

Airborne Pollen and Spore Survey in Relation to Allergy and Plant Pathogens in Nsukka, Nigeria

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Abstract

Airborne bio-particles of allergic significance were recorded at a height of 15m in Nsukka during September 1999 – February 2000. Spores of fungi and pollen grains, which are important part of the exposure that may lead to allergic discomfort and plant diseases, dominated the particles. Other primary sources of the allergens counted include spores of ferns, diatom frustules, algal fragments, insect parts and charred gramineae cuticles. Thirty plant families consisting of 33 pollen types were identified to 10 generic and 23 specific levels. The common pollen grains counted include those of Poaceae, *Elaeis guineensis*, *Casuarina equisetifolia*, Asteraceae, Combretaceae / Melastomataceae, *Alchornea cordifolia* and Amaranthaceae/ Chenopodiaceae. The pollen grains of anemophilous plants such as those of Poaceae, *Elaeis guineensis*, Asteraceae and *Casuarina equisetifolia* that are regarded among the important causes of allergic rhinitis were commonly counted in the atmosphere. Thirteen spore types of fungi were identified and they belong to three classes: Deuteromycetes, Ascomycetes and Basidiomycetes. The spores of species of *Nigrospora*, *Ustilago*, *Cladosporium*, *Drechslera*/ *Helminthosporium*, *Fusarium*, *Alternaria*, *Puccinia* and *Curvularia* were among the important allergic and pathogenic fungal spore types counted in the study.

Keywords: Airborne Pollen, Spore, Pollen Allergy, Plant Pathogens, Nigeria

Introduction

Atmospheric surveys conducted in Nigeria by Agwu and Osibe (1992), Agwu (1997, 2001); in West Africa by Melia (1984), Dupont and Agwu (1992) and in other parts of the world, Spieksma *et. al.*, (1985), Frankland (1987), Singh (1987), Chakraborty *et. al.*, (1999), Burge (2002) and Jato *et. al.*, (2002) indicate that the atmosphere contains numerous organic and inorganic particles such as pollen grains, spores of fungi and ferns, diatom frustules, plant fragments, insect parts, dust, oil droplets, siliceous materials, salt crystals and other inhalable materials most of which are important causes of pollinosis and/ or plant diseases. Many factors influence the composition and abundance of these airborne particles. These include geography, vegetation, meteorology, human activity and plant life cycles (Jato *et. al.*, 2002).

Each rainy season and dry/harmattan season, some of these minute and offensive particles (pollen and spores) are released and dispersed from trees, shrubs, weeds, grasses and fruiting bodies of ferns and fungi

into the atmosphere. Some other particles are released from dead organic matter, industrial activities, dust storm and other anthropogenic activities. The air quality in some circumstances depends on the types and concentration of these particles in the atmosphere that have impact on public health, crop survival and yield.

Primarily, the role of pollen grains is to effect pollination on compatible female gametophyte and accomplish fertilization of the egg and endosperm nuclei. For some flowering plants insects accomplish the pollen transfer, while other plants rely on wind transport. The spores of fungi and terrestrial ferns (asexual spores) are more or less exclusively transported by wind to, perhaps a suitable substratum where they germinate and develop to form new plants. Unfortunately, these airborne particles in the course of their transportation in the air enter human nostrils, throat and eyes, eliciting a sensitization that triggers off allergic rhinitis called pollinosis. These allergens have also been reported to exacerbate asthmatic condition in sufferers and cause skin reactivity that results to itching (Spieksma *et.*

al., 1985, Vittal and Glory 1985, Royes, 1987).

Airborne fungal spores and the hyphal/ mycelial fragments are major sources of inocula in plants infected by pathogenic fungi. Airborne fungal pathogens are of particular interest because of their associated plant diseases that cause agricultural losses. Spores of *Ustilago*, *Nigrospora*, *Drechslera* / *Helminthosporium*, *Puccinia*, *Cladosporium*, among others that cause seeds and cereals diseases have been commonly isolated in Nigeria (Iloba, 1996; Lawal, 1996; Olajide, 1996; Adeoti and Marley, 1995). Some species of *Aspergillus*, *Torula*, *Helminthosporium*, *Fusarium*, *Curvularia*, *Alternaria*, *Rhizopus* and *Actinomyces*, among others, have also been implicated in biodeterioration of equipment and storage materials (Pandey and Trivedi, 1981; Adeoti and Marley, 1995; Lawal, 1996 and Iloba, 1996).

The potency of pollen and spore allergens to cause a sensitization has been attributed mostly to the type of chemical composition (protein, glycoprotein and protease enzymes) of the pollen grains and spores and not actually on the quantity of the allergens in contact with the individual. However, the tendency of an individual to become susceptible to allergic reaction is inherited; hence, the sensitivity will vary from person to person depending on the genetic make-up. In consideration of these problems arising from these airborne particles, this work was aimed at identifying the various forms of airborne bio-particles especially pollen grains and spores floating in the atmosphere during the six months study period lasting from September 1999 to February 2000.

Materials And Methods

The investigation was conducted with the Agwu M6-5 atmospheric sampler, standard microscope slides, cover slips, a transparent tape, Vaseline jelly, double faced adhesive tape, 98% ethanol and a "Leica" binocular microscope. The sampling instrument (Agwu M6-5) is a passive sampler consisting of a plastic cylindrical casing with total surface area of about 125.71cm² and a volume of 123.2cm³. The cylinder has 84 openings, each of which is about 0.28cm in diameter. It has a movable base that controls the size of the opening through which specimens slides

are positioned on the plat form. Details of the efficiency of the sampler have been published in Agwu (1988) and Agwu and Osibe (1992), (Fig. 1).

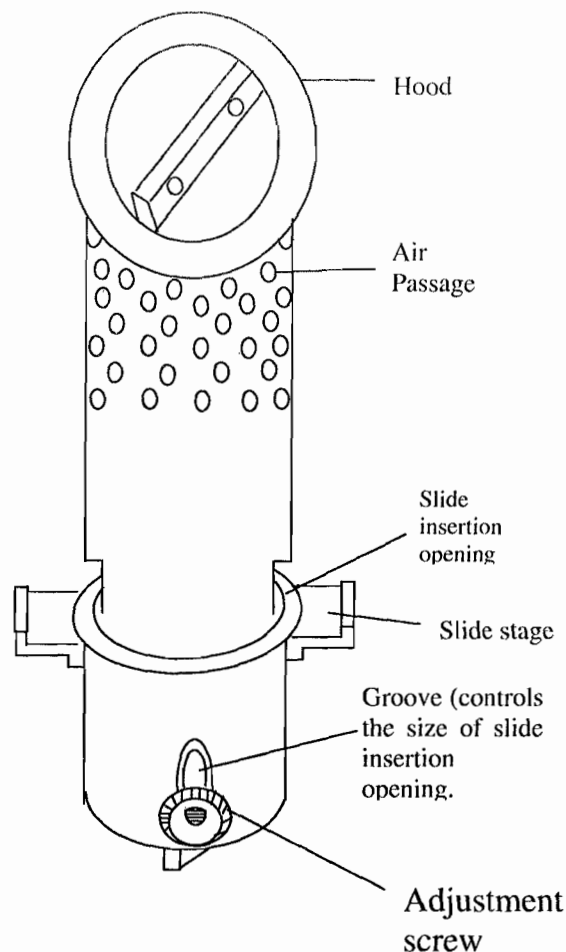


Fig. 1: Agwu M6 – 5 Pollen Sampler

Preparation of specimen slides: A piece of transparent tape 3.0cm x 2.0cm cleaned with 98% ethanol was affixed onto a sterilized standard microscope slide using a double-faced adhesive tape. Vaseline jelly was smeared lightly on the transparent tape to serve as a trapping surface. The prepared slide was then inserted in the sampler and mounted at the flat roof of the Faculty of Biological Sciences, University of Nigeria, Nsukka at a height of about 15m above ground level.

The slide was replaced every week for six months. The transparent tape on the recovered slides were each detached and immersed in gevatol on a new cleaned microscope slide and covered with a cover slip (2.4cm x 5cm). The slides were turned upside down and allowed to stand in that position for two weeks (Agwu, 2001). This was done to ensure even spread of the

trapped particles near the surface of the cover slip. The entire surface area of 6cm² containing the trapped particles for each slide was studied with a "Leica" binocular microscope at x400 magnification. The unidentified pollen and fungal spores were classed under varia / indeterminate and fungal spores, respectively. This was done to ensure that the counted pollen and fungal spores were recorded for quantitative purposes.

Results

The airborne bio-particles counted in this study consist of pollen grains, spores of fungi and pteridophytes, diatom frustules and algal fragments. Others include insect parts, charred gramineae cuticles, as well as other plant materials (Table 1). The inorganic fractions were ignored. Of the bio-particles counted, pollen grains and spores of fungi predominate in the samples. A total of 2676 pollen grains were counted, out of which thirty families were classified. Ten and 23 pollen types were identified to generic and specific levels, respectively.

The monthly pollen counts indicate that the highest airborne pollen was recorded in December (751), followed by October (597), January (513), February (346), September (242) and least in November (227). A high frequency of the pollen of *Elaeis guineensis*, Poaceae, Amaranthaceae / Chenopodiaceae, *Lannea* spp, *Berlinia grandiflora*, *Azadirachta indica*, Combretaceae / Melastomataceae and *Casuarina equisetifolia* were counted in almost all the samples examined. The most common pollen records in September were Poaceae, *Elaeis guineensis* Amaranthaceae/ Chenopodiaceae, *Casuarina equisetifolia* and *Berlinia grandiflora*. That of October belongs to Poaceae, *Elaeis guineensis*, *Casuarina equisetifolia* and *Cyperus*; November- Poaceae; December- Poaceae, *Elaeis guineensis*, *Alchornea cordifolia*, *Casuarina equisetifolia* and Combretaceae/ Melastomataceae; January- Poaceae, *Elaeis guineensis*, *Casuarina equisetifolia* and Combretaceae/ Melastomataceae, and February- *Elaeis guineensis*, Poaceae and Amaranthaceae/ Chenopodiaceae.

A total of 3317 spores of fungi were counted and of this number, 13 spore types were classified to six families and belong to twelve genera. The identified spores belong

to three classes, Deuteromycetes (imperfect fungi), Ascomycetes and Basidiomycetes. The Deuteromycetes spore types identified were those of *Alternaria*, *Asperisporium*, *Fusarium*, Johnson's grass smut, *Nigrospora*, *Tetraploa* and *Pithomyces*. The ascospores of *Venturia* and the basidiospores of *Puccinia* and *Ustilago* were identified for these two classes (Ascomycetes and Basidiomycetes).

The spores of *Cladosporium*, *Curvularia*, *Fusarium*, *Nigrospora* and *Ustilago* were fairly distributed in most of the monthly samples. The dominant spore types recorded for September were those of *Nigrospora* and *Cladosporium*; October- *Nigrospora*; November- *Drechslera Helminthosporium* and *Nigrospora*; December- *Nigrospora*, *Cladosporium*, *Ustilago*, *Venturia*, *Curvularia* and *Puccinia*; January- *Curvularia*, *Alternaria*, *Fusarium* and *Ustilago*, and that of February were *Ustilago*, *Nigrospora*, and *Drechslera/ Helminthosporium*. With the exception of *Nigrospora* that was present all through the months, the majority of fungal spore types were substantially present in the air only during December – February. The fern spores were sporadically distributed. Of the 29 fern spores counted 8 monolete and 21 trilete spores were distinguished. The diatom frustules, algal fragments, insect parts, charred gramineae cuticles, trichomes and vascular particles were not classified, rather each group was identified as an individual unit.

Discussion

Airborne pollen investigation conducted in Nsukka by Agwu and Osibe (1992) and Agwu (1997) from February to April and November to January, respectively, during the dry season were characterized by pollen of several plant taxa released from the immediate and surrounding vegetation at fluctuating rates. Among the dominant contributors were *Milicia excelsa*, *Elaeis guineensis*, *Musanga cercopoides*, *Hymenocardia acida*, Poaceae, Asteraceae, Amaranthaceae/ Chenopodiaceae, *Casuarina equisetifolia* and *Alchornea cordifolia*. These results compare favourably with that of the present work in which the common pollen grains include those of *Elaeis guineensis*, Poaceae, Amaranthaceae / Chenopodiaceae, *Asteraceae*, *Berlinia*

Moraceae	<i>Milicia excelsa</i>	-	1	3	*	10	-	*	-	*	-	8	*	-	6	2	-	-	-	2	-	1	2	1	2	
	<i>Treculia africana</i>	-	-	-	*	-	-	*	-	*	-	-	*	-	-	-	-	-	-	-	2	7	-	-	-	
Clacaceae	<i>Olax cordix</i>	-	-	-	*	-	-	*	-	*	-	-	*	-	-	4	-	-	-	-	-	4	-	-	7	1
Onagraceae	<i>Ludwigia leptocarpa</i>	-	-	-	*	-	-	*	-	*	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-	
Pinaceae	<i>Pinus sp</i>	-	-	1	*	3	-	*	-	*	-	-	*	-	-	1	-	-	-	-	-	-	-	-	1	
Rubiaceae		-	-	-	*	-	-	*	-	*	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-	
Rutaceae	<i>Fagara type</i>	-	1	-	*	2	1	*	-	*	-	1	*	-	-	2	1	-	-	4	4	1	1	1	-	
Ulmaceae	<i>Celtis grandifolia</i>	-	-	-	*	-	-	*	-	*	-	-	*	-	-	-	-	-	-	-	1	-	2	2	-	
Verbanaceae	<i>Gmelina arborea</i>	-	-	-	*	-	-	*	-	*	-	-	*	-	19	-	-	-	-	-	-	-	-	-	-	
Verifridelaminata		12	1	5	*	7	1	*	-	*	-	3	*	3	-	5	2	1	4	1	1	1	2	-	-	
	TOTAL	68	57	117	*	385	4	*	208	80	44	103	116	190	297	64	108	187	128	130	68	47	98	70	131	
Dermatiaceae	<i>Alternaria sp</i>	-	-	-	*	2	-	*	1	-	-	-	-	-	-	5	30	64	53	2	3	1	64	-	2	
Dermatiaceae	<i>Asperisporium sp</i>	-	-	-	*	2	-	*	-	-	-	1	2	2	5	5	53	-	-	-	-	-	4	3	2	
Dermatiaceae	<i>Cladosporium sp</i>	14	9	-	*	3	5	*	-	5	2	-	70	44	-	70	44	-	70	-	-	-	156	-	-	
Dermatiaceae	<i>Curvularia sp</i>	-	1	3	*	-	4	*	-	-	18	-	70	51	13	-	-	-	-	56	8	5	13	-	-	
Dermatiaceae	<i>Drechslera / Helminthosporium</i>	-	-	-	*	-	-	*	-	3	60	-	4	-	-	4	-	-	-	55	46	-	-	-	31	
Tuberulariaceae	<i>Fusarium sp</i>	-	-	3	*	3	-	*	2	2	-	-	1	46	24	-	43	2	1	3	1	3	3	2	2	
Tilletiaceae	<i>Johnson's Grass smut</i>	5	5	1	-	-	-	-	2	3	-	-	6	-	3	1	-	-	-	-	-	-	-	42	-	
Dermatiaceae	<i>Nigrospora sp</i>	11	12	3	*	29	-	*	300	3	14	5	12	270	-	15	4	2	6	5	4	4	14	3	14	
Dermatiaceae	<i>Puccinioses</i>	-	-	-	*	1	-	*	-	-	13	-	40	58	-	-	-	-	-	58	-	7	-	2	1	
Pucciniaceae	<i>Tetraria sp</i>	1	2	2	*	-	-	*	3	1	1	-	4	52	26	46	-	-	-	-	-	-	-	-	-	
Dermatiaceae	<i>Tetraploa sp</i>	1	2	3	*	-	-	*	-	1	-	-	4	40	13	24	-	-	-	40	23	36	18	27	51	
Ustilaginaceae	<i>Ustilago sp</i>	3	1	1	*	-	-	*	-	-	-	-	30	70	40	9	40	9	40	23	36	18	27	51	34	
Venturiaceae	<i>Venturia sp</i>	-	1	5	*	-	-	*	-	-	-	4	10	38	90	-	-	-	-	-	-	-	-	-	-	
	Other Fungal spores	1	4	-	*	-	-	*	1	15	*	-	19	78	152	63	14	-	-	5	12	11	-	8	17	
	TOTAL	36	37	21	*	38	9	*	307	33	109	24	235	807	402	368	144	43	223	114	41	114	108	104	104	
ALGAE	Algal cells	3	4	1	*	14	7	*	-	9	15	8	-	10	9	12	3	-	-	3	1	13	-	4	4	
	Diatoms	-	7	4	*	7	-	*	7	-	-	-	1	15	3	2	-	-	-	5	-	5	22	7	14	
	TOTAL	3	11	5	*	21	7	*	7	9	15	8	1	25	12	14	3	-	-	8	1	35	7	18	18	
PTERIDOPHYTES	Ferns (Monolete)	-	-	-	*	5	-	*	-	1	3	2	-	1	-	-	-	-	-	-	-	-	-	-	1	
	(Triete)	1	-	-	*	11	-	*	-	-	3	2	-	-	-	-	-	-	-	4	-	-	-	-	1	
	TOTAL	1	-	-	*	16	-	*	-	1	3	2	-	1	-	-	-	-	-	4	-	-	-	-	1	
PHYTOLITHS	Trichome	4	1	4	*	6	-	*	1	2	7	-	25	16	3	-	-	-	-	-	-	9	24	-	16	
	Charred Gramineae Cuticles	1	2	-	*	-	-	*	-	-	-	-	2	-	18	47	4	8	6	4	8	6	4	38	13	
	TOTAL	5	3	4	*	6	-	*	1	2	7	-	25	18	3	18	47	4	8	6	4	8	6	4	60	
Vascular particles		21	17	3	*	18	-	*	30	22	15	14	-	-	-	-	-	-	-	-	-	-	-	42	116	
Insect parts		3	2	2	*	4	-	*	1	1	-	-	7	4	4	5	5	-	-	7	7	2	1	3	3	
	TOTAL	24	19	5	*	22	-	*	30	23	15	16	7	4	4	5	5	-	-	7	7	2	1	4	119	
	GRAND TOTAL	69	70	35	*	105	16	*	345	88	149	50	268	855	477	405	194	51	298	130	47	254	128	453	453	

grandiflora, *Azadirachta indica*, *Milicia excelsa*, *Combretaceae* / *Melastomataceae* and *Casuarina equisetifolia* (Table 1). All these plants are commonly found in the indigenous vegetation of the study area with the exception of *Casuarina equisetifolia* and *Azadirachta indica* which are exotic species.

Of significance in the whole study is the dominance of pollen grains of anemophilous plant species. The flowers of these anemophilous trees, weeds and grasses are usually inconspicuous and produce large quantity of pollen grains that are usually small, light, dry and buoyant. These features enable them to be readily transported by wind (Singh 1987), and the large scale production, consequently contribute to their major abundance in the atmosphere. This accounts for their high record among offensive pollen allergens trapped in the air.

A total of 30 plant families consisting of 33 genera were counted. This number of plant taxa is an indication of the heterogeneous species present in the area, though it is merely a fractional representation of the entire vegetation characteristics and diversity of the region (cf Keay 1989). In absolute terms, it is known that pollen count is an approximate and fluctuating measure, yet it has served as an indication of the species diversity of a vegetation zone. The number of plant taxa recorded may be comparable to the findings of Agwu and Osibe (1992), who identified 32 families and 40 genera and Agwu (1997); 34 families and 52 genera during the dry season. These findings not only portray the heterogeneous nature of airborne pollen, but a reflection of the number of plants species in anthesis during the study period. Incidentally, this coincides with the findings of Keay et al., (1960, 1964), Hutchinson and Dalziel (1954-72) that most plant species of the savanna flower during the dry season in Nigeria.

Much has been reported about the abundance and cosmopolitan nature of fungal spores, and the associated plant diseases and allergies to man. Asthmatic provocations, perennial or seasonal rhinitis (runny nose, watery eyes) and itching of the skin have been reported to be associated with such identified allergic and pathogenic fungi as *Cladosporium*, *Alternaria*, *Fusarium*, *Asperisporium*, *Curvularia* and *Drechslera/Helminthosporium* (Gallup et al., 1987 and Frankland 1987). Pathologically, species of

Alternaria, *Curvularia*, *Drechslera/Helminthosporium*, *Nigrospora*, *Fusiclinia* and *Ustilago* among others trapped have been implicated in various diseases of cereals, leguminous crops, fruit rots and other economic crops in Nsukka and other regions of Nigeria (Adeoti and Marly 1995, Lawal 1996, Olajide 1996 and Iloba 1996). Their prevalence in the air is still a reflection of the serious threat to agricultural crops.

Interestingly, it has been shown in this work and other previous works in Nsukka that three groups of fungi produce the predominant spore types circulating in the atmosphere. These are the spores of Deuteromycetes, ascospores (Ascomycetes) and basidiospores (Basidiomycetes). Hurtado and Riegler-Goihman (1987), studying in Venezuela, trapped a large variety of fungal spores, majority of which are from these three groups. Their dominance in the atmosphere may be attributed to their capacity to produce large quantity of spores and high adaptability to tropical environment.

The spore counts of fern have been very poor in most aero-studies in Nigeria (Agwu and Osibe 1992, Agwu 1997). The monolete and trilete fern spores recorded, occurred sporadically in the samples. This may be due to the herbaceous nature of most ferns. They shrivel during dry season and so do not sporulate except those inhabiting humid environment. The other bio-particles counted such as charred gramineae cuticles, diatom frustules, trichomes, vascular particles, other algal fragments and insect parts are indications of the multitude of organic particles circulating in the atmosphere especially during the dry periods. Majority of these particles are allergenic and also constitute environmental hazards to human health. The charred gramineae cuticles that were prevalent from second week in December to February were released from annual bush fires heralding the onset of farming activities and hunting expeditions, while the other particles may have been re-floated from the dry environment by wind storm.

Conclusion: The airborne study indicates that numerous bio-particles especially pollen grains and spores are constantly floating in the atmosphere. Common among these are the pollen grains of *Milicia excelsa*, *Elaeis guineensis*, *Poaceae*, *Combretaceae/Melastomataceae*, *Asteraceae*,

Amaranthaceae/ Chenopodiaceae and *Casuarina equisetifolia*. Critical of all the bio-particles counted are fungal spores which include the spores of *Cladosporium*, *Alternaria*, *Drechslera* /*Helminthosporium*, *Nigrospora*, *Ustilago* and *Puccinia*. These fungal spores apart from their damage to agricultural crops and produce are potential causes of allergy either alone or in synergy with themselves or with allergic pollen grains. Further studies need to be conducted so as to have seasonal calendar of these bio-particles in the air.

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