Teacher's notes 1.2

Talbotiella gentii: genetic variation and conservation

David Boshier, Daniel Dompreh and Mike Swaine
Acknowledgements

The editors of this Forest Genetic Resources Training Guide wish to thank Jarkko Koskela and Barbara Vinceti for their contribution in identifying the need for the guide and for their continuous support during its preparation. We acknowledge the important advice from a reference group of scientists at Bioversity International - Elizabeth Goldberg, Jozef Turok and Laura Snook - who at various stages supported this project.

This training guide was tested during several training events around the world. We would like to acknowledge the valuable feedback received from many students and their teachers, in particular Ricardo Alía and Santiago González-Martínez from the National Institute of Agriculture and Food Research (INIA), Spain and Peter Kanowski from the Australian National University.

We would like to give special thanks to Thomas Geburek, Department of Genetics, Federal Research and Training Centre for Forests, Natural Hazards and Landscape (BFW), Vienna, Austria, for his review of the Case studies presented in this module. His valuable feedback led to important improvements of the module.

The photos in the PowerPoint presentation are the copyright of Kenneth Anyomi, David Boshier, Daniel Dompreh, Thomas Geburek, William Hawthorne, Mike Swaine, ‘New Scientist’, Royal Botanic Gardens-Kew and United Nations Environment Programme (UNEP).

Finally, the production of the Forest Genetic Resources Training Guide would never have been possible without the financial support of the Austrian Development Cooperation through the project, ‘Developing training capacity and human resources for the management of forest biodiversity’, implemented by Bioversity International during 2004-2010. We would also like to thank the European Commission funded “SEEDSOURCE” project for additional financial support.

All cover illustrations were drawn by Rosemary Wise and the layout was created by Patrizia Tazza. We are grateful for their beautiful work.
MODULE 1
Species conservation strategies

Teacher’s notes 1.2

Talbotiella gentii: genetic variation and conservation

David Boshier, Department of Plant Sciences, University of Oxford
Daniel Dompreh, Mike Swaine, University of Aberdeen

Introduction

These Teacher’s notes aim to assist teachers in using the Case Study 1.2 Talbotiella gentii: genetic variation and conservation in classes. The notes:

• describe the key concepts covered in the case study, with references to forest genetic resources textbooks where explanations can be found (full references at the end of these notes).
• give tips on how to prepare and run the exercise and discuss the main learning points (genetic and other) that students should be able to derive from the case study.
• give an outline commentary to the PowerPoint presentation which is used to introduce the case study to the students. The presentation contains pictures of the species, reserves where it occurs, relevant land-use issues in the area, and figures/tables from the exercise.

The PowerPoint presentation, along with the Case Study can be found on the accompanying DVD, or at the Forest Genetic Resources Training Guide webpage at www.bioversityinternational.org

Key concepts to cover/introduce in this case study

General conservation

Genetic concepts
• Genetic processes associated with small/fragmented populations – increased genetic drift, bottlenecks, increased inbreeding and consequently homozygosity and inbreeding depression: see FAO et al. (2004a) pp 43-44; Finkeldey (2005) pp 75-76.

How to run the exercise

The exercise can be run in a number of ways depending on the time available and size of the class. The exercise works best if students work in groups of 4-5 (no more
than six). It is best if the students have already read the case study before they start the exercise. This way valuable class time is not lost with students reading the paper during the class. So give the case study out in a previous class with instructions to read it before the next class. It perhaps goes without saying that it is vital that the teacher and any assistants are fully familiar with the whole text.

NB: the exercise is set in the context prior to the year 2008, both in terms of the species status and country profile. As such, more recent information and changed contexts are not included as they are not relevant to the exercise.

Ideal number of students: 4-15

Ideal length of class: 3 hours, broken down as follows:

**Introduction**: use the PowerPoint. The commentary below gives the main points when presenting to the students - approx 20 minutes.

**Group work**: suits 1-3 groups of 4-5. Each group devises a strategy, but tends to take a different approach, such that different issues are raised and overall, most points are covered. Students discuss the case study amongst themselves, responding to the specific points and developing their strategy. The teacher should be around to answer any queries the groups have. However, it is not essential that all of the time is spent with the whole class together with the teacher. Once the teacher and groups are happy they understand the assignment and issues, each group could meet, discuss and prepare the strategy outside of class time - 1.5 hours.

**Presentations**: each group presents its strategy verbally to the class (supported by main points written on large paper or in a PowerPoint presentation) – 10 minutes per presentation, with 5 minutes after each presentation for questions/comments by the rest of the class and teacher.

**Final discussion**: led by the teacher. Allows the teacher to make general comments about what was good, what was missed, etc. – 10 minutes.

**Background information**

**PowerPoint**: about 20 minutes to go through. This allows explanation of some specific issues raised in the case study students’ notes.

**Slide 2** - shows flowers, fruit, leaves of *T. gentii* and its ability to sprout from the trunk.

**Slide 3** - aerial photograph and map show Ghana’s forest reserves and location of *T. gentii* populations.

**Slide 4** - map from exercise showing species distribution and location of specific populations. Draw attention to extinct and extant populations and the increased gap in the distribution produced by human deforestation.

**Slide 5** - general view looking to the southern scarp of the Akwapim-Atewa hills.

**Slides 6 to 7** - photos of specific *T. gentii* populations. They illustrate the isolation of the forest on the hills and lack of forest on the plains.

**Slide 8** - shows the population at Yongwa in flower, light pink crowns just below the cliffs, not the red flowered trees. Also an internal view of the forest.

**Slides 9** - summarises the loss of stands/populations of *T. gentii* in recent years.
Photo shows cutting for firewood at Sapawsu.

**Slide 10** - shows deforestation in one population of *T. gentii* in recent years.

**Slide 11** - shows the Botanical Gardens at Aburi in Ghana.

**Slide 12** - covers alternative objectives for conservation. The teacher must stress the need for students to first define an objective for their conservation strategy. It is very common for students to not define a strategy or to forget to explain what it is. Without this, it is impossible to judge the efficacy of a conservation strategy. Objectives should be pertinent and realistic and avoid the "operation was a success, but the patient died" syndrome, i.e. we carried out all the activities successfully, but it did nothing to help.

**Slides 13 to 14** - cover the issue of population size – relate these to population sizes of remnants in Table 2 and how overlapping generations mean the effective population size will be lower than the census number.

**Slides 15 to 18** - summarises different approaches to conservation and associated problems. The emphasis should be on their complementary nature, rather than either/or. The emphasis will, however, shift depending on the characteristics of the species and the population of concern.

**Slide 19** - allows explanation of the correspondence analysis diagram in the exercise, i.e. which populations are more closely related genetically to each other (broken lines enclose samples from the same population). The text mentions a high level of genetic differentiation between the populations. The teacher can point to the idea that this can help in prioritising which populations to conserve. However, the teacher should emphasise the point made in the text that failure of samples from some locations to group genetically with other geographically close locations is likely to reflect sampling/bottleneck effects from both the small sample and actual sizes of some remnant stands, rather than any true genetic differentiation.

**Slide 20** - allows explanation of a genetic bottleneck, where alleles/diversity are lost through sampling, effects, but also through the increased impact of genetic drift. Differentiation increases between populations that were previously similar.

**Slide 21** - allows explanation of the significance of Table 3 for the exercise i.e. larger populations show a higher percentage of polymorphic loci (e.g. Abiriwapong 16.9%, Yongwa 13.6%, Chalet 8.4%) compared with smaller populations. Although the data are clearly biased by the unequal and low sample sizes, in the case of some small populations this represents a 100% sample (e.g. Botriansa 2.1%, Senkyeso 2.1%, Hospital 0.8%) and is therefore a true reflection of low genetic diversity in these remnants, most of which no longer exist.

**Slide 22** - allows explanation of the significance of Figure 3 for the exercise i.e. controlled pollinations between trees from different populations show increased fitness (expressed as fruit set in Fig. 3) with increased geographic and genetic distance between parents. This is key to deriving a conservation strategy. The improved seed/seedling performance with increasing genetic/geographical distance supports the idea that *T. gentii* populations have been affected by increased fragmentation and reductions in size, leading to increased selfing and inbreeding depression.

**Slide 23** - allows the teacher to go over what the students should be doing in the exercise. The following should be stressed: a) the need to be specific in what the strategy includes. Students tend to be too general in their recommendations; b) the need to prioritise. Students tend to recommend doing everything, failing to
recognise resources for activities are extremely limited; c) Students should indicate what information/evidence they have used to justify each recommended action; d) students need to present a convincing case that would sway a donor/government to give them funds and/or enact policy/legislation to conserve the species.

Important points to cover in students’ strategies and to draw out in discussion

Comments about the questions

• How is human disturbance likely to have shaped levels of genetic diversity in T. gentii?
See Distribution section and maps. Students should be able to identify that there is no evidence that the overall distribution of the species was much larger than currently is shown in Figure 1. However, genetic diversity has been lost through the extinction of specific populations. The species also now shows a higher degree of fragmentation due to human deforestation with great reductions in population sizes leading to the low genetic diversity seen within the small populations.

• What are the mating system, seed and pollen dispersal mechanisms? What do these mean for conservation?
See Phenology section and page 9. Controlled pollinations show T. gentii has a mixed mating system (mixture of selfing and outcrossing). Short distance wind-dispersal of T. gentii pollen and an apparent absence of insect pollination suggest limited outcrossing and limited pollen flow between trees within and between populations. T. gentii seeds are explosively dispersed over short distances leading to germination of related groups of trees and increased likelihood of inbreeding between related trees. The resultant loss of genetic diversity and inbreeding depression may negatively affect demography through the selective fruit abortion, low seed set and regeneration that are evident and thus threaten the conservation of this endangered species.

• What are the levels of genetic variation and how is it distributed across populations? Which are different?
See Genetic variation in natural populations section. Larger populations show a higher percentage of polymorphic loci (e.g. Abiriwapong 16.9%, Yongwa 13.6%, Chalet 8.4%) compared to smaller populations. Table 3 shows the low levels of genetic diversity that are expected for stands/populations that are very small. The populations fall into three main genetic groups associated with geographic location (i.e. western group, eastern group, and Yongwa). The high population differentiation value (0.941) suggests the populations of T. gentii are highly structured with high similarity between individuals within each population and low gene flow between them. The Okpe stand (Fig. 1b), which was not surveyed, may also be genetically distinct, given its spatial separation.

• To what extent does sample size limit the conclusions that can be drawn from the genetic marker data?
The data are biased by the unequal and low sample sizes. The levels of genetic diversity are as much a reflection of the sample size, as the actual size of the population (see column 3, Table 3). In the case of some small populations, this represents a 100% sample (e.g. Botriansa 2.1%, Senkyeso 2.1%, Hospital 0.8%, polymorphic loci) and is therefore a true reflection of low genetic diversity in these remnants. The high population differentiation value and failure of some stands to group genetically with other geographically close locations reflect sampling/bottleneck effects from both the small sample and actual sizes of remnant stands, rather than any true genetic differentiation. Thus, classification of each stand as a population is artificial and unlikely to reflect original biological populations. The molecular genetic data should not therefore be used as conclusive evidence on which to base a conservation strategy.
What non-genetic marker information in the study can be used to guide genetic conservation?

Geographical spatial separation, the size of remaining stands, and the information on fruit set from controlled pollinations, can provide good guidance to develop sound genetic conservation, without reliance on the molecular marker data. Stands/populations that are geographically closer to each other are likely to have been historically most closely related genetically (isolation by distance). The spatial distribution (Figs. 1a and 1b) allows a priori grouping of the populations, e.g. a) Ajena, Chalet, Hotel, Hospital, Sapawsu, Yongwa; b) Dorrkper, Nayo, Yogaga; c) Okpe; d) Krobo; e) Abiriwapon, Boobene, Koware. These could be adjusted by any information showing large environmental differences between stands (e.g. acidic vs. alkaline soil) that would suggest strongly different selection pressures. The size of the remaining stands indicates those that are threatened (see next paragraph Which populations are too small?) and for which restoration of connectivity between remnant stands is important. The controlled pollination study shows the importance of restoring connectivity or facilitating movement of material between geographically close stands to increase reproductive success and the possibilities for regeneration.

List problems by type

Genetic

Which populations are too small?

Most populations are too small – only Yongwa with some 134 ha is likely to have >500 trees (estimate is 1000). Effective population sizes are likely to be much lower due to overlapping generations. Isolation of *T. gentii* populations by increased fragmentation preventing genetic flow, leads to genetic drift and inbreeding with lower levels of genetic diversity. This may threaten the survival of particular populations and over the long term that of the species, due to deleterious effects on fitness and reduced adaptive capacity under changing environmental conditions. Controlled long-distance pollinations that restore gene flow can remove these threats, resulting in higher fruit set, seed set and improved seed quality in terms of mass, germination and survival of seedlings.

Others

What are the threats to *T. gentii* (short-term and long-term)?

Short-term threats are mainly deforestation, fire and a lack of knowledge about *T. gentii*. Long-term, the small population size of almost all populations and the lack of gene flow between them, threatens the species in terms of regeneration capacity, stochastic threats and evolutionary adaptability.

For which populations is action a priority and of what type should this be?

Priority should be to conserve those populations that have the greatest chance of viability (genetically and in the social context), that together cover the range of genetic diversity within the species. Data on the distribution of genetic variation among populations are important for management decisions, e.g. an efficient sampling strategy for *ex situ* conservation, or which populations are priorities for conservation actions (see *in situ* section below for detail).

What are the limiting social factors to conservation and planting?

Constraints, ranging from economic to scientific and organizational, will affect *in situ* conservation. Factors also include human pressures on remaining forests and trees and specifically, exploitation of *T. gentii* for charcoal/fuelwood, periodic bush fires and farming activities. Charcoal burning is done *in situ* and many seedlings are killed by heat from this process. While the country profile shows there is much goodwill amongst local communities for conservation, the stark reality of there being few livelihood alternatives means that they are forced to use the reserves’ resources.
Students' strategies should indicate

Which conservation methods? The strategy should use a combination of in situ and ex situ methods:

- **In situ** actions should focus on conserving the natural *T. gentii* populations with the largest chance of long-term viability. There are 12 extant populations: five in Forest reserves (Abiriwapong, Yongwa, Sapawsu, Nayom, Dorrkpor), four in protected areas under the Volta River Authority and community members (Chalet, Ajena, Volta Hotel, Okpe), and two in sacred forest (Krobo Mt, Yogoga). The remaining population (Kuwere) is under the protection of community members of Nyamebekyere (see Table below for specific population suggestions). Each population has different threats/status and therefore may require different conservation solutions. It is important to improve the conservation status of the entire habitat to enhance maintenance of the populations. Both the genetic differentiation between the disjunct populations and the limited number of 'eggs in the basket' require immediate action to spread the risk of losing this rare species. Given the inevitable limitations of resources, priority should go to strengthening the in situ conservation status of the largest and genetically distinct populations i.e. Abiriwapong, Yongwa, Okpe, Sapawsu, Ajena, Chalet. Table 1 shows that all populations face similar threats from fire, illegal clearance and other chance events. Ex situ conservation, as discussed below, is likely to be more cost-effective for the remaining reserves given their size and threats.

In addition to the issues and actions described below for in situ conservation, (see above and Table 1 these notes) genetic limitations can be addressed through in situ planting of seedlings. Gene flow and effective population size in these reserves can be increased through the in situ planting of seedlings from other populations. Seedlings from populations can be raised from seed in a nursery (see ex situ below) and then planted in other populations of *T. entii* to improve the genetic exchange among populations. Transfers would only be within the western or eastern regions (see ex situ below). For this action to be cost-effective requires that the incidence of fire and other threats, such as grazing, are well controlled.

- **Ex situ** conservation via seed storage offers a stopgap measure for species under immediate threat. Seed storage of most tree legumes is relatively straightforward and therefore likely to be effective for *T. gentii*. Given the problems with storage facilities (see country profile), duplicates of the collection should be held in two different cold stores (i.e. FORIG and Plant Genetic Resources Centre). Given the high genetic differentiation, it is important that seeds are collected from all populations to conserve most of the species’ remaining genetic diversity. Seed should be collected from at least 10 trees spread out across each population.

Good performance of *T. gentii* seedlings in plantation trials suggests it can also be successfully conserved through ex situ planting of the seed collections. For establishment of ex situ conservation stands, consideration should be given to the suitability of an area in respect of similar climate, weather and soil conditions to where the species is native. Given the genetic differentiation and physical separation of the populations, the ex situ plantings should be made in different 'secure' locations as regional collections i.e. western region (seed from Abiriwapong, Kuwere, Boobohene), eastern region (Yongwa, Sapawsu, Ajena, Chalet, Nayom, Dorrkpor, Krobo Mt, Yogoga). Okpe could be kept separate or combined with the eastern region. This would facilitate inter-population crosses, allowing *T. gentii* to recover from inbreeding depression with improved fruit set, seed set, seed germination and seedling survival. Outcrossing between these regions is avoided as it may result in dilution of locally adapted genomes with foreign genes and lower progeny fitness.
(outbreeding depression). The seed produced from these ex situ stands could also be used in both in situ plantings (see above) and in restoration initiatives.

There is potential for the establishment of *T. gentii* in the Ghanaian botanical gardens at Aburi, Bunso and Legon. The role for botanical gardens in ex situ genetic conservation has inherent limitations, such as the small numbers conserved. However, such an activity may offer more general benefits in terms of support to conservation of the species and the reserves through the potential for education and publicity, given the number of visitors each year. There may also be possibilities of developing the species as an ornamental, as has already occurred with *Psychotria ankasensis*. Again, while the potential role for ex situ genetic conservation via this approach is limited, it can have both publicity and educational value.

**What do end-users need to know and how will you communicate that?** Education programmes for local people are needed to help reduce the uncontrolled burning and removal of *T. gentii* for charcoal and fuelwood. This requires production of targeted information (e.g. posters/leaflets), but must go hand in hand with actions that provide viable alternatives for local communities to take pressure off the reserves, e.g. planting of community wood lots.

**Table 1. Recommendations for actions by population of *Talbotiella gentii***

<table>
<thead>
<tr>
<th>Population</th>
<th>Recommendation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kuwere</td>
<td>Population highly diverse, educate community about the need to protect the species. Ex situ conservation recommended.</td>
</tr>
<tr>
<td>Abirivapong</td>
<td><em>In situ</em> conservation recommended.</td>
</tr>
<tr>
<td>Boboohene</td>
<td>Population is within grassland vegetation, high threat of extinction by fire. <em>Ex situ</em> conservation recommended.</td>
</tr>
<tr>
<td>Yongwa</td>
<td>Population highly diverse, <em>in situ</em> conservation recommended, Yongwa community must be involved in conservation strategies. Fire belt around population recommended.</td>
</tr>
<tr>
<td>Krobo Mt.</td>
<td><em>Ex situ</em> conservation recommended due to inaccessibility in terms of permission to protect the population.</td>
</tr>
<tr>
<td>Sapawsu</td>
<td>Collaboration (Forestry Commission &amp; VRA*) to protect the population. <em>In situ</em> conservation recommended.</td>
</tr>
<tr>
<td>Hotel</td>
<td>Collaboration (Forestry Commission &amp; VRA) to protect the population. Population is highly diverse, <em>ex situ</em> conservation recommended due to its location.</td>
</tr>
<tr>
<td>Nayom</td>
<td>Both <em>in situ</em> and <em>ex situ</em> recommended. <em>In situ</em> trial is ongoing carried out by the Forestry Commission technical officer in charge of the site.</td>
</tr>
<tr>
<td>Doorkper</td>
<td>Both <em>in situ</em> and <em>ex situ</em> recommended. <em>In situ</em> trial is ongoing by Forestry Commission Technical Officer in charge of the site.</td>
</tr>
<tr>
<td>Yogoga</td>
<td><em>Ex situ</em> conservation recommended due to inaccessibility in terms of permission to protect the population.</td>
</tr>
<tr>
<td>Ajena (Oninwi)</td>
<td>Protection laws must be tightened. VRA should collaborate with Forestry Commission in protection of population. Both <em>in situ</em> and <em>ex situ</em> conservation recommended.</td>
</tr>
<tr>
<td>Okpe</td>
<td>VRA should collaborate with Forestry Commission in protection of population.</td>
</tr>
<tr>
<td>Chalet</td>
<td>Protection laws must be tightened, VRA should collaborate with Forestry Commission in protection of population.</td>
</tr>
</tbody>
</table>

* Volta River Authority
Who will do what and where? Chiefs, local people and government bodies (Volta River Authority, Forest Research Institute of Ghana-FORIG, Forestry Department-FD) should be involved in in situ conservation to link their ongoing conservation interests, e.g. construction of fire belts around *T. gentii* populations at Sapawsu and Chalet reserves. Management plans documenting appropriate interventions to safeguard the forest habitat are a high priority (FD). Monitoring of populations to check their status is needed (FD). Seed collection should be undertaken and also establishment of *in situ* and *ex situ* plantings (FORIG) in conjunction with the relevant authority and community for the reserves and with others for *ex situ*.

How will you pay for it? It is vital students understand that resources for conservation are limited and therefore require a prioritisation of actions. Conservation efforts need to be targeted where scarce resources can be most effective. It is not practical, nor cost-effective to recommend all activities for all populations. The main actions outlined above are limited, but feasible. Seed collection would require additional funds, while communication of the importance of *T. gentii* requires a modest budget that could benefit from redirection of existing resources.

Further Information


FAO, FLD, IPGRI. 2004b. Forest genetic resources conservation and management. Vol. 3: In plantations and genebanks (*ex situ*). International Plant Genetic Resources Institute, Rome, Italy.


Forest Genetic Resources Training Guide

MODULE 1  Species conservation strategies
1.1  *Leucaena salvadorensis*: genetic variation and conservation
1.2  *Talbotiella gentii*: genetic variation and conservation
1.3  *Shorea lumutensis*: genetic variation and conservation

MODULE 2  Trees outside of forests
2.1  Conservation of tree species diversity in cocoa agroforests in Nigeria
2.2  Devising options for conservation of two tree species outside of forests

MODULE 3  Tree seed supply chains
3.1  Genetic bottlenecks in the restoration of *Araucaria nemorosa*
3.2  Tree planting on farms in East Africa: how to ensure genetic diversity?

MODULE 4  Forest management
4.1  Impacts of selective logging on the genetic diversity of two Amazonian timber species
4.2  Does selective logging degrade the genetic quality of succeeding generations through dysgenic selection?
4.3  Conserving *Prunus africana*: spatial analysis of genetic diversity for non-timber forest product management

MODULE 5  How local is local? – the scale of adaptation
5.1  Selecting planting material for forest restoration in the Pacific north-west of the USA
5.2  Local adaptation and forest restoration in Western Australia

*Other modules to be published among the following: Plantation forestry, Tree domestication, Forest restoration, Genetic modification*