MATERIAL AND METHODS

In the Present investigation spore trapping was done by using Tilak's Continuous Air Sampler. This air sampler (Tilak and Kulkarni 1970) is indigenous one for which Prof. Tilak was awarded President's medal by the "Invention promotion board" New Delhi in 1972.The technical description and method of working of this Tilak's Continuous air sampler is as below –

The equipment and its working:-

The volumetric Tilak's Continuous air sampler is an electrically operated device, consisting of a cubical tin box of 10.4"x10.4"x8" dimension with an elevated round cap over the closing lid at the top. The Sampler requires an electric power supply (AC-230V) and provides continuous sampling of air for 7 days. The electric clock fitted in an instrument is synchronized with the drum. Air is sucked through the orifice of projecting tube at the rate of 5 Litres / minute or $0.17Ft^3$ / minute. As the air rushes in, it impinges on the transparent cellotape of the rotating drum coated with the thin layer of petroleum jelly and thus entraps the biocomponents from the air.

The rotating drum completes one circle in 7 days, thus giving the trace of catches of 7 days. Glycerine jelly was used for mounting of cellotape. Scanning was done by dividing the cellotape into equal sized 14 strips which were mounted on 14 separate clean glass slides. The cellotape faces the orifice of projecting tube 0.5 cm. away from it. The disc rotates in clockwise direction giving a continuous trace for 7 days. At the end of 7 days, the cellotape was divided into 14 equal parts as marked on the drum, each measuring 4.2 cm. in length. Each piece of cellotape obtained, represent the 12 hours sampling area for a day or night accordingly. The cellotape for 12 hours was mounted on a slide in glycerine jelly.

The air was sucked through the tube with the help of a small fan having 3 prongs and fixed in the circular opening in the cover of air sampler so as to force air out of collection chamber causing a negative pressure. An exhausts area measuring 6 x 2.7cm. was kept in a lid of the apparatus.

This sampler is modified from the spore clock model of Panzer's (1957) 24 hours slide spore collector, when it was compared with other spore traps, it was found that the Rotorod Sampler (Perkins 1957) is useful only for spot sampling, although its collection efficiency is 85%. The Hirst trap (Hirst 1953) with minimum 45%

collection efficiency has the disadvantages of the capital cost, power requirement and unsuitability both for culture and trapping splash dispersed spores. The Panzer's slide spore collector (Panzer 1957) with 70% collection efficiency has less retention capacity and requires attention after every 24 hours, whereas the Tilak's Continuous Air Sampler has 75% collection efficiency, greater retention capacity and is also economical besides it provides data for one week.

Sampling method:-

Sampling was carried out with the help of Tilak's Continuous Air Sampler with the collection efficiency 75%. The sampler was kept at a constant height of 1 meter above from the ground level in the wheat and groundnut fields in the respective seasons. Air was sampled at the rate of 5 Litres / min. and the transparent cellotape coated with petroleum jelly was changed every 7 days at about 12 hours. The petroleum jelly was used as an adhesive on cellotape. The exposed cellotape was cut into 14 equal parts, each part representing 12 hours trace area, of a day or night accordingly. The pieces of cellotape were mounted on glass slides using glycerine jelly as a mountant which has the best optical properties for visual examination. It was prepared as follows-

1) Gelatin - 40gms
2) Glycerine - 120ml
3) Distilled water - 140ml
4) Phenol crystals - 0.5gms

The required amount of Glycerine and distilled water were mixed in a beaker and heated in a water bath for 1-2 hours. During heating this mixture, gelatin was added slowly by stirring to avoid the clumping. After complete dissolution of gelatin, phenol crystals were added as preservatives and metabolic inhibitors. This glycerine jelly was used for preparation of permanent slides.

Calculations to obtain the conversion factor:-

1) Sampled area = 8.4cm x 1 cm
   = 8.4 sq.Cm.
   = 84,00,00000sq.Microns

2) Scanned area = 20 x 20 x 24

= 9600 sq.Microns

= Micron x Micron xHrs.

3) Volume of air sampled per minute = 5 litre

4) Volume of air sampled in 24 hours = 5 x24 x 60

= 7,200 Litres

5) To convert 1 litre air into cubic meter

Multiply by = 0.001000028

6) Volume of air sampled in 24 Hrs.

In terms of cubic meter = 7200 x 0.01000028

= $7.3m^3$

7) Volume of air sampled in the

Scanned area in 24 Hrs. = $\frac{9600 \times 7200}{10,00,000}$

= 69.12 litres

8) Volume of air sampled in the

Scanned area during 24 Hrs. = $\frac{1000}{69.12}$

= $14.2m^3$ ($1m^3$= 1000U)

Hence the conversion factor is 14.

The number of spores thus scanned, multiplied by conversion factor would gives the number of spores in m^3 of air.

Scanning:-

Scanning of total spore counts was done regularly under10x and 45x eyepiece objective combination of the microscope. The timing are given in Indian Standard Time (I.S.T.).The identification of spores trapped was based on

- Microscopic characters
- Comparison with the parasitic and saprophytic fungal material collected in and around the field was studied microscopically by preparing reference slides.
- Comparison with cultural characters.

The number of spores per unit volume of the air was computed with the help of conversion factor 14 and efficiency 75%. Assuming the trapping efficiency to be 75%, the number of spores counted on the tape of wheat and groundnut fields were readily converted into an estimated number of spores per cubic meter of air.

Identification of the spores was done on the basis of colour, shape, size and other diagnostic features of the spores at the generic level. The following books were referred for identification of spores –

a) Air borne pollen and fungal spores by S.T.Tilak.
b) Aerobiology by S.T.Tilak.
c) Aeromycology by S.T.Tilak.
d) Illustrated genera of imperfect fungi by N.L.Burnett.

Sampling sites:-

The present investigation deals with the aeromycological studies over wheat and groundnut fields from Karad region. Karad is a Taluka place of Satara district of Maharashtra state. Karad is situated between 17°43'north latitude and 74°11' east latitude. Height from the main sea level (MSL) is 1874 feet.

Karad lies at the confluence of Koyana and Krishna River, popularly known as 'Pritisangam'. The rivers meet exactly opposite to each other forming shape as letter "T". Moreover Krishna and Koyana originate at Mahabaleshwar and meet at Karad. It will be interest to know that their length from originating point to meeting point is almost same. That is uniqueness in the world. Karad is well known for Sugar production and known as "sugar – bowl" of Maharashtra owing to the presence of many sugar factories in and around Karad. Karad is also called as the "City of Education" as there are many colleges. It has Krishna hospital and medical research center.

The present investigation was carried out in Taluka Seed Farm located at Station road, Vidyanagar, Karad, which covers an area of about 32 acres of which an area nearly of two acre used for the cultivation of wheat and groundnut crops. The surrounding area of plot was used for the cultivation of sugarcane, fodder grass and other crops. The Eucalyptus, Tamarind and Coconut trees are present in the investigation area.

The studies were carried out by using Tilak's Continuous Air Sampler for two crops wheat in rabbi and groundnut in summer season.

1) Wheat - *Triticum aestivum* Linn.

Family – Poaceae, Variety – Kalyansona (H.D. 1953)

2) Groundnut – *Arachis hypogaea*

Family – Fabacae, Variety – J.L.24

The sampler was located in the center of selected crop field at a height of 1 meter above from the ground level. Continuous sampling was done during the investigation period.

Period of investigation:-

Sr. No.	Crop	Sampling Started	Date of Plantation	Date of Harvesting	Sampling Stopped	Total Period of Sampling
1	Wheat	1st Nov. 2007	5th Nov. 2007	25th Feb. 2008	28th Feb. 2008	119 Days
2	Groundnut	1st March 2008	5th March 2008	10th June 2008	15th June 2008	117Days

Meteorological data:-

During the investigation period, daily records of temperature, humidity and rainfall were obtained from Agricultural Research Station Karad as it affects the airspora of any location. During the investigation period (1st Nov.2007 to 15th June 2008), the maximum temperature was 33.01°C, minimum temperature 16.13°C, maximum relative humidity 86.25%, minimum relative humidity 41.94% and the total rainfall was 27.22 mm.

Fungal spore types, hyphal fragments, insect scales and parts and unclassified group of fungal spores were recorded separately.

PLATE - III

Fig. 4 Tilak's Continuous Air Sampler in Wheat Field

PLATE - IV

Fig. 5. Tilak's Continuous Air Sampler in Groundnut Field