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The Park Grass Experiment 1856–2006: its contribution to ecology

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Summary

- 1** The Park Grass Experiment, begun in 1856, is the oldest ecological experiment in existence. Its value to science has changed and grown since it was founded to answer agricultural questions. In recent times the experiment has shown *inter alia* how: plant species richness, biomass and pH are related; community composition responds to climatic perturbation and nutrient additions; soil is acidified and corrected by liming. It also provided one of the first demonstrations of the evolution of adaptation at a very local scale and contains a putative case of the evolution of reproductive isolation by reinforcement. The application of molecular genetic markers to archived plant material promises to reveal a whole new chapter of genetic detail about the long-term dynamics of plant populations.
- 2** Over the range of values observed at Park Grass, biomass (productivity) has a negative effect upon species richness. Any positive effect of species richness on productivity could only be weak by comparison. The experiment provides support for both the competitive exclusion and pool size hypotheses for determination of species density.
- 3** Instantaneous comparisons of species richness between plots do not accurately reflect temporal rates of loss which may be multiplicative rather than additive. This suggests that comparisons among sites, nutrient inputs, especially N treatments, or soil acidity may in general underestimate the threat posed to plant species diversity by long-term changes in plant nutrient availability, both enrichment and depletion.
- 4** Differences between plots at the community level are maintained despite a flow of propagules between plots. There is no strong evidence for a spatial mass effect.
- 5** Guild (grass/legume/other) compositions of plant communities have equilibrated, but the species composition within guilds is more dynamic and continues to change over time, suggesting that species and guild abundances are independently regulated.
- 6** At least some members of all the major trophic levels, including predators (spiders), herbivores (leafhoppers) and detritivores (springtails) are treatment-specific in their distributions.
- 7** Plant populations on Park Grass are subdivided by treatments which, to some degree, have led to plots becoming genetically isolated from one another and decoupled demographically. This subdivision has created a metapopulation structure in each species, characterized by species-specific rates of local colonization and extinction.
- 8** Inverse clines in flowering time occur in the grass *Anthoxanthum odoratum* across some plot boundaries. These suggest that reproductive isolation between plots has been reinforced by natural selection.
- 9** Drift as well as selection may have taken place in *A. odoratum*, especially on plots where effective population size is restricted by population bottlenecks caused by drought.
- 10** Park Grass illustrates how long-term experiments grow in value with time and how they may be used to investigate scientific questions that were inconceivable at their inception. This is as likely to be true of the future of Park Grass as it has proved to be of its past.

Key-words: long-term experiment, permanent grassland, biodiversity, plant population dynamics, natural selection, plant nutrition, soil fertility, stability, ecological genetics

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Introduction

The Park Grass Experiment (Fig. 1), begun by John B. Lawes and Joseph H. Gilbert in 1856, at Rothamsted in Hertfordshire, England, is the longest running ecological experiment in the world (Tilman *et al.* 1994) and over 170 publications have arisen from it to date. The early results from Park Grass quickly answered the question that motivated Lawes and Gilbert to start the experiment, namely how different fertilizers would improve the yield from hay meadows (Lawes & Gilbert 1859). Today, Park Grass has acquired new relevance for the study of fundamental ecological processes and for nature conservation. It has inspired new ecological theory (the resource ratio hypothesis (Tilman 1982)) and has helped ecologists to recognize the value of long-term experiments in ecological studies, notably at Cockle Park in Northumberland, UK (Arnold *et al.* 1976), Wageningen in the Netherlands (Elberse *et al.* 1983), Cedar Creek, Minnesota, USA (Tilman 1987) and Nash's Field, Silwood Park, UK (Edwards *et al.* 2000). This paper aims to illustrate the modern relevance of Park Grass and its unique and increasing value to various branches of ecology.

In the temperate climate and on the silty clay loam soil at Rothamsted, processes of soil development and change are slow and require time to play out. Over 150 years the different treatments have variously resulted in soil acidification, changes in soil organic matter, and P and K enrichment or depletion, and these have all affected the fragmentation of plant populations. Fortunately, some of these treatments have experimentally reproduced two of the principle extrinsic impacts that threaten plant biodiversity in natural habitats more generally, namely plant nutrient enrichment and acidification of the soil, and therefore offer a unique opportunity to study their long-term effects on extinction. The numerous long-term treatments reveal relationships between nutrient availability and grassland biodiversity (Silvertown 1980; Crawley *et al.* 2005). They have also provided a test of the influence of species richness on the stability of plant communities in relation to climate (Dodd *et al.* 1994b; Silvertown *et al.* 1994b) that is highly relevant to the diversity–stability debate (Loreau *et al.* 2001).

A variety of patterns of long-term plant population dynamics related to life history have been discovered in the experiment that are not detectable in shorter time series (Dodd *et al.* 1995; Silvertown *et al.* 2002). Due to its well-documented 150-year history, Park Grass is a fertile arena for the study of natural selection (Snaydon 1970) and a promising subject for ecological genetics

(Silvertown *et al.* 2005). Soil and hay samples from the experiment have been archived from the beginning and these have been used in retrospective studies of atmospheric pollution (Zhao *et al.* 1998; Blake *et al.* 1999; Warneke *et al.* 2002) and population genetics (Biss *et al.* 2003). In the sections which follow, we provide a brief overview of the experiment, selectively review results under the main topics of soil processes, plant dynamics, evolution and fauna and then attempt to draw some of these threads together into a synthetic overview that provides some general pointers for future research.

The Park Grass Experiment

THE EXPERIMENTAL SITE

The site of the experiment is about 2.8 ha of almost level land that had certainly been in grass for more than a century when the experiment began in 1856. The soil was slightly acid (pH 5.4–5.6) and the nutrient status poor. The top 23 cm of soil is a silty clay loam overlying clay-with-flints and is moderately well drained. At the start, the species composition of the herbage appeared uniform across the whole site. In terms of the modern National Vegetation Classification (Rodwell 1992) the vegetation of the meadow would have been mesotrophic grassland of MG5 type (Dodd *et al.* 1994a).

EXPERIMENTAL TREATMENTS AND MANAGEMENT

Fertilizer and liming treatments

A plan of the experiment and amounts and forms of N, P and K tested are given in Fig. 2, together with the average nutrient content of the organic manures; more details are in Table S1 in the Supplementary Material. Plots 1–13 were started in 1856; plots 14–17 were added in 1858 while three others started later: plot 18 (1865) and 19 and 20 (1872). Before 1905, treatments to a limited number of whole plots were changed (Warren & Johnston 1964). Since then, modifications have been made at various times by dividing the existing plots, but, the original treatment has always continued on one subplot. A regular application of lime on half-plots to test soil pH effects was started in 1903 (Warren & Johnston 1964). By 1959 this liming had *gradually* created different pH values in both the 0–23 cm and 23–46 cm soil depths for each treatment (Warren & Johnston 1964) (Fig. 3). The difference in soil acidity with the various nutrient inputs had a major effect on species composition by 1964



Fig. 1 An aerial view of the Park Grass Experiment looking due north, taken on 23 May 2005. Note the sharp plot boundaries, many of which are clearly demarcated by differences in vegetation.

(Thurston *et al.* 1976). Consequently, to extend the range of pH values, most plots were divided into four subplots (a–d, Fig. 2) in 1965, with the aim of achieving surface soil pH values of 7, 6 and 5 where possible by applying appropriate amounts of lime, preserving the fourth subplot as an unlimed control (Warren *et al.* 1965). Important modifications since then have tested changes in nutrient inputs by dividing plots 9, 14, 13 and 2 (see Fig. 2 for details).

Achieving and maintaining the desired pH in the surface soil on each subplot has taken many years, the rate of change has been variable and very different amounts of lime have been required. The current pH of the top 23 cm of soil is given in Table S1. Soils with no nutrient inputs (unmanured) have changed little from an estimated value of 5.5 in 1856 (Fig. 3); the higher pH in 1923 compared to 1876 is due to the small amount of lime added to all plots in the 1880s. The higher values on plots given sodium nitrate must be because sodium ions (Na^+) have been lost as balancing cations in preference to Ca^{2+} . Where N is applied as ammonium sulphate without lime, the soils have become progressively more acid, reaching an apparent equilibrium at about pH 3.6. The same amount of chalk was added to soils receiving either ammonium sulphate or sodium nitrate until 1964 (Fig. 3). After that date, the $\text{NO}_3\text{-N}$ and the $\text{NH}_4\text{-N}$ plots required, respectively, 14 and 63 t ha^{-1} CaCO_3 in total between 1965 and 2003 to bring or maintain them both to pH 7 by 2000 (mean 1995–2005) (Fig. 3). The difference in the amount of chalk needed was due to the greater acidifying effect of ammonium sulphate.

Hay cutting and data collection

Herbage has always been cut and made into hay on the plots, usually in June. After hay harvest, the subsequent regrowth was grazed by sheep for 15 of the first 19 years; since 1875 a second cut has been taken and removed from the plot. Thus, since 1875 species composition has not been affected by grazing.

Yields have been recorded, and samples kept for analysis, from each plot and every harvest since 1856 and these data are lodged in the online Electronic Rothamsted Archive (ERA: www.era.iacr.ac.uk/parkgrass_1.html). Quantitative (weight of each species present in hay samples) and qualitative (presence/absence of species) records of the botanical composition of plots have been made at varying intervals. The first comprehensive, quantitative record was made in 1862 and then at 5-year intervals until 1877. The most recent quantitative botanical analyses on all plots were made annually between 1991 and 2000 (Crawley *et al.* 2005). Numerous quantitative surveys of subsets of plots were made between 1877 and 1976 (Williams 1978). A separation of the hay by weight into three guilds: grasses (Poaceae), legumes (Fabaceae) and ‘other species’ (non-legume, broadleaved herbs) was made annually for various plots for long periods between 1858 and 1948 (Williams 1978; Silvertown 1987). Qualitative records of the presence/absence of species were taken twice a year from every plot for 60 years between 1920 and 1979 (Dodd *et al.* 1995) as well as less regularly at other times before and since (Crawley *et al.* 2005). Botanical survey data are lodged in the ERA.

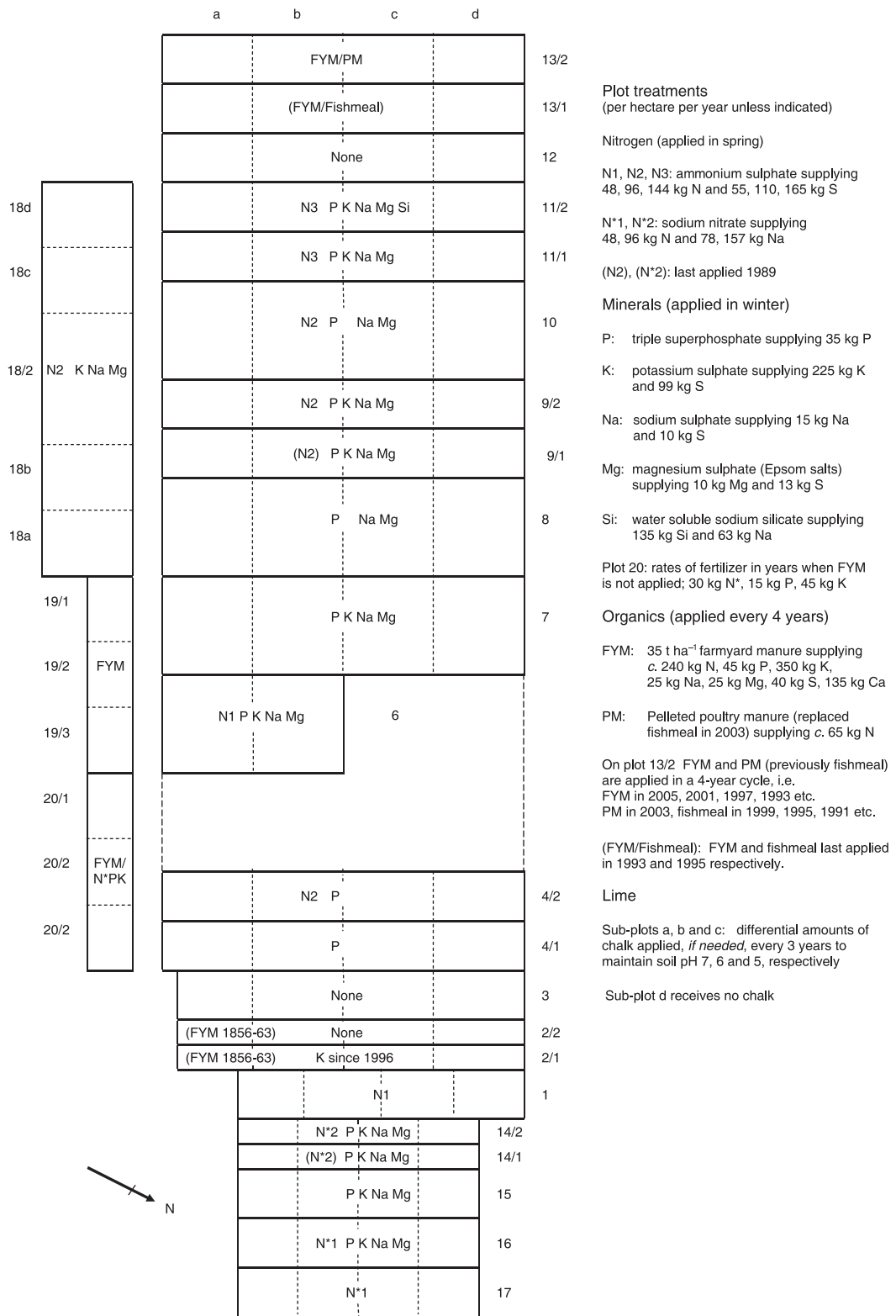


Fig. 2 Plot layout and current treatments of the Park Grass Experiment.

Soil processes

N INPUTS, SOIL ACIDITY AND ACIDIFICATION

Soil acidification has greatly affected species composition of the sward. Soils have acidified as a result of applying

ammonium sulphate and because of atmospheric N inputs. Fertilizer N inputs have remained constant. However, the input of atmospheric N (as wet and dry deposition) varies, both within and between years, depending on emissions to the atmosphere and rainfall. Goulding *et al.* (1998) calculated that the annual atmospheric N input totalled c. 10 kg ha⁻¹ at the start of the experiment,

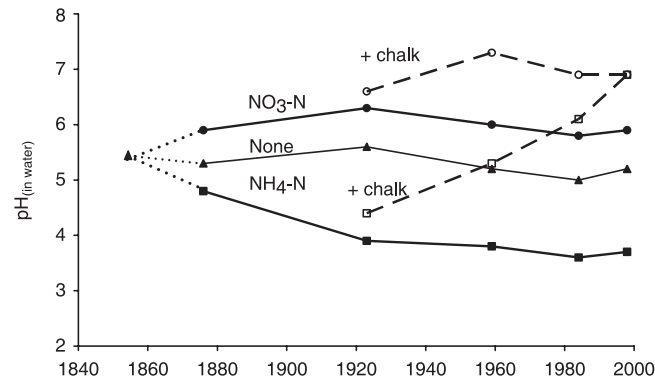


Fig. 3 Surface soil, 0–23 cm, $\text{pH}_{(\text{in water})}$ since 1856; ▲ = plot 3 (unmanured); ● = plot 14 (N^*2PKNaMg); ○ = plot 14 plus chalk since 1920; ■ = plot 9/2 ($\text{N}2\text{PKNaMg}$); □ = plot 9/2 plus chalk since 1903.

increased gradually to $c. 30 \text{ kg ha}^{-1}$ by the 1960s before peaking at $c. 45 \text{ kg ha}^{-1}$ in the mid-1980s. Since then there has been a decline to $c. 35 \text{ kg ha}^{-1}$. Acidification also occurs from inputs of sulphur dioxide which reached a maximum of 65 kg S ha^{-1} each year in 1980 but have since declined dramatically (McGrath *et al.* 2002) as a result of declining emissions from industrial sources.

ORGANIC MATTER AND P AND K STATUS OF THE SOIL

Data from Park Grass has been crucial in extending the use of the soil organic matter model, RothC (Jenkinson 1990). Originally developed to look at the turnover of soil organic matter in the plough layer of arable soils, organic C and ^{14}C measurements on archived Park Grass soils were used to demonstrate its validity for use in permanent grassland (Jenkinson *et al.* 1992). Additional organic C and ^{14}C analyses of recent and archived soils sampled, by horizons, to a depth of 90 cm on Park Grass and on arable and woodland sites are being used to further extend RothC such that the turnover of the very significant proportion of C stored in subsoils can be taken into account. RothC and the Hadley Centre's coupled climate–carbon cycle general circulation model are now being used to predict the effects of global change on soil carbon stocks (Jones *et al.* 2005).

In 2002, in the 0–23 cm soil layer of the unlimed soils receiving no N or N as sodium nitrate ($\text{pH } 5.0\text{--}5.6$) there was 3.0% C, whilst those receiving ammonium sulphate ($\text{pH } 4.1\text{--}3.6$) contained 4.8–6.6% C. Most of this difference is due to the accumulation of C in the mat of undecomposed material on the most acid soils. This mat may affect the ability of the roots of germinating seeds to grow into the underlying mineral soil. Where soils have a pH of 6 or 7 a mat did not develop, or was broken down, and percentage C is similar on all treatments, ranging from 3.7% to 4.2% C.

Current levels of plant-available soil phosphorus (Olsen P) and potassium (exchangeable K) in the 0–23 cm soil layer are either very small or very large. On soils not given P or K, Olsen P is about 4 mg kg^{-1} and exchangeable K about 60 mg kg^{-1} (range 44–71) and

species composition will depend on the ability of plants to survive where there is so little plant-available P and K. Where P and K have been applied there is sufficient in the soil to meet the needs of any plant (Fig. 2).

Plant dynamics

COMMUNITY CHANGE

The various fertilizer treatments initiated in 1856 had so dramatic an effect upon the botanical composition of different plots that Lawes & Gilbert (1859) wrote that in less than two years 'the experimental ground looked almost as much as if it were devoted to trials with different seeds as with different manures'. Although the initial changes were rapid, the communities continued to change over the following 40 years, not reaching an equilibrium until the early 20th century (Silvertown 1980; Dodd *et al.* 1994a). Even then, the equilibria were only attained at the aggregate level (Micheli *et al.* 1999), measured in terms of the proportions of grass/legume/other species (Fig. 4). These proportions were distinctively different between treatments; grasses consistently dominated ($c. 90\%$) N-fertilized plots; legumes were abundant ($> 30\%$) on plots receiving P and K but no N, and an intermediate ratio of the three guilds occurred on unfertilized plots (Silvertown 1987).

While the guild composition of communities had equilibrated by 1910, the species composition within guilds was more dynamic and continued to change over time, suggesting that species and guild abundances are independently regulated (Silvertown 1980). Species changes are discussed in a later section. The equilibrium in guild composition is a dynamic one, continually perturbed by climate. Annual rainfall averages 698 mm, but ranges widely from 380 mm in 1921 to 974 mm in 2000. Silvertown *et al.* (1994) found that above-average spring rainfall directly increased hay biomass (see also Cashen 1947), but altered guild proportions with a one-year time lag, increasing grass abundance at the expense of the other two guilds. They suggested that the lag was the result of interspecific competition for light which, because of positive feedback between growth and light

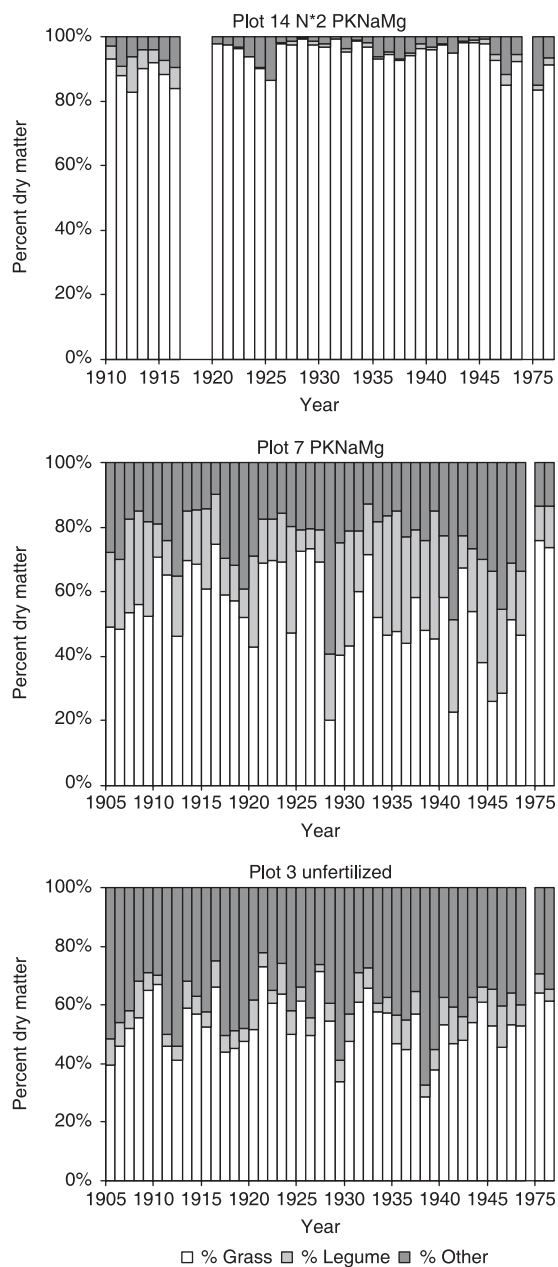


Fig. 4 Relative proportions by dry weight annually between 1910 and 1948 of grasses (white), legumes (light grey) and other broad-leaved herbs (dark grey) on un-limed sections of plots receiving (a) N*, P, K, Na and Mg; (b) P, K, Na and Mg; (c) no fertilizer. Proportions in 1975 and 1976 are shown for comparison. (after Silvertown 1987; with permission and redrawn with data from the Electronic Rothamsted Archive).

capture, would amplify climatic effects on grass growth and thereby increase the competitive suppression of legumes and other species. Dunnet & Grime (1999) subsequently demonstrated such an amplification effect in experimental grassland microcosms.

Changes in fertilizer treatment on plots that have recently been split (see section 'Experimental treatments and management') have afforded the opportunity to observe how quickly grassland communities change in response to a 'press' type of perturbation (Bender *et al.* 1984). Comparing subplots where N fertilization had ceased with controls, Henman (2001) found that the speed

of community change depended upon soil pH. New communities differed from those on plots that had the same current fertilization treatment, but no history of perturbation. This is an important observation: halting the application of N while leaving soil acidity uncorrected is unlikely to cause any major change in sward composition.

RESOURCE COMPETITION AND COMMUNITY STRUCTURE

The clear effects of different fertilizer treatments on plant community composition in the Park Grass Experiment were used by Tilman (1982) in support of the resource ratio hypothesis when he gave the hypothesis its first full exposition. The hypothesis proposes that the number and relative abundances of plant species in a community at equilibrium are determined by competition for essential resources such as plant nutrients and light. All plant species require a common suite of such resources, but each species has its own characteristic ratio of essential resources at which resource availability limits growth. According to the hypothesis, when two or more species consume the same essential resource the winner in the competition for that resource will be the one able to tolerate (i.e. continue growing at) the lowest concentration.

In general, legumes, such as *Lathyrus pratensis* for example, can continue to grow at a lower resource concentration of N than can grasses such as *Agrostis tenuis*. When supplied with sufficient N, grasses have the upper hand against legumes in competition for P. Thus, grasses dominate N-fertilized communities like Plot 14 (Fig. 4a) while legumes are favoured in Plot 7 (Fig. 4b) that receives P, but no N. Coexistence is possible in intermediate situations if there is spatial heterogeneity in N : P ratios. Tilman's (1982) graphical application of the resource ratio hypothesis to competition between *L. pratensis* and *A. tenuis* in Park Grass plots is reproduced in Fig. 5(a).

Tilman (1982) also proposed that differences between Park Grass communities in the relative abundances of four grasses and *L. pratensis* could be explained by the resource ratio hypothesis if the species' relative requirements for light and N were negatively correlated. This trade-off is indicated by the ranking on axes for light and N of zero net growth isoclines in Fig. 5(b). The isoclines and consumption vectors shown in Fig. 5 and in the many other Park Grass examples used by Tilman (1982) are hypothetical, but are consistent with observations made in the experiment. Despite two decades of research inspired by the resource ratio hypothesis (Craine 2005), a recent review (Miller *et al.* 2005) concluded that important work remains to be done in measuring isoclines and consumption vectors in the field in order to test the hypothesis fully.

DIVERSITY

Determinants of species richness

The relationship between species richness and productivity has been the subject of heated debate (Huston

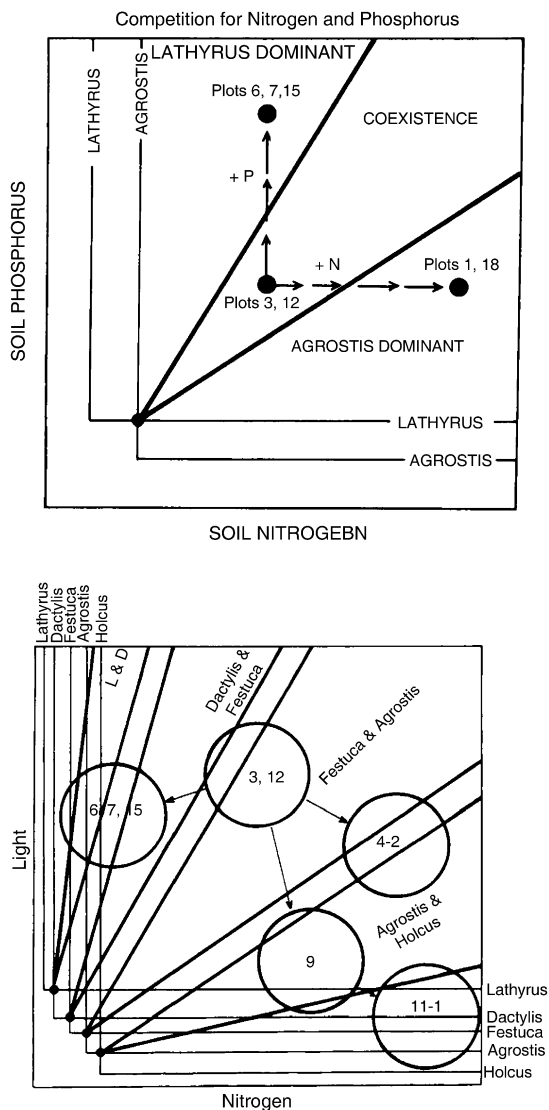


Fig. 5 Application of the resource ratio hypothesis to competition (a) for P and N between *Lathyrus pratensis* and *Agrostis tenuis* and (b) for light and N between *L. pratensis* and the grasses *A. tenuis*, *Dactylis glomerata*, *Festuca rubra* and *Holcus lanatus*. Thin lines are zero net growth isoclines and thick lines are consumption vectors for species. Arrows indicate compositional change caused by nutrient addition from the starting point on unfertilized control plots 3 and 12 towards the composition in 1948/49 of communities on the fertilized plots indicated. In (b), light availability at the soil surface was assumed to decline as biomass increased with N-fertilization (from Tilman, 1982; with permission, Princeton University Press).

1997; Loreau *et al.* 2001). The Park Grass data show unequivocally that, over the range of values observed, biomass (productivity) has a negative effect upon species richness (Crawley *et al.* 2005). Consequently, any positive effect of species richness on productivity could only be weak by comparison, or can occur only in very different circumstances to those prevailing in mesotrophic grassland (Keddy 2004). Modern species numbers vary from 3 to 44 per 200 m² among the plots (Crawley *et al.* 2005). Historically, the primary determinants of this variation were hay biomass and soil pH, which operated in an addi-

tive manner (Silvertown 1980). By the 1990s plots acidified by fertilization with ammonium sulphate had lost so many species that they no longer showed variation in species density in response to biomass (Crawley *et al.* 2005).

All plots, including the unfertilized ones, have lost species with time (Silvertown 1980). On non-acidified plots, for example, about 18 species per plot were lost over the 130 years from 1862 to 1991 (Dodd *et al.* 1994b). Some loss due to the subdivision of plots might be expected, but species–area relationships suggest that a plateau is reached at 75 m², which is the smallest plot size (Crawley *et al.* 2005). Other possible causes of widespread species loss are external. These include varying degrees of soil acidification, inputs of N and S as manures, fertilizers and from aerial deposition, and soil enrichment or depletion of P and K. On plots where soil pH remained at or above its estimated starting value, because of treatment or liming, acidification cannot have been a cause of species loss.

According to the multivariate model of species density variation of Crawley *et al.* (2005), 50 kg N ha⁻¹ year⁻¹ added as fertilizer reduces species number by about 6.5 species, compared to the no N plots. Since the start of the experiment, N-deposition slowly increased to reach a maximum of c. 45 kg N ha⁻¹ year⁻¹ to all plots in the 1980s but the amount is now nearer 35 kg N ha⁻¹ year⁻¹. This input alone could therefore account for a significant, experiment-wide loss of species. Such an effect has already been demonstrated for acid grasslands in the UK (Stevens *et al.* 2004).

Note, however, that the model predicts only about a third of the species loss that was actually observed between 1862 and 1991. This could be because the model is based upon between-plot variation in the 1990s, while the total loss of species took place over the previous 130 years when species number was greater. This difference could be important for three reasons. First, the most vulnerable species may already have been lost by the 1990s. We may call this a ‘sensitivity effect’, and it would result in a lower rate of species loss caused by N addition in the 1990s than previously. The existence of a sensitivity effect is supported by the results of Dodd *et al.* (1994b) who found that the slope of the species richness vs. biomass relationship on non-acid plots was significantly steeper, by 30%, in 1862 than it was in 1991.

The detected effect might explain some of the difference between modelled and expected species losses, but the observed discrepancy is very large and other explanations should be considered. A second reason for the discrepancy could be that the model used only the amount of fertilizer N applied. However, atmospheric and soil derived N (which could vary with past treatment and soil pH) could have increased the total amount of N available to the herbage in any one year. The third and perhaps a more important consideration is that instantaneous comparisons of species richness between plots do not accurately reflect temporal rates of loss which may be multiplicative rather than additive. This is an important possibility because it suggests that comparisons among sites or N treatments

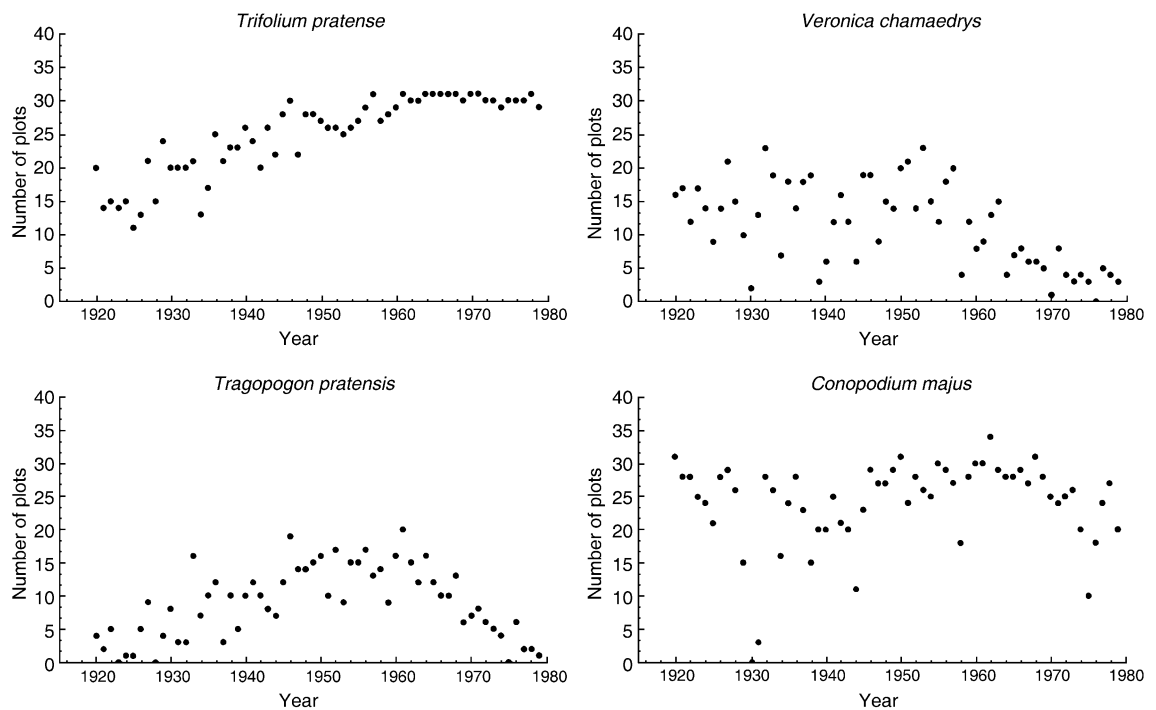


Fig. 6 Metapopulation dynamics of some representative species on non-acidified plots of the Park Grass Experiment: (a) increase in *Trifolium pratense*; (b) decrease in *Veronica chamaedrys*; (c) outbreak in *Tragopogon pratensis*; and (d) fluctuation in *Conopodium majus*. From Dodd *et al.* (1995).

may in general underestimate the threat posed to plant species diversity by long-term changes in plant nutrient availability, both enrichment and depletion.

Diversity and stability

The long-established differences in species diversity between adjacent plots and the very long time series of data on hay biomass make the experiment an ideal system in which to address the long-debated question of how diversity and stability are related (May 1973; Tilman & Downing 1994). Plots with low biomass (which also tend to have greater species richness) and the acid plots have the most variable hay biomass (Dodd *et al.* 1994b). Dodd *et al.* (1994b) regressed the variability of hay biomass, measured in an 11-year moving window, upon biomass itself and species number per plot for 42 different years between 1862 and 1991. In many years (29/42) species richness was positively correlated with stability of hay biomass, although the correlation was significant in only three years. Species richness may therefore have some stabilizing effect upon hay biomass, but the effect would appear to be weak.

METAPOPULATION DYNAMICS

Plant populations on Park Grass are subdivided by treatments which, to some degree, have led to plots becoming genetically isolated from one another (see section 'Evolution', below) and decoupled demographically. This subdivision has created a metapopulation structure in each species, characterized by species-

specific rates of local colonization and extinction. Dodd *et al.* (1995) discovered that between 1920 and 1979 some species occurred in increasing numbers of plots, some declined and another group which they called 'outbreak' species first increased and then decreased again (Fig. 6). Comparing the two groups of species that initially increased (i.e. the increasers and outbreakers) with species that did not change, the former were more ruderal (*sensu* Grime *et al.* 1988) than the latter. Silvertown *et al.* (2002) suggested that the initial increases were triggered by drought which reduced interspecific competition, allowing more ruderal species to increase and confirmed experimentally that the outbreak species had a higher net reproductive rate than other species when released from competition.

Among the two groups that initially increased, Dodd *et al.* (1995) found that those species that decreased again (the outbreakers) had selfing mating systems when compared with the increasers that were able to sustain themselves on plots that they had colonized. This correlation with mating system was confirmed by Silvertown *et al.* (2002) who estimated selfing rates in populations on Park Grass and found that, as expected, outcrossing and genetic diversity were positively correlated. Silvertown *et al.* (2002) proposed that selfing species may have declined after their initial spread because of one or more ecological handicaps created by selfing: inbreeding depression, an inability to adapt to the environment of new plots because of limited genetic variation, or susceptibility to pathogens.

These ideas have not yet been tested, though the outbreak species *Tragopogon pratensis*, which has no

detectable genetic variation, is highly susceptible to rust disease. Results from an annual sampling of plant density and rust infection from 1995 to 1998 and 2000–04 (G.R. Edwards *et al.*, unpublished data), showed that within years, there was a significant negative correlation between plant density per plot and percentage rust infection in 4 of 9 years. In 4 out of 7 years there was a significant correlation between the severity of rust infection on plots and a decrease in plant density the following year. These results suggest the operation of delayed density dependence which might cause population cycling or even chaotic dynamics.

Evolution

LOCAL ADAPTATION

The classic study by Snaydon & Davies (1972, 1976, 1982; Snaydon 1970; Davies & Snaydon 1973a,b, 1974, 1976) of evolution in *Anthoxanthum odoratum* growing on Park Grass plots was an early demonstration of the now well-known tendency towards local adaptation in plants (Linhart & Grant 1996). Even though *A. odoratum* is outcrossing (self-incompatible) and wind pollinated and therefore considerable gene flow across plot boundaries must occur, heritable differences in many traits were found between closely adjacent populations growing on the plots receiving different treatments. Reciprocal transplants between plots receiving different treatments demonstrated that selection coefficients against non-native genotypes could be over 70%, based on plant survival over an 18-month period (Davies & Snaydon 1976).

Snaydon & Davies (1976) found heritable flowering time differences between plants on adjacent plots that could potentially reduce gene flow. At the boundary between the then limed halves of plots 8 (pH 7) and 9 (pH 5.3) there was an inverse cline in flowering date, with plants on the border flowering several days earlier than those either side. Over 30 years after Snaydon and Davies' samples were collected, Silvertown *et al.* (2005) found an inverse cline in flowering time on the border between the unlimed d subplots of plots 8 and 9/1. There was also significant genetic differentiation at ISSR marker loci between populations either side of the boundary and genetic evidence of greatly reduced gene flow via pollen. Silvertown *et al.* (2005) suggested that reproductive isolation across the boundary between plots 8d and 9/1d had been reinforced by natural selection.

Reinforcement is a controversial evolutionary mechanism that may facilitate sympatric speciation. Few, if any, incontrovertible examples from the field are known (Servedio & Noor 2003). A critical requirement for reinforcement to occur is that hybrids between populations should have lower fitness than within-population crosses. This is yet to be tested at Park Grass, but seems highly likely given the very strong selection against alien genotypes demonstrated by Davies & Snaydon (1976).

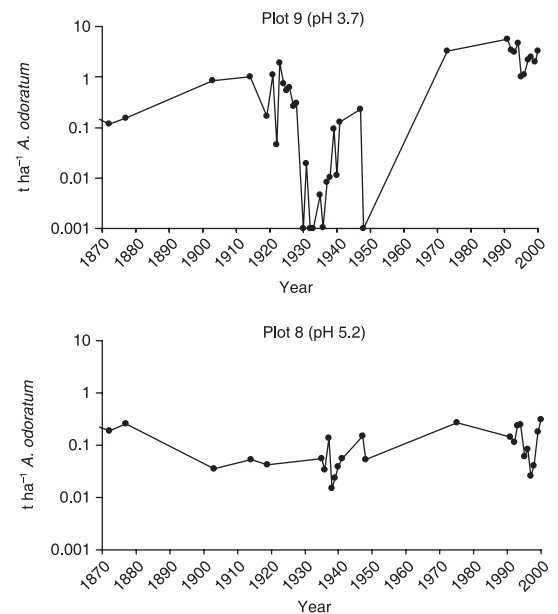


Fig. 7 Time series, 1870–2000, of *Anthoxanthum odoratum* abundance (t ha^{-1} dry matter) on plots with contrasting pH (measured 1995–2005).

Local adaptation has been tested in only one other plant species found on the plots, *Holcus lanatus*. Naylor *et al.* (1996) tested for the frequency of arsenate tolerance on different plots but found no correlation with soil conditions. In a glasshouse study, no differences between *H. lanatus* growing on a plot with (14/2c) or without (15c) N fertilizer was found for root growth, overwinter seedling survival or leaf shape (F. van denBerg, pers. comm.). Why one species, *A. odoratum*, should show local adaptation in many traits while another, *H. lanatus*, exposed to the same selection pressures does not is unclear. However, this situation is quite common (Bradshaw 1991), and Park Grass is well suited to its further investigation.

GENETIC DRIFT

Drift as well as selection may have taken place at Park Grass, especially on plots where effective population size (N_e) is restricted by population bottlenecks. In a fluctuating population, N_e is approximately equal to the harmonic mean of effective population size in each separate generation (Wright 1969). Low values have a highly disproportionate effect upon a harmonic mean, and so a population crash caused by drought has the potential to create a genetic bottleneck that will leave an imprint upon genetic structure long after the population has recovered its former abundance. Very acid plots, such as plot 9/2d (pH 3.8), have the greatest variance in biomass (Dodd *et al.* 1994b) and so ought to be the most susceptible to the loss of genetic variation through drift (Fig. 7).

Chloroplast microsatellite markers (Biss *et al.* 2003; Provan *et al.* 2004) were used to haplotype leaves of *Anthoxanthum odoratum* from archived hay samples

taken from the crops on Plots 8d (pH 5.2), 9/2d (pH 3.8) and 12d (pH 5.2) and from fresh leaves sampled from these plots in 2001. Between 1870 and 2001 haplotype diversity (measured by the Shannon information function H') more than halved on plot 9/2d, while on the less variable plots 8d and 12d it did not change significantly.

Fauna

The degree to which species in one trophic level influence the composition and abundance of species in other trophic levels is an active area of ecological research (Hunter & Price 1992; Shurin *et al.* 2006). Variation in plant species composition among plots has been used as a basis for investigating bottom-up (plant \rightarrow herbivore) effects at Park Grass. Morris (1992) sampled the Auchenorrhynca (leafhoppers) of 13 pairs of b and d subplots at Park Grass and found that the species composition of this group varied with amounts of applied N and soil pH. The species richness of Auchenorrhynca, though not their total abundance, was correlated positively with soil pH and negatively with N fertilization. Since plant and leafhopper species richness were affected by N and pH in the same direction, this suggests that plant species number may be the underlying cause of variation in herbivore species number, although Morris (1992) did not measure the direct correlation between these variables.

Edwards *et al.* (1976) did find a direct positive correlation between the species richness of plants and Collembola (springtails). Collembola are mostly detritivores, so the link between their diversity and plant species richness may be indirect. Edwards *et al.* (1976) found that the total spider fauna at Park Grass included species normally associated with several different grassland types, reflecting the heterogeneous nature of the vegetation. However, the effects of specific plot treatments on the surface invertebrates sampled by Edwards *et al.* (1976) were weaker than those affecting the Auchenorrhynca studied by Morris (1992).

The range of plant communities in the Park Grass Experiment allows hypotheses predicting how plant communities affect insect abundance and in particular the intensity of herbivory to be tested. Two different but not mutually exclusive hypotheses reflect observations that insect herbivore loads are reduced in more diverse plant assemblages (Root 1973). The 'resource concentration' hypothesis (RCH) predicts that insect herbivores are more likely to find and remain in areas where their host plants are concentrated in time and space. The 'enemies hypothesis' (EH) predicts that natural enemies of insects are expected to be more abundant in polycultures and thus suppress herbivore population densities more efficiently than in monocultures. Both predict higher herbivore densities in monoculture but differ in their predictions for polyphagous herbivores. In the 'microclimate hypothesis' differences in the structural complexity of vegetation affect microclimate and may alter herbivore abundance through lowered mortality rates or increased fecundity (Crawley 1983). The 'exploitation

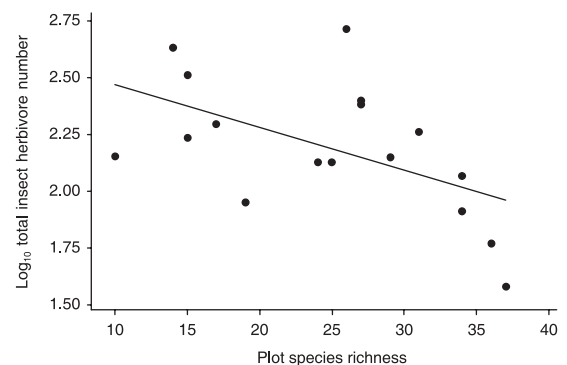


Fig. 8 Relationship between \log_{10} total insect herbivore number and species richness across plots ($F_{1,15} = 6.08$, $P < 0.05$; $R^2 = 29\%$).

ecosystem hypothesis' (EEH) developed by Oksanen *et al.* (1981) suggests that the intensity of herbivory varies systematically with productivity because of increasing top-down control at higher productivities, and predicts peak herbivore impact at intermediate levels. Thus many factors can affect insect herbivore impact. These are rarely addressed simultaneously in field studies, but this has been done for insect herbivores in the Park Grass Experiment by Heard (1999).

Heard (1999) used field observations of insect abundance and herbivore damage to pot individuals (phytometers) of five plant species (two grasses and three forbs), introduced into the plots to test the various hypotheses. As expected, results showed considerable variation in intensity of herbivory (0–74% leaf area) across plant species. In general, damage decreased with increasing neighbourhood plant species richness ($F_{1,16} = 5.11$, $P < 0.05$), consistent with EH. More detailed observations on *Ranunculus acris* showed that mean leaf damage per plot decreased with increased plant diversity (Shannon–Weiner index, $F_{1,12} = 6.32$, $P < 0.05$; $R^2 = 35\%$), also supporting EH. However, there was also partial support for EEH because increased plant productivity resulted in increased intensity of leaf damage although there was no sign of a hump in the relationship. In contrast, the effect on *Rumex acetosa* phytometers provided support for both the EH and RCH hypotheses. Insect sampling showed decreased herbivore abundance with increased plant species richness (Fig. 8) also consistent with EH. Whilst the study demonstrates systematic variation in invertebrate herbivory across community types, there was little evidence that one hypothesis satisfactorily accounts for the whole range of variation observed. The herbivores of some plant species obviously have highly individualistic responses to habitat variables. The impacts of plant species richness and productivity are clearly important, but the negative correlation between these parameters among Park Grass plots makes it difficult to separate their relative impacts and importance. Further analysis of the invertebrate food web structure may elucidate the impacts on the abundance and relationship of specific functional groups.

The soil fauna at Park Grass is strongly influenced by N treatment and pH and its variation among plots has ecosystem effects. In particular, earthworm density decreased with increasing levels of N application and with decreasing pH (Edwards & Lofty 1975). On soils with pH 4 and below, earthworms were absent. Earthworms perform an important ecosystem function by burying dead leaf material on the soil surface. The absence of earthworms and decreased microbial activity on the most acid plots (e.g. plot 9/2d), has led to the accumulation of a layer of undecomposed, peat-like organic matter on the soil surface. The failure of plant roots to penetrate from the organic layer into the mineral soil underneath appears to be the cause of the severe drought sensitivity of plant populations on plot 9/2d (Fig. 7).

Discussion

The Park Grass Experiment is unique, not just because of its duration but also because of the range of investigations that have been carried out there. Even though different investigators have pursued independent lines of enquiry, the cumulative result has the potential to tell us about more than the sum of the parts. Before a synthesis is attempted, what have we learned about the parts? First, that the plant communities in the experiment are at dynamic equilibrium (Silvertown 1987), where the average guild structure of the vegetation on a plot is determined by nutrient availability, particularly N, but also P and K, and lime, and is continually perturbed by climate (Silvertown *et al.* 1994). Although there is an equilibrium at the guild level, individual species within guilds change abundance and distribution among plots in a more dynamic manner which is at least partly related to species' life-history traits (Dodd *et al.* 1995; Silvertown *et al.* 2002). Some of the changes within plots may also be driven by changes in the availability of nutrients (Tilman 1982). For example, *Taraxacum officinale* responds strongly to K and liming (Tilman *et al.* 1999).

The potential for compositional change, some of which is held in check by regulatory processes, is further illustrated by the fact that seedlings of certain species turn up in plots (e.g. *Taraxacum officinale* and *Rumex acetosa* in 18d) where adults of the species are absent (G.R. Edwards, unpublished data). Even though seeds disperse across plot boundaries, this does not result in any strong spatial mass effect altering community composition at plot edges (Kunin 1998). Just as at the population level genetic differentiation between plots has occurred in spite of gene flow, differences between plots at the community level are maintained despite a flow of propagules between plots. Park Grass is perhaps the clearest example there is of the need to study community variability at a hierarchy of levels (Micheli *et al.* 1999). Second, we have learned that the Park Grass Experiment provides support for both the competitive exclusion and pool size hypotheses for determination of species density. Species richness is negatively correlated with total plot biomass, which is itself controlled mainly

by N (Crawley *et al.* 2005). One way in which biomass reduces species richness is by limiting seedling recruitment (see above). The pool size effect is seen most clearly in the effect of pH on species richness. Few species in the Rothamsted flora, other than *Anothoxanthum odoratum* and *Holcus lanatus*, can tolerate the high levels of plant-available aluminium found on the very acid plots. Decreases in soil pH lower species richness and operate additively with the effect of biomass (Silvertown 1980). These results are relevant to recent debates on the functional role of species diversity in plant communities. They indicate that in mesotrophic grasslands productivity will suppress species diversity rather than species diversity promoting productivity. Species diversity also has a minimal effect on the variance of biomass over years (Dodd *et al.* 1994b). Invertebrate communities at Park Grass merit further study, but the results discussed indicate that at least some members of all the major trophic levels, including predators (spiders), herbivores (leafhoppers) and detritivores (springtails) are plot-specific in their species distributions. In most cases the distribution of invertebrates is probably related to either the species composition or the physical structure of the vegetation. Either mechanism might cause climate-driven changes in the vegetation to propagate upwards through the food web. Voigt *et al.* (2003) found that higher trophic levels were more sensitive to climate than lower ones in grassland communities. Park Grass would be an ideal system in which to investigate tritrophic interactions and the degree of coupling between trophic levels over time.

In at least one very important case fertilizer treatment has had a direct effect. Lack of microbial activity and the absence of earthworms on the very acid soils given ammonium sulphate and not limed has led to the development of a surface mat of peat-like material that is a few centimetres thick. This mat is greatly affected by both freezing and drought which, in turn, has had an effect on the species growing on these soils. These are the two grasses, *Holcus lanatus* and *Anthoxanthum odoratum*, and their susceptibility to drought can, in some years, produce a population bottleneck (Fig. 7). The causal relationships between fertilizer treatment, soil pH, microbial activity and earthworms, drought and population bottlenecks illustrates one way, amongst many possible ways, that Park Grass studies lend themselves to synthesis (Fig. 9). The preliminary evidence on chloroplast haplotype variation presented in an earlier section suggests that population bottlenecks caused by drought on highly acidified soils may lead to loss of genetic variation through drift.

What the long-term consequences of this may be for plant population viability in a world where extreme climatic events are predicted to recur with increasing frequency (Meehl & Tebaldi 2004), we do not yet know. One scenario is that genetic bottlenecks can tip a population into an extinction vortex if effective population size (N_e) and census population size are coupled by positive feedback (Frankham 2005). The archive of hay samples at Park Grass contains genetic material

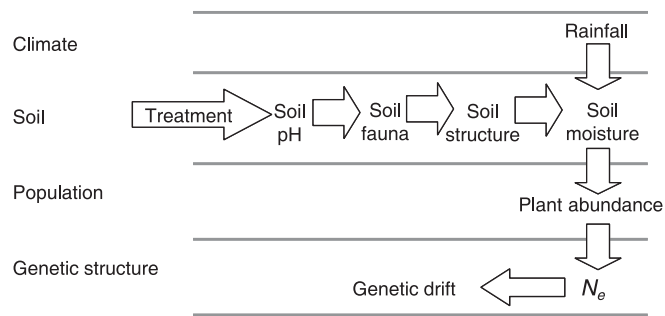


Fig. 9 A schematic model of the proposed mechanism by which climatic variation in rainfall, mediated by the influence of plot treatment on soil invertebrates and of these on soil structure and hence soil moisture, influence plant abundance and population genetic structure (N_e).

belonging to multiple populations that have gone locally extinct during the history of the experiment, offering the possibility that the genetics of recorded extinctions could be analysed retrospectively.

Synthesizing the results from another perspective, Park Grass is of increasing relevance to grassland management for nature conservation. The experiment now contains several grassland types recognized by the National Vegetation Classification, including remnants of the original MG5 hay meadow which is now a grassland type that is an object of conservation efforts in its own right. Nutrients and soil pH control community composition and species diversity and there are regulatory mechanisms that keep these in equilibrium, but these equilibria can be shifted to new positions. A changing climate could potentially do this because its effects can be amplified by interspecific competition. This implies the possibility of rapid community change in future, causing a decrease in species diversity.

Significant numbers of species have been lost from all Park Grass plots. We can account for a proportion of these losses through the effects of nutrient treatments, acidification and the traits that make certain species more vulnerable than others, but it is worrying that models do not predict reductions in species diversity as large as those actually observed since 1856. The general message must be that we do not yet fully comprehend the causes of local extinction in plants, even where the losses are as well documented as they are at Park Grass.

As already mentioned, the invertebrate fauna of Park Grass is little-studied, but even less is known about microbial and fungal communities. Old grasslands can harbour high diversities of waxcap fungi (Griffith *et al.* 2004). G. W. Griffith (pers. comm.) found patterns of fruiting of macrofungi in 2004 that were remarkably similar to those reported by Gilbert in (1875); including the presence of several *Hygrocybe* spp. on untreated plots. An increasing body of experimental evidence supports the view that the fungal and plant communities in grassland are closely coupled (e.g. Vandenkoornhuyse *et al.* 2003). Three-way interactions among plants, their fungal mutualists and natural enemies could theoretically influence plant community dynamics (Bennett *et al.* 2006), although whether the effects are strong enough to be detected in the

presence of the kind of powerful effects of soil nutrients and climate shown at Park Grass remains to be seen.

The first 150 years of the Park Grass Experiment have taught us a great deal, but there are still whole parts of the communities it contains, like the fungi, that have yet to be studied and there are many unanswered questions. These gaps notwithstanding, if ecosystems are like multidimensional jigsaw puzzles, then Park Grass is beginning to show us what the picture on the lid of the box must look like. This should provide a useful crib for those trying to solve similar ecological puzzles in other grassland ecosystems.

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References

- Arnold, P.W., Hunter, F. & Gonzalez-Fernandez, P. (1976) Long-term grassland experiments at Cockle-Park. *Annales Agronomiques*, **27**, 1027–1042.
- Bender, E.A., Case, T.J. & Gilpin, M.E. (1984) Perturbation experiments in community ecology. *Ecology*, **65**, 1–13.
- Bennett, A.E., Alers-Garcia, J. & Bever, J.D. (2006) Three-way interactions among mutualistic mycorrhizal fungi, plants, and plant enemies: hypotheses and synthesis. *American Naturalist*, **167**, 141–152.
- Biss, P., Freeland, J., Silvertown, J., McConway, K.J. & Lutman, P. (2003) Successful amplification of rice chloroplast microsatellites from century-old grass samples from the Park Grass Experiment. *Plant Molecular Biology Reporter*, **21**, 249–257.
- Blake, L., Goulding, K.W.T., Mott, C.J.B. & Johnston, A.E. (1999) Changes in soil chemistry accompanying acidification over more than 100 years under woodland and grass at Rothamsted Experimental Station, UK. *European Journal of Soil Science*, **50**, 401–412.
- Bradshaw, A.D. (1991) The Croonian Lecture, 1991 – Genostasis and the limits to evolution. *Philosophical Transactions of the Royal Society of London Series B, Biology Sciences*, **333**, 289–305.

- Cashen, R.O. (1947) The influence of rainfall on the yield and botanical composition of permanent grass at Rothamsted. *Journal of Agricultural Science*, **37**, 1–10.
- Craine, J.M. (2005) Reconciling plant strategy theories of Grime and Tilman. *Journal of Ecology*, **93**, 1041–1052.
- Crawley, M.J. (1983) *Herbivory. The Dynamics of Animal–Plant Interactions*. Blackwell Scientific Publications, Oxford.
- Crawley, M.J., Johnston, A.E., Silvertown, J., Dodd, M., de Mazancourt, C., Heard, M.S., Henman, D.F. & Edwards, G.R. (2005) Determinants of species richness in the Park Grass experiment. *American Naturalist*, **165**, 179–192.
- Davies, M.S. & Snaydon, R.W. (1973a) Physiological differences among populations of *Anthoxanthum odoratum* L. on the Park Grass Experiment, Rothamsted. I. Response to calcium. *Journal of Applied Ecology*, **10**, 33–45.
- Davies, M.S. & Snaydon, R.W. (1973b) Physiological differences among populations of *Anthoxanthum odoratum* L. on the Park Grass Experiment, Rothamsted. II. Response to aluminium. *Journal of Applied Ecology*, **10**, 47–55.
- Davies, M.S. & Snaydon, R.W. (1974) Physiological differences among populations of *Anthoxanthum odoratum* L. on the Park Grass Experiment, Rothamsted. III. Response to phosphate. *Journal of Applied Ecology*, **11**, 699–708.
- Davies, M.S. & Snaydon, R.W. (1976) Rapid population differentiation in a mosaic environment. III. Measures of selection pressures. *Heredity*, **36**, 59–66.
- Dodd, M.E., Silvertown, J., McConway, K., Potts, J. & Crawley, M. (1994a) Application of the British National Vegetation Classification to the communities of the Park Grass Experiment through time. *Folia Geobotanica and Phytotaxonomica*, **29**, 321–334.
- Dodd, M.E., Silvertown, J., McConway, K., Potts, J. & Crawley, M. (1994b) Stability in the plant communities of the Park Grass Experiment: the relationships between species richness, soil pH and biomass variability. *Philosophical Transactions of the Royal Society of London Series B, Biology Sciences*, **346**, 185–193.
- Dodd, M., Silvertown, J., McConway, K., Potts, J. & Crawley, M. (1995) Community stability: a 60-year record of trends and outbreaks in the occurrence of species in the Park Grass Experiment. *Journal of Ecology*, **83**, 277–285.
- Dunnett, N.P. & Grime, J.P. (1999) Competition as an amplifier of short-term vegetation responses to climate: an experimental test. *Functional Ecology*, **13**, 388–395.
- Edwards, C.A., Butler, C.G. & Lofty, J.R. (1976) The invertebrate fauna of the Park Grass plots. II. Surface fauna. *Rothamsted Experimental Station Report For*, **1975 Part 2**, 63–89.
- Edwards, C.A. & Lofty, J.R. (1975) The invertebrate fauna of the Park Grass plots. I. Soil fauna. *Rothamsted Experimental Station Report For*, **1974 Part 2**, 133–154.
- Edwards, G.R., Bourdot, G.W. & Crawley, M.J. (2000) Influence of herbivory, competition and soil fertility on the abundance of *Cirsium arvense* in acid grassland. *Journal of Applied Ecology*, **37**, 321–334.
- Elberse, W.T., van den Bergh, J.P. & Dirven, J.G.P. (1983) Effects of use of mineral supply on the botanical composition and yield of old grassland on heavy-clay soil. *Netherlands Journal of Agricultural Science*, **31**, 63–88.
- Frankham, R. (2005) Genetics and extinction. *Biological Conservation*, **126**, 131–140.
- Gilbert, J.H. (1875) Note on the occurrence of ‘fairy rings’. *Journal of the Linnean Society – Botany*, **15**, 17–24.
- Goulding, K.W.T., Bailey, N.J., Bradbury, N.J., Hargreaves, P., Howe, M., Murphy, D.V., Poulton, P.R. & Willison, T.W. (1998) Nitrogen deposition and its contribution to nitrogen cycling and associated soil processes. *New Phytologist*, **139**, 49–58.
- Griffith, G.W., Bratton, J.L. & Easton, G. (2004) Charismatic megafungi: the conservation of waxcap grasslands. *British Wildlife*, **15**, 31–43.
- Grime, J.P., Hodgson, J.G. & Hunt, R. (1988) *Comparative Plant Ecology*. Allen & Unwin, UK.
- Heard, M.S. (1999) *The Role of Invertebrate Herbivory and Plant Competition in Structuring Mesic Grassland Communities*, PhD Thesis, Imperial College, University of London, Silwood Park.
- Henman, D.F. (2001) *Rapid Transient Dynamics In Response To Altered Nutrient Inputs At The Park Grass Experiment*. PhD Thesis, Imperial College, University of London, Silwood Park.
- Hunter, M.D. & Price, P.W. (1992) Playing chutes and ladders: heterogeneity and the relative roles of bottom-up and top-down forces in natural communities. *Ecology*, **73**, 724–732.
- Huston, M.A. (1997) Hidden treatments in ecological experiments: Re-evaluating the ecosystem function of biodiversity. *Oecologia*, **110**, 449–460.
- Jenkinson, D.S. (1990) The turnover of organic carbon and nitrogen in soil. *Philosophical Transactions of the Royal Society of London*, **329**, 361–368.
- Jenkinson, D.S., Harkness, D.D., Vance, E.D., Adams, D.E. & Harrison, A.F. (1992) Calculating net primary production and annual input of organic-matter to soil from the amount and radiocarbon content of soil organic-matter. *Soil Biology and Biochemistry*, **24**, 295–308.
- Jones, C., McConnel, C., Coleman, K., Cox, P., Falloon, P., Jenkinson, D. & Powlson, D. (2005) Global climate change and soil carbon stocks; predictions from two contrasting models for the turnover of organic carbon in soil. *Global Change Biology*, **11**, 154–166.
- Keddy, P.A. (2004) The ecology of plants. *Quarterly Review of Biology*, **79**, 55–59.
- Kunin, W.E. (1998) Biodiversity at the edge: a test of the importance of spatial ‘mass effects’ in the Rothamsted Park Grass experiments. *Proceedings of the National Academy of Sciences USA*, **95**, 207–212.
- Lawes, J.B. & Gilbert, J.H. (1859/1893) Report of experiments with different manures on permanent meadow land. *Journal of the Royal Agricultural Society of England*, **XIX**, Part II and **XX**, Parts I & II, 1–111.
- Linhart, Y.B. & Grant, M.C. (1996) Evolutionary significance of local genetic differentiation in plants. *Annual Review of Ecology and Systematics*, **27**, 237–277.
- Loreau, M., Naeem, S., Inchausti, P., Bengtsson, J., Grime, J.P., Hector, A., Hooper, D.U., Huston, M.A., Raffaelli, D., Schmid, B., Tilman, D. & Wardle, D.A. (2001) Ecology – Biodiversity and ecosystem functioning: current knowledge and future challenges. *Science*, **294**, 804–808.
- May, R.M. (1973) *Stability and Complexity in Model Ecosystems*. Princeton University Press, Princeton, NJ.
- McGrath, S.P., Zhao, F.Z. & Blake-Kalff, M.M.A. (2002) History and outlook for sulphur fertilizers in Europe. *Proceedings No. 497. The International Fertilizer Society*, York, UK, 1–24.
- Meehl, G.A. & Tebaldi, C. (2004) More intense, more frequent, and longer lasting heat waves in the 21st century. *Science*, **305**, 994–997.
- Micheli, F., Cottingham, K.L., Bascompte, J., Bjornstad, O.N., Eckert, G.L., Fischer, J.M., Keitt, T.H., Kendall, B.E., Klug, J.L. & Rusak, J.A. (1999) The dual nature of community variability. *Oikos*, **85**, 161–169.
- Miller, T.E., Burns, J.H., Munguia, P., Walters, E.L., Kneitel, J.M., Richards, P.M., Mouquet, N. & Buckley, H.L. (2005) A critical review of twenty years’ use of the resource-ratio theory. *American Naturalist*, **165**, 439–448.
- Morris, M.G. (1992) Responses of Auchenorrhynca (Homoptera) to fertilizer and liming treatments at Park Grass, Rothamsted. *Agriculture Ecosystems and Environment*, **41**, 263–283.
- Naylor, J., McNair, M.R., Williams, E.N.D. & Poulton, P.R. (1996) A polymorphism for phosphate-uptake arsenate tolerance in *Holcus lanatus* L – Is there a correlation with edaphic or environmental factors? *Heredity*, **77**, 509–517.

- Oksanen, L., Fretwell, S.D., Arruda, J. & Niemela, P. (1981) Exploitation ecosystems in gradients of primary productivity. *American Naturalist*, **118**, 240–261.
- Provan, J., Biss, P.M., McMeel, D. & Mathews, S. (2004) Universal primers for the amplification of chloroplast microsatellites in grasses (Poaceae). *Molecular Ecology Notes*, **4**, 262–264.
- Rodwell, J.S. (1992) *British Plant Communities*, Vol. 3. *Grasslands and Montane Communities*. Cambridge University Press, Cambridge.
- Root, R.B. (1973) Organization of a plant–arthropod association in simple and diverse habitats: the fauna of collards (*Brassica oleracea*). *Ecological Monographs*, **43**, 95–124.
- Servedio, M.R. & Noor, M.A.F. (2003) The role of reinforcement in speciation. Theory and data. *Annual Review of Ecology Evolution and Systematics*, **34**, 339–364.
- Shurin, J.B., Gruner, D.S. & Hillebrand, H. (2006) All wet or dried up? Real differences between aquatic and terrestrial food webs. *Proceedings of the Royal Society B, Biology Sciences*, **273**, 1–9.
- Silvertown, J. (1980) The dynamics of a Grassland Ecosystem – Botanical equilibrium in the Park Grass Experiment. *Journal of Applied Ecology*, **17**, 491–504.
- Silvertown, J. (1987) Ecological stability: a test case. *American Naturalist*, **130**, 807–810.
- Silvertown, J., Dodd, M.E., McConway, K., Potts, J. & Crawley, M. (1994) Rainfall, biomass variation, and community composition in the Park Grass Experiment. *Ecology*, **75**, 2430–2437.
- Silvertown, J., McConway, K.J., Hughes, Z., Biss, P., Macnair, M. & Lutman, P. (2002) Ecological and genetic correlates of long-term population trends in the Park Grass Experiment. *American Naturalist*, **160**, 409–420.
- Silvertown, J., Servaes, C., Biss, P. & Macleod, D. (2005) Reinforcement of reproductive isolation between adjacent populations in the Park Grass Experiment. *Heredity*, **95**, 198–205.
- Snaydon, R.W. (1970) Rapid population differentiation in a mosaic environment. I. The response of *Anthoxanthum odoratum* populations to soils. *Evolution*, **24**, 257–269.
- Snaydon, R.W. & Davies, M.S. (1972) Rapid population differentiation in a mosaic environment. II. Morphological variation in *Anthoxanthum odoratum* L. *Evolution*, **26**, 390–405.
- Snaydon, R.W. & Davies, M.S. (1976) Rapid population differentiation in a mosaic environment. IV. Populations of *Anthoxanthum odoratum* L. at sharp boundaries. *Heredity*, **36**, 9–25.
- Snaydon, R.W. & Davies, T.M. (1982) Rapid divergence of plant populations in response to recent changes in soil conditions. *Evolution*, **36**, 289–297.
- Stevens, C.J., Dise, N.B., Mountford, J.O. & Gowing, D.J. (2004) Impact of nitrogen deposition on the species richness of grasslands. *Science*, **303**, 1876–1879.
- Thurston, J.M., Williams, E.D. & Johnston, A.E. (1976) Modern developments in an experiment on permanent grassland started in 1856: effects of fertilisers and lime on botanical composition and crop and soil analyses. *Annales Agronomiques*, **27**, 1043–1082.
- Tilman, D. (1982) *Resource Competition and Community Structure*. Princeton University Press, Princeton.
- Tilman, D. (1987) Secondary succession and the pattern of plant dominance along experimental nitrogen gradients. *Ecological Monographs*, **57**, 189–214.
- Tilman, D., Dodd, M.E., Silvertown, J., Poulton, P.R., Johnston, A.E. & Crawley, M.J. (1994) The Park Grass Experiment: insights from the most long-term ecological study. *Long-term Experiments in Agricultural and Ecological Sciences* (eds R.A. Leigh & A.E. Johnston), pp. 287–303. CAB International, Wallingford, UK.
- Tilman, D. & Downing, J.A. (1994) Biodiversity and stability in grasslands. *Nature*, **367**, 363–365.
- Tilman, E.A., Tilman, D., Crawley, M.J. & Johnston, A.E. (1999) Biological weed control via nutrient competition: potassium limitation of dandelions. *Ecological Applications*, **9**, 103–111.
- Vandenkoornhuysen, P., Ridgway, K.P., Watson, I.J., Fitter, A.H. & Young, J.P.W. (2003) Co-existing grass species have distinctive arbuscular mycorrhizal communities. *Molecular Ecology*, **12**, 3085–3095.
- Voigt, W., Perner, J., Davis, A.J., Eggers, T., Schumacher, J., Bahrmann, R., Fabian, B., Heinrich, W., Kohler, G., Lichter, D., Marstaller, R. & Sander, F.W. (2003) Trophic levels are differentially sensitive to climate. *Ecology*, **84**, 2444–2453.
- Warneke, T., Croudace, I.W., Warwick, P.E. & Taylor, R.N. (2002) A new ground-level fallout record of uranium and plutonium isotopes for northern temperate latitudes. *Earth and Planetary Science Letters*, **203**, 1047–1057.
- Warren, R.G. & Johnston, A.E. (1964) The Park Grass Experiment. *Rothamsted Experimental Station Report for 1963*, 240–262.
- Warren, R.G., Johnston, A.E. & Cooke, G.W. (1965) Changes in the Park Grass Experiment. *Rothamsted Experimental Station Report for 1964*, 224–228.
- Williams, E.D. (1978) Botanical composition of the Park Grass plots at Rothamsted, 1856–1976. *Rothamsted Experimental Station*, Harpenden, UK.
- Wright, S. (1969) *Evolution and Genetics of Populations*, Vol. 2. The Theory of Gene Frequencies. Chicago University Press, Chicago.
- Zhao, F.J., Spiro, B., Poulton, P.R. & McGrath, S.P. (1998) Use of sulfur isotope ratios to determine anthropogenic sulfur signals in a grassland ecosystem. *Environmental Science and Technology*, **32**, 2288–2291.

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Supplementary material

The following supplementary material is available online from www.Blackwell-Synergy.com

Table S1 Manorial history of the Park Grass plots, Rothamsted 1856–2005 and the pH (in water) of the top 23 cm of soil in 1995/2005.