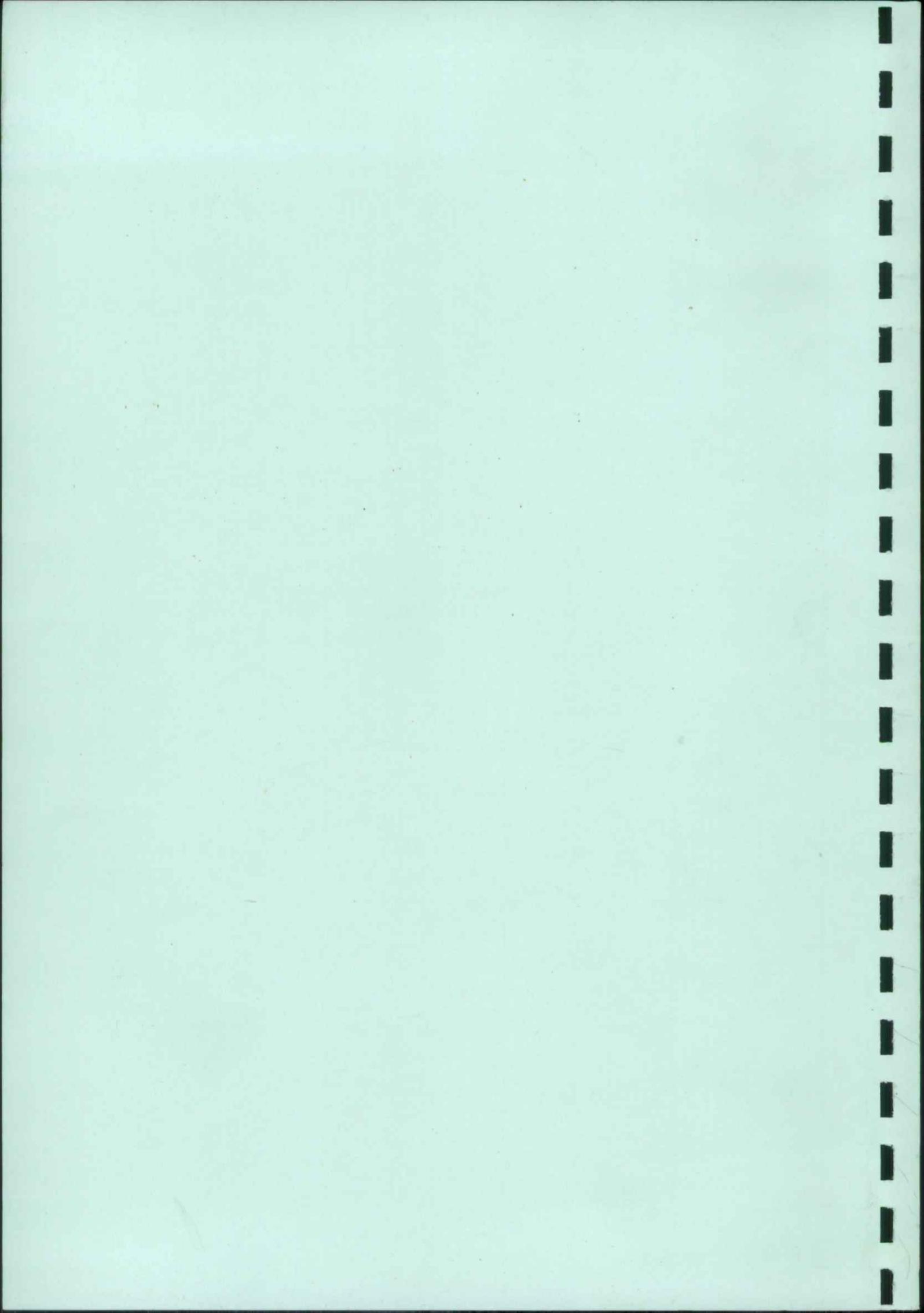


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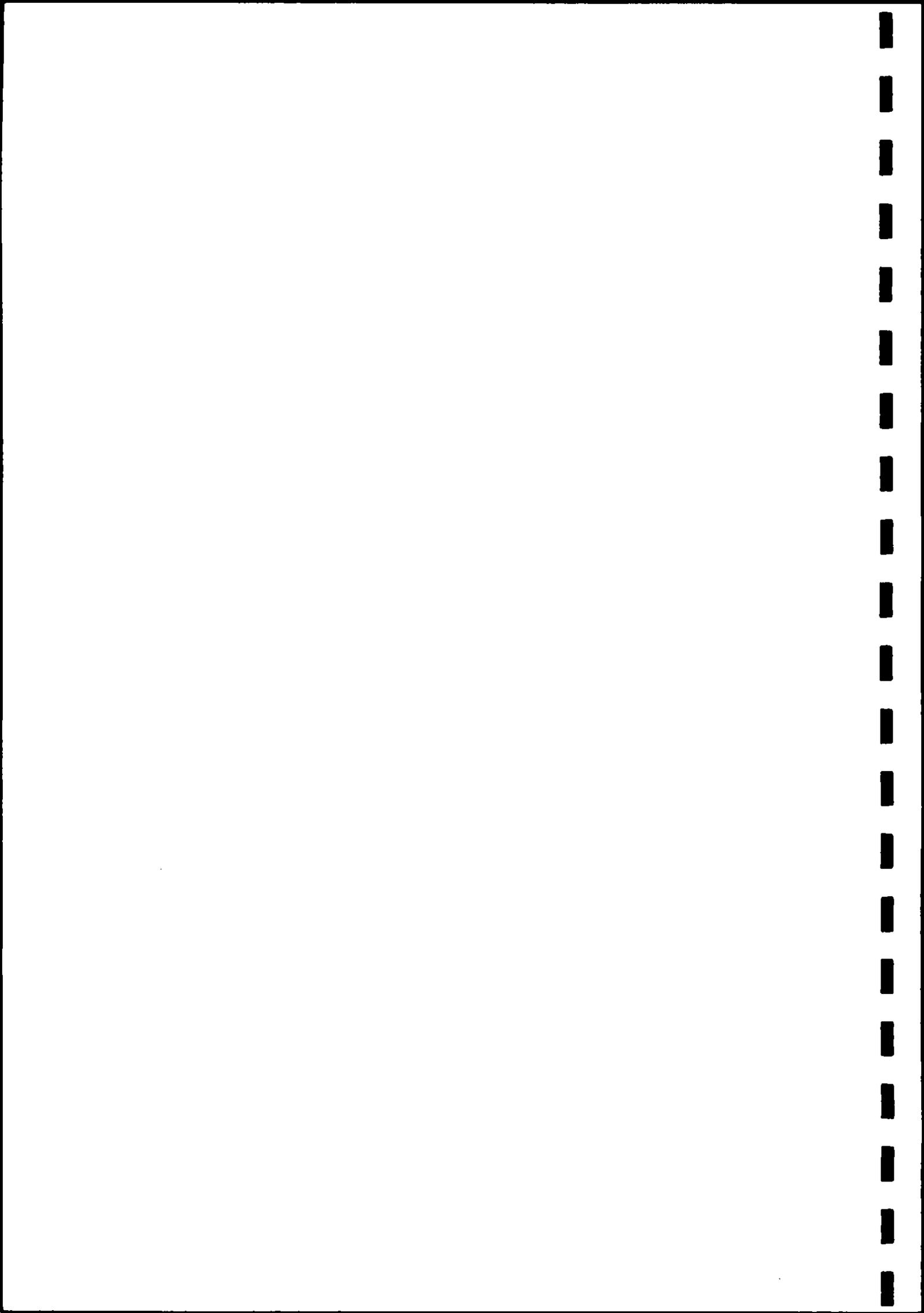
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EFFECTS OF ACID PRECIPITATION ON VEGETATION IN WALES

T W ASHENDEN

Bangor Research Station
Penrhos Road
Bangor
Gwynedd

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1 SUMMARY

The work described is the first phase of a study to assess the potential threat of acidic pollutants to vegetation in Wales. In this part of the project, the primary objective is to determine the effects of simulated wet acid deposition on a range of plant species growing on a range of Welsh soil types to identify vulnerable species.

Five species were grown on soils ranging from those considered to be sensitive, to those considered to be non-sensitive to acid precipitation. The species were exposed to simulated rainfall treatments at pH's of 2.5, 3.5, 4.5 and 5.6. The species investigated were Lolium perenne L. (perennial ryegrass), Trifolium repens L. (white clover), Hordeum vulgare L. (winter barley), Betula pendula Roth. (silver birch) and Picea sitchensis (Bong.) Carr. (Sitka spruce).

Ryegrass, white clover and winter barley were harvested after 22-25 weeks of exposure to the polluted rainwater treatments and dry weights obtained for different plant fractions. For ryegrass, there were higher yields for root and shoot dry weight fractions but not total plant dry weight at the most acid (pH 2.5) treatment. In contrast, white clover showed lower shoot dry weights which resulted in reduced whole-plant weights at the pH 2.5 treatment. Winter barley proved to be the species most sensitive to the acid rainwater treatments and showed 30% reduction in the yields of all dry weight fractions and the numbers of ears produced at pH's 3.5 and 2.5 as compared to the "unpolluted" pH 5.6 rainwater treatment. Mathematical equations were developed which related the dry weights of plant fractions of winter barley with the pH of artificial rain applied.

For birch and Sitka spruce the rainfall treatments are being continued but, after 1 year of exposure, measurements of plant height were taken. These revealed no effects of treatments on Sitka spruce but a greater height of birch seedlings in the most acid treatment. For all five species studied, it was noted that there was a large within-treatment variation and this may have reduced the magnitudes of differences between the treatments.

In addition to the main experiment, several smaller studies are described. The first was a germination trial for winter barley which revealed that the germination rate of this species was not affected by the acidity of rainfall. The second investigated the possibility of an interaction effect on yield of defoliation and acidity of rainfall using Lolium perenne as the plant material. This study revealed that the yields of Lolium perenne were unaffected by the pH of rainfall treatments. The final study assessed the potential for discriminating between the foliage of plants subjected to different rainfall acidities using radiometric techniques. Birch was used as the plant material and the technique was found to be extremely good at discriminating between all four acidities of the rainwater treatments.

The results obtained are discussed in relation to the acidities of rainfall found at sites in Wales and those aspects of the work requiring further investigation are identified.

2 INTRODUCTION

The possible threat of ambient levels of acidic pollutants to natural ecosystems in Britain has been a matter of growing concern in scientific circles for the past decade (Bleasdale, 1973; Ashenden & Mansfield, 1977). More recently, the problem has been highlighted by extensive media coverage; reports of disappearing fish stocks in rivers and lakes, dying trees and reductions in crop yields as a result of 'acid rain' have caused much public concern. This project was initiated to assess the potential threat of acidic pollutants to natural vegetation and crops growing in Wales.

Sulphur dioxide (SO_2) and nitrogen oxides (NO_x) are the most commonly occurring gaseous pollutants in Britain and they are toxic to plants when present separately (see reviews by Law & Mansfield, 1982; Bell, 1982) and may act synergistically in combination (Ashenden & Mansfield, 1978; Ashenden & Williams, 1980; Ormrod, 1982). After release into the atmosphere, SO_2 and NO_x are partially hydrolysed to form the secondary pollutants sulphuric and nitric acids. In these forms, the pollutants dissolve in clouds or remain as an aerosol and may be transferred large distances before they are deposited through rain and snow. The time taken for the removal processes results in a much wider distribution than for primary pollutants. Thus areas with heavy rainfall may be subjected to large amounts of secondary pollutant deposition even though they are remote from major industrial sources.

Without additions of pollutants, rainwater would be slightly acidic since water in equilibrium with atmospheric concentrations of carbon dioxide has a pH of 5.6 ($2.5 \mu \text{Equiv. H}^+ \text{l}^{-1}$). Thus, by strict definition, 'acid rain' is snow, hail or rain which is more acid (*viz.* lower pH) than pH 5.6. However, the term is now usually used to incorporate both wet and dry depositions of acid pollutants. Since vegetation in Britain is unlikely to be subjected to one form of pollution without the other, this broader use of the term is more useful and realistic when evaluating the effects of pollutants on the environment. However, an understanding of the separate and interactive effects on vegetation of the two forms of deposition is equally important since the balance of wet to dry deposition differs markedly for different regions (contrast rural with urban areas).

The effects of gaseous acidic pollutants on plant growth have been the subject of many laboratory investigations and it is known that low concentrations of SO_2 may cause substantial reductions in the yields of plants (Bell & Clough, 1973; Ashenden, 1978). Indeed, reductions of over 60% have been reported for *Lolium perenne* L. in overwinter fumigations with as little as $43 \mu\text{g}\cdot\text{m}^{-3} \text{SO}_2$ (Bell, Rutter & Retton, 1979) and such a concentration is comparable with that found in most agricultural areas of the UK (Warren Spring, 1980; Fowler & Cape, 1982). Similarly, low concentrations of NO_2 have been found to cause reductions in the yields of some species (Ashenden, 1979a), affect physiological processes (Capron & Mansfield, 1976; Ashenden 1979b) and alter plant enzyme levels (Wellburn *et al.*, 1976). Moreover, it has been shown that when SO_2 and NO_2 are present in the atmosphere together they may act synergistically in causing visible plant injury (Tingey & Reinert, 1975) or plant yield reductions (Ashenden & Mansfield, 1978). Hence the necessity of considering pollutant mixtures when defining the potential threat of pollution to an environment.

In the form of wet deposits, the most publicised effects of acidic pollutants are those on lakes and streams. Gradual acidification of freshwaters in Scandinavia and America has been linked with an increase in algae, a decrease in zooplankton and the death of fish (see Hendry et al., 1976; Schofield, 1976). There have been recent reports in the media of similar acidification and declining fish populations for some lakes and streams in Wales. However, whether these changes are primarily caused by direct acid precipitation or afforestation is still a matter for debate. It is known that as rain passes through forest canopies it washes down dry deposits of pollutants and leaches nutrients from within tree tissues thus increasing in acidity prior to reaching the ground (Lakhani & Miller, 1978; Mayer & Ulrich, 1978; Hornung, work funded by the Welsh Office).

The direct effects of wet acid deposition on vegetation are not well defined. Scandinavian governments claim that declining conifer production in southern Sweden and Norway over the past two decades is a result of acid rain and snow but, in a comprehensive study of these regions, Abrahamsen et al. (1976) were unable to obtain firm evidence to support this view. However, they did find an effect of pH on the germination and establishment of spruce in laboratory experiments using realistic levels of acidity. Harcourt & Farrar (1980) have shown simulated acid rain to reduce the growth of Phaseolus vulgaris L. and Ferrebough (1976) found effects on the histology of leaves but, in both of these studies the levels of acidity needed to obtain an effect were unrealistic (pH 2.5 and below) in terms of the acidities of rainfall normally measured in field situations. At slightly higher pH's of 3.1 and 3.4 several authors have found simulated rain to induce leaf lesions in both woody and herbaceous species (Evans, Gmur & Da Costa, 1977 and 1978; Jacobson & van Leuken, 1977) and, recently, additions of simulated acid rain of pH 4.0-4.1 to field-grown crops have been found to cause yield reductions (Evans et al., 1982 and 1983).

Most laboratory experiments aimed at assessing the effects of wet acid deposition on plants have been conducted over short periods and it has been argued that long-term effects are more critical. Ferguson & Lee (1980) have suggested that the disappearance of Sphagnum spp. from the blanket peat of the southern Pennines during the past 200 years is a result of acid rain. Similarly, it was suggested by Ulrich (cited in a review by Pearce, 1982) that extensive damage and tree death in thousands of hectares of forests in Germany over the past eight years was due to wet acid deposition although, more recently, it has been argued that ozone is at least partially responsible for the damage there (see Dunnett, 1983). In more controlled conditions, Proctor (1983) found decreased fruit production in apple trees the year following two consecutive years of applying simulated acid rain of pH's 3.0 and 4.0 compared with a control of pH 5.6. He suggested some sort of carry over mechanism of injury similar to that found the following year after winter fumigations of trees with SO₂.

3 OBJECTIVES

The overall objective of this project is to assess the potential threat of acidic depositions of pollutants to the vegetation in Wales. The first phase of the work has been confined to studies on the effects of purely wet acidic pollutants on plants grown under controlled conditions. In this phase, the primary objective has been to determine any effects of wet acid deposition on a range of plant species growing on a range of Welsh soil types to allow the identification of any vulnerable plant species and/or soils. Supplementary studies have aimed to determine any interactive effects on plants of wet acid deposition with other environmental stresses commonly exerted on vegetation in Wales, e.g. sulphur dioxide pollution and defoliation.

The final objective has been to conduct pilot studies to identify any other plant responses which may alter in relation to differences in the concentrations of wet acids being deposited.

It is hoped that the results in phase 1 of the project will allow the development of a strategy for conducting a field survey to identify those areas of Wales which are being adversely affected by acidic pollutants. The object of phase 2 would be to implement this survey.

4 SCREENING SPECIES GROWING ON DIFFERENT WELSH SOILS

This has been the main study in phase 1 of the project. The aim is to find a higher plant species which can act as an indicator of wet acid deposition. To achieve this aim a range of species have been exposed to simulated rainfall at different acidities under controlled conditions. Growth responses are measured and related to rainfall acidity. In a field situation, growth responses could be differentially affected by soil conditions and thus the species were screened growing on a range of Welsh soil types.

4.1 The exposure system

This was a polythene tunnel which was erected on site at the ITE Bangor Research Station. It was divided internally by polythene sheeting into four treatment bays on each side of a central path, to provide a total of eight treatment bays. This allowed duplicate blocks of four different simulated rain treatments. Each tray was fitted with three "eintal" spray nozzles which were positioned to give the best possible distribution of droplets over the ground area. Treatment solutions were made up in 120 litre tanks and pumped to the exposure bays from an adjacent building.

4.2 The soils

A range of local soil types was collected ranging from those considered to be sensitive (by leaching of nutrients or release of aluminium) to those considered to be non-sensitive to acid precipitation. The soil types were: Newport series and Newborough dune sand (low cation exchange capacity and thus sensitive); Denbigh and Arfon series (intermediate sensitivity); Ddol and Cottam series (non-sensitive); Manod series (high levels of aluminium on exchange complex); and John Innes No.2 potting compost (control soil). All soils were collected at a depth of between 15 and 45 cm from undisturbed sites (see Table 1). At Bangor, the soils were thoroughly mixed, large stones were removed by hand and samples were taken for determination of pH and ammonium acetate extractable cations following the methods of Allen (1974). The ammonium acetate extracts were analysed for sodium, potassium, calcium, magnesium and manganese using a Perkin-Elmer atomic absorption spectrophotometer.

4.3 Plant material

The size of the exposure system and the large range of soils limited the number of species which could be studied to just five in the first experiment. It was decided to choose one species from each of five broad categories: a cereal, a grass, a broad-leaved herb, a deciduous tree seedling and a coniferous tree seedling. Winter varieties of Hordeum vulgare L. (barley) are the most economically important cereal grown in Wales and thus a recommended variety, Igr1, was chosen as the cereal. For the grass, it was decided to use the Aberystwyth S23 variety of Lolium perenne L. (perennial ryegrass) since this has been widely used in studies of atmospheric gaseous pollutants. Trifolium repens L. var. grasslands hula (white clover) was chosen as the broadleaved herb because of its economic importance in improving

Table 1 Types of soil used and their sources

Soil Type	Expected Sensitivity to acid leaching	Brief Soil Description	Source (Grid Reference)	Vegetation at source
NEWPORT SERIES	SENSITIVE	BROWN SAND	SJ 106 717	Rough pasture
NEWBOROUGH SAND	SENSITIVE	DUNE SAND	SH 432 640	Dune top vegetation
DENBIGH SERIES	INTERMEDIATE SENSITIVITY	BROWN EARTH	SH 772 574	Beech woodland
ARFON SERIES	INTERMEDIATE SENSITIVITY	BROWN EARTH	SH 608 686	Old pasture
DDOL SERIES	NON-SENSITIVE	CALCAREOUS BROWN SAND	SJ 125 718	Young Sycamore
COTTAM SERIES	NON-SENSITIVE	BROWN EARTH WITH GLEYING	SH 619 813	Scrub/rough pasture
MANOD SERIES	SENSITIVE WITH HIGH ALUMINIUM	BROWN PODZOLIC SOIL	SH 757 577	Beech woodland
JOHN INNES NO. 2	NON-SENSITIVE	POTTING COMPOST	-	-

grassland swards. Betula pendula Roth. (silver birch) was chosen as a typical, common, deciduous tree species and the seed for this study was taken from a local tree. As a coniferous tree species, Picea sitchensis (Bong.) Carr. (sitka spruce) was chosen and a British strain of seed was obtained from the Forestry Commission.

4.4 Experimental procedure

Seedlings of the five species were raised in trays of John Innes potting compost and transferred to 7.5 cm diameter pots of the different soils in December, 1983. There were 15 replicates of each species for each soil x rainwater treatment making a total of 2400 pots in the experiment. The pots were placed in random positions within each treatment bay and left for four weeks to become established prior to the start of rainwater treatments on 18 January, 1984. Subsequently, simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6 (made up by additions of sulphuric and nitric acids in a ratio of 7:3 by volume to tapwater) was applied at a rate of 3 cm per week.

Vigorous growth of winter barley, perennial ryegrass and white clover during the early summer meant that these plants were in danger of becoming pot-bound and were beginning to shade each other. Therefore, they were harvested at 21 weeks for winter barley, 23 weeks for perennial ryegrass and 24 weeks for white clover after the start of the rainwater treatments. At harvest, the roots were washed free of soil and plants were separated into different portions for dry weight determination.

Small samples of soil were taken from each pot when the plants were harvested. The samples for each rainwater treatment for each species were bulked and thoroughly mixed. Subsequently, duplicate subsamples were taken and determinations of pH made. For the soils from the pots of clover, ammonium acetate extracts were obtained and cations determined as for the original soils described in 4.2. These analyses were made to allow at least some comparison to be made of soil nutrient status before and after the experiment.

In order to determine any direct effects of the treatments on plant nutrient status, the white clover plants grown on John Innes compost were subjected to further analysis. The leaves of the plants were separated, ground in a mill and ashed at 500°C in a furnace to determine percentage ash contents. The soluble fractions of the ashed samples were taken up in hydrochloric acid for cation determinations (Allen, 1974). The resultant solutions were analysed for total sodium, potassium, calcium, magnesium, manganese and aluminium using a Perkin-Elmer atomic absorption spectrophotometer.

The slow rates of growth of birch and Sitka spruce have allowed the exposures to continue to the present time and they will remain in the different rainwater treatments until the end of summer 1985. However, non-destructive estimates of growth in the form of simple height measurements were obtained for both of these species in January 1985 - one year after the start of the rainwater treatments.

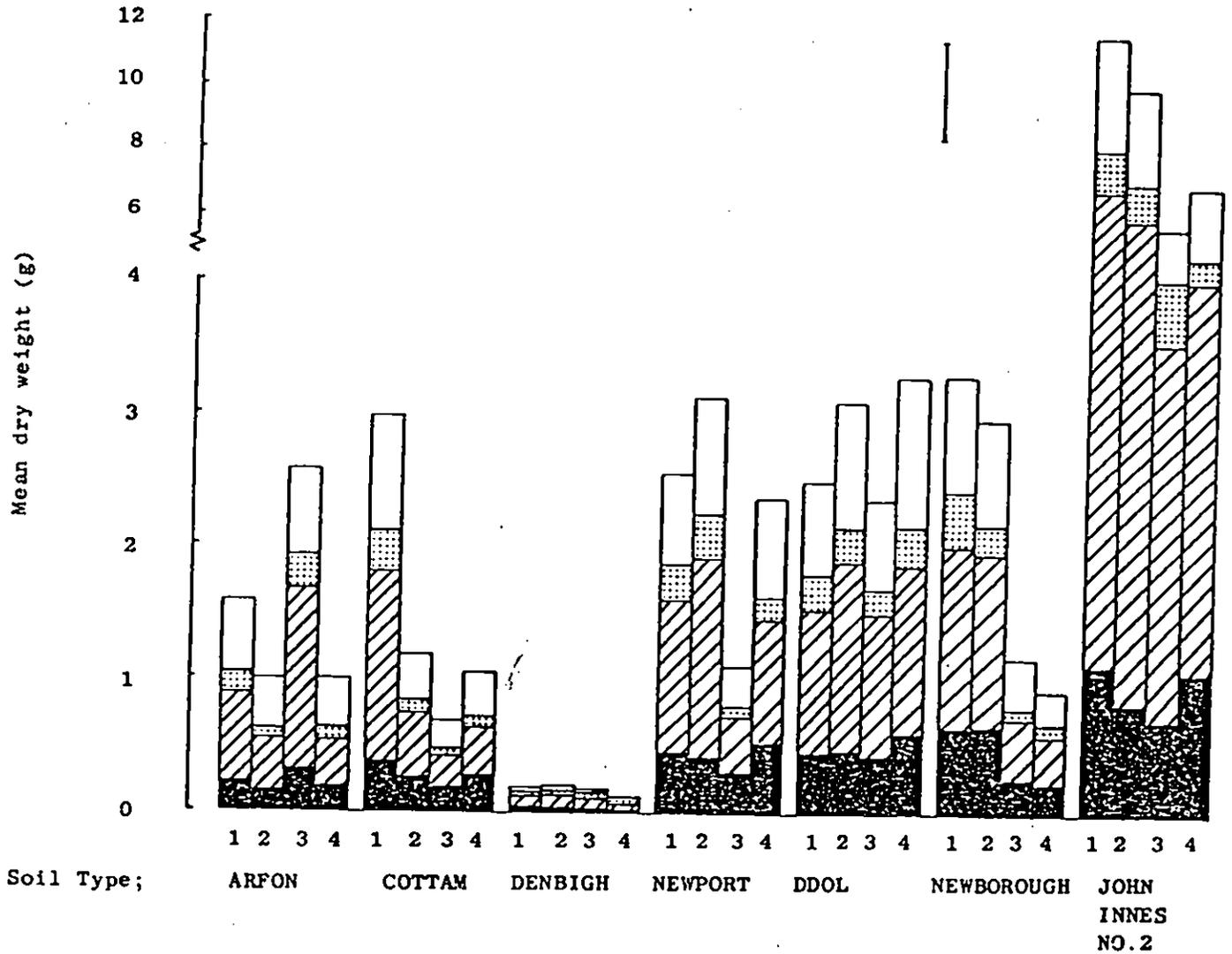
4.5 Results

In the early stages of the experiment, there were no visible effects of the different acid rain treatments on the growth of the seedlings. However, after 18 weeks visible damage appeared on the leaves of white clover and birch seedlings in all soil types for the pH 2.5 treatment. The damage was found mainly on the margins of leaves; a slight chlorosis followed by bleaching and death of the leaf cells. At about 20 weeks, some seedlings of Sitka spruce in all the soil types, except John Innes potting compost, exhibited a brown discoloration of crown needles in the pH 2.5 treatment. This discoloration was sometimes followed by death of the growing point. For both Sitka spruce and birch, the symptoms of visible injury became less apparent with further exposure and there was a tendency towards recovery. Plants of barley and ryegrass did not exhibit visible leaf damage in any treatment but, after 21 weeks of the experiment, it was quite clear that there was less plant material in the pH 2.5 treatment bays as compared to the other treatments. This visual impression was subsequently confirmed.

The mean total dry weights and their component fractions of roots, stems, leaves and ears for winter barley at all four rainwater treatments for all soils except Manod are shown in Figure 1. All plants died in the Manod soil at all rainwater treatments. It is immediately apparent that the yields of plants in all soils except the John Innes potting compost were poor and that there was a general trend for larger yields at the less acid rainwater treatments. However, in analyses of variance of the data for individual soils there were no significant effects of treatments. This was considered to be due to a large amount of variation between individual plants within a treatment. Therefore, further analyses of the data were carried out taking into account all soils and treatments. In the full analyses of variance, it was confirmed that there were highly significant effects of soils ($P \leq 0.001$) for all dry weight fractions with consistently higher yields in the John Innes potting compost. In addition, there were overall significant effects of rainwater treatments on the dry weight of stems ($P \leq 0.05$), leaves ($P \leq 0.01$), ears ($P \leq 0.01$) and the whole plant ($P \leq 0.05$) and the number of ears produced ($P \leq 0.05$). Comparisons of the grand means of the rainwater treatments revealed that there were significantly greater yields of all of these dry weight fractions ($P \leq 0.05$) in the pH 5.6 treatment than in the pH 3.5 and pH 2.5 treatments. In addition, the dry weights of leaves and ears were higher ($P \leq 0.05$) in the pH 4.5 as compared to the pH 3.5 treatment but not the pH 2.5 treatment. The numbers of ears produced (Table 2) were significantly greater ($P \leq 0.05$) in both the pH 5.6 and pH 4.5 treatments as compared to the more acid rainfall regimes. There were no significant effects of rainfall on the dry weights of roots or root/shoot ratios and no interactive effects of soils x rainwater treatments could be detected.

Typically, analysis of variance techniques assume that all treatments are discretely different and not related. For this experiment, this is not the case since log hydrogen ion concentration (pH) fits a linear scale. This relationship between treatments may be taken into account by considering rainfall as a covariate with the different growth parameters. If the subsequent analyses are significant, it is possible to develop equations to describe the relationship between pH and growth measurements. Table 3 shows the relationships calculated for winter barley.

Figure 1. Mean dry weight yields of winter barley grown on a range of Welsh soils and exposed for 21 weeks to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.



The vertical bar indicates the least significant difference ($P < 0.05$) between total plant dry weights for any treatment on any soil.

Table 2 Numbers of ears produced by plants of winter barley grown on a range of Welsh soils and exposed to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.

Soil	pH				Soil mean
	2.5	3.5	4.5	5.6	
Arfon	1.00	1.67	1.33	1.67	1.42
Cottam	1.07	1.07	1.27	1.73	1.28
Denbigh	0.13	1.00	0.87	0.93	0.73
Newport	1.47	1.20	2.00	1.80	1.62
Ddol	2.00	1.40	2.07	1.53	1.75
Newborough	1.20	1.20	1.93	2.00	1.58
John Innes No. 2	3.53	3.00	4.73	5.33	4.15
Treatment Means	1.49	1.50	2.03	2.14	

L.S.D. between soil means = 0.65 ($P \leq 0.05$)

L.S.D. between treatment means = 0.49 ($P \leq 0.05$)

Table 3 Equations to describe the relationship between pH of rainwater treatments and measurements of growth for winter barley

<u>Growth parameter</u>	<u>Equation</u>
Stem weight	$y = 0.1512 + 0.273x$ ($P \leq 0.01$)
Leaf weight	$y = 0.0265 + 0.268x$ ($P \leq 0.01$)
Weight of ears	$y = 0.2346 + 0.137x$ ($P \leq 0.01$)
No. of ears	$y = 0.8160 + 0.242x$ ($P \leq 0.01$)
Total shoot weight	$y = 0.4083 + 0.47x$ ($P \leq 0.01$)
Total plant weight	$y = 0.6975 + 0.5x$ ($P \leq 0.01$)

where y = growth parameter and
 x = pH

No linear relationship could be predicted for pH and the weights of roots

Figure 2 shows the mean total dry weights and their component fractions of roots and shoots for perennial ryegrass at all four rainwater treatments for all soils. Once again, analyses of the separate results for each soil did not reveal differences between the treatments. In the larger analysis for all soils and rainwater treatments, there were significant effects of soils ($P < 0.001$) but no effects of treatment or the interaction of soils x treatments for total plant weights and the effect on soils was due entirely to an overall higher yield in the John Innes soil. However, in the analysis of the root fraction dry weights, there were additional significant effects of rainwater treatments ($P < 0.01$) with the overall largest mean weight of roots being found in the pH 2.5 treatment. This was significant ($P < 0.05$) in comparison with the pH 3.5 treatment. Similarly, for the shoot dry weight fraction, there were significantly higher yields in both the pH 3.5 and pH 2.5 as compared to the pH 5.6 treatment. For root/shoot ratios, the soil differed markedly from each other (see Table 4) and there was a generally lower measure for the pH 2.5 treatment which was significant ($P < 0.05$) with respect to the pH 5.6 and pH 3.5 treatments. The number of seed heads per plant was not significantly affected by rainwater treatment (Table 5) but there was a greater overall production ($P < 0.001$) in the John Innes soil. Considering rainfall as a covariate with the different growth parameters only produced a significant relationship ($P < 0.01$) for root dry weight of ryegrass. The equation was:

$$y = 1.2371 - 0.082x$$

where y = root dry weight and x = pH.

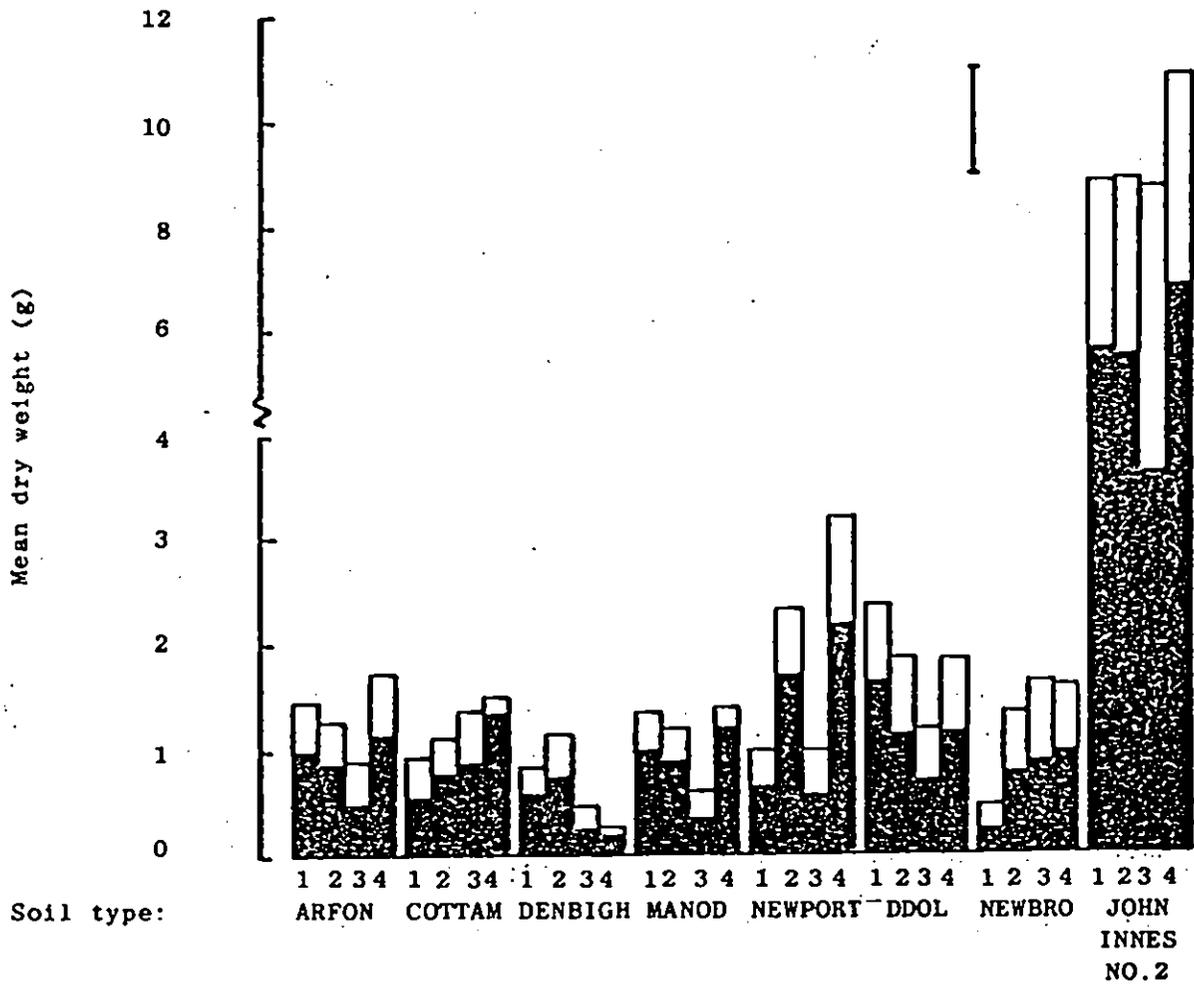
The mean total yields of white clover and their component fractions of roots and shoots at all rainwater treatments for all soils are shown in Figure 3. Analyses of the data for the separate soils again failed to reveal significant differences between the treatments. In the full analysis of variance for all soils and rainwater treatments, it was immediately apparent that there were large differences between the soils for root, shoot and total dry weights. For all measurements there were significant differences ($P < 0.05$) between yields for the soils on a gradient - John Innes Newport Arfon the other soils. In addition, there were significant effects of rainwater treatments for shoot ($P < 0.001$) and total plant ($P < 0.05$) dry weights. For both parameters, there was a lower overall yield in the pH 2.5 as compared to all other treatments. A significant interactive effect of soil x treatment ($P < 0.05$) revealed that the overall treatment difference observed was a result of poorer yields at the pH 2.5 treatment only for plants grown on John Innes, Arfon and Newport soils. The plants grew so poorly on the remaining soils that differences between treatments were negligible. For white clover, there were no effects of treatments on root dry weights or root/shoot ratios. Considering rainfall as a covariate with the different growth parameters produced significant relationships ($P < 0.01$) for both shoot and total dry weights. The equations were:

$$\begin{array}{ll} \text{for shoot dry weight} & y = 0.8013 + 0.109x \\ \text{for total plant dry weight} & y = 1.098 + 0.112x \end{array}$$

where y = growth parameter and x = pH.

The pH's of the different soils before and after use in the experiments are shown in Table 6. Treating rainwater and soils as main effects in an analysis of variance, it was found that the pH 2.5 treatment had

Figure 2. Mean dry weight yields of *Lolium perenne* L. (ryegrass) grown on a range of Welsh soils and exposed for 23 weeks to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.



Key

- | | |
|----------|----------|
| 1 pH 5.6 | □ Shoots |
| 2 pH 4.5 | |
| 3 pH 3.5 | ■ Root |
| 4 pH 2.5 | |

The vertical bar indicates the least significant difference ($P < 0.05$) between total plant dry weights for any treatment on any soil.

Table 4 Mean root/shoot ratios of *Lolium perenne* L. (ryegrass) grown on a range of Welsh soils and exposed to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6

SOIL	pH OF RAINWATER TREATMENT				SOIL MEANS
	5.6	4.5	3.5	2.5	
ARFON	0.851	0.853	0.943	0.626	0.818
COTTAM	1.017	0.694	1.022	1.131	0.966
DENBIGH	1.275	1.532	1.564	0.466	1.209
MANOD	1.043	1.560	1.057	0.419	0.770
NEWPORT	1.126	0.711	0.975	1.106	0.980
DDOL	1.013	1.131	0.892	1.130	1.042
NEWBOROUGH	1.419	1.557	1.503	1.201	1.420
JOHN INNES NO. 2	0.943	0.968	1.903	0.913	1.182
TREATMENT MEANS	1.086	1.001	1.232	0.874	

L.S.D. between soil means = 0.242 ($P \leq 0.05$)

L.S.D. between treatment means = 0.171 ($P \leq 0.05$)

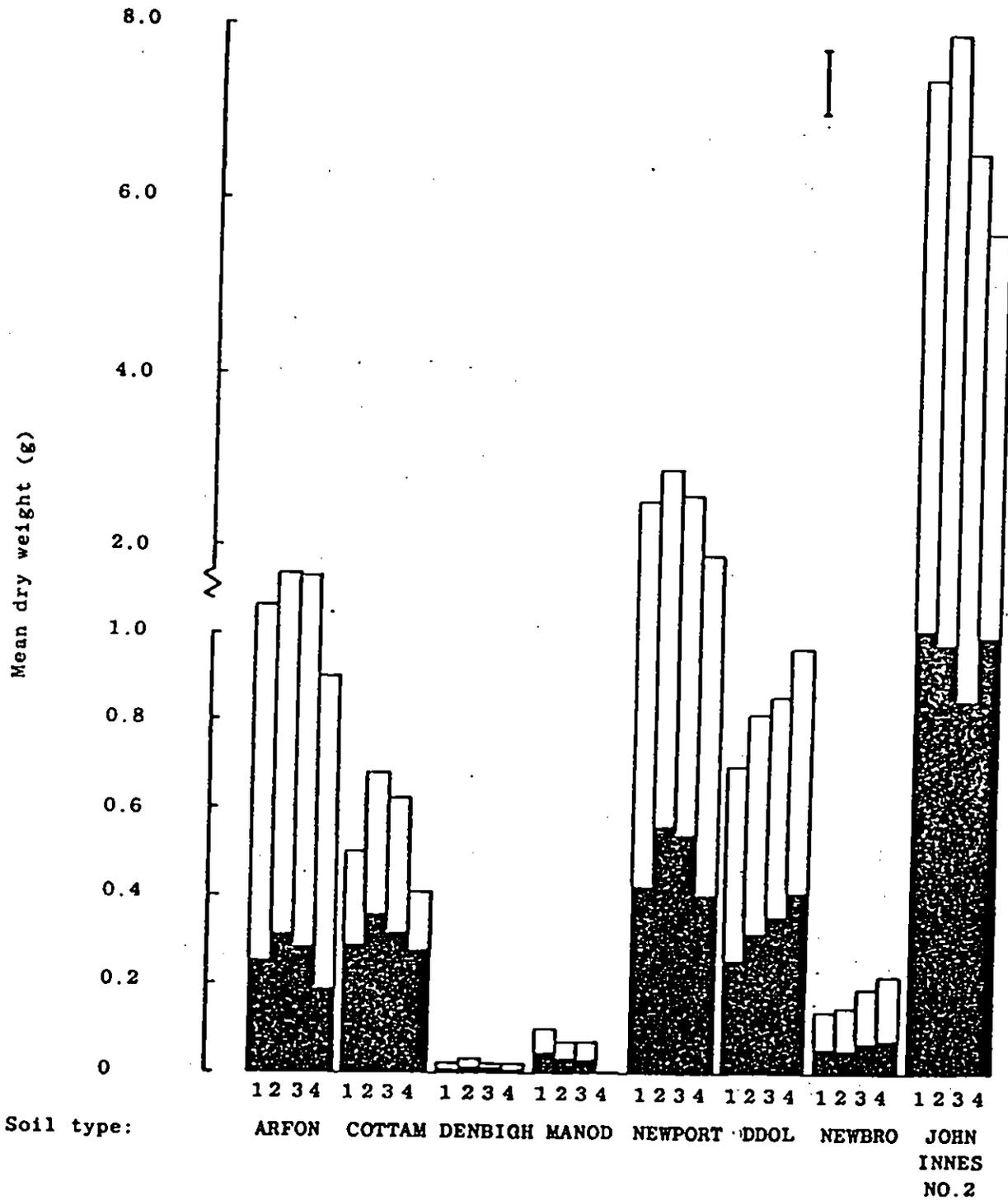
Table 5 Mean numbers of seed heads per plant of Lolium perenne L. (ryegrass) grown on a range of Welsh soils and exposed to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.

SOIL	pH OF RAINWATER TREATMENT				SOIL MEANS
	5.6	4.5	3.5	2.5	
ARFON	1.20	0.93	0.13	1.53	0.95
COTTAM	0.33	0.87	0.73	1.00	0.73
DENBIGH	0.33	1.00	0.33	0.13	0.45
MANOD	0.67	0.47	0.20	2.13	0.87
NEWPORT	0.53	2.27	0.33	1.87	1.25
DDOL	2.40	1.40	0.40	0.67	1.22
NEWBOROUGH	0.00	0.87	0.27	0.93	0.52
JOHN INNES NO. 2	6.20	7.27	4.20	7.93	6.40
TREATMENT MEANS	1.46	1.88	0.83	2.03	

L.S.D. between treatment means = Soil 1.339 ($P \leq 0.05$)

L.S.D. between treatment means = Rain 0.947 ($P \leq 0.05$)

Figure 3. Mean dry weight yields of *Trifolium repens* L. (white clover) grown on a range of Welsh soils and exposed for 24 weeks to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.



Key

- 1 pH 5.6
- 2 pH 4.5
- 3 pH 3.5
- 4 pH 2.5

- Shoots
- Roots

The vertical bar indicates the least significant difference ($P < 0.05$) between total plant dry weights for any treatment on any soil.

Table 6 pH's of a range of Welsh soils before and after supporting the growth of (a) winter barley, (b) ryegrass and (c) white clover during exposure to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.

SOIL TYPE	INITIAL SOIL pH	SPECIES GROWN	RAINWATER TREATMENTS			
			5.6	4.5	3.5	2.5
ARFON	5.48	Barley	5.14	5.01	5.19	4.69
		Ryegrass	5.10	5.03	5.03	4.25
		Clover	4.69	4.61	4.60	4.14
COTTAM	5.05	Barley	5.03	4.90	4.84	4.31
		Ryegrass	4.77	4.79	4.68	4.13
		Clover	4.65	4.56	4.52	4.01
DENBIGH	4.40	Barley	4.20	4.15	4.18	4.03
		Ryegrass	4.20	4.21	4.24	4.13
		Clover	3.94	4.08	3.89	3.74
MANOD	4.04	Barley	3.94	3.85	3.91	3.70
		Ryegrass	3.79	3.80	3.67	3.51
		Clover	3.63	3.55	3.58	3.52
NEWPORT	8.00	Barley	7.45	7.44	7.43	7.44
		Ryegrass	7.36	7.37	7.46	7.32
		Clover	7.49	7.42	7.37	7.43
DDOL	7.40	Barley	7.31	7.31	7.39	7.36
		Ryegrass	7.40	7.37	7.37	7.29
		Clover	7.36	7.34	7.36	7.34
NEWBOROUGH	8.25	Barley	7.70	7.73	7.72	7.71
		Ryegrass	7.67	7.56	7.50	7.24
		Clover	7.88	7.92	7.83	7.74
JOHN INNES	6.39	Barley	6.09	6.07	5.90	5.13
		Ryegrass	6.27	6.28	6.19	4.51
		Clover	5.61	5.54	5.26	5.18
TREATMENT MEANS			5.78	5.74	5.71	5.41

L.S.D. between treatment means = 0.118 ($P \leq 0.05$)

caused an overall reduction in soil pH ($P \leq 0.01$) as compared to the other treatments. Data for the individual soils reveal that the reductions in pH occurred for all soils except the three most alkaline: Ddol, Newport and Newborough. Further analyses of the soils from the clover experiment to determine extractable cation contents are shown in Table 7. The quantities of some cations in some soils at the end of the study as compared to the pre-experiment samples appear to be less than might be anticipated. However, an analysis of variance revealed that there were no significant differences caused by the different rainwater treatments in the levels of cations present in any of the soils.

Table 8 shows the percentage contents of nutrient cations in leaves of white clover at the end of the exposure period. It can be seen that there were no significant differences between the treatments for potassium, sodium, magnesium and aluminium. However, there were significantly larger amounts of manganese ($P \leq 0.05$) in the pH 2.5 treatment as compared to the others and a larger amount of calcium in the pH 2.5 as compared to the pH 3.5 treatment. Within the three least acid treatments, there was the opposite trend for larger quantities of nutrients with decreasing acidity.

The results of the non-destructive growth measurements of birch and Sitka spruce are shown in Figures 4 and 5. For birch, an analysis of variance revealed highly significant effects of both soils ($P < 0.001$) and treatments ($P < 0.001$). Overall plants grew better in the John Innes soil ($P < 0.001$) and worse in the Ddol and Newborough soils ($P < 0.001$) as compared to the others. Furthermore, there was an overall greater height ($P < 0.001$) in the pH 2.5 treatment and an overall lower height in the pH 5.6 treatment ($P < 0.05$) as compared to the two intermediate pH treatments. These results were in contrast to those obtained for Sitka spruce where there were no significant effects of treatment on plant height. For Sitka spruce, plant heights were generally lower in the Ddol soil and more than double in the John Innes compost ($P < 0.001$) as compared to other soil types.

4.6 Discussion

It is readily apparent from the data presented that increasing acidity in simulated rainfall may induce visible damage in some plants. For the five species studied in this experiment, there were obvious visible leaf lesions or discolorations at the pH 2.5 treatment in white clover, birch and Sitka spruce. Except in the case of Sitka spruce, this damage occurred for all soil types indicating that it was a direct effect of acidified water passing over the leaves rather than an indirect effect of the exposure treatments on the soils. The lack of visible damage for Sitka spruce growing on John Innes compost may be related to the greater rate of growth of the plants in this soil. Sitka spruce was the slowest growing species in the study and for soils other than John Innes the maximum mean height after one year was 3.4 cm - about one third of that for plants grown in John Innes compost. It is known, from studies with gaseous pollutants, that plants are more susceptible to pollution injury under conditions of slow growth (see reviews by Ormrod, 1978; Bell, 1982).

In the assessment of effects of simulated acid rainwater treatments on dry weight production, it is apparent that there was much variation between plants within a treatment. In previous work, the use of 15

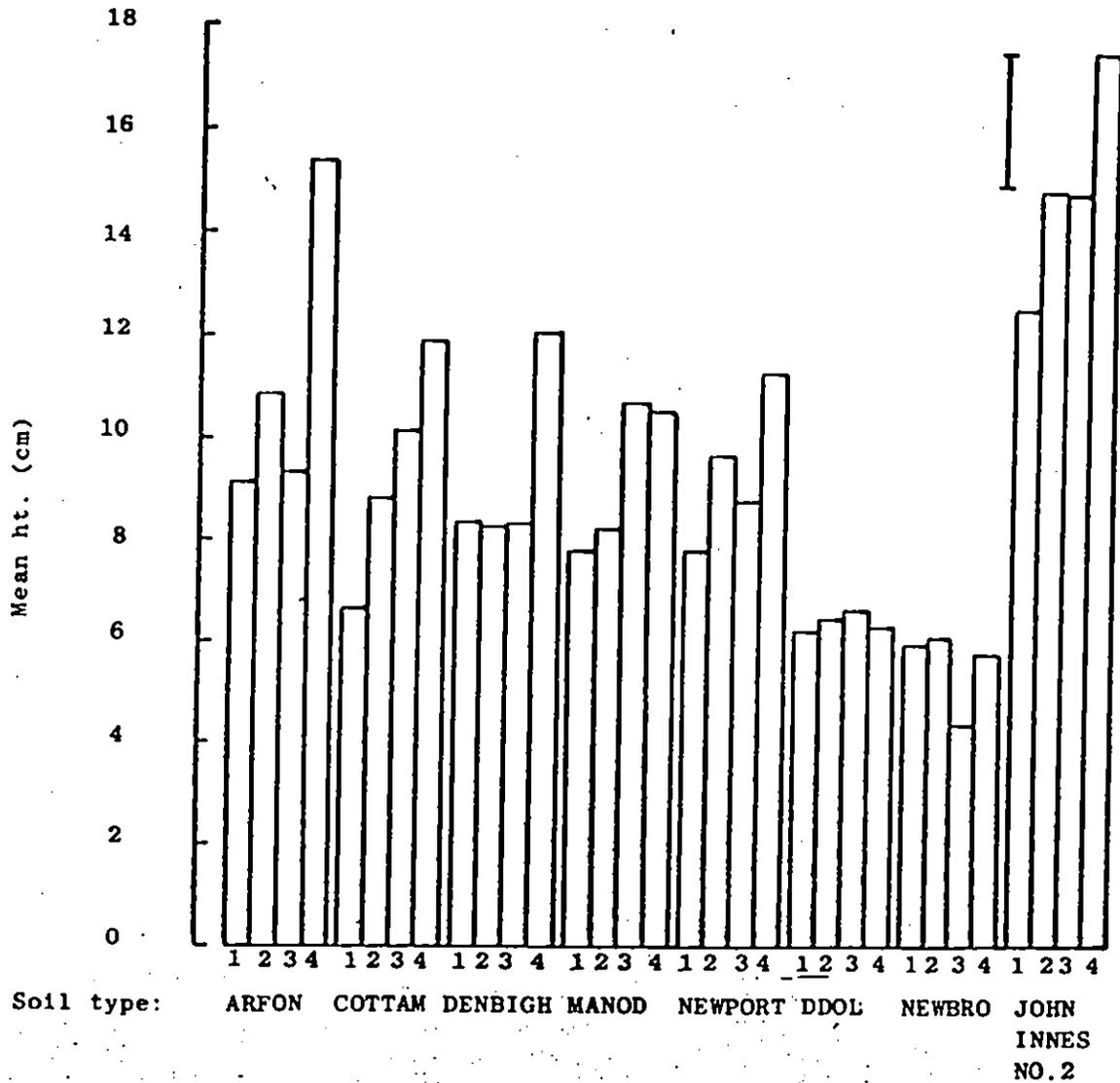
Table 7 Soil cation contents (meq/100g air dry soil) for a range of Welsh soils before and after supporting the growth of white clover exposed to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6

SOIL	CATION	INITIAL CONTENT	CONTENT AFTER GROWTH OF CLOVER IN pH's			
			5.6	4.5	3.5	2.5
ARFON	K	0.459	0.464	0.426	0.422	0.424
	Na	0.296	0.165	0.183	0.237	0.177
	Mn	0.307	0.264	0.157	0.391	0.483
	Ca	4.60	5.04	4.40	4.76	3.40
	Mg	0.556	0.477	0.373	0.360	0.307
COTTAM	K	0.175	0.174	0.164	0.165	0.182
	Na	0.383	0.176	0.200	0.190	0.148
	Mn	0.120	0.116	0.136	0.130	0.191
	Ca	4.28	4.12	4.36	3.96	2.48
	Mg	1.439	1.288	1.341	1.282	0.674
DENBIGH	K	0.326	0.188	0.165	0.170	0.214
	Na	0.261	0.205	0.165	0.167	0.170
	Mn	0.653	0.394	0.275	0.394	0.596
	Ca	1.48	1.60	1.12	1.40	1.08
	Mg	0.477	0.314	0.229	0.249	0.170
MANOD	K	0.237	0.286	0.177	0.155	0.135
	Na	0.331	0.226	0.235	0.181	0.141
	Mn	0.102	0.103	0.097	0.048	0.074
	Ca	0.84	0.68	0.68	0.80	1.04
	Mg	0.464	0.504	0.451	0.392	0.183
NEWPORT	K	0.296	0.106	0.222	0.111	0.115
	Na	0.209	0.118	0.113	0.136	0.146
	Mn	0.020	0.026	0.020	0.019	0.017
	Ca	22.16	24.40	23.20	25.20	23.20
	Mg	0.438	0.235	0.216	0.242	0.249
DDOL	K	0.265	0.156	0.109	0.113	0.080
	Na	0.244	0.174	0.172	0.240	0.176
	Mn	0.033	0.033	0.033	0.049	0.041
	Ca	1.86	1.56	1.08	1.20	1.00
	Mg	1.027	0.759	0.896	0.772	0.844
NEWBOROUGH	K	0.118	0.047	0.049	0.029	0.039
	Na	0.209	0.169	0.197	0.183	0.139
	Mn	0.038	0.030	0.033	0.033	0.022
	Ca	17.60	20.40	15.60	15.60	7.20
	Mg	0.327	0.222	0.229	0.209	0.144
JOHN INNES	K	0.816	0.377	0.602	0.272	0.714
	Na	0.505	0.209	0.188	0.204	0.181
	Mn	0.320	0.358	0.233	0.429	0.287
	Ca	11.04	8.00	8.00	8.00	8.40
	Mg	1.406	1.059	0.857	0.974	1.190

Table 8 Percentage contents of nutrient cations (per unit dry weight) in leaves of white clover exposed to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6. (All plants were grown in John Innes No. 2 potting compost)

CATION	RAINWATER TREATMENT (pH)				L.S.D. (P<0.05) Between treatments
	5.6	4.5	3.5	2.5	
POTASSIUM	2.261	2.384	2.143	2.812	1.221
SODIUM	0.2129	0.1753	0.1405	0.2038	0.100
MANGANESE	0.067	0.054	0.040	0.093	0.025
CALCIUM	3.918	3.619	2.334	4.520	1.058
MAGNESIUM	0.492	0.477	0.384	0.533	0.152
ALUMINIUM	0.0804	0.0832	0.0871	0.0605	0.037

Figure 4. Mean heights of *Betula pendula* Roth. (birch) seedlings grown on a range of Welsh soils and exposed for 1 year to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.

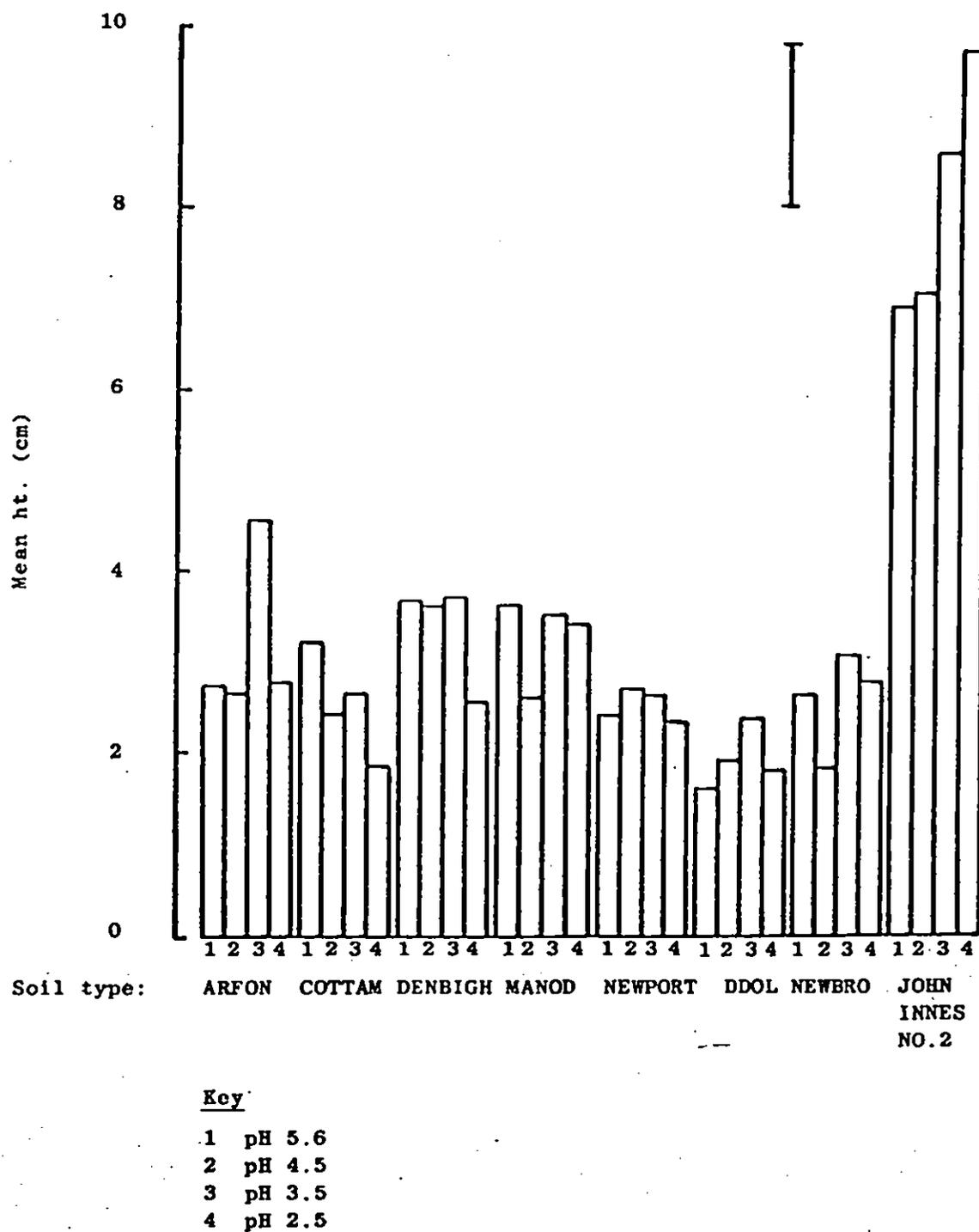


Key

- 1 pH 5.6
- 2 pH 4.5
- 3 pH 3.5
- 4 pH 2.5

The vertical bar indicates the least significant difference ($P < 0.05$) between plant heights for any treatment on any soil.

Figure 5. Mean heights of *Picea sitchensis* (Bong.) Carr. (Sitka spruce) seedlings grown on a range of Welsh soils and exposed for 1 year to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6



The vertical bar indicates the least significant difference ($P < 0.05$) between plant heights for any treatment on any soil.

replicates per treatment has been found adequate for studying the effects of a range of environmental stresses on plants (Ashenden, 1974; Ashenden & Mansfield, 1978). Here separate analyses (*viz.* 15 replicates per treatment) for individual soil types failed to show effects of the treatments on the growth of barley, ryegrass or clover. It was only in the larger analyses of the effects of the rainwater treatments over the whole range of soils that significant differences were found.

The effects of rainwater treatments on dry weight yields varied for the three species. The least sensitive species was ryegrass. This produced increased shoot dry weights at the two more acid treatments but, these increases were not reflected in statistically significant higher plant yields. Therefore, the suggestion must be that the treatments caused a shift in the proportioning of assimilates between the roots and shoots of the plants. The apparent promotion of shoot growth for ryegrass in this study is in complete contrast to the observations of Amthor & Bormann (1983) who found dry mass accumulation in leaves to be inhibited by increased precipitation acidity in long-term exposures. More recently, however, there has been some suggestion (Amthor, 1984) that ozone may have contributed to the yield reductions which they observed.

White clover was more sensitive to the different rainwater treatments than ryegrass. Overall, there were significant reductions in the dry weight yields of the shoots which caused corresponding reductions in total plant dry weights for the pH 2.5 treatment. As compared to control (pH 5.6 treatment) plants, the yields for both parameters were an average of 0.33 g per plant less which was equivalent to a 21% reduction in total plant yield.

The most sensitive species of those taken for dry weight determinations was winter barley where there were overall significant reductions in the dry weight yields of stems, leaves, ears and whole plants for both the pH 3.5 and pH 2.5 treatments as compared to the control (pH 5.6) treatment. The magnitudes of the reductions were 44% for leaves, 46% for stems, 34% for ears and 37% for whole plant yields for the most acid (pH 2.5) treatment and slightly larger for the pH 3.5 treatment.

For winter barley, there were also significant differences between the pH 4.5 and pH 3.5 treatments for the dry weights of leaves and ears which confirm the general trend for decreasing yields with increasing acidities of simulated rainfall. The decreased numbers of ears found for the two most acid rainfall treatments are no doubt, at least partially, responsible for the lower yields of the ears dry weight fractions. In grasses, reductions in the numbers of flowering tillers have been reported as a response to gaseous acidic pollutants (Whitmore & Mansfield, 1983).

While significant differences were not found for dry weight yields between the two least acid rainfall treatments (pH's 4.5 and 5.6) for any species, there were definite indications of higher values of all growth parameters at the unpolluted (pH 5.6) treatment for barley. Furthermore, for all three species, it proved possible to devise equations which related different growth parameters to the pH of simulated rainfall applied and all of these equations gave the relationships with a very high degree of confidence (99.9%). The relationships most sensitive to pH are those found for winter barley where the coefficients of x are largest, thus indicating larger gradients in the straight line relationships between growth parameters

and pH. Therefore, it is this species which, with observations on larger number of plants, would be most likely to allow discrimination between all four treatments.

In the non-destructive measurements of growth for trees, it was interesting to find that the only difference between the rainfall treatments was a stimulation in the growth of birch at the most acid treatment. This was particularly surprising since this was the only treatment to cause leaf damage earlier in the experiment. The most probable explanation for the growth enhancement is that the sulphur and nitrogen in the acids used to make up the rainwater treatments are being utilised as plant nutrients. After one year, the soils supporting the plants may have become deficient in these essential nutrients. In studies with pollutant gases, it is known that sulphur dioxide and nitrogen oxides may improve the yields of plants growing in nutrient deficient soils (see review by Cowling & Koziol, 1982).

It is apparent that there were large differences between the soils used in this investigation. While this is useful in that it allows an interpretation of the effects of the different simulated rainfall treatments to be averaged over a broad spectrum of soils, it may have also decreased the magnitudes of differences between the treatments (and certainly pro rata the numbers of plants handled). It was apparent from the data for white clover that the overall effect of the pH 2.5 treatment was due to the effects on plants growing in only 3 of the 8 soils. The plants in the other soils grew so poorly that the differences between the treatments were negligible. Similar differences between soils were found for winter barley, where all plants died in the Manod soil and growth was generally extremely poor in the Denbigh soil. No doubt these effects for winter barley were associated with initial soil pH; these were the two most acid soils and barley is known to fail as a crop in acid soils. In this connection, it is interesting to note that birch, which is an acid-loving plant, grew better in the two most acid soils than in some of the more alkaline ones.

As an averaged effect over all the soils, the most acid rainwater treatment caused a drop in soil pH (Table 6) which was apparent in the more acid soils. Such decreases in soil pH will enhance the rate of leaching of soil nutrients and subsequently decrease soil fertility. The fact that these changes were recorded in under six months suggests that soils may quickly deteriorate with additions of hydrogen ions. While the effect was observed only for the pH 2.5 treatment, it seems likely that similar shifts in soil pH would occur with less acid treatments over longer periods of time or with larger amounts of rainfall. The controlling influence must be the total quantity of hydrogen ions added to the soil.

In the more detailed analyses of the soils from the clover study, it is surprising to note that the lower pH of soils in the most acid rainfall treatment was not associated with a significant reduction in soil nutrients. A study of the data in Table 7 certainly reveals that the amounts of calcium and magnesium in some of the soils are much less in the pH 2.5 than the other rainfall treatments. These are the two cations which would be leached initially from the exchange complex by acid washing. The fact that there are not significant differences between the different rainwater treatments may be due to the use of only duplicate samples and further study in this area may be necessary.

The effects of the different rainfall treatments on the nutrient contents of leaves of which clover grown in John Innes compost were the opposite of what would have been predicted. Most reports on the effects of acid rain on plant nutrient status show increasing acidities of rain to increase the leaching of leaf cations and result in lower concentrations within the plant (see reviews by Abrahamsen, Hornvedt & Tveite, 1976; Rorison, 1980). One possible explanation of the increases in calcium and manganese found at the pH 2.5 treatment in this study is that the higher acid input to the soil increased the availability of these two nutrients to the plant. Of the nutrients measured, manganese and calcium, in the form of calcium carbonate, are the most easily mobilised by additions of acid. In the making of John Innes compost, calcium carbonate is one of the substances added. If the experiment had been continued, it is possible that the increased availability of these two nutrients would revert to a deficiency in this treatment.

5 EFFECTS ON GERMINATION

A possible drawback to the studies in Section 4 is that the plants were not exposed to the rainwater treatments during germination or at emergence. These could be critical stages when plants may be more sensitive to acidic pollutants. This experiment investigates that possibility.

5.1 Methods

Winter barley was chosen as the plant material for this study because it was known to be sensitive to soil pH and is not recommended by ADAS as a crop in fields with a pH below 5.8. A total of 10 germination trials was carried out in which 50 seeds were sown at a depth of 1 cm in each of 4 trays of John Innes No. 2 potting compost. The first 6 germination trials were carried out in an unheated glasshouse during winter 1983 and spring 1984 and the others in controlled environment cabinets set at a temperature of 5°C and a 12h photoperiod. In all trials, the trays were watered with 3 x 0.6 cm per week of simulated acid rain adjusted to pH's of 2.5, 3.5, 4.5 and 5.6 by 7:3 additions (by volume) of sulphuric and nitric acids. At each watering, the numbers of seedlings which had emerged were recorded.

In November 1984, a further study was carried out on seeds sown individually at a depth of 1 cm in 7.5 cm - diameter pots of John Innes No. 2 potting compost. These pots were placed in the polythene tunnel exposure system described in 3.1 and sprayed with 3 cm per week of the 4 different simulated acid rain treatments - pH's 2.5, 3.5, 4.5 and 5.6.

5.2 Results

The results of the different germination trials are shown in Table 9. In the tray experiments, there appears to be a tendency towards a greater rate of germination in the less acid treatments. However, an analysis of variance of germination rate failed to detect significant differences between the treatments. In the experiment where seeds were sown individually in pots, there were no differences between the rainwater treatments.

5.3 Discussion

Previous work has shown that some species are more susceptible to wet acid pollutants during germination and establishment (Abrahamsen *et al.*, 1976). In the initial studies conducted here (experiments 1-4), it did appear that the germination of winter barley was reduced by watering with simulated rain at the higher levels of acidity. However, these results were not confirmed in later studies.

Experiments 5 and 6 were carried out in a glasshouse during warm weather and it was considered that the higher temperature had increased the speed of germination and thus nullified the effects of the rainwater treatments. Similarly, in experiments 7 and 8 a temporary fault in the controlled environment cabinets caused a brief (24h) elevation in temperature. However, the later pot experiment was conducted in

Table 9 Total numbers of winter barley seedlings emerging in germination trials during exposure to different acidified watering treatments

Experiment	pH treatments			
	2.5	3.5	4.5	5.6
Tray experiments - 50 seeds sown per treatment				
1	30	37	36	49
2	29	39	10	49
3	32	24	40	42
4	19	22	30	24
5	50	50	48	48
6	49	45	46	48
7	42	35	44	44
8	44	39	46	46
9	18	35	31	35
10	28	32	42	40
Treatment Means	26	32	32	40
Pot experiment - 100 seeds sown per treatment				
	96	98	97	99

November 1984 and temperature conditions in the exposure chamber were deemed to be comparable with those in the field at normal sowing times (September) for this crop. The lack of any effect in this experiment must lead to the conclusion that the germination of winter barley is not affected by the acidity of rainfall.

6 INTERACTIVE EFFECTS WITH DEFOLIATION

This experiment was carried out because of the possibility that wet acid deposition may be more toxic to plants in the presence of additional stresses. It is known that gaseous acid pollutants are more toxic when additional stresses are operative (see Davies, 1980; Ormrod, 1978). In much of Wales, the most commonly occurring stress on vegetation is defoliation either by mechanical means or grazing animals.

6.1 Methods

Seeds of Lolium perenne L. (ryegrass) were sown at a rate of 0.25 g (135 seeds) per pot in 96 x 7.5 cm diameter pots containing John Innes No. 2 potting compost on 19 December, 1983. The pots were maintained in an unheated greenhouse and, after 6 weeks, the plant material in half of the pots was cut back to 2.5 cm above soil level and the clippings collected for dry weight determination. From the day of this initial cut, simulated rainfall treatments commenced. There were 4 treatments - pH's 2.5, 3.5, 4.5 and 5.6 - with 3 cm being applied per week to each of 12 cut and 12 uncut pots. Subsequently, the plants which had been cut were defoliated to a height of 2.5 cm every two weeks for 12 weeks and the clippings collected for dry weight determination. At the final cut, all plants ("cut" and "uncut") were cut back to 2.5 cm above soil level and then to soil level and the dry weights of clippings obtained.

6.2 Results

The results of this experiment are shown in Table 10. It can be seen that there was a large difference between the defoliated and non-defoliated regimes and, in an analysis of variance this difference was highly significant ($P < 0.001$). Within the defoliated plants, there were significant differences ($P < 0.05$) in the dry weights of clippings obtained from plants exposed to the different acid rain treatments at several different cuts, but no particular pattern emerged. The yield from the pH 2.5 treatment was lower than the pH 5.6 treatment ($P < 0.05$) at the second cut but higher than all other pH treatments ($P < 0.05$) at the third and sixth cuts. At the end of the study, the only difference in the summed yields of the cuts was that plants at the pH 4.5 treatment had yielded less than those in the pH 2.5 treatment ($P < 0.05$). There were no significant differences between rainwater treatments for the yields of the 0-2.5 cm fraction of the defoliated plants, either of the two plant fractions measured in the uncut plants or total shoot weights of both cut and uncut plants.

6.3 Discussion

It is apparent from the data presented that there are no interaction effects of acid rainwater and defoliation stresses on the yields of Lolium perenne. The differences in yields found between the rainwater treatments for defoliated plants at individual cuts are most likely to have been caused by experimental error in the technique used to determine the height of cutting. Thus, a lower yield at the third cut became a higher yield at the fourth cut for the most acid as compared to the least acid treatment.

Table 10 Mean dry weights (g) of *Lolium perenne* L. (ryegrass) (a) defoliated to a height of 2.5 cm at 2 week intervals and (b) not defoliated during exposure to simulated acid rain at pH's of 2.5, 3.5, 4.5 and 5.6.

	2.5	3.5	4.5	5.6	L.S.D. both treatments ($P \leq 0.05$)
<u>Defoliated</u>					
Pre-treatment cut	0.042	0.038	0.042	0.041	0.005
1st cut	0.068	0.068	0.064	0.063	0.006
2nd cut	0.064	0.072	0.069	0.079	0.010
3rd cut	0.133	0.111	0.093	0.104	0.018
4th cut	0.078	0.073	0.067	0.083	0.010
5th cut	0.073	0.063	0.063	0.078	0.016
6th cut	0.090	0.066	0.062	0.067	0.015
Total for all cuts	0.548	0.489	0.458	0.513	0.063
Final cut to soil level	0.771	0.753	0.807	0.918	0.171
TOTAL PLANT WEIGHT	1.318	1.243	1.266	1.432	0.204
<u>Not defoliated</u>					
Cut to 2.5 cm above soil	1.130	0.988	0.945	0.983	0.200
Subsequent cut to soil level	1.007	0.983	0.931	1.075	0.158
TOTAL PLANT WEIGHT	2.137	1.972	1.876	2.058	0.178

7 RADIOMETER STUDY

The objective of this study was to assess the potential for discriminating between the foliage of plants that have been subjected to different levels of acid precipitation, using multispectral remote sensing.

7.1 Methods

The plant material used in this study was the birch seedlings described in 4.3 and exposed to simulated acid rain adjusted to pH's of 2.5, 3.5, 4.5 and 5.6 as described in 3.4. Only the plants grown in John Innes No. 2 potting compost were used and the observations were made in the first week of September, 1984 - 34 weeks after start of the experiment treatments.

From each of the four acid treatments, two groups of seven birch seedlings were assembled for observation. The plants in each group were placed close together, and all background soil below the leaves was masked with black paper to minimise extraneous reflected light. A radiometer and 35 mm camera were positioned 70 cm directly above the plants for making each observation. The radiometer, a Milton multiband, measured radiance in four wavebands (green, 0.5-0.6 μ m; red, 0.6-0.7 μ m; near infra-red, 0.76-1.1 μ m; and mid infra-red, 1.35-1.75 μ m). Black and white 35 mm photography recorded the canopy structure of each group of plants to enable subsequent correction for gross differences in leaf area. One photograph, and three readings in each radiometer band were taken from each of the eight groups of plants. The plants were illuminated by diffuse daylight, and radiometer readings were taken from a Kodak grey card to allow correction for variation in the level of illumination. A video camera and BBC-B microcomputer were used to digitise black and white photographs of the birch seedling canopies. From the resulting data, relative area estimates were calculated for each of the eight canopies, and these were applied to the radiometer data to correct for gross differences in leaf area.

In radiometer studies, bidirectional reflectance (BDR), which is expressed as the ratio of target radiance to grey card radiance, is normally computed to reduce variation in radiance that may be due to changes in the level of illumination. Each observation of the seedling canopies thus produced four BDR values, one for each of the radiometer wavebands, and these values were entered into the subsequent statistical analysis. Discriminant analysis is presented with classes and variables. In this experiment, the acid treatments form the classes and the BDR values for each of the four wavebands form the discriminating variables. The analysis identifies linear combinations of the variables in a discriminant function which maximises separation between classes. A second function frequently helps to discriminate between classes that were not clearly separated by the first function.

7.2 Results

In general, reflectance from the foliage increased with increasing acidity of the rainwater treatments. This was found to be the case for each of the four radiometer bands. Function one discriminated clearly between the two extremes of pH with a high degree of significance

($P \leq 0.001$). This discrimination can be seen in the direction of the x-axis on Figure 6. Radiometer bands one and two (green and red) contributed more than bands three and four (near-IR and mid-IR) to this first discriminant function (Table 11).

Function two discriminated between the intermediate treatments (pH 3.5 and pH 4.5) with a high degree of significance ($P \leq 0.001$). This discrimination can be seen in the direction of the y-axis on Figure 6. Radiometer bands two and three (red and near-IR) contributed more than bands one and four (green and mid-IR) to the second discriminant function. The respective contribution of each radiometer band to the two discriminant functions is detailed in Table 11. Using both functions, it has been possible to discriminate between the reflectance of all four seedling canopies in two-dimensional feature-space.

7.3 Discussion

It is immediately apparent from the data presented that radiometer studies of this nature are able to discriminate between the canopies of birch seedlings exposed to different acid precipitation treatments with a high degree of statistical significance. A multifactorial approach to the statistical analysis of reflectance data has utilised the full potential for discrimination between the treatments. To a greater or lesser extent, all of the radiometer bands had some utility in detecting differences between the rainfall treatments.

These encouraging results suggest that more detailed radiometer studies, including other species, should be carried out. It may then prove possible to develop mathematical models to describe the effects of acidic pollutants on radiance. If the discrimination between canopies subjected to different levels of acidic pollutants is consistently sensitive for a wide range of species it might prove worthwhile to analyse data collected by the NERC aeroplane and airborne Thematic Mapper sensor. In the long term, the integration of radiometer studies with aircraft and satellite data may lead to an operationally useful system for monitoring the effects of acidic pollutants on vegetation.

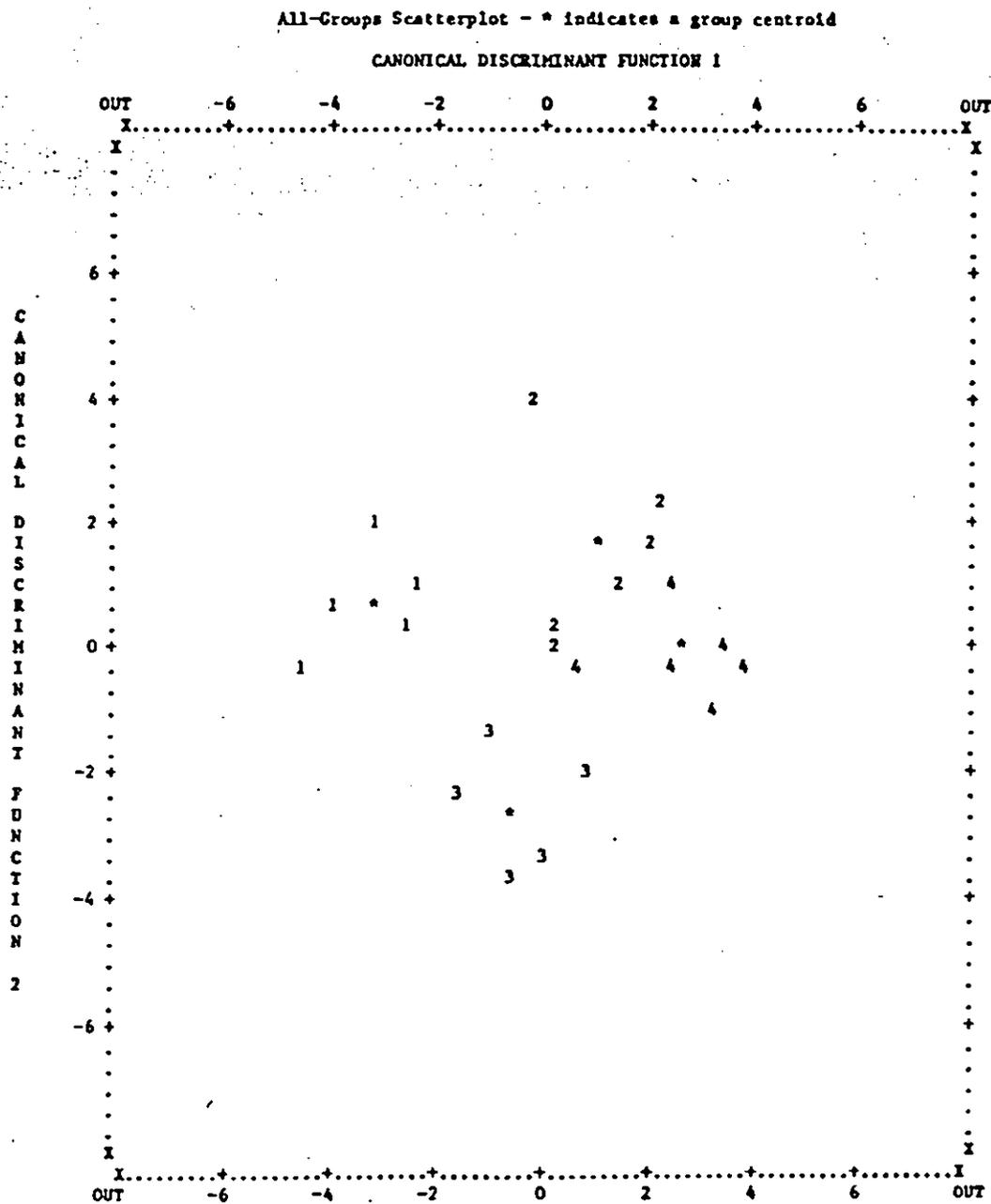
Table 11 Discriminant function coefficients of the radiometer bands used to differentiate between canopies of birch seedlings exposed to different simulated acid rain treatments.

	Function 1	Function 2
Band 1	-3.43810	-2.37017
Band 2	2.61210	4.06243
Band 3	0.28280	-4.09139
Band 4	1.28520	2.46561

Key

Band 1 Green light
Band 2 Red light
Band 3 Near infra-red light
Band 4 Mid infra-red light

Figure 6. Scatterplot of canonical discriminant function coefficients obtained from a radiometric study on birch seedlings exposed to different simulated acid rain treatments.



Symbols used in plots

Symbol	Group	Label
1	1	pH 2.5
2	2	pH 3.5
3	3	pH 4.5
4	4	pH 5.6
*		Group centroids

8 CONCLUDING REMARKS

The overall objective of this project is to assess the potential threat of acidic pollutants to the vegetation in Wales and thus the data should be viewed in relation to the acidities of rainfall known to prevail. Unfortunately, no large survey has yet been conducted of the rain in Wales although the indication is that it is acidic. Martin & Barber (1978) found a mean pH of 4.1 (70μ Equiv. H^+l^{-1}) for a mid-Wales site and ITE data for Plynlimon (Reynolds, personal communication) has revealed pH values in the range 3.8-4.6. Thus, the most important treatments in this study are those between pH 5.6 (the acidity of rain without any additions of pollutants) and pH 3.5 (slightly more acid than the readings so far obtained).

During the screening of plants growing on a range of Welsh soils, the only visible damage to plants occurred at the most acid (pH 2.5) rainfall treatment. This is outside of the range of rainfall acidities found for Welsh sites and thus, while wet deposits of acid pollutants may cause visible lesions on leaves of higher plants, they are unlikely to be doing so in the natural environment in Wales.

In determinations of the effects of the four simulated rainfall treatments on plant dry weights, there was a large amount of within treatment variation which may have masked differences between the treatments. None the less, it was apparent that winter barley was more sensitive to the acid rain treatments than ryegrass or white clover. Applications of rainfall at pH's 3.5 and 2.5 caused more than 30% reductions, compared with the control rainfall treatment of pH 5.6, in the yields of all dry weight fractions and the number of ears produced for winter barley. Furthermore, there were significantly greater yields of the leaves and ears dry weight fractions for the pH 4.5 as compared to the pH 3.5 rain treatment. This gradation of response in winter barley, of decreasing yields for the different dry weight fractions with increasing acidities of rain applied, allowed the generation of mathematical equations to describe the relationship with a very high degree of confidence ($P < 0.01$). The fact that these reductions in yield are occurring, over the critical range of acidities of rain recorded for sites in Wales, must be of some concern and work is required to quantify the effects more accurately. It is interesting to note that, while the growth of winter barley is sensitive to the acidity of rainfall its rate of germination is not.

The promotion of plant height in birch seedlings by the most acid rain treatment is interesting in that it suggests a fertilizer effect. The implication is that, for an acid tolerant species, growing in a nutrient deficient soil, the inputs of nitrogen and sulphur in acidified rain may be beneficial. However, these data should be viewed with caution since the beneficial effects may prove to be short-lived and within a natural ecosystem different processes may be involved. It is likely that continued high inputs of acid to a soil would gradually alter the composition and structure of the soil with subsequent adverse effects on the plants.

The data obtained in the radiometer study are extremely interesting in that they allowed discrimination between all four acid rain treatments. At present, observations have only been made on one species and screening of others must be carried out to see if the relationship holds

between light reflectance from leaves and acid watering treatments. In addition, the possible interactive or modifying effects of other environmental variables on reflectance would need to be considered.

These aspects including the mechanism of reflectance change will be investigated by ITE under its remote sensing program if finances are available.

9 FUTURE WORK

Studies on the effects of simply wet deposits of nitric and sulphuric acids on plants will be continued in the polythene tunnel exposure system using the present simulated acid rain treatments of pHs 2.5, 3.5, 4.5 and 5.6. In particular, the long-term experiment (initiated in January 1984), in which seedlings of birch and Sitka spruce growing in a range of Welsh soils have been exposed to 3 cm per week of the four rainwater treatments, will be extended for another growing season. Additional experiments to clarify and quantify the effects of acidic rainwater treatments on the growth of winter barley will be initiated. These experiments will aim to overcome the problems of the large within-treatment variation which prevented clear discrimination between the four rainwater treatments. Larger numbers of plants growing in one standard soil (John Innes compost) will be used in these experiments.

In order to increase the likelihood of identifying a suitable species to act as a bio-indicator of rainwater acidities in the field, additional species will be studied in the polytunnel exposure system. In particular, the suggestion that legumes may be more susceptible to acid rain will be investigated and another major cereal, winter wheat, will be screened. The importance of including winter crops in these studies is because it is known that plants are more susceptible to pollutants under conditions of slow growth and higher levels of pollutants may be expected in winter months. Further species to be screened will be lichens; these have been successfully used as bio-monitors of sulphur dioxide pollution but their responses to wet acidic pollutants have not yet been quantified.

The term "acid rain" is usually used to incorporate both wet and dry depositions of acidic pollutants. Since one form of pollution is rare without the other in Wales, this broader term is more useful and realistic when defining the polluted environment. Studies on the effects of typical urban concentrations of sulphur dioxide on plants have been conducted for several years but the effects of rural concentrations are less well documented. Similarly, studies on the effects on plants of nitrogen oxides have received little attention despite the fact that they are now known to occur at almost equal concentration to sulphur dioxide at many locations. In order to assess the impact of acidic pollutants on the Welsh environment it is essential to include studies on the effects of ambient concentrations of nitrogen oxides and sulphur dioxide on plants in addition to wet acidic inputs. It is intended to develop this theme in 1985/86. Field and greenhouse fumigation chambers and a sulphur dioxide air monitor are already available at Bangor and it is hoped to expand the facilities to allow the handling of nitrogen oxides. At a later stage, it is hoped that the equipment will be utilised to carry out the urgently required monitoring of these gaseous pollutants in crucial areas of Wales.

Following the encouraging results obtained in the initial radiometer study, it is hoped to expand this work by measuring the reflectance of light from the foliage of other species exposed to different levels of both wet and gaseous forms of acidic pollutants. The possibility of utilising this technique in field surveys will be assessed.

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