ABSTRACTS FROM THE IRISH BOTANISTS' MEETING, BELFAST, MARCH 29–31, 1999

Edited by

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INTRODUCTION

The Irish Botanists' Meeting (IBM) is an annual conference for all plant scientists in Ireland. In 1975 Professor Eric Simon (now deceased) of the Queen's University of Belfast and Dr (now Professor) Matt Harmey of University College Dublin met at a plant science meeting in Wageningen, the Netherlands. They resolved to bring the plant scientists of Ireland together at a meeting closer to home. In 1976 the first Irish Botanists' Meeting, hosted by University College Dublin, took place in the Royal Dublin Society rooms, with delegates from University College Cork, University College Galway, the Queen's University of Belfast and Trinity College Dublin as well as University College Dublin. Following the success of the first meeting, participants agreed to continue to meet at a different venue each year. Today the meetings are rotated annually between the constituent universities of the National University of Ireland (NUI Galway, NUI Maynooth, University College Cork and University College Dublin), the Queen's University of Belfast, Trinity College Dublin and the University of Ulster, Coleraine.

Not only does the IBM provide a valuable opportunity for postgraduates and postdoctoral students to present their work, but a guest speaker is usually also invited to contribute to a debate on a topical issue, thus providing a wealth of new scientific material and encouraging lively discussions. Contributors to the IBMs are also actively involved in their organisation and running.

The IBMs aim to provide a relatively informal setting for the presentation of work in progress, thus promoting communication among plant scientists on this island and leading to the formation of a larger pool of expertise and experience. The following Abstracts from the 1999 IBM demonstrate strong commitment to research in all aspects of plant science, including molecular genetics, mycology and environmental and conservational matters.

As the number of papers submitted to the IBMs has grown rapidly over the past two decades, many must now be presented in poster form. In the following alphabetical (by author) list, no distinction is made between oral and poster contributions.

THE INFLUENCE OF REDUCED FERTILISER INPUTS ON N AVAILABILITY AND CO_2 PRODUCTION IN SOIL UNDER DIFFERENT CROP ROTATIONS

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This study investigated the effect of a two-year grass break (GR) compared with an arable break (AR) in crop rotations on soil nitrogen (N) availability and carbon dioxide (CO_2) production at the Agricultural Research Institute of Northern Ireland. Both rotations were under potatoes in 1995 and winter wheat in 1996. The AR plots went into winter barley in September 1996 and oilseed rape in September 1997. After the wheat harvest in September 1996 the GR plots remained fallow prior to the sowing of spring barley in March 1997, followed by winter barley in September 1997. Each rotation had three levels of input. Current farming practice (CFP) used levels of agro-chemicals comparable to the best commercial practice in Northern Ireland, low input approach (LIA) used 50% of the CFP levels and a minimum input (MIN) received no inorganic fertiliser. Nitrogen availability and CO_2 pro-

duction was measured in the top 15cm of soil in aerobic incubations, at soil temperature, at two-week intervals from March 1995 to September 1998. A two-year grass break in an arable rotation increased both soil available N and CO_2 production compared to an arable break and the N effect was still significant after 4 years. Lowering the level of input had no significant effect on either the release of soil N or CO_2 production.

STUDIES IN HEATHLAND ECOLOGY AND SOIL EROSION IN THE FALKLAND ISLANDS

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Heathlands cover a large part of the Falkland Islands and are dominated by the dwarf shrub *Empetrum rubrum*. The underlying peaty soils are often very dry and are susceptible to erosion following mismanagement, such as overgrazing and burning. Prior to the literature review by D. Broughton (unpublished M.Phil. thesis, the Queen's University of Belfast, 1999) the heathlands of the Falkland Islands had been poorly investigated in terms of their ecology. This project aims to monitor existing erosion in the islands as well as use locally available treatments to revegetate eroded areas. Three treatments (grass mulch, sheep dung and ground peat) were applied to clay patch surfaces, both singly and in combination. After three months, all treatments were found to be successful in improving plant growth in the clay patches when compared to the control areas (i.e. without treatments). The treatments containing dung promoted plant growth most significantly ($p < 0.001$), with an average cover of 12.5% on treatment areas with dung compared to 9.2% cover in areas without dung. Dung also significantly ($p < 0.001$) increased the number of species growing in the treatment areas—0.83 species per plot in those areas without dung added compared to 4.00 species in those areas receiving applications of dung. Further investigations into a greater range of treatments are planned for 1999/2000, as are experiments to monitor the seed rain in clay patches and the surrounding vegetation in order to better understand the processes underlying revegetation.

PHYSIOLOGICAL TESTS AS INDICATORS OF DORMANCY STATUS AND QUALITY IN SEEDLINGS OF BEECH (*FAGUS SYLVATICA* L.) AND OAK (*QUERCUS ROBUR* L.)

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In Ireland the demand for broad-leaved tree species is increasing, particularly for beech (*Fagus sylvatica* L.) and oak (*Quercus robur* L.). However, the field mortality of beech is often quite high, while in oak early growth rates and plant form are frequently poor, although survival is quite good. The physiological quality of seedlings may be a contributing factor to this poor performance. In this study the physiological status, particularly the dormancy development, of beech and oak seedlings was determined at two- to four-week intervals from October 1997 (December for beech) to May 1998. It was tested using a number of physiological indicators, including root electrolyte leakage, root growth potential, water potential, dry weight fraction and days to bud break. In addition to the physiological changes during the lifting season, stress due to the storage of seedlings (cold 2°C and warm 15°C storage) was tested with the same parameters. The changes gave a good indication of dormancy status of the plants but may be poor indicators of plant vitality after storage.

TEXTURAL AND HISTOLOGICAL VARIABILITY BETWEEN PERENNIAL, HYBRID AND ITALIAN RYEGRASSES

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Herbage intake in grazing dairy cows is influenced by a number of sward and plant factors. Textural properties of leaf laminae have a potential influence on the cow's ability to ingest and process herbage; tensile properties may affect herbage intake at the prehension stage, and shear properties influence herbage degradation in the rumen and affect rate of flow through the rumen. A number of ryegrass (*Lolium*) species (perennial, hybrid and Italian), with perennial ryegrass varying in ploidy level, were studied to assess textural properties which may be relevant to herbage intake. Tensile and shear properties of first fully-developed leaf lamina, taken at random from a population sample of each of the ryegrass varieties, were determined using a Lloyd Texture Analyser and related to histological components. Transverse cross-sectional area (CSA) and sclerenchyma and vascular tissue content and distribution were determined from embedded leaf sections using image analysis. Tensile and shear strengths varied between varieties (range 6.60–12.28 N mm^{-2} and 1.14–2.37 N mm^{-2} respectively) and index of masticatory load (IML), a measure of shear force required to cut a mass of material into particles small enough to pass out of the rumen, also varied significantly between varieties (range 1.48–1.79 kg g^{-1} DM, S.E. 0.07). Tensile and shear strength were related to the amount of sclerenchyma in the leaf and to the distribution of the sclerenchyma in terms of relative bundle size and number per CSA. This work highlights the variability in textural properties within the *Lolium* genus and their relationship with histological components of leaf laminae. It provides evidence to justify an in-depth study of textural variability and its effect on herbage intake between varieties and species of ryegrass.

WHERE HAVE OUR NATIVE WOODS GONE?

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Ireland, both north and south of the border, is one of the least forested parts of Europe. Since the Gulf Stream gives Ireland mild temperatures with high rainfall—which should favour luxuriant woodlands—the question must be asked why there is such a scarcity of ancient mature native trees. The answer can be found in early literature, including poetry, records of battles, commercial and religious records and state papers. It appears that the greatest loss of native trees occurred between 1600 (when there was about 12.5% cover) and today (Northern Ireland now has only 1300ha of deciduous state forest). Timber exploitation over the last 400 years has created a dearth of trees. The main uses were timber export, fuel for factories (glass, silver), charcoal for iron-smelting, bark for leather tanning, domestic heating and ship-building. Today the state forest services in Northern Ireland and in the Republic contribute to timber production, but native woodlands are still seriously depleted. Much education of public authorities and the general public remains to be done. Reverence for mature trees is superseded by reverence for large houses and modern conveniences. The only legal controlling system, the 'Tree Preservation Order', to protect mature and otherwise significant trees—has been and still is largely symbolic.

WILLOW: THE INSECT DIMENSION

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The willow is as old as any of the earliest recorded plants and, with the exception of a few insignificant equatorial regions, it has practically a world-wide distribution. It is undoubtedly for these reasons that willow has been grown and utilised for many different purposes over the ages. During its evolution, many associations with insects have developed so that today, in the British Isles, willow rivals oak in terms of the number of species that have become associated with it. As such, willow plantations are potentially a valuable wildlife reservoir for the environment. However, monocultures are notoriously vulnerable to damage by over-exploitation, by disease or by insect species. Willow as a plant is highly disposed to cross-breeding and numerous varieties and clones have been developed that differ in characteristics such as susceptibility to disease or

herbivory by insects. In 1997 a replicated experiment was established at Loughgall, Co. Armagh, comprising a new planting of five clones of willow to examine variation in the impact of insects on the productivity of short rotation coppice willow. Three treatments were imposed—uncontrolled pest attack, spraying when attack was warranted by monitoring and manual defoliation. During the establishment year and the first production year the only significant pest infestation was by *Phratora vulgatissima* ('Blue' Willow Beetle). Spray applications of deltamethrin were timed to coincide with population peaks of the beetle in May/June, when the overwintering population had entered the crop, and in late July, when the new generation of beetles emerged. Although no significant damage due to invertebrate pests could be detected in either year, the manual defoliation produced significant reductions in productivity in both the establishment phase and the first production growth year, indicating the potential for serious damage should climatic conditions permit large beetle populations. One variety showed a trend towards lower rust infection and infestation with invertebrate pests. Future work is planned to investigate the possibility of interaction of clone, rust infection and insect colonisation.

AN EXAMINATION OF THE PARTITIONING AND TRANSPORT OF CARBON IN MYCORRHIZAL CUCUMBER PLANTS

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Previous studies have shown that arbuscular mycorrhizal (AM) colonisation imposes a sink demand for carbon (C) in roots. It has also been hypothesised that an AM-mediated improvement in phosphorus (P) status stimulates the assimilation of C in leaves. The objective of this study was to distinguish between a sink effect and the effect of an improved P status. Gas exchange, ^{14}C-labelling and nutrient analyses of leaves and phloem exudates were conducted to compare how C is partitioned within leaves and translocated in AM, non-mycorrhizal (NAM) and non-mycorrhizal P supplemented (NAM + P) cucumber plants. ^{14}C-labelling data revealed that leaves of AM and NAM + P with P concentrations higher than in NAM plants showed a shift from the accumulation of starch to the synthesis of exportable sugars. However, analysis of phloem exudates showed that increased sugar loading into the phloem and transport of sugars from leaves to roots in AM plants was independent of leaf P status. In conclusion, the results suggest that mechanisms involved in regulating C partitioning within cucumber leaves are dependent on P status and leaf age, while enhanced sink strength, associated with AM colonisation, increases C transport to the roots.

GROWTH OF *SCHIZOPHYLLUM COMMUNE* IN LOW-OXYGEN ATMOSPHERES

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The fungus *Schizophyllum commune* is now commonly found colonising big-bale silage in Ireland. Silage bales are routinely wrapped in four layers of polythene, creating a microaerobic environment favourable to microbial fermentation and silage preservation. M.A. Sprague (Oxygen disappearance in alfalfa silage (*Medicago sativa* L.). In *Proceedings of the 12th International Grassland Congress*, vol. 3 (1974), 651–6) showed that within 30 minutes of sealing a bale of alfalfa silage, the oxygen (O_2) concentration had decreased to 0.5%. An anaerobic environment is probably rarely achieved as some O_2 diffusion can occur through the wrap and bales are often damaged. Oxygen concentrations in older intact bales are typically in the range 0.5–2.0%. The present investigation was undertaken to examine the ability of *S. commune* to grow in low O_2 environments such as might occur in big-bale silage. *Schizophyllum* growth in various gas mixtures was assessed using both radial and biomass measurements of colonies cultured in potato-dextrose media at 27°C for nine days. Experiments were carried out using a gas-tight incubation chamber, through which gas flowed at 50ml min^{-1}. *Schizophyllum* grew at 0.1% O_2 and 0.01% O_2 (both in N_2). The mean mycelial dry weight of three *Schizophyllum* isolates was 91.6 ± 10.9mg at 0.01% O_2 relative to 170.4 ± 4.9mg at 21% O_2 ($n = 27$). It was concluded that *Schizophyllum* can grow well at the O_2 concentrations present in big-bale silage.

THE DWARF SHRUB HEATHLANDS OF THE FALKLAND ISLANDS: AN OVERVIEW

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The vegetation of the Falkland Islands is a diverse shrub heath dominated by the woody shrubs *Empetrum rubrum* and *Baccharis magellanica* and the grass *Cortaderia pilosa*. Current land use is for extensive sheep grazing, and there is a potential to increase cattle production. The heathlands are important in terms of their agricultural, nature conservation, scientific and cultural value. It is therefore important to understand and quantify the dwarf shrub vegetation in terms of the environment that has helped shape it and the effects of current management practices on the heathland environment. This habitat is also important for the conservation of biodiversity within the Falkland Islands and in a larger biogeographical context. The baseline information is now available that should aid in the development of new management plans based on scientific principles and allow the heathland resource to be managed in a sustainable way, compatible with the maintenance of high biodiversity.

THE FLORA OF SAUNDERS ISLAND, FALKLAND ISLANDS

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Previously unpublished data on the ecology and distribution of the vascular flora of Saunders Island in the north-west of the Falkland Islands archipelago has been synthesised to produce a combined checklist and ecogeographic conspectus for the flora of the island. This checklist represents the first regional Flora produced for the archipelago and is therefore an important advance. The flora is a relatively rich one, with 170 species present, representing 61% of the Falkland Islands flora. The species count includes ten of the fourteen taxa endemic to the archipelago. The two endemic taxa *Gamochaeta antarctica* (Hook. f.) Cabrera and *Plantago moorei* Rahn, previously only known from their type localities, are tentatively identified as being present in the flora. The alien taxa *Ribes uva-crispa* L. and *Sambucus nigra* L. are present and represent 'new' additions to the Falkland Islands flora. Ecogeographic studies are predictive and can be used as a guide for future collecting missions and in determining areas of importance for (a) *in situ* conservation and (b) collection for *ex situ* conservation.

THE DETERMINATION OF THE HULLABILITY AND KERNEL CONTENT OF OATS WITH A LABORATORY DEHULLER

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Oat variety samples obtained from the 1997 Northern Ireland Recommended List (NIRL) trials were test milled, using a laboratory dehuller, to investigate the effects of the duration of dehulling and the moisture content of the grain on the hullability, or ease of husk removal, and the potential for determining the kernel content of an oat lot. Increased duration of dehulling from 20 to 120 seconds resulted in a lower proportion of grain remaining unhulled and a lower mechanically determined kernel content. Significant variety differences in the proportion unhulled were detected at shorter dehulling durations. However, this declined until no significant differences were detected at 120 seconds. Reducing grain moisture content from 13.3% to 10.7% resulted in a significant reduction in the proportion of grain remaining unhulled, but the kernel was not significantly affected. Variety samples from the 1996, 1997 and 1998 NIRL trials were milled for 40 seconds, after moisture contents had been equilibrated. Significant differences were detected between varieties in terms of the proportion of grain remaining unhulled. These were consistent across years, Barra having the lowest proportion unhulled and Gerald the highest. Significant differences in kernel contents were detected between varieties and between the conventional hand and mechanically determined kernel contents. Mechanically determined kernel contents were consistently 2% lower than the hand method, as reflected in the non-significant variety-by-method interaction. Results obtained for Barra and Gerald, two commonly milled oat varieties in Ireland, reflect millers' observations in both ease of husk removal and extract yield obtained, with Barra being the preferred milling variety.

XYLEM DIFFERENTIATION IN PLANTS: A SIGNALLING ROLE FOR HYDROGEN PEROXIDE

E. Burbridge

Oxalate oxidase generates the toxic radical hydrogen peroxide (H_2O_2). Peroxidase uses H_2O_2 to fuel its reactions while at the same time converting H_2O_2 to water and oxygen. Both enzymes are produced normally in very small amounts in plants. Horseradish peroxidase and wheat oxalate oxidase have been transformed into tobacco, and hybrids between the peroxidase transgenic plants and the oxalate oxidase transgenic plants have then been formed. Peroxidase is the enzyme that polymerises monolignols in the cell wall in the presence of H_2O_2 to give lignin. Oxalate oxidase is a source of H_2O_2 and its role in plants has not been defined. Germin, the only well known oxalate oxidase is present during germination and is therefore thought to have a role in early development. The results of studies of the effects of their over-production on plant physiology, mainly the deposition of lignin in secondary cell walls and the differentiation of cells into tracheids are presented and contribute towards a better understanding of the role of H_2O_2 in these processes. Earlier work has been undertaken on molecular analysis of these plants to determine whether they have the gene and express the proteins of interest. Lignin levels of wild-type tobacco and peroxidase transgenics have been compared. The over-expressing peroxidase plants have shown a decrease in lignin content compared to wild-type plants.

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EFFECTS OF LIGHT INTENSITY ON THE NITRATE CONTENT OF PROTECTED LETTUCE

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New EU legislation imposes limits on nitrate content of lettuce: 3500mg kg^{-1} fresh weight in summer and 4500mg kg^{-1} in winter. Two experiments were carried out to investigate the effects of light on nitrate content. The first endeavoured to quantify the effect of light intensity on nitrate content of protected lettuce by imposing reduced light levels (shade) during summer daylight hours. The second experiment studied diurnal variation in nitrate content of lettuce. Lettuce seed cv. Clinton was sown into peat blocks and subsequently planted in soil filled containers. Four light levels, 100%, 76%, 57% and 43%, of natural daylight were obtained by shading with muslin cloth. Two nitrogen (N) fertilisers, calcium ammonium nitrate (CAN) and calcium cyanamide were included at two rates to supply 100mg N l^{-1} and 200mg N l^{-1}. These treatments were combined in a split plot design with ten replications. Plants receiving full light had a higher nitrate content (2602mg kg^{-1}) than those under shade, where levels of 2161mg kg^{-1}, 2303mg kg^{-1} and 2142mg kg^{-1} (S.E. 79.4) were recorded. Shading had no effect on fresh weight but dry weight was reduced, being 4.62g, 4.56g, 4.02g and 3.89g at 100%, 76%, 57% and 43% daylight respectively (S.E. 0.12). Calcium cyanamide produced lower nitrate levels (2211mg kg^{-1}) than CAN (2393mg kg^{-1}, S.E. 46.0). In the second experiment, seven plants of cv. Troubadour were randomly harvested from a block at 8.00 hours, 12.00 hours and 16.00 hours. There was no difference in nitrate content between plants harvested at different times.

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MONITORING OF ENVIRONMENTALLY SENSITIVE AREAS IN NORTHERN IRELAND: AN OVERVIEW

A. Cameron, J. Campbell and J.H. McAdam

The Environmentally Sensitive Areas (ESA) scheme commenced in Northern Ireland in 1988 with the launch of the Mournes and Slieve Croob ESA. The programme was expanded until 1992, and there are currently five ESAs, covering 20% of the land area. The scheme is voluntary and is aimed at safeguarding areas of the countryside where the landscape, wildlife or historic interest would benefit through farmers engaging in, or continuing with, environmentally sensitive farming practices. There has been an uptake of 70% in some areas, which represents a considerable investment. The ESA monitoring programme was initiated in 1993 to determine the effectiveness of the ESA scheme in fulfilling its stated objectives. Baseline biological and landscape surveys were

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carried out in all ESAs, and these were followed by re-surveys after three years. Biological monitoring was based on plant and invertebrate communities as indicators of biodiversity. Landscape monitoring was based on a stratified random survey of 183 0.25km squares, covering 2% of the land area. Data were then extrapolated to provide estimates of landscape character for each ESA. Data for land under ESA agreement and land not under ESA agreement were split, and comparisons made between years. Generally, biodiversity and landscape character increased on ESA agreement farms and remained static or declined on non-agreement farms.

THE EFFECTS OF BURNING AND FLAILING ON HEATHLAND REGENERATION IN NORTHERN IRELAND

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Moorland has been traditionally managed by rotational burning of small patches of older heather to create a mosaic of uneven aged stands. A combination of mature heather and young developing heather is the most desirable combination for production of biomass and maintenance of species diversity. Older stands of heather provide shelter for sheep and cover for grouse and other ground nesting birds, while younger stands have a higher proportion of new shoots and are consequently more palatable and of higher nutritive value. Uneven aged stands have been shown to have a greater invertebrate species diversity, due to the greater range of structure and ages. More recently, flailing has been introduced as a management tool with the same objectives as burning. There is little history of controlled heather management in Northern Ireland, and there is concern over some of the practical limitations attached to both techniques. Burning is labour intensive and very dependent on the availability of appropriate weather conditions. Flailing is a more attractive option for some farmers but it is important to establish whether the heath regeneration and species composition vary significantly from those obtained through burning. Monitoring of a number of recently burned and flailed sites was initiated in 1997, and these sites were re-monitored in 1998. Percentage cover of plant species within four equidistant $1m^2$ quadrats along a 20m transect through each site, and within adjacent unmanaged controls was recorded. Transects were marked with a magnetic bearing, taken from a permanent marker, to allow relocation the following year. Soil samples were taken for analysis from each site, and measurements of heather age and height were noted, along with any indications of grazing. The resulting plant species data and specific management details provide the basis for a long-term comparative study. This survey will supply information on the suitability and effectiveness of these management practices on blanket heath in Northern Ireland.

THE EFFECTS OF OIL SPILL DISPERSANTS ON CONIDIAL GERMINATION AND ULTRASTRUCTURE IN THE MARINE FUNGUS *DENDRYPHIELLA SALINA*

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The effect of three modern dispersants (Corexit 9527, Enersperse 1583 and Slickgone LTS), with and without added nutrients, on conidial germination and ultrastructure in the marine fungus *Dendryphiella salina* was investigated. In general, conidial germination decreased significantly as the concentration of each dispersant increased. Germ-tube lengths also decreased as the concentration of each dispersant increased. The addition of nutrients significantly enhanced conidial germination in the controls (seawater only) at 5°C, 10°C, 15°C and 20°C and at 10°C and 15°C in all concentrations of each of the three dispersants. However, at 5°C at 100ppm and 1000ppm of each dispersant, and at 20°C at all concentrations of each dispersant, there were no significant differences in conidial germination. The addition of nutrients did not generally enhance germ-tube lengths. Following immersion of *D. salina* conidia for 24 hours at 16°C in (a) seawater and (b) 1000ppm Corexit 9527, studies carried out using a transmission electron microscope revealed new evidence of ultrastructural damage.

WHY HAS *MYCELIS MURALIS* BECOME SO ABUNDANT IN THE BURREN?

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Mycelis muralis is most abundant in Ireland on open limestone pavements in the Burren, despite being regarded as a plant of woodland and shaded habitats throughout most of its range. Although acclimation to high irradiances at leaf level is limited, highest biomasses and reproductive outputs occur in high-light habitats. A combination of low whole-plant growth rates and high allocation to roots may be advantageous in low-resource environments, such as limestone pavements, although these traits may lead to reduced competitive ability in many high-resource habitats. In addition, population differentiation may be important, particularly as Burren populations produce greater amounts of leaf anthocyanins, which could play a role in photoprotection in exposed habitats.

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MADS-BOX GENES IN SITKA SPRUCE (*PICEA STICHENSIS*)

K. Clarke and T.F. Gallagher

The MADS-box gene family encodes a conserved group of proteins found in plants, animals and fungi. In angiosperms these genes have evolved as a large gene family whereas in animals and fungi the gene family has not evolved to the same extent. In angiosperms many members of this family function as transcription factors, where they are involved in the formation of the floral meristem and the subsequent specification of floral organ identity. There is little information on this gene family in plant groups other than angiosperms. The aim of this project is to characterise the MADS-box gene family in Sitka spruce and to examine its expression within developing reproductive structures. Primers were designed against the conserved MADS-box domain. This allowed a 180-base-pair region to be amplified using PCR. Sequence analysis confirmed that this represented a MADS-box domain. Current work involves extending this sequence using techniques such as thermal asymmetric interlaced (TAIL) PCR and RT-PCR to characterise the rest of the gene and the encoded mRNA.

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CONSERVATION GENETICS OF NORTHERN IRELAND HAWTHORN POPULATIONS

S. Clarke, G. Hoppé, C. Fleming and J.H. McAdam

Hedgerows are an important feature of the landscape and a habitat of high conservation status in Ireland. Hawthorn (*Crataegus monogyna*) is the commonest hedgerow species. Concern has been expressed that alien introductions of hawthorn (for over 200 years) may have resulted in the genetic erosion of native stock. This paper describes the impact of imports of hawthorn of European (mainly Dutch) origin on the genetic constitution of the Northern Ireland populations. Samples of hawthorn were collected from 15 sites across Northern Ireland and compared with those of known Dutch origin. The genetic diversity was analysed using a range of molecular genetic techniques. Detection of genetic diversity utilised RAPDs. Variation in disease resistance was assessed at specific sites on the genome. Phylogenetic trees were constructed and statistically tested. Partitioning of genetic variation within and between populations was described and statistically tested. There has been a high degree of introgression of non-native genes into the native Northern Ireland gene pool. Introgression varied between sites, with one population possibly representing a relict of truly native genotypes. The future loss of genetic diversity in hawthorn through the increased importation of hawthorn into Ireland from non-native sources, particularly Eastern Europe, is of concern.

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GENETIC DISTANCE APPLICATIONS FOR PLANT BREEDING

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Plant Breeders Rights have been used to protect new varieties for the plant breeding industry for over forty years, by allowing the awarding of rights to accessions that have been proven to be phenotypically unique. Recent developments in technology have provided plant breeders with the means of creating new and improved varieties with greater precision and in less time. Genetic engineering, recombinant DNA technologies and marker assisted breeding are but a few of the methods available, and these are becoming increasingly more popular as a way to produce novel varieties. In 1997 the problem of possible exploitation of the existing system was addressed and the concept of Essential Derivation was introduced by the Union for the Protection of Varieties of Plants (UPOV). Essential Derivation recognises the fact that any proposed accession may have been derived from a protected variety as opposed to being bred as part of an independent breeding programme. However, the assessment of Essential Derivation requires a measure of genetic distance to expose the extent of breeding effort that produced the accession in question. The aim of this project is to develop an understanding of how such a measure could be accomplished with specific reference to *Lolium sp.*, where particular difficulties arise due to its inherent intra-varietal variation resulting from the out-breeding nature of the species. Using information from morphological observation of the phenotype to genotypic studies, a clearer picture of how the legal concept of Essential Derivation might be applied to the science of plant breeding is being evaluated. This project is examining a core set of twelve accessions, carefully chosen to indicate similarity and distinctness. Each accession has undergone both laboratory and field testing procedures. Genetic variation has been easily identified between all test accessions, and the expected clustering of similar groupings has been apparent, particularly with PAGE testing of storage proteins and analysis of morphological characteristics.

PRODUCTION OF A PHARMACEUTICALLY ACTIVE SECONDARY COMPOUND, ALLANTOIN, IN TISSUE CULTURES OF *SYMPHYTUM* SPECIES

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Plant tissue cultures can be used for the production of commercially valuable secondary metabolites. One plant species, *Symphytum* (Comfrey) has been used traditionally in various types of pharmaceutical preparations. *Symphytum* is well known for its impressive anti-inflammatory and wound-healing properties. These medicinal properties are due to the presence of the ureide, allantoin. Through tissue culture it may be possible to produce allantoin (Aln) *in vitro*, which eventually may be used for the industrial production of allantoin based pharmaceuticals. Since *Symphytum orientale* is also an endangered species, tissue culture offers the possibility of increasing plant numbers through somatic embryogenesis. Callus cultures of three species *S. x uplandicum*, *S. officinale* and *S. tuberosum* have been established. Preliminary TLC studies made of cell extracts from the cultures are presented here. It is proposed that full nutrient analysis, screening and selection of cell lines and bioreactor studies will be investigated, with the aim of producing the highest yield of allantoin *in vitro*. An *in vitro* method for the detection of the anti-inflammatory activity of allantoin will also be developed.

NEW COPROPHILIC SPECIES OF *SPOROTHRIX*, ANAMORPH OF *PHOMATOSPORA COPROPHILA*

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The genus *Phomatospora* belongs to the Ascomycotina and comprises some twenty species which form non-stromatic, black, immersed perithecia with short necks. Most species have not been linked to anamorphic states. Only two coprophilic species are known, *P. coprophila* and *P. hyalina*, both of which are rarely found due to their minute size, submerged habit and relatively long development periods. *P. coprophila* has not previously been recorded in Ireland but was found on several sheep- and cow-dung collections from November 1992 to April 1995, at Calary, Co. Wicklow (O2213), Trim, Co. Meath (N7613) and The Curragh, Co. Kildare (N8057). The

genus has hitherto been represented in Ireland only by *P. argentina* and *P. berkeleyi*. A previously unknown anamorph of *P. coprophila* was discovered when fragments of crushed ascomata were cultured on malt agar supplemented with asparagine. The conidial state was identified as a species of *Sporothrix* Hektoen and Perkins, distinguished by its hyaline, denticulate conidiogenous cells that form on undifferentiated hyphae and from intercalary hyphal cells. Each denticle produces a single conidium before proliferating sympodially to form the next and succeeding conidia. Conidia are falcate, 9–11 × 1μm, hyaline, one-celled, smooth, apex narrowed and base truncate. The fungus is not referable to any known *Sporothrix* species and is recognised as new.

THE PROSPECT OF BIOLOGICAL CONTROL WITHIN NORTHERN IRELAND BRAMLEY ORCHARDS

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Surveys conducted within Northern Ireland Bramley orchards indicate that pest damage is normally inconsequential, even though pest numbers regularly exceed recommended spray thresholds. It is thought that natural enemy fauna within the orchards may be capable of limiting pest populations during times when damage is likely. The predatory mite *Anystis baccarum* is common in the Bramley orchards and is a voracious generalist predator feeding on any prey it can overpower. It may therefore contribute to biological control of pest species in the orchards. The aim of this work was to evaluate the impact of *A. baccarum* as a bio-control agent of apple pests. In the laboratory *A. baccarum* ate, on average, either 6.3 *Panonychus ulmi* per day, 5.2 *Bryobia rubrioculus* per day or 1.2 *Rhopalosiphum insertum* per day. In the field, the impact of *A. baccarum* on *Aculus schlechtendali* was studied. Since *A. baccarum* is mobile on the branches, whereas *A. schlechtendali* is sedentary on the leaves, sticky-tape wrapped around branches was used to trap *A. baccarum* but did not affect *A. schlechtendali*. Treatments of sticky-tape trap, sticky-tape trap plus two applications of demeton-s-methyl (acaracide) and untreated control were allocated to randomly selected branches on each of six trees within an orchard. *A. schlechtendali* populations were assessed weekly throughout the summer and early autumn of 1998. Treatments that affected *A. baccarum* populations usually had greater mean *A. schlechtendali* numbers than treatments where the *A. baccarum* population was unaffected. For example, samples collected on 17 August had mean *A. schlechtendali* populations per 4 leaves of 1138 (sticky trap plus acaracide, S.E. 91.1), 862 (sticky trap, S.E. 45.3) and 597 (control, S.E. 78.4). In analysis of variance on $\log_{10}$ transformed data, $F = 20.28$; d.f. = 2,10; $P < 0.001$. In conclusion, it has been shown through laboratory and field experiments that *A. baccarum* eats the pests of apple and may, therefore, contribute towards biological control within the Bramley orchards.

SHORT ROTATION COPPICE WILLOW IN NORTHERN IRELAND 1973–99

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Interest in growing short rotation coppice (SRC) willow (*Salix*) began in the early 1970s. Initially this interest was generated through the Paper Industry Research Association in response to a projected world shortage of cellulose for paper manufacture. Following the Middle East oil crisis in the mid-1970s the emphasis of the work turned to the potential of SRC willow as an energy source. This was both in terms of diversification of supply and as a response to escalating prices of crude oil. Early trials investigated basic agronomic parameters. A planting density of 20,000 trees per hectare gave 10–12 tonnes of dry matter annually, using a three-year harvest cycle. In the mid-1980s the clonal nature of the crop led to a serious outbreak of foliar rust which became the major limiting factor in production. Currently, new varieties coming from breeding programmes in the UK and Sweden, incorporated into mixed variety planting, are being evaluated as a disease control strategy. A further area of research is the use of willow coppice in the bio-remediation of waste streams, such as the effluent from water treatment works or the leachate from landfill sites. Willow can act as an efficient biofiltration system while at the same time benefiting directly through nutrient recovery. Investigations into the utilisation of willow have resulted in the development of small scale, down-draft gasification for electricity and heat production.

ANALYSES OF IRON DEPOSITS ON THE ROOTS OF *AGROSTIS STOLONIFERA* AND *JUNCUS EFFUSUS* (WETLAND PLANTS) GROWING IN A MINE TAILINGS POND AT GLENDALOUGH, CO. WICKLOW

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At Glendalough, Co. Wicklow, run-off water from the mountain slopes drains through abandoned lead/zinc mine spoils. This water becomes enriched with heavy metals as a result and subsequently flows into a tailings pond and marsh. Wetland plants, including *Agrostis stolonifera* and *Juncus effusus*, grow in the tailings pond where anaerobic conditions prevail. Ferrous (Fe^{2+}) iron is the predominant form found in such environments, and by oxidising this to ferric (Fe^{3+}) iron in the rhizosphere, wetland plants develop so-called iron plaques. This research investigates the occurrence and distribution of the iron plaque on the roots of wetland plant species by employing light microscopy, cryo-electron microscopy, X-ray microprobe analysis and chemical extraction techniques. Results to date indicate that the distribution of iron plaque varies greatly within and between plant species.

ECOLOGY OF *SCHOENOPLECTUS TRIQUETER* IN THE SHANNON ESTUARY

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A study of the ecology of *Schoenoplectus triqueter* in the Shannon estuary commenced in September 1998. *S. triqueter* was found to be more widely distributed in the upper part of the estuary than first anticipated. It occurs in the muddy riparian zone but is confined to the lowest reaches of the zone, with *Bolboschoenus maritima* and *Schoenoplectus tabernaemontani* growing just above it. It occurs in bands of varying width (1.0m–10.0m), in substrates of varying composition, with a substrate pH of between 7.4 and 7.8. The density and stature of *S. triqueter* are greater towards the upper part of these bands, where the periods of inundation are shorter and there is less chance of grazing by waterfowl. Stands of *S. triqueter* are inundated for periods of up to 16 hours per day. The species only appears to occur in the upper parts of the estuary where the salinity is quite low.

THE GENETIC BASIS OF RADIAL PATTERN IN THE *ARABIDOPSIS* ROOT

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Multicellular organisms are composed of patterned arrays of tissue types, and each tissue in turn comprises populations of differentiated cells. During the course of development from zygote to mature organism, the basic elements of the body plan are progressively laid down. The acquisition of a molecular understanding of the mechanisms underpinning this process in plants is the main focus of our research. We use the *Arabidopsis* root as a model system because of the simple organisation of cells and tissues and also because *Arabidopsis* is amenable to genetic analysis. The primary root is composed of concentric pattern elements (tissues) that are laid down during embryogenesis. At the core of the root is the stele, which is surrounded by rings of ground tissue, epidermis and root cap. The epidermis is composed of a patterned array of two cell types, whose respective fates are positionally determined. Hair cells in the epidermis are derived from meristematic trichoblasts and located over the anticlinal (radial) walls of two cortical cells. Non-hair cells are derived from meristematic 'atrichoblasts' and located over the outer periclinal (tangential) walls of underlying cortical cells. We have used a genetic approach to identify genes that are involved in (1) specifying epidermal identity during embryonic pattern formation (2) specifying cell identity within the epidermis and (3) root hair growth. Our progress towards a genetic mechanism of epidermal development in roots will be presented.

A LANDSCAPE APPROACH TO THE MANAGEMENT OF ESKER VEGETATION AT CLONMACNOISE

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Eskers are raised river-beds formed by the deposition of material under melting glaciers and found extensively in the midlands of Ireland. This study centred on the Clonmacnoise area, where a unique esker grassland flora known as the *Antennarietum hibernicae* sub-association is found. The aim of this study was to identify environmental and cultural factors that influence this grassland type and suggest beneficial management regimes for its survival. The species-rich patches are very fragmented in the landscape, and a GIS approach was used to measure their isolation and surrounding land-use matrix. Samples of grassland types present on the eskers were analysed using ordination methods (TWINSPAN, DCA, CCA). From TWINSPAN analysis, four main vegetation types were identified: (i) a grass-dominated, mesotrophic group on steep slopes, (ii) a mixed acidic group on thin soil, (iii) a calcareous group on thin soil on esker crests and (iv) a nutrient-enriched flora on deeper soils. In CCA, environmental factors were analysed, and slope, soil depth, presence of grazing/poaching and bare soil were found to be the main explanatory variables. Butterfly presence and abundance were used as indicators of grassland quality. Butterfly species diversity was higher in more species-rich patches ($p < 0.01$) and species were also more abundant in these patches ($p < 0.01$). Adjacent land-use to species-rich patches were analysed using ARCVIEW, and graded in terms of perceived threat to their survival. This work will enable us to propose an overall management plan for the site.

THE USE OF SPENT MUSHROOM COMPOST AS A NUTRIENT SOURCE IN CEREAL PRODUCTION

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The object of this experiment was to assess the suitability of spent mushroom compost (SMC) as a soil amendment and nitrogen (N) source in winter wheat by studying its effects on yield, quality and N uptake. The experimental design was a split plot with four replications of 8 × 15m plots receiving five rates of SMC application (0, 8t ha^{-1}, 16t ha^{-1}, 32t ha^{-1} and 64t ha^{-1}), containing 8.09kg N t^{-1}. Each plot was split into three subplots receiving three rates of N fertiliser application (0kg N ha^{-1}, 80kg N ha^{-1} and 160kg N ha^{-1}). The SMC was ploughed in, and winter wheat cv. Brigadier was sown. The addition of SMC (comparing means of the 0t ha^{-1} with the 64t ha^{-1} treatments) increased shoot N uptake from 197kg ha^{-1} to 223kg ha^{-1} (S.E. 5.5), grain protein content from 8.92% to 9.47% (S.E. 0.137) and lodging percent from 15.0 to 36.6 (S.E. 2.7). Grain yield was not significantly affected, being 9.93t ha^{-1} and 10.40t ha^{-1} (S.E. 0.154). The addition of fertiliser N (comparing means of the 0kg N ha^{-1} with the 160kg N ha^{-1} treatments) increased shoot N uptake from 175kg ha^{-1} to 239kg ha^{-1} (S.E. 2.99), grain protein content from 8.32% to 9.89% (S.E. 0.074), lodging percent from 14.0 to 35.5 (S.E. 1.65) and grain yield from 9.63t ha^{-1} to 10.40t ha^{-1} (S.E. 0.154). It was concluded that SMC is an inefficient source of N, with only 5.0% of added N being taken up by the current crop compared to 40.4% of chemical N.

REHABILITATION MANAGEMENT OF INDUSTRIAL CUTAWAY ATLANTIC BLANKET BOG

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Commercial extraction of peat from the Bellacorick Atlantic blanket bog complex, north-west Co. Mayo, has produced 1200ha of cutaway bog. When peat fields are abandoned the remaining peat is dry, compressed, bare, acid, nutrient-poor, and exposed to rainfall exceeding 1600mm per annum and frequent strong winds. The success of vegetation establishment on the cutaway is variable, depending on peat depth, time since abandonment, nutrient status and hydrology. Cutaway areas that have been hydrologically isolated from adjacent production bog can sustain embryonic bog communities. The water-table in these areas remains at or above the peat surface throughout the year, promoting the establishment of typical bog species. Vegetation is slow to

establish on areas where the underlying acid glacial till has been exposed. Trials have been set up to determine the effectiveness of a number of management options, including (a) transplantation of birch onto areas denuded of peat, (b) ploughing and terracing and (c) dam construction. This work will contribute to a management plan for Bellacorick that is designed to accelerate vegetation establishment and restore functionally intact peat-producing ecosystems where suitable habitat conditions prevail.

THE ROLE OF ECTOMYCORRHIZAS IN THE PROTECTION OF LODGEPOLE PINE GROWING ON OLD MINE SPOIL

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The acid metalliferous mine spoils at Old Avoca Mines, Co. Wicklow, are unfavourable environments for the establishment and growth of trees. Nursery-grown Lodgepole pine has been planted on mine spoil but exhibits chlorosis and poor growth due to the high magnesim (Mg) and copper (Cu) content in the spoil. Although colonisation by naturally-regenerated pines in these habitats has also been slow, they appear to be coping adequately with the toxic conditions. Differences in ectomycorrhizal populations have been observed between planted nursery seedlings and those that have regenerated naturally. Several mycorrhizal fungi have been isolated in pure culture from fruiting bodies collected in naturally-regenerated forests. All of these cultures formed mycorrhizas on Lodgepole pine. The mycorrhizal fungi of *Paxillus involutus*, *Suillus bovinus* and *Scleroderma citrinum* enhanced the growth of seedlings in limed mine spoil in a pot study. The inoculation with another symbiont (*Lactarius rufus*) produced similar root and shoot lengths to uninoculated controls. It was concluded that certain mycorrhizal fungi occurring naturally at Avoca have the ability to ameliorate Mg and Cu toxicity.

ASSESSMENT OF THE VIRUS, VIROID AND PHYTOPLASMA STATUS OF A COLLECTION OF HISTORICAL IRISH APPLE CULTIVARS

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The Lamb Clarke historical Irish apple collection was established in 1996 to preserve for posterity many old, Irish bred cultivars. The collection currently contains 187 different cultivars. The object of assessing the virus status of the collection was to enable any viruses, viroids or phytoplasmas that were present to be removed, after which all trees could be propagated and disseminated. Once all cultivars were of an equal status their merits and characteristics could be compared. Phytoplasmas were detected using nested primer PCR. The viruses—apple mosaic, apple chlorotic leafspot, apple stem grooving and apple stem pitting—were tested for using RT-PCR. Apple scar skin viroid was tested for using RT-PCR. No trees were found to be positive for any of the viruses or viroid tested. Sixteen cultivars were found to be initially positive for phytoplasmas; however, when tested again in the autumn, all cultivars gave negative results. As viruses can be present at very low titres, and in specific areas of a tree, the collection will require repeat testing in the following two years.

GENE EXPRESSION IN DIFFERENT MORPHOTYPES OF *CYMBALARIA MURALIS*

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Two morphotypes of Ivy Leafed Toad Flax (*Cymbularia muralis*), a red anthocyanin-containing form and a green non-anthocyanin-containing form, can be found growing in the same environment. The molecular basis for this difference is not known but appears to be a single gene mutation. This study aims to investigate the molecular basis for this difference. Northern blot analysis is currently being used to analyse the red and green forms in relation to the genes involved in the phenylpropanol (PAL) pathway. This approach will establish whether the lesion in the non-anthocyanin containing form occurs in this enzymatic pathway. Future work will examine the response of the two forms to elevated UV and will use Representational Difference Analysis (RDA) to search for differences in gene expression between the two forms in areas other than the PAL pathway.

ANALYSIS OF THE REGULATION OF THE *ARRO-1* APPLE GENE

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The molecular mechanisms of adventitious root formation are poorly understood, particularly in woody plants. A*RRO-1* (Adventitious Root Related Oxygenase-1), a gene highly up-regulated during adventitious root formation in apple (Jork 9) stem discs, has been isolated in this laboratory. The aim of this project is to characterise the expression of this gene in detail during adventitious root formation. The 5′ upstream region of *ARRO-1* has been isolated using TAIL-PCR (Thermal Asymmetrical Interlaced PCR), and the resulting 944bp fragment has been cloned and sequenced. A further aim of this project is to obtain an understanding of the expression of *ARRO-1*. This will be achieved by a functional characterisation of this promoter in transgenic *Arabidopsis* and apple plants. It will involve the fusion of the promoter to a GUS reporter gene or a Green Fluorescent protein (GFP). The availability of this material will enable the localisation of gene expression in specific cell types and the dynamics of *ARRO-1* expression during adventitious root formation. These investigations will give us a further understanding of the role and regulation of the *ARRO-1* gene.

AN EVALUATION OF PHYSIOLOGICAL PARAMETERS OF HYBRID LARCH (*LARIX* × *EUROLEPIS*) (HENRY)) SEEDLINGS AS INDICATORS OF STRESS RESISTANCE

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Hybrid larch is gaining favour in Ireland as a minor species, particularly for planting in mixtures. However, the dormancy development and levels of stress resistance achieved during the nursery lifting season in the mild conditions of Ireland are largely unknown. The poor field performance of hybrid larch in recent years has prompted research into the role of physiological changes in maintaining stress resistance levels that occur in the plant during the lifting period from November to March. Transplants of hybrid larch were lifted from the nursery at regular intervals from October to May and dispatched to the university (UCD) for evaluation of physiological status at the time of lifting and following periods of cold (2°C) storage. A number of different parameters were evaluated, including cold hardiness development of the shoot, root electrolyte leakage, root growth potential and dry weight fraction of the shoot tip. Clear seasonal patterns were identified for most parameters. These were further examined in response to prolonged cold storage with the aim of assessing deterioration of the plant during storage. The potential of these parameters as indicators of stress resistance in hybrid larch is discussed.

THE EFFECTS OF ELEVATED CARBON DIOXIDE LEVELS ON THREE SPECIES FOUND IN SIMILAR WETLAND HABITATS

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Carbon dioxide (CO_2) levels have increased steadily since the industrial era, mainly due to human activities. The effects of this elevated CO_2 concentration on wetland plants or plants found in areas of high water availability have not been fully investigated. In an effort to examine this in more detail, three plants found in wetland areas, and which have similar life forms, were studied, and the results are discussed here. *Gunnera tinctoria*, an unusual wetland nitrogen-fixing species, was chosen because previous reports indicated that stomatal opening is relatively insensitive to instantaneous changes in CO_2 concentration. The responses of *G. tinctoria* were compared to those of *Petasites fragrans*, a species with a similar ecology, and *Tussilago farfara*, a species that can survive in wet or dry habitats. *G. tinctoria* plants grown in elevated CO_2 concentrations (680μmol mol^{-}1) had lower maximum photosynthetic rates (Pm) than those grown in ambient concentrations (350μmol mol^{-1}). *T. farfara* and *P. fragrans* plants grown in elevated CO_2 levels had lower maximum photosynthetic rates until week eleven when values rose to those of plants grown in ambient CO_2 concentrations. No stimulatory effect of high CO_2 on plant biomass was observed on *G. tinctoria* and *T. farfara* plants by week seven, with *P. fragrans* showing only a slight increase in dry matter production. However, changes in biomass partitioning occurred at elevated CO_2

levels. An increase in percentage leaf allocation was observed in *T. farfara* plants at elevated CO_2 levels. In *P. fragrans* this was coupled with an increase in the percentage of roots.

IMPACT OF INVASION BY THE NITROGEN-FIXING SPECIES *GUNNERA TINCTORIA* (MOLINA) MIRBEL ON VEGETATION DYNAMICS

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In areas invaded by the nitrogen-fixing herbaceous plant *Gunnera tinctoria*, the plant community changed from a wet grassland type (Phytosociological class Molinio-Arrhenatheretea, order Molinietalia), to one with similarities to the *Salix cinerea-Galium palustre* woodland association. Previous studies have shown that changes in species composition can be explained by increases in soil nitrogen (N) following invasion by N-fixing plants. In this study, changes in species composition were not associated with *G. tinctoria*-induced increase in nitrogen, despite the annual decay of most of its above-ground plant parts. Likewise, there was no evidence to suggest that the alterations were due to other soil parameters measured. Investigations to determine if canopy shade was responsible for the changes in composition proved to be inconclusive. While suppression of some native species was evident, shading did not fully account for the absence of some species from the colonised plots, and this may be a result of reduced seed germination.

SEED BANK DIVERSITY OF HAY MEADOWS IN THE FERMANAGH ENVIRONMENTALLY SENSITIVE AREA

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The species-rich grasslands within the West Fermanagh and Erne Lakeland Environmentally Sensitive Area (ESA) are of national and international importance. This project assesses the soil seed bank resource within a range of hay meadow grassland types with a view to restoring and enhancing biodiversity. Six field sites were sampled in 1995, representing the range of species-rich, species-poor and species-variable hay meadow types within the ESA. Soil samples (5 locations × 5 cores per site) were collected and seeds separated and identified. Herbage species composition was compared with the below-ground seed bank flora. The highest seed bank diversity was recorded within the species-variable hay meadows. Seed abundance (number) was highly variable both within and between hay meadow types. There was a poor correlation between the floristic composition of the vegetation and the seed bank in the species-rich and species-variable sites. However, the correlation in one of the species-poor sites was highly significant. The study indicates the potential for further species enrichment of these hay meadows; the species-variable sites offering the greatest potential for restoration and enhancement of diversity.

IMPROVEMENT OF QUALITY AND SHELF LIFE OF VEGETABLE BRASSICAS BY GENETIC TRANSFORMATION

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The objective of our ongoing research is to improve the quality and shelf life of cauliflower and broccoli through *Agrobacterium*-mediated transformation. To achieve this we used two strategies. First, we introduced a prokaryotic ammonium-specific asparagine synthetase gene (*asn*A) under control of the CaMV-35S promoter, and have been evaluating the effect of this new ammonium assimilation pathway on vegetable quality. It is expected that the altered nitrogen metabolism will have beneficial effects on the nutritional and organoleptic qualities of the transformed plants. The second approach is based on the observation that cytokinins delay the onset of senescence in plants. We introduced an isopentenyl transferase gene (*ipt*) for cytokinin biosynthesis, under control of a senescence-specific promoter ($pSAG_{12}$), and have demonstrated that this auto-regulatory system resulted in altered cytokinin levels and delayed leaf senescence in cauliflower.

INVESTIGATION OF ANTHOCYANIN GENE EXPRESSION AND POPULATION DIFFERENTIATION IN *MYCELIS MURALIS*

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This study investigated the molecular control of anthocyanin accumulation in two populations of *Mycelis muralis*. Northern blot analysis has been used to monitor the expression of PAL, CHS and CHI genes for flavonoid precursors. This was done to determine if there is any difference in the levels of mRNAs encoding anthocyanin genes between the populations. Fifty genotypes from each population were grown under elevated UVA/UVB, elevated UVB and white light treatments. Total RNA was extracted from leaves harvested at different times over a two-week period. Northern analysis showed up-regulated expression of PAL, CHS and CHI in the Burren population, while the Durrow population did not respond to any of the UV treatments. Using RAPD-PCR with twenty random primers indicates that the Burren and Durrow plants can be divided into two separate populations. Dendrograms created using Unweighted Pair Groupings Method using Averages (UPGMA) analysis divided both *Mycelis* populations into two distinct branches.

THE EFFECTS OF MOISTURE TENSION FOR TRIGGERING IRRIGATION ON THE GROWTH OF *VIBURNUM TINUS* IN CONTAINERS

E. Kehoe, M.J. Maher and A. Hunter

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Nursery stock plants grown in small peat-filled containers require irrigation. At present Irish growers irrigate manually, which may be sub-optimal. The object of these experiments was to study the use of digital tensiometers, placed in the growing medium, to control the scheduling of irrigation under two systems—overhead spraylines and ebb-and-flood irrigation. Eighteen independent growing beds (5×2m) were used. The experiments were conducted over two growing seasons using *Viburnum tinus* in two-litre pots. A digital tensiometer set to trigger an irrigation cycle at a tension of 50hPa, 100hPa or 200hPa was placed in a pot in each of the beds. The treatments were combined in a 3×2 factorial design with three replications. Plots irrigated at tensions of 50hPa, 100hPa and 200hPa were watered 30, 19 and 10 times (S.E. 1.3) respectively during the first season and 54, 31 and 21 times (S.E. 1.7) respectively during the second season. There was no difference between the irrigation systems. At the end of the second season the fresh weights of the respective plants given 50hPa, 100hPa and 200hPa tension treatments differed, with mean yields of 174.3g, 162.9g and 151.5g per plant respectively (S.E. 3.17). Measurements of stomatal resistance were taken on a number of occasions using a portable porometer (AP4 Delta-T). Resistance tended to be higher in plants in the high-tension treatments, a typical series showing values of 4.7s cm^{-1}, 5.4s cm^{-1} and 8.2s cm^{-1} (S.E. 0.69) at the 50hPa, 100hPa and 200hPa tensions respectively. It was concluded that a tension of 50hPa was best for initiating irrigation periods and that higher tensions increased water stress and reduced plant weight.

THE EFFECT OF ELEVATED UV ON 'SUN' AND 'SHADE' POPULATIONS OF *MYCELIS MURALIS*

G.J. Lanigan and B.A. Osborne

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Seedlings from populations *of Mycelis muralis* from an open and a woodland site, as well as seedlings of *Sonchus oleraceus*, were subjected to elevated UVA + UVB, UVA and white light only. After two weeks, various morphological, biochemical and photosynthetic parameters were measured. Total biomass, specific leaf mass, total leaf area, leaf nitrogen concentration and a number of photosynthetic traits were all significantly reduced under the elevated UVA + UVB treatment compared to the other two treatments for both populations and *S. oleraceus*, although *S. oleraceus* was generally affected to a lesser extent. Total chlorophyll per unit mass and chlorophyll a/b ratio remained constant for the two populations of *M. muralis* but increased under elevated UVA + UVB for *S. oleraceus*. While the responses of most measured parameters to the various spectral treatments were similar for the two populations of *M. muralis*, leaf anthocyanin concentrations were significantly higher for the Burren population.

THE SUITABILITY OF WILLOWS (*SALIX* SPP) FOR ENERGY, SHELTER, BROWSE, BIOREMEDIATION AND AMENITY USE IN THE FALKLAND ISLANDS

J.H. McAdam, W.M. Dawson and R. Reid

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The climate of the Falkland Islands is cool and windy; soils are acid, infertile peats; vegetation is dwarf shrub heath; and there are no indigenous trees. Energy resources are based on imported oil and some local peat use. Agriculture is low-productivity, extensive sheep farming limited by poor pastures and exposure. To develop agriculture and expand cattle production there is a need for trees for shelter, to improve survival and enable crops to be grown. Trees can also be valuable for amenity planting around settlements. Coppicing species have potential for energy use (chipping and burning) and bioremediation of sewage waste. A series of experimental shelter belts based on Lodgepole pine have been planted, but there is a need to diversify the range of tree species planted. Willow (*Salix* spp) exhibits a huge range of tolerances to soil and climatic extremes, has attractive foliage and bark, coppices well for renewable energy cropping, is increasingly used in bioremediation systems and may offer potential in the Falkland Islands. *Salix viminalis* is occasionally grown locally for hedging, and *S. alaxensis*, *S. lanata*, *S. phylicifolia* and *S. lasiandra* are being tried by the Department of Agriculture (the last two have been used as browse fodder for sheep and cattle elsewhere). In 1990 cuttings of 'Bowles Hybrid', 'Germany', 'Q83', 'Matsudana' and 'Dasyclados' were taken from the Northern Ireland Horticultural and Plant Breeding Station, Loughgall, Co. Armagh, and planted into a damp sheltered site in the Falklands. *Salix* 'Dasyclados' performed best, with 90% of long cuttings surviving. Mean growth was 0.6m, and one plant grew over 1.2m in the season after planting. There is a need for expert evaluation of the potential of willows for a range of situations, for more comprehensive trials to be initiated in the Falkland Islands and for advice on harvesting and utilisation.

BRAMLEYS AND SPENT MUSHROOM COMPOST

S. Mac An tSaoir and J. Mansfield

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For the last three growing seasons, spent mushroom compost (SMC) has been used successfully as a mulch (10cm deep) for weed control in a mature Bramley orchard. SMC contains many nutrients, so it has the potential to promote the quality of the crop. The yields of three plots (9 trees/plot), each receiving a different treatment, were recorded for three seasons:

Treatment	*Yield (kg/plot)*			*Fruit bud/m*
	Season			*Season*
	1996	*1997*	*1998*	*1998*
Full herbicide	638	877	341	8.0
No mulch or herb.	551	676	233	7.5
Mulch+reduced herb.	531	751	506	9.5

During the first two seasons the mulch reduced yield. However, during the 1998 season, when the blossom had been destroyed by frost, the mulch treatment produced a much higher yield than the other treatments. Preliminary investigations of this phenomenon suggest that the mulch-treated plots have a higher quality/number of fruit buds due to the slow release of nutrients during the latter half of the season when fruit bud formation occurs.

DEALING WITH RUST: THE GREATEST LIMITATION TO THE GROWTH OF SRC WILLOW

A. McCracken

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Rust of willow is caused by *Melampsora epitea* var. *epitea*. It was first detected on short rotation coppice willow in Northern Ireland in 1986. Its effect was serious, resulting in yield loss and death of stools. Within a few years, the then commonly grown variety *Salix burjatica* Korso could no longer be grown. It was shown that the rust could be controlled using fungicides,

but this approach was unacceptable for practical, environmental and financial reasons. Hence, in the late 1980s varietal mixtures were used as a disease control strategy. When planted in mixtures of four to six varieties, the onset of the rust was delayed by up to four weeks in some cases, the build-up of disease slowed and the final levels reduced. Current research is examining the effect of numbers of varieties (up to twenty) within a mixture as well as the effect of planting density and planting configuration on disease development. Of particular importance are the changes occurring within the rust population, with new pathotypes of *M. epitea* var. *epitea* frequently being detected. It is a concern that the use of varietal mixtures may encourage the development of super-races of the pathogen that could infect all of the varieties currently being used. However, data suggest that this is not the case. Varietal mixtures, incorporating new varieties, should contribute to disease management and yield sustainability.

INTEGRATION OF NON-LINKED GENES INTO THE *N. TABACUM* PLASTOME VIA PEG-MEDIATED PROTOPLAST COTRANSFORMATION

N. McGrath, P. Medgyesy, and P.J. Dix

Polyethylene glycol mediated co-transformation of chloroplasts in tobacco leaf pieces with an equal volume of two independently targeted genes was carried out. This was done to determine whether targeted insertion of the non-selectable passenger gene could be achieved without linkage to the selectable marker gene. Selection of transformants was on the basis of greening of protoplast-derived colonies on an antibiotic medium followed by screening for herbicide resistance. Analysis included RFLP, fluorescence and thermoluminescence measurements. Efficient co-transformation will make it possible to transform the plastome without the need for physical linkage of the targeted region and the selectable marker gene.

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MOLECULAR SYSTEMATICS OF THE RED ALGAL GENUS *POLYSIPHONIA* (GREVILLE)

L.M. McIvor, C.A. Maggs and M.J. Stanhope

Polysiphonia is the largest genus in the Rhodophyta with over two hundred described species, but its generic circumscription has been in an almost constant state of flux since its original proposal (as *Hutchinsia*) by C. Agardh in 1817. The species currently placed in *Polysiphonia* have traditionally been divided into two groups, based on the number of periaxial cells, with *Oligosiphonia* having four periaxial cells, and *Polysiphonia* five or more. Sequence data for the *rbcL* gene has been obtained for 30 samples, representing twenty species (including most species found in the British Isles) that exhibit a wide range of morphology. In addition, sequences have been obtained for other closely related genera, such as *Boergeseniella*, *Pterosiphonia*, *Lophosiphonia* and *Aphanocladia*. The genus *Polysiphonia* is paraphyletic, consisting of four strongly supported clades, with the type species *P. stricta* forming a distinct clade no more closely related to other species of *Polysiphonia* than to other genera such as *Pterosiphonia*. The genus *Boergeseniella* is in the same clade as multiaxial species of *Polysiphonia*. The position of *P. atlantica* is unresolved. Although the *Polysiphonia* group is monophyletic, *Oligosiphonia* is paraphyletic.

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OPTICAL PROPERTIES OF MARINE PHYTOPLANKTON

D. McKee, B.A. Osborne and G. Tarran

The optical properties of marine phytoplankton can be observed on a variety of scales. Mesoscale processes occurring in the oceans are monitored by satellite-borne ocean colour sensors, while individual particles can be classed using flow cytometry. In order to fully utilise the potential of these measurement platforms, it is essential that the pathway between individual particle and bulk optical properties is adequately understood. In this presentation we will assess the effect of size distribution and the presence of other types of particles on the optical properties of laboratory-grown cultures. Implications for flow cytometry operations, especially with regard to remote-sensing applications, will be discussed.

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ATLAS 2000 IN NORTHERN IRELAND: ORIGINS, HISTORY AND PROGRESS OF THE BSBI PLANT DISTRIBUTION MAPPING SCHEME

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The origins of distribution atlases in their modern form lie in schemes carried out by Scandinavian botanists in the 1950s, although the study of phytogeographical distributions in these islands can be traced back to the work of H.C. Watson in the 1830s. The most recent BSBI Atlas (published in 1962) is based on work carried out by members and others in the 1950s. The new scheme, launched in 1995, aims to produce a new Atlas in both printed and CD-Rom formats in 2001 and involves re-surveying the entire British Isles by the end of 1999. Existing historical information will also be incorporated. It is expected that the results will provide an up-to-date picture of distributions of British and Irish vascular plants and highlight changes in frequencies that may be valuable in terms of conservation strategies. Within Northern Ireland, the scheme is mainly funded by the Environment and Heritage Service of the Department of the Environment.

AN ANALYSIS OF THE EXPRESSION OF MADS-BOX GENES DURING ADVENTITIOUS ROOT FORMATION IN *MALUS DOMESTICA*

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The formation of adventitious roots in woody plants is poorly understood but is a vital aspect of micropropagation. Much research has been carried out on physiological aspects of adventitious roots. However, the molecular biology of root initiation is poorly characterised. Relatively few adventitious rooting related genes have been isolated e.g. *ARRO-1*, *RSI-1*, and *LRP-1*. However, one class of genes that is involved in root formation are MADS-box genes. This class of genes is traditionally associated with the development of floral organs and meristems of plants. Recently *Arabidopsis* MADS-box genes (*AGL-12*, *AGL-14* and *AGL-17*), whose expression is restricted to roots, have been identified. The *ANR-1* mutant, which showed altered root development, was identified as a MADS-box gene. The aim of this project is to characterise root-specific MADS-box genes in apple and to examine their role in adventitious root formation. RNA isolated from Jork 9 apple stem discs, which were initiated to form adventitious roots, was used in the construction of a cDNA library. Any MADS-box genes isolated from this library are potentially expressed during adventitious root formation. Primers have been designed using sequence alignments of apple MADS-box and rooting related *Arabadopsis* MADS-box genes. To date, 60 bp and 120 bp MADS-box fragments have been isolated and cloned from genomic apple DNA. These will be used as probes to screen the cDNA library. RT-PCR has been used to synthesise cDNA from RNA that has been isolated from the roots of germinating apple seeds. MADS-box specific fragments will be amplified from the cDNA and also used to screen the cDNA library.

WHEN IS AN OAK IN IRELAND NOT AN IRISH OAK?

J. Martin, G. Douglas, T. Hodkinson, C. Kelleher and D. Kelly

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It is desirable that Irish foresters plant appropriate Irish oak (*Quercus petraea* and *Quercus robur*) material for regeneration and reforestation, as this will help conserve Irish genetic diversity and utilise trees that are adapted to the Irish environment. However, it first has to be possible to distinguish Irish oak populations from non-native ones. This project is currently investigating Irish oak biodiversity by using genetic, morphological and ecological techniques. The genetic investigation is split into the analysis of nuclear DNA and chloroplast DNA (cpDNA). Chloroplast DNA is maternally inherited and relatively conserved in terms of sequence level variation (cpDNA changes very slowly with time). This means that the original provenance of a tree can usually be identified as the effects of hybridisation and introgression between different populations or species are removed. When placed in the context of a wider European study, the results of this investigation should also reveal the postglacial recolonisation routes of oak into Ireland.

A REVISION OF *UVARIA* L. (ANNONACEAE) IN INDIA AND INDOCHINA: A REPORT OF RECENT FIELDWORK IN SOUTHEAST ASIA

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Uvaria L. is a large genus of *c.* 150 species distributed from West Africa through Asia to New Caledonia and Fiji. It is the third largest genus in the primitive angiosperm family Annonaceae. All species are climbers and occur in lowland tropical rainforest or seasonally wet mixed tropical forest. This revision is focused on the *c.* 40 species that inhabit continental Asia. A recent field expedition to Thailand and Vietnam has provided new data on the ecology, growth, distribution and medicinal uses of Uvarias in these two countries, including the discovery of previously undescribed species. Fresh material was also collected in silica gel from 40 species of Annonaceae for DNA sequencing analysis at the Plant Molecular Systematics laboratory at Trinity College.

THE UPTAKE OF HEAVY METALS BY MARINE MACROALGAE: BIOREMEDIATION OF CONTAMINATED WATERS

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Contamination of natural waters by heavy metals is an issue under constant legislative review as new discoveries about the lethal and sublethal effects are made. Seaweeds have shown a high capacity for removing metals from the environment, even at low trace concentrations. Much research has focused on the use of processed seaweed biomasses or extracts for the remediation of contaminated waters. For process design this non-viable biomass approach has several advantages; it has, however, eclipsed much of the research on uptake kinetics, optimisation and even the toxicology of metals on viable algae. Preliminary experiments suggest that the brown seaweeds show great promise as agents for metals bioremediation. Tank experiments have shown rapid and efficient uptake kinetics, comparable with that of chelation resins, as suggested in the literature. Culture and analysis techniques are being developed to allow for the monitoring of this uptake and its effect on growth and health. The effects and capacities of a sustainable metals removal system will be investigated, as will its characterisation. Consideration of the reclamation of contaminants from the biomass must also be made.

EVALUATION OF REDUCED RATES OF FUNGICIDE USAGE ON CEREALS IN NORTHERN IRELAND

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Trials over three years on winter wheat, winter barley and spring barley indicated scope for reduction in rates of fungicide application but depended on the crop/disease combination. For example, good control of disease and improved yield were obtained with an eighth rate of fungicide applied to spring barley for mildew control, but the same rate for control of *Septoria tritici* proved largely ineffective. The use of disease-resistant cultivars also allowed for a greater reduction in the use of fungicide. This was particularly marked for spring barley.

POST-HARVEST TREATMENT OF APPLES: POTENTIAL FOR REDUCTION IN PESTICIDE USAGE

S.H. Mitchell and S. Mac An tSaoir

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After harvesting, apples may be stored for considerable periods of up to 10 months. Disease prevention is essential during this storage period and is generally achieved using post-harvest treatment e.g. dipping the apples in fungicides, such as Ridomil mbc 60 WP (Ciba), which contains carbendazim and metalaxyl. Experiments were performed to measure the effects of the temperature of the dip and the concentration of the fungicide on the keeping quality of the fruit and the presence of incurred residues. HPLC with UV detection was used to analyse apple flesh and peel before and after storage of treated and untreated samples. The apples were stored under controlled atmosphere conditions of 4.5°C, 5% CO_2 and 1% O_2. Preliminary results indicated that the fruit in the temperature range 12–180°C absorbed the greatest amount of fungicide. The results of these experiments show that the initial fungicide is present on the surface of the fruit. However, during storage the fungicide penetrates the flesh, with a significant reduction in the level of residues in the peel. There was no significant difference in the level of incurred residues in flesh or peel when the fungicide was used at half the recommended rate. In addition, there was no significant difference in scores for the indicators of keeping quality when half and full rate treatments were compared. This suggests that the amount of fungicide used to treat apples prior to storage could be significantly reduced without impairing the efficacy of the treatment. This is of importance to the apple and related fruit storage industry, as production and waste disposals costs could be reduced.

EFFECTS OF MYCORRHIZAL ASSOCIATIONS ON ROOT ELECTROLYTE LEAKAGE OF NURSERY-GROWN DOUGLAS FIR SEEDLINGS

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Root electrolyte leakage (REL) is recognised as an accurate test to determine seedling viability, by assessing the condition of the root system. This method determines the leakage of ions from root tissue by measuring conductivity. With damaged tissue, the ions leak at a faster rate, thus giving a higher REL value. To date, many studies have been reported on the relationship between REL and the planting quality and survival of tree seedlings. As yet, there have been no studies carried out to investigate how mycorrhizal associations may affect REL. The objective of this study was to investigate the effect of cold storage on the REL of three-year-old (2 + 1), nursery grown Douglas fir seedlings from the Coillte Nursery at Ballintemple, Co. Carlow. Prior to REL determination, the root systems of these seedlings were separated into mycorrhizal types. There were three mycorrhizal types colonising these seedlings, which were formed by *Mycelium radicis atrovirens*, *Tuber* sp. and *Amphinema byssoides*. The components, e.g. mantle thickness, of these mycorrhizas were measured. Preliminary baseline results on non-cold stored seedlings showed significant differences in REL between these mycorrhizal types. Results will be presented to elucidate the effects of mycorrhizal associations on the REL of Douglas fir seedlings subjected to cold storage.

INFLUENCE OF WATER AVAILABILITY ON WOOD DECAY BY *DONKIOPORIA EXPANSA*

C. Moore and H. Fuller

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Oak timbers in historic buildings are subject to decomposition by the white rot fungus *Donkioporia expansa*. The water requirements for wood decay by *Donkioporia* were investigated in a series of experiments, the first of which examined decay of medieval and modern oak at relative humidities (RH) in the range 75–100%. Pre-colonised wood blocks were exposed for sixteen weeks at 22°C in the variously humid atmospheres in rectangular jars. Significant weight losses occurred only in wood blocks incubated in 100% RH, but not for all isolates; mean percentage wood block moisture content (% MC) ranged from 51–60%. Mycelial growth did not occur on wood blocks incubated in atmospheres below 97% RH (*c.* 28% MC). The effect of direct

contact of wood blocks with water was investigated in an experiment in which oak blocks were placed on sterile moist sand. Fungal growth was extensive on all blocks after incubation for sixteen weeks at 22°C. Humidity within the chambers was high, and percentage MC of wood blocks ranged between 80 and 90. While weight losses, up to 6.2%, were significant and higher than those recorded in previous experiments, they do not reflect the known potential of *Donkioporia* to cause extensive decay of oak in buildings. The results presented indicate that oak wood in contact with water over prolonged periods of time is more likely to be decayed by *Donkioporia*.

COMPARATIVE ECOPHYSIOLOGY OF DIFFERENT POPULATIONS OF *RAPHANUS RAPHANISTRUM*

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Two subspecies of *Raphanus raphanistrum* occur in Ireland. *R. raphanistrum* ssp. *raphanistrum* occurs very rarely in some inland locations, whilst *R. raphanistrum* ssp. *maritimus* occurs in a number of coastal habitats. Preliminary work presented here has focused on comparing intra-subspecies differences, and future work will examine the possibility of inter-subspecies differences. The methodology used will involve greenhouse growth experiments to determine what characteristics associated with the different populations enable them to grow under different environmental conditions. The principal factors on which research will focus are salinity, temperature, pollinators and moisture availability. Preliminary work comparing seeds of two populations of subspecies *maritimus* has established that there can be differences in seed weight between populations, and this has been related to soil nitrogen and phosphorous status. We are now investigating whether this is due to phenotypic or genotypic factors by examining a range of plant traits in material derived from seed collected from two coastal sites.

THE ALGAL HERBARIUM OF THE ULSTER MUSEUM

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The algal herbarium of the Ulster Museum is over 200 years old and world-wide in geographical coverage. It contains over 11,000 specimens and is continually being extended by collection, donation, exchange and purchase. It provides voucher specimens for publications, both old and recent, as well as reference specimens that have been borrowed for research. All have now been catalogued and can therefore be accurately referenced in publications. The herbarium contains type material and specimens collected by some of the most famous phycologists of the past.

ASSESSING GENETIC DIVERSITY IN IRISH OAK (*QUERCUS* SPP) USING RIBOSOMAL DNA AND MICROSATELLITE LOCI

G. Muir, C. Schloetterer, C. Fleming and M. Stanhope

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The examination of genetic diversity in plant species is of increasing importance for conservation purposes, assessing population structure and addressing phylogenetic problems. Molecular techniques offer an effective means of delivering this information provided that appropriate plant genes are targeted. In a study of Irish and European oak species, rDNA and microsatellite genes were investigated to determine their applicability as a general tool for estimating genetic diversity in *Quercus*. A number of non-species specific rDNA haplotypes were found in oak species, suggesting that rDNA may not be the most suitable marker for population genetic studies in the *Quercus* species complex. Variation at microsatellite loci proved more effective in discriminating oak gene pools with both northern Irish and central Irish *Quercus* exhibiting differences from southern Irish populations, which appeared to have a greater genetic affinity with other European *Quercus* populations.

THE COMMERCIAL FEASIBILITY OF VEGETATIVE PROPAGATION OF ASH (*FRAXINUS EXCELSIOR*) VIA PHOTOAUTOTROPHIC MICROPROPAGATION (PAM)

N. Nakhshab, M.J. Hennerty and R. Long

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The possibility of the commercial feasibility of vegetative propagation of ash via photoautotrophic micropropagation was investigated. While *in vitro* multiplication of shoots from seedling was successful, multiplication of shoots from old trees proved to be difficult because of fungi and bacterial contamination. One possible means was to provide an alternative *in vitro* establishment procedure. Non-axenic cultures of ash cuttings in a carbon dioxide (CO_2) enriched environment (CDE) induced shoot regeneration. These new shoots (microcuttings) were successfully used for direct establishment of ash under CDE conditions. *In vitro* stool beds of ash microplants were established, and by renewing the medium weekly, shoot elongation of microplants reached a suitable length for repruning after three to four weeks. Different media were tested to identify optimal nutrient content for use in PAM system. Enshi, full DKW and half-strength DKW media were used successfully without resulting in noticeable differences in the shoot growth rate of microplants. The multiplication rate was one microcutting per plant after the first pruning; however, it increased to two to three microcuttings per microplant as a result of repeated hedging yielding more lateral shoots. The vigorous growth rate and regeneration capacity of these microplants, hedging microplant shoots at regular intervals and using the excised nodes as inocula for subsequent generation, have now provided a valuable alternative *in vitro* establishment procedure.

DETERMINATION OF THE SHORT-TERM EFFECT OF EXOGENOUSLY APPLIED COPPER ON SOIL RESPIRATION RATE: PRACTICAL PROBLEMS

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Most studies on the effect of copper (Cu) on soil respiration rate in the past have tended to focus on the short-term addition of copper sulphate ($CuSO_4$) to small soil samples maintained within the laboratory. Such laboratory-oriented studies have involved, in the main, significant soil pre-treatment, where sieving and removal of plant material from air-dried soils is common. Results presented here illustrate the danger in such pre-treatment and present an alternative laboratory assay for soil respiration where maintained, stable rates of carbon dioxide evolution are possible at constant soil water potential. Using this assay, results are presented relating the concentration of bio-available Cu to soil respiration.

DEVELOPMENT OF A NOVEL TISSUE CULTURE SYSTEM FOR THE ENHANCED PRODUCTION OF AN ANTIBIOTIC COMPOUND IN SELECTED RANUNCULACEAE SPECIES

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Members of the Ranunculaceae are well known to have vesicant properties due to the presence of an antibiotic substance, protoanemonin. Protoanemonin is found predominantly in the aerial parts of the plant, and a previous report has shown that cultured cells of petioles produce good yields of the antibiotic *in vitro*. It is therefore proposed that a number of different *in vitro* studies will be applied to further enhance the biosynthetic capacity of cultured *Anemone* and *Clematis* species. These studies will include genetic transformation of cultured shoots to produce shooty teratomas and examination of the possible biotransformation of a precursor compound to the antibiotic by cultured cells. Both of these studies may provide a greater knowledge of the natural biosynthetic pathways employed by Ranunculaceae in the production of protoanemonin. An *in vitro* bioassay on the antibiotic activity of protoanemonin will also be developed.

CHLOROPHYLL FLUORESCENCE AS A RAPID MEASURE OF COLD HARDINESS AND FREEZING TOLERANCE IN DOUGLAS FIR SEEDLINGS

M. Perks, B.A. Osborne and D. Mitchell

Chlorophyll fluorescence, as measured by pulse modulated or integrated fluorometry systems, can be used to detect water deficits, freezing and heat damage in conifer seedlings. We present preliminary results of an investigation into the potential application of fluorescence measurements for rapid detection of freezing injury and to determine the development of cold acclimation. A pulse modulated system was used, alongside a 'conventional' visual injury assessment scale, to predict cold hardiness of three-year-old Douglas fir seedlings after controlled freezing tests on two occasions in early 1999. Cold storage regimes (temperature and duration) have also been investigated, in conjunction with infra-red gas analysis, to elucidate the physiological status of stored nursery stock. Results will be presented in the context of nursery management practices, and conclusions will be drawn concerning the value of this technique (i) for rapidly (and non-destructively) assessing seedling vitality and (ii) as a supplement, or replacement, for traditional stock quality assessment methods.

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A STUDY OF CELL MORPHOGENESIS IN THE *ARABIDOPSIS* ROOT EPIDERMIS

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The *Arabidopsis* root epidermis consists of root hair cell files and non-root hair cell files, which form a distinct pattern, with hair files always overlying the cortical anticlinal cleft. This study concentrates on the morphogenesis of the root epidermis from the meristem through to mature, fully elongated root hair producing cells. It also includes the characterisation of the *kojak* (*kjk*) mutation. Roots with the *kjk* mutation fail to produce root hairs, resulting in a bald root. This study concentrates on the cytological events that occur prior to, during and after root hair formation in wild-type and *kjk* roots using a variety of microscopic techniques including LM, TEM, Cryo-SEM and CLSM. Epidermal cells in the meristem are small with a large nucleus and a dense cytoplasm. Cells specified to be root hair cells undergo more division than non-root hair cells, and vacuolation is also reduced, resulting in shorter, less vacuolated cells. The final step in differentiation is the production of a root hair. This process begins with a localised bulging at the apical end of the hair cell. From this bulge a tip-growing root hair emerges. In the *kjk* mutant there is an excessive initial bulge, and subsequently each hair cell bursts at the site of the bulge. This results in dead hair files, giving the root its bald appearance. This implies a role for the *kojak* gene in root hair initiation.

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TOWARDS THE PRODUCTION OF AN EDIBLE ORAL VACCINE USING PLANT BIOTECHNOLOGY

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Oral vaccines have considerable advantages over parenteral delivered vaccines, including ease of administration and improved safety. A novel, self-replenishing approach to the development of oral vaccines is the production of transgenic plants, which provide the antigen in an edible form. We are developing a model system for the production of a plant-based oral vaccine to influenza virus and for testing immunogenicity in mice. The influenza virus haemaglutinin (HA) gene is currently being cloned into an *Agrobacterium tumefaciens* binary vector to facilitate transfer to two crop plants, tomato (*Lycopersicon esculentum*) and oilseed rape (*Brassica napus*). Expression of the HA gene will be controlled by a high-level constitutive promoter, and plant tissues expressing the influenza HA antigen will be assessed for immunogenicity in a murine model system. The HA gene was cloned into the plasmid vector pET !21(d) for protein expression in *E. coli* to verify that the open reading frame is correctly translated and that the antigen can be immunologically detected in a heterologous host.

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PHOTOKARST IN HELL: THE ROLE OF ENDOLITHIC ALGAE IN THE DEVELOPMENT OF INTERTIDAL KARST

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Striking karst features, such as fretted pinnacles and intertidal notches, along limestone coasts have long been a topic of debate. Since seawater is completely saturated with respect to calcium carbonate, direct dissolution of limestone cannot occur. It has been proposed that locally the water may become undersaturated through mixing with rainwater or through the effects of biological respiration, but such arguments remain unconvincing. Observations in Hell, a partly drowned cave at Doolin, Co. Clare, reveal that endolithic algae are a major factor in the development of marine karst. In the darkest parts of this system, original solutional features, linked unequivocally to dissolution by flowing fresh water, remain in pristine condition, despite several thousand years' exposure to sea water. The limestone is deeply fretted in well lit parts of the cave, where the roof has collapsed, but this passes into a zone of small, light-orientated limestone pinnacles in more dimly lit regions. The poorly lit sides of the pinnacles appear colonised by endolithic cyanophytes, while the pits in between, facing directly into the light, appear to favour red algae. The much more reflective nature of the chalk in Northern Ireland allows the development of light-orientated tubes rather than the pinnacles that develop on the grey Carboniferous limestone at Doolin.

ATTITUDES OF IRISH CONSUMERS TO GENETICALLY MODIFIED FOODS

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A survey of the attitudes of Irish consumers to genetically modified foods was carried out in March 1999. The survey respondents comprised rural and urban samples of adults and secondary school students. The level of respondents' familiarity with the term 'genetic engineering' was assessed. Respondents' perceptions of the risks and benefits associated with these foods were also appraised. In addition, respondents were asked their opinions on fertilisers and organically produced foods, so that a comparison could be drawn between these technologies and genetic engineering. In the final section of the questionnaire, the likelihood of respondents purchasing various genetically modified foods was investigated. Respondents were also asked which sources of information on GMOs they thought were most reliable.

GROWTH AND TISSUE ION CONCENTRATIONS OF THREE *PLANTAGO MARITIMA* POPULATIONS GROWN IN VARYING SODIUM CHLORIDE CONCENTRATIONS

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Soil and water salinity are major limiting factors to crop production in arid and semi-arid regions of the world. With the global population expected to reach ten billion by the year 2050, increasing food demands will necessitate the use of more marginal lands for crop production. One response to the salinity problem is the development of salt-tolerant crops through breeding and genetic engineering. More recently, the focus has broadened to incorporate the domestication of halophytes to combat the salinity problem. It is beneficial to study the physiological reactions of such halo-tolerant plants to saline conditions in order to characterise salinity responses. *Plantago maritima*, a perennial herb, has a widespread distribution and is frequently found growing on seashores and salt marshes. Plants from two salt marsh populations and a sea-cliff population were grown in various sodium chloride (NaCl) concentrations. Growth rates and tissue ion concentrations were presented for each population, demonstrating that increasing salinity levels decreased growth in all populations, and that the pattern of ion accumulation differed between marsh and sea-cliff populations.

SORBITOL ACCUMULATION IN ROOTS AND SHOOTS OF A SALT MARSH POPULATION OF *PLANTAGO MARITIMA* IN RESPONSE TO EXTERNAL NACL CONCENTRATIONS

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Osmotic stress, generated either by drought or salinity, represents the most common severe environmental stress limiting plant growth and productivity. In response to such stress conditions, many plants accumulate compatible metabolites in the cytoplasm of their cells in an attempt to combat the water problem. These compounds are soluble, of low molecular weight and do not interfere with the biochemical reactions of the cells. Amino acids (e.g. proline), quaternary ammonium compounds (e.g. glycinebetaine) and sugar alcohols (e.g. sorbitol) all act as compatible metabolites. It has been suggested that *Plantago maritima* accumulates the sugar alcohol sorbitol in response to increased external sodium chloride (NaCl) concentrations. An experiment using a salt marsh population of *Plantago maritima* was established to investigate this relationship. The results are presented here. Long-term exposure of *Plantago maritima* to higher salt levels has resulted in the accumulation of sorbitol to concentrations considerably higher than those reported in the literature. Sorbitol content appears to be correlated with sodium chloride (NaCl) concentration but to a lesser extent than would be expected if this metabolite were the primary means of osmotic adjustment.

THE MEASUREMENT OF HEATHLAND PRODUCTIVITY AND UTILISATION BY GRAZING HERBIVORES IN THE EASTERN MOURNES ASSI

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There has been a reduction in the level of heather cover in the Eastern Mournes Area of Special Scientific Interest (ASSI) in recent years, and the Department of the Environment has instigated a studentship to investigate the problem of overgrazing. The project has three main aims: (1) to measure the biomass and productivity of various heathland types; (2) to measure the extent and patterns of utilisation of heather by grazing herbivores; and (3) to apply the Macaulay Land Use Research Institute (MLURI) Aberdeen Hill Grazing Management Model, which will calculate a sustainable grazing density for the study area. The Grazing Model works by calculating the production of each of the vegetation types over a two-year period. It then simulates utilisation by grazing herbivores over a similar period. A utilisation level above 40% on young heather is seen as detrimental. The results from the computer model are comparable field measurements. In our study there was a poor correlation between heather age and physical characteristics. Utilisation samples were taken before the growing season began and biomass samples were taken at the end of the growing season, from exclosure cages. Altitude had a significant impact on biomass and productivity. Biomass utilisation within the study area was found to be above the suppressive levels. The MLURI Model can be used to forecast the impacts of a change in the grazing regime, i.e. a decrease in sheep numbers should reduce the impacts on heather. It can also be used to assess the impact of a management programme on the study area.

THE GENETIC TRANSFORMATION OF *MALUS DOMESTICA* CULTIVAR GREENSLEEVES

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The economic importance of woody fruit species has led to their selection and breeding over thousands of years. Recent developments in biotechnology have provided an alternative approach to woody fruit tree improvement, through the introduction of genes of interest by genetic transformation. This allows us to bypass the long periods required for conventional breeding. *Agrobacterium tumefaciens*-mediated transformation protocols have been worked out for the apple cultivar Greensleeves. This work involved the introduction of two different plasmids into the apple cultivar Greensleeves. The genes of interest are uidA (GUS reporter) and nptII (kanamycin resistance) in the first plasmid, and pSAG12 ipt, nptII and uidA in the second. ipt is involved in cytokinin biosynthesis in plants; it has an established role in senescence. When constitutively

expressed in transgenic plants its overproduction causes a delay in senescence along with deleterious developmental and morphological changes. The pSAG12 ipt fusion has been introduced into *Nicotiana tabacum*, creating a system in which senescence is delayed without cytokinin overproduction. The aim of this work was to introduce such a system into the apple. The results presented describe the characterisation of transformants expressing the nptII and uidA genes. Preliminary results from the first lines transformed with the ipt construct were also presented.

WETLAND PLANT-ASSOCIATED EFFECTS ON METAL MOBILITY IN PB-ZN MINE TAILINGS

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Promoting vegetative cover on mine tailings ponds is one proposed remediation method of these sites, but the effect of plants on metal mobility in stagnant, waterlogged, Zn-Pb mine tailings is poorly understood and documented. Specialised lysimeter chambers (root zone containers) allowed examination of the effects of wetland plant presence on the sediment under these conditions. At specific locations relative to plant roots, sediment redox potential (Eh) was measured, and porewater samples (analysed for pH and metal concentrations) were collected repeatedly and nondisruptively over the course of a year. Statistical analysis (split-plot ANOVA) highlighted significant effects of plant presence within and between the experimental treatments. In comparison with the unplanted control, *Typha latifolia* caused decreased pH and increased soluble Zn to 1cm lateral radius and below the roots, while *Glyceria fluitans* did not significantly affect the sediment chemistry. These results form the basis of continuing research on the implications of wetland cover over stagnant mine tailings ponds and subsequent sediment heavy metal retention.