Nature’s barcode: the simplest way to track wood

DNA fingerprinting becomes a viable option for verifying existing wood-tracking systems

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Stockpiled DNA: Sampled logs await loading onto a barge. *Photo: A. Suchayo, DoubleHelix*

Using the genetic data inherent in wood DNA as a natural barcode has long been proposed as an elegant and simple way to track wood products through the supply chain. Until recently, however, such an approach has been considered too costly and complicated, the primary barrier being the expense of setting up a comprehensive genetic database. This paper describes a DNA ‘fingerprinting’ methodology that does not rely on the establishment of a genetic database. It could be the holy grail of wood-tracking.

Wood-tracking systems

A necessary requirement of all certification and legality verification systems is a system to track wood from the forest source to export, with the aim of guaranteeing that wood and wood products are derived from legal and sustainable sources and that wood derived from unknown and illegal sources is excluded. Any wood-tracking system should also enable independent monitoring to assure all interested parties that the system is working as planned and is credible (Anon. 2007).

The DNA methodology described here is based on physically matching samples taken from the same log at different stages of the supply chain in conjunction with existing paper-based chain-of-custody (CoC) documentation. A relatively simple DNA fingerprinting test is used to confirm whether the samples originate from the same log, validating or invalidating the CoC documentation.

Why DNA fingerprinting?

Two significant changes have taken place to unlock the potential of DNA for wood-tracking: one in thinking and the other in technology. Early thinking held that DNA tracking would require a comprehensive genetic database of any target tree population, but no such databases existed. It was concluded, therefore, that DNA fingerprinting was unlikely to be a solution for wood-tracking, although in the future it might be used to establish the origin of suspected illegally harvested logs (Dykstra et al. 2003).

New thinking, however, envisages tracking based on the matching of samples of individual trees (Lawson 2007), which would not require an existing genetic database. This concept is similar to a human paternity test, in which DNA samples taken from two individuals are tested against each other to see if they match. Participants in paternity tests do not require their DNA profile to be included in a pre-existing database.

The technological breakthrough comes courtesy of the Human Genome Project, which was completed in 2003. The drive to sequence the human genome was akin to the space race of the 1960s - it fuelled huge advances in technology and genetic sequencing know-how. It took the Human Genome project 13 years and US$3 billion to sequence a human genome. To indicate the resultant advances that have been made in genetic sequencing, in 2012 the cost of sequencing a human genome was about US$6000 (Wetterstrand 2013). Thus, the cost and ease of extracting, sequencing and matching DNA (i.e. DNA fingerprinting) are such that DNA wood-tracking is now a commercially viable option.

A 21st century approach to chain of custody

The genetic matching of individual logs along a supply chain supports, rather than replaces, existing paper-based methods of wood traceability. DNA analysis can be used to validate existing CoC documentation, which is acknowledged to be vulnerable to falsification - particularly between the logging concession and the mill, where most illegally logged wood is introduced into the supply chain (Zahnen 2008).

In 2009, ITTO supported a project through its Biannual Work Programme to evaluate the scientific viability of integrating DNA wood-tracking with an existing CoC system. The project was conducted on a merbau (*Intsia* spp.) supply chain, with logs harvested in Papua, Indonesia, and transported to a mill in Java for processing into flooring and decking. The project demonstrated reliable and accurate differentiation between individual trees (and the logs derived from those trees) and showed it was possible to validate (or otherwise) the transportation documentation from concession to mill. The results of this project are reported in detail in Lowe et al. (2010) and summarized below.

Methodology

Forest concession samples were taken from 2627 merbau logs from particular batches (barge-loads) of raw wood, at either a primary log pond or the point of loading, between 14 November 2009 and 11 March 2010. Samples were referenced to painted markings on the logs indicating the log number, the petak (the harvesting area), the species and dimensions, and the year of the cutting licence under which the tree was felled. Logs were then loaded onto a barge, shipped to Java and transported by truck to the mill. This process was tracked with standard Indonesian government paperwork known as SKSKB transport documents and associated log lists.

At the mill, a second set of samples was taken from 741 logs and again referenced against the painted log markings showing log and petak numbers. Concession and mill samples were collected following strict quality-control protocols designed to maintain the freshness of wood samples by minimizing the loss of moisture content and thus preserving the DNA stored in the sample.

Following guidelines of the International Organization for Standardization (ISO) for sampling procedures for quality-control inspection (ISO 2859), 32 samples were chosen randomly from the samples collected at the mill and matched with the samples collected from the concession using the SKSKB log transport documentation and log lists. The sample pairs were forwarded to a laboratory, where DNA was extracted and used to amplify 14 genetic markers (one chloroplast microsatellite and 13 nuclear microsatellites). For each of these markers, the success of DNA extraction and amplification was recorded for both samples of each of the 32 logs. Population allele frequencies were used to calculate the probability that an individual genotype (or genetic profile) occurred within the logging concession (Lowe et al. 2004); this was done as an additional check on the possibility that logs had been substituted along the supply chain.

Results

The study found that while the ability to extract and analyse DNA from logs decreased slightly between the forest concession and sawmill samples, overall sufficient data were obtained for 27 of the 32 logs to provide exact genotype matches between forest and sawmill samples. For four of the five samples that failed, the sawmill sample failed to amplify any microsatellite loci; for the fifth sample, non-overlapping genetic markers were amplified between the forest and sawmill samples, making it impossible to determine whether the samples matched.

Of the 27 samples for which the forest and mill DNA genotypes matched, it was possible to calculate the probability of an identical genotype being present within the forest concession, providing a test of the likelihood that an illegally substituted log would have the same genotype as the forest sample. The probability of illegal substitution was very low (1 in 100 000 or lower) for 18 samples, low (1 in 100 to 1 in 10 000) for 7 samples, and moderate (1 in 10) for 2 samples (see table following page).

Towards a more practical approach

Clean-slate approaches

Most technologies for tracking wood on the market aim to replace existing government-regulated marking and paperwork systems entirely in a 'clean-slate' approach, on the basis that most existing government systems are insufficiently robust and are open to abuse. There are problems with the clean-slate approach, however. Starting from scratch makes new systems slow to implement, while the need to use advanced technologies to overcome fraud makes them more expensive than the traditional government controls they replace. Perhaps more importantly, a clean-slate approach is short-sighted because it fails to support efforts to improve official controls by simply bypassing them. While a clean-slate approach allows progressive individual concessions – such as those with strong links with sensitive markets – to establish their own systems on a voluntary basis, it does little to counter the broader problem of illegal logging. Ideally, similar robust and advanced technologies would be taken up nationally by governments, but this requires a rare willingness to consider radical change and the capacity to pay for it, which is also often lacking.

Results of DNA fingerprinting on 32 randomly selected merbau logs harvested in Papua, Indonesia, and transported to a mill in Java for processing

Log	No. of loci matched[a]	% confidence no log substitution[b]	Verdict
1	6	99.99995	Match
2	4	99.998	Match
3	6	99.999999999	Match
4	4	99.999	Match
5	4	99.9998	Match
6	3	99.99998	Match
7	12	100	Match
8	10	100	Match
9	0	0	No result
10	4	99.9999	Match
11	12	100	Match
12	4	99.9999	Match
13	3	99.9	Match
14	3	99.99	Match
15	1	99	Match
16	1	67	Moderate chance of substitution
17	4	99.998	Match
18	4	99.995	Match
19	10	99.999999997	Match
20	5	99.98	Match
21	5	99.9999999996	Match
22	1	96	Low chance of substitution
23	4	99.994	Match
24	4	99.97	Match
25	0	0	No result
26	1	99	Low chance of substitution
27	1	89	Moderate chance of substitution
28	4	99.998	Match
29	0	0	No result
30	3	99.98	Match
31	0	0%	No result
32	0	0%	No result

Note: a = the number of loci (genetic markers) that amplified and matched from both forest and mill samples; b = the resulting confidence level that another log with the same genetic profile was not substituted. For example, for log 1 there is 99.99995% confidence that there was no log substitution between concession and mill and that the CoC was intact.

Incremental approaches with high-tech checking

An alternative to the clean-slate approach is to support existing systems with technologies that can double-check the veracity of those systems and to build on them incrementally. This approach has the advantage of being quick and cheap, and it is more likely to be viable for national uptake. It also supports rather than sidelines government efforts to tackle illegal logging.

A DNA-based system is ideal for this kind of incremental approach. DNA match-sampling can be carried out at given stages of the supply chain and used to cross-check the veracity of painted markings and paper-based CoC systems. Such systems may remain susceptible to fraud, but DNA-matching technology would catch enough fraudsters to make cheating uneconomic.

There are benefits in terms of cost as well. Other technological solutions for tracking wood must cover every stage of the supply chain and are only as strong as their weakest link. In a supply chain verified using DNA, however, only the beginning and end of the chain need be secured. Basic paper-based (or, in some places, computer-based) systems would still be needed for matching samples with their source logs, but the incentive to abuse such systems would be removed by the risk of exposure by DNA matching. This would reduce the burden on independent auditors or verifiers by removing the need to examine the intermediate stages of the supply chain (Lawson 2007).

An incremental approach using existing systems combined with DNA matching can complement and even replace audits conducted in person at critical points in the supply chain. By relying more on scientific data, it may be possible to reduce the overall cost of certified wood products and to reduce the cost burden of monitoring the supply chain, making certified products cheaper. A comprehensive cost–benefit assessment of the various wood-tracking options is recommended to enable comparisons, assess the cost-effectiveness of certification with and without DNA testing, and evaluate the most efficient combination of DNA testing and physical auditing.

If the cost-savings associated with a reduced need for auditing outweigh the costs of DNA testing, the cost of maintaining a DNA–CoC system will be less than a traditional paper-based and audited system. With the increasingly low cost of DNA testing, this is a probable outcome.

An incremental approach using existing systems combined with DNA testing can also be integrated into the legality assurance systems (LASs) being developed by countries participating in the European Union Forest Law Enforcement, Governance and Trade (FLEGT) voluntary partnership agreements (VPAs). It is likely that LASs will involve improvements and additions to existing government systems. DNA could be used either as part of a standard in-country verification process, or jointly by the European Union and source-country governments as an enforcement and verification tool.

Implementation

The ITTO project described above provides pointers for further development. To test the methodology on a small scale, the project was implemented using specific batches of wood and therefore on only a limited part of the supply chain. Samples were taken at the primary log pond rather than the point of harvest - it is at the log pond where the buyers of raw wood are decided and the final destinations of the logs determined. A next step would be to apply this methodology to an area-based certification system where samples are taken by the concessionaire at the point of harvest or during the forest inventory. Sampling would also extend further along the supply chain through processing to the finished product, with a focus on the links in the chain that are most at risk from log or lot swapping.

The scientific validity of the methodology has been demonstrated. Further improvements can be made to the DNA extraction protocols to improve the reliability and quality of DNA extraction to obtain an even higher success rate. This would reduce the need to repeat tests and would further reduce the cost of the testing process.

At the far end of the supply chain, the methodology is currently limited to solid wood products such as flooring, decking and furniture, where the extent of processing (heat and chemical treatment) and therefore the impact on the wood's DNA is relatively low. Improvements to DNA extraction protocols may enable the application of the technology to further-processed products such as plywood.

DNA testing is not designed to replace existing paper-based systems; rather, it is designed to support, simplify and strengthen them. Genetic mismatches highlighted by DNA testing can act as a 'red flag' to auditors, who can then conduct more thorough investigations. We believe that this DNA tracking methodology is now suitable for industry uptake to track certified wood and check for illegal substitutions along solid-wood-product supply chains. The methodology will not only complement paper-based CoC methods, it will contribute to future methods that use databases on genetic structure (e.g. Deguilloux et al. 2003; Lowe et al. 2004; Lowe 2008; Lemes et al. 2010).

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