The Natural News

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Tips and tickles from a u.k. macrofungi course ~ Robin Garnett Heathland plants and their fungal associations ~ Sarah Lloyd Wildlife at Westbury Reserve ~ Sarah Lloyd

This well decorated spider's web is suspended between the leaves of a rush, *Juncus* sp. at Black Sugarloaf, Birralee. Photo S. Lloyd

Tips and tickles from a U.K. Macrofungi course *Robin Garnett Photos by Phil Collier*

Before Covid-19 lockdown restricted our movements, Phil and I took part in a Field Study Centre (FSC) fungi course based at FSC Preston Montford in Shropshire, near the border of Wales and England. It was led by outstanding British mycologist Geoffrey Kibby, who has nearly finished his magnum opus - a three volume set of books on "*Mushrooms and Toadstools of Britain & Europe*". He is describing and illustrating over 2,200 species: Volumes 1 and 2 are in print and beautifully produced; Volume 3 is forthcoming.

Our group of thirteen people spent the mornings going to damp, mushroomy places under Geoffrey's guidance to see what we could find; the afternoons in the lab looking through microscopes and consulting reference books to try to put names to our collections; and the evenings listening to lectures from Geoffrey. Geoffrey passed on lots of good advice from his many years as a mycologist and I would like to pass some of this on in the hope that it is new and useful.

Making collections

In the field, we noted down the trees associated with each fungus, as often there are distinct associations. For example, there is a Brown Birch bolete, *Leccinum scabrum*, and an Oak milkcap, *Lactarius quietus*. Some people used their cameras to record nearby trees. Others put leaves of nearby trees with their fungi collections. We also had many discussions about the smells of freshly picked fungi to help with identification. Radish was a commonly recognised smell. *Lactarius quietus* is said to have "a sweet oily smell like bedbugs". I have made a mental note to be more careful about recording tree associations and



Lycoperdon exculpiforme, a type of puffball fungus.



Mycena rosea

fungi smells when collecting in Tasmania. Our fellow course participants tasted and spat out tiny samples of the gills of *Russula* or the milky exudate of *Lactarius*, which helps identification. We, with our Aussie concerns about poisoning, were reluctant to taste. The one



Common stinkhorn Phallus impudicus

Lactarius I tasted, *Lactarius turpis*, nearly burnt my tongue off! We weren't terribly keen on the "Kiss Test" for viscosity either. In that test, one holds the cap of a fungus against one's lips to judge how sticky it is.

Most people carried plastic boxes with many compartments with damp paper towel or moss in each section to keep their collections moist.

Spore prints

Back in the lab, we laid out the caps of our collections to make spore prints on coverslips rather than glass slides and covered them with cups to retain moisture. This meant that when we inverted the coverslip onto a glass slide with a drop of Melzer's stain, the spores were closer to our eyes and sharper for photography than spores collected directly onto a slide.

We left the spore print for 15 - 20 minutes rather than several hours, which usually gave us enough (but not too many) spores to see under the microscope. If spores were too clumped together to see clearly, a drop



Above: Stinkhorn fungi like this Common stinkhorn *Phallus impudicus* first appear as an 'egg' and have a phalloid shape when fully emerged. Stinkhorn or phalloid fungi is a group of fungi whose members have a strong, fly-attracting stench. Flies land on the spore mass and spread the spores that adhere to their feet to other areas. *impudicus* is derived from the Latin for "shameless" "immodest".

of detergent would separate them. Geoffrey suggested that we make a colour reference collection of spores for species such as Russulas by scraping a dense pile of spores together on a glass slide, covering them with a coverslip and then sealing the edges with nail polish or sticky tape. He also showed us an effective way to "tickle" ascomycetes to stimulate them to release their spores: hold a coverslip just above a mature ascomycete and gently tickle the spore-bearing surface with a small paintbrush. The spores will puff out onto the cover slip. Then mount the spores on a slide stained with Cotton blue in lactophenol. Warm the slide gently with a cigarette lighter or by placing it on a warm light bulb in order to make the stain penetrate the spores. Now they are ready for viewing.

Measuring spores

We learnt that it was important to measure spores from a spore print rather than from a gill edge as dropped spores are mature whereas the gill edge often includes larger, immature spores. Once we had focused spores at 1000x magnification, in order to select twenty spores randomly to measure their lengths and widths, we twitched the position of the microscope stage and measured the spore closest to the cross hairs of the graticule. We only measured single spores that were obviously side-on with an apiculus visible. We also noted any spore ornamentation such as spines, ridges or lumps. These show up particularly well in Melzer's Reagent.

To see whether the spores were formed in clusters of two or four on each basidium, we looked at the side of a dry gill under a dissecting microscope. The four-spored fungi had obvious patterns of four dots like the end of a domino whereas the two-spored gill edge had spores that appeared to be placed randomly.

Looking at tissue under the microscope

To search for identification clues, we looked at a tiny fragment of the gill edge under the microscope. A useful tip was to take the sample from an inner half-gill rather than a large main gill as it is more likely to be protected from damage, particularly in Mycena species. We put a drop of stain next to the gill sample and dragged the stain across to the tissue using a pin rather than blasting a drop straight onto the delicate tissue. And after putting on a coverslip, we put gentle pressure on the tissue by pressing and twisting with the rubber end of a pencil rather than tapping which tends to smash up the preparation and drive cells further from their original positions. The size and shape of cystidia were often vital identification features. We were amazed at the variety of cystidia shapes: fingers or skittles, antennae or clubs. We also looked at the cells on the cuticle of the cap. With a razorblade we cut three sides of a tiny square on the top of the cap and then peeled off the square using



Ramariopsis tenuiramosa a coral fungus

forceps. The thinnest area for microscopy was usually the uncut, torn edge. After mounting and staining the peel, we looked to see if the cuticle cells were like spaghetti or balloons. The cuticle peel was also a good place to look for clamp connections. The orientation and length of the cells in the middle of the gills of Hygrocybe species is important for identification. We practised lying a hygrocybe gill on a glass slide and using another glass slide as our cutting edge to make very thin slices with a sharp razor blade. It was quite hard to make a clean slice when the gill was moist and easier when it had dried out a bit.

Stains

A useful summary of stains by Robin Dean can be found at

http://fungus.org.uk/nwfg/chemdec99.htm As well as those listed we used Patent Blue as a stain and found it effective in staining fungi cells such as cystidia and gill issue. It is one of the components that people use to reveal dental plaque. Dental plaque disclosing liquid or tablets can themselves be used as a fungi stain. It is safer to use than Congo Red, which



Illustration of fungus showing basidium and cystidium on the edge of a gill. Drawing: Robin Garnett

is categorised as a carcinogen. A tip to reveal whether cells had intra cellular granules or an encrustation around the outside was to mount the tissues in a concentrated sugar solution (without staining).

Some of our team-mates carried their bottles of stains in padded boxes designed to hold aromatherapy bottles. Others thought that this was such a neat idea that they ordered boxes online for themselves.

Are there clamp connections?

It is harder to be certain that clamp connections between hyphae are NOT present than to say that they are. Where should we look for them and how long should one go on looking? Geoffrey said that we are more likely to find clamp connections, if they are present, in rapidly growing tissue such as the mycelium at the base of the fungus.

Drying fungus for long-term collection

DNA analysis is becoming increasingly important in establishing the identification of fungi and the relationships between different species. (Thankfully there are no fungi hybrids apart from perhaps one *Coprinus*.) We heard that it is best to dry fungi for a long time (e.g. 12 hours or more) at 40 degrees C than to try to dry them quickly at a higher temperature because high temperatures can destroy the DNA.

Drawing pictures of macrofungi

In past years I have always drawn pictures of fungi using pencil and coloured crayons so I was fascinated to watch Geoffrey Kibby doing beautiful drawings on his iPad. He uses an inexpensive software package developed in Tasmania called "Procreate", which was launched in 2011 and by 2018 was the overall best-selling iPad app in the world. Learning to draw fungi with Procreate is certainly a challenge that I want to take up in the future – and maybe you do too. Good luck with your fungi forays. I hope these notes have added some new skills to your repertoire.

Heathland plants and their fungal associations Text and photographs by Sarah Lloyd

Heathlands are characterised by low growing impenetrable tangle of shrubs, sedges, rushes and herbs that are especially beautiful in early spring when their colourful flowers attract numerous insects and birds. Australian heaths have a rich array of species that rivals the diversity of tropical rainforests. The Kwongan sandplains of Western Australia (WA), for example, support 5710 species of which nearly 80% are endemic to that state. (Kwongan is an Aboriginal term now widely used for the low heath-like vegetation common over a wide area in the south-west.)

I wrote this before I visited WA, so it was fascinating to see the plants flourishing in sandy soils that are considered to be so deficient in nutrients as to be "effectively lethal for domestic plants". In many cases, it's an association with fungi that enables their survival.

When plants first colonised the land some 600 million years ago they had two options: they could develop an extensive, fine root system of their own, or they could enter into a relationship with fungi and thereby increase their ability to obtain nutrients and water from soil via the fungal hyphae—the microscopic thread-like structures that are the living component of most fungi.

Some plants, including members of the saltbush family (Chenopodiaceae), cabbage family (Brassicaceae), sedges (Cyperaceae and Restionaceae) and rushes (Juncaceae) adopted the first strategy and rarely form mycorrhizal associations. However, the vast majority—perhaps 95%—of plants, including ferns, mosses, lycopods and most families of vascular plants opted for symbiotic (i.e. mutually beneficial) partnerships with fungi; relationships that are especially important in nutrient deficient soils. There are several different types of mycorrhizal associations:

Ectomycorrhizae

(Also known as ectotrophic mycorrhiza) occur in about 3% of plant species and are common in conifers, eucalypts, and deciduous trees such as beech, oak and birch. In ectomycorrhizae the fungus does not penetrate the hosts' cells but forms both a filamentous sheath that envelopes the root and a net (known as the hartig net) consisting of tightly packed hyphae between the outer 3 - 4 cell layers of the root. The fungi involved are occasionally ascomycetes but are usually basidiomycetes that produce the frequently seen fruit bodies such as mushrooms, puffballs, coral fungi and truffles from genera such as Amanita, Russula, Cortinarius, Tricholoma and Boletus.

Endomycorrhizae

(Also known as endotrophic mycorrhiza) occur in approximately 80-85% of plant species. The fungal hyphae penetrate into the root cells rather than forming an external sheath of mycelium. Unlike ectomycorrhizae, they generally do not produce large fruit bodies. There are three types:

1. **Arbuscular endomycorrhizae** (also called vesicular-arbuscular mycorrhizae or VAM) are the most common and widespread. They occur in natural environments such as tropical rainforests, alpine meadows and deserts and in agricultural systems including crops of cereals, grasses, legumes, citrus, coffee, cotton, oil palms, rubber, sunflower and tea. They are obligate parasites - i.e. they do not survive for long in the absence of their hosts. Arbuscular mycorrhizas penetrate the cells in the outer layer (epidermis) and cortex of the root and produce highly branched structures called ar-



Ectomycorrhizae are common in conifers, eucalypts, and deciduous trees such as beech, oak and birch.



Orchid endomycorrhizae are formed in all members of the orchid family such as *Cyanicula deformis*.



White-cheeked Honeyeater in one of the many *Banksia* species that thrive in the sandy soils.

buscles (derived from the Latin word for "little bush"). There is an extensive interface between the arbuscles and the hosts' cell membranes, which enables water and other substances to be transferred between plant and fungus.

2. Orchid endomycorrhizae are formed in all members of the orchid family and in their natural habitats orchids cannot grow without their fungal partners. The fungus forms coiled structures in the cells of the root cortex and transfers carbon and other nutrients to the orchid. It gets nutrients by either breaking down organic matter in the soil, or by become mycorrhizal on other plants. The underground orchid of WA, for example, is linked via fungal hyphae to a *Melaleuca* species.



Astroloma microdonta and other members of the Epacridaceae family have ericoid endomycorrhizae.

3. Ericoid endomycorrhizae are a group of fungi associated with members of the Ericaceae family of the Northern Hemisphere and their closely related southern counterparts in the Epacridaceae family. Plants of this family are common in the nutrient deficient soils of coastal heaths and alpine bog ecosystems and include such familiar species as cranberry heath *Astroloma* spp. common heath *Epacris* spp., swamp heath *Sprengelia* spp., and beardheaths *Leucopogon* spp. The fungus forms snake-like hyphal coils within the epidermal cells of the hair roots resulting in most of the cell volume being occupied by the fungus.

It is through these various mechanisms that an exchange of nutrients takes place between



Members of the Proteaceae family have proteoid roots that consist of hundreds of densely packed extremely hairy rootlets that grow off the main roots. Left: *Isopogon* sp. Right *Banksia ilicifolia*



Leguminous plants including the pea family and wattles have mycorrhizal associations as well as root nodules (see caption p. 9). Wattles ranging from tall trees to prostrate plants are common throughout Australia.

the organisms. Fungi, which are unable to photosynthesize, gain carbon compounds and probably also amino acids, vitamins and other nutrients from their host. The plant also provides the fungus with a habitat that is relatively free from other soil micro-organisms.

The plant benefits in several ways: the fine microscopic fungal hyphae that grow out from the infected plant can penetrate extremely small spaces, effectively extending its root zone. Thus the fungus supplies the plant with water and soil nutrients, particularly phosphorus and nitrogen.

Plants have evolved various other strategies

to ensure their survival in harsh conditions:

Members of the Proteaceae family including *Banksia* and *Isopogon* spp. have proteoid roots that consist of hundreds of densely packed extremely hairy rootlets that grow off the main roots. They perform a similar function to mycorrhizal fungi in extending the root zone and increasing nutrient and water uptake.

Leguminous plants including the pea family (Fabaceae) and wattles (Mimosaceae) are especially prevalent in heaths. As well as having mycorrhizal associations, they have root nodules with special bacteria that are able to fix atmospheric nitrogen and make it available



Peas and wattles have root nodules with special bacteria that are able to fix atmospheric nitrogen and make it available to the plant. Left: *Gompholobium* sp. Right: *Bossiaea spinescens*



Insectivorous herbs such as sundews (*Drosera* spp.) have sticky tentacles that attract, capture and absorb small insects using digestive enzymes secreted from the glands to dissolve and absorb compounds.

to the plant.

Insectivorous herbs such as sundews (*Drosera* spp.) abound on nutrient poor soils. Their sticky tentacles attract, capture and absorb small insects using digestive enzymes secreted from the glands to dissolve and absorb nitrogenous compounds. Similarly, fairy's aprons or bladderworts (*Utricularia* spp.), small herbs of wet places, capture tiny insects in intricate traps or bladders that resemble minute bubbles on threadlike segments of their leaves that lie at or below the soil surface.

Many heathland plants also have aboveground characteristics that enable them to withstand hot and dry conditions, namely sclerophyllous leaves; evergreen leaves that are small, hard, thick and leathery. However, their most important adaptations are hidden from view in that marvelous subterranean world that most of us seldom think about.

Wildlife at Westbury Reserve *Sarah Lloyd*

Many animals—like many people—have favourite places to hang out. For instance, every time I visit Westbury Reserve on warm sunny days, I see two or three Tasmanian Tree Skinks (*Carinascincus pretiosus*) on a log near the entrance.

On the rocky slope above Brushy Rivulet, there's a family of White's skinks (*Liopholus whitii*), fast moving reptiles that dart to their burrows whenever they sense danger. I detected such a movement during a visit in early January and when I returned several days later, a skink was resting in exactly the same place and obligingly allowed close observation.

White's skink is one of our most recognisable reptiles. It is a medium-sized lizard boldly patterned with stripes and spots along the length of its stocky body. It lives for about 8-9 years, but it grows slowly and doesn't attain its full snout-vent length of 80 mm until it's about four years old.

White's skinks are viviparous, i.e. they give birth to live young rather than laying eggs like some other reptiles. Their dual-entrance burrows (for ventilation and escape) are large enough to accommodate the family group comprising a pair of adults plus 3 or 4 young. The latest offspring may stay close to their parents for up to a year.

White's skinks feed on spiders, millipedes, ants and other insects and occasionally plant material. Interestingly, they are unusual among Tasmanian lizards in regularly defecating at the same place.

The insects have been fascinating to observe. One of my visits coincided with masses of cranefly *Leptotarsus macquarianus* and I watched a succession of butterflies with bright copper *Paralucia aurifera* in early spring, followed by huge numbers of common brown *Heteronympha merope*, first the males and then the females; then Klug's zenixa *Geitoneura klugii* and shouldered brown *H. penelope*.

During a recent visit to the northern end to continue documenting the huge-hollow-bearing eucalypts scattered throughout the reserve, I observed numerous unmistakable platelets caused by Painted Button-quail when they forage. The 'circular scrapes', are formed when the bird stands on one leg and pirouettes to reveal seeds and invertebrates under the leaf litter.



White's skink (Liopholus whitii)



Kangaroo grass Themeda triandra



Cranefly Leptotarsus (Macromastix)



White-spotted swift spider Nyssus albopunctatus

Sugar Gliders in Tasmania

The has been much discussion about whether the sugar glider (*Petaurus breviceps*) is a long term resident of Tasmania or a recent introduction. Research in 2018 using molecular and historical data has settled this debate.

The first account of sugar gliders being introduced as pets into Launceston occurred in 1835. Records of sightings and carcasses around Launceston were presented to a meeting of the Tasmanian Society in 1845. In 1863 there was a statement of introduction by ornithologist John Gould.

The first museum record in Launceston was in 1846 and the first museum record near



Slime mould Cribraria sp.



Ovipositing Braconid wasp

Hobart was in 1883. There was an exponential growth in records from 1960.

Analysis of divergence between Tasmanian individuals and those in southern Australia suggest a recent introduction into Tasmania from southern Australia.

Sadly, the tree with the impressive sugar gliders' nest featured in TNN #77 was flattened during recent logging.

Ref: Campbell CD, Sarre SD, Stojanovic D, et al. When is a native species invasive? Incursion of a novel predatory marsupial using moleular and historical data. Divser Distrib 2018; 24:831-840

Walks and other events

Bring food, water, clothes for all weather, hand lens, binoculars, note book and curiosity

Monday 26 April Brushy Rivulet (aka Westbury)Reserve—A joint Birdlife Tasmania and CNFN bird walk. Meet at 8 am at Egmont Reserve, 4 km north of Westbury, and we will travel from there. Please let Sarah Lloyd know by email or phone if you are planning to attend. Ph 6396 1380

Sunday 2 May—Fungi at Dip Falls. Meet at 10 am at Dip Falls Car park which is approximately 135 km from Devonport (i.e. approximately a 1hour 45minutes drive). Heading west along the Bass Highway look for the Mawbanna Road/C225 turn off in Black River (near Peggs Beach Conservation area). Follow C225 to Dip Falls Road in Mawbanna. Helen Robertson and fellow enthusiast Pat Harrison will be our guides. Contact: June Hilder (0424350183)

Sunday 6 June— **Fingerpoint.** Linda Barker's place, 29 Cummings Rd Harford. Meet at 10 am. Directions: Turn onto Lades Rd, (first gravel road on left after Rubicon River Bridge, or first right after Narawntapu turnoff). Drive approx 350 m, Cummings Rd starts at a white boom gate. Travel approx 2 km to the house, veering left at the V in the road when you come to my neighbour's (i.e. the first house.) Approximately 300m further on is my house, where we will park (there will be signs up on the day). Activities include: bird counts; looking for evidence of devils or wombats; exploring the wetlands while they are wet!; having lunch in the camp shed with a big open fire. Please stay out of the very sensitive gully area with the burrowing crayfish. Leader: Linda Barker (0409025422)

Sunday 4 July— Food Plants International Meet at 10 am outside the Centre, 109 Wilson Street, Burnie. This is a Christian not-for-profit organisation that aims to provide information about edible plants to help improve food security and regular supply for the world's poor. Founder, Bruce French commenced researching and documenting nutritious local plants in Papua New Guinea during the 1970s. Bruce has offered to open the Centre for our outing and we are welcome to eat our lunch under cover at the Centre. For members wishing to undertake an afternoon walk in addition to visiting the Centre, we could visit the Fern Glade Reserve for a short easy riverside walk in the afternoon. There is so much to see at the Centre, we may not be able to tear ourselves away. Leaders: June Hilder 0424350183 and Hazel Britton 64252785.

Sunday 1 August—AGM CNFN Patron Dr Peter McQuillan will give a short presentation. Details in the next e-news.

We welcome articles for inclusion in the natural news. Please send unformatted word documents and photos attached to the email to blacksugarloaf@gmail.com. Check the website for more details on how to contribute:

https://www.disjunctnaturalists.com/articles3/contributions.htm

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