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A COMPARISON BETWEEN THE SUMMER AND AUTUMN AIR-SPORAS AT LONDON AND LIVERPOOL

By

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In 1961, from June to October inclusive, automatic volumetric spore traps (*Hirst* 1952) were continuously operated at London and Liverpool to investigate and compare the changing concentrations of the spore content of the atmosphere in these densely populated areas.

The London trap was placed on the 25 metre high flat roof of the nurses' home at St. Mary's Hospital, Paddington (altitude 29 metres) where catches would typify a wide area and be unbiased by any pollen or spore sources in the immediate locality. Although the hospital lies in a densely built-up part of Central London there is a great deal of vegetation in the neighbourhood. Within a kilometre radius of the trapping site are more than 500 plane trees (*Platanus acerifolia*) many of which line grassed areas, and a kilometre to the South lie Kensington Gardens and Hyde Park where there are over 7,000 trees and 2.8 sq. kilometres of park land which is mainly grass clad. Less than 2 kilometres to the North West, in the area of Regent's Park, there are over 5,000 trees and approximately 2.4 sq. kilometres of grassland.

In Liverpool, the trap was sited on the 12 metre high roof of the Liverpool Chest Hospital, which lies on the western slope of Mount Pleasant Hill at an approximate altitude of 42 metres. The neighbourhood is more densely built-up than that adjacent to the London site; there are only 45 trees (33

Platanus) within a kilometre of the hospital and the nearest sizeable park (approximately 72 hectares) lies about 2 kilometres away.

Both traps were continuously operated at a suction rate of 10 litres per minute seven days a week throughout the period of the investigation and were stopped each day only for the few minutes necessary when slides were changed. The slides were coated with petroleum jelly and after exposure mounted in glycerine jelly as used by *Hamilton* (1959). The deposits were scanned under an oil immersion objective and the daily mean concentrations of pollen grains and fungal spores estimated from counts along a single narrow traverse parallel to the direction in which the slide moved while in the trap.

This method of scanning samples the whole of the exposure period and is most convenient for comparing the spore content of the air in two different areas. The counts were not corrected for anisokinetic errors in sampling and the efficiency of the method, which averages about 80% and thus underestimates the true concentration, is described by *Gregory & Hirst* (1957).

Classification of the pollen grains and spores caught was based on microscopic morphology and the pollen grains were examined unstained since the natural colour of the fungal spores must be retained for their identification. Fungal spores were assigned to a series of categories or "Form Groups", mainly those used by *Gregory & Hirst* (1957) and *Hamilton* (1957 and 1959) and whose relationship to well established taxonomic genera will be described below.

POLLEN

The amount of pollen in the air is primarily dependent upon the seasonal cycle of plant flowering. This is governed by climate and, in England, according to *Blackley* (1873) and *Hamilton* (1959), the grass pollen season usually extends from the beginning of June to the end of the third week in

July. The actual amount of pollen in the air, however, is affected by short term weather variations and rain for instance, has been shown by *Blackley* (1873), *Hyde* (1950) and *Hamilton* (1957) to remove pollen from the atmosphere.

The mean daily estimates of grass pollen in the air at London and Liverpool are given in Fig. 1 which illustrates the enormity of the daily variation. To smooth out the curves and make it easier to study the effects of long spells of weather and phenology, and also render the data from the two cities more easily comparable, the results for the common pollen grains are presented as ten day running means, Fig. 2.

It may be seen (Fig. 2) that for Liverpool in 1961 the curve for grass pollen ascends 9 days later than that for London, the season was 9 days shorter than London's and in June and July the total Liverpool catch equalled only 66% of the number of grains trapped in London, yet on 24th and 30th June, however, the mean daily estimates for Liverpool were 440 grains per cubic metre, whilst the highest concentration recorded in London, on one day only, was 370 grains per cubic metre on June 15th.

Although there are no comparable data for previous seasons at Liverpool, *Hamilton* (1959) reports that in London in 1954 and 1955 the highest grass pollen counts were obtained on June 22nd in both years; in view of the fact that in 1961

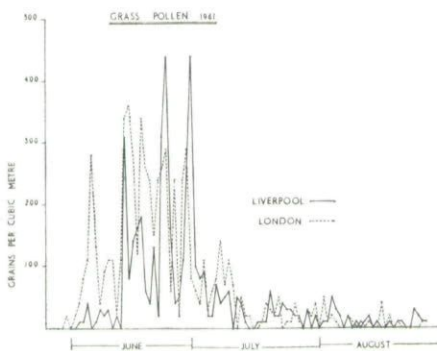


Fig. 1.

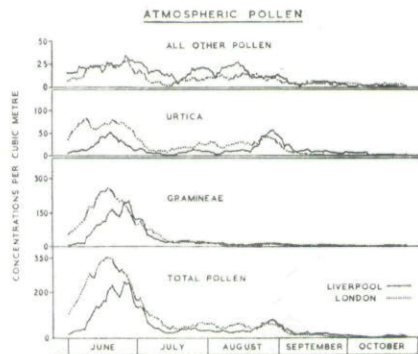


Fig. 2.

the highest concentration occurred on June 15th, it must be concluded that the date of maximum grass pollen production varies from year to year.

Urtica (nettle) not only occurs in the atmosphere throughout the summer, but appears to have two cycles of flowering, the numbers of airborne grains rising to a first peak in June and then to a second peak in late August in both cities (see Fig. 2). In June the maximum concentrations were 20 grains per cubic metre on the 6th in London and 10 grains per cubic metre on the 20th in Liverpool. During the August peak, however, the maximum concentration in Liverpool was greater than that in London and these were 9 and 17 grains per cubic metre on the 29th and 28th respectively. During the period June to October inclusive, although the total Liverpool catch was only about half that of London, in September it was four times London's.

Most of the anemophilous tree pollens were shed by June 1st, so that the grains included in "all other pollens" (Fig. 2) are predominantly those of *Artemisia*, other *Compositae*, *Chenopodium*, *Rumex* and miscellaneous pollen.

The extent to which the total pollen content of the atmosphere in the Summer in England is predominated by the *Gramineae* and *Urtica* is clearly shown in Fig. 2. In June and July, in London, grass pollen accounted for 62% and *Urtica* 26% of the total catch, whilst at Liverpool, the proportions were 70% and 16% respectively.

FUNGAL SPORES

Over forty different categories of fungal spores may be recognized in the deposits on slides exposed in a Hirst trap but only those which were very common or consistently present in the atmosphere of both cities will be considered here.

Fungi Imperfecti:

Cladosporium.—comprises a spore category of a distinctive and easily recognisable morphology in which the conidia, seen

on the slides singly, or in groups or digitate clusters, are undoubtedly those of the genus *Cladosporium* Link ex Fries. Although many of the spores cannot be ascribed to a species, those of *C. macrocarpum* Preuss and *C. herbarum* Link ex Fries are recognisable and the latter species predominates.

In London the season commenced earlier than it did at Liverpool and rapidly rose to a daily mean peak concentration (16,800 spores per cubic metre) on July 20th. In Liverpool the seasonal rise in June and July was much smaller than in London (Fig. 3), and the maximum mean daily concentration (14,300 spores per cubic metre) was not attained until September 2nd. In June and July when *Cladosporium* comprised 70% of the entire spore catch in London it was only 35% of that in Liverpool. From late August onwards, however, the *Cladosporium* concentration at Liverpool rose to almost equal that of London.

"*Penicillium*".—The spore group "*Penicillium*" includes the conidia of the whole of the *Aspergillaceae*; the spores are small, round or ovoid phialospores which when trapped from the air may appear on the slides either singly or in short chains. The group is designated "*Penicillium*" because cultural studies have shown the spores of this genus outnumber by far those of *Aspergillus* in the air of Britain. Three times as many "*Penicillium*" spores were trapped in Liverpool as were caught in London (Fig. 4).

Botrytis Pers. ex Fries.—(Fig. 5) produces characteristic

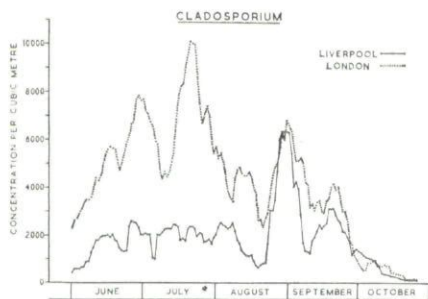


Fig. 3.

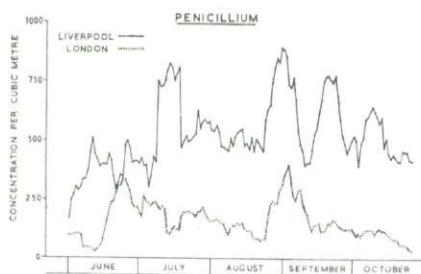


Fig. 4.

ovate almost hyaline one-celled conidia which when trapped from the air occasionally occur in clumps of four or five. The fungus occurs as a common saprophyte and is also pathogenic to a wide variety of plants. It may be seen (Fig. 5) that the greatest numbers were trapped from the air in July and late August and September, and there tends to be an autumnal peak particularly at Liverpool. From June to October inclusive the London trap caught only 72% of the total number of *Botrytis* conidia trapped in Liverpool.

Ascomycetes:

The genera *Chaetomium*, *Leptosphaeria*, *Pleospora*, *Sordaria* and *Venturia* are but a few whose characteristic ascospores may be recognised in a slide deposit; others such as *Epichloe*, *Claviceps* and *Ophiobolus* produce long filiform ascospores. Hamilton (1959) classified airborne ascospores into *Filiform*, *Fusiform*, *Leptosphaeria* and a *Miscellaneous group*, but restricted *Leptosphaeria* to include only multiseptate fusiform yellow spores with one markedly swollen cell, although in many species of *Leptosphaeria* Ces de Not. the ascospores have no swollen cell (Holm 1952). In the present study the ascospore classification of Hamilton (1959) is adopted with the exception that the spore group designated *Leptosphaeria* has been broadened to include hyaline, yellow to brown, cylindrical or fusiform ascospores with transverse

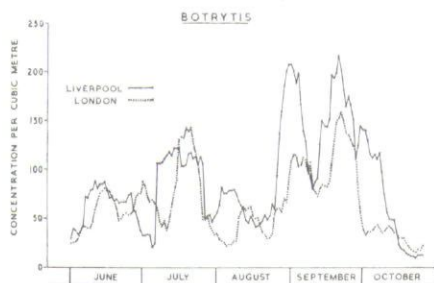


Fig. 5.

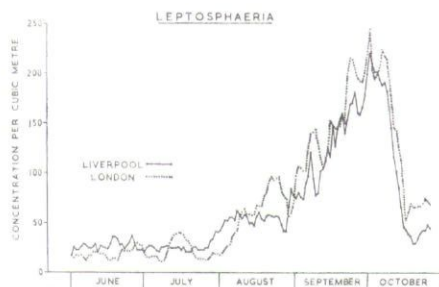


Fig. 6.

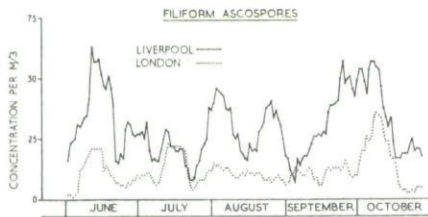


Fig. 7.

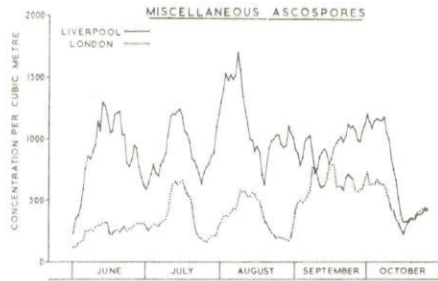


Fig. 8.

septae only. The *Fusiform* group is retained for fusiform thin walled ascospores with thickened transverse septae only, but since both these and *Venturia* ascospores were not consistently caught during the period of this survey they are included in the *Miscellaneous* group illustrated in Fig. 8.

Leptosphaeria.—These “leaf-spot-fungi” most commonly occur on plant leaves and stems, and Last (1955 b) has described ascospores of the *L. eustoma* group in concentrations of 65,000 per cubic metre in the air of a barley crop.

The atmospheric concentrations of “*Leptosphaeria*” ascospores increased during August to reach a peak in late September at both stations (Fig. 6).

Filiform Ascospores.—These comprise a group described by Hamilton as being produced mainly from lesions on plant leaves and having a ratio of length to breadth greater than 20:1. They may be septate or aseptate and include the ascospores of such genera as *Claviceps*, *Epichloe* and *Ophiobolus*.

The aerial concentrations of filiform ascospores at Liverpool were higher than those at London during June to October 1961 (Fig. 7). In view of the density of urban development at Liverpool, it is most likely that these spores originate from the open countryside away from the cities.

Miscellaneous Ascospores.—This group includes such morphologically recognisable ascospores as those of *Chaetomium*, *Sordaria* and *Venturia* together with others which have not yet been classified. The general trend in London concentrations is

similar to that at Liverpool (Fig. 8) and the Liverpool catch was higher throughout the period studied.

Basidiomycetes:

The basidiospores from toadstools, bracket fungi etc. are usually asymmetrical and characterized by a distinct apiculus. A few are sufficiently distinctive to be recognizable and assigned to genera and even species in the *Agaricaceae*. The spore form genera of *Ganoderma*, *Tilletiopsis* and *Sporobolomyces* will each be considered separately since they were regularly encountered at both stations; all other basidiospores have been simply classified according to their colour into two groups—hyaline or coloured.

Coloured basidiospores.—(Fig. 9) were trapped in rather higher concentrations at Liverpool than London, the total London catch forming 93% of that obtained at Liverpool (Fig. 9).

Hyaline basidiospores (Fig. 10) were trapped in far greater numbers in Liverpool than London, the total London catch being only 51% of that at Liverpool.

Both the hyaline and coloured basidiospores showed the same seasonal rise to a peak concentration in late September and early October.

Ganoderma.—A bracket fungus which grows on the trunks of trees. The spores of *G. applanatum* are brown with an outer hyaline wall pierced by projections from an inner coloured

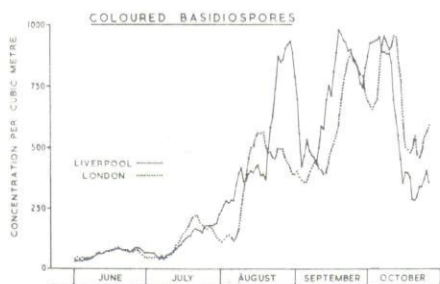


Fig. 9.



Fig. 10.

wall, and consequently are easily identified in the deposits on Hirst trap slides.

The concentration of *Ganoderma* spores in the atmosphere of London was consistently higher than at Liverpool (Fig. 11). This is clearly correlated with the numbers of trees in the areas; there are more than 13,000 trees within a 2 kilometre radius of the London trap and less than 1,300 within a similar radius of the one at Liverpool.

Sporobolomyces.—"pink mirror yeast" produces characteristic hyaline basidiospores. This fungus, together with *Bullera*, "white mirror yeast", and *Tilletiopsis* occurs abundantly on the leaves of cereals as wheat and barley (Last 1955 a, b). The numbers of *Sporobolomyces* trapped are shown in Fig. 12 and probably include some *Bullera*, a less common fungus but which has spores indistinguishable from *Sporobolomyces*. Since the concentration can rise extremely rapidly, during rain as on July 12th at Liverpool, when from a previous daily mean concentration of 6,500 per/M³ the count reached 26,440 per/M³, the mean daily concentrations have been given for this fungus. The London total was 41 % of that recorded in Liverpool.

Tilletiopsis—produces minute sickle-shaped basidiospores which when trapped by impaction from the air are usually folded hinge-like into two unequal parts. Hamilton (1959) reports the peak occurrence of *Tilletiopsis* spores occurred in September and October, and that their numbers in air were not

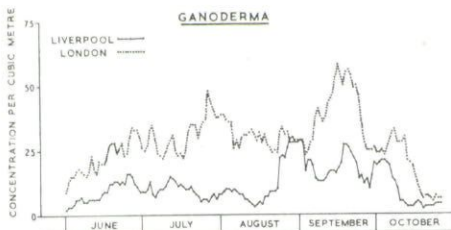


Fig. 11.

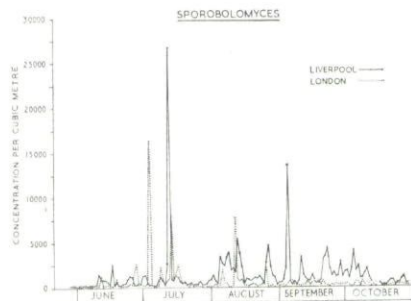


Fig. 12.

significantly increased when rain was falling. In this study, the concentrations of *Tilletiopsis* spores in the air of London were far less than at Liverpool where they rose to a distinct peak in late September and early October.

Total—All Spores.—Before Mid-August the total fungal spore concentrations at London were higher than those at Liverpool, but from then on the Liverpool concentrations were the higher. (Fig. 14). The contribution of each spore category to the total air-spores together with the dates of highest daily concentrations during the period studied is given in Table 1.

DISCUSSION AND CONCLUSIONS

The constituents of the air-spores may be classified into groups according to their season, in Britain, *Cladosporium*, *Ustilago* and *Sporobolomyces* have summer maxima, the spores of *Botrytis*, such ascomycetes as *Leptosphaeria* and certain basidiomycetes occur in highest concentration in the autumn.

The far higher total spore count at London during the summer was mainly due to a preponderance of *Cladosporium* (Figs. 3 and 14). In the public parks in the proximity of the London trap there is approximately five square kilometres of grass (mainly *Poa*, *Holcus* and *Festuca* spp.) which is mown weekly and the cuttings left lying on the sward. Since *Cladosporium* predominates the primary flora which colonizes moribund grass tissue (*Webster* 1956, *Hudson & Webster* 1958)

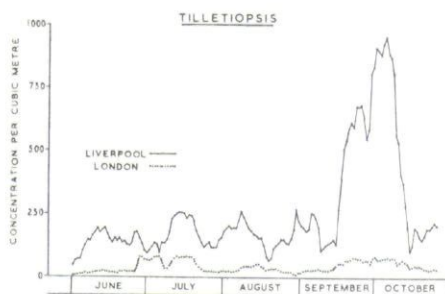


Fig. 13.

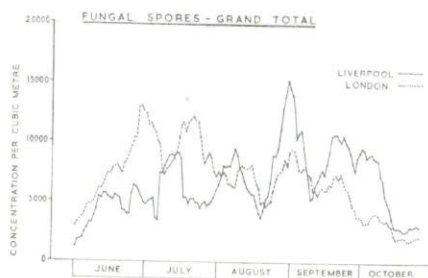


Fig. 14.

this area of substrate, presented weekly, must be important in contributing to the *Cladosporium* concentration in the London atmosphere during the summer months. Sampling in the park was therefore initiated in 1962 with rotorod samplers (*Perkins*

TABLE 1
Contributions of Spore Categories to the Total Air Spora.

London			Liverpool			
Highest daily concentration		% of total air spora	Spore category	% of total air spora	Highest daily concentration	
Date	Number M ³ of air				Number M ³ of air	Date
<i>Ascospores</i>						
27.IX 11.VII & 16.VIII 13.IX	580 90 1,940	11.04 0.18 5.79	Leptosphaeria Filiform Miscellaneous	1.0 0.48 13.52	360 230 2,590	21.IX 16.VI 18.VII
<i>Basidiospores</i>						
10.X 20.IX 1.VIII 3.VII 3.VII	1,890 1,100 110 16,310 360	5.30 3.79 0.42 7.87 0.61	Coloured Hyaline Ganoderma Sporobolomyces Tilletiopsis	5.90 7.67 0.17 20.11 3.81	1,520 2,540 120 26,440 2,580	28.VIII 4.X 29.VIII 12.VII 4.X
29.VIII 21.VII 20.VII 10.IX 31.V 8.V 1.VIII 7.X 24.IX 30.VIII 22.VI 5.IX 30.VIII	220 420 16,860 150 140 1,060 100 270 270 60 2,800 490 810	0.41 0.96 61.81 0.26 0.43 2.35 0.10 0.31 0.31 0.12 3.46 2.30 2.27	Alternaria Botrytis Cladosporium Epicoccum Erysiphe Penicillium Polythrincium Pullularia Torula Uredospores Ustilago Other Spores Mycelium	0.19 0.35 28.40 0.14 0.25 7.98 0.20 0.18 0.17 0.17 2.18 4.40 1.75	240 830 14,300 90 270 1,590 150 110 240 80 2,800 1,630 1,250	29.VIII 12.VII 2.IX 1 & 9.IX 25.VII 29.VIII 29.VIII 31.VIII 29.VIII 29.VIII 30.VI 22.IX 1.IX
3.VII	34,040		Total Spores		41,880	12.VII

1957)¹ and the concentrations there compared with those recorded by the Hirst trap on the 25 metre high roof a kilometre away. The results given in Table II are typical of those obtained when samples were taken between 1300–1400 hours.

TABLE 2
Variation in Cladosporium concentrations at different sites. Samples were taken simultaneously and counts are given per cubic metre.

<i>June</i>				
Day	25	26	27	29
Kensingtons Gardens	92,400	123,180	49,200	1,900
Roof (25 m. high)	25,530	1,116	1,600	700
Streets	372	900	120	180
<i>July</i>				
Day	2	6	8	9
Kensingtons Gardens	16,590	17,250	17,500	44,544
Roof (25 m. high)	4,700	2,500	5,200	533
Streets	3,500	136	—	155

Since the park concentrations are so much higher than at the other sites it must be concluded that in the summer months, the grassland of the public parks is an important source of atmospheric *Cladosporium* in central London. By contrast, in Liverpool where there is no comparable area of grassland near the trapping site, airborne *Cladosporium* concentrations in June and July 1961 were only half that in London. From late August onwards, the *Cladosporium* concentrations at Liverpool rose and approached that of London. This may be explained by the commencement of the autumnal seasonal decay of vegetative matter throughout the British countryside, and by the fact that as the summer temperatures decline, the rate

¹ Trapping efficiency of the rotorod varies with particle size, for *Cladosporium* it was taken as 50 % since under still air conditions in the laboratory its efficiency varied between 40–50 % when compared with a slit-sampler (Bourdillon *et al.* 1941).

at which grass grows and the frequency of mowing and provision of fresh substrate for colonization in the park diminishes.

Peak concentrations for *Cladosporium* in London, but not Liverpool, may be correlated with the highest summer temperatures. *Bockmann* (1933) recorded temperatures between 21–26° C as optimal for the development of *Cladosporium* in culture and *Richards* (1956) found a correlation between temperature and the number of conidia in the air. Since highest *Cladosporium* counts in Liverpool were not obtained until the autumn however, the availability of substrate for colonization appears to have a greater effect on the *Cladosporium* content of the air than does temperature.

The conclusion of *Richards* (1956) based on culture plate exposures, that the number of *Cladosporium* spores in the air are inversely related to the degree of urban development of the trapping site environs, is here substantiated, despite the fact that *Gregory & Stedman* (1953) have shown the trapping efficiency of the exposed Petri dish to be no higher than 5% and that of the Hirst trap is approximately 80%.

Three times as many "*Penicillium*" spores were trapped in Liverpool as were caught in London (Fig. 4).

Ambler & Vernon (1951) and *Richards* (1956), in New Zealand and Britain respectively, found *Penicillium* to be more frequently deposited from the air of large towns than in less built up places.

It is noteworthy that *Brown* (1936) described moulds as the weeds of the home, and *Davies* (1960) not only found *Penicillium* to be the mould genus most frequently isolated from house dust, but present in larger numbers in dust from densely built up areas than that from less urban districts. Given adequate conditions of humidity, both *Penicillium* and *Aspergillus* readily colonize house dust, whilst *Cladosporium* is not only unable to colonize house dust, but rapidly loses viability in it and can be rarely isolated (*Davies* 1960). In view of these findings and the fact that the present study shows the "*Penicillium*" spore type to be far more abundant in the more dense-

ly built up area, it must be concluded that "*Penicillium*" is characteristic of urban areas in contrast to *Cladosporium* which is more common in the air of the open country.

The spores of *Botrytis*, *Leptosphaeria*, *Tilletiopsis* and the hyaline and coloured basidiospores were trapped in their highest concentrations in the autumn. Among the ascospores those of *Leptosphaeria* were trapped in similar numbers at both cities, the other groups were more common in the air at Liverpool; apart from *Ganoderma* more basidiospores were trapped in Liverpool than London. The number of *Ganoderma* spores trapped in London must be correlated with the fact that there are over 13,000 large trees within 2 kilometres of the trapping site and less than 1,300 in the comparable area of Liverpool. Most of the fungi in these groups are uncommonly found growing in city areas; *Leptosphaeria*, *Sporobolomyces* and *Tilletiopsis* occur abundantly on cereals (Last 1955 a, b), filiform ascospores are produced by pathogens of the *Gramineae* such as *Claviceps*, *Epichloe* and *Ophiobolus*, the ascospores of the miscellaneous group are mainly from saprophytic or coprophilous fungi and the association of the higher basidiomycetes with forest trees is well known. Clearly the origin of such spores must mainly lie in the countryside adjacent to the cities. The fact that the trapping sites at London and Liverpool respectively, were approximately 15 and 5 kilometres away from open country with mixed vegetation and land under crops, most probably accounts for the higher ascospore and basidiospore catches at Liverpool. In this survey, the ascospores and basidiospores together comprised 54% of the total spore catch at Liverpool and 29% at London, and yet, as allergens their role has been scarcely investigated. *Feinberg* (1945) and *Frankland* (personal communication) have reported that positive skin reactions may be obtained with some of the fungi in these groups and they are worthy of further investigation. The lack of knowledge of these fungi lies in part with the fact that most air-spore surveys have been made with culture plate exposures and as noted by *Hamilton* (1959) only the spores of the *Fungi Imperfecti* and *Mucorineae* were recorded. Under

conditions of artificial culture 32% of the spores trapped at Liverpool and 21% at London, would fail to produce an identifiable colony, even if no competition was presented by the other fungal constituents of the air-spora.

The effect of buildings on wind flow and the pollen and spore concentrations in city streets does not appear to have been investigated although preliminary studies (see Table II) suggest street levels more akin to indoor concentrations than those of wide open spaces.

The sampling of atmospheric allergens is beset with numerous problems, concentrations not only vary from hour to hour but with the patient's immediate environment and the weather. At present, the roof top continuous sampler which provides a visual record of the entire air-spora seems the most practical approach to the city wide survey; since concentrations will be higher in the adjacent countryside and lower in the central streets, the record should be supplemented with "spot" samples taken in every conceivable situation. Detailed studies on the preponderance of *Penicillium* and *Aspergillus* spp. in urban areas must employ volumetric cultural methods such as those described by Bourdillion *et al.* (1941) and Andersen (1958). The ultimate aim of all such sampling must be to record the range of pollen and spore concentrations to which the patient is exposed, it is for the clinicians to decide their allergic importance.

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SUMMARY

The summer and autumn air-sporas at London and Liverpool were compared by continuous automatic sampling from May 28th to October 31st 1961. The grass pollen season at London rose to a peak 9 days earlier than at Liverpool where the season was shorter. Higher spore concentrations at London

during the summer were due to a preponderance of *Cladosporium* and this was correlated with the amount of grass park land in Central London. Ascospores and basidiospores were trapped in higher concentrations at Liverpool and related to the relative distances of the trapping sites from open countryside. Preliminary studies of spore concentrations in open spaces, at roof top and ground level in the streets, show street concentrations to be low and akin to those indoors.

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