Tree Transgenesis

M. Fladung D. Ewald (Eds.)

Tree Transgenesis

Recent Developments

With 19 Figures, 8 in Color, and 23 Tables

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Preface

A continuous development in plant biotechnology including gene technology has been observable during the past 20 years. Different methods elaborated with model plants were also applied to forest trees on a larger scale. Whereas in the beginning the meaning of the term “plant biotechnology” embraced a wide variety of meanings like, e.g., regeneration of plantlets via tissue culture, embryo rescue, somatic embryogenesis and gene transfer, the focus of this term has changed more and more. Nowadays, it is the transfer of genes which comes into mind when plant biotechnology is discussed, including of course the evaluation of all challenges and risks related to gene transfer methods.

Compared with annual plants, especially in the field of agriculture, the work and the progress with transgenic trees is still in its infancy. Nevertheless, but often unnoticed by the scientific community, there are a few countries which already allow the commercial use of a restricted number of transgenic tree clones after different critical steps of approval. This and the ongoing improvement in transgenic research in trees led to the idea of preparing a summary of the present state of the art from different points of view. With the help of a number of authors directly or indirectly involved in tree transgenesis, this book was produced. Based on scientific results it is aim of this book to inform the reader about the present state of the art and to stimulate discussion concerning problems of biosafety and risk assessment and the necessary experimental tasks in the future, as well as to support decision-making processes in politics.

In view of the availability of the whole genomic sequence of poplar (http://genome.jgi-psf.org/Poptr1/Poptr1.home.html) and also, in the near future, of *Eucalyptus* (http://www.ieugc.up.ac.za/ieugc_Main.htm#), gene technology is a valuable scientific tool to down-regulate or over-express single genes and, thus, study their role in plant growth and development. Such a functional genomics approach will allow us to unravel the basic principles of plant growth regulation one day. Thus it will soon be possible to improve transgenic trees mimicking natural strategies including their use for a sustainable application.

However, trees also need our special attention as unbred and long living individuals. In most cases, forests consist of wild populations of trees with great importance regarding both the climate and the sustainable provision of wood. Therefore, it is justified to take special care concerning risk assessment and biosafety issues to prevent an undesired environmental release of transgenic trees by chance.

There is still an urgent need for ongoing research in the field of biotechnology in the near future. All aspects have to be included in these research strategies not only to estimate the risks properly but also to come to a critical evaluation concerning chances and challenges of transgenic trees to meet the future growing demand of the renewable resource wood.

April 2006

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Part A Transgenic Trees in the World

1 Field Trials with Transgenic Trees – State of the Art and Developments

MARCEL ROBISCHON

1.1 Introduction

Research and development on transgenic trees differs from such work carried out on herbaceous model systems first in that it necessarily involves field trials if data on aspects of the mature plant are required. Second, in contrast particularly to field tested transgenic agricultural crops, GM tree field trials are bound to be extended with the same plant individuals over longer than one single vegetative period and can last many years.

Given this, and taking into account the fact that in all cases a large amount of work has to be done before beginning any work beyond the test tube stage in the growth room and a potted plant in a greenhouse, the development of field trials and field releases worldwide is expected to be an indicator for overall development in the field of forest biotechnology.

1.2 Transgenic Trees in Test Tube and Field Trials

A field trial is expected to document in itself a well-developed research project that has led past various testing phases in lab based work to a stage in which the tree can be taken to the next round of tests in the field. It is, however, not just the success of the primary lab-based work that, under consideration of all the other factors, influences what happens in the field. The success of the field trials will also determine whether in the future more work is invested in the lab-based work. The dimensions of field releases of transgenic trees in trials can therefore only with great care be seen as a direct, simple function of the progress made in development in the lab. Many other factors come into the equation.

The closer research and development with transgenic trees gets to the field trial or release and thus the closer it gets to structures of primary production in "classical" forestry, the more it carries on some of the burdens of technical "peculiarities" and socio-economic involvements that are typical for forestry

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worldwide. On the technical and economics side, long production times can be identified that on the one hand cause a low return – whether in a classical management scenario or in the development of a new GM-tree based product – and on the other hand delay the progress of research and development considerably (Speidel 1984). On the social side the involvement of many stakeholders is typical and is found in either, e.g. the afforestation of a stand near a settlement or the start of a field trial (Köpf 2002).

This latter point is well reflected in the fact that a transgenic tree, if studied as a "tree" rather than a "seedling-like plantlet in a test tube" with the release of a transgenic plant in the long term and some potential environmental implications causes a great deal of concern to the public, as documented in a flood of non-technical and newspaper articles, media reports, political and lobbying activities and in some cases vehement protests (Arthur 1999; Highfield 1999; Miller 2003).

Keeping in mind the aforementioned limitations, the number and type of field trials, and the development of these data over time give an impression of both, work on GM trees already carried out successfully at an earlier stage of the development process, but also gives an impression on what further research may build up and, if in the context of present economic and political developments, into what directions future work may be pushed. In the following it is attempted to provide an overview of past and present field trials worldwide, with the aim of developing an image that allows some insight into the future developments that may shape work on transgenic trees.

The global situation regarding releases of transgenic trees to the field is nowhere documented completely and in detail. The main reason for this is a distinct lack of data and information. This is partly due to the nature of some data as "confidential business information" as in recent years a large percentage of field releases were carried out by the Research and Development labs of large forest companies, particularly outside Europe. Some companies, when approached by researchers or journalists, clearly stick to a "no-information" policy, leaving requests ignored and questions unanswered.

Obtaining information is in many cases a particular challenge, as the respective companies are often joint ventures between various other firms, often in the pulp and paper industry, or other industrial branches and are subject to frequent change by merger, takeover, sale, closure, re-naming etc. or partners leaving the joint venture. Under such circumstances it can also be difficult to trace back the historic continuity of work carried out by individual companies. This is true not only specifically for firms that carry out GM work on trees, but also for other companies in the field of industrial and plantation forestry (Carrere and Lohman 1996).

Some insight however is possible due to the legal and administrative structures in some countries that require permission for field trials and list releases together with some limited information in publicly accessible databases.

Therefore, for the following overview several sources that are quite different in nature have been used. For the US there is a detailed database listing all applications for permission, and respective notifications of a field release of a transgenic organism, which also includes trees (http://www.aphis.usda.gov/brs/status/BRS_public_data_file.xls). Equally detailed is the Canadian database published for all field releases of transgenic plants online at http://www.inspection.gc.ca/english/plaveg/bio/triesse.shtml. The same field releases are also partly covered in a database that lists the equivalent applications for Europe, US, Canada, Australia and New Zealand that is provided by OECD (Organisation for Economic Co-operation and Development) (http://www.olis.oecd.org/biotrack.nsf). The situation in the EU is separately documented in an EU database (http://biotech.jrc.it/deliberate/dbcountries.asp and http://gmoinfo.jrc.it/gmp_browse_geninf.asp).

While these databases are thought to be comprehensive, they do not give any specific information on the size of the respective field trial nor on whether this trial has in the end actually been carried out, or indeed at what point in time it has actually been terminated. They also do not show, whether an application for or a notification of a field trial is for a completely new experiment or simply the continuation of an earlier experiment with plants of the same type – or even the same plant specimens.

However, the regulative frameworks in many countries are at present still being developed. In these cases information was sought on work carried out in the respective country via academic networks. This data is backed up with information from scientific publications, non-technical publications and newspaper articles, environmentally concerned publications as well as personal communication with researchers and persons involved in environmental NGOs (non-governmental organization). It is an inherent problem of the evaluation of a range of diverse sources that the information obtained may in some cases not match or even be contradictory.

In this overview, first work on forest trees is covered. This includes species whose traditional use falls in either of the three classical functions of managed forests: production of timber and non-timber forest products, protection of the landscape, and the recreational function (Dieterich 1953). Trees whose main function is the production of fruit are discussed in a separate section. In addition there are also a few examples of genetically engineered trees in field trials, whose potential economic application is in the production of an entirely new product or service that is only tenuously linked to the traditional use of trees in forestry and fruit farming.

There are a few examples of transgenic trees that have been genetically modified to improve their use for ornamental, landscape or environmental purposes, which however do overlap with the function of creating a more productive forest crop. There is one field trial documented for *Amelanchier*, which has mainly ornamental use. In numbers such trials however are completely irrelevant and are mentioned here solely for completeness.

1.3 Transgenic Trees for Improvement of Forestry

1.3.1 Northern America

The region in which the largest number of field trials on transgenic trees has been carried out is North America.

Even though a country with traditionally strong research in forest biology, the share of field releases of transgenic trees in Canada is small. The Canadian database lists for 1997 a poplar with an antibiotic resistance released in Quebec (the only one found also in the OECD database), for 1998 a submission for herbicide tolerant poplar in Alberta, and from 2000–2004 two submissions for black spruce with selectable marker genes, an insect-resistant white spruce and a poplar with a selectable marker. These trials are well covered in the non-technical media. A "National Post" article of 2003, for example, covered a planned field trial with transgenic trees (Jack 2003). The trial comprises 400 transgenic spruces and poplars planted out in a forest near Val Cartier, Quebec. The article pointed out that there were as yet no commercial plantings of transgenic trees in Canada, but that the development by now had reached a point at which use in commercial plantations was within reach. This work was also publicised in CBC (Canadian Broadcasting Corporation) News (2003), quoting Armand Séguin of the Canadian Forest Service, according to whom this was the only field trial with transgenic trees in Canada.

The vast majority of field trials in North America to date took place in the US, for which the database (March 2005) documents about 185 applications respectively notification for releases of genetically engineered forest trees (Table 1.1).

Table 1.1. Present number of field trials with transgenic fruit and forest trees comparing Europe and North America

	Fruit trees Europe	Forest trees Europe	Fruit trees North America	Forest trees North America
Marker	–	7	3	45
Herbicide resistance	–	3	–	45
Insect resistance	–	1	13	15
Disease resistance	11	1	47	12
Sterility	–	1	3	28
Lignin	–	8	–	27
Developmental	6	4	3	27
Heavy metal	–	3	–	6
Fruit quality	–	–	21	–
Other	4	2	3	5
Total	21	30	94	212

For comparison in this paper these trials were grouped according to the nature of the altered trait (herbicide tolerance, insect resistance (Chap. 12), disease resistance (Chaps. 10 and 11), sterility or altered fertility (Chap. 2, Sect. 2.4), lignin content (Chap. 5), developmental traits, heavy metal tolerance (bioremediation, Chap. 7), or other traits. A clear change over time in the type of traits for which field trials were applied for respectively notified could be observed.

The work on *herbicide resistance*, for example, so far “peaked” in 1999 (Table 1.2) with the number of trials for this trait decreasing since. With the long term investments that forestry naturally involves (Speidel 1984), the altered trait has to be of potentially high economic significance. This may partly explain, for example, the reduction of experiments on transgenic trees with herbicide resistance. The then director of Weyerhaeuser forest biotechnology was quoted in a 2002 article in *Science* (Mann and Plummer 2002) with the comment that herbicide application in the forest industry “*is not that large of an expense*”. Shifting to different herbicides if necessary may therefore, in the long run, be more economic than generating trees resistant to one particular to allow its extended use. Furthermore, the use of herbicides is a classical environmental issue and hence likely to form a focal point of public criticism.

Table 1.2. Applications and notifications of field trials using transgenic forest trees in the US. The category “other” includes work on gene stability and thus reflects also work on safety aspects. In most cases a release of a plant with a specific trait of interest is accompanied by a release of plants with markers or plants may have more than one trait, including the (visual) marker

Year / Type of trait	‘89	‘90	‘91	‘92	‘93	‘94	‘95	‘96	‘97	‘98	‘99	‘00	‘01	‘02	‘03	‘04	‘05
Marker	1	–	–	–	–	–	–	–	1	1	4	6	10	4	21	31	8
Herbicide resistance	–	–	–	–	–	1	1	1	2	7	13	7	3	7	2	2	1
Insect resistance	–	1	1	–	1	–	–	–	2	3	1	5	1	3	–	11	–
Disease resistance	–	–	–	–	–	–	–	–	2	3	2	1	–	–	1	4	–
Sterility	–	–	–	–	–	–	1	–	1	–	–	2	4	–	6	17	–
Lignin	–	–	–	–	–	–	–	–	–	2	–	2	–	–	6	10	4
Developmental	–	–	–	–	–	–	–	–	–	1	1	–	–	1	4	16	4
Heavy metal	–	–	–	–	–	–	–	–	–	–	–	–	1	–	2	3	–
Other	–	–	–	–	–	–	–	–	1	1	–	–	–	1	–	7	1
Total	1	1	1	–	1	1	2	1	9	18	21	23	19	16	42	10	18

Insect resistance has remained a trait worked on quite continuously with some of the earliest trials being on this trait, but still in 2004 a large number of field trials were carried out with trees modified for insect resistance. This can be easily explained with pest damage being a continuous problem in forestry, in particular given the steady stream of exotic species being introduced into new environments as novel pests (Schedl 1936) and the enormous cost arising from this ongoing "biological globalisation" (Scigliano 1999). In an attempt to make an informed guess of future development work on this trait, it has to be taken into account that a large proportion of the earlier work on insect resistance in trees was carried out with the Bt (*Bacillus thuringiensis* toxin) genes. However, in case it turns out that extended use of Bt-transgenic plants leads to the formation of resistance in pests, for which indication has already been found (ABC News 19th April 2001), the concept of achieving insect resistance may have to be revised. This may lead to more research and more field trials being required in the future.

Work on *disease resistance* seems to follow a similar development, without however the very earliest trials in the early 1990s. After the 1990s the number of trials with this trait seemed to decline. However, even in 2003 and 2004 some trials for this trait appeared again. This is partly due to transgenic methods now under discussion with the aim of healing the wounds that disastrous epidemics have torn into stands of chestnuts in the forests of North American East Coast, or to bring back elms to the suburban streets after they was all but wiped out from natural and cultural landscapes in America and Europe (Campanella 2003). So far there is one field trial with transgenic elm and two field trials with transgenic American chestnut documented in the US database. It is likely that work on this will continue, given the high importance these tree species once had in the Eastern North American landscape (Dr. R.C. Kellison, personal communication[1]). Furthermore, there is also work on transgenic lines of the Chinese Elm (Aziz et al. 2003) suggesting a substantial interest in this problem.

Notably, work to generate transgenic, disease resistant elm has also been carried out in Europe (Gartland et al. 2000) and has attracted considerable attention from the media (Kelbie 2001), as yet however without any field trials. More details on this topic are given in a chapter later in this volume. Work on transgenic lines has also been published for European Chestnut (da Costa Seabra and Pais 1999). It has, however, as yet not lead to field trials, possibly because the problem of chestnut decline does not have the same dimensions as in the US (Prof. Dr. O. Holdenrieder, personal communication[2] and Dr. U. Heiniger, personal communication[3]).

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An increased interest in research in trees with altered *lignin composition* may be interpreted in two ways. First, there is a rise in the production of pulp and paper worldwide. A FAO (United Nations Food and Agriculture Organisation) forecast predicted a growth of global paper consumption to 400 million tonnes by 2010. For comparison, the consumption in 1970 was estimated at 125 million tonnes and 1995 at 276 million tonnes (Enskilda Futures 1997).

Second, the production of paper is a procedure with extremely severe environmental effects. Even though technical progress, including a number of biotechnical developments (Bajpai and Bajpai 1998), aims at improving the situation, it is expected that with the rising paper productivity worldwide these problems will increase, making the development of trees with lower lignin content an economically and environmentally interesting topic (Dr. K. Holt, personal communication[4]).

The importance of work linked to *bioremediation* is obvious with large areas of land being polluted by industrial waste products, including heavy metals (Raskin and Ensley 2000). Since 2001, field trials have been notified for work on transgenic trees with the ability to tolerate heavy metal contamination of the soil. From 2001 to 2004 their number increased from 1 to 3.

The work on *sterility or altered fertility* of trees has increased clearly over recent years. There are two possible reasons for this. First, still in the context of increased demand for wood products, particularly pulp and paper, a reduced fertility is expected to increase the productivity of the tree. Second, environmental reasons are likely to play an increasing role for research in this trait. It is in the interest of both, publicly funded institutions as well as private companies to work on methods to reduce unwanted gene flow from a transgenic crop into natural populations or to prevent uncontrolled spreading of transgenic material.

The somewhat widely defined category "developmental traits" includes work with the aim of increasing yield, but also work that is involved in basic research, onto which more applied projects may build (e.g. nitrogen metabolism (Chap. 8) or disease resistance traits (Chaps. 10 and 11)). Finally, field trials established to study biosafety-related issues like gene or genome stability (Chap. 14) or horizontal gene transfer (Chap. 15) fall in this category. Such research is important in the frame of elevating public acceptance.

It can be concluded that it is evident from the sources used that the number of field trials in North America, as well as the number of traits worked on, has been growing since the first trials at the beginning of the 1990s. There has also been a clear shift in the importance of individual types of traits. This shift can be linked well to the economic and political context. Overall in the work documented in the database for the US, a trend towards the development of a more "sophisticated" and more elaborate use of molecular biological

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methods for potential use in plantation forestry, that also takes into account environmental concerns, can be observed.

1.3.2 Europe

In the EU, according to the EU and OECD databases, there have been about 30 applications for field trials with transgenic forest trees to date. This included 18 on poplar, 4 on eucalyptus, 2 on pine and 2 on spruce. The distribution according to types of traits is given in Table 1.3.

Insect resistance, herbicide tolerance and disease resistance are much less an issue in research in Europe than in the US. Work in field trials with trees modified with these aims appears to phase out around the same time as work with herbicide resistant trees. This can be interpreted in the context of a completely different structure of the forest industry in Europe and the US, with central aspects being the absence of large (i.e. "American dimension") forest companies in Europe (with the exception of Scandinavia). Also large areas of fast growing monoculture plantations to be clear cut after the rotation time for production of pulp and paper are more of an exception in wood production in Europe, with the exception of some fast growing eucalyptus plantations in Spain and Portugal.

Nevertheless, studies on lignin formation that were linked in the previous section – in an US-American scenario – to plantation forestry for pulp and paper, increased in numbers also in Europe over the years. Notably a 1997

Table 1.3. Applications and notifications of field trials using transgenic forest trees in the EU

Type of trait	'92	'93	'94	'95	'96	'97	'98	'99	'00	'01	'02	'03	'04
Marker	–	1	1	–	2	2	–	–	–	–	–	–	–
Herbicide resistance	–	1	–	1	1	–	–	–	1	–	–	–	–
Insect resistance	–	–	–	1	–	–	–	–	1	–	–	–	–
Disease resistance	–	–	–	–	–	–	–	–	1	–	–	–	–
Sterility	–	–	–	2	–	–	–	–	–	–	–	–	–
Lignin	–	–	–	2	2	1	–	1	–	–	–	1	–
Developmental	–	–	–	–	1	–	2	1	1	–	–	1	1
Heavy metal	–	–	–	–	–	–	–	–	–	1	–	–	–
Other	–	–	–	–	–	–	–	–	–	–	–	2	–
Total	–	2	1	6	6	3	2	2	4	1	–	4	1

In contrast to Table 1.2, in this Table marker genes are not listed as a separate trait, unless it is the only trait worked on in the specific experiment trial on lignin was carried out by a large company involved in this type of forestry in Portugal (cf. Table 1.4). In other cases this interest in lignin formation may be due on the one hand to basic research with interest in the basic processes of lignin formation. On the other hand a driving force in Europe may be the interest to contribute in the long run to biotechnical mechanisms to reduce pollution caused by the paper industry (Dr. K. Holt, personal communication).

As in the US the work on developmental traits has set in relatively late but appears to be continuously an important topic worked on, potentially as a basis for future, more applied research.

With reference to work linked to bioremediation (Chap. 7), a trial carried out on transgenic poplar with altered glutathione level in Germany is of particular interest. These trees are supposed to help mopping up heavy metals from the soil (Dr. A. Peuke, personal communication[5]) and reflect a growing interest in environmental applications of transgenic trees in Europe.

The most obvious difference between the development in Europe and the US is that there is a much smaller number of trials and also no apparent trend

Table 1.4. Field trials on transgenic trees applied for or notified by industrial companies in the US and Europe

	‘93	‘94	‘95	‘96	‘97	‘98	‘99	‘00	‘01	‘02	‘03	‘04	‘05
USA total	–	–	–	1	1	6	9	11	15	11	33	65	10
Arborgen	–	–	–	–	–	–	–	–	3	10	30	60	9
Applied Phytogenetic	–	–	–	–	–	–	–	–	–	–	2	4	1
Westvaco	–	–	–	–	–	2	9	10	9	1	1	1	–
Int. Paper	–	–	–	–	–	–	–	1	3	–	–	–	–
Monsanto	–	–	–	–	–	1	–	–	–	–	–	–	–
Weyerhaeuser	–	–	–	–	1	–	–	–	–	–	–	–	–
Union Camp	–	–	–	1	–	3	–	–	–	–	–	–	–
Europe total	1	1	1	1	1	–	–	–	–	–	–	–	–

For the US a specification of the companies involved is given
In Europe the 1993 and 1995 trials have been carried out by Shell Forestry, now dissolved
The 1994 trials was by *Celulosas de Asturias* in collaboration with Advanced Technologies Cambridge
The 1996 trials was by Zeneca
The 1997 trial was by Stora Celbi – i.e. the five industrial trials in Europe carried out by four companies
The 152 industrial trials in the US were carried out by basically 7 companies
In recent years there is also an increasing centralization with the vast majority of field trials being carried out by one single company
Notably ArborGen is a joint venture of Westwaco, Fletcher Challenge, International Paper and Monsanto founded in 1999

[5] Dr. Andreas Peuke, Institut für Forstbotanik und Baumphysiologie, Universität Freiburg, Am Flughafen 17, D-79085 Freiburg

of an increase in their number. The development in Europe mirrors the trends observed in the US in as much as there has been over the years generally a growing number of different traits that have been worked on in transgenic trees. As in the US in recent years, work with traits that may be important for environmental purposes, namely resistance to heavy metals, have emerged.

As a consequence of the different structure of forest industry, a potentially different attitude in the public and the different legal environment, it can be expected that in the nearer future more companies will choose to do work in the US rather than in Europe. In this context it is of interest to compare the trials carried out to date by industry in Europe and in North America.

Of the trials documented in the databases for North America, 162 were, as far as is evident from the applications as listed in the databases, run by industrial companies. All of these were based in the US. The 152 industrial trials in the US were carried out by no more than 7 companies. In recent years there is also an increasing centralization with the vast majority of field trials being carried out by a single company (Table 1.4). Notably, ArborGen is a joint venture of Westwaco, Fletcher Challenge, International Paper and Monsanto. After its foundation in 1999 the applications and notifications of the mother companies phase out.

Of the 30 field trials with forest trees in Europe, as far as it is evident from the applications documented in the databases, only 5 trials were run by industrial companies (Table 1.4). All of these were early trials (between 1993 and 1997) on eucalyptus apart from one case with poplar. Of these industrial trials two carried out in 1993 and 1995 were run by Shell Forestry, now dissolved. The 1994 trial was by Celulosas de Asturias (CEASA) in collaboration with Advanced Technologies Cambridge. The 1996 trial was by Zeneca in a project that was more of an academic nature and run jointly with the French national institute for agronomy research INRA (Institut National de la Recherche Agronomique). The 1997 trial was carried out by Stora Celbi, i.e. the five industrial trials in Europe were carried out by a mere four companies. All the field trials in Europe were relatively short lived. The two trials with eucalyptus in England lasted for three months each. Also the trial at CEASA lasted for three to four months only. The trial by Astra Zeneca and INRA however was worked on for four years and was destroyed by activists shortly before the planned date of termination (Dr. C. Halpin, personal communication[6]). Future work on transgenic trees is not part of the business portfolio of Zeneca at present (Dr. K. Holt, personal communication). On the Stora-Celbi trial there is no further information available. Generally the interest in industry to conduct field trials in Europe appears to have faded away in the past. All field trials on transgenic forest trees carried out at present in Europe are part of academic studies.

[6] Dr. Claire Halpin, Plant Research Unit, School of Life Sciences, University of Dundee at SCRI, Invergowrie, Dundee DD2 5DA, UK

This development is illustrated in the history of Shell Forestry. According to Dr. J. Purse (personal communication[7]), it became clear in the late 1990s, that developing a GM tree crop was too expensive and not cost effective for one single company. Therefore the company initially tried to get involved into joint ventures – as for example with Sappi (South African Pulp and Paper Industries Ltd.) in South Africa (see below). The plan to transform the research branch, after the company's decision to withdraw from work on GM trees in order to obtain FSC (Forest Stewardship Council) certification and to provide the development of GM trees for other companies, failed due to lack of customers. The company withdrew not only from field trials but then from molecular work on trees and was eventually dissolved by the Shell concern altogether (Dr. J. Purse, personal communication).

1.3.3 Latin America

In the past there have been field trials of transgenic eucalyptus in Latin America, namely in Uruguay and Chile. These are however not documented in any of the databases mentioned above, nor is there any scientific publication on any of them. They are however covered in publications produced by environmental groups. The "World Rainforest Movements" (WRM) bulletin comments on field trials with transgenic trees (no species is mentioned, but from other sources it is documented as eucalyptus) run by 'Florestal Oriental', a forest company that then was then jointly owned by Shell and UPM (United Paper Mills Ltd.)/Kymene in Uruguay. According to Pérez (2000) these trials were run only over a period of two years and ended in 1999. All trees were according to this source destroyed. This information was confirmed by Dr. J. Purse, formerly of Shell Forestry. The company was at that time aiming at FSC certification, which would have been precluded if these experiments had been carried on longer. In 2000 there were no transgenic trees in the country (Pérez 2000).

Field trials with transgenic trees were also carried out in 1999 in Chile by a company called 'Forestal y Agricola Monte Aguila S.A.' that then belonged to the same group and was thus basically in-house research of Shell Forestry (Dr. J. Purse, personal communication). It involved a mere 60 eucalyptus plants that were resistant against the herbicide glyphosate. Notably these field trials in Latin America have attracted interest worldwide and references to these are found frequently in NGO literature concerned with environmental issues (Manzur 2000).

The traits of herbicide resistance and altered lignin formation relate as in the case of similar work in other parts of the world to plantation forestry with short rotation times for pulp and paper production. The small scale, short life and the fact that they were terminated without being continued or repeated

[7] Dr. John Purse, Prima Bio, Kent, UK

in this region, however, suggests that these trials were merely an initial test to try out for the first time the new technology by an individual company, but not part of a longer-term development.

In January 2001, Shell produced a press release (Royal Dutch Shell Petroleum 2001) stating that it had received FSC certification for its forest business in Latin America. FSC certification excludes not only the use of GM trees by a forest company in its plantations, but also involvement of the respective company in research. Shell has since withdrawn from this work and from the forest industry entirely.

It may be expected that, for the successor companies, the FSC certification is of importance, in particular with respect to the European market. With the dissolving of Shell forestry and their research branch, the direct link to the technical side of the development has vanished. It therefore does not seem likely that these or similar trials are going to be revived soon.

However, Chile actively supports the development of the biotech sector. This suggests that in the future this country may attract other investors for GM work on trees or may become involved in research with its own institutions.

1.3.4 South Africa

In the past at least one field trial with transgenic trees has existed in South Africa. This experiment was carried out with roundup-ready resistant eucalyptus planted in 1997 in a project run jointly by Sappi, Shell and Monsanto (Dr. J. Purse, personal communication). Dr. Arlene Bayley of Sappi[8] (personal communication) stated that this field trial was terminated after a year, due to a temporary limited permission. The plants herbicide tolerance was tested at an age of 6 and 10 months. According to Dr. Bayley (personal communication) Sappi was, however, at the time of these works, more interested in testing the technology and collecting experience with the legal and administrative processes than in a commercial use.

The situation with regards to transgenic field trials in South Africa is, however, not as easy to assess as in the case of the US and Europe. While a 2002 newspaper article from South Africa (Friedman 2002) quotes NGO Biowatch with the statement that, in 2001, permission was given for field trials on (among other crops) eucalyptus and apples[9], Mrs. M. Vosges[10], South African

[8] Dr. Arlene Bayley, Sappi Forest Research, PO Box 472, Kwambonambi 3915, South Africa

[9] Notably the above quoted article reports an NGO going to court to enforce a clearer information policy on releases of genetically engineered organisms in South Africa, which in itself reflects a lack of sufficient information and confirms the above described somewhat unclear situation. No comments however were received from Biowatch South Africa

[10] Mrs. Michelle Vosges, Directorate Genetic Resources, Department of Agriculture, Harvest House Room 261, 30 Hamilton Street, Arcadia, Pretoria, 0002, South Africa

Department of Agriculture as well as Prof. M. J. Wingfield[11], Director of the Forestry and Agricultural Biotechnology Institute of the University of Pretoria confirmed in personal communications (2005 and 2004, respectively) that there were to their best knowledge at present no field trials with transgenic forest trees in South Africa. This is supported by the fact that all forest companies in South Africa are FSC certified (Prof. M. J. Wingfield, personal communication[8], Dr. S. Verryn, personal communication[12]) which prevents not only commercial use but also research and development work with genetically engineered organisms.

For other parts of Africa there is no information on any cases of transgenic trees being released into the field. For the illegal release of material in Kenya, for example, that was recently reported in some newspapers and online magazines (N.N. 2004a,b) no convincing evidence has been produced as yet. According to the Director of ISAAA (International Service for the Acquisition of Agri-biotech Applications) AfriCenter, Dr. S. Wakhusama, personal communication[13] (2004), there are at present no genetically engineered trees in the country.

1.3.5 Australasia

1.3.5.1 New Zealand

Field trials on transgenic trees have also been run in the past in New Zealand. The OECD database lists two field trials on transgenic pine, probably radiate pine, applied for in 1997 and 2000. The first of these was carried out by the New Zealand Forest Research Institute, the second by the private company Carter Holt Harvey Pulp and Paper.

Work on transgenic *Pinus radiata* has a long tradition in New Zealand. The group of Dr. Christian Walter developed transformation methods for *Pinus radiata* over the last decade. The first transgenic plants were planted in a field trial in 1998. This trial was ended in 2003 due to expiry of the permission that covered only five years. At present, however, there are two further field trials with transgenic radiata pine going on in New Zealand, which are supposed to run from 2003 to 2023. The trees planted in this trial carry marker genes, herbicide resistance genes and genes linked to reproductive development. The research work is aimed at understanding environmental effects and there are no plans for commercialisation (Dr. C. Walter[14], personal communication).

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[11] Prof. Dr. Michael J. Wingfield, Mondi Professor of Forest Pathology, Director, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria 0002, South Africa

[12] Dr. Steve Verryn, Council for Scientific and Industrial Research (CSIR), Meiring Naude Road, Brummeria, Pretoria, South Africa

[13] Dr. Sam Wakhusama, ISAA, Africenter, PO Box 25171, Nairobi 00603, Kenya

[14] Dr. Christian Walter, ForestResearch, Private Bag 3020, Rotorua, New Zealand