Response of grassland fungi to an agricultural change

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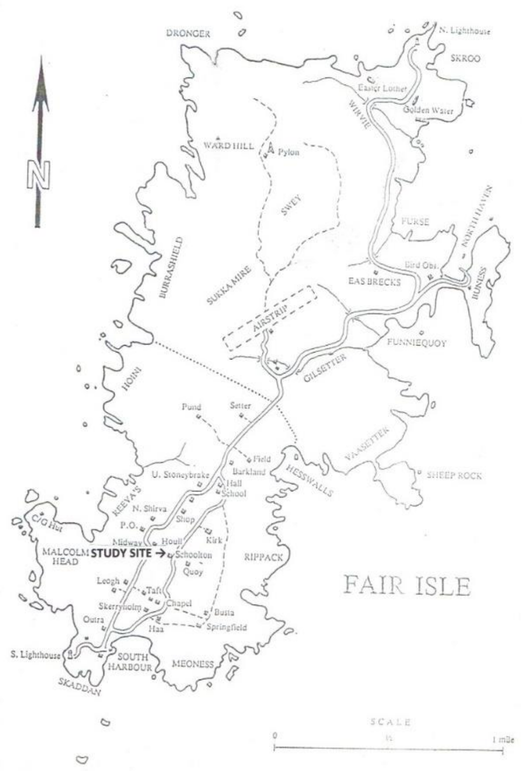
The island of Fair Isle is splashed with colour throughout the summer. As autumn sets in the flowers give way to waxcaps and other grassland fungi which adorn grassy places from late August to October. Even wet areas have their specialists and the only grassland generally lacking them is the improved fields and leys of the in-bye (=crofting area).

The most intensively grazed fields and silage/hay parks of the in-bye receive a light dusting of agricultural fertilisers every May. This practice to enhance the nutrient value and growth rate of the grass began in the early 1970s. Prior to that, the cost was prohibitive for an economically constrained community. The fields at Schoolton were amongst those receiving fertiliser when the first author and his wife took over the tenancy of Schoolton croft (Figure 1) in 1990. Initially the only change to field management was a small reduction in livestock numbers. In 1996, however, a decision was made to cease the application of artificial fertilisers.

Figure 1. Location of study site within island, Schoolton, Fair Isle.



In September 2004, waxcap (*Hygrophoraceae*) fruiting bodies were noted for the first time in the permanent pasture and there was a small increase the following autumn. Grassland fungi are recognised as a priority group for conservation (e.g. Long & Ward 2005, Genney *et al*. 2009). The group is in serious decline in Europe and to a lesser extent in the UK. A driver in this decline has been habitat loss through agricultural changes, which include grassland 'improvement'. This background was the catalyst to contact the second author who had already carried out a 3 year study of the fungi of Mainland Shetland (Watling, 1992). Here was a fortuitous opportunity to observe the response of a group of basidiomycetous fungi of conservation importance to the ending of artificial fertiliser application.

The appearance of these fungi was a by-product of an objective to return the southern part of Schoolton park to a flower-rich meadow. Dating back to *circa* 1980, the land had received 25 kg agricultural fertiliser (NPK at proportions of 21-8-11) per hectare every spring. The last application was on 2nd May 1995. Since then, no fertiliser of any type, other than natural droppings from the sheep, has been applied to any part of Schoolton croft. Change in the vegetation community was slow, with the first yellow rattle *Rhinanthus minor* colonising the southern part of the park, cut over in late August for silage, in 2004. Concurrently the first waxcap was noted in the northern part of the field, given over to permanent grazing. The occurrence of a few more waxcaps in 2005 prompted the two authors to set up a sampling regime starting in 2006, two years from the first waxcap fruiting body and eleven years after the last NPK application.

The sampling regime

The sampling was conducted over the six-year period, 2006 to 2011, and comprised five collections per annum of grassland fungi fruiting bodies at approximately 10-day intervals between 21st August and 10th October in sheep-grazed permanent grassland immediately north of Schoolton crofthouse, Fair Isle. The target group was 'grassland fungi', defined for our purposes as members of the *Hygrophoraceae* (waxcaps) and *Entoloma* (pink-gill) groups. We also included praticolous *Clavariaceae* (fairy clubs). To make the study manageable, a 40 x 30 m (1200 m²) representative parcel of permanent pasture, centred on HZ 2047 7055, was selected in the north-east portion of the field.

Each field collection comprised two persons, one collecting and one processing. Processing involved labelling and maintaining each sample in separate containers. Thirty minutes were allocated to each collection after which no samples were taken, irrespective of numbers left in the study plot. Attempts were made to sample from all parts of the study plot in the allotted time. Immediately following collection, the samples were labelled, dated, individually photographed and placed in an airing cupboard to dry. Once dry, the fungi were sent to the second author for identification. As an adjunct to the sampling activity, an inventory of plants was made along with details of the structural complexity of the vegetation, topography and soil conditions (depth, pH values) in the study plot.

The study plot (Figure 2)

The study plot was part of a larger field area of 2.05 hectares, lightly grazed by up to 17 sheep (headage 8.3 per hectare). Grazing pressure increased from the end of May to mid August (headage 26 per hectare) while the southern part of the field was fenced off to fulfill its purpose as a silage park. The density of livestock was sufficient to suppress grass growth and maintain vegetation cover in the study area as a low, tightly grazed sward.

Figure 2. Location of study site within its immediate environment, Schoolton, Fair Isle.

The study site comprised a somewhat diverse plot in terms of topography, soil depth, acidity and plant species (35 vascular plants, 10 mosses, 5 liverworts). The plot sloped gently from west to east and was predominantly a tight sward of low sheep-grazed grassland. A rocky substrate was generally close to the surface with occasional exposed rock, including a 2 m high vertical rock face in the west, giving way to a band of deeper soils in the eastern part but none deep enough for agricultural disturbance such as ploughing. Vegetation height was largely determined by soil depth with a tight ground-hugging sward on thin soils giving way to a band of low but taller grassland in the north, east and south-east portions. The overall characteristics of the site are best described as reverting from improved to semi-improved low to moderate nutrient grassland (pH 4.5 to pH 5.5 in deeper soils).

The results

The grassland fungi

During the six year study a total of 26 waxcap *Hygrophoraceae* taxa, 17 from the pink-gill group and 3 fairy club fungi was recorded within the study plot (Tables 1 and 2).

Table 1. Waxcap *Hygrophoraceae* taxa: total number of encounters from the five collections per annum during the study period, 2006-2011.

TAXA (sensu Orton, 1960)	**2006**	**2007**	**2008**	**2009**	**2010**	**2011**
Hygrocybe cantharellus	1		1	2		
Hygrocybe chlorophana sensu stricto	3		2	2	1	4
Hygrocybe coccinea	1		2	1	3	3
Hygrocybe insipida	4		1	2	2	3
Hygrocybe pratensis	2	1	3	2	2	5
Gliophorus (Hygrocybe) psittacina	3	1	1	3	1	4
Hygrocybe turunda	1					
Hygrocybe flavescens sensu Orton		1	2	2		3
Hygrocybe miniata sensu Orton		2	1			
Hygrocybe quieta		1	1	2		3
Hygrocybe reidii		2		1	2	
Hygrocybe strangulata		1	3			2
Hygrocybe ceracea			1	2	2	5
Hygrocybe irrigata			1			1
Hygrocybe laeta			1	2	1	2
Hygrocybe nivea			1			
Hygrocybe obrussea			1			
Hygrocybe reai			1		1	
Hygrocybe berkeleyi				1		
Hygrocybe aurantiosplendens					1	
Hygrocybe coccinea					1	
Hygrocybe helobia					1	3
Hygrocybe fuscescens						1
Hygrocybe glutinipes						3
Hygrocybe russocoriacea						1
TAXA YEAR TOTALS	7	7	16	14	12	15
TAXA ACCUMULATIVE TOTALS	7	12	18	19	22	25

Note 1: For each taxon, the number in each column refers to the number of sampling visits in which the taxon was encountered, the maximum possible score per annum being five.

Note 2: *Recent DNA studies have shown that *H. psittacina* now sits in a separate genus, *Gliophorus* (Ainsworth *et al.*, 2013).

Table 2. *Entoloma* (Entolomaceae) and Clavariaceae: total number of encounters from the five collections per annum during the study period, 2006-2011.

TAXA (sensu Orton, 1991)	2006	2007	2008	2009	2010	2011
Entoloma (Nolanea) conferendum	3		1	1		2
Entoloma (Nolanea) infula	1					
Entoloma (Leptonia) lividocyanula	1			1		
Entoloma (Leptonia) pseudoturci	1					
Entoloma (Leptonia) sodale	1		1			1
Entoloma (Nolanea) clandestinum			1			
Entoloma (Leptonia) corvina			1	1		
Entoloma (Nolanea) juncina			1			
Entoloma (Nolanea) papillata			1			
Entoloma (Nolanea) sericea			1	1		
Entoloma sericea var. nolaniformis				1		
Entoloma (Leptonia) atrocaeruleum						1
Entoloma (Nolanea) farinolens						1
Entoloma ?inutile						1
Entoloma ?ochromicaceum						1
Entoloma (Leptonia) serrulata						1
Entoloma (Eccilia) undata						1
Clavulinopsis corniculata		1	1			
Clavulinopsis fusiformis						1
Clavulinopsis helvola	3	1	1		2	3
TAXA YEAR TOTALS	6	2	9	6	2	10
TAXA ACCUMULATIVE TOTALS	6	7	12	13	13	20

Trends

The results indicate a year on year increase in number of waxcap taxa. The trend was not as smooth for the *Entoloma* group, suggesting a slower recovery. Nevertheless, the accumulative number of taxa more than tripled over the six years.

This study sampled the fruiting bodies. Fruiting depends on a number of factors including weather conditions during and prior to the production of the above-ground structure and some fungi are notoriously shy about fruiting if the conditions are not right. This may explain the 'no-show' of *Entoloma* fruiting bodies in 2007 and 2010 (Table 3). However, Newton *et al.* (2003) and Griffith *et al*. (2013) have demonstrated that the pink-gills respond to environmental conditions differently than the waxcaps. At Schoolton, only *Hygrocybe pratensis* and *Hygrocybe psittacina* were recorded in all six years. Both are widespread on the isle away from agricultural improvement.

Implications of the findings

Because of the conservation interest in waxcaps, diversity amongst the group has been used as an indicator of habitat quality. This was quantified for Denmark by Rald

(1985) who used the number of species to evaluate levels of grassland conservation value (Table 3). Vesterholt *et al*. (1999) added the category of 'international importance' once a threshold of 22 species was achieved, while Nitrare (1988) incorporated Clavariaceae, Geoglossaceae, *Entoloma* and *Dermoloma* – collectively shortened from their group names to 'CHEGD' (Rotheroe *et al*., 1996) – as further indicators of grassland conservation value.

Table 3. Levels of conservation value, after Rald (1985).

Conservation value	Total number of *Hygrocybe* species
of national importance	17-32
of regional importance	9-16
of local importance	4-9
of no importance	1-3

Based on these categories, the Schoolton study area attained national importance status, at least for *Hygrophoraceae*, in 2008 and international importance by the end of the study – even applying a more conservative taxonomic treatment.

The use of species or taxa totals to assess habitat conservation value has its limitations (see for instance Newton *et al*., 2003). Scandinavian researchers refined the system by giving weighted scores to rare or threatened species (Jordal and Gaarder, 1993; Jordal, 1997; Vesterholt *et al*. (1999), an approach used in Britain and Ireland too (e.g. Rotheroe, 1999; Evans *et al*., 2001; McHugh *et al*. 2002).

The majority of the taxa in the study site are commoner species. However, the list includes two rarely recorded *Hygrocybe* and a further two appearing on European Red Lists, *Hygrocybe quieta* and *Hygrocybe insipida* – although the former is rather common and widespread in Scotland. Nine (53%) of the *Entoloma* taxa are classified as rarely recorded and one, if determined correctly, is very rare with only two records, both in Scotland. The UK status of each taxon, where known, is summarised in Tables 4 and 5.

Table 4. Study site *Hygrocybe* taxa: UK status.

TAXA (following Orton, 1960)	**UK Status**
Hygrocybe cantharellus	Occasional; widespread
Hygrocybe chlorophana sensu stricto	Common; widespread
Hygrocybe coccinea	Common; widespread
Hygrocybe insipida	Uncommonly reported; European Red List
Hygrocybe pratensis	Common; widespread
Hygrocybe psittacina	Common; widespread
Hygrocybe turunda	Rarely recorded; apparently widespread
Hygrocybe flavescens sensu Orton	*Common; widespread*
Hygrocybe miniata sensu Orton	Occasional; widespread
Hygrocybe quieta	Uncommonly reported; European Red List
Hygrocybe reidii	Uncommonly reported; widespread

Hygrocybe strangulata	*Relatively common; widespread*
Hygrocybe ceracea	Common; widespread
Hygrocybe irrigata	Occasional; widespread
Hygrocybe laeta	Occasional; widespread
Hygrocybe nivea	Common; widespread (as *H. virginea*)
Hygrocybe obrussea	*Occasional; widespread*
Hygrocybe reai	*Common; widespread*
Hygrocybe berkeleyi	Occasional; widespread
Hygrocybe aurantiosplendens	Occasional; widespread
Hygrocybe coccinea	Common; widespread
Hygrocybe helobia	Rarely recorded; widespread
Hygrocybe fuscescens	Occasional; localised
Hygrocybe glutinipes	Occasional; localised
Hygrocybe russocoriacea	Common; widespread

Note: UK status following Orton (1960) in italics; all others, Legon & Henrici (2005)

Table 5. Study site *Entoloma* and Clavariaceae: UK status.

TAXA (following Orton, 1991)	**UK Status**
Entoloma (Nolanea) conferendum	Very common; widespread
Entoloma (Nolanea) infula	Uncommonly reported; apparently widespread
Entoloma (Leptonia) lividocyanulum	Rarely recorded
Entoloma (Leptonia) pseudoturci	Rarely recorded
Entoloma (Leptonia) sodale	Rarely recorded
Entoloma (Nolanea) clandestinum	Rarely recorded; mainly Scotland
Entoloma (Leptonia) corvinum	Occasional; widespread
Entoloma (Nolanea) juncinum	Rarely recorded
Entoloma (Nolanea) papillatum	Frequent; widespread
Entoloma (Nolanea) sericea	Common; widespread
Entoloma sericeum var. nolaniformis	*Common; widespread*
Entoloma (Leptonia) atrocaeruleum	Rarely recorded; apparently widespread
Entoloma (Nolanea) farinolens	Rarely recorded; apparently widespread
Entoloma ?inutile	Rarely recorded
Entoloma ?ochromicaceum	Very rare; two records (Scotland)
Entoloma (Leptonia) serrulata	Frequent (common in Scotland); widespread
Entoloma (Eccilia) undata	Rarely recorded; apparently widespread
Clavulinopsis corniculata	Common; widespread
Clavulinopsis fusiformis	Common; widespread
Clavulinopsis helvola	Common; widespread

Note: The taxa follow Orton (1991) but this author spread the species, now placed in *Entoloma* by Legon & Henrici (2005), over several separate genera; they are indicated in brackets above (UK status in italics follow Orton, 1960); all others correspond with Legon & Henrici (2005)

Conservation research

Unimproved grassland loss in Western Europe over the last 75 years is estimated at 90% (Lovegrove *et al*, 1995; Hewins *et al*., 2005; Stevens *et al*., 2009). CHEGD grassland species are major casualties. More than 75% of the UK species are now included in Red Data Lists for at least one European country (Ing, 1993; Griffith *et al*., 2013). This has prompted a number of studies into the various causes and implications of decline. They include field studies to locate and assess the ecology and conservation status of mycologically important grasslands (e.g. Newton *et al*., 2003; Griffiths *et al*., 2013), Landsat and field mapping, and experimental work to investigate the impact on fungi of fertiliser treatments, including plots with none (see Griffiths *et al*., 2004). Recently a restoration experiment has been launched to establish waxcaps at a new location by translocation of spores from an existing site as a development mitigation measure (Wright, 2015).

However, the speed and extent that grassland fungi populations establish or, in the case of loss, recover remains largely unknown. Russell (2004) made reference to a few *Hygrocybe* species appearing after 10 years of 'abandoned' fields and Griffith *et al*. (2004) considered that it may take some years before visual evidence of success is displayed, while other opinions were of 'populations failing to recover for decades after cessation of fertilization' (https://sites.google.com/site/scottishfungi/conservation/grassland-fungi).
At Schoolton, the on-site presence and local knowledge of a field observer constantly observing over a long period has provided a rare opportunity to provide novel information on fungal recovery after the cessation of chemical fertilizer application; and the results run counter to assumptions that '*once damaged, these habitats* [grasslands] *are extremely difficult, if not impossible, to restore'* (Genney *et al*., 2009). This study suggests that, with the right management, conditions and perseverance, restoration can occur, and more quickly than feared by some.

It should be stressed that the Schoolton study does not have the full rigour of a planned scientific experiment. It is purely an opportunistic study stimulated by the appearance of basidiomes when, according to the literature, fertiliser suppresses or removes grassland fungi from the ecosystem. The results would be far more powerful if replicate plots had been set up and comprehensive soil quality testing done. Other limitations include the possibility that the results merely reflected grassland fungi response to subtle changes in grazing pressure, weather conditions, seeding by spores from elsewhere on the isle or indeed from farther afield rather than a direct relationship to the cessation of agricultural chemical applications. Nevertheless, the sudden appearance and unforeseen proliferation of CHEGD grassland fungi on land walked most days by the resident crofter (NJR) is of sufficient note to suggest a new avenue of research for long-term investigation.

Scotland: status, conservation, appreciation

Scotland is of exceptional importance for grassland fungi (Newton *et al*., 2003). The UK Department for Environment, Food and Rural Affairs (DEFRA) and Scottish Natural Heritage (SNH) funded research into the classification and ecology of fungi from unimproved grassland habitats (so-called 'waxcap grasslands'). The research, by CABI, Royal Botanic Gardens Kew and the University of Aberystwyth, recognised them as fragile ecosystems containing a high diversity of fungi. It concluded that these grasslands were especially vulnerable to 'improvements' in the form of agricultural fertilisers causing the loss of many fungal species, with populations failing to recover for decades even after cessation of fertilisation (https://sites.google.com/site/scottishfungi/conservation/grassland-fungi). A SNH commisioned report flagged up the CHEGD group has as indicators of environmentally high-quality grassland and recommended them as a priority group for conservation (Genney *et al*., 2009), a position accepted by the Scottish Government. Yet the Schoolton study field was refused entry to the Scottish Rural Stewardship Scheme whose management strategy – ostensibly for the whole of Scotland – appeared to be based on "remove livestock for two months of summer". Grass grows luxuriantly on Fair Isle in summer. Without summer grazing pressure, vegetation cover would limit opportunities for grassland fungi fruiting bodies. An appeal to the Scottish Executive Environment and Rural Affairs Department prompted a reply signed by the Minister himself that if we did not remove them 'the sheep may tread on the nests and chicks of birds'.

It seems that agri-environmental schemes cater only for birds and colourful flowering plants. The implications are that this is all the general public wants. The failure to recognise other priority groups is a serious flaw in conservation management and exposes shortcomings in biodiversity conservation generally. This needs to change. Scotland is of exceptional importance for grassland fungi and should be leading the way. A wider lack of familiarity and appreciation may be at the root of failures in conservation management for this group and the habitats it requires. This study sends a message that restoring high-quality grasslands and the fungi community it supports can be achieved under the right conservation management – but also requires appreciation, commitment and pro-active measures by the conservation bodies representing the general public.

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