Tree Seeds 2008

Trees, Seeds and a Changing Climate

22 – 25 September 2008
Royal Botanic Gardens Kew, Wakehurst Place
& University of Sussex, Brighton, UK

CONFERENCE PROGRAMME & ABSTRACTS

Organisers:
Hugh W. Pritchard, Kate Hardwick, Matt Daws

Seed Conservation Department, Royal Botanic Gardens Kew,
Wakehurst Place, Ardingly, West Sussex RH17 6TN

Further details:
Hugh W. Pritchard. E-mail: h.pritchard@kew.org

INTRODUCTION TO THE SPONSORS

Botanic Gardens Conservation International:
Botanic Gardens Conservation International (BGCI) was founded in 1987 to link botanic gardens as a co-operating global network for effective plant conservation. It now links over 2500 institutions in over 120 countries, all working together to preserve and promote plant diversity for people and the planet. We work in three key areas, with the aim of securing plant diversity for the well-being of people and the planet: (1) enabling people and botanic gardens; (2) securing plant diversity; (3) influencing decision making and policy. Our vision is "*A world in which plant diversity is valued, secure and supporting all life*" and to create this world our mission is: "*To mobilise botanic gardens and engage partners in securing plant diversity for the well-being of people and the planet*".
[http://www.bgci.org/]

International Seed Testing Association:
Founded in 1924, with the aim to develop and publish standard procedures in the field of seed testing, ISTA is inextricably linked with the history of seed testing. With member laboratories in over 70 countries world wide, ISTA membership is truly a global network. Our vision is ‘uniformity in seed testing world wide. ISTA achieves its vision by producing internationally agreed rules for seed sampling and testing, accrediting laboratories, promoting research*, and providing international seed analysis certificates, training and dissemination of knowledge in seed science and technology to facilitate seed trading nationally and internationally. *Relevant technical committees include: Forest Tree and Shrub Seed, and Seed Storage. [http://www.seedtest.org/en/home.html]

International Tree Foundation:
The International Tree Foundation is the oldest tree charity in the UK, founded in the 1920's by Richard St. Barbe Baker OBE. We work with communities 'on the ground' both in the UK and overseas, creating tree based projects that are sustainable and enhance lives. Internationally, we work with non-governmental organisations in developing countries, to support sustainable projects run by and for local communities. These projects involve promoting awareness of arboriculture and the need to maintain biodiversity, protecting existing trees, and planting trees that provide food and income for local people. We are dedicated to extending the world's forests for the benefit of our children and our planet. [http://www.internationaltreefoundation.org/]

International Union of Forest Research Organisations:
IUFRO – The Global Network for Forest Science Cooperation. IUFRO is a non-profit, non-governmental international network of forest scientists, which promotes global cooperation in forest-related research and enhances the understanding of the ecological, economic and social aspects of forests and trees. It unites more than 15,000 scientists in almost 700 Member Organizations in over 110 countries, and is a member of ICSU. Scientists cooperate in IUFRO on a voluntary basis. Our mission is: (1) to promote global cooperation in forest-related research and to enhance the understanding of the ecological, economic and social aspects of forests and trees; (2) to disseminate scientific knowledge to stakeholders and decision-makers; (3) to contribute to forest policy and on-the-ground forest management. [http://www.iufro.org/]

Millennium Seed Bank Project, Royal Botanic Gardens Kew:
The Millennium Seed Bank Project, which is managed by Kew's Seed Conservation Department, is the largest *ex situ* conservation project ever conceived. Its partners will have banked seed from 10% of the world's wild plant species by the end of the decade. These will not be just any plants, but will include the rarest, most threatened and most useful species known to man. Our scientific objectives are to increase knowledge and understanding of seed diversity, particularly in relation to four key traits: seed maturity (development); seed desiccation tolerance; seed storability; and seed dormancy (germination). These traits have direct relevance to improving the seed conservation practices of seed 'collecting', 'processing', 'storing' and 'monitoring and using'. The group's work is divided into seven themes including Climate and Reproductive Biology. [http://www.kew.org/msbp/]

Royal Botanic Gardens Kew:
RBG Kew currently employs over 680 staff, over 60% of whom are specialist plant or fungal scientists or botanical horticulturists. The mission of the Royal Botanic Gardens, Kew is: To inspire and deliver science-based plant conservation worldwide, enhancing the quality of life. Our business aim is to produce basic and applied information about plant- and fungal-related topics and to manage and communicate this to all our stakeholders. This aim is carried out through science and research in systematics, biological interactions, economic botany, conservation and horticulture. This is underpinned by our extensive collections of living and preserved plants and fungi, associated artefacts, literature and archives, and is interpreted to the public through the Gardens at Kew and Wakehurst Place. These provide an amenity for the public, offering the opportunity to learn about plants and our wider work. Our ability to increase our outreach and global impact depends on the effectiveness of our communications. Our education programmes, dissemination activities, our national and international partnership networks are fundamental in ensuring the transfer of our specialist knowledge to the global community. [http://www.kew.org/]

University of Sussex:

The University of Sussex was the first of the new wave of universities founded in the 1960s, receiving its Royal Charter in August 1961. Forty years on, the University has become a leading teaching and research institution, characterised by a number of academic strengths including research excellence, internationalism and interdisciplinarity. As a research intensive university, Sussex has a dynamic and thriving research agenda. Our strengths range across the arts, social sciences, science and medicine, with excellence demonstrated both within individual subjects and across thematic areas. Sussex has an international reputation for its innovative styles of teaching and for the quality and range of its research work. It has academic links with every continent, with 2,300 international students and teaching/research staff from over 120 nations. [http://www.sussex.ac.uk/]

PROGRAMME:

Mon 22 Sept. 2008	**Univ Sussex**	**Conference Centre (3rd floor, Bramber House)**
Afternoon	Arrival	
16.00 –18.00 h	Registration	Conference Centre, 3rd floor Bramber House
18.00 h	Drinks	Lounge Bar, Conference Centre, 3rd floor, Bramber House
18.30 h	Dinner	Downs Restaurant, Conference Centre, 1st floor, Bramber House

Tues 23 Sept	**Univ. Sussex**	**Conference Centre**
07.30 h	**Breakfast**	Downs Restaurant, 1st floor, Bramber House
08.00 h	Registration	Conference Centre, 3rd floor, Bramber House
08.45 h		**Hugh W. Pritchard** (UK) Don C. MacIver (Canada) : Welcome and introduction

CLIMATE CHANGE: PREDICTED AND OBSERVED IMPACTS

09.00 h	Invited speaker	**Douglas Gibbs & Sara Oldfield** (UK): Threatened trees in a changing climate
09.45 h	Invited speaker	**Tim Wheeler** (UK): Using climate information for biological predictions
10.30 h	**Refreshments**	**Posters**
11.00 h		**Hugh W. Pritchard & Matt Daws** (UK): Thermal time for seed development and tree seed quality across Europe
11.30 h		**A Tsiroukis and Costas Thanos** (Greece) Field seed germination of horse chestnut (*Aesculus hippocastanum*) in Greece and climate change impacts
12.00-12.15 h		General Discussion
12.30 h	**Lunch**	**Gallery 2** (3rd floor, Bramber House)
14.00 h	Invited speaker	**Ghillean T. Prance** (UK) Trees and other organisms show that climate change is real
14.45 h	Invited speaker	**Monique S.J. Simmonds** (UK) Climate Change: Tree-insect interactions
15.30 h	**Refreshments**	**Posters**
16.00 h		**Rose Newton** (UK) The relationship between rainfall, water source and growth of *Widdringtonia cedarbergensis*, an endangered tree in south-western Cape
16.30 h		**E Tillman-Sutela & A Kauppi** (Finland) Seed structures and predicted success of tree-line conifers in a warming climate.
17.00 – 17.15		General discussion
18.30 h	**Dinner**	Downs Restaurant, 1st floor, Bramber House

Wed 24 Sept	Wakehurst Place	Wellcome Trust Millennium Building
07.00 h	**Breakfast**	Downs Restaurant, 1st floor, Bramber House
08.15 h	Coach departure	From Brighton to Wakehurst Place, Ardingly
09.25 h		**Paul Smith** (UK): Welcome

FOREST FRAGMENTATION AND RESTORATION

09.30 h		**J. L. C. Camargo, B.A. Santos, M.R. Mesquita, H.D. Brum, J.B.P. Costa & Isolde D.K. Ferraz** (Brazil): Does forest fragmentation alter the phenological patterns of trees in Central Amazon?
09.55 h		**Shelagh McCartan, P. Gosling, M. Broadmeadow & J. Clarke** (UK): Tree seed biology, climate change and local seed source.
10.20 h		**G. Peter Buckley & D Blakesley** (UK): Origins and supply of native tree seed in the UK.
10.45 h		**Lars Schmidt** (Denmark): Advances in tropical tree seed procurement.
11.10 h	**Refreshments**	
11.30 h		**Jens-Peter Barnekow Lillesø** (Denmark): Support to smallholder tree planting in the tropics – some issues
11.55 h		**David Cutler** (UK) Sustainable fuelwood supplies and climate change: research in dry areas of Zimbabwe and NE Brazil'.
12.20 h		**David Blakesley & S Elliott** (UK): Framework species and forest restoration
12.45 h		General Discussion
13.00 h	**Lunch**	
14.00 h	Tours	Guided tours of the Wellcome Trust Millennium Building. A chance to look behind the scenes at the science and facilities of the Seed Conservation Department, including the world-leading conservation work of the Millennium Seed Bank Project
15.15 h	Free time	Time to explore the beautiful gardens at Wakehurst Place, one of the leading visitor attractions in the south of England
17.30 h	Coach departure	Public car park (Wakehurst Place)
18.30 h	**Drinks**	Marble Bar, Downs Restaurant, 1st floor, Bramber House
19.00 h	**Conference dinner**	Downs Restaurant, 1st floor, Bramber House

Thurs 25 Sept	**Univ. Sussex**	**Conference Centre (3rd floor, Bramber House)**
7.30 h	**Breakfast**	Downs Restaurant, 1st floor, Bramber House

TREE SEED SCIENCE

09.00 h	Invited speaker	**Patricia Berjak & N. Pammenter** (RSA): Seeds: the insurance against the ravages of climate change
09.45 h	Invited speaker	**Ilse Kranner & H.W. Pritchard** (UK): Tree seeds in a changing climate: stress concepts and case studies
10.30 h	**Refreshments**	**Posters**
11.00 h		**Christian Mong, V. Vandvik & M.I. Daws** (Norway)**:** Germination and establishment requirements of conifers: evidence based on germination tests in laboratory and semi-natural establishment trials.
11.30 h		**Dave Kolotelo** (Canada): Dormancy, seed pretreatment, and quality assurance in British Colombia conifer seedling production.
12.00 h		General discussion
12.30 h	**Lunch**	**Gallery 2** (3rd floor, Bramber House)
14.00 h		**C.V. Vieira, E.A. Amaral da Silva, A. Alves Alvarenga, E. Mauro de Castro, & Peter E. Toorop** (Brazil): Desiccation tolerance of germinated seeds of *Tabebuia impetiginosa*
14.30 h		**Kim Hamilton, S.E. Ashmore & H.W. Pritchard** (Australia): Effects of natural distribution of Australian wild citrus on phase transition of seed oils and cryopreservation.
15.00 h		**CT Chien, BSP Wang & SY Chen** (Taiwan): Bigleaf mahogany (*Swietenia macrophylla* King) seeds in Taiwan exhibit orthodox storage behaviour
15.30 h	**Refreshments**	**Posters**
16.00 h		**P. Chmielarz & C. Koziol** (Poland): Cryopreservation of *Quercus robur L.* plumules.
16.30 h		**Steve W. Adkins & Y.M.S. Samosir** (Australia): Embryo transplant: a potential technique for propagation of high value, endosperm-mutant coconuts
17.00 h		General discussion
18.30 h	**Dinner**	Downs Restaurant, 1st floor, Bramber House
Fri 26 Sept	Departure	

ABSTRACTS: Oral communications

Douglas Gibbs & Sara Oldfield

Threatened trees in a changing climate

Botanic Gardens Conservation International, Descanso House
199 Kew Road, Richmond, Surrey, TW9 3BW, UK

Approximately 10 percent of the world's tree species are threatened with extinction according to the IUCN Red List Categories and Criteria. The impact of climate change has rarely been taken into account in evaluating the conservation status of trees but predictive modelling is now indicating the scale of this over-arching threat to plants in general. This presentation will outline the impacts of a changing climate on tree species; Red Listing efforts for tree species and how the resulting information can be used to reinforce tree conservation efforts. BGCI's work on *Magnolias* will be used as a case study to highlight information collection and integrated conservation planning. The importance of botanic gardens working individually and collectively will be emphasised taking into account both *ex situ* and *in situ* conservation approaches. The need for further research will also be discussed.

Tim Wheeler

Using climate information for biological predictions

Walker Institute for Climate System Research, Department of Agriculture,
University of Reading, Earley Gate, Reading RG6 6AR, UK.

There is increasing concern about the possible impacts of human-induced climate change on terrestrial ecosystems. The fourth assessment report of the Intergovernmental Panel on Climate Change published in 2007 documented numerous examples of changes in natural ecosystems that have been already been observed and that in many cases have been linked to changes in climate. Robust predictions of possible changes in the future due to climate are vital for our understanding and management of both natural and managed ecosystems over the coming decades.

Predictions of the behaviour of biological systems can be provided using numerical models of those systems. Assessment of the impacts of climate change on a biological process that is sensitive to climate, such as the rate of seed germination or seedling emergence, involves two quite different models: a climate model and the biological model. Understanding the nature of each type of model is crucial to understanding the skill of biological predictions, and the uncertainty of the predictions, under scenarios of climate change. Important research problems that need to be addressed include the different spatial and temporal scales of the models; the underlying assumptions of different scenarios of climate change; and how to best represent the uncertainties of biological predictions under climate change. This talk explores how to tackle these challenges in climate change impacts research.

Hugh W. Pritchard & Matt Daws

Thermal time for seed development and tree seed quality across Europe

Seed Conservation Department, Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, West Sussex RH17 6TN, UK.

Long-lived perennials, especially trees, are at the greatest threat of species extinction from habitat change, as long life cycles reduce the opportunity for adaptation to changing environmental conditions. Moreover, one of the earliest predicted impacts of an increase in the global average temperature is a reduction in seed yield. Consequently, we investigated the effects of temperature across Europe on the biochemical, physiological and physical aspects of tree seed quality throughout the growing season. We selected two species (*Aesculus hippocastanum* and *Acer pseudoplatanus*) with pan-European distributions, stretching from their native southern origins to their introduced, northern ranges. Using principal component analysis we found in both species predictable and consistent patterns in all traits, which correlated with heat sum during seed development; for example, improved germinability. The heat sum in southern Europe was about 50% greater than in northern Europe. At the point of natural dispersal the embryonic axis moisture content was about 15% higher in northern compared to southern populations, indicating for both species more immature seeds in the north. Whilst the critical moisture content for viability loss was systematically improved with developmental heat sum, *Aesculus hippocastanum* seeds always had high desiccation sensitivity levels typical of recalcitrant seeds. However, southern European populations of *Acer pseudoplatanus* had tolerance levels far greater than those of recalcitrant seeds, revealing a phenotypic variation in seed storage response. We exploited the additional desiccation tolerance of southern *Acer pseudoplatanus* and stored the fruits at sub-zero temperatures, enabling us to demonstrate the ex situ seed conservation potential of mature fruits from southern Europe. At a continental scale, temperature clearly has profound impacts on seed quality and the pattern of response now allows us to predict the potential impact of future climate changes.

A. Tsiroukis[1] and Costas A. Thanos[2]

Field seed germination of horse chestnut (*Aesculus hippocastanum*) in Greece and climate change impacts

[1]Technological Education Institute of Larissa, Department of Forestry, Karditsa, Greece
[2]National and Kapodistrian University of Athens, Department of Botany, Panepistimiopolis, Athens 15784, Greece

Despite its wide cultivation throughout the temperate world, the horse chestnut tree (*Aesculus hippocastanum* L., Sapindaceae) is considered as a Tertiary relict and is currently a Balkan endemic (mainly restricted to Greece) with a highly fragmented, natural geographical distribution. In regard to conservation status (according to the new IUCN criteria), the species has been recently assessed as Critically Endangered [B2ab(iv,v)]. Horse chestnut seeds are large (the European champions in seed mass) and recalcitrant; moreover, they are dormant when shed in the autumn and, apparently, lack any specialized dispersal agent. All these traits contribute negatively to the species natural regeneration.

Several previous works (with seeds obtained from trees cultivated outside the natural range of the species) have shown that seed dormancy is relieved by prolonged chilling, thus enabling germination at gradually lower temperatures and with higher percentages and rates. In this work we confirm similar germination behaviour for seeds collected from indigenous trees, growing in several sites within the natural area of the species. This strategy is obviously adapted towards timing germination and seedling establishment in the spring, therefore avoiding the winter frost. On the basis

of both observations and experiments in the field, we report that this is the case indeed: germination takes place early in spring (March) almost coinciding with snow melting. Germination data are supplemented and correlated with temperature and precipitation data, obtained from both dataloggers and conventional meteorological stations. Finally, we present and discuss the potential impacts of climate change on seed germination, natural regeneration and conservation status of horse chestnut native populations.

Ghillean T. Prance

Trees and other organisms show that climate change is real

Lyme Regis, Dorset UK

Some examples of the effect of climate change on various organisms will be given. This will be based on the author's travels mainly in the tropics. Even the indigenous peoples such as the Guaraní of South America are noting changes in the climate that are affecting their daily life. Efforts to conserve biodiversity will be in vain if we do not get climate change under control and so this is a call to address that issue with urgency.

Monique S.J. Simmonds

Climate change: Tree-insect interactions

Jodrell Laboratory, Royal Botanic Gardens Kew, Richmond, Surrey TW9 3AB, UK

E.C. February[1], Rose J. Newton[1,2] and A.G. West[1]

The relationship between rainfall, water source and growth of *Widdringtonia cedarbergensis,* an endangered tree in south-western Cape

[1]Department of Botany, University of Cape Town, Private Bag X3, Rondebosch, 7701, South Africa
[2]Seed Conservation Department, Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, West Sussex, RH17 6TN, United Kingdom (present address)

It is now widely accepted that the impact of humans on the environment since industrialization has led to significant changes in climate. The fynbos, a fire-prone shrub land confined to the nutrient poor soils of the south-western parts of South Africa, is a biodiversity hotspot threatened by the effects of global warming. The Clanwilliam cedar, *Widdringtonia cedarbergensis* Marsh., is one of the few trees that occur in the Fynbos Biome. This rare conifer is endemic to a single mountain range in the Western Cape, with a patchy distribution over approximately 250 km^2. Trees are confined by regular fires to marginal rocky outcrops and ravines between 900 and 1400 metres above sea level. Here we attempt to develop an understanding of the effects that future changes in climate may have on *W. cedarbergensis* by examining the relationship between radial trunk growth and water source. Our results show no significant relationship between monthly radial growth and rainfall. The dendrometer bands reflect a slow but steady growth of the trees at the study site. There is, however, a significant relationship between the $\delta^{18}O$ isotopic composition of the xylem water and the rain $\delta^{18}O$ values. We speculate that *W. cedarbergensis* utilises water that accumulates within the bedding planes of the rocks on which they grow, and that this water is regularly recharged by rainfall. Thus, while growth is not correlated with monthly rainfall amount, it appears to be dependent on reliable access to available

water that is recharged by rainfall. If climate change predictions for the region are realized and rainfall is reduced this may have implications for the growth and fitness of this iconic species. In addition to water availability, fire and seed predation are additional pressures that threaten this already endangered species. The extinction of *W. cedarbergensis* in the wild would fundamentally affect both the species composition and vegetation structure of the region.

E. Tillman-Sutela[1] and A. Kauppi[2]

Seed structures and predicted success of tree-line conifers in a warming climate.

[1]Metla, Muhos Research Station, Kirkkosaarentie 7, 91500 Muhos, Finland
[2]Dept. of Biology, University of Oulu, P.O. Box 3000, 90014 Oulun yliopisto, Finland

Rigorous climate conditions and short growing seasons are considered the main reasons for irregular seed crops and poor regeneration success of the conifers at both the boreal and the alpine tree-line. Adequate temperature during the growing season is known to favour the conifer pollen and embryo growth, whereas maturation of the surface structures is less dependent on the temperature and relies also on the photoperiod. Consequently, the differences in the schedule of maturation among the surface structures and the megametophyte and the embryo can be several weeks. The extent of maturation stage in seeds prior to the unfavourable season varies among the conifers and even among the species within the same family as an adaptation to the ways of dispersal. Both structural and chemical maturity of seed structures is indispensable for the wind-dispersed species at the boreal tree-line e.g. *Pinus sylvestris*, because seeds stay in trees during the cold winter period. Meanwhile the bird-dispersed seeds of alpine tree-line species, e.g. *Pinus albicaulis*, *P. cembra* or *P. sibirica*, are buried in the soil and covered by snow and therefore do not encounter very low temperatures. When buried in soil for one or more years *Cembrae* seeds also continue structural maturation and differentiation to attain readiness to germinate.

Warming of climate is regarded to increase seed production in boreal and alpine areas due to improved embryo growth and maturation. It is uncertain, however, if the surface structures that serve as a protection against desiccation, fungi, and predators, will benefit equally of the predicted higher temperatures, because the day-length will stay unchanged. The warming climate results also into smaller amounts of snow and presumably into deeper freezing of the soil in both the boreal and alpine tree-line. Freezing of the soil to a great depth can injure the roots and impair stress-tolerance of trees against fungi and predators, which would diminish florescence and seed production. Shorter periods and smaller amounts of snow melts can cause increased drought particularly in early summer, which would impair germination success and survival of young seedlings. The diminishing distribution of these tree stands would disturb the biodiversity of both the flora and fauna, because conifer seeds are rich in lipids and have great food values for many birds and mammals including the bears.

**J. L. C. Camargo[1], B. A. Santos[1], M. R. Mesquita[2],
H. D. Brum[2], J. B. P. Costa[1] and I. D. K. Ferraz[2]**

Does forest fragmentation alter the phenological patterns of trees in Central Amazon?

[1]Biological Dynamics of Forest Fragment Project (Brazilian National Institute of Amazon Research (INPA)/ Smithsonian Tropical Research Institution (STRI)
PO Box 478, 69011-970, Manaus, AM, Brazil
[2]Department of Tropical Forest Science (INPA)

Forest fragmentation alters the microclimatic conditions in the forest remnants. In general, a forest fragment has a more open canopy and drier and hotter micro-environmental conditions in comparison to the continuous forest. Such changes may be analogous to foreseen changes caused by global climatic changes and may alter tree reproduction cycles, with consequences to forest dynamics. In the unflooded forest in the North of Manaus, the 18 most common tree species (representing 23% of trees in this forest) were marked. The reproductive cycles of 20 trees of each species were observed in both continuous forest and in a 10-ha forest fragment. Flowering and fruiting were monitored monthly during February 2003 and January 2006. During this 3-yrs period, 80.7% of the potentially reproductive trees showed some reproductive event in the continuous forest, while in the forest fragment this number dropped to 56.5%. The probability of occurrence of any reproductive event was different for each species: decreased significantly in the forest fragment for five tree species, moderately for nine species and increased moderately for four species. In the continuous forest trees showed a more consistent pattern for seasonality of flowering and fruiting; this was not the case in the fragment, where synchronous reproductive events were reduced by 18%. The environmental changes caused by forest fragmentation appear to diminish flower and seed/fruit production. We hypothesize that even populations of common trees isolated in forest fragments may face short-term reductions in seedling establishment and rapidly changes on biotic interactions with pollinators, seed dispersers and predators.
Financial support CNPq/MCT/PPG-7/FAPEAM

Shelagh McCartan, Peter Gosling, Mark Broadmeadow and Jo Clark

Tree seed biology, climate change and local seed source

[1] Forestry Commission Research Agency, Alice Holt Lodge, Wrecclesham, Farnham, Surrey, GU10 4LH, UK

There are currently a number of groups throughout the European Union encouraging the collection and propagation of “native trees from local seed sources” – the laudable aim being to preserve locally adapted genotypes. However, if the climate is likely to change significantly in the next 50 years then perhaps we need to study where today’s local seed sources may be best suited to in the future, and plan for their migration.

This paper examines whether seed dormancy, pretreatment requirement and hence germination of European trees is likely to be affected by climate change. It shows that some tree seeds are “deeply dormant” – that is they are dispersed in autumn with a complete inability to germinate until they have received a lengthy, moist-chilling period (prechill) over the next and subsequent winters. Current winter temperatures and durations overcome their dormancy and equip them with the ability to germinate. But if winters become either warmer, or shorter or both they may no longer receive a sufficient prechill to be able to germinate. Other trees produce “shallowly dormant” seeds – on dispersal they are unable to germinate at today’s autumn temperatures (10-15 deg C) but they do germinate in slightly warmer conditions (15-25 deg C). If climate change brings about warmer, extended autumns the seeds of these species may germinate too soon and their highly vulnerable seedlings will be much more likely to be killed by subsequent winter conditions.

The paper explores the potential effects of climate change on natural regeneration and aims to identify where today’s seed sources may do best in the future.

G.Peter Buckley[1] and D Blakesley[2]

Origins and supply of native tree seed in the UK

[1]Peter Buckley Associates, Mersham, Ashford, Kent TN25 7HD

[2]Wildlife Landscapes, Bearsted, Maidstone, Kent ME15 8JJ

Current forest restoration strategies in the UK aim to recover ancient woodland from former commercial plantations; plant new native woodlands on former agricultural land; and increase connectivity between existing woodland fragments. In each of these objectives the overiding assumption is that native plants of indigenous or local provenance will be used, but in practice this may be hard to achieve and arguably difficult to justify in view of the rapidity of climate change.

Our study, part of a Franco-British Interreg IIIA Programme, investigated the ability of British nurseries to supply native seed or plants of local provenance to the forest, amenity and conservation industries. Forest seed supply is partly controlled by European Forest Reproductive Regulations and moderated in this country by the Forestry Commission, who designate a scheme of seed collection zones. However, there are issues relating to a) whether the stands collected from are indeed indigenous to these zones, b) there is good availability of seed in a given harvesting season, and c) if collection protocols are rigorous enough to ensure adequate genetic diversity.

A questionnaire survey revealed that more than a third of native tree seed supplied by the nurseries is currently non-British, to some extent reflecting customer indifference to origin and sensitivity to price. Seed collection (and natural regneration) was often from relatively few fruiting trees and nurseries experienced difficulty in locating suitable native woodland seed sources. We consider how the system might be improved, and whether the present emphasis on 'local provenance' material from seed collection zones should be relaxed in order to permit a wider mix of provenances, including material imported from outside the UK.

Lars Schmidt

Advances in tropical tree seed procurement.

Faculty of Life Sciences, Forest and Landscape, University of Copenhagen, Hoersholm Kongevej 11, DK-2970 Hoersholm, Denmark.

Focal areas for international support to forest seed sectors in tropical countries have shifted from technical support to commercial plantation species, to diversified species including many agroforestry species. Much of Danish involvement in tropical tree seed has been linked to tree improvement. Present support is primarily to institutional / sector support. Technological development compiled in three Danish seed handling guidebooks: A guide to forest seed handling (Willan 1975); A guide to handling of tropical and sub-tropical forest tree seed (Schmidt 2000); and Tropical forest seed (Schmidt 2007). Support to forest seed sectors currently face the dilemma between low technology adapted to prevailing technological standard in resource poor countries, and high tech input aimed at pioneering and bringing the seed sector up to an international standard. Current technical and economic progress in most developing countries gives access to better simple adapted technology. Many practical problems in seed procurement have been overcome by applied research. A few still remains. Parallel progress in vegetative propagation have by-passed much arduous seed propagation. However, small differences in seed propagation costs tend to favour the cheapest species independent of long term objectives. A major obstacle to adoption of methods and hence improvement of germplasm quality in the actual plantation / afforestation activities is lack of access to material and knowledge for a large part of seed users.

Jens-Peter Barnekow Lillesø

Support to smallholder tree planting in the tropics - some issues

Forest and Landscape, University of Copenhagen, Hørsholm Kongevej 11, 2970 Hørsholm, Denmark.

During many years organised support to tree planting in the tropics to a large extent evolved around National Tree Seed Centres (NTSCs). It appears that the current role of the NTSCs in smallholder tree planting is much less than what could be desired and at the same time NGOs and projects have been unable to adequately support smallholder tree planters with quality material. The presentation attempts to provide an explanation for this situation and will outline how appropriate public and private sector intervention potentially could lead to increased productivity of smallholder plantings for millions of poor farmers in Sub-Saharan Africa

David F.Cutler [1,2]

Sustainable fuelwood supplies and climate change: research in dry areas of Zimbabwe and NE Brazil'.

[1]Jodrell Laboratory Royal Botanic Gardens Kew, Richmond, Surrey TW9 3AB
[2]Linnean Society, Piccadilly, London

As early as 1988 a small international research team recognised that the climate in Zimbabwe was becoming dryer, and this could lead to fuelwood poverty, particularly in the Communal Lands. Four species preferred for fuelwood that grow in both dryer and hotter parts, and less heat and water stressed areas, were selected for study. These are *Acacia karroo, A. tortilis, Combretum apiculatum* and *Colophospermum mopane.* The initial experiments, funded by the EU, were set up to determine whether in dry and hot habitats drought and heat tolerance had evolved in the four selected species in localities where gene flow between them and the less stressed trees was highly improbable. The experiments and results, which did indicate the inheritance of such tolerances, will be described.

This research was followed by experiments in Zimbabwe on fuelwood harvesting times and methods for the four species. Control trees were designated, and the cutting treatments were pollarding, coppicing or crown thinning. Political problems have intervened, and the outcome is uncertain, but some valuable observations have been made.

The experimental model was taken to NE Brazil in 2001, to see if changes could be made to traditional methods of harvesting wood for charcoal production that had less environmental impact, and were potentially more sustainable. There the species studied are *Caesalpinia pyramidalis, Mimosa ophthalmocentra, Mimosa tenuiflora* and *Croton sonderianus.* The field work in this project, jointly conducted by teams in Kew and Brazil, will be completed towards the end of 2008, but some interesting observations made following the harvesting of half of selected the tree specimens in 2005 will be reported.

David Blakesley[1] and Stephen Elliott[2]

Framework species and tropical forest restoration

[1]Wildlife Landscapes, Bearsted, Maidstone, Kent ME15 8JJ
[2]Forest Restoration Research Unit, Biology Department, Faculty of Science, Chiang Mai University, Chiang Mai, Thailand 50200.

Chiang Mai University's Forest Restoration Research Unit (FORRU-CMU) is located in the seasonally dry tropical forest of Doi Suthep-Pui National Park, in northern Thailand. Since its foundation in 1994, FORRU's research programme has developed methods to accelerate biodiversity recovery to restore forest ecosystems on degraded land, within protected areas in northern Thailand, particularly to promote biodiversity recovery and environmental protection.

FORRU-CMU has adapted the 'framework species method' to the ecological and socio-economic conditions of northern Thailand, to successfully to restore evergreen forest to deforested land. This method involves planting 20-30 indigenous forest tree species, which enhance natural forest regeneration and accelerate biodiversity recovery. FORRU-CMU initially carried out a study of the flowering and fruiting phenology of indigenous forest tree species to determine optimal seed collection times. Dormancy, seed germination and seedling growth were monitored in a nursery to help screen more than 420 native tree species for their suitability for planting in degraded areas. Working with such large numbers of species creates logistical problems for nursery managers, particularly in relation to dormancy and germination. Field trials to compare field performance among species and their responses to silvicultural treatments were carried out in close collaboration with the Hmong hill tribe villagers of Ban Mae Sa Mai, where a community tree nursery was established to test the practicability of the techniques developed. Through working with villagers, FORRU-CMU has also gained an insight into motivational factors that influence villagers' participation in forest restoration projects.

Patricia Berjak and N.W. Pammenter

The insurance against the ravages of climate change

Plant Germplasm Conservation Research, School of Biological & Conservation Sciences
University of KwaZulu-Natal (Westville Campus) Durban, 4001 South Africa

Although the details of the projected effects of climate change on parameters such as surface warming, sea level rise and precipitation vary according to the model used and on which SRES Emission Scenario[1] a model is based, all are unanimous that climate change is inevitable and already well underway. Vegetation, including importantly, the development and post-shedding responses of seeds will be impinged upon by changes in climate, particularly higher average temperatures, and especially the increased frequency of very warm or heat-wave conditions; and changes in rainfall. These parameters are likely to have marked effects on development and post-harvest responses of seeds, depending on the combinations in which the climatic factors occur, on the provenance of the plants (trees), and on the categorisation of the seeds themselves – i.e. whether they are orthodox or non-orthodox and, in the latter case, whether truly recalcitrant or not.

In order to ameliorate the effects of climate change and manage effective environmental restoration on an ongoing basis, it is essential to have the widest possible knowledge and most comprehensive understanding of reproductive biology, often on a species basis – from the nature of the pollen, pollinating agents and pollination, through the events of seed development and the post-

[1] Summarised in 2007 from the IPCC (Intergovernmental Panel on Climate Change) Special Report on Emission Scenarios.

shedding responses of the seeds. It is in aspects of this wide context that the seed biology of selected temperate and tropical species will be discussed, with particular emphasis on non-orthodox/recalcitrant types.

The basis of genetic resources conservation with the objective of the amelioration of the effects of climate change in terms of environmental restoration, depends critically on being able to store seeds successfully. This, in turn, depends on the most comprehensive understanding of post-shedding seed responses and, in the case of recalcitrant seeds, on strategies to achieve successful cryopreservation and subsequent production of vigorous plants. The basis of both limitations and successes in achieving this, will be discussed.

Ilse Kranner and Hugh W. Pritchard

Tree seeds in a changing climate: stress concepts and case studies

Seed Conservation Department, Royal Botanic Gardens Kew, Wakehurst Place,
West Sussex, RH17 6TN, UK.

Global climate change fundamentally impacts on plant life. It entails alterations in temperature and water availability and thereby increases environmental stress. As all plants, trees are affected by such abiotic stresses. Trees they are extremely important components of key ecosystems, including tropical rain forests. Hence, particular attention should be directed towards their protection and conservation. In this lecture, stress concepts will be reviewed and suggestions made how those from other disciplines can be applied to plants and plant seeds in particular. One of the widely accepted stress concepts in humans, proposed by Hans Selye in 1936, is the "General Adaptation Syndrome". It involves three stages of response, "alarm", "resistance" and "exhaustion". Correspondingly, in plants, stress signalling, involving cross-talk between reactive oxygen species (ROS) and plant hormones could be viewed as part of the "alarm" response. Acclimation, adaptation, and repair could be considered as the "building blocks" of the "resistance" response. When protection and repair mechanisms eventually fail, cell death is the result, corresponding to "exhaustion" and ultimately, cell death. Case studies will be presented that outline mechanisms of cell death, and others that refer to the stresses that accompany cryopreservation of recalcitrant seeds. The latter are found in an estimated 10% of all seed-bearing plants although in some habitats such as tropical forests, almost half of the flowering plants, mainly trees, produce recalcitrant seeds. Due to their sensitivity to desiccation and low temperatures, recalcitrant seeds are particularly challenging to store, compromising the conservation of the germplasm of recalcitrant-seeded plants. Production of ROS accompanies the first stages of cryopreservation, which are excision of the embryonic axis, *i.e.*, wounding, and desiccation. Although often viewed as deleterious, the extracellular production of ROS appears to play an important role in the stress response of recalcitrant seeds. Fundamental roles of ROS in stress response will be discussed with a view of manipulating ROS production as a key strategy for the optimization of cryopreservation techniques.

Christian E. Mong[1+2], V. Vandvik[2] and M.I. Daws[1]

Germination and establishment requirements of conifers: evidence based on germination tests in laboratory and semi-natural establishment trials.

[1]Seed Conservation Dept., Royal Botanic Gardens Kew, Wakehurst Place, Ardingly,
W Sussex, RH17 6TN, UK
[2]Department of Biology, University of Bergen, PO Box 7800, 5020 Bergen, Norway

Mechanisms regulating the distribution of coniferous boreal forests vs. broad leaf temperate forests are poorly understood. In a context of climate warming, one may expect changes to the distribution of these two major forest types. This study focus on the requirements of conifer seeds for germination and establishment. Target species were *Abies procera, Larix* spp., *Picea abies, Picea sitchensis, Pinus sylvestris* and *Tsuga heterophylla.* Some of these species are native to the west coast of Norway and some introduced, but all are currently increasing in abundance. Due to reduced agricultural activity in outfields, cultural landscapes are being encroached by both conifers and deciduous trees. What will happen when coniferous and broad leaf forest types meet?

Germination experiments show that seeds of conifers only require water to germinate, and that species display a relatively tolerant response to germination inhibiting factors such as acidity and water stress. In addition, we are exploring the effects of seed size, substrate type and leaf litter on germination and establishment to test the hypothesis that large seeds (e.g. *Abies procera*) will successfully establish through litter from broad leaf trees more frequently than medium sized seeds (e.g. *Picea abies*), small seeds (e.g. *P. sitchensis*) and very small seeds (e.g. *Tsuga heterophylla*).We also hypothesise that in substrates covered with broad leaf litter fragments, the advantage of larger seeds will be less pronounced, and this advantage will be even smaller in substrates covered with narrow leaf litter from native pine forests.

David Kolotelo

Dormancy, seed pretreatment and quality assurance in British Columbia conifer seedling production

BC MFR Tree Seed Centre, Tree Improvement Branch, Victoria, British Colombia, Canada.

The major reforestation species in British Columbia (BC) are Pinus contorta, Pseudotsuga menziesii, Thuja plicata and a species complex of Picea glauca, engelmannii and hybrids which in total account for approximately 93% of the reforestation in the province. An additional 15 species (varies by year) account for the remainder of reforestation. The seed of these species will be briefly reviewed as well as some background on seed use in the container seedling production system used in BC.

The importance of our quality assurance (QA) system is to ensure seed pretreatment results and advancements in lab testing are realized in operational seed preparation and seedling production. Operational seed pretreatments are based on naked stratification in polyethylene bags using the target moisture content concept. A sample of seedling requests will have grams added to perform QA tests on moisture content and germination prior to shipping. Nursery germination results are generally received on all these requests allowing for a comparison of germination % results of the current lab test , QA results on operational quantities of seed and results actually realized in nursery operations. This allows for a comparison of results (falldowns versus lab results) and a prioritization of species and areas requiring improvements. The importance of monitoring operational quantities of seed will be presented with Pinus monticola (one of our most dormant species) and the importance of moisture content monitoring presented with work on Pinus contorta.

Seed dormancy is absent from Thuja plicata and this species is pelletized for precision sowing in BC. We perform a pellet assessment based on the contents of 8 replicates of 25 pellets divided into the following categories: one seed per pellet (what we want), more than 1 seed per pellet, debris-filled and empty. Pelleting is a very efficient process with approximately 98% of pellets containing a single seed. At the opposite end of the spectrum deeply dormant species are Pinus monticola, Chaemacyparis nootkatensis, Abies amabilis, Abies procera and Abies lasiocarpa. These species acount for approximately 2% of reforestation, but their seed pretreatments are quite intensive with the targetting of stratification moisture content, the use of a dryback treatment for Abies species and the regular monitoring of seed condition and equilibration of seed condition performed on a set schedule. The remaining spcies are considered to have low to medium levels of dormancy with large variability exhibited between seedlots within a species.

Results of our QA moisture content and germination tests will be presented and discussed in our standard five-year rolling average format. An additional aspect of our QA program is the use of returned or unused seed. This is seed that has been stratified, but not used at the nursery and by legislation it must be returned to the Tree Seed Centre. We try and avoid returned seed by adjusting grams allotted to nurseries to increase seed-use efficiency, but some returned seed is unavoidable. This program is primarily geared to low to mid-dormancy species. The seed is dried to storage moisture content, tested for germination and given a new seedlot identification number. It will then be available by the owner for sowing and if selected will be re-stratified for operational use. Results with this seed have been quite good with only a 2% decrease in germination realized at the nursery.

Carlos Vinicio Vieira [1]; Edvaldo Aparecido Amaral da Silva [2]; Amauri Alves Alvarenga [1]; Evaristo Mauro de Castro [1]; Peter E. Toorop [3]

Desiccation tolerance of germinated seeds of *tabebuia impetiginosa*

[1] Setor de Fisiologia Vegetal – Dept. de Biologia – Univ. Federal de Lavras
[2] Lab. de Sementes Florestais – Dept. de Ciências Florestais – Univ. Federal de Lavras
[3] Seed Conservation Dept., Royal Botanic Gardens, Kew, Wakehurst Place, Ardingly, West Sussex, RH17 6TN, UK.

Like for many species, desiccation tolerance in seeds of *Tabebuia impetiginosa* is lost upon completion of germination. Loss of desiccation tolerance can be reverted by applying osmotic stress, described previously for *Cucumis sativus* and *Impatiens walleriana* (Bruggink and van der Toorn, 1995) and *Medicago truncatula* (Buitink et al. 2003). The success of such a treatment depends on the survival rate, which varies. The aim of this study was to investigate (i) if desiccation tolerance could be re-induced in germinated seeds of *Tabebuia impetiginosa* and (ii) which factors can further improve survival of germinated seeds.

Polyethylene glycol (PEG) was capable of re-inducing desiccation tolerance in radicle tips of germinated seeds with a primary root length of up to 3 mm. An osmotic potential of -1.7 MPa provided the best result. A short cold shock or heat shock, administered just prior to osmotic treatment, further improved survival. Addition to the osmotic solution of abscisic acid (ABA), which is involved in the stress response in seeds, also resulted in improved survival of radicle tips. Upon re-imbibition, growth resumption of primary roots was unaffected by the osmotic treatment of germinated seeds with 1 or 2mm long radicles. ABA added to the osmotic solution improved growth resumption and thus ABA addition has no negative residual effect on post-germination growth, but rather stimulated growth.

It was concluded that osmotic treatment, ABA addition, cold shock and heat shock have a positive effect on survival upon desiccation of germinated *Tabebuia impetiginosa* seeds and also on post-germination growth. Seedling establishment may benefit from routine application of such treatments, which has implications for restoration ecology.

<u>Kim N. Hamilton</u>[1], S.E. Ashmore[1] and H.W. Pritchard[2]

Effects of natural distribution of Australian wild citrus on phase transition of seed oils and cryopreservation.

[1]Centre for Forestry and Horticultural Research, Griffith University, Nathan, Brisbane, Qld 4111, Australia
[2]Seed Conservation Dept., Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, W Sussex, RH17 6TN, UK.

Environmental effects are known to influence seed chemistry, particularly oil composition, which in turn can affect seed storability as a result of their thermal behaviour. Differential Scanning Calorimetry (DSC) provides a useful tool to investigate the characteristic thermal properties of phase transitions in oil rich seeds of such as citrus. We investigated the relationship between the thermal transitions in seeds, cryopreservation and geographical origin for rare and threatened northern Australian citrus species, *Citrus inodora* (FM Bailey) and *C. garrawayi* (FM Bailey), and southeastern Australian species *C. australasica* (F Mueller), which is cultivated as a 'bushfood'.

Thermal analysis of phase transitions in cotyledon tissue revealed differences between species in the crystallization and melt onset temperatures of *in vivo* seed oils, suggestive of differences in the proportion of saturated fatty acids. These differences appeared to be associated with geographic gradient, i.e. an increased mean onset temperature of lipid melt coincided with latitude (SE to Far N Queensland) and climatic zone (warm subtropical to hot tropical) of the natural distribution range. In addition, lower melt / crystallization onset temperature appeared to relate to the temperature limit for germination. Tolerance to cryopreservation was demonstrated in all three species after drying, with a mean % germination of 75±2, 71±7 and 42±12 for *C. australasica*, *C. inodora* and *C. garrawayi*, respectively. All three species have edible fruits and are valuable as crop wild relatives (CWR) of citrus. Seed cryopreservation now offers a strategy for the long-term *ex situ* conservation of this germplasm, enabling its improved use in restoration and breeding programs.

<u>C.T. Chien</u>[1], B.S.P. Wang[2] and S.Y. Chen[3]

Bigleaf mahogany (*Swietenia macrophylla* King) seeds in Taiwan exhibit orthodox storage behaviour

[1]Division of Silviculture, Taiwan Forestry Research Institute, 53 Nan-Hai Road, Taipei 10066, Taiwan
[2]Emeritus Research Scientist, Petawawa Research Forest, Natural Resource Canada, Chalk River, ON K0J 1P0, Canada
[3]Division of Forest Biology, Taiwan Forestry Research Institute, 53 Nan-Hai Road, Taipei 10066, Taiwan

Bigleaf mahogany (*Swietenia macrophylla* King) is an exotic tree species and has adapted well to the growing environments in Taiwan. This species is widely planted in Taiwan and seeds have been always treated as an orthodox and stored in sealed plastic bags at 3°C. In view of recently published information on the species that showed intermediate seed storage behaviour, continued planting program of bigleaf mahogany in Taiwan requires confirmation of its seed storage behaviour. Seeds used in this study were drawn from a bulk collection lot and they were dried to 3.7% moisture content (fresh weight) and stored at 3°C for two years with an initial germinability of 82.7%. The two-year-stored seeds were then transferred to -20° and -196°C for continuous storage of another two years.

Result showed that the seeds stored at either -20°C or -196°C retained their original germinability. Thus, seeds of *S. macrophylla* harvested in Taiwan exhibited orthodox storage behaviour.

P. Chmielarz[1],and C. Koziol[2]

Cryopreservation of *Quercus robur L.* plumules.

[1] Institute of Dendrology in Kórnik, Polish Academy of Sciences,
Parkowa 5, 62-035 Kórnik, Poland
[2] Forest Gene Bank Kostrzyca, State Forests, Miłków 300, 58-535 Miłków, Poland

Cryopreservation was conducted on the plumules (growing tips for the shoots), isolated from the embryonic axes of Pedunculate Oak (*Quercus robur* L.), which produces seeds sensitive to desiccation and low temperatures (kat. r*ecalcitrant*). Two methods of desiccation and two methods of freezing in the liquid nitrogen were used. Survival after unfreezing was evaluated in the *in vitro* cultures. The plumules were exsiccated in the stream of gaseous nitrogen – N – or in Petri dishes over the silica gel – G. The next step was freezing by shooting the plumules into the chilled nitrogen (~ -210°C) – S – or directly, in vials filled with the liquid nitrogen (-196°C, LN) - V, after freezing the samples were stored for 24 hours. In case of all the variants, the same method of cryoprotection of plumules was used, alternately in solutions of sucrose and glycerol.

Survival of unfrozen plumules after the storage in LN was from 21% to 67%. The method of desiccation had a significant influence on the percentage of the survival of plumules. An interaction between the method of freezing and the method of desiccation was observed as well. The highest level of survival of plumules frozen in LN was only gained after desiccation over the silica gel (G) and freezing in vials filled with liquid nitrogen. Survival of plumules exsiccated in the stream of gaseous nitrogen (N) was at the significantly lower level (21% - direct freezing (V) and 37% - freezing by shooting into nitrogen (S). Freezing by shooting into nitrogen (S) enabled the survival of 37-49% of samples (statistically irrelevant differences) regardless on the method of the plumules desiccation. Average regeneration of unfrozen plumules used in the experiment into the shoot was 14%.

The project was carried out in frames of the common research project of the Institute of Dendrology, Polish Academy of Sciences in Kórnik, and the Kostrzyca Forest Gene Bank. According to the researches, it was proved that there is a possibility of storage with use of the cryopreservation method in case of the Pedunculate Oak.

Stephen W Adkins and Yohannes M S Samosir

Embryo transplant: a potential technique for propagation of high value, endosperm-mutant coconuts

School of Land, Crop and Food Sciences, The University of Queensland,
Brisbane 4072, Australia

Low income, smallholder coconut farmers have been facing difficulties for decades due to the falling price of copra, their traditional cash product from coconut. These farmers are now looking towards new, higher value products from coconut to make their industry viable over the longer term. There are a number of elite coconut types that have either a soft, jelly-like endosperm or a flavoursome, aromatic drinking water. Both kinds are naturally occurring mutants which cannot germinate in nature as their endosperm cannot support the germination of the embryo. For propagation, the embryo has to be removed from the fruit and grown in vitro to produce a seedling. This can take many months to do and is very inefficient. Thus, a new and innovative technique has been developed for the re-establishment of coconut seedlings, from the isolated zygotic embryos of these mutants, using a surrogate fruit host. This technique is simple to undertake, cheap, rapid and reliable. In the latest form of the technique, embryos are carefully removed from the dehusked donor fruit, in an enclosing endosperm cylinder using aseptic technique. At this point in the procedure these cylinders enclosing the embryo can be moved nationally or internationally using a series of previously published protocols. At the home base, similarly prepared surrogate fruits are used to receive the endosperm plugs. The surrogate fruits, with the transplanted embryos, are then allowed to germinate naturally, under protected environmental conditions. We have now obtained the first embryo-transplanted coconut seedlings using this technique.

ABSTRACTS: Poster cocmmunications

Xia Ke [1], W. Stuppy[2], M.I. Daws[2], Z.K. Zhou[1] and H.W. Pritchard[2]

Comparative Fruit Biology of Chinese Evergreen Oak Seeds (*Quercus* subgen. *Cyclobalanopsis*, Fagaceae)

[1]Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, PR China.
[2]Seed Conservation Dept., Royal Botanic Gardens, Kew, Wakehurst Place, Ardingly, RH17 6TN, UK.

While the fruit / seed biology of *Quercus* subgenus *Quercus* has received considerable attention, much less is known about subgenus *Cyclobalanopsis* which is distributed mainly in tropical and sub-tropical Asia. Consequently we investigated both germination and responses to desiccation for 12 such species from S China. Similar to subgenus *Quercus* (e.g. *Q. robur*) we found that all 12 species had desiccation-sensitive (recalcitrant) seeds. For the 12 species which had seed masses spanning 0.58 to 25.0 g, there were wide differences in drying rates. These differences were independent of seed mass and appeared to be related to pericarp morphology. In addition, in comparison to 8 European species of *Quercus* subgenus *Quercus* (including *Q. robur* and *Q. suber*), seeds in subgenus *Cyclobalanopsis* germinated more slowly and had maximum germination at higher temperatures. For example, across the European *Quercus* (subgenus *Quercus*) species the average time to 50% of maximum germination, at 15°C, was 36d compared with 108 d for subgenus *Cyclobalanopsis*, germination at 25°C took an average of 27 and 44 days for subgenera *Quercus* and *Cyclobalanopsis*, respectively.

Efisio Mattana[1], M.I. Daws[2] and G. Bacchetta[1]

Germination ecology of *Rhamnus persicifolia* (Moris), an endemic tree species of Sardinia (Italy)

[1] Centre for the Conservation of Biodiversity (CCB), Department of Botany, University of Cagliari, 09123, V.le S. Ignazio da Laconi 13, Cagliari, Italy.
[2] Seed Conservation Dept., Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, W Sussex, RH17 6TN, UK

Rhamnus persicifolia Moris is an endemic small tree or shrub referred to the *R. catharticus* L. group. It occurs in the mountains of Central-East Sardinia, growing in scattered groups or single trees, in alder scrub along water streams and in holm oak woods. The full extent of its distribution in Sardinia is unknown and no studies have been carried out to investigate its ecology and seed biology before.
Here we present data for the effects of temperature, light and prechilling on *R. persicifolia* germination. In particular, the effects of a range of constant temperatures (5-30°C) and two alternating temperature regimes (15/5 and 25/10°C) were investigated at two different photoperiods (8 h light / day and darkness); the results achieved at the best conditions were also compared with those obtained at the same parameters after a pre-chilling period (5°C for 3 months).

R. persicifolia germination occurs at warm temperatures (above 20°C), with no differences between constant and alternating regimes and light or darkness photoperiods; furthermore the pre-chilling period improves the final germination percentage and the germination speed (calculated by t_{50} value).

These studies provide new data on the seed biology of this endemic species previously unknown, that could be helpful to understand its ecology and to assess its real conservation status.

Jayanthi Nadarajan[1*,2,a], M Mansor[2], B Krishnapillay[2], H Staines[1]
E Benson[3] and K Harding[3]

Applications of differential scanning calorimetry in developing cryopreservation strategies for *Parkia speciosa*, a tropical tree species

[1]University of Abertay Dundee, Bell Street. Dundee DD1 1HG, Scotland, UK
[2]Forest Research Institute of Malaysia (FRIM), Kepong, 52109 Kuala Lumpur, Malaysia
[3]Research Scientists, Conservation, Environmental Science and Biotechnology, Damar, Drum Road, Cupar Muir, Fife, KY15 5RJ, Scotland, UK
*Author for correspondence
[a](Current contact address) Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, West Sussex RH17 6TN, UK

Shoot-tips of *Parkia speciosa,* a recalcitrant seed producing tropical leguminous tree withstood cryopreservation using encapsulation-vitrification in combination with trehalose preculture. Differential Scanning Calorimetry (DSC) revealed that trehalose moderated the thermal characteristics of the shoot-tips. A 30 min PVS2 treatment had the lowest glass transition temperature (Tg) (-50.21 ± 1.07ºC) when applied in combination with 5% (w/v) trehalose. The Tg increased to -40.22 ± 0.95ºC as the sugar's concentration was decreased to 2.5% (w/v). Tg heat capacity for shoot-tips treated with 2.5% and 5% (w/v) trehalose and exposed to PVS2 for 30 min increased from 0.17 ± 0.05 to 0.23 ± 0.01 $J.g^{-1}$ respectively. Enthalpies of the melt-endotherm varied in proportion to trehalose concentration, for the 30 min PVS2 treatment, whereas the melt enthalpy for control shoots was >150 $J.g^{-1}$ and decreased to ca. 60 $J.g^{-1}$ with 2.5% (w/v) trehalose. For 5% and 10% (w/v) trehalose treatments, enthalpy declined to ca. 24 and 12 $J.g^{-1}$ respectively and freezing points were depressed to

–75ºC and –85ºC with 2.5% and 5% trehalose (w/v), respectively. DSC elucidated the critical points at which vitrification occurred in germplasm exposed to trehalose and PVS2. A 60 min PVS2 treatment supporting ca. 70% survival was found optimal for stable glass formation during cooling and on rewarming.

Thomas Roach.[1,2], L. Colville.[1], M. Ivanova [3], R. Beckett[3], F. Minibayeva [4] and I. Kranner.[1]

Mechanisms of extracellular superoxide production in embryonic axes of *Casatanea sativa* seeds

[1]Seed Conservation Dept., Royal Botanic Gardens Kew, Wakehurst Place, Ardingly, W Sussex, RH17 6TN, UK
[2]University College London, London, WC1E 6BT, UK
[3]School of Biological and Conservation Science, University of KwaZulu-Natal, Pietermaritzburg, Scottsville 3209, Republic of South Africa.
[4]Kazan Institute of Biochemistry and Biophysics, Russian Academy of Sciences, P.O. Box 30, Kazan 420111, Russian Federation.

Mechanisms of extracellular superoxide ($O_2^{\bullet -}$) production were studied in *Castanea sativa* seeds in response to embryonic axis excision and desiccation. Axis excision led to an immediate burst of $O_2^{\bullet -}$ production, demonstrated by a colorimetric assay using epinephrine, electron spin resonance and staining with nitroblue tetrazolium. Isolated axes subjected to variable levels of desiccation stress showed a decrease in viability and vigour. Superoxide production displayed a bell-shaped pattern in response to increasing desiccation stress. The production of $O_2^{\bullet -}$ was sensitive to sodium azide, and could be stimulated by H_2O_2 and NADH, suggesting that the apoplastic enzymes involved in $O_2^{\bullet -}$production include peroxidases. Cell wall fractionation of embryonic axes revealed that the peroxidases potentially involved in $O_2^{\bullet -}$ production were mainly bound to the cell wall by strong electrostatic forces. The chemical composition of the leachates produced by excised axes was investigated using GC-MS. Individual compounds found in the leachates were tested for their capability to stimulate $O_2^{\bullet -}$ production, suggesting that phenolic acids rather than NADH are among the likely natural reductants used by these peroxidases.