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YEAR TO YEAR VARIATION IN THE FUNGUS SPORE CONTENT OF THE ATMOSPHERE

By

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INTRODUCTION

Fungus spores were first shown to cause "bronchial catarrh" when *Charles Blackley* (1873) inhaled the spores of *Penicillium glaucum* and *Chaetomium*, but it was *Feinberg* (1935, 1936) of Chicago who first proved that they were of considerable importance in asthma. *Hyde, Richards & Williams*, (1956) report that "allergy to mould spores in all probability accounts for some 5 % of all cases of asthma at Cardiff and may well play a subsidiary part in many more", while *Gregory & Hirst* (1952) state that the basidiospores of the toad-stools and bracket-fungi occur in the air in such numbers that they "obviously need investigating in connection with seasonal asthma".

Many surveys of fungus spores have been conducted by the culture plate method, (*Bernstein & Feinberg*, 1942; *Hyde & Williams*, 1949; *Richards*, 1956) or by exposing sticky slides, horizontally, in a suitable shelter (*Durham*, 1944; *Hyde & Williams*, 1946). *Gregory* (1950) has shown that both methods are inefficient, especially for sampling small particles, and neither gives the volumetric spore content of the atmosphere. Culture plates record only those fungus spores that will grow on the medium used and these will be shown to have been less than half the spores recorded in this survey. The only means, therefore, of investigating the entire air-spora is by counting the spores deposited on a sticky slide that has been

exposed in an efficient trap for 24 hours. *Pady* (1957) and *Kramer et al.* (1959) exposed culture plates and slides in a Slit Sampler, through which a known volume of air was drawn, but only for half an hour each day. Although sampling was efficient and volumetric counts were obtained, the short exposure did not sample adequately spore types that have been found to exhibit a definite diurnal periodicity. The Hirst Automatic Volumetric Spore Trap (*Hirst*, 1952) was chosen for the present survey because it catches small spores (down to 3 microns) as efficiently as large ones, samples continuously throughout the 24 hours and is much less affected by changes in wind speed than the two early methods of sampling. Since its invention *Hirst* (1953) has studied diurnal periodicity of fungus spores and the effects of weather at Harpenden, England and with *Gregory* he has sampled the air over crop plants throughout the summers of 1951 and 1952 (*Gregory & Hirst*, 1952, 1957). *Gregory & Sreeramulu* (1958) have surveyed the air over an estuary in Southern England, *Hamilton* (1959) has compared the pollen and spore counts at London and Harpenden during the summer months of 1954 and 1955 and *Lacey* (1962) has studied the summer air-spores of two contrasting adjacent rural sites. From 1 January 1958 to date a continuous survey of the pollen and fungus spore content of the air over London has been made (*Hamilton & Denny*, 1960; *Davies*, 1962). The present investigation forms part of the long term survey of pollen and fungus spores as allergens which was begun at Cardiff in 1942 (*Hyde & Williams*, 1944; *Hyde & Adams*, 1960) and was undertaken to determine, not only the day to day changes in the air-spores during the summer months but, to what extent the daily concentrations differed between winter and summer and from year to year.

METHOD

The trap was situated on the roof of the National Museum of Wales, 60 feet above ground level. The Museum stands at an altitude of 45 feet in an area which is built up to the west, south and east and is suburban in character to the

north. The country around Cardiff is mainly agricultural with grass, arable land and scattered woods. Within the city there are numerous parks containing herbaceous plants, shrubs and trees, and the suburban streets are tree lined.

The Hirst trap and its operation has been described elsewhere (Hirst, 1952). A known volume of air is drawn through a narrow slit which is always directed into the wind, and the particles carried are impacted on to a microscope slide coated with glycerin jelly. The slides, which move past the intake slit at the rate of 2 mm per hour, were changed at 0900 hours and mounted daily. A traverse, 110 μ wide, was microscopically scanned lengthwise, with a 1/7" oil immersion fluorite objective together with compensating eye-pieces. From this sample of known volume, the daily mean concentrations in spores/m³ was obtained for each spore type. The 1/7" oil immersion objective simplified the scanning, particularly of small hyaline (colourless) spores, which had been carried out with a 1/6" objective before November 1958.

Cladosporium only, was counted from May 1954 when the trap was first installed; six additional spore types, namely *Alternaria*, *Epicoccum*, *Erysiphe*, *Polythrincium*, Rust uredospores and *Ustilago* were counted from January 1956 and a complete survey of all fungus spores was made from January 1958 to December 1961. In addition, at the height of the seasons of the 17 most important spore types, at least seven consecutive slides were scanned at two hourly intervals across the deposited trace to determine the time of day of the maximum catches of each type.

By the courtesy of the Medical Officer of Health, City of Cardiff, the meteorological information was obtained from the Penylan Observatory, Cardiff. This station is situated 1½ miles north-north-east of the Museum and the readings were taken once a day at 0900 hours.

COMPOSITION OF THE CATCH

The spores caught on the exposed slides were derived from all four classes of fungi, namely Phycomycetes, Ascomycetes, Basidiomycetes and Fungi Imperfecti.¹ Their relative abundance and variation in quantity from year to year for the years 1958-61 are illustrated in Fig. 1 and the average mean daily concentrations, in spores per cubic metre over twelve months, for the most important spore types in these classes are shown in Table 1 for the same years; but no indication

¹ In addition to the fungus spores the following were caught in small numbers each year. Small colonies of a blue-green alga; spores of myxomycetes, mosses, bracken, ferns and horse-tails. These, together with pollen grains, insect and plant fragments and inorganic particles are outside the scope of this paper.

FUNGUS SPORES AT CARDIFF 1958-61
 PROPORTIONS OF MAJOR GROUPS
 AVERAGE MEAN DAILY COUNTS OVER 12 MONTHS
 SPORES PER CUBIC METRE

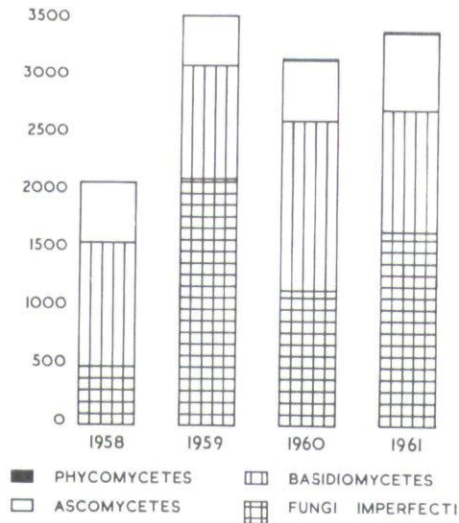


Fig. 1.

Proportions of the main classes of the Fungi, 1958-61. The catch of the Phycomycetes was too small in 1958 and 1959 to show clearly.

is given here of the abundance of the hourly, daily and seasonal catches that are registered by each type. Some indication of the sources of the spores belonging to these four classes is given below:

1. Some *Phycomycetes* are parasitic on insects, while others cause destructive mildews and blights on crop plants and rot fruit and vegetables in store.

2. The *Ascomycetes* or Sac Fungi form a large group, the members of which differ greatly in appearance from one another, but they nearly all consist of a fruiting body, embedded in or growing on the host, which encloses a number of sac-like structures called asci; there are usually eight ascospores in each ascus. Many *Ascomycetes* are inconspicuous parasites which cause diseases in both wild and cultivated plants and

Fungus sports over Cardiff, 1 January 1958-31 December 1961. Average daily mean concentrations in spores per cubic metre.

TABLE I

	1958	1959	1960	1961	Average 1958-61	% of total catch over 4 years, 1958-61
1. <i>Phycomycetes</i>	1.0	1.5	2.0	3.5	2.0	0.06
2. <i>Ascomycetes</i>						
a) <i>Erysiphe</i>	2.0	5.0	4.0	6.0	4.2	0.1
b) <i>Ascospores</i> (coloured)	92.0	48.3	76.5	87.5	76.1	2.5
c) <i>Ascospores</i> (hyaline)	432.5*	382.5	459.0	598.0	468.0	15.2
3. <i>Basidiomycetes</i>					548.3	17.8
a) <i>Agaricales</i> Basidiospores (coloured)...	189.0	176.5	292.5	239.5	224.4	7.3
<i>Agaricales</i> Basidiospores (hyaline) ...	498.0	360.5	805.5	493.5	539.4	17.5
b) <i>Lycoperdales</i> (Puff Balls)	1.0	0.2	0.3	0.6	0.5	0.01
c) <i>Sporobolomycetaceae</i>						
<i>Sporobolomyces</i>	295.0	298.0	256.5	235.5	271.2	8.8
<i>Tilletiopsis</i>	22.0*	17.5	21.0	13.0	18.5	0.6
d) <i>Uredinales</i> (rusts)	6.0	10.0	3.5	7.0	6.6	0.2
e) <i>Ustilaginales</i> (smuts)	61.0	125.6	86.5	67.0	85.0	2.7
4. <i>Fungi imperfecti</i>					1145.6	37.1
a) <i>Alternaria</i>	6.2	8.5	9.0	8.0	8.0	0.3
b) <i>Botrytis</i>	27.5	25.0	34.5	30.0	29.2	0.9
c) <i>Cladosporium</i>	409.0	1964.0	979.0	1465.0	1204.2	39.1
d) <i>Epicoecum</i>	9.0	36.5	7.0	9.0	15.4	0.5
e) <i>Penicillium</i> and <i>Aspergillus</i>	14.5	36.0	73.5	75.0	49.8	1.6
f) <i>Polythrincium</i>	7.5	9.0	7.5	6.5	7.6	0.2
g) <i>Pullularia</i>	14.0	26.0	31.0	38.5	27.5	0.9
h) <i>Torula</i>	4.5	13.5	7.5	9.5	8.7	0.3
i) <i>Minor types</i>	8.0	9.0	9.7	17.9	11.0	0.4
5. <i>Undetermined and damaged</i>					1361.4	44.2
Total.....	18.3	20.9	29.0	30.5	24.7	0.8
Total.....	2118.0	3574.0	3195.0	3441.0	3082.0	100.0

* Re-examination of 1958 slides has shown that a hyaline ascospore was, in error, counted as *Tilletiopsis*. The average daily mean concentrations of both spore types have been adjusted in accordance with these findings.

often destroy them. Others, which produce large, often brightly coloured, fruiting bodies live on the soil or decaying logs and leaves.

Most of the obvious Ascospores could not be identified with certainty, so the coloured spores were separated from the colourless (hyaline) ones.

3. *Basidiomycetes*.

(a) The *Agaricales* (gilled and pored fungi) are the familiar mushrooms, toadstools and bracket fungi. Although some of the Basidiospores are readily identifiable, the bulk of them have to be grouped according to colour.

(b) The *Lycoperdales* include the common puff balls and earth stars.

(c) *Sporobolomycetaceae*. The false yeasts *Sporobolomyces* and *Tilletiopsis* are microscopic fungi growing on dead and living leaves of cereals, ferns and flowering plants. They do not appear to be either harmful or beneficial to their hosts.

(d and e) The rusts and smuts are both microscopic parasitic fungi and cause great losses to cultivated crops, particularly cereals.

4. *Fungi Imperfecti*. With the exception of *Polythrincium*, which causes "Black Blotch" disease of clover, the Fungi Imperfecti listed here are common moulds found on living plants or dead organic substances and are recorded regularly during culture plate surveys.

PHENOLOGY

The calendar (Fig. 2), which shows the time of year when the 17 most important spore types were caught, has been compiled from the *average dates* over the four years 1958-61 on which each type first reached a daily mean concentration of at least one spore/m³, 200 spores/m³, 2,000 spores/m³ and 10,000 spores/m³, and the last dates when such concentrations were reached. The Hyaline and Coloured Ascospores and Basidiospores, *Cladosporium*, *Sporobolomyces* and "*Penicil-*

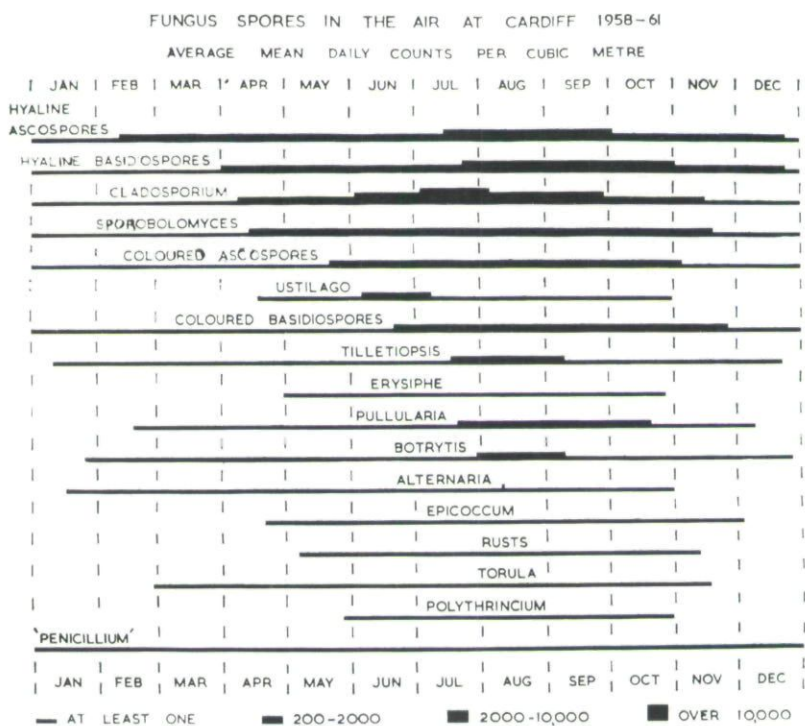


Fig. 2.

Fungus spore calendar, 1958-61. 'Penicillium' excepted, the spore types are arranged in the order in which they first became abundant. The concentrations indicated were attained during the periods shown, although not necessarily every day. (See text).

lium" were caught from 1 January to 31 December in all four years but *Tilletiopsis*, *Pullularia*, *Botrytis* and *Alternaria* were recorded as early as 1 January or as late as 31 December in some years only. With the exception of *Torula* the remaining six types did not appear in the air in any year until the end of March and most of them disappeared regularly by the end of November. The Phycomycetes are not included in this diagram because the catches were very small and intermittent.

As a measure of abundance and great abundance the, purely arbitrary, daily mean concentrations of 200 spores/m³ and 2000 spores /m³ have been chosen. Eleven spore types were

regularly caught in abundance. The Hyaline Ascospores and Basidiospores (both composite groups) had the longest periods at this level and they were sometimes caught in these numbers from January to December, while *Tilletiopsis*, *Botrytis* and *Alternaria* had abundant catches in some years only. The "*Penicillium*" group recorded 200 spores/m³ on many days each year, particularly during the winter months; but there was no well defined period, such as was found in the other eleven spore types.

Cladosporium, the Hyaline Ascospores and Basidiospores (both composite groups) were the only types regularly caught in great abundance (daily mean concentration, 2,000–10,000 spores/m³). *Sporobolomyces* and *Ustilago* reached this level very occasionally in some years.

Only *Cladosporium* regularly reached a daily mean concentration of over 10,000 spores/m³ and it did so some time between 16 June and 15 October. The length of this period varied and was as long as 15 weeks in 1959. (see the section on *Cladosporium* and Figs. 13 and 14.)

INCIDENCE OF THE PRINCIPAL TYPES OF SPORES

Each spore type listed in Table 1 is dealt with separately and an account given of its hourly, daily, monthly and yearly variations. An attempt has been made to correlate weather conditions and the sources of supply of some spores with these variations.

1. PHYCOMYCETES

This composite group consisted mainly of the sporangia of *Peronospora*, *Cystopus* and *Entomophthora*, together with very few *Phytophthora* and spores that resembled *Mucor* and *Rhizopus*.

Spores were caught in all months except January; one-third arrived in September and one-third in July and August together (Fig. 3). Half the spores caught resembled those of *Peronospora* and *Cystopus*; these, together with *Phytoph-*

PHYCOMYCETES

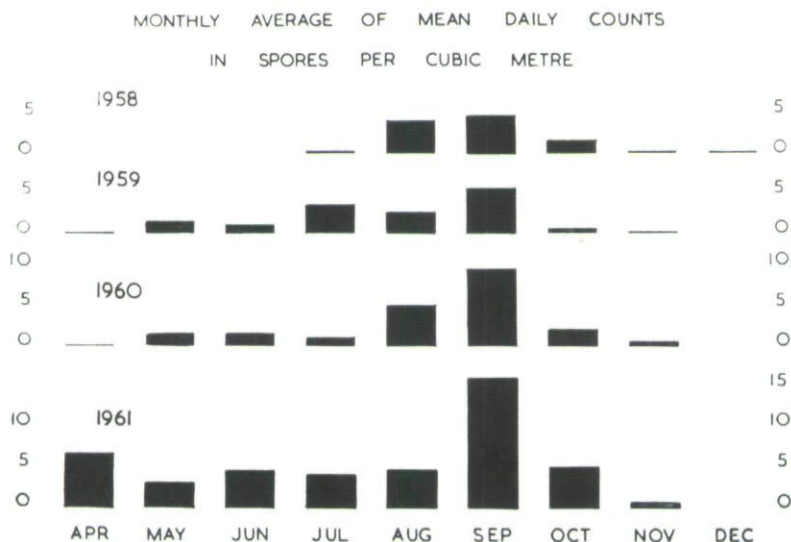


Fig. 3.

Variation in the incidence of miscellaneous Phycomycetes, April to December, 1958-61. The highest daily mean count was only 99 sporangia/m³ in September, 1961. (See text).

thora, were observed on crops growing near Cardiff by the officers of the National Agricultural Advisory Service in all years.¹ The sporangia of the insect attacking fungus *Entomophthora* accounted for a further 40%. The numbers of spores caught were, at all times, too low to give a reliable periodicity count or to show clearly the weather conditions favourable for the dispersal of the sporangia but on those days when the concentrations were relatively high the rainfall was nil or slight and the humidity 70-96%. *Entomophthora* sporangia appeared mainly between 0500 and 1400 hours, while those of *Peronospora* and *Cystopus* were scattered throughout the day. The highest counts recorded were 180 sporangia/m³ at 1000 and 1200 hours.

¹ I am indebted to Dr. J. Bradley-Jones of the National Agricultural Advisory Service for reports on crop diseases in South Wales and for material to make standard slides of a number of spore types.

2. ASCOMYCETES

a) *Erysiphe*.

The conidia of *Erysiphe* (largely *E. graminis*, wheat mildew) grow in chains on the surface of the host plant and, although they were usually found singly on the slides, short chains of up to four conidia were observed.

98 % of the six years' catch arrived between May and October, with 64 % in June and July (Fig. 4). The daily mean concentration rarely exceeded 200 spores/m³ and the counts were highest during 1957, 1959 and 1961, being 217, 109 and 130 % of average. *Erysiphe graminis* was recorded on cereal crops near Cardiff in June and July every year but in 1957 it was recorded also during April, May and August and, in addition, other species of *Erysiphe* were recorded on other

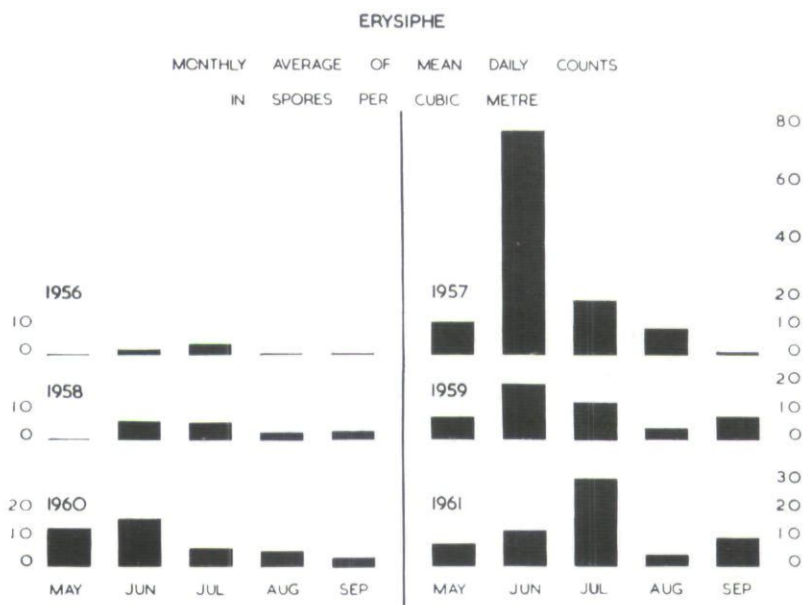


Fig. 4.

Variation in the incidence of *Erysiphe* conidia from May to September, 1956-61. No spores were caught between December and March. The highest daily mean count was 210 spores/m³ in June, 1957. (See text).

crops; this, no doubt, accounted for the outstanding catch of that year. There were more conidia in the air when it was dry, the temperature high (Maximum 70° – 85° F) and the humidity low (50–65 %). This type of weather occurred more frequently during the early summers of 1957, 1959, 1960 and 1961 than in the other two years. Maximum concentrations were recorded between 1100 and 1900 hours G.M.T.; the highest count being 483 spores/m³ at 1700 hours.

b) *Coloured Ascospores.*

Only 14 % of all ascospores caught were coloured. They were recorded throughout the year but 60 % arrived in August, September and October (Fig. 5). Catches were highest in wet weather, when the humidity was over 80 % and the maximum temperature 55 – 70° F. Counts were only 63 % of average in the hot, dry late summer and autumn of 1959 and 121 % in 1958, which was generally warm, wet and humid. Concentrations of over 200 spores/m³ were regularly recorded (Fig. 2) and a large proportion of the spores observed were of two distinct types. The yellow-brown, fusiform and generally 3-septate spores belonged to the genus *Leptosphaeria*; the most common ones were like *Leptosphaeria do-liolum* (as described by Dennis, 1960). The second group were much smaller, pale fawn, sausage-shaped and non-septate. They resembled the genus *Diatrype*; a fungus which is very common on the dead wood of deciduous trees. Spores which resembled those of *Chaetomium*, *Pleospora*, *Venturia*, *Perisporium*, *Ascobolus*, *Geoglossum*, *Melanomma*, *Xylaria* or *Daldinia*, *Melogramma*, and *Cryptosphaeria* were observed in smaller numbers, together with many unknown types.

Coloured ascospores showed a clear diurnal periodicity; maximum concentrations were observed between 2300 and 0900 hours and the highest count recorded was 5,700 spores/m³ at 0500 hours.

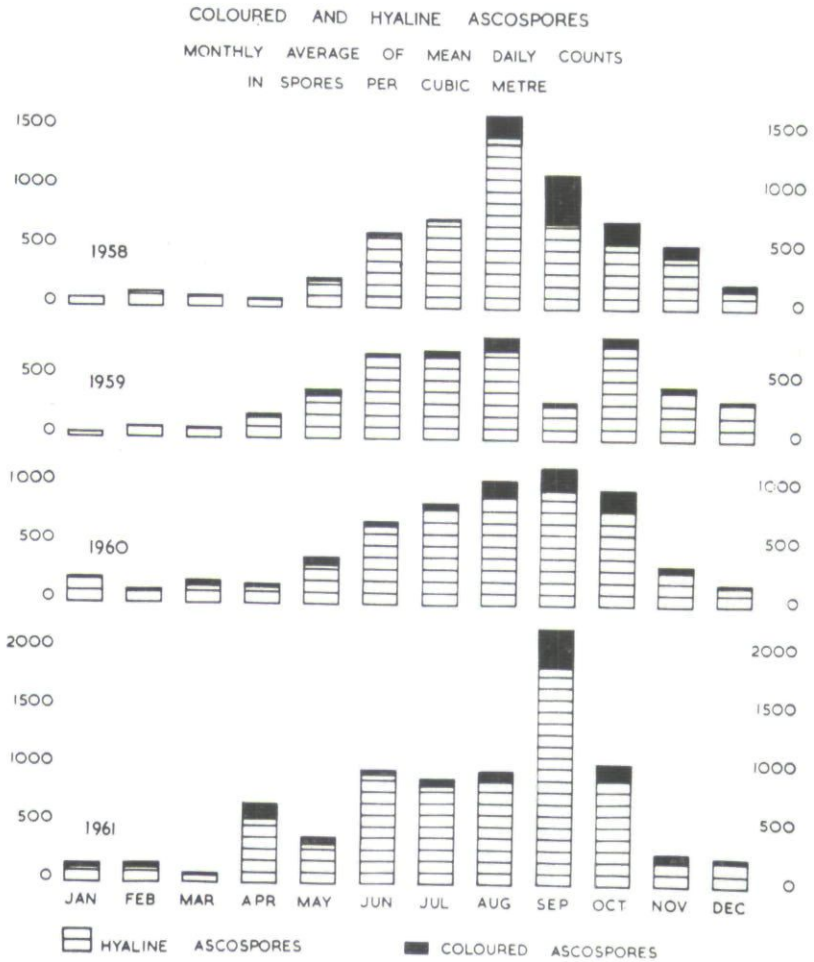


Fig. 5.

Seasonal variation in the incidence of Ascospores, 1958-61. The highest daily mean counts: Coloured Ascospores, 1700 spores/m³ in September, 1958. Hyaline Ascospores, 7,600 spores/m³ in September, 1961. Unusually high catches in late April and early May, 1961 were also recorded in London (*Davies, 1962*) and Aberystwyth (*Hyde & Adams, unpublished*). (See text).

c) *Hyaline Ascospores.*

The majority of these spores were caught between June and October (Fig. 5). Maximum concentrations were greater and

spread over a longer period than were those of the coloured spores (Fig. 2). Catches were highest in wet weather, when the humidity was over 80 % and the maximum temperature below 70° F. After a dry period high concentrations of spores were observed on the slides within a few hours of rain falling. The catch was abnormally low in the hot, dry September of 1959.

Over thirty different types of hyaline ascospores have been observed regularly; small, rod-like, non-septate spores predominated. Hyaline rods formed a large proportion of *Hamilton's* (1959) ascospore catch, and *Lacey* (1962) suggests that they might be of the Discomycete genus *Orbilina*. The large numbers of elliptic-cylindric or fusiform, one-septate spores which varied in size from $6 \times 1.5\mu$ to $24 \times 5\mu$ were obviously derived from a number of genera; the larger ones probably from the common fungus *Nectria cinnabarina*. Long filiform spores (which resembled *Epichloe typhina*, a very common parasite on many grasses) were caught in small numbers between July and October but were completely absent in the hot, dry August and September of 1959. *Gregory & Sreeramulu* (1958) caught these spores immediately after rain and found that they were completely absent in dry weather.

Maximum concentrations of all hyaline ascospores were found during the night and early morning, 0100–0900 hours; the highest catch recorded being 15,000 spores/m³ at 0400 hours.

3. BASIDIOMYCETES

a) The Agaricales.

Coloured Basidiospores.

These spores were recorded throughout the year and formed about one third of all basidiospores caught. 80 % arrived between July and November and, September 1959 excepted, the maximum concentrations were in August, September and October, (Fig. 6). The highest counts were recorded a few days or a week after a period of substantial rain. On those

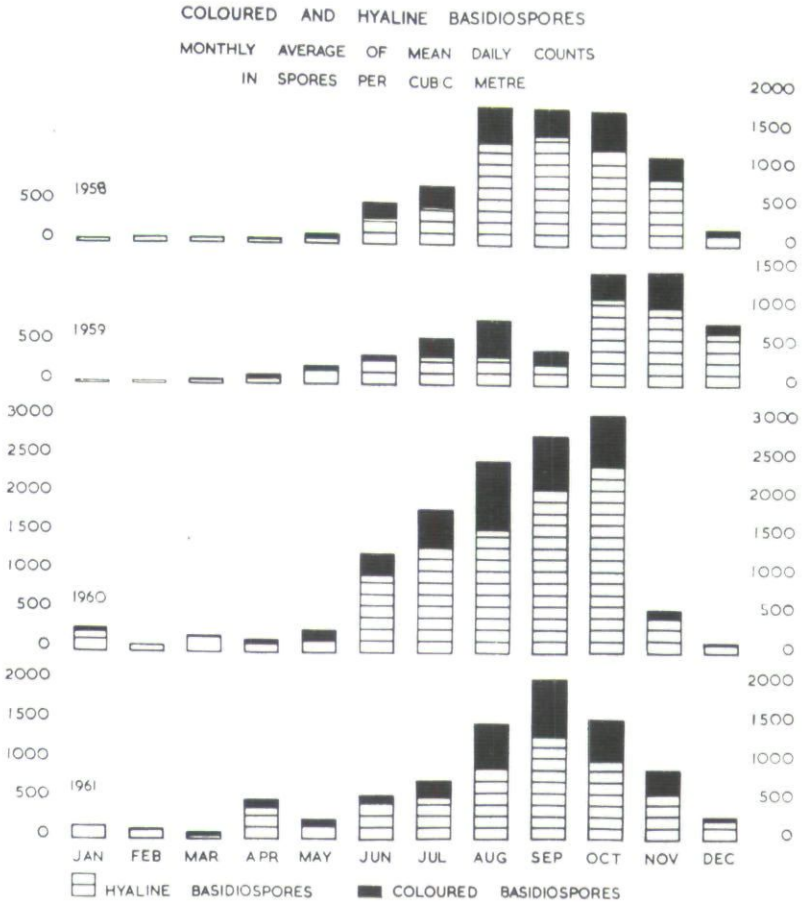


Fig. 6.

Seasonal variation in the incidence of the Basidiospores, 1958-61. The highest daily mean counts: Coloured Basidiospores, 3000 spores/m³ in September, 1961. (The only day the catch exceeded 2000 spores/m³). Hyaline Basidiospores, 8300 spores/m³ in June, 1960 (one outstanding high count). September and October, 1960 had relatively high daily counts throughout. (See text).

days of maximum concentration it was warm (mean temperature 56°-66° F), the humidity was usually over 80 % and there was little or no rain. *Gregory & Hirst* (1957) found that their peaks occurred during warm spells about two weeks after rain. Counts were 78 % of average in the hot, dry late

summer of 1959 and 130 % in 1960 when rainfall and humidity were above normal and the temperature was favourable. *Ganoderma*, a bracket-fungus growing on the trunks of old trees, has easily recognisable spores which were counted separately. Their average daily mean concentration over the four years was 19 spores/m³; 8.5 % of all Coloured Basidiospores. Other distinctive types observed were *Boletus* spp., *Hypholoma* spp., *Coprinus* spp., *Psilocybe* spp., *Entoloma* spp., *Nonalea staurospora* and *Inocybe asterospora*. In addition, at least 40 other different types of Coloured Basidiospore were seen.

The spores were caught at all times during the 24 hours but the majority of them arrived between midnight and 0400 hours; the highest concentration recorded was 4500 spores/m³ at 0400 hours.

Hyaline Basidiospores.

The majority of these colourless spores resembled those of the Bracket Fungi and the Tremellales, (Gregory & Hirst, 1952); fungi which grow on trees and logs. Only the spores of the cap fungi, *Russula* and *Lactarius* (ornamented with spines and ridges), were distinctive but they formed less than 1 % of the whole group. Although in the air throughout the year 87 % were caught between June and November and, September 1959 excepted, the maximum concentrations were recorded from August to October (Fig. 6). The highest counts were obtained when it was wet, the humidity was high (usually over 85 %) and the mean temperature 50°–63° F. The response to rainfall and increasing humidity was rapid; after a hot, dry period a high concentration of spores appeared on the slides within a few hours. This behaviour was observed by Gregory & Hirst (1952) and Gregory & Sreeramulu (1958). Counts were only 66 % of average in the hot, dry, late summer of 1959 and 150 % in 1960 when the weather was favourable. This range is greater than that recorded by the coloured spores.

Although some spores were caught during the day the ma-

jority were recorded at night and the maximum concentrations were usually between 0200 and 0600 hours; the highest count recorded was 15,500 spores/m³ at 0000 hours.

b) *Lycoperdales*. (Puff Balls).

96 % of the very few spores observed were recorded in August, September and October. The highest catches were made in the wet August and September of 1958 when the average humidity was 85 % and 84 % respectively and the temperatures were above normal for the time of year. The highest daily mean count recorded was only 126 spores/m³; the numbers were too low to give a reliable time of day of maximum concentration.

c) *Sporobolomycetaceae*.

Sporobolomyces.

85 % of the minute, hyaline spores of *Sporobolomyces* arrived in the five months June to October (Fig. 7). The mean daily concentrations during this period oscillated violently from day to day and occasionally reached over 2000 spores/m³. This fungus appeared to be particularly sensitive to changes in weather conditions, and there was a rapid increase in spore concentration when rain, high humidity (over 80 %) and moderate temperature (maximum 60°–70° F) followed a hot, dry period; as is exemplified in Table 2.

During the unusually hot, dry month of September, 1959, the catch was consistently low.

Although some spores were found on the slides during the day the bulk of them were recorded between 2300 and 0700 hours with the maximum concentration about 0500 hours (see Fig. 21); the highest recorded was 108,000 spores/m³ at 0500 hours. This catch was small compared with that obtained by Gregory & Sreeramulu (1958) in Chichester Harbour, who at times recorded about a million spores/m³. Their apparatus was only 6 ft. above ground level as compared with 60 ft. in

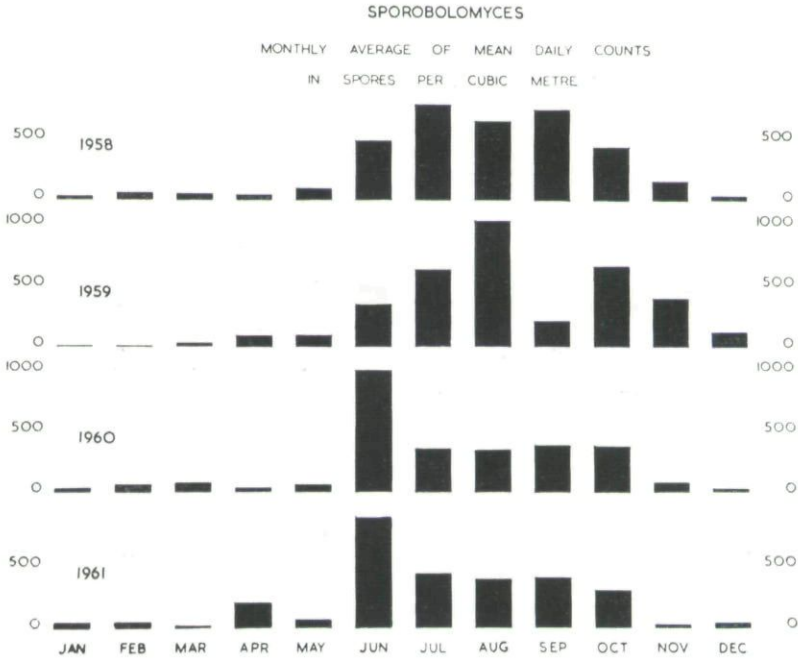


Fig. 7.

Seasonal variation in the incidence of *Sporobolomyces*, 1958-61. The unusually high daily mean concentrations of 24,500 spores/m³ in June, 1960 and 10,700 spores/m³ in June, 1961 accounted for the high averages for those months; other daily counts did not exceed 5000 spores/m³. (See text).

this survey. They state that, in the early morning when conditions are favourable for maximum concentrations, "thermal and frictional turbulence are low and the spores may remain concentrated in the layer of air near the ground".

TABLE 2

1960	Maximum temp. °F.	Rainfall in inches	Relative humidity	Daily mean concentration in spores/m ³
June 21.	78	0	62%	120
June 22.	79	0.47	76%	530
June 23.	64	0.18	96%	24,500
June 24.	68	0	96%	356
June 25.	75	0	75%	5

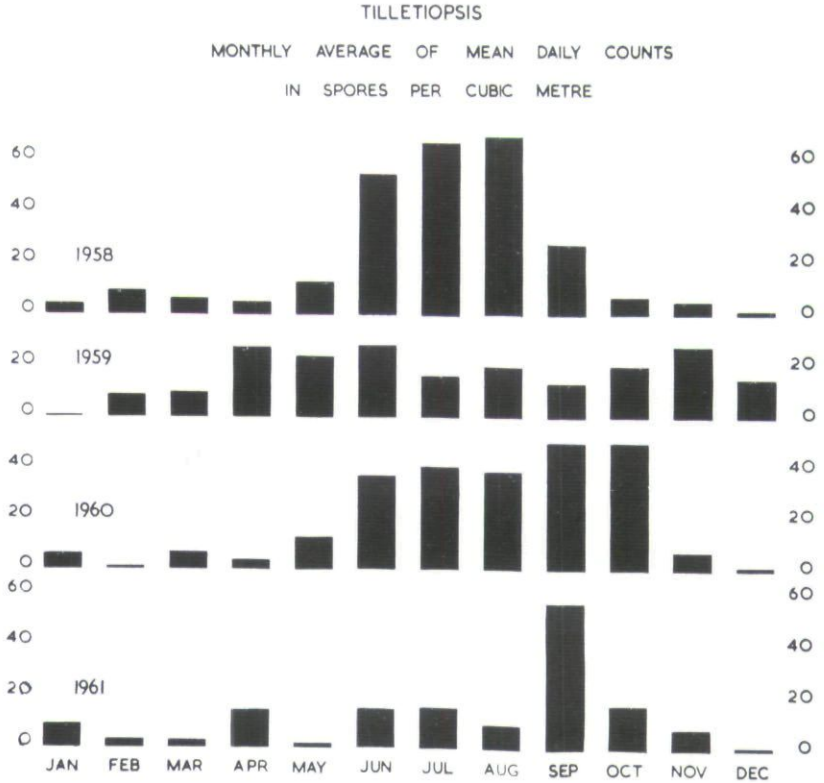


Fig. 8.

Seasonal variation in the incidence of *Tilletiopsis*, 1958-61. The highest daily mean count was 576 spores/m³ in September, 1961. The very low catches from June to August, 1961 were caused mainly by low relative humidity during these months. (See text).

Tilletiopsis.

The hyaline, "hinged" spores were caught in all months but the majority were usually recorded from June to October (Fig. 8). With the exception of 1959, daily mean concentrations of over 200 spores/m³ were registered each year. Catches were highest when it was warm (maximum temperature 60°-70° F), and the relative humidity was greater than 80 % but the rainfall was not critical. Relatively high concentrations

were recorded even on dry days if the humidity was high and the maximum temperature below 70° F, as is exemplified in Table 3.

TABLE 3

1958	Maximum temp. °F.	Rainfall in inches.	Relative humidity	Daily mean concentration in spores/m ³
July 8.	73	0	75%	54
July 9.	69	0	81%	396
July 10.	70	0	68%	27
July 11.	71	0.44	56%	27
July 12.	64	0.17	97%	171

Low daily mean concentrations were recorded in the months, July to October 1959 and June to August 1961; the relative humidity during these periods was usually below 80 % and the maximum temperature above 70° F.

Very few spores were caught between 0900 and 2100 hours; maximum concentrations were found between 2300 and 0300 hours, the highest count recorded being 2800 spores/m³ at 0300 hours.

d) *Uredinales* (Rusts).

The uredospore stage of *Puccinia glumarum* (Yellow Rust of Wheat) and *P. coronata* (Brown Rust of grasses) were the most common rust spores observed; *P. graminis*, *P. triticea*, *Uromyces* spp. and *Melampsora* spp. were also caught. 40 % of the small catch was recorded in September and a further 45 % in July, August and October (Fig. 9). The daily mean concentration rarely exceeded 100 spores/m³. Most spores were caught on dry days; temperature and humidity varied. In 1959, when the catch was 200 % of average, September was a very dry month and rusts were reported to be present on a number of crops near Cardiff. In August and September 1956 (catch only 44 % of average) the rainfall was above normal and no outbreaks of rust diseases were reported.

Maximum concentrations of Rust uredospores were from

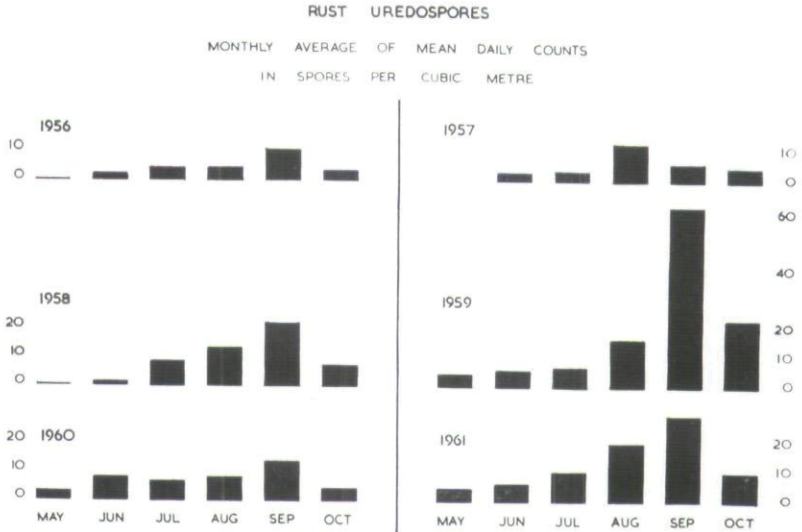


Fig. 9.

Variation in the incidence of miscellaneous Rust Uredospores from May to October, 1956-61. Spores were rarely recorded between December and April. The highest daily mean count was 162 spores/m³ in September, 1959. (See text).

1300-2100 hours, and only a very few spores were observed at night. The highest catch recorded was 576 spores/m³ at 17 hours.

e) *Ustilaginales*. (Smuts).

The spores of *Ustilago avenae* (Loose Smut of oats) together with, probably, *U. nuda* (Loose Smut of wheat and barley) and *U. hordei* (Covered Smut of barley and oats) formed the bulk of the catch of the *Ustilaginales*. They were caught singly or in clumps from mid-April to mid-November but 90 % arrived in June and the first half of July in all years (Fig. 10). Spore concentrations were highest when the maximum temperature was over 70° F, there was little or no rain and the relative humidity was 50-70 %. These conditions occurred frequently in June 1957, 1959 and 1960 when the catch was 152, 131 and 118 % of average; *Ustilago avenae*, *U. nuda* and *U. hordei* were seen on crops near Cardiff in

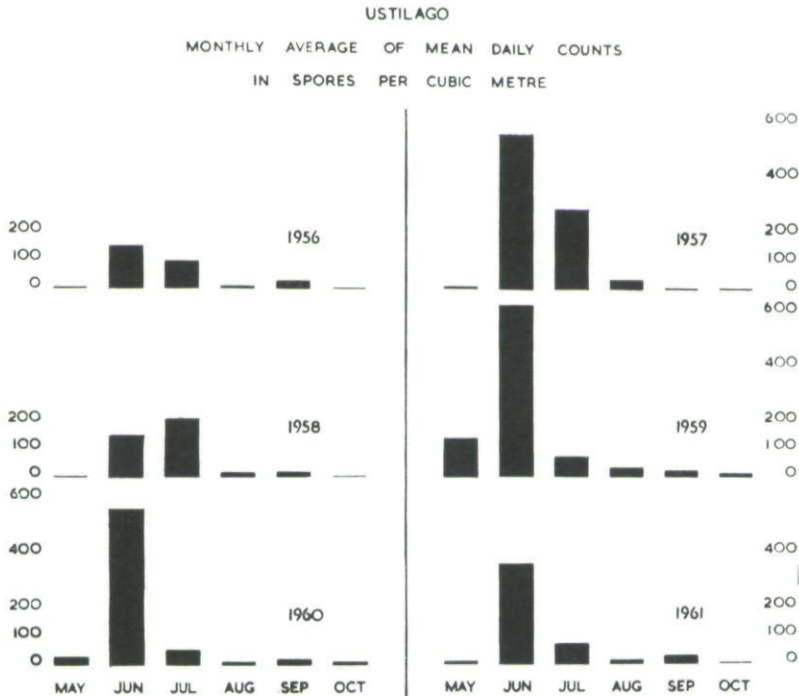


Fig. 10.

Variation in the incidence of *Ustilago* from May to October, 1956-61. Only an occasional spore was recorded between December and March. The highest daily mean count was 5200 spores/m³ in June, 1959. (See text).

these years. Outbreaks of these diseases were not reported in 1956 and 1958 and the weather did not favour high catches. There was a very mild outbreak of *U. nuda* only in 1961. With the exception of 1956 and 1958 a daily mean concentration of over 2,000 spores/m³ was recorded on one or two days each year in June.

The bulk of the daily catch arrived between 1500 and 2100 hours, G.M.T. and the highest count recorded was over 15,000 spores/m³ at 2000 hours.

Tilletia and *Urocystis* were recognised and recorded together with some unknown smuts, but they formed a very small proportion of the catch of the *Ustilaginales*.

4. FUNGI IMPERFECTI

a) *Alternaria*. Culture plate and slide surveys have shown that *Alternaria* spores occur with great frequency in the air in the U.S.A. (Durham, 1937, 1938; Morrow, Lowe & Prince, 1942; Pady, 1957 and Kramer, Pady & Rogerson, 1959b) but they accounted for only 0.2 % of Hamilton's (1959) catch at Harpenden, 1954, 1 % of all colonies recorded by Richards (1954) in his culture plate survey at Cardiff, 1951-53, and 0.3 % of the total catch in this survey. Durham (1938) thought that the most important source of the spores was the straw of wheat and other cereals infected with the fungus. Blackening of wheat ears due to *Alternaria* is known to occur sometimes on crops near Cardiff (Hyde & Williams, 1946) but it is not an important disease in the district. (Bradley-Jones, personal communication). The large, brown, beaked spores were caught in all months, but 46 % arrived in August and a further 35 % in July and September (Fig. 11). Hyde & Willi-

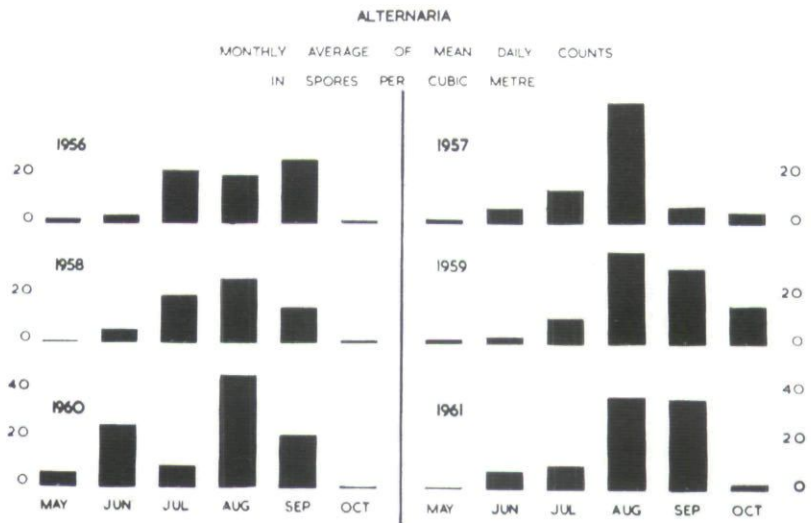


Fig. 11.

Variation in the incidence of *Alternaria*, May to October, 1956-61. Few spores were caught between November and April. The highest daily mean count was 400 spores/m³ in August, 1960. (See text).

ams (1946) working with gravity slides near Cardiff in 1942 and 1943, recorded similar results; August 48 %, July and September 40 %.

Daily mean concentrations were never high; 200 spores/m³ were recorded on one or two days only each year. Catches were highest when it was dry, the maximum temperature 70°–80° F, and the humidity below 85 %. Counts were generally low in the wet, humid Augusts of 1956, 1958 and 1960; one unusually high daily mean concentration in August 1960 accounted for the high average catch for that month.

Spores were caught all through the day and night but the maximum concentrations were between 1300 and 1600 hours; the highest count recorded was 760 spores/m³ at 1500 hours.

b) *Botrytis* is a grey mould found on a wide variety of plants and, under humid conditions, is particularly troublesome on soft fruits. The spores, which resembled those of *B. cinerea*, were caught in all months but 80 % were recorded from June to September (Fig. 12); the concentrations in these four months differed widely from year to year.

Outbreaks of *Botrytis* on various crops near Cardiff were reported from May to August, 1958–60; in 1961 the disease was not observed until September.

Concentrations were highest on warm, humid days after rain (Mean temperature over 60° F and relative humidity greater than 75 %) and when the winds were light (1–3 Beaufort Scale). Hamilton (1959) found that *Botrytis* spores in London and Harpenden were reduced during high winds.

In August and September 1958, September 1960 and 1961, when maximum counts were recorded, the relative humidity was above average for the month and usually over 80 %. Conversely, during June to September 1959, July 1960 and June to August 1961, when catches were below normal, it was dry and the humidity was below average for each month, and less than 75 %. Counts were never high; a daily mean of over 200 spores/m³ was recorded on a few days only each year. An extraordinarily high catch on one day in June 1960 accounted for that month's high average.

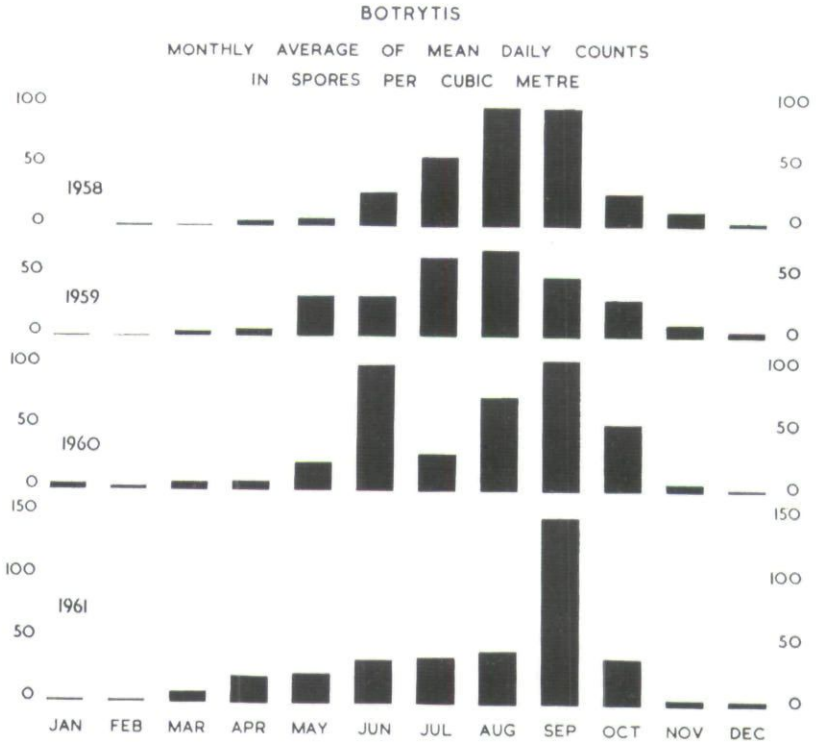


Fig. 12.

Seasonal variation in the incidence of Botrytis, 1958-61. The highest daily mean count was 810 spores/m³ in June, 1960. (See text).

Spores were caught at all times of the day and night but the bulk of them arrived between 1200 and 1900 hours; the highest catch was 1750 spores/m³ at 1700 hours.

c) *Cladosporium* grows on a great variety of dead organic substances all over the world and many species are now recognised. (*De Vries*, 1952). *Harvey* (1960) in Cardiff and *Kramer, Pady & Rogerson* (1959) in Kansas, both working with petri plates, found that *C. herbarum*, *C. cladosporioides* and *C. macrocarpum* made up 44.6 (53), 30.6 (29) and 17.1 (16.5) % of their *Cladosporium* catch. Although, while counting the spores on the slides, no attempt was made to discrimi-

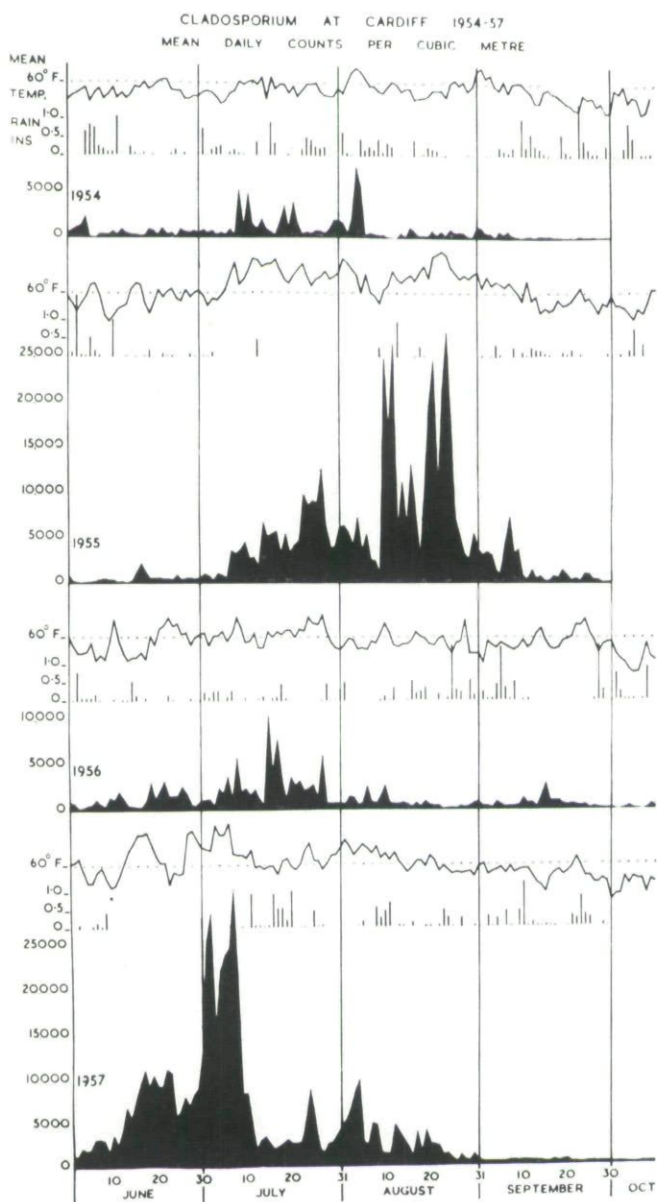


Fig. 13.

Daily counts of *Cladosporium* spores (1 June to 30 September, 1954-55 and 1 June to 10 October, 1956-57) together with daily mean temperatures and rainfall. High concentrations were usually recorded when the mean temperature was over 60° F and the rainfall was nil or slight.

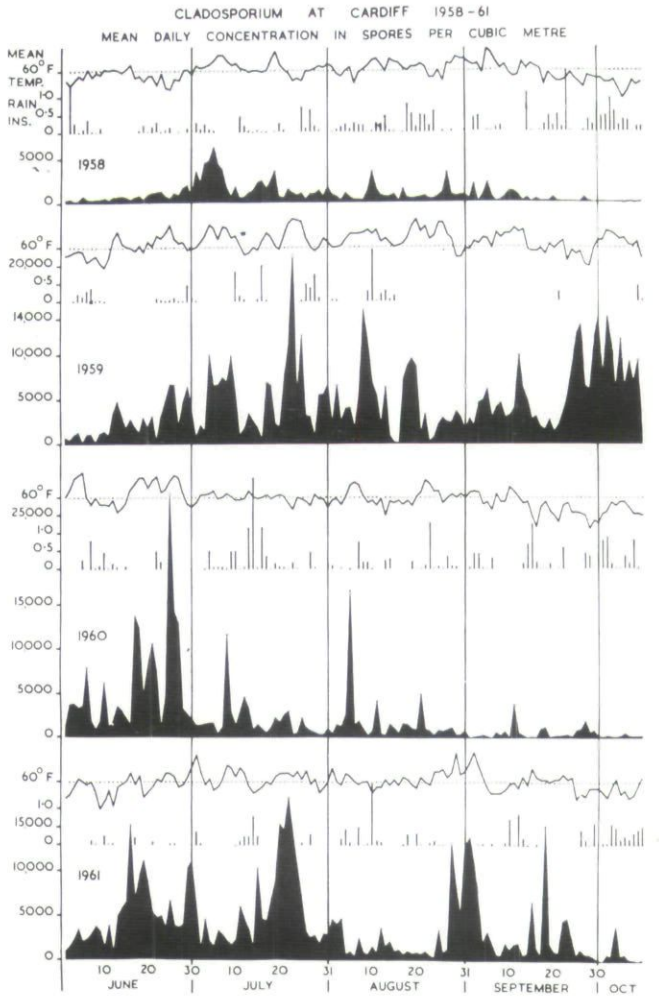


Fig. 14.

Daily counts of *Cladosporium* spores, 1 June to 10 October, 1958-61, together with the daily mean temperatures and rainfall. High concentrations were usually recorded on those days when the mean temperature was over 60°F and the rainfall was nil or slight. The catch in late September and early October, 1959 was abnormally high (for explanation, see text).

nate between the various species, it was observed that spores resembling those of *C. herbarum* and *C. cladosporioides*, which arrived either singly or in clumps, appeared to make up the bulk of the *Cladosporium* catch; the larger spores of *C. macrocarpum* were also seen.

Over the years 1958–61 *Cladosporium* formed 39 % of all fungus spores caught (Table 1). This compares with *Harpenden* 1952, 46.5 % (Gregory & Hirst, 1957); *Harpenden* 1954, 39 % (Hamilton, 1959) and *Kansas* 1956–58, 41 % (Kramer *et al.* 1959).

Figs. 13 and 14 show the day to day catches, together with mean temperature and rainfall, from 1 June to 10 October for the eight years 1954–61. Although *Cladosporium* was caught in all months it had a distinct summer maximum (see Calendar, Fig. 2) and over 80 % was recorded from June to September. The catch varied considerably in quantity from year to year during these months. Table 4 shows the average mean daily concentration for each month and the percentage of the 8 years average for each year; this last varied from 27 % in 1954 to 172 % in 1957. Maximum concentrations of *Cladosporium* spores were recorded when the mean and maximum temperatures were over 60° F and 70° F respectively and there was little or no rain (see Figs. 13 and 14). The summers of 1954, 1956, 1958 and 1960 (except June) were generally wet and, except for 1958, the average mean temperatures for each month were below 60° F; catches during these years were only 27, 44, 34 and 86 % of average. The summers of 1955, 1957, 1959 and 1961 were generally hot and dry and catches were 144, 172, 152 and 141 % of average. The average mean daily concentration during September 1959 was extraordinarily high (Table 4). During this month the maximum temperature averaged 70° F and the mean temperature over 60° F (for the only year during the eight years under review) and a very little rain fell on two days only. The high catches continued with the warm, dry weather until 9 October. On 10 October the temperature dropped (mean temperature 56° F) and rain fell; the mean daily concentration of

spores dropped from nearly 10,000 spores/m³ on the 9th. to 700 spores/m³ on the 10th.

TABLE 4
Cladosporium. Cardiff. 1954-61.
Average mean daily concentration in spores/m³.

Year	June	July	August	September	Average June to Sept.	% of 8 year average
1954	592	1358	954	261	791	27
1955	557	4728	10,315	1216	4204	144
1956	1172	2618	771	623	1298	44
1957	6277	10,538	2897	420	5033	172
1958	672	1966	907	459	1001	34
1959	2441	5517	4367	5471	4449	152
1960	5942	1784	1735	569	2507	86
1961	5259	5401	2690	3133	4121	141
Average 1954-61 (8 Years)					2923	100

For *Cladosporium* a mean temperature of over 60° F and little or no rain seem to be essential for mass spore production and liberation. This confirms the findings of *Hyde & Williams* (1953) and *Richards* (1956) who worked with petri plates in Cardiff, and *Hirst* (1953) and *Hamilton* (1959) who used a Hirst Trap in Harpenden and London. The highest catches of the year may be any time between late June and early October (Figs. 13 and 14), depending on the time of the most favourable weather conditions.

Cladosporium spores were caught all through the 24 hours but the majority of them arrived in the afternoons and the peak for the day was usually between 1200 and 1500 hours (Fig. 21). Maximum concentrations recorded were 62,000 spores/m³ at both 1500 and 1600 hours in 1955, and 46,000 spores/m³ at 1300 hours in 1957.

d) *Epicoccum* appears as dark spots on dead leaves and stems of numerous plants. It is common on decaying wood, paper and textiles, and has been isolated from soil, woodpulp, insects and human skin and sputum (*Schol-Schwarz*, 1959).

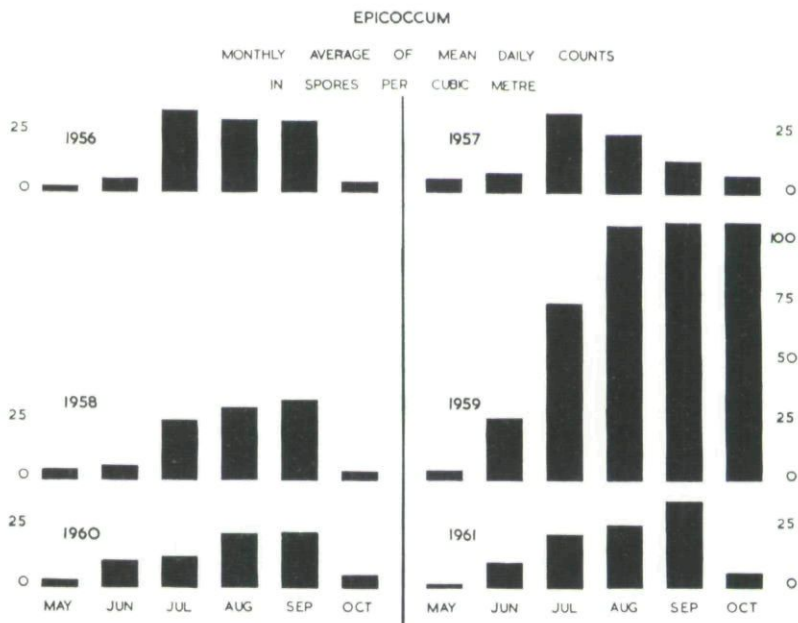


Fig. 15.

Variation in the incidence of *Epicoccum* spores, May to October, 1956-61. This fungus was rarely recorded from December to mid-April. The highest daily mean count was 288 spores/m³ in October, 1959; the catch in this month was abnormal (for explanation, see text).

The dark brown, warted conidia were observed from mid-April to mid-December but, 1959 excepted, over 75 % of the catch was recorded from July to September (Fig. 15). Maximum concentrations were observed on dry days when the relative humidity was below 80 per cent and the maximum temperature was 70°-80°F. In 1959, then the catch was 280 per cent of average, the unusual hot dry weather continued into October. Rain in the middle of that month brought the long run of relatively high catches to an abrupt end. Mean daily concentrations very rarely exceeded 200 spores/m³.

The spores were caught throughout the day and night but maximum counts were between 1200 and 1500 hours; the highest recorded being 612 spores/m³ at 1400 hours.

e) *Penicillium* and *Aspergillus* are widely distributed on a

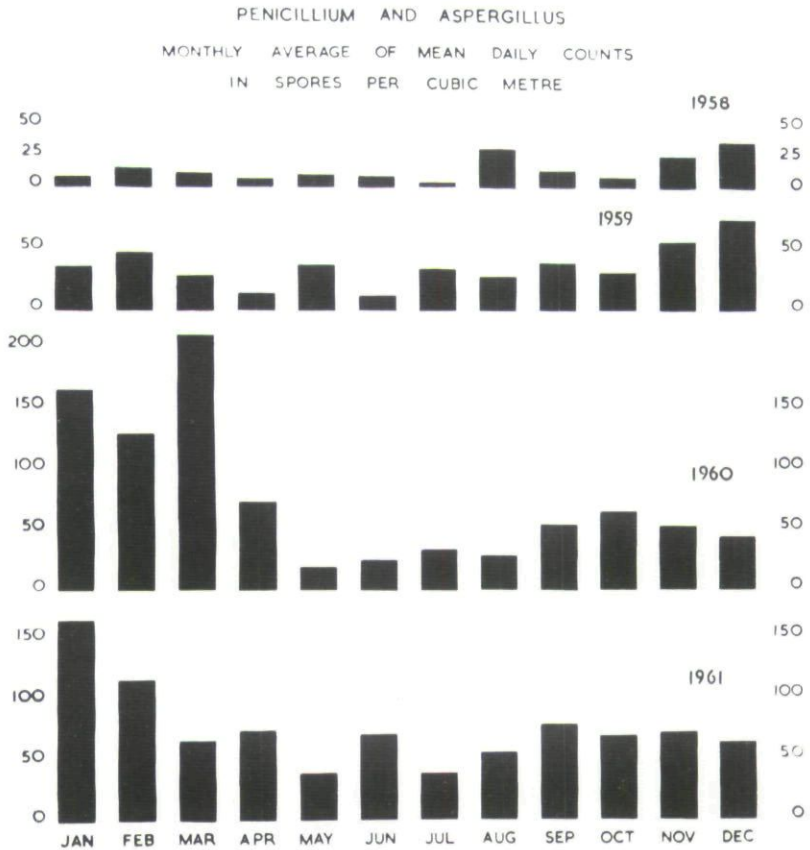


Fig. 16.

Variation in the seasonal incidence of *Penicillium* and *Aspergillus* combined, 1958-61. Maximum concentrations were usually recorded during the winter months and the highest daily mean count was 2000 Spores/m³ in January, 1961.

variety of substrates and cause the well-known blue-green mould found on foodstuffs, damp leather and textiles. It was impossible to distinguish between the spores of *Penicillium* and *Aspergillus* respectively and they have therefore been placed together under one heading. The group may have included a few other fungi whose spores also grow in chains, are spherical and less than 5 μ in diameter. Fig. 16 shows that spores were caught in all months but that, over the four years,

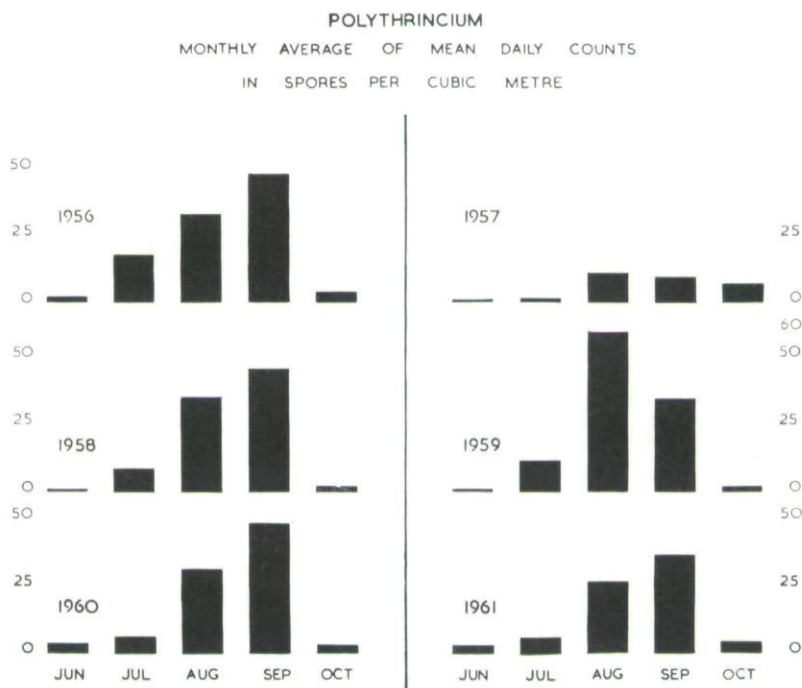


Fig. 17.

Variation in the incidence of *Polythrincium*, June to October, 1956-61. This fungus was never recorded from November to mid-May. The highest daily mean count was 315 spores/m³ in September, 1960. (See text).

73 % of them were recorded between October and April. Richards (1954) who worked with petri plates in Cardiff during 1951-53, reported that *Penicillium* varied little throughout the year but that *Aspergillus* had a marked winter maximum. The highest concentrations were on dry days, when the maximum temperature was 45°-60°F and the minimum 34°-50°F. The catch from January to April, 1958 was well below average. January and February were wet months and there were 31 days from January to March when the minimum temperature was below freezing point.

The daily mean concentration exceeded 200 spores/m³ on many days during the winter months of 1959-61 but only

intermittently. Few spores were caught during the night and the highest counts were recorded between 1200 and 2200 hours: the maximum was 5000 spores/m³ at noon.

f) *Polythrincium trifolii* is the conidial stage of "Black blotch" disease of clover and its incidence, therefore, is likely to be related to the conditions affecting its host. The light brown, two-celled, pear-shaped spores were caught from the end of May to October but 83 per cent of the six years' catch was recorded in August and September (Fig. 17). The daily mean concentration exceeded 200 spores/m³ on three days only during the six years. The highest catches were recorded on dry days; no other meteorological factor seemed to affect the number of spores caught

It is difficult to account for the very low catches of 1957; weather conditions were as favourable then as in other years. One possible explanation which, unfortunately, cannot be confirmed is that the clover on the vacant land near the trap, which would be one of the chief sources of the spores, was mown early that year.

Maximum concentrations were recorded between 0900 and 1500 hours; very few spores were observed during the evening and night. The highest count recorded was nearly 700 spores/m³ at 1500 hours.

g) *Pullularia* is a sooty mould that forms a black coating on the surfaces of leaves that have often been covered with the honeydew of aphides. Smit & Wieringa (1953) found that *Pullularia* occurred on freshly opened tree buds and that it was almost exclusively the only mould isolated from yellow leaves that had fallen from many trees.

The dark brown, thick walled, old mycelial cells were found in clumps on the slides in all months but the bulk of them were recorded from July to October (Fig. 18). Concentrations were usually highest when there was little or no rain; otherwise little correlation between the catch and the weather was observed. Daily mean concentrations of over 200 cells/m³ were recorded intermittently between June and November and the bulk of the catch arrived in the afternoons.

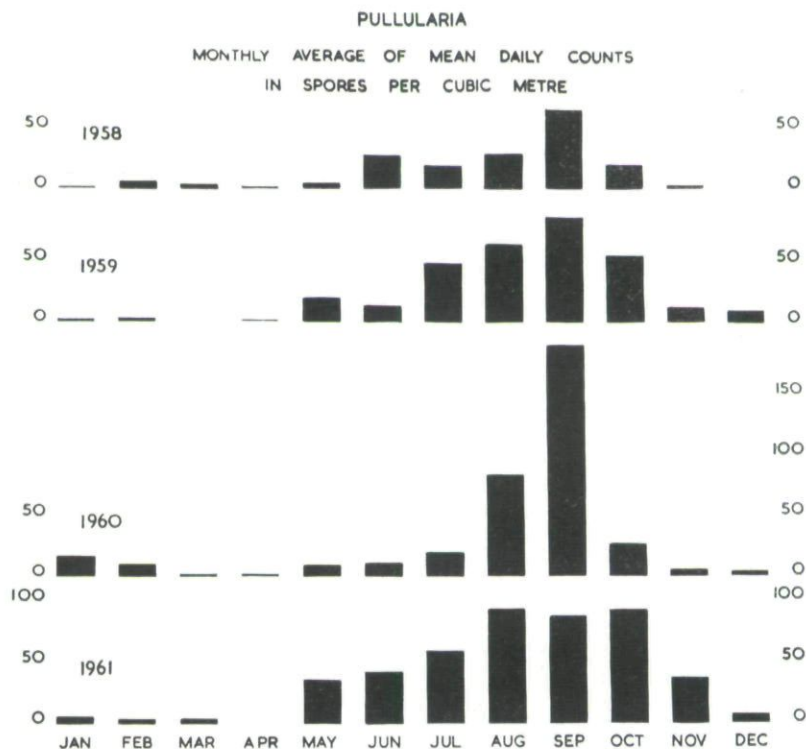


Fig. 18.

Seasonal variation in Pullularia, 1958-61. The highest daily mean count was 1500 cells/m³ in October, 1961. (See text).

h) *Torula* is found on decaying wood and soil and produces chains of brown, warty spores which, when mature, break into short lengths. 75 % of these chains, of usually four or five spores, were observed on the slides from May to September and, except in 1958, there was an early summer and September maxima. (Fig. 19). *Hamilton* (1959) recorded this double maxima in Harpenden and London. The maximum concentrations (never greater than 180 spores/m³) were recorded on completely rainless days with ten or more hours of bright sunshine. Rising temperatures and decreasing humidity increased the counts. Catches were 155 % of average in the

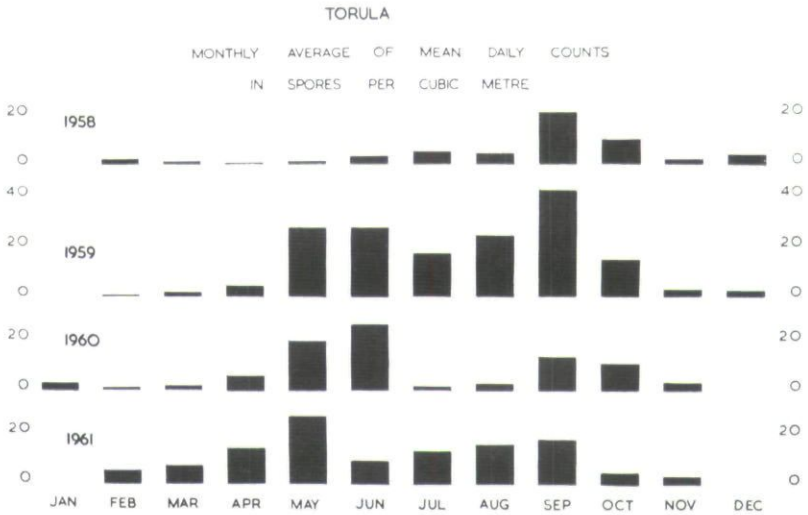


Fig. 10.

Seasonal variation in *Torula*, 1958-61. There was a tendency towards early summer (May, June) and September maxima. The highest daily mean count was 180 spores/m³ in August, 1959. (See text).

hot, dry summer of 1959 but 51 % in 1958 which was generally sunless and wet.

The bulk of the spores arrived between 1100 and 1900 hours and they were completely absent at night; the highest catch recorded was 470 spores/m³ at noon.

i) *Minor types*. Eighteen additional genera were observed on the slides in small numbers and formed only 0.8 % of all spores of the Fungi Imperfecti; together their average daily mean concentration over the 4 years was 11 spores/m³. They were *Hormiscium* (spores like *Torula* but caught in much longer chains), *Helicosporium* type, *Scopulariopsis*, *Periconia*, *Spicaria*, *Macrosporium*, *Geotrichum*, *Bispora*, *Helminthosporium*, *Trichothecium*, *Stemphylium*, *Cercospora*, *Piricularia*, *Acrothecium*, *Tetraploa*, *Echinobotryum*, *Triposporium*, and *Asterosporium*.

5. UNDETERMINED AND DAMAGED

Spores included in this group were:

- (a) Damaged or partially obscured by debris;
- (b) Not identified with reasonable certainty;
- (c) Unknown.

DISCUSSION

The trap used in this continuous survey was efficient (Hirst 1953) and enabled one to estimate the numbers of spores per cubic metre of air for any desired period. Microscopic examination of a small proportion of the deposited trace gave a reliable estimate of those spores which were recorded in large numbers and these types are of particular interest to the clinicians; estimates given here of the spore types which occur in the air in small numbers or large clumps are not so reliable. It was difficult to classify some spores visually and only a few of the obvious Ascospores and Basidiospores could be named with any certainty; these groups await further study. In a culture plate survey, the spores of these two main classes (with the exception of a few Ascomycetes), which made up 55 % of all spores caught in this survey, would not have been recorded.

There were very wide differences in average frequency between the various spore types; the daily mean concentration over four years varied from 1,204 spores/m³ for *Cladosporium* to 0.007 spores/m³ for *Asterosporium* and the highest daily mean count recorded varied from 32,000 spores/m³ for *Cladosporium* to 9 spores/m³ for some Ascomycetes and many of the minor Fungi Imperfecti.

All spore types varied in quantity from year to year; some more than others. Thus *Epicoccum* varied from 237 to 45 % of average while *Sporobolomyces* only varied from 110 to 86 %; all other spore types fall between these extremes. This variation appeared to depend on the weather and the presence or absence of diseases on crops in the neighbourhood. As nearly all the spores caught (with the exception of the Agari-

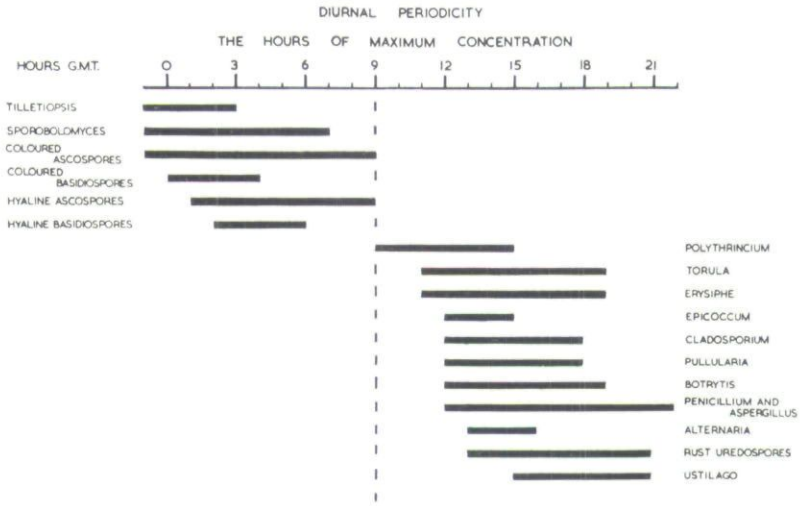


Fig. 20.

Diurnal periodicity. The highest counts of the various spore types were recorded between the hours shown. (See text).

cales) are derived from fungi growing on living or dead vegetation, the condition of the vegetable substrates in the district must play an important part in determining the quantity of spores produced and liberated.

All spore types (with the exception of the *Penicillium* and *Aspergillus* group) showed a marked seasonal incidence; the highest counts being recorded in the summer and/or autumn months (Figs. 2-19). Within this season the timing of the peak counts varied with the weather and there was a difference of up to seven weeks between the peaks of one year and the next, e.g. *Cladosporium*, 1955 and 1957 (Fig. 13); the *Penicillium* and *Aspergillus* group showed a poorly defined winter maximum (Fig. 16).

Most spores showed a clear diurnal periodicity in addition to a marked seasonal incidence; some reached their maximum during the day (usually after noon) while others were highest at night (Fig. 20). The six spore types with a night and early morning maximum recorded their highest daily mean concen-

SPORES PER CUBIC METRE OF AIR

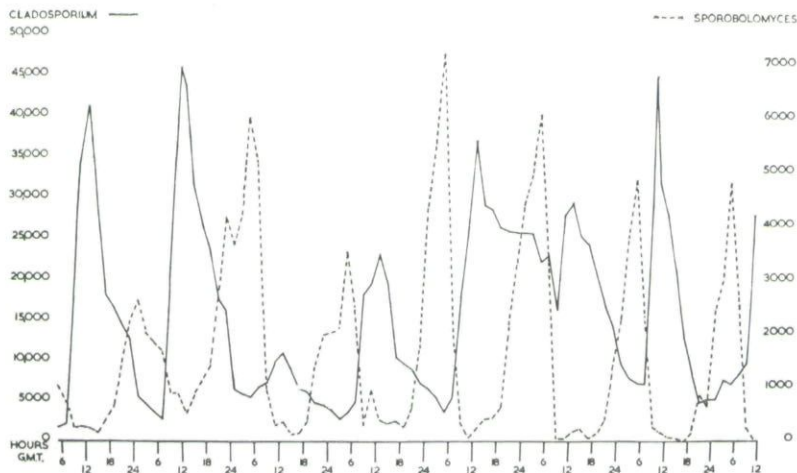


Fig. 21.

Two hourly count of *Cladosporium* (1-8 July, 1957) and *Sporobolomyces* (27 July-3 August, 1959). These two weeks were chosen for investigation because the daily mean counts were high. Similar results were obtained for *Cladosporium* 23-29 July, 1956. Maximum contractions then were from 1200 to 1500 hours and minimum concentrations, 0100 to 0700 hours.

trations when the weather was wet, warm and the humidity was high. With the exception of *Botrytis*, the spore types which showed an afternoon and early evening maximum liberated more spores when it was dry and the humidity was decreasing. *Botrytis* was at its highest in warm, damp, humid weather but Paul (1929) found that its spores were formed more freely in the light than in the dark and this, no doubt, accounts for its diurnal periodicity. Fig. 21 shows 2-hourly counts of *Cladosporium* (1-8 July 1957) and *Sporobolomyces* (27 July-3 August, 1959) and demonstrates that this diurnal periodicity can, sometimes, be most marked and regular.

The results of this survey clearly indicate that continual sampling, with a volumetric trap, is necessary to give a complete picture of the incidence of fungus spores. This enables

one not only to obtain a volumetric count of the daily concentration, but to show the time and magnitude of the maximum concentrations of spores to which patients are exposed for short periods.

SUMMARY

1. A Hirst Automatic Volumetric Spore Trap, situated 60 feet above ground level, was used to make a continuous survey of fungus spores over Cardiff.
2. Seven spore types made up 93 % of the total catch. They were *Cladosporium*, 39.1 %; Hyaline Basidiospores, 17.5 %; Hyaline Ascospores, 15.2 %; *Sporobolomyces* 8.8 %; Coloured Basidiospores, 7.3 %; Coloured Ascospores, 2.5 % and *Ustilago*, 2.7 %.
3. Each spore type had a marked seasonal incidence. *Cladosporium*, June to September; Basidiospores, July to November; Ascospores, June to October; *Sporobolomyces*, June to October; and *Ustilago*, June and the first half of July.
4. Some spore types, e.g. *Cladosporium*, were present all through the year, while others e.g. *Ustilago*, were recorded only in the summer months.
5. Each spore type varied in quantity from year to year. This variation depended on the weather, the presence of diseases on crops and the general condition of the vegetation in the neighbourhood.
6. Most spore types displayed a diurnal periodicity. Generally those recorded most frequently in damp, humid weather had their maximum concentrations at night, while those whose highest counts were recorded when it was dry and the humidity was decreasing had an afternoon maximum.

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