EVALUATING POULTRY WORKERS SUSCEPTIBILITY AGAINST Fungal aeroallergens using Anderson and Rotorod sampler

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The assessment of airborne fungi was done in intramural and extramural environment. Fungal spores were trapped using Anderson two stage sampler and Rotorod volumetric sampler. Poultry feedsamples were also analyses through baiting technique of two selected poultry farms. The dominant cultures identified were Aspergillus fumigatus (16.49%) and Aspergillus flavus (12.14%) in Anderson sampler while Aspergillus niger (11.3%), Aspergillus flavus (10.5%) and Rhizopus sp. (10.5%) were obtained from baiting technique. Aspergilli group (23.3%), Alternaria sp. (11.3%) and hyphal fragments (7.3%) were found dominant in Rotorod volumetric sampler. Twenty workers were tested for Skin Prick Test with antigen of Aspergillus flavus and Aspergillus fumigatus out of which 30% were found positive with Aspergillus flavus and 15% with Aspergillus fumigatus. Than antibodies of allergic workers was checked for IgG level using ELISA techniques. IgG level of workers was very low in case of 31-70 years of age group. Among the sites the Fcal value obtained is 237.5. The degree of freedom is 9.28 that is higher than the calculated value means their is acceptance of alternative hypothesis.

Keywords: Poultry; Sampling; Skin Prick test.

Introduction
The microbial population of atmosphere at any place constitute its airspora. The micropopulation of the atmosphere is referred to airspora which varies in time, place, season and weather both in number of particles and their species composition. Airborne dust in poultry confinements was analysed to determine the correlation between airborne fungi and symptoms of allergic diseases. The objective of this study is to examine the casual relationship between the composition of airborne fungal spores in poultry house and allergic symptoms in poultry workers.

Material and Methods
Trapping of the airborne fungal spores in the intramural and extramural environment of two poultry farm near Jabalpur was done using Anderson two stage sampler and Rotorod volumetric sampler. Air samples for viable fungi were collected for 10 minutes into sterile petridishes (of 7 cm and 9 cm diameter) containing Sabourauds dextrose agar media (SDA) two times in a month by keeping the sampler at a height of 1 meter from the ground level. Petridishes containing the samples were sealed with cello tape for transport to the laboratory and incubated for 2-3 days in the BOD incubator at 28°C. Colonies were counted and identified microscopically using reference slides and available literature.

Simultaneously Rotorod Sampler was also operated for 10 minutes at 2300 rpm fortnightly. Adhesive transparent cellophane tape was cut into strips of 4x6cm which is applied on the surface of rod. After rotation strips was carefully removed and placed on the glass slides and mounted in glycerine jelly for further study. Analysis of variance (ANOVA) between sites and months two way classification for Rotorod trapped spores. Among sites Fcal value obtained is 237.5. The table value at degree of freedom 3,3 is 9.28. This is lower than the calculated value means there is acceptance of alternative hypothesis.

Result and Discussion
The aerobiological survey of poultry shed indicates the concentration and varieties of fungi prevailing at extramural and intramural sites of two poultry farm from May to August 2005. Total 22 types of fungal spores were identified in Rotorod volumetric sampler (Fig. 1). Some other bioparticles like hyphal fragments, insect part were also enumerated. Seven types of spores identified in Ascomycotina, 3 in Basidiomycotina and 12 in Deuteromycotina. The dominant spores found were 23.3% of Aspergilli group followed by Alternaria (11.3%) and hyphal fragments (7.3%).

In Anderson sampler (Fig. 2) 27 fungal cultures were

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Fig. 1. Percentage contribution of fungal spore types on Rotorod strips during May-August 2005.

Fig. 2. Fungal colonies isolated from poultry farm using Anderson sampler during May-August 2005.

Fig. 3. Fungal colonies isolated from poultry using Baiting technique during May-August 2005.
identified of which 3 of Zygomycotina 1 of Ascomycotina & 23 of Deuteromycotina. The dominant cultures obtained were Aspergillus fumigatus (16.49%), Aspergillus flavus (12.14%) and Cladosporium sp (11.05%).

The cultures obtained as a result of baiting technique (Fig. 3) are 2 to Zygomycotina 1 to Ascomycotina 20 to Deuteromycotina. The dominant cultures were Aspergillus niger (11.3%) followed by Aspergillus flavus (10.65%) and Rhizopus sp. (10.5%).

The analysis of variance (ANOVA) between sites and months two way classification for Rotorod trapped spores Among sites the Fcal value obtained is 237.5 The table value at degree of freedom 3,3 is 9.28. This is lower than the calculated value means there is acceptance of alternative hypothesis.

Table 1. SKIN PRICK TEST. Number of person showing positive reaction with Skin Prick Test against Fungal antigen.

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Antigen</th>
<th>No. of workers surveyed</th>
<th>21-30</th>
<th>31-40</th>
<th>41-50</th>
<th>51-60</th>
<th>61-70</th>
<th>Total</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aspergillus flavus</td>
<td>20</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>-</td>
<td>2</td>
<td>30</td>
</tr>
<tr>
<td>2</td>
<td>Aspergillus fumigatus</td>
<td>30</td>
<td>-</td>
<td>2</td>
<td>1</td>
<td>-</td>
<td>-</td>
<td>3</td>
<td>15</td>
</tr>
</tbody>
</table>

Table 2. Comparison of IgG level in fungal allergic and non allergic individual ELISA (490 nm).

<table>
<thead>
<tr>
<th>Age group</th>
<th>Person I</th>
<th>Person II</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>21-30</td>
<td>1.081</td>
<td>1.212</td>
<td>1.819</td>
</tr>
<tr>
<td>31-40</td>
<td>0.288</td>
<td>0.444</td>
<td>1.773</td>
</tr>
<tr>
<td>41-50</td>
<td>0.406</td>
<td>0.834</td>
<td>1.731</td>
</tr>
<tr>
<td>51-60</td>
<td>0.647</td>
<td>0.385</td>
<td>0.918</td>
</tr>
<tr>
<td>61-70</td>
<td>0.685</td>
<td>0.201</td>
<td>0.708</td>
</tr>
</tbody>
</table>

Table 3. Meterological Record of Jabalpur during May-August 2005.

<table>
<thead>
<tr>
<th>Month</th>
<th>Humidity (%)</th>
<th>Temperature (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>7 May</td>
<td>72</td>
<td>38.3</td>
</tr>
<tr>
<td>21 May</td>
<td>30</td>
<td>42.6</td>
</tr>
<tr>
<td>4 June</td>
<td>30</td>
<td>42.8</td>
</tr>
<tr>
<td>18 June</td>
<td>34</td>
<td>42.4</td>
</tr>
<tr>
<td>2 July</td>
<td>90</td>
<td>28.3</td>
</tr>
<tr>
<td>16 July</td>
<td>93</td>
<td>27.4</td>
</tr>
<tr>
<td>30 July</td>
<td>88</td>
<td>29.9</td>
</tr>
<tr>
<td>13 August</td>
<td>74</td>
<td>29.5</td>
</tr>
</tbody>
</table>
| 27 August    | 87           | 28.4             | For Anderson sampler isolated colonies among sites the Fcal value obtained is 3.6. The tab value at degree of freedom 3,3 is 9.28. This is higher than the calculated value by means rejecting the alternative hypothesis.

Twenty workers were tested with fungal antigens (Aspergillus flavus and Aspergillus fumigates). 30% of the workers were found positive with Aspergillus flavus & 15% with Aspergillus fumigatus (Table 1) than the antibodies of allergic workers were taken for IgG level using ELISA techniques. The IgG level is low in case of 31-70 years of age group that is 0.288% (Table 2).

As Aspergillus species is dominant in both the sampler as well as in baiting technique. Barter and Paster reported that the fungal growth on feeding stuffs can cause severe losses to the poultry by decreasing the nutritional value of diets. Chute and Barden reported that Aspergillus flavus constituted more than half of the fungal flora of hatcheries. The present study also showed Aspergillus flavus to be dominating fungi which is reported responsible under certain set of conditions for a serious toxicity in Turkey. Some report shows the dominance of Aspergillus in different working environment in Calcutta. Our result exhibited dominance of Aspergillus group and Alternaria recorded in all the sites. The hyphal fragment also recorded 7.3% from poultry sheds. Tilak et al. caught 3.6% from poultry sheds. Some report showed dominance of Aspergillus fumigatus and Aspergillus flavus from the lung and liver pieces of the effected birds, both the species was observed dominant from both the sites in the present study. Survey was done in Pachpedi region at Jabalpur and showed the dominance of Aspergillus flavus 24%. A worker reported the dominance of
Cladosporium sp. and Aspergillus sp. from indoor poultry which is quite similar to our findings\textsuperscript{18}. As our present result shows 30\% allergenicity with Aspergillus flavus and 15\% with Aspergillus fumigatus Verma and Vidy\textsuperscript{a}\textsuperscript{19} studied the sensitivity to TB patients by Skin prick test and found (65\%) allergenicity with Aspergillus flavus and 5\% with Aspergillus fumigatus.

The comparison of IgG level with non allergic individual and allergic individual reveals that IgG level of the worker is very low in case of 31-70 years of age group. Verma and Jain\textsuperscript{20} studied ELISA positivity against and individual antigen among 9 patients sera of 4 cancer patients (44.44\%) showed positivity against Aspergillus fumigatus antigen sera of 6 patients (66.66\%) were found positive against Aspergillus flavus antigen. Sera of 4 patient showed (44.44\%) positive reactivity against Aspergillus niger antigen.

Jacob\textsuperscript{22} worked on the meterological conditions in Aerobiology. An aeromycological study was also conducted in relation to meterological factors (Table 3). Among sites the Feal value is obtained lower than the calculated value means there is acceptance of alternative hypothesis.

References