Mycological Research News

This issue of Mycological Research News features: Sudden Oak Death (Phytophthora ramorum) discovered on trees in Europe; and ‘Fusarium graminearum’ comprises nine separate species.

Eleven research papers appear in this part. Molecular studies reveal that the perithecioid Porinaceae are nested within ostropalean fungi; and that Gelasinospora species are nested with those of Neurospora, necessitating a broader concept of Neurospora. Also investigated are Armillaria species in the Czech Republic, and the status of Tricholoma bunionum. A new phytopathogenic Phytophthora is described with three subspecies causing diseases in Alnus; as are two new kickxcellaean fungi; and a new Streptopodium on papaya. Variation in and distributions of Duddingstonia flagrans and Eutypa leptoplaca genotypes are also reported.

Methods for study and assessing the occurrence of anaerobic chytridiaceous fungi in sheep and cow are presented, and used in a gut-long study of their occurrence in sheep.


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IN THIS ISSUE

It is now generally recognized that traditional classifications of the ascomycetes into main groups based on ascorna types is unsound. New sequence data on a range of representatives now reveals that the perithecioid Porinaceae, a family over whose position there has been considerable uncertainty, is nested within the apostecioid ostropalean fungi; this is apparently due to a neotenic ontogeny, the ascomata ceasing development and maturing at a much earlier phase (pp. 1111–1118).

A radical new interpretation of the circumscription of Neurospora follows the discovery that species traditionally included in the genus are intermixed with those of Gelasinospora in phylogenetic trees; the genera were formerly separated by ascospore ornamentation types which in reality intergrade, and the genera are synonymized here, with 49 species being recognized and keyed in Neurospora (pp. 1119–1142). Molecular characterization of isolates of six species of Armillaria in the Czech Republic using three restriction endonucleases and PCR-RFLP on the ITS discriminated between the species and clustered them into three groups (pp. 1153–1161). Studies on the ITS region of of Tricholoma sulphureum and T. bunionum, together with examinations of the mycorrhizas formed and other factors, suggest that the two taxa should be synonymized (pp. 1162–1171).

Several novel fungi are recognized here. Most important is the formal naming of the hybrid Phytophthora and three variant subspecies responsible for the death of alder trees (pp. 1172–1184). In addition a new Streptopodium is described on papaya from Brazil, with a treatment and key to other powdery mildews on the same host plants (pp. 1185–1194). The

1 Mycological Research News is compiled by David L. Hawksworth, Executive Editor Mycological Research, The Yellow House, Calle Aguila 12, Colonia La Maliciosa, Mataelpino, ES-28492 Madrid, Spain (tel/fax: [+34] 91 857 3640; e-mail: myconova @terra.es), to whom suggestions for inclusion and items for consideration should be sent. Unsigned items are by the Executive Editor.
occurrence and host range of _Eutypa leptoplaca_, a pathogen of grapevine in the USA, has been investigated molecularly and epidemiologically (pp. 1195–1204). _Duddingtonia flagrans_, a fungus with potential in nematode biocontrol, appears to be clonal with little molecular variation across wide geographic areas and is posited to have arisen around 16–23,000 yr ago. The zygomycete order _Kickxellales_ has few specialists, and a new genus and two new species are reported here from Japan (pp. 1143–1152).

The last two papers in this number concern anaerobic rumen fungi. The first is methodological and describes techniques and behaviour of these fascinating fungi both in culture and in the rumens of sheep and a cow (pp. 1215–1226), while the second reports on their occurrence along the entire digestive tract if sheep using SEM and chitin assays and suggests that these fungi constitute up to a remarkable 20% of the total microbial biomass in the rumen (pp. 1227–1233).

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**SUDDEN OAK DEATH (PHYTOPHTHORA RAMORUM) DISCOVERED ON TREES IN EUROPE**

_Physophtora ramorum_ is the cause of rapid mortality, _via_ stem bleeding cankers, of a range of native oaks (_Quercus_ and _Lithocarpus_ spp.) in California and Oregon, USA. It also causes leaf blights or dieback of many canopy trees and understory shrubs and ornamental nursery stock such as rhododendron (e.g. Goheen _et al_. 2002, Davidson _et al_. 2003). In Europe, it has been spreading on rhododendrons and other shrubs, in nurseries and in woodland gardens (e.g. Lane _et al_. 2003, Werres _et al_. 2003). Surveys show that _P. ramorum_ has been introduced into at least eight European countries by movement of stock. Many European native and non-native trees, including some plantation species, are potentially susceptible (Brasier _et al_. 2002). Since European and American populations of _P. ramorum_ exhibit behavioural and molecular differences (e.g. Brasier 2003, Hansen _et al_. 2003, Ivors _et al_. 2004), the pathogen is thought to have been separately introduced into each continent. Recently, the European type has appeared in nurseries in the Pacific north-west (Hansen _et al_. 2003).

Since the last report in this column (Mycological Research 107 (3): 258–259, March 2003) the pathogen has been discovered on trees in Europe. In October–November 2003, it was isolated from stem bleeding cankers on two American northern red oaks, _Quercus rubra_, in woodlands in The Netherlands (Hans de Gruyter, pers. comm.); and from a bleeding canker girdling an American southern red oak, _Q. falcata_, in Sussex, southern Britain (Brasier _et al_. 2004a). Subsequently, the fungus has been isolated from stem bleeding cankers on several European beech (_Fagus sylvatica_), Turkey oak (_Q. cerris_), southern beech (_Nothofagus obliqua_), and a horse chestnut (_Aesculus hippocastanum_); and from leaf and shoot symptoms on _Q. ilex_ (holm oak), _Castanea sativa_ (sweet chestnut) and _Drymis winteri_, at three wooded or semi-wooded sites in Cornwall, south-west Britain (Forest Research and Central Science Laboratories, unpubl.); around 20 infected trees have been found in Cornwall to date. All the UK sites have infected rhododendrons, and at two of the Cornish sites these are heavily infected.

In November 2003 there was a further development. A second new _Physophtora_, informally called _Physophtora taxon C_, was discovered causing bleeding cankers on beech at the two most affected Cornish sites (Brasier _et al_. 2004b). Like _P. ramorum_, _P. taxon C_ is spreading on rhododendrons and is also aerial or splash-dispersed _via_ caducous sporangia. As with _P. ramorum_, the rhododendrons probably act as a platform from which the pathogen infects the trees. Surveys of rhododendrons indicate _P. taxon C_ may be confined to the area around the two main Cornish sites, and is therefore a relatively recent introduction. It has not been found elsewhere in Europe. So far it has been obtained from bleeding cankers on 13 beech trees, sometimes causing extensive bark necrosis from near ground level to 11 m; and from a severely bark-infected _Liriodendron tulipifera_ (tulip tree, _Magnoliaceae_; Forest Research, unpubl.). It has also been isolated from necrotic foliage of _Magnolia_ and from _Gevuinea avel-lana_ (Chilean hazelnut, _Proteaceae_) (CSL and Forest Research, unpubl.). Its potential host range on UK trees and shrubs has yet to be determined.

Limited tests indicate that neither _P. ramorum_ nor _P. taxon C_ produce significant numbers of sporangia on intact outer bark surfaces of infected mature trees, in contrast to their behaviour on rhododendron (Forest Research, unpubl.). A key to their eradication, therefore, is the removal of affected rhododendrons or other foliar hosts on which they sporulate freely. The longer these remain the more trees that may become infected. Attempts are therefore underway to eradicate both _P. ramorum_ and _P. taxon C_ from affected sites in the UK. Eradication of _P. ramorum_ across Europe as a whole is also highly desirable. In both instances, eradication would prevent further spread of these pathogens from Europe to hosts or ecosystems that might be at risk in other, climatically suitable parts of the world. The current records suggest that Southern Hemisphere ecosystems with _Fagaceae_ and _Proteaceae_ are a possible example; as well as some of Europe’s special ecosystems, such as the Canary Island cloud forests or the _Q. ilex_ maquis woodlands of Iberia.
Another reason to contain their spread is to prevent hybridisation with other Phytophthora pathogens. In Cornwall: (1) P. taxon C and P. ramorum are sometimes obtained from adjacent lesions on the same beech tree; (2) another aerial Phytophthora, P. ilicis, is also very active at these sites, causing bleeding cankers and foliar necrosis of native and introduced holly (Ilex spp., Aquifoliales); and (3) well-known, established soil-borne Phytophthora tree pathogens such as P. cambivora are already present on these sites (Forest Research, unpubl.). Both invasive and established Phytophthora pathogens are therefore coming into close proximity, sometimes occupying the same niche. This raises the possibility of interspecific hybridisation, a process that can give rise to entirely new organisms, or to genetically modified pathogens exhibiting new host ranges. A good example is the swarm of hybrid Phytophthora’s now killing alder trees across Europe (Brasier, Cooke & Duncan 1999, Brasier et al. 2004c).

An obvious assumption is that P. ramorum, P. taxon C, and also P. ilicis, have been introduced into Europe and America as unintended byproducts of the international plant trade. However, clues to their geographical origins remain limited and circumstantial. From their biology and temperature-growth relationships, these taxa appear adapted to aerial dispersal in temperate or Mediterranean-type climates. P. ramorum exhibits a wide and ever growing host range in America and Europe, especially among Fagaceae and Ericaceae. Its closest known phylogenetic relative is P. lateralis (Martin & Tooley 2003, Ivors et al. 2004), an invasive pathogen now killing Chamaecyparis lawsoniana (Cupressaceae) in its native range in southern Oregon (Hansen 2003), but believed to come from Asia (Everett M. Hansen, pers. comm.). So far, the hosts of P. taxon C in the UK are confined to Fagaceae, Magnoliaceae, Proteaceae and Ericaceae. Its nearest phylogenetic relative, on ITS sequence, is P. boehmeriae (Brasier, David L. Cooke & Kelvin Hughes, unpubl.), a species recorded mostly on trees and cotton in China and Australia (Erwin & Ribeiro 1996). P. ilicis appears confined to Ilex spp. and its European distribution is apparently limited to the UK, where it has probably spread across the country over the past 20–30 years. P. ilicis is also present in Canada and Oregon USA, where it occurs on imported ‘English holly’ in nurseries (Erwin & Ribeiro 1996). This restricted distribution suggests P. ilicis has been introduced into all three countries. The nearest known relative of P. ilicis is P. psycrophila, a very rare taxon obtained from soil around oak trees in Germany and Poland (Jung et al. 2002).

It seems likely that all three aerial species come from forested areas in Asia where, being coevolved with their native hosts, they may be relatively benign. In the case of P. ramorum and P. taxon C, the native host need not necessarily be rhododendron. In the US and European nursery trades rhododendrons tend to be susceptible to many Phytophthora’s they encounter (cfr Erwin & Ribeiro 1996) and the genus is therefore something of a ‘universal suspect’. Indeed, rhododendron leaves make good Phytophthora baits. Rhododendrons may therefore tend to act as a carrier of these pathogens within the trade. If P. ramorum and P. taxon C do have native tree hosts other than rhododendron, they may tend to cause only leaf and twig blights in the canopy, rather than the massive stem bleeding cankers they can cause on susceptible Quercus, Lithocarpus or Fagus spp. in America and Europe. Bark of the latter species might be particularly susceptible because they possess similar constitutive defence mechanisms to those present in the pathogen’s native hosts, such as specialised polyphenols, that the pathogen can to tolerate or degrade, but lack additional co-evolved defence mechanisms.

In the search for the geographic origins of P. ramorum, P. taxon C and P. ilicis (or indeed searches for other new Phytophthora’s that could potentially escape and damage the world’s forests in future) likely starting points include Yunnan, Taiwan, and the eastern Himalayas. Yunnan, in particular, has a very wide range of forest vegetation types and climatic regimes. It also has a wide range of unique Fagaceae, Ericaceae, Magnoliaceae, Cupressaceae and Aquifoliales, including numerous species of Rhododendron. For these same reasons, Yunnan has long been a popular destination for plant collectors. Their activity may well have introduced more than interesting plant species.

**Fusarium graminearum** comprises nine separate species

Molecular and incompatibility studies are increasing revealing the existence of cryptic species within currently recognized single species. Where plant pathogens are involved, it is essential to unravel the biological situation for effective plant resistance breeding and control measures. 

‘Fusarium graminearum’ (teleomorph Gibberella zeae) is the causal agent of the devastating Fusarium head blight disease of wheat and barley, and also a producer of mycotoxic trichothecenes and estrogenic compounds posing threats to human and animal health and food safety. This single ‘species’ has been known to be a complex for some years (O’Donnell et al. 2000), but additional data has now led to the recognition of nine species within ‘F. graminearum’ by O’Donnell et al. (2004). These researchers obtained sequences from portions of 11 nuclear genes, including the mating-type locus, and found agreement between the mating-type loci and seven other genes. The nine cryptic species all have contiguous **MAT1-1** and **MAT1-2** idiomorphs consistent with a homothallic mode of reproduction, but only one of these was found in five other putatively sexual or heterothallic species. The **MAT** genes are evidently under strong selective pressure, and the phylogenetic trees support a monophyletic origin of homothallism in the *F. graminearum* clade.

Eight species in the complex are formally described as new: *F. acaciae-mearnesii*, *F. asiaticum*, *F. australo-americanum*, *F. boothii*, *F. brasilicum*, *F. cortaderiae*, *F. meridionale*, and *F. mesoamericanum*. While the new species are primarily characterized by molecular differences, there are also morphological correlations with conidial characters; these include width, curvature, symmetry, the apical beak, and location of the widest point, and can be used to separate three of the new species and three species pairs. The new species are known from different geographical regions and hosts, especially in Africa, Asia, Australasia, and Central and South America, but the endemic areas are not always clear from the strains so far studied. The name *F. graminearum s. str.*, however, is wisely retained for the most cosmopolitan species of the complex, although the critical issue of the typification of that name is not discussed here.

Plant pathologists and those concerned with safety aspects of ‘*F. graminearum*’ will now need to examine the strains they work with more critically in order to identify the species they are dealing with, and caution will be needed in interpreting the earlier literature – especially where voucher cultures are not available for study.

In combining comprehensive molecular studies with critical morphological analyses, and examining strains from a wide range of localities and hosts, this study provides a model to be emulated in tackling parallel problems in other economically important fungi.