

DNA fingerprinting of maire *Nestegis cunninghamii* tissue

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The Tongariro/Taupo Conservancy initiated court proceedings regard the felling of a mature maire, on DOC administered land, for firewood. The defendants asserted that the firewood came from their own property. The NZFRI were asked to investigate whether the firewood under contention had come from the DOC tree. The NZFRI reported accordingly. [Ed]

We have completed the DNA fingerprinting assay on the eight maire (*Nestegis cunninghamii*) samples supplied by Department of Conservation. I have been asked to comment with respect to the following four questions:

1. What method is best used to determine the genetic composition of plant tissue from maire (*Nestegis cunninghamii*)?
2. Have similar methods been used in criminal investigations elsewhere and what is the applicability to other plants for similar compliance or other fingerprinting matters?
3. Can it be proven that a sample of maire firewood (seized by Department of Conservation) does not match a maire from a farm not on land administered by the Department of Conservation?
4. What is the probability that the sample matches the tree (on the land administered by the Department of Conservation) which the Department alleges the firewood came from?

I will address these questions in turn.

1. The most sensitive fingerprinting method currently available for use on maire is random amplified polymorphic DNA (RAPDs). This technique reveals the differences between individual trees at the level of the DNA. Importantly, RAPD markers can be used to fingerprint species where there is no *a priori* DNA sequence information, and only small amounts of tissue are required for the analysis. I have successfully used RAPDs to fingerprint approximately a dozen plant species and use them extensively for fingerprinting and gene mapping studies in radiata pine. RAPDs are similarly applied by other scientists worldwide.
2. To the best of my knowledge, there has only been one use of plant DNA fingerprinting data in a criminal case. This occurred in the USA in April 1993 (Moon 1993). The DNA fingerprinting methodology which was deemed admissible in that case was the same technique as was used here. Because RAPDs are generally applicable to species which have been little studied, there may well be additional criminal cases where they have been used that I am not aware of. Certainly they are generally useful for fingerprinting nearly all species studied to date, though there are exceptions and every species must be independently evaluated.
3. Demonstrating that RAPD profiles (or other "DNA fingerprints") from two samples do not match is clear evidence that the samples are not derived from the same specimen (provided that the tests are carefully carried out so as to avoid artefacts). From the 20 RAPD profiles pro-

duced we detected 27 bands that differed between samples 9 and 10. This provides indisputable evidence that sample 9 is not derived from the same tree as sample 10.

4. "Proving" that two samples match is not as straightforward as demonstrating that they do not. Two samples may "look" identical and yet still be derived from different individuals. The DNA fingerprints of identical twins are an obvious example.

To calculate the probability that two samples that match are not derived from the same individual one multiplies the population frequencies of each matching band. Essentially what this means is that if two samples share a match that is very uncommon in the population there is a greater likelihood that the two samples are derived from the same source than if they share a match that is more common. This calculation generates the "matching probabilities" used in human forensic science and we use the same approach here.

Our results indicate that there is approximately a 1-in-67 426 chance that samples 6 and 10 are derived from different individuals. In other words, there is a 99.9999% "chance" that the two samples are from the same tree. It is never possible to achieve a 100% "chance".

Reference

Yoon, C. K. 1993 Botanical witness for the prosecution. *Science* 260 : 894-854.

Appendix 1.

Synopsis of RAPD fingerprinting procedure.

DNA from the 3 primary samples and 5 maire sampled from the population near Whakapapa was extracted from 1-2 leaves. Twenty different RAPD profiles were generated for each of the 8 samples. Appendix 2 shows examples of 4 of the 20 different primers. Within each primer each vertical lane is a different tree, labelled by sample numbers provided by DOC. The "bands" are fragments of DNA. These banding profiles were examined to detect matches and mismatches among the samples.

Appendix 2.

RAPD profiles produced by 4 primers.

