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Forest Genetic Resources Working Papers

Feasibility and proposed outline of a global review of forest biotechnology

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Background

This paper was prepared for and presented to the 13th Session of the FAO Panel of Experts on Forest Gene Resources, 10-12 November 2003, Rome, Italy. The report of the Panel is available on line at http://www.fao.org/forestry/site/3484/en .

Disclaimer

The Forest Genetic Resources Working Papers report on issues and activities in related to the conservation, sustainable use and management of forest tree genetic diversity. The purpose of these papers is to provide early information on on-going developments in the field, and to stimulate discussion.

Comments and feedback are welcome.

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EXECUTIVE SUMMARY

Developments and applications of forest biotechnology are rapidly expanding. They are becoming both more complex and more accessible throughout the world. There are fundamental differences in public perception and potential applications of biotechnology in forestry relative to agriculture. These differences stem from the multifaceted cultural and utilitarian role forests play, and their relatively recent history of domestication compared to that of agricultural crops.

In this report, we present a modified definition for biotechnology and propose five major categories of forest biotechnological tools. These are: (1) markers, (2) propagation, (3) functional genomics, (4) marker-aided selection/breeding, and (5) genetic modification. It should be emphasized that only one out of these five categories deal with genetic modification and its state-of-the-art is only experimental. However, common perception does not differentiate among categories and all are considered as genetic modification. This report represents an attempt to inventory and describe these activities. It is the aim of this document to reflect on the relevance, feasibility and operational aspects of a global review of biotechnology applications in the forestry sector, including present uses and foreseeable compilation and synthesis of forest biotechnology into a unified report detailing current global capability, development, applications, deployment and implications of each tool.

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1. INTRODUCTION

The current working definition of biotechnolology used by the FAO refers to any means of developing or using living organisms to produce or alter or improve a product or organism for a specified purpose (UNEP 1992; Schmidt 1997), which would include prehistoric plant and animal domestication. A more current perspective specifies biotechnology having commercial applications, featuring deliberate manipulation of the genetic components of living organisms or their products (IBPGR 1991; Iowa State University 1994).

These definitions, although accurate for the specific purposes for which they were intended, contribute to the prevalent confusion surrounding biotechnology and in particular forest biotechnology. In some cases, biotechnology is associated with genetic modification, and in others it can be used to define a broad spectrum of modern methods applicable to forest science. We propose a compromise between the two; more specific than the former, yet less utilitarian than the latter:
"*the use of the whole or targeted portions of organisms to provide quantitative information and/or desired products, including the isolation and/or manipulation of specific genetic components of that organism*".

This would encompass both conventional plant or animal breeding as well as more modern developments which focus on only a portion of a biological system. The main reason for the proposed new definition is to clearly separate the part that is based on the generation of information and/or products without the intentional deliberate genetic manipulation from those methods that are exclusively require genetic modification. This approach is helpful in-light of Stone (2002) termed as the "global war of rhetoric" between supporters and opponents of food and agriculture biotechnology. It should be highlighted here that, technical advances in forest sciences, in general, followed the footsteps of that of agriculture, so it would not be a surprise to see the same controversial issues repeated in forestry.

Many recent publications, including working groups and conferences of the FAO, have dealt with various aspects of biotechnology in the forestry context (e.g., Krutovskii and Neale 2001; Yanchuk 2001; FAO 2002). During the past 15 years, this field has seen rapid development of new technology and a large increase in the number of groups involved in research and applications around the world (Ellis et al. 2001; Campbell et al. 2003). Research and testing of modern biotechnology in the initial stages requires up-to-date laboratory facilities. While the technological tools for forest biotechnology are typically adapted from an agricultural background, the subsequent applications and impacts differ substantially when applied to trees (Owusu 1999; FAO 2002). Biotechnology in forestry, for example its use in intensively managed plantations, engenders a host of issues not addressed by agricultural models, center to those is the relative long rotation required for forest crops to reach its economic maturity. The majority of these differences originate from the fundamental life history characteristics of trees as sessile, long-lived, out crossing organisms which can disperse pollen and seed across very long distances, and would likely be planted within potential mating proximity of wild, compatible populations or related species (DiFazio et al. 1999). Other differences between agriculture and forestry which must be taken into account stem from public perception: people often regard trees as essential components of forest ecosystems which perform many functions and provide many tangible and metaphysical values simultaneously, unlike most crop plants (Owusu 1999; FAO 2002).

Although anthropological influences are tractable in most of the world's forests, and managed forests have been the norm in parts of Europe for centuries, trees have only been partly domesticated in the past half century. In effect, very few of these trees are only removed one or two generations from their wild congeners (Libby 1973). Since trees have fairly long generation times, breeding and deployment populations and even clonal lines produced through various vegetative propagation methods including biotechnological methods are still very similar to their wild contemporaries, unlike many agricultural crop plants (Campbell et al. 2003).

For example, teosinte has been transformed through millennia of traditional and intensive breeding into a narrow range of maize or corn lines, with a drastically different mating system, range, phenology, physiology and associated genetic characteristics.

2. TECHNOLOGICAL CATEGORIES OF FOREST BIOTECHNOLOGY

Recent literature varies with respect to the categorization of forest biotechnology (e.g., Haines 1994; Yanchuk 2001; FAO 2001). Due to the gradual evolution of our understanding of genetics, and the consequent development of new tools and technologies by refining and combining new knowledge with prior research, these categories overlap to some degree. We propose five major categories, each of which can be applied to a characteristic subset of applications (see section 3, and Tables 1 and 2):

1. *Markers (biochemical and molecular)*
2. *Propagation & multiplication*
3. *Genomics (functional, structural, comparative, associative, statistical)*
4. *Marker-aided selection & breeding*
5. *Genetic modification*

These tools and their utility are applicable at different scales, from the individual cell or plant to the landscape level (Table 1). The only category which results in genetic manipulation of living trees is (5); although development of some markers and DNA libraries for genomics may involve transformation of bacteria in the laboratory. The impact of these biotechnologies will therefore vary with the specific use, as will the associated benefits and risks (Table 2, Arntzen et al. 2003). Certification of forest products, forest companies and lands is also potentially affected by the application of technology. There are many certification agencies around the world, and some, such as the Forest Stewardship Council, have specifically excluded genetically engineered trees from certifiability, or the land on which they grow, or other products obtained from stands containing these trees (FSC 1996). Some clonal forestry operations are certified, but most agencies have no explicit guidelines concerning genetic applications. Industrial processes which utilize genetic transformation of enzymes to chemically break down lignin in the harvested wood, e.g., pulping using enzyme digestion, have been certified on the merit that this reduces toxic chemical use and discharge. Thus, certification standards vary among agencies, countries, products, processes and applications. Public opinion regarding genetic engineering is more amenable to downstream (post-harvest) compared to upstream (pre-harvest) applications; the main cause for concern appears to be the potential for release of transgenic trees into the environment, which is only an issue for the latter (Pew Initiative 2001; Gartland et al. 2002).

Developed and developing countries have different priorities and applications for this technology (Table 2, Anonymous 2003). DNA-based applications (some markers, genomics and genetic modification) require a large initial outlay of resources, may have continuing high costs depending on the project, and entail a highly trained work force (Ritland and Ritland 2000; Industry Canada 2001). Developed nations have so far been leaders in developing and applying these technologies for a variety of purposes (breeding, commercial and conservation; public and private uses), however, it should be stated, based on the agriculture model, that barriers to the flow of capital and expertise associated with the introduction of biotechnology to new territories are virtually non-existent.
In other words, the introduction of forest biotechnology tools to developing countries could be fast as long as economic opportunities are present. Government agencies, in developed countries, typically provide much of the funding and infrastructure for the development of basic biotechnology research, however, it should be stated that research and development associated with any potential commercialization are mainly driven by the private sector. The shift of funds supporting biotechnology from the public to the private sectors requires a new way of viewing the introduction of these tools into the forest sector.

Therefore, published information indicate that the majority of known forest biotechnology activities in developing countries have been more restricted in their scope, primarily using markers and propagation, however, other tool could be introduced into the private sectors and information on the type of biotechnology application, their place of introduction and their frequency are unknown. The FAO Biotechnologies in Developing Countries (BioDeC) program (http://www.fao.org/biotech/inventory_admin/dep/default.asp) could be a useful storage system for forestry biotechnology developments.

The flow of some biotechnology through teams including partners from both affluent and developing countries, such as CAMCORE, a collaboration between the U.S.A., central and South American and central African nations, have provided increased access to these resources and tools for less wealthy regions, e.g., southeast Asia, Africa and eastern Europe.

Risk assessment is a critical application of many of these new biotechnological tools, and must, in turn, be applied to innovations as they are developed (Government of Canada 1985; Owusu 1999). Targeted gene manipulation is of key concern, both with respect to inserting and eliminating gene function. Gene flow assessments are being conducted for poplars (e.g., DiFazio et al. 1999), but much more information is required to allay public concerns and ecological hazards surrounding the potential for gene escape (Pew Initiative 2001; FAO 2002; Giles 2003). This applies to pollen and seed dispersal, as well as the ability of species to sprout via roots or stumps, become vigorous weeds, or hybridize with wild sympatric congeners (Government of Canada 1985; Crawley et al. 2001; Pilate et al. 2002; Dalton 2002; Adam 2003). Escapes and introgression of agricultural genes have been documented, as has unauthorized planting of genetically modified crop seeds, leading to assessment and regulatory problems. The relative fitness effects conferred by each novel trait must be evaluated in the environmental and genetic context of the field in order to properly assess risks (Johnson and Kirby 2001; Pilate et al. 2002; Anonymous 2003; Dalton 2003). Promoter genes and other biosensors are being investigated as means of tracking transgenic material, a necessary precondition prior to approval for field testing or deployment. Experts have widely concurred that a case-by-case examination is necessary for approval of transgenic trees (Government of Canada 1985; Heron and Kough 2001; FAO 2002; Arntzen et al. 2003).

3. APPLICATIONS

The high cost of biotechnology will likely steer its primary use towards short-rotation plantations as opposed to less intensively managed forests, but some applications are suitable for all forest types and can guide forest conservation activities (see Tables 1 and 2) (Sedjo 1999; Yanchuk 2001). An attempt indicating the status and trends of each tool will be made in the following section.

3.1 Markers (biochemical and molecular)

Biotechnology has progressed from phenotype to genotype, and has been trying to quantify relationships between the two ever since. Early work focused on biochemistry (e.g., terpene analysis), progressed to biosynthetic constitutive or induced protein expression (isoenzymes) and is now utilizing DNA-based tools (e.g., microsatellites, quantitative trait loci - QTL) since the advent of PCR technology.

Molecular markers can either be dominant - only the dominant allele is expressed if both parents are genetically different at a given locus, or codominant – the genotypes of both parents are quantifiable at a locus. Dominant markers (amplified fragment length polymorphism – AFLP, randomly amplified polymorphic DNA – RAPD) require larger sample sizes for statistical analysis than codominant markers (isoenzymes, microsatellites/simple sequence repeats – SSR, restricted fragment length polymorphism – RFLP, sequence-tagged sites – STS, expressed sequence tags – EST, single nucleotide polymorphisms – SNP).

The different markers all have varying benefits, drawbacks, costs and ease of development, and all can be applied to a limited range of optimal applications, generally focused on neutral or non-adaptive genetic variation (see Table 1, Ritland and Ritland 2000; Krutovskii and Neale 2001, and others for reviews). Molecular tools have been widely used to measure gene flow and genetic diversity of natural and artificial populations of forest trees and associated species, and the impacts of anthropogenic disturbance on their evolutionary potential (Haines 1994; Ritland and Ritland 2000; Yanchuk 2001). The long-term viability of species and endemics subject to influences like pollution or climate change can be modeled using genetic data. There are still no highly repeatable, easily assayed markers directly linked to quantitative traits.

Although progress has been made identifying QTLs for some adaptive traits in some species, a good genomic linkage map is essential, as are breeding and identification and sequencing of candidate genes for quantitative traits of interest. The application of markers technology in forestry is by far the most extensive among other tools. It is not restricted to neither developed nor developing countries, however, the type of markers, the speed of its use, and frequency of utilization differed due to the status of markers development and the biometrical methods needed for data analyses (Table 1 and 2).

3.2 Propagation & Multiplication

Clonal forestry using both hardwoods and conifers is gaining popularity due to the resulting uniformity and ease of silviculture, harvesting and processing. More importantly, it allows the capture of level of genetic gains that can not be attained through sexual reproduction. This technology is restricted to organogenesis and somatic embryogenesis, which require extremely detailed understanding of cell biology, multiplication, biochemical signaling, differentiation and the production of copies or clones in a laboratory setting. Both of the above methods capitalize on plant species' ability to regenerate an entire genetically identical individual from a single cell or group of differentiated or undifferentiated cells via *in vitro* tissue culture. Individuals with selected traits or the highest performers from testing trials or breeding programs can be replicated at a large scale, capturing both additive and non-additive genetic variation from traditional breeding. The advantage of these methods is the ability to indefinitely store regenerated tissues in liquid nitrogen or under laboratory conditions (i.e., hold the genotype constant) while field testing is underway to identify elite lines. This allows selected lines with desirable attributes to be further multiplied for operational deployment. Currently some operational scale productions are underway in both developed and developing countries, however, it should be stated that the scale of production in hardwoods is larger than that of softwoods.

3.3 Genomics

Our understanding of fundamental biology and evolution is also being enriched through collaborative efforts using model species in genomics research around the world (Krutovskii and Neale 2001). Evolutionary synteny among taxa, even those as distantly related as algae and angiosperms, can be quantified and candidate genes or groups of proteins (microarrays) involved in biochemical pathways of interest can be identified easily: this process of identifying putative genes controlling certain traits is called gene discovery.

Expression of key gene families or microarrays and their relative up- or down-regulation following stress or environmental changes can be gauged, possibly leading to identification of compounds of significant importance, and their interactions with genotype and environment. Characterization of genetic components of disease or pest resistance is a rapidly expanding field (Ellis et al. 2001; Gartland et al. 2002). Genomics is a fairly new field with many sub-disciplines (Krutovskii and Neale 2001), which requires substantial investment for start-up and maintenance: high-technology automata, costly supplies and chemicals, PCR and other machines, highly trained laboratory staff, marker (EST, SNP) development, as well as vast bioinformatics and associated statistical capacity are necessary. At present, the majority of activity in this area is at the research level and is restricted to developing countries.

3.4 Marker-aided selection & breeding

While the phenotype is the desired end result of breeding, variation associated with differences in expression, genotype-by-environment interaction and non-additive or epistatic genetic variability necessitates large, costly and time-consuming and field trials for trees with traits of interest (Haines 1999; Yanchuk 2001). Mendelian inheritance and our increasing ability to understand and target specific genes have enabled the development of marker-aided selection to complement traditional breeding (Haines 1994; Campbell et al. 2003).

This technique has been used with tremendous success in agricultural crop breeding for various genes and traits over the past two decades, and is increasing in importance as more comprehensive genetic maps, and the locations of QTLs on those maps, are developed for each species. If marker-assisted selection can be used to characterize and select tree genotypes, substantial cost savings may be realized by a much shorter breeding cycle (i.e., more rapid turnover between generations) (Haines 1994; Sedjo 1999). Although based on tools developed using markers, it requires the use of material of known pedigree and integrates the study of specific structural, functional and morphological attributes of species' genomes. Although this work is experimental at this stage, several studies in both developed and developing on hardwoods and softwoods are underway. Exact account of these studies and their state-of-the-art are sketchy due to their mixed funding nature (i.e., public and private).

3.5 Genetic modification

Research and the potential for deployment of genetically modified trees have caused widespread public concern. Regulatory agencies have called for more research on gene flow, likelihood of horizontal and vertical gene transfer via escapes, hybridization and introgression, and a range of ecological impacts of transgenic material in the field (Owusu 1999; Johnson and Kirby 2001; Pew Initiative 2001; Gartland et al. 2002; Anonymous 2003). Concerns around transgenic trees share similarities with agricultural crops, but while the latter are grown and used for research and consumer products throughout many developed and developing nations, transforming trees has only recently begun, and is currently only at the experimental stage. In the laboratory, transformation has been achieved using ballistics and *Agrobacterium* species for three conifer and ten hardwood genera of commercial importance. Specific genes and regions, primarily conferring insect or disease resistance, sterility, and wood quality attributes, have been the focus of nearly all of the research (Pilate et al. 2002; Campbell et al. 2003). Some promoter and marker genes have also been tested. In some regions, pulp and paper processing has achieved dramatic reductions in the amount of highly toxic chlorine and other chemicals by employing enzymes genetically modified to digest lignin (Ellis et al. 2001; Pilate et al 2002; Campbell et al. 2003). The use of genetic engineering during industrial processing does not currently appear to cause public concern, especially when the outcome includes substantial environmental benefits. Accurate account of this type of work is very difficult to document since the majority of work is being conducted by the private sector and its associated growing negative publicity.

4. THE NEED FOR GLOBAL ACCOUNT OF BIOTECHNOLOGY IN FORESTRY

There is, to date, no single database collating the different forest biotechnology activities around the world. This is necessary to gauge current concerns and predict future trends, impacts and needs of this rapidly expanding field. International regulation of biotechnology is inconsistent, and even varies between jurisdictions. Research and development being done in the private sector subject to confidentiality or intellectual property agreements, patent protection or corporate competition preclude or prevent dissemination of some types of information. Other forestry biotechnology applications occur on private lands, or in areas where there is no requirement or framework to report their implementation or extent. An easily accessible global compilation and synthesis would be an invaluable aid for regulatory bodies in adopting relevant and informed legal and approval frameworks around forestry biotechnology.

The amount of information required, the means to obtain it from the appropriate range of sources (e.g. governments, academia, private sector, etc.), and the required depth and breadth of the topic is beyond the capacity of one individual or organization that is based in a single location to obtain and amalgamate in a timely manner. We propose to subdivide this project into parallel, congruent subsections by region and application, which will be accomplished by several independent individuals or agencies with specific expertise from different disciplines and regions. When complete, these components could be collated and cross-referenced into a unified detailed synthesis, eliminating redundancies and comparing and contrasting the different regions. A summary and links to the complete report would be made available through the FAO website, similar to the BioDeC resource, or as a parallel link, possibly incorporating a similar searchable database.

The global review would include separate state-of-the-arts that provide clear trends and applications for the five proposed categories. Additionally, section on the safety and regulatory concerns associated with genetic engineering of forest tree species that covers the approval process of genetically modified trees testing, enforcement, and ultimately approval for deployment is needed.

5. CONCLUSION

This document is prepared to highlight the urgent need for a global study on the status and trends of biotechnology applications into the forestry sector. Technical development of forestry, in most cases, mirrors that of agriculture and our experiences from agriculture indicate that there is no need to let history repeat itself. In fact, the ambiguity surrounding the use of biotechnology tools into forestry has already caused confusion. As stated above, only one out of five possible biotechnology categories in forestry involves the deliberate genetic manipulation and its state-of-the-art is only experimental, however, public perception may preclude or color the usefulness of the other categories into forestry. It should be highlighted that the FAO is the most suitable organization to take the lead role in this important undertaking due to its neutrality and global presence.

The diversity of topics and the variable level of competence in the field of forestry biotechnology require cross-functional team effort. Wide contribution that covers species and geographic representation is required to ensure accuracy and balance.

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Table 1: Anticipated contribution to and scale of impact of each broad area of biotechnology on elements of natural and artificial forest populations

| Applicable Forestry component | Spatial Scale | Development Elements Relevant to Biotechnology | Broad Technologies | | | | | | |
| | | | Molecular Applications | | | | | | Cloning/ Regeneration |
			Bioinformatics	Diversity Measurement	Gene Discovery	Genetic Manipulation	Simple Biosensors	Product Verification	
Natural Populations	tree - population	Genetic Resources Characterization	X	X	X				
	population	Mating System/Gene Flow		X					
	population - landscape	Conserving Diversity		X					
	population - landscape	Silvicultural Impact Assessment		X					
Breeding Populations	tree	Selection	X	X	X				
	tree - population	Mating Designs		X					X
	tree	Progeny Testing	X	X	X			X	X
	tree	Attribute Assessments	X	X	X		X		
	population	Diversity Management		X	X				
Production Populations	population	Mating System	X	X					X
	population	Gene Flow/Contamination	X	X				X	X
	population	Seed Orchard Management	X	X				X	
	population	Seed Orchard Design	X	X				X	X
	population - landscape	Silvicultural Impact Assessment		X			X		X
Regeneration	stand	Natural		X	X			X	X
	stand	Artificial		X	X	X		X	X
Domestication	population	Native Species Diversity	X	X	X		X	X	
	population - landscape	Native Species Growth/Yield		X			X	X	
	population	Exotic Species Risk Assessment	X	X	X		X	X	
	population - landscape	Exotic Species Growth/Yield		X			X	X	
Gene Conservation	population	Diversity Assessment	X	X					
	population	Gene Flow/Contamination		X			X		X
	population	Effective Population Size		X					
	tree	Reproduction		X	X		X		X
Forest Health	tree - stand	Risk/Hazard Assessment	X	X			X		
	tree	Resistance Screening	X	X	X	X	X		X
	stand	IPM Options					X	X	
	tree - landscape	Other Pest Control		X	X	X	X		X
Processing/ Value Added	stand	Pulp Processing	X		X		X	X	
	stand	Wood Treatment	X		X		X	X	
Marketing	tree	Chain of Custody	X		X			X	
	stand - landscape	Certification		X		X	X	X	X
	stand	Product Description	X	X	X			X	

Table 2: Current global applications and projected future importance and trends of forest tree biotechnology
A = developed countries; B = developing countries; on a scale of 0-3: 0 = nil, 3 = common.

Broad Technologies	Components	Current Applications A	Current Applications B	Projected Trends
Bioinformatics	*Large databases:*			The storage, retrieval, analysis, and interpretation of large amounts of biological data will cross boundaries of all broad biotechnologies. Capability and application range of this tool will continue to increase dramatically. Mining the massively increasing amounts of data at all scales, and integrated analyses and syntheses of these data will greatly increase power to detect genes and understand their functions. The databases developed for sequence and microarray data in particular must be co-ordinated regionally, nationally and internationally, and accessible to the public. Bioinformatics research requires resource-intensive, multidisciplinary teamwork, naturally leading towards more international cooperation over a range of study scales and systems. Opportunities are opening up for developing countries to become involved in these projects.
	Targeted DNA Sequence	3	0	
	Proteomic	2	0	
	Gene Mapping & Markers	3	2	
	Microarray	2	0	
	Phenotypic	3	3	
	Integrated Applications	1	0	
Diversity Measurement	**mtDNA**	2	0	The use of molecular markers for studying natural and artificial forest tree populations has undergone unprecedented expansion due to the vast array of population genetics applications they have enabled. These topics include measurement of genetic diversity within and among populations, comparisons among taxa, historical reconstruction and prediction of species' range shifts, gene flow, assessment of natural and artificial (e.g., seed orchard) population mating system parameters, introgression and hybridization. Markers have also been used to evaluate the impacts of domestication and silviculture. Genomic and QTL mapping have recently expanded due to the development of unlimited numbers of markers. Anticipated development cost reductions for SNPs will trigger an increase in their use in all forest genetics resources applications.
	cpDNA	2	0	
	RAPD, AFLP, RFLP	3	2	
	Microsatellite (SSR)	1	0	
	SNP, ESTP	3	1	
Gene Discovery	**Phenotypic Traits**	3	3	Phenotypic and quantitative trait measurements represent the backbone of all conventional tree breeding. Their proven efficacy and ease of use has resulted in significant gains in many species worldwide.
	QTL Mapping	3	1	QTLs will likely increase in importance, particularly for important or hard to measure traits, especially those which require older material for assessments (e.g., all wood properties, disease and insect resistance). The focus is shifting from flanking region markers to markers within the actual gene or QTL of interest.
	Genome & EST Sequencing	2	0	Massive, redundant conifer genomes will restrict sequencing to a few regions of interest. EST sequencing for commercially important species will accelerate within 5 years. Whole genome sequencing has begun for *Populus*; the data will be freely available; other economically and ecologically important species will follow. International collaboration is important for species whose ranges cross international borders.
	Microarray Analysis	2	0	As microarrays become available for gene discovery for growth and yield, wood quality and adaptive attributes (e.g., disease and insect resistance and stress tolerance), more reliable oligo-based will likely replace cheaper clone-based arrays. The potential benefits and challenges will spur international collaboration. This new field is likely to prove most cost-effective for understanding gene function, rapid production technology development for advanced breeding programs and in high yield plantations.
	Proteomic Analysis	2	0	Notes for microarray analysis also apply here. Proteomics airms to elucidate protein variation beyond simple transcriptional regulation, including levels of expression, interactions and post-translational modification.
	Metabolomics	1	0	Still in the initial stages, metabolomics asseses the presence/absence of non-protein structural precursors of essential components in biochemical pathways.

Broad Technologies	Components	Current Applications		Projected Trends
		A	B	
Molecular Genetic Modification	**Gene Insertion/ Sequence Modification**	2	1	Inserting foreign targeted genes into tree genomes has profound potential. This method will enable cross-species gene transfer in cases where it is not possible via conventional breeding. Transformation is widely used in agriculture, but is new to forestry and has engendered major public contention and strict biosecurity protocols for testing and deployment. Some countries and organizations have restricted testing and planting genetically modified trees. Potential for gene escape into wild populations needs further study for risk assessment. Reproduction must be eliminated or postponed past rotation age. This technique could potentially improve fibre yield and quality as well as other important qualitative, quantitative and adaptive attributes.
	Gene Targeting/ Knockout	1	0	Silencing gene function is the complement of gene insertion. This technique will expand as the results of microarray and proteomic analyses accrue. Similarly, there is a broad spectrum of potential functional genomics applications and associated ethical issues.
Product Verification	**Pedigree Verification**	3	3	Development of larger-scale, lower cost application platforms will increase accessibility by genetic improvement programmes to this technology, expanding the prevalent uses: retroactive pedigree verification and verifying clonal identies in seed orchards. Other potential applications include determining the efficacy of seed orchard management techniques on a large scale, and consequent growth and yield determination of resulting crops.
	Quality Control/ Quality Assurance	1	0	Rapid developments in product description systems, commonly used in food manufacturing facilities, will result in increasing support for research and application of automated product verification systems. The main technology is PCR-based, e.g., DNA markers to detect mislabelled products (e.g., clones). Functional genomics is expected to yield suites of gene markers that can be used to check process efficiencies during mass production of clonal seedlings.
Cloning	**Organogenesis**	1	1	Micropropagation (organogenesis) and gametic or somatic embryogenesis require treating tissue explants with growth regulators to induce bud or shoot formation. Most of the shortcomings of organogenesis can be overcome using somatic embryogenesis, but methodology and success are species dependent. These methods are currently in production and cloning will be a significant element in high yield plantation forestry.
	Somatic Embryogenesis	1	0	
Biosensing	**Simple Biosensors**	1	0	Measuring and monitoring components of clonal production will increasingly employ sensors comprising physical, chemical and molecular markers to detect specific biological processes, e.g., expression-tagged genetic markers to quantify or detect presence/absence of metabolic pathway components of interest.