected for Bulked BSOs are mixed as described above. A higher number of families are required for this type of BSO because family control is not maintained and reduced diversity will not be observable. It is therefore recommended to start out with at least 100 families in bulk BSOs in order to mitigate the risk of reducing diversity following the phenotypic selections.

4.2 Calculation of effective number of seed trees

When preparing for the BSOs in the nursery it is also important to mix the progenies in approximately equal quantities in order to maintain a high effective population size (*Ne*) in the *BSOs without family control*.

If seed trees contribute with different amounts of seed to the total seed lot, then the effective population size will be smaller than the actual number of seed trees. In such cases a simple way to calculate the effective population size due to unbalanced contribution from seed trees is:

$$N_{ef} = 1/\ \Sigma\ p_i^2$$

Where, p_i is the percentage of seed (or seedlings) that each seed tree contributes to the total seed lot (planting stock).

For example: assume that 10 single tree collections contribute as follows:
tree 1: 20%, tree 2: 15%, tree 3: 12%, tree 4: 10%, tree 5: 10 %, tree 6: 8%, tree 7 : 8%, tree 8: 7%, tree 9: 5%, and tree 10: 5%

$\Sigma p_i^2 = 0.20^2 + 0.15^2 + 0.12^2 + 0.10^2 + 0.10^2 + 0.08^2 + 0.08^2 + 0.07^2 + 0.05^2 + 0.05^2$
$= 0.04 + 0.0225 + 0.144 + 0.01 + 0.01 + 0.0064 + 0.0064 + 0.0049 + 0.0025 + 0.0025 = 0.1196$
and
$N_{ef} = 1/ \Sigma p_i^2 = 1/0.1196 = 8.36$

The effective population size calculated this way should preferably not be less than 40, and collection from 50-60 trees or more is recommended.

For some species it may be difficult to find the recommended number of superior seed trees. In such cases specific decisions must be made as a trade-off between the desire for genetically broad material (future options), and the short term demand for selected planting material.

The actual level of genetic diversity (N_e) of the seed lot will be higher, because the seed from each single tree collection is sired by a number of fathers (cf. discussion in section 3.2 above). A fair approximation can be obtained from the formula:

$N_e = 4*N_{ep}*N_{ef}/(1+ N_{ep})$[11],

Where N_{ep} is the average, effective number of pollen donors per seed tree. Seed trees are collected carefully in the mobilisation phase, and N_{ep} will probably be larger than for farmland seed sources. Still, it will depend on the species and location of the selected mother trees. If one assumes $N_{ep} = 5$, then $N_{ef} = 40$ will correspond to $N_e = 4*5*40/6 = 133$. Alternatively $N_{ep} = 2$ will result in $N_e = 4*2*40/3 = 107$, i.e. a high level of diversity that can meet the requirement for diversity in the initial phases of a domestication programme.

The effective population number of the BSO will normally decrease during the subsequent selective thinnings. This is because the number of trees are reduced, and because some families may be more subject to thinning than others (families with lowest breeding value and therefore less superior phenotypes). In the Bulked BSOs this decrease cannot be monitored or controlled, because the family identity is not maintained. This calls for higher levels of genetic diversity (effective population number) in seed collections for Bulked BSOs in order to ensure that diversity is not reduced to a critical level during selection in present and future generation BSOs.

In Family BSOs, the decrease in genetic diversity can be calculated by applying the above formulas on the percentage of trees from different families remaining after thinning (including removal of whole families through 'between-family selection'). In this way, the effect of 'between-family-selection' can be evaluated before specific families are removed in all (or some of) the blocks (cf. section 4.6 below).

11 The effective population number N_e also depends on the actual number of trees to be planted in the BSO, but unless one is dealing with a very small BSO, the given formula should give a quite good estimate of *Ne*

Table 3. Six examples on design in BSOs given different (planned) thinning scenarios

A. Family BSO

Assumptions
Families = 100 Families to remain after thinning = 50
Trees left pr ha at age 10 = 200 Trees planted pr ha = 3300
Seed harvest starts 4 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	4	3300	1.5 x 2	Plant and check survival after 1. season	
2	4	3300	1.5 x 2	Assess and check maps	
4	1	825	≈ 3 x 4	Assess and leave only 1 best tree per plot	X
6	1	412	≈ 6 x 4	Remove 50 worst families based on calculation on both 4 and 6 yrs data	X
8	1	412	≈ 6 x 4	Maintain	X
10	1	206	≈ 7 x 7	Remove ≈ 50% worst trees maintaining 50 families	X
15+	1	100	≈ 9 x 9	Remove ≈ 50% worst trees maintaining 50 families	X

'Different scenario: no removal of families – seed harvest begins late (age 8)'
Assumptions:
Families = 50 Families to remain after thinning = 50
Trees left pr ha at age 10 = 200 Trees planted pr ha = 3300
Seed harvest starts 8 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	8	3300	1.5 x 2	Plant and check survival after 1. season	
2	8	3300	1.5 x 2	Assess and check maps	
4	4	1650	≈ 3 x 2	Leave only 4 best trees per plot	
6	2	825	≈ 3 x 4	Assess and leave only 2 best trees per lot	
8	1	412	≈ 6 x 4	Leave only 1 best tree per plot	X
10	1	206	≈ 7 x 7	Remove ≈ 50% worst trees maintaining 50 families	X
15+	1	100	≈ 9 x 9	Remove ≈ 50% worst trees maintaining 50 families	X

'Different scenario: Seed harvest begins early (at 2 years) and trees growth large within 10 years'
Assumptions:
Families = 50 Families to remain after thinning = 50
Trees left pr ha at age 10 = 100 Trees planted pr ha = 3300
Seed harvest starts 2 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	4	3300	1.5 x 2	Plant and check survival after 1. season	
2	1	825	≈ 3 x 4	Assess and leave only 1 best tree per lot	X
4	1	412	≈ 6 x 4	Reduce ≈ 50% worst trees maintaining 50 families	X
6	1	206	≈ 7 x 7	Maintain	X
8	1	206	≈ 7 x 7	Maintain	X
10+	1	100	≈ 10 x10	Remove ≈ 50% worst trees maintaining 50 families	X

B. Bulk BSO

Assumptions:
Trees pr ha at age 10 = 200 Trees planted pr ha = 3300 Seed harvest starts 4 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	No plots	3300	1.5 x 2	Plant and check survival after 1. season	
2	No plots	2475	≈ 2 x2	Remove ≈ 25% worst trees	
4	No plots	1650	≈ 3 x 2	Remove ≈ 33% worst trees	X
6	No plots	825	≈ 3 x 4	Remove ≈ 50% worst trees	X
8	No plots	412	≈ 5 x 5	Remove ≈ 50% worst trees	X
10	No plots	206	≈ 7 x 7	Remove ≈ 50% worst trees	X
15+	No plots	100	≈ 10 x 10	Remove ≈ 50% worst trees	X

'Different scenario: Slower growth'
Assumptions:
Trees pr ha at age 16 = 200 Trees planted pr ha = 3300 Seed harvest starts 8 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	No plots	3300	1.5 x 2	Plant and check survival after 1. season	
4	No plots	1650	≈ 3 x 2	Remove ≈ 50% worst trees	
8	No plots	825	≈ 3 x 4	Remove ≈ 50% worst trees	X
12	No plots	412	≈ 5 x 5	Remove ≈ 50% worst trees	X
16	No plots	206	≈ 7 x 7	Remove ≈ 50% worst trees	X
25+	No plots	100	≈ 10 x 10	Remove ≈ 50% worst trees	X

'Different scenario: Selection for fruit trait can only be done after 8 years as the trees need to start fruiting before theys can be evaluated and best trees selected'
Assumptions:
Trees pr ha at age 12 = 160 Trees planted pr ha = 3300 Seed harvest starts 8 years

Age Years	Trees per plot	≈ Trees Pr ha	Spacing (m)	Operations	Seed collection
0	No plots	3300	1.5 x 2	Plant and check survival after 1. season	
2	No plots	1650	≈ 3 x 2	Remove ≈ 50% according to vigor and health	
8	No plots	750	≈ 3.7 x 3.7	Remove ≈ 50% according to fruit set and size	X
10	No plots	325	≈ 6 x 5	Remove ≈ 50% according to fruit set and size	X
12	No plots	160	≈ 8 x 8	Remove ≈ 50% according to fruit set and size	X
15+	No plots	100	≈ 10 x 10	Remove ≈ 50% according to fruit set and size	X

Important note: BSOs should be designed and thinned in ways that fit the individual species. Fast growing species can require earlier and stronger thinnings compared to slower growing species. Species selected for fruits can require delay of the thinning until fruiting starts in order to allow selection for fruit characteristics, and so on. Therefore, the above six scenarios are only examples for inspiration. It is strongly recommended that the seed source establisher, prior to establishment, considers how to thin the seed source before it is established so the seed source can be designed accordingly. This is especially important for seed sources with family control. Lack of seedlings can also lead to different scenarios reducing the intensity of thinning (see text for details).

4.3 Plot size, shape and spacing in the seed orchards.

Family BSOs are established with progenies from selected single trees in replicated plots. Selection within plots reduces the number of seedlings per family to one, but between-family-selection is only anticipated in the Family *BSOs* (*with family control, design for within and between-family-selections*).

The plot size in the *BSOs with family control - within and between-family-selections* is a trade off between precision in family estimates (that points towards small plots) and gain from selection within families (that are increased in large plots), see e.g. Barnes (1984).

4.4 Calculation of plot size in Family BSOs

In the Nepali Family BSOs, only moderate between-family-selection is planned. This directs/points towards large plots. As an extreme, the maximum plot size will be limited by the following management decisions:

1) Number of initial families and number of families after genetic thinning.
2) Target number of trees per ha to remain for seed production after thinning
3) Initial spacing

For example: Assuming that 100 families are to be established, and 40 families should remain after the genetic thinning. Also assuming that 200 trees/ha should remain for seed production. Initial spacing is assumed to be no less than 2m x 1.5m = 3 m^2 per tree.

Calculation: 200 trees per ha from 40 families remaining after genetic thinning => this give 5 trees per family => as each family should be represented once in each block, a minimum of 5 blocks is required => 1 ha should consist of 5 replications (blocks). Each replication should therefore be = 10.000 m^2/5 = 2000 m^2. Initially, 100 families are established per block, i.e. each block should contain 100 plots. Plot size will therefore be 2000 m^2 /100 = 20 m^2. With spacing of 3 m^2 per tree this will allow approximately 7 trees per plot.

There may be two important practical reasons to establish smaller plot sizes:

Too few seedlings. Sometimes the number of seedlings is a limiting factor. This is a typical problem in species having large fruits. A dense spacing of the seedlings from the selected trees may result in a small seed orchard that cannot produce sufficient amounts of seed. As a consequence, it may be preferred to establish a larger seed production area – even if it compromises the optimal layout for *within-family-selection* (and therefore reduces the potential gain from selection at this stage).

Early seed production. Some species begin seed production after only a few years (as e.g. *Bauhinia purpurea* starting after 2 - 3 years). With e.g. 16 tree plots (48 m^2/plot, at 1.5 x 2.0 m spacing), the number of trees must e.g. be reduced

Inventoring the Dalbergia sisso, intermediate level BSO in Kapil Bastu district of western Nepal. Photo: Lokendra Dhakal

to 1/16 before seed collection. If seed production is initiated after 2 years, then a reduction by 15/16 of the stem number will be required in order to be able to collect the seed (only one tree per family must remain in each plot to avoid inbreeding). Sudden opening-up of space might also cause danger of uprooting the young trees during a storm. Furthermore, a small number of trees per ha may result in an inefficient utilisation (diminished seed production) of the area as the trees cannot cover the area. In such cases, the plot size should be reduced (and the number of replications instead increased).

Environmental heterogeneity. Big plots are useful as they allow strong *within-family-selection* by comparing trees grown close to each other and therefore grown under very similar environmental conditions. However, if plots get too big then heterogeneity in soil *etc.* within the plot can cause environmental variation within the plot to an extent that weakens the effect of selection. Thus plot sizes should be sufficiently small to avoid any significant environmental variation within a plot.

Yet other factors may prove important for some species like the dioceous[12] *Choerospondias axillaris*. This species is widely planted on farmland for income generating fruit production. It is not possible to directly thin for better fruit quality (nor for sex) before the trees are mature, and therefore (all other things being equal) such species should be planted initially at a larger distance between families.

Uncertain future management regimes. Family BSOs are established with plots that contain half-sib families. Consequently, seed should not be collected before the BSOs are thinned to 1 tree per plot as described above. It can be argued that single tree plots should be considered, when the future management regime for some reason seems uncertain, because seed then can be collected without thinning at all. This means that BSOs based on single tree plots still can be used if e.g. the plot demarcation is not maintained or thinnings for some reason are not performed. The BSO type where seed from selected plus trees are mixed prior to establishment (Bulked BSOs) could also be considered as an option in situations where future management regimes are uncertain.

[12] With separate male and female trees

4.5 Block and plot shape depending on contours

The size and shape of the blocks are important - especially when designing Family BSOs where between-family-selection is planned. In this type of seed orchard, it is important to design the blocks in such a way that they cover an area as uniform as possible. Variation should be *between* blocks and not *within* blocks.

Plots should also be as homogeneous as possible. Site variation should to the extent possible be excluded from the plots. Low environmental variance in the family plots is a pre-requisite for obtaining moderate to high heritabilities, and therefore a pre-requisite for obtaining gain from the *within-family-selection*.

A compact, square plot can result in low within-plot variation on flat lands (at least 3 m^2 per tree is to be maintained). However, many of the future BSOs in Nepal are to be established on sloping land. In such situations it will often be recommendable to establish line plots following the contours of the landscape and with each block having minimum possible altitude differences.

Left: Chlorospondia axilliaris BSO two years after establishment (25 progenies, selection criteria: fruit) Right: Michelia champaca BSO two years after establishment (40 progenies, selection criteria: timber). Both BSOs are established on sloping land at Thankot, close to Kathmandu. Square plots are often to be preferred in flat landscape (compact and therefore minimum heterogeneity), but line plots will often provide more homogeneous plots in sloping landscapes. Photo: Erik D. Kjær.

4.6 Genetic thinning and formation of future generations

All seed orchards must be carefully thinned by selecting for the best traits of an individual tree (as described above). Criteria depend on the given species and especially its use.

Some species are difficult to thin at an early age. An example is the fruit tree species where superior trees cannot be identified until fruit production has initiated.

For most agroforestry species, phenotypic selection will be based on so-called ideotypes. These 'ideal tree types' are identified together with farmers who have experience in using the different species. For example, trees with many branches are often preferred by farmers in comparison to more straight trees, as the branchy types in general give a larger amount of fodder. Fodder value of the leaves is of course important for many species but is difficult to select for, as it would require expensive laboratory testing of all trees.

Selection *between-the-families* in the Family BSOs must be based on 1) formal assessment, 2) analysis of data and 3) calculation of breeding value. Assessment and analysis of data can easily become a bottleneck in seed source management activities, and care should therefore be taken not to establish too many BSOs that later will require resource consuming assessment, analysis and genetic thinning. If so, the seed orchards may not be timely thinned, and both gain and seed collection opportunities therefore lost. As described above, it is much easier to perform the *within-family-selection* as this is simply done by selecting the best trees within each plot. At present, *between-family-selection* is applied only for *Dalbergia sissoo*. Establishment of BSOs for *between-family-selection* might be established for other species in the future if important, low-heritability traits are identified.

4.7 Selection for next generation BSOs

A BSO can be said to be only the first step in a breeding programme. In such cases the BSO is called a *first generation* BSO, indicating that successive generations of BSOs can be established in the future based on material from previous generation BSOs. Generation turnover requires that trees in the first generation BSOs flower (produce progenies). Establishment of second generation BSOs – in order to form a new and more improved seed source – must therefore wait until flowering of the first generation BSOs.

This kind of thinking – establishment of new BSOs based on seed collection in previous generation BSOs – is based on the idea of so-called recurrent selection that is the core basis for most breeding programmes. The idea is that a moderate, but substantial, gain can be achieved by selection in each generation. However, these 'gains per generation' will accumulate over generations allowing a progressive gain over time.

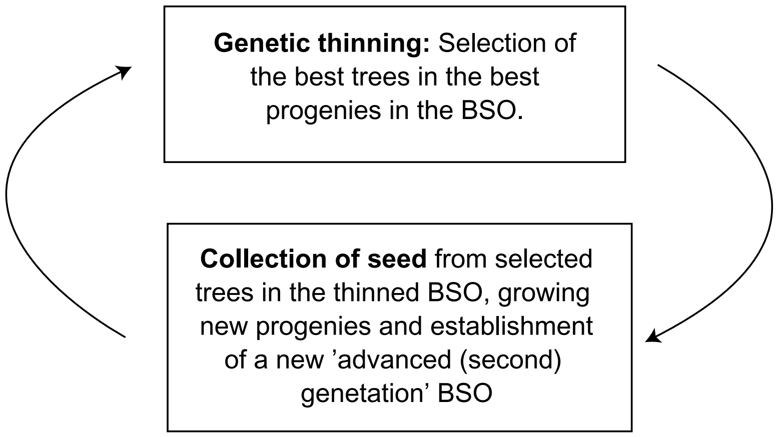

Figure 3. Recurrent selection

BSO of Albizzia lebbeck two years after establishment (36 families) established on flat lands at Teelkane, Chitwan. Photo: Erik D. Kjær.

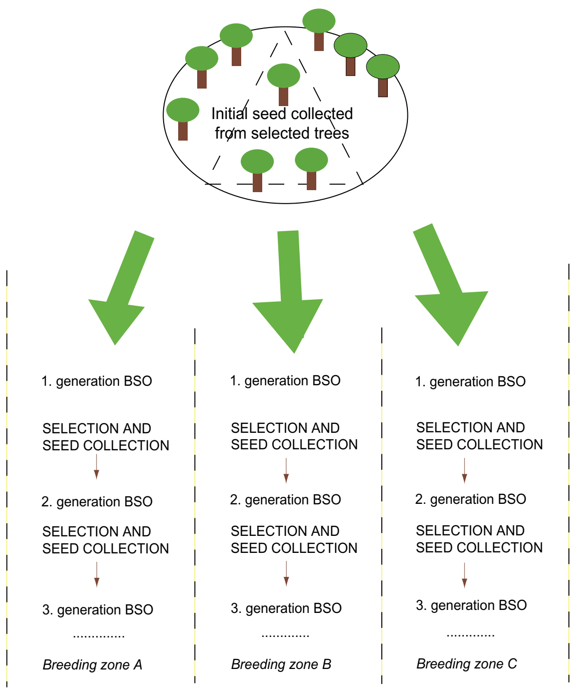

Figure 4. Development of multiple breeding populations from a core of selected plus trees

Introduced Leucaena leucophala x diversifolia hybrid clones (two years old) growing at Teel Kane, Chitwan. Fast growing species such as leucaena requires different thinning regimes than slow growing species. It is a good idea to consider the expected thinning regimes prior to establishment of the seed source in order to adapt initial spacing and plot sizes accordingly. Seed collection from clones (such as the present leucaena clones) is a complicated matter to be considered carefully. One therefore needs to look into the number and origins of the clones before deciding on their suitability for seed collection. Photo: Erik D. Kjær.

Gains are obtained in every breeding cycle (generation), and the progress in terms of genetic improvement will therefore continue over time. 'Gain per time' will not only depend on the gain per selection round, but also on the length of generations. Consequently, gain per time can be especially high for species flowering at an early age. Still, there are limits to the generation length other than flowering age. First of all, the superior trees cannot be selected until they reach an age where important traits are revealed. Secondly, very rapid generation turnover will be very resource demanding in terms of testing and seed source management.

For many species the 1st generation Nepali BSOs established in the different breeding zones have been based on the same initial collections (*i.e.* the same plus trees). However, selection taken place during the first generation growth in the different BSOs may have caused local adaptation to the different environments. Thus, second generation BSOs should be planted in the same breeding zone as the first generation BSOs from which improved seed is harvested (see figure 2). This is the basic idea of a multiple population improvement programme, where each population develops into a unique breeding population adapted to local conditions.

The considerations concerning diversity discussed in section 3 above are also valid for generation turnover, but additional concern should be given to relatedness between trees in future generations. An important advantage of *BSOs with family control* (with family identity) compared to *BSOs without family control* (without-family-identity) is that the build of relatedness over the generations is easier to control when family identity is maintained.

The generation turnover is discussed specifically for *Dalbergia sissoo* in Dhakal *et al.* (2004).

4.8 Design of advanced generation Seed Orchards

The design of the next generation BSOs follow the ideas of the 1. generation BSO, only will it now be important to incorporate any family structure in order to reduce any effect of inbreeding. Consequently, progenies from the same single trees (i.e. that originate from the same 1st generation family) should not be planted next to each other in the new BSOs. When selecting mainly within-families, the number of trees that originate from the same families will be minimised, but co-ancestry will eventually build up over the generations, and it is therefore important to keep track of the relatedness in a systematic manner. This can be done by assigning an identity number to each selected seed tree, and ensure that their relationship is carefully recorded (relevant for *BSOs with family control*). An example of a practical recording system is found in Kjær (1999).

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