5. DISCUSSION

5.1 The density and concentration of airborne bacteria and fungi and their variations

The quantitative results of airborne bacteria and fungi were dependent on the air sampling techniques used. The density of bacteria and fungi in this study meant the average CFUs in each opened plate in one minute because the volume of air could not be obtained during the process of sedimentary air sampling. The concentration meant the CFUs per cubic meter of air when inertial method (air sampler) was used.

The average density of airborne bacteria was 4.8 CFUs/dish/min. with a range of 0.4 and 10.3 CFUs/dish/min.from grid to grid in the rainy season. The mean concentration of bacteria was 720.0 CFUs/ m^3 in the rainy season . In the cool season, the mean bacterial concentration was 2364.4 CFUs/ m^3 with a range of 1600 and 3613.3 CFUs/ m^3 from grid to grid . The average coefficient of variation (C.V.) of bacterial density was 106.25% but the mean C.V. of bacterial concentration was 35.3% (Table 3).

The mean density of airborne fungi was 1.3 CFUs/dish/min.with a range of 0.6 and 2.8 CFUs/dish/min. from grid to grid in the rainy season. The average concentration of fungi was 1690.0 CFUs/ m^3 in the rainy season. In the cool season, the average of fungal concentration was 2842.2 CFUs/ m^3 with a range of 2146.7 and

4000 CFUs/ m^3 from grid to grid. The average C.V. for fungal density and concentration were 69.2% and 25.3% respectively (Table 6).

The different C.V. among the density and concentration of bacteria and fungi indicated that the precision of quantitative study of airborne bacteria and fungi can be improved by using inertial method (volumetric air sampler) and the results of airborne fungi (density or concentration) were better than those of airborne bacteria in terms of precision when the same method was used.

Although there was a wide range of means of density and concentration of bacteria and fungi from grid to grid, the data analysis (analysis of variance, ANOVA) still indicated that these differences among grids were not statistically significant (Table 10). One reason to explain this result was that the variation of density and concentration of bacteria and fungi within a grid was too high . The other reason was the small sampling size within a grid (n = 3).

The relationship between bacterial and fungal density indicated that the density of bacteria was higher than that of fungi in each grid except grid 1 and grid 6 in the rainy season but the relationship between bacterial and fungal concentration in the cool season was opposite to that between bacterial and fungal density in the rainy season i.e. the concentration of bacteria was lower than that of fungi in each grid except grid 1 and grid 12 (Table 8). It was possible to explain this difference between two seasons as a result of different sampling techniques because the concentration of bacteria (720.0 CFUs / m^3) in the rainy season was still lower than that of fungi (1690.0 CFUs / m^3) by inertial method when the density of bacteria was higher than that of fungi. This result indicated that the concentration of bacteria was lower than that of fungi if inertial method was used. When the sedimentary method (open plate) was used to take air samples, the airborne bacteria on the dusts might have more chance to fall down on the plates than airborne fungi because the airborne dusts were heavier than individual fungal spores. Wind speed was another possible reason to explain this difference. When air sampler was used, there were less effects from the weight of the dusts or spores and wind speed.

The results from data analysis demonstrated the differences among grids without statistical significance but the results from LSD test (Least Significant Difference test in ANOVA) showed that the lowest groups of means were still significantly different from the highest means. This indicated that the concentrations of bacteria and fungi at some grids in the study area were higher than those of other grids although generally speaking there was no significant difference among all grids.

5.2 The comparison of concentration of bacteria and fungi between rainy and cool seasons

In Chiang Mai city , the dry season consists of cool season and summer. The air sampler was used to take samples in the whole area in the cool season but only used to take samples at grid 3 and 4 in the rainy season due to the limitation of materials at that time. The comparison of seasonal differences in bacterial and fungal concentrations was based on the results at grid 3 and 4. The t-tests indicated that the differences of bacterial and fungal concentrations between the rainy and cool seasons were significantly different with p value of 0.004 for bacteria and 0.02 for fungi respectively (Tables 12,13). The concentrations of bacteria and fungi in the cool season were much higher than those in the rainy season.

These differences could be mainly explained by the effects of the weather and amount of airborne dust. In the rainy season, the water content of the air (relative humidity, RH) was higher than that in the cool season so the weather condition was favorable to the growth of bacteria and fungi. However there was another effect of rain i.e. the rain washed air frequently in the rainy season therefore the concentration of airborne dust in the rainy season was not as high as that in the cool season. The airborne dusts were the carriers of airborne bacteria and fungi especially the former. Some studies showed that the highest colony count was during the months of winter (Mullins *et al.*, 1984; Lawande and Onyemelukwe, 1984 and Abdel-Hafez 1984). Other reports indicated the peak concentration of airborne bacteria and fungi in summer (Martinez et al 1986, D'Amato et al 1984, Murgia 1984). Air temperature and sun radiation were important also. Unfortunately, the manner of interaction of temperature, radiation and others to airborne bacteria and fungi was not well understood (Lynch and Hobbie,1988).

The results from the comparison of mean densities of bacteria and fungi between two sampling batches in the same season (the rainy season) indicated the differences in the mean densities without statistical significance (Table 11).

5.3 The identification of bacteria and fungi

The resultes of bacterial identification provided useful information on the spectrum of airborne *Bacillus* species but the final identification of specimens should be obtained after the confirmation tests are done by referent laboratories or animal inoculation. There were possibilities to have the variation in biochemical reactions for the same species. The quality in isolation and the inoculation of single isolated colonies was another possibility to make results uncertain. The operator's working experience in microbiology laboratory was an important factor to affect the results.

The identification of fungi depended on the characteristics of colonies and spores. A good reference book with color pictures is indispensible. Unfortunately, several specimens can not be identified due to the following reasons. First, no spores produced in SDA media. Second, the author's mycological knowlege and laboratory skill are limited. *Aspergillus* spp., *Penicillium* spp., *Fusarium* spp., and Zygomycetous fungi were found in the rainy and cool seasons in this study. *Trichosporon* sp. and *Curvularia* spp. were only found in the rainy season. *Cladosporum* spp. was only found in the cool season (Table 7).

5.4 The comparison of the two air sampling techniques

The efficiency of air sampling method was expressed as the number of CFUs per minute. Table 9 shows that the sampling efficiency of the inertial method was much higher than that of the sedimentary method (12 times in bacteria and 60 times in fungi). In addition, the most important advantage for inertial method is that the air volume could be obtained during the sampling process so the concentration of airborne bacteria and fungi could be calculated.

The variation of results was expressed in terms of C.V. On average, the C.V.s of bacterial and fungal density obtained by sedimentary method were greater than those of bacterial and fungal concentration obtained by inertial method (Table 3, 6). The main reason was that the effects from weather, weight of the dusts and spores in inertial method was less than those in sedimentary method.

The sedimentary method is cheap and easy to operate but the quantitative results are easily influenced by the weather and other factors. The efficiency of the air sampling is low also. In the qualitative aspect, the results might gave a potential bias i.e. the heavier spores or airborne dusts have more chance to fall down to opened plates than the light spores (Booth, 1971). The inertial method (air sampler) is a kind of volumetric techniques in this field. The concentration of airborne bacteria and fungi could be obtained with relatively less variation (generally from 20% to 50%). The efficiency of air sampling is higher than that of sedimentary method. The disadvantages of air sampler includ high cost and difficulties in transportation to and operation in the field.

5.4 The risk assessment of airborne bacteria and fungi to public health

The risk assessment indicates that the high risk was shown at grid 2 (Table 15, Fig 21). This area with the high concentration of airborne bacteria and fungi possibly was the potential risk to the health of local people and tourists because of the following reasons. First, Chiang Mai is an international tourist city in tropical county even in winter the air temperature is still favorable for the growth of bacteria. Second, there are many open food stands at the road side. This operational style increases the chance of contaminating foods.

It was important to select suitable methods to classify the original data. The risk level of each grid might be changed if the original data were classified by other methods. The best way to classify original data was to adopt the standards for environmental quality. It was unfortunate that the standards of air quality on airborne bacteria and fungi were not available when this study was done.

The composition of airborne bacteria and fungi is important also when the risk of airborne bacteria and fungi is considered. In this study, most of the airborne bacteria were Gram positive rods, which were mainly composed of *Bacillus* spp. The food poisoning caused by *B.cerecus* , a toxin-mediated disease, rather than infection, has been increasing in frequency in recent years (Koneman , 1994). The genus of Gram positive cocci included *Staphylococcus* spp., which could cause staphylococcal food poisoning.

Mycotic diseases caused by common airborne fungi have become a growing, global public health problem because the normal integumentary and immunological defense mechanisms of some people were impaired by chronic diseases, antibiotic therapy and others . Airborne fungi play increasingly important role in both human and animal health (Pitt,1991 and Mishra *et al.*, 1992).

Aspergillus spp., *Fusarium* spp., and *Penicillium* spp. were found in the rainy and cool seasons in this study. These fungi are able to produce mycotoxins (Pitt, 1992 and Hocking, 1991). The significance of low-level, chronic exposure to mycotoxins has been considered in recent years (Pitt, 1991). For example, about 33.49% of *Aspergillus flavus* were aflatoxin-producing strains (Yang *et al.*, 1989). Some of the mycotoxins are possible carcinogens or mutagens (Pitt,1991).

Allergy caused by fungal allergens is another risk to public health. The composition of airborne fungi is different around the world from tropical areas to arctic zones (Cole and Kendrick, 1981). There are many foreign tourists from temperate areas visiting Chiang Mai city each year. These tourists may never have a chance to contact some kinds of fungal spores before. The allergy may happen to some susceptible individuals. Therefore it is a potential risk to foreign tourists who have high susceptibility to fungal allergens. Even the local people living in this city for a long time, there is still a possible risk for the vulnerable persons.

Hospital acquired infection is a common problem to admitted patients. This is an important aspect of the risk of airborne bacteria and fungi to human health. The outdoor air is the source of indoor air, so the common room in the hospital is exposed to outdoor air without significant decrease in microbial air contaminants if there is no air-conditioning system in the room (Kodama and McGee, 1986). The baseline information about airborne bacteria and fungi is very useful to prevent and control some hospital acquired infections. This information is helpful to design air supply systems for operating rooms, Intensive Care Unit (ICU) and other rooms in the hospitals.