Growth and fruit yield of seedlings, cuttings and grafts from selected son tra trees in Northwest Vietnam

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Sammy Carsan and Nguyen La work for the World Agroforestry Centre (ICRAF).
Summary

This study examined tree growth and fruit production of son tra (Docynia indica), an indigenous fruit tree. Eight phenotypically superior trees with high fruit yields and good fruit appearance were selected in 2005 at Ngoc Chien commune, Muong La District, Son la Province, Northwest Vietnam. Three types of planting material were raised from these trees: grafts from the selected trees onto unselected seedling rootstocks, seedlings raised from seeds collected from the superior trees and cuttings raised from seedlings. The grafts, seedlings and cuttings were planted out in 2006 in adjacent blocks in a field trial established at Chieng Bom experimental station in Son La Province. Thirty trees of each type were monitored for a 3-year period commencing in January 2012, when the trees were 6 years old. Survival of all three types from planting was excellent, remaining above 90% at the end of 2014. Grafted trees grew fastest, attaining a mean height of 7.0m and crown width of 4.4m, while trees raised from cuttings grew significantly slower (height 5.0m and crown width 3.4m, with seedling-derived trees intermediate. Mean fruit yield at 8 years was significantly higher for grafts (38.7kg per tree) compared to trees raised from seedlings (30.7kg per tree) and those from cuttings (28.9kg per tree). Fruits from the grafted trees were judged to be of superior quality, being predominantly yellow in colour and of uniform large (> 3cm) size, while fruit from most of the seedling and cutting-derived trees were judged to be of moderate quality with inferior size and colour. Considering the excellent survival of grafts and their superior fruit yield and fruit quality, grafting can be recommended as a better way to propagate selected son tra trees, compared with using seedlings raised from seed collected from the selected trees or cuttings raised from these seedlings.

Keywords: Son tra, Docynia indica, grafting, cutting, seedlings, domestication, Vietnam
Acknowledgements

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Acronyms

ACIAR  Australian Centre for International Agricultural Research

AFLI   Agroforestry for Livelihood Improvement

CGIAR  Consortium of International Agricultural Research Centres

CSIRO  Commonwealth Scientific and Industrial Research Organization

DBH    diameter at breast height

FSCNW  Forestry Science Centre for Northwest

FTA    Forests, Trees and Agroforestry

ICRAF  World Agroforestry Centre

NWFSC  Northwest Forest Science Centre of Vietnam

TBU    Tay Bac University
Introduction

The Agroforestry for Livelihood Improvement (AFLI) project managed by the World Agroforestry Centre and funded by the Australian Centre for International Research is supporting the domestication of son tra (*Docynia indica*), an indigenous fruit tree species native to the North West and North Central ecoregion of Vietnam. The use of selected, genetically superior planting material of son tra could increase fruit yields and improve quality of fruit from plantations and home gardens. Screening to locate superior genotypes in natural forests and plantations has been undertaken. Over the period 2012-2014, the North West Forest Science Centre, Vietnamese Academy of Forest Sciences, has screened over 2000 candidate trees from natural forests and plantations in three northwestern provinces for vigour, health, fruit yield and fruit quality. Thirty trees ranked highest for a combination of these traits are now being evaluated in grafted clonal trials established in farmers’ fields (Tiep et al., 2016).

In fruit tree domestication, it is common to produce planting stock by clonal propagation of superior selected trees rather than by raising planting stock using seeds of the selected trees (Leakey and Akinnifesi, 2008). This is because the genetic identity of the tree crown is retained in the graft, so the fruit characteristics of the selected tree are maintained. If seeds are collected from the same selection, the offspring will differ genetically from the mother tree owing to variation introduced during sexual reproduction, and will not have the exact characteristics of the selected tree. Selected clones of temperate and tropical fruit tree species such as apple, pear, citrus varieties and mango are therefore often grafted onto seedling root stocks to produce grafts for planting in commercial fruit tree orchards.

Apple, pear and quince, which are easily grafted, are closely related to son tra, belonging to the same plant family, *Rosaceae*, so there is good reason to expect that a domestication strategy involving grafting of selected and tested clones would be suitable for son tra. In addition to maintaining the genotype, grafting has the advantage that scion material taken from the mature crown and grafted onto a seedling rootstock will typically produce a tree that bears fruit earlier than a tree of the same species raised from a seedling. Clonal propagation by rooted stem cuttings is another option, however it is typically found that cuttings take longer to bear fruit than do grafts, and there may be problems of root system and crown development in trees raised from cuttings.

An experiment which evaluated production of trees from grafts, cuttings and seedlings of selected, phenotypically superior son tra trees was established by the Northwest Forest Science Centre of the Forest Science Institute of Vietnam in 2006. The initial
experiment was a short-term (2-year) project which established these three types of planting stock in a field trial at Chieng Bom Silviculture Experimental Station in Son La Province. The experimental planting survived and grew successfully and now constitutes a valuable experimental resource which can provide insights into the long-term performance of vegetatively propagated trees, relative to trees raised from seedlings.

AFLI supported ongoing monitoring of the growth and fruit production of the three different types of son tra planting materials in this trial, over the period 2012-2014. The objective of this study was to compare growth, fruit yields and fruit quality) of son tra trees established from grafts, seedlings and seedling-derived cuttings all originating from the same selected trees.
Materials and methods

Original trial

The trees under study were established in the course of a project implemented by the Northwest Forest Science Centre of Vietnam (NWFSC) titled, “The application of biotechnology to propagate Son tra in Son La”, conducted in 2005-2006. The planted trees were established at the Chieng Bom Silviculture Experimental Station, Northwest Forest Science Centre, located at Chieng Bom Commune, Thuan Chau District, Son La Province (21°23’02” N, 103°38’51” E, 1109m elevation).

The project first selected eight phenotypically superior son tra trees in Ngoc Chien Commune, Muong La District, Son La Province. Three types of planting material were developed from these selected trees, as follows:

- Treatment 1: Seedlings (planting stock raised from seeds collected from the eight selected trees)
- Treatment 2: Cuttings (planting stock raised from rooted cuttings taken from seedlings collected from the eight selected trees)
- Treatment 3: Grafts (planting stock raised by grafting scion material from selected trees, grafted onto seedling rootstocks)

Details of the development of the planting stock for the three treatments are shown in Table 1.

Table 1: Genetic origins, methods and timelines to develop planting stock of grafts, cuttings and seedlings of son tra for the field trial at Chieng Bom Station.

<table>
<thead>
<tr>
<th></th>
<th>Seedlings</th>
<th>Cuttings</th>
<th>Grafts</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Origin</strong></td>
<td>Seeds were collected from 8 phenotypically superior trees, bearing heavy crops of superior fruit, in Ngoc Chien commune, Muong La district, Son La.</td>
<td>Cuttings were prepared from seedlings propagated from seeds from the 8 selected trees in Ngoc Chien commune, Muong La district, Son La.</td>
<td>Scion material was collected from crowns of the 8 superior trees in Ngoc Chien commune, Muong La district, Son La.</td>
</tr>
<tr>
<td><strong>Preparation of planting stock</strong></td>
<td>Seedlings sown in October 2005</td>
<td>Cuttings set in October 2005</td>
<td>Seedlings sown in November 2005</td>
</tr>
</tbody>
</table>
Planting stock of all three treatments was raised in the NWFSC nursery at Son La City. Planting stock of the different treatments was of similar size at the time of field planting in June-July 2006.

Three adjacent treatment blocks (0.4 ha of seedlings, and 0.3 ha each of cuttings and grafts) were planted out at the Chieng Bom station on 1 ha of land with roughly uniform topography (10-15° slope) and uniform soil. Spacing between trees was 3m x 3m, giving a stocking at planting of approximately 1100 trees per ha. Fertilizer was applied to each tree as follows:

1) At planting – 0.5kg mineral fertilizer (N 5, P₂O₅ 10, K₂O₃) + 3kg composted animal manure per tree
2) 2007-2011 – annual top dressing of 2.5kg composted animal manure per tree
3) 2012-2014 – annual top dressing of 0.5kg NPK mineral fertilizer per tree

Tree crowns were not pruned. Tree survival in all the three experimental treatments was good, with over 90% survival up to 2012, when the experimental monitoring reported here commenced.

**Monitoring son tra trees growth and fruit yields (2012-2014)**

Monitoring commenced in January 2012, and continued for a period of three years. Within each of the three treatments, 30 trees were selected at random for monitoring. The selected trees were numbered and labels were painted on tree trunks for ease of identification during field assessments. No deaths occurred during the monitoring period.

Growth was measured once every year in May, early in the growing season. Stem diameter at breast height (DBH) was determined as the average measurement of north-south and east-west diameters, measured at 1.3m above ground using calipers. Tree height was measured using a measuring stick. Crown diameter was measured using a measuring stick, in two directions (north-south and east-west) and the average value calculated. Pest and disease status was monitored in the course of visits made throughout the year.

Fruit production was determined by harvesting when fruit was of suitable ripeness for collection. Either one or two fruit harvests were conducted per tree. Total weight of fruit harvested per tree was recorded.

Fruit harvested from each tree was examined and fruit quality was assigned to one of the three following categories:
1) High quality – fruit predominantly uniform-size with diameter >3 cm and yellow in colour;

2) Moderate quality – fruit diameter range 2-3 cm and predominantly yellow in colour;

3) Low quality – fruit predominantly uneven-size, diameter <3 cm and brown or greyish in colour.

**Data analysis**

Univariate one-way analysis of variance was carried out for each growth variate and fruit yields in each year, to test whether there were significant differences among treatments, using the analytical software package Genstat (release 16). The critical difference at $p=0.05$ for comparing treatments was calculated as the standard error of the difference of the treatment means multiplied by student’s $t$-value for the residual degrees of freedom in the analysis of variance (87 residual degrees of freedom, student’s $t$ value = 1.99).
Results

Differences among treatments were significant \((p < 0.001)\) for all growth traits at each year of measurement. Grafted trees grew fastest, with the largest mean dbh, height and crown width in each of the three years of monitoring (Table 2). Cuttings grew slowest and seedlings were intermediate in their growth. By age 8 years, grafts had a mean height of 7.0m and crown width of 4.4m, while cuttings had a mean height of 5.0m and crown width of 3.4m.

Treatment rankings for fruit production were the same as those for growth (Table 2). Mean fruit yield of grafted trees increased from 5.7kg per tree at age 6 years to 38.7kg per tree at age 8 years. The corresponding yields for seedlings were 4.1kg per tree at age 6 years to 30.7kg per tree at age 8 years, with fruit yields from cuttings slightly lower than those of seedlings (Table 2). Differences in fruit yield among treatments were not significant at age 6 years, but were significant \((P < 0.001)\) at ages 7 and 8 years. The higher fruit yield of the grafted trees is the cause of the significant treatment effects at ages 7 and 8 years (Table 2).

Table 2: Mean growth and fruit yield for the three categories of planting material over 3 years

<table>
<thead>
<tr>
<th></th>
<th>2012</th>
<th>2013</th>
<th>2014</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Seedlings</td>
<td>Cuttings</td>
<td>Grafts</td>
</tr>
<tr>
<td>Height (m)</td>
<td>4.6</td>
<td>4.6</td>
<td>6.6</td>
</tr>
<tr>
<td>Dbh (cm)</td>
<td>8.6</td>
<td>5.5</td>
<td>10.9</td>
</tr>
<tr>
<td>Crown width (m)</td>
<td>2.9</td>
<td>2.7</td>
<td>4.1</td>
</tr>
<tr>
<td>Fruit yield (kg)</td>
<td>4.1</td>
<td>4.1</td>
<td>5.7</td>
</tr>
</tbody>
</table>

*\(p = 0.05\) Critical difference for comparing treatment means.
The percentage of trees bearing fruits increased markedly from 2012 (age 6 years), when fewer than 50% of trees bore fruit, to over 80% of trees bearing fruit in 2013 and 100% bearing fruit in 2014. The three treatments did not differ significantly in the percentage of trees bearing fruit (Figure 1).

Variation in fruit yield within the three treatments, in 2013 and 2014 are summarized in Table 3. As less than half of the trees bore fruit in 2012, little useful information on tree-to-tree variation in yield could be obtained for that year. The coefficient of variation is the standard deviation divided by the treatment mean, expressed as a percentage. Standard deviations of fruit yield were in the range 4-5kg in both 2013 and 2014. Because grafts had the highest fruit yields, the coefficients of variation were lowest for grafts, reducing to 14% in 2014 (age 8 years), while those for seedlings and cuttings remained slightly higher at 18% and 17% respectively.
Table 3: Variation of fruit yield within treatments in 2013 and 2014

<table>
<thead>
<tr>
<th>Year</th>
<th>Treatment</th>
<th>Mean yield (kg)</th>
<th>Standard deviation (kg)</th>
<th>Coefficient of variation (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>Seedlings</td>
<td>11.0</td>
<td>5.2</td>
<td>47</td>
</tr>
<tr>
<td></td>
<td>Cuttings</td>
<td>8.4</td>
<td>4.1</td>
<td>49</td>
</tr>
<tr>
<td></td>
<td>Grafts</td>
<td>15.2</td>
<td>5.4</td>
<td>36</td>
</tr>
<tr>
<td>2014</td>
<td>Seedlings</td>
<td>30.7</td>
<td>5.6</td>
<td>18</td>
</tr>
<tr>
<td></td>
<td>Cuttings</td>
<td>28.9</td>
<td>4.8</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>Grafts</td>
<td>38.7</td>
<td>5.4</td>
<td>14</td>
</tr>
</tbody>
</table>

Quality of fruits from all fruiting trees of all the three treatments was assessed as moderate in 2012 (Table 4). In 2013 (age 7 years), six out of 30 grafted trees were classed as having good fruit quality, along with only three trees from cuttings and two trees from seedlings. By 2014 (age 8 years), fruit of all 30 grafted trees was assessed as being of good quality while all 30 trees from seedlings and 25 trees from cuttings had fruit of moderate quality.

Table 4: Numbers of trees in three fruit quality categories (2012-2014)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Year</th>
<th>Fruit quality</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Good</td>
</tr>
<tr>
<td>Grafts</td>
<td>2012</td>
<td>13</td>
</tr>
<tr>
<td>Grafts</td>
<td>2013</td>
<td>6</td>
</tr>
<tr>
<td>Grafts</td>
<td>2014</td>
<td>30</td>
</tr>
<tr>
<td>Cuttings</td>
<td>2012</td>
<td></td>
</tr>
<tr>
<td>Cuttings</td>
<td>2013</td>
<td>3</td>
</tr>
<tr>
<td>Cuttings</td>
<td>2014</td>
<td>3</td>
</tr>
<tr>
<td>Seedlings</td>
<td>2012</td>
<td></td>
</tr>
<tr>
<td>Seedlings</td>
<td>2013</td>
<td>2</td>
</tr>
<tr>
<td>Seedlings</td>
<td>2014</td>
<td></td>
</tr>
</tbody>
</table>
Discussion

Monitoring the three experimental treatments in this trial over the period 6-8 years after planting has given clear findings relevant to the domestication of son tra. Trees raised from seedlings, grafts and cuttings all grew and survived well at the study site, with over 90% survival at the age 8 years. This shows that the species is easily grafted, with good long-term survival of grafted trees and no problems of graft union failure. Cuttings taken from seedlings also survived well.

Grafts of scion material taken from selected trees grew significantly faster than seedlings and seedling cuttings raised from seeds collected from the same trees, and their fruit yields were significantly higher at ages 7 and 8 years. The higher yield of grafted trees was the result of higher fruit yield per tree, rather than a higher proportion of trees bearing fruit. By age 8 years, all 30 monitored trees in all three treatments were bearing fruit. The mean fruit yield of 38.7kg per tree for grafted trees in year 8, at a stocking of about 1000 trees per hectare, equates to a fruit yield of about 4000kg per hectare. The relatively high doses of fertilizers, applied on an annual basis, should be kept in mind when interpreting the fruit yields obtained. The influence of fertilizer application on yield is not yet known.

Grafts of selected trees grew faster and yielded more fruit than did seedlings, and rooted stem cuttings derived from seedlings, raised from the same selected trees. Assessments of fruit quality (colour and size) suggested that quality was higher from grafts than from the trees raised from seedlings and cuttings. It is important to note that the distinction between good and moderate fruit quality was based on fruit size, uniformity and colour only. It remains to be determined whether this would translate into better prices and market demand for the high quality fruit. Nonetheless this result shows that the grafts of selected trees can bear fruit judged to be of improved quality.

Information on variation within treatments is useful for planning future experiments that will compare growth and fruit yields of candidate son tra clones. Standard deviations and coefficients of variation can be used to determine an appropriate level of replication (number of clonal ramets to be tested) to enable detection of specified differences among clones in traits such as fruit yield. The grafted treatment in this study comprises a total of 30 trees representing up to eight clones. The exact number of clones represented may be somewhat less than eight. Grafts of a single clone would be expected to have a lower coefficient of variation for fruit yield than that reported here for the grafted treatment shown in Table 3. For a single clone, the grafted crowns would be genetically identical and variation in yield among trees would be caused
only by stock effects (seedling stocks of the grafts would differ genetically), graft performance effects and local environmental differences.

Unfortunately, some genetic information was lacking from the initial experiment. Eight selected individual trees of superior phenotype (in terms of vigour, fruit yield and fruit quality) were selected from Ngoc Chien Commune, and supplied scion material for producing the grafted trees and seeds for producing the seedlings and cuttings under test. However, the number of scion genotypes (clones, derived from the ortets) represented among the 30 selected grafted trees evaluated in the grafted treatment is not known, because clone identities of individual grafts planted out in 2006 were not retained. It is possible that some of the 8 selected trees are not represented in the sample of 30 grafted trees. Similarly, the maternal identities of the 30 seedling-derived and cutting-derived trees are not known. Another uncertainty is the intensity of the selection used to select the 8 trees. These uncertainties make the superior growth and fruit yield of the grafts difficult to interpret: are they simply a consequence of grafting per se, or are they a consequence of genetic selection, or of both? Probably both factors contribute to the better performance of the grafted trees. The effectiveness of genetic selection is supported by the indication of superior fruit quality from the grafts. Further information on the results of selection will be obtained from new clone trials established under the AFLI project (Tiep et al., 2016).

The field trial was not set out with replication, there being only a single block planted for each of the three treatments. Therefore there is some need for caution in interpretation of the treatment differences, because local site differences may have contributed to the treatment differences. However, the relative uniformity of the one-hectare trial site and relatively low coefficients of variation in fruit yield within treatments suggests that site differences would not be a major confounding effect.
Conclusion

This trial has yielded useful information on the performance of grafted son tra relative to seedling-derived and the gains in fruit yield and quality potentially available from selection. The results show that grafts of selected trees will grow, survive and give high fruit yields. Grafting can be recommended as a better way to propagate selected son tra trees, compared with using seedlings raised from seed collected from the selected trees or cuttings raised from these seedlings. Information obtained on fruit yield, including tree-to-tree variation in yield, will be useful for planning future field trials for son tra domestication.
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