

INTRAMURAL AEROMYCOLOGICAL

STUDIES OF

A

LIBRARY.

AEROMICROBIOLOGICAL STUDIES OF SATARA REGION

1) Historical Introduction :

Aerobiology is the study of passively air borne micro-organisms, their identity, behaviour, movement and survival. It is mainly concerned with the distribution of the micro-organisms in the air. The aerobiology deals with the study of dispersal of fungal spores, pollen grains, insect scales, protozoan cysts, algal components etc., which are transported from place to place. The air we breath varies from place to place. This fact was recognised many centuries before industrialised man assumed the right to pollute the atmosphere with poisonous chemicals and radioactive isotopes. Extensive data on the composition of the "air spora" has been accumulated during last fifty years. The aerobiological investigation deals with a variety of problems which can be grouped under two categories - i) External aerobiology and ii) Internal aerobiology. The former deals with the distribution of micro-organisms in the open atmosphere (outdoor) and the latter with closed atmosphere (Indoor). The importance of the study of air spora was realised during last 50 years and since then lot of research work was done on their existence and their effects on plant and human beings. It was found to be useful in understanding the problems of air-borne plant diseases as well as the consequences on human health. At present there are number of investigators engaged with the study of air spora from different regions.

It is a more or less established fact that the street dust is not so allergenic in comparison with the house dust. Most of the diseases of man are caused by bacteria and viruses while those of plants are caused by the fungi. The air borne pollen grains of different types of plant are allergenic in nature and cause various kinds of metabolic disorders in man. There are also number of fungal spores which cause different types of diseases like asthma, bronchia catarrh, hay fever etc.

In old days classical writers believed that the wind sometime brought sickness to man, animals and crops. Hippocrates the Father of Medical science, held that men were attacked by epidemic fevers when the inhaled air infected with "such pollutions are hostile to the human race". Lucretius in 55 B.C. held a quite and modern view. He observed the sparking of small particles on a sunbeam in a dark room and concluded that their movement must result from bombardment by innumerable, invisible moving atoms in the air. This brilliant intuition enabled him to account many interesting phenomena including the origin of pestilences. It is now known that the bodies which transmit human diseases through the air are larger than those which Lucretius thought of atoms. He thus touched on some of the main problems existing in plant pathology and allergy today.

After Lucretius, more than 1500 years passed before man even began to be aware that the air is full of microscopic living organisms. But for this discovery he had to wait

almost until the invention of the microscope. The hand made lenses of Anton Van Leeuwenhoek rendered visible the world of minute organisms whose existence had only been imagined. He doubted the belief that flies, mites and moulds were generated spontaneously by decaying animal and vegetable matter. It was P. Micheli (b 1679, d 1737) botanist to public gardens at Florence, who first illustrated "seeds" of many fungi including mushrooms, cup fungi, moulds and slime moulds. He showed that spores of some common moulds were indeed "seeds" of the fungi. He noted however that, some of his control slices also become contaminated and concluded that the spores of moulds are distributed through air (Buller, 1915).

Louis Pasteur showed that food could be preserved only in the presence of oxygen and that preservation depends on the destruction by heat of "something" contained in the air. Pasteur demonstrated visually the existence of an air-spore, and pointed out that it should be measured while in suspension and not after the deposition on the surfaces. He made first rough visual measurements of the concentration in the atmosphere of the city of Paris, and concluded that several thousands of micro-organisms were carried in suspension per cubic meter of air.

Recent studies in Aerobiology, however, had its own origin about a century ago, when Louis-Pasteur (1861) provided in his classical experiments in combating theory of spontaneous generation of life and in developing germ theory of diseases, that air is the carrier of many common germs. But over existing

knowledge regarding the composition of air spora can be said to have started accumulating during 1870's with Ehrenberg's (1872) first published information on the micro-organisms which he had collected from atmospheric dust and Cunningham's (1873) analysis of micro-organic contents of air over presidency Jails Calcutta. Other contributions followed by Miquel (1883) who elaborated techniques to analyse the microbial population of air. However, the credit for establishing the subject of microbiology of atmosphere as a special branch of study goes to Meir et.al (1933) of United States and Stepanov (1935) of U.S.S.R.

Mention may be made of the work done in India by Padmanabhan et.al (1952), Rajan, Nigam and Shukla (1952), Sanghevi, Sethi and Kasliwal (1957), Konger and Baruah (1958), Shree-ramulu and his co-workers (1958 onwards), Ganesan and Raghavan (1960), Shivpuri et.al (1960), Sena Gupta and Chattopadhyay (1963), Ramlingam (1966), Mehrotra (1969), Mishra and Shrivastava (1969), Shukla (1971) and Tilak and his coworkers (1966 onwards), Kulkarni and Kulkarni (1979) and Patil and Kulkarni (1980). Feinberg and associates (1935-1937) were the first who made a systematic and comprehensive study of fungal allergy. Further researches of Feinberg and Durham (1944), Sheldon et.al (1953), Naranjo, P. (1958), Hyde et.al (1956) have established beyond doubt that fungal spores play an important role in the etiology of nosebronchial allergy.

Blackley (1880) from his findings it was clear that the air borne fungal spores were an etiological elements in hay fever and asthma. Leeuwen (1924) from Holland presented a work in which he claimed that asthma was particularly prevalent in the lowlands and it was due to higher concentration of fungal spores in damp and low lying places. Codham (1924) reported three cases of uredinous fungus asthma in the wheat district of Canada. Hansen (1928) reported several cases of asthma in Germany due to the spores of Aspergillus and Penicillium. Since then there were many reports on hyper sensitivity to fungal spores by Hopkins et.al (1930), Flood (1931), Prince et.al (1934), Feinberg (1935), Wittich and Stalkman (1937), Wittich (1939), Chobot et.al (1940), Hampton and Lowe (1945) Prince and Morrow (1959). The choice of fungi is undoubtedly of great importance in the study of fungal allergy hence a knowledge of the fungi present in the air and their seasonal variation must be investigated. Durhaman (1938) and Bernstein and Feinberg (1942) showed that Alternaria and Cladosporium are the most common fungi in America. According to Jimenez-Dias et.al (1960) the most common air fungi are Cladosporium and Penicillium and the yeast from Madrid and Valencia. In Cardiff the most common fungi during the summer months are Cladosporium, Pullularia Pullulans, Alternaria, Botrytis, and Epicoecum during the winter months Aspergillus and Oospora (Hyde and Adam, 1960). In Copenhagen Cladosporium, Pullularia pullulans

and Penicillium are most common fungi according to Flensburg and Samson-Jansen (1950). These investigations relate to outdoor frequency scale (Nilaby, 1949, Jimenez-Oais et.al 1960 and Flensburg and Samson-Jansen, 1950). Thus it was found that there was a higher frequency of Penicillium from indoor than outdoor with lower frequency of Cladosporium. Rennfelt (1947) investigated the indoor and outdoor fungal spore contents by keeping glass dishes with fungal media for every 14 days. He found that Cladosporium, yeast and Pullularia pullulans were the most common from outdoor while Penicillium, Cladosporium and yeast were from indoor.

In India studies on Air-spores in relation to phytopathological problems were initiated by Prof.K.C.Mehta of Agra University during 1940. After Mehta's pioneering work (1952) systematic studies on air-spores were initiated by several workers. According to Shivpuri et.al (1966-68) and Vishwanathan (1966) allergy is a common disease in India. There is a great need for undertaking aerobiological and clinical studies in the various parts of this country to find out the various aeroallergens present in the atmosphere of the library.

The present investigation deals with the study of the microbial contents inside the library of Koregaon College (Satara Region). Much of the work done in Maharashtra is on exteramural aerobiology, specially from Marathwada region. Satara district is rather dry region with less humidity.

Since no aerobiological investigations were carried out in this region it was felt necessary that such investigations would be useful in understanding the composition of the air borne microbes, their seasonal variations and their correlation with meteorological conditions. Climate of Satara district is variable in different seasons. Maximum temperature is about 35°C while the minimum temperature is $13^{\circ} - 15^{\circ}\text{C}$. This area receive moderate amount of rainfall (659.4 to 1067-9 mm per annum). As Koregaon (Satara) is surrounded by cultivated patches of the fields and the factories, the atmosphere of this city is polluted. It is likely that the atmosphere within the library might have been equally contaminated with air borne microbes which may be harmful to human beings. Taking this important fact into consideration, the author undertook the study of air spora inside the library (intramural) of the local college. The detail observations on the presence of the fungal spores, insect scales, hyphal fragments, Algal Filaments, xylem Fibers, etc. are presented in this part with special emphasis on allergenic fungi.

2) Material and Methods :

The Rotorod sampler was used for this study. The Rotorod sampler was fully described by Dr. W.A. Perkins (1957). The device relies upon the high efficiency with which small air borne particles are deposited on narrow cylinders oriented at right angles to high velocity winds. A small constant speed,

battery operated motor is used to whirl thin-sticky coated brass rods about its axis at a constant-high speed. It has been developed into a cheap and portable and high efficiency sampler with high sensitivity. It is well fitted to use in the field and relatively independent of external wind speed. Collecting arms of the model are made up of 0.159 cm (1/16 inch), square section brass rods slightly bent inwards. The vertical arms are 6 cm. long and 4 cm from the axis. According to Gregory (1951) the width should give more than 60-70% efficiency of deposition for 20 μ diameter spores at wind speed above 4 m.p.h. (2 mm/sec). The model employs D.C. controlled speed motors of the type used for record players with the rods in position, the motor gives 23000 r.p.m.

3) Sampling Rate :

The sampling rate is the volume swept by the collecting surface per unit time. The dimension selected make this 2 (arms) \times 0.159 cm \times 6 cm \times 8 \times 2300 $\times 10^{-3}$
 $48.0 \times 10^{-3} \times 2300$ litres/min.
 approximately 110 litres/min.

4) Sampling Method :

Sampling was carried out by operating Rotord air sampler. The collection efficiency of this model is 85%. The petroleum-jelly is used as a adhesive on cello tape.

The Rotord sampler has been used for a wide variety of air borne particles. After the application of jelly to

the cello tape the edges of the cello tape are trimmed back to the width of the rods with sharp razor blade (The alternative would be to apply the transparent cello tape strip and then coat with adhesive). The cello tape is cut into four equal parts 1.5 cm. length before adhesive is applied and after applying the adhesive these are exposed for an hour and then mounted beneath a cover glass with suitable mountant like glycerine jelly which has the best optical properties for visual examination. It was prepared as follows :

Gelatine	=	1 gm
Glycerol	=	7 gm
Water	=	6 ml
Phenol	=	1%

5) Scanning :

The total spore counts obtained on the known areas during morning hour and evening hour were scanned under 10 x, 45 x eye pieces objective combinations of the microscope regularly. The number of spore/unit volume of the air was computed with the help of conversion factor and efficiency.

Considering the counted efficiency to be 85% with the help of conversion factor the number of spore counted on the tape of known area was readily converted into an estimated number of spore per cubic meter of air. All timings are given in Indian Standard Time (I.S.T.). The identification of the spore caught was based on - (i) Microscopical characters

(ii) Comparison with parasitic and saprophytic fungal material collected around the campus and studied microscopically, and (iii) comparison with the cultural characters. In all possible cases specific and generic counts were made which are based on the colour, shape and other diagnostic features of the spore.

6) Sampling site :

Rotored air sampler was kept inside the library of D.P. Shesale College, Koregaon (Satara) at the height of 1 meter from the ground level. Koregaon is situated between 17.87° north latitude and 74.00 east latitude. Height from main sea level (M.S.L.) is 657 meters.

7) Period during investigation :

Air spora of the library was investigated for a period of six months from 1st April to 30th September 1981. Daily two counts were taken during morning (11 a.m.) and evening (4 p.m.).

8) Weather :

During the period investigation daily record of temperature, rainfall was obtained from Agricultural department of Satara Zilha Parishad, Satara. During this period the total rainfall of Koregaon was (659 mm). During this period the minimum temperature was 13.7° C and maximum temperature was 39.0° C.