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Bacterial Distribution Analysis in the Atmospheric Air of the Primary and Secondary Schools

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Abstract. Bacterial distribution analysis of the atmospheric air of fourteen primary and sixteen secondary school classes at Ibb City in Yemen was performed during the period between February and May, 2002.

The average number of blood agar grown bacteria in non-ventilated, semi-ventilated, and well-ventilated school classes were 492, 269, and 246 cfu/50 liters of air sample respectively. Primary schools showed a higher value of bacterial counts, compared to the secondary schools. The concentration of the bacteria inside the school classes was higher than those of school square. Inside the classes, in spite of approximate values of student per unit area, i.e. $0.64m^2$, the total number of bacteria per cubic meter was observed to be inversely proportional with the class volume.

Introduction

Quantitative and qualitative microbiological measurements are acquired in premises where safe environment depends on the air's content of microorganisms being kept at a very low level e.g. schools, hospital wards and premises where certain food and pharmaceutical materials are prepared.

In schools in which there is outbreak of cross-infection, it may be required to examine the air for its content of a particular pathogen. The type and number of the microorganisms in the indoor school ambient air at any time depended on a variety of factors, the most important of which are number of persons present, the extent of their body movement and the amount of disturbance of their clothing [1].

It has been reported that the major group of infection that have proved largely insusceptible to control by environmental sanitation are those of the respiratory tract, e.g. common colds, soar throats, influenza, whooping cough, pneumonia, tuberculosis,

measles and chickenpox [2]. Ayliffe *et al*, 1992 [3] have shown that the organisms that enter and leave the body via respiratory tract may be transmitted by a variety of means, including contact and air-borne secretion droplets. It is probably this versatility in their means of spread, as well as the frequency with which urban dwellers share breathing indoor air polluted by other, that explain the continuing high prevalence of respiratory infections.

Since most of the data available in the field of air pollution microbiology concerned with the fungi [4-6], while no, or little, data available concerning bacterial concentration in the atmospheric air of schools, therefore, the present work aimed, as an attempt, to study the analysis of the distribution of some medically important bacteria in the atmospheric air of the primary and secondary school classes.

Materials and Methods

Sample sites and collection

The atmospheric air samples were collected during the period between February and May, 2002, from fourteen primary and sixteen secondary schools distributed at different sectors at Ibb City, Yemen. At each school, the samples were collected from school classes and school squares. Three replicates were applied from each site. Microbiological Air Sampler (Merck, model MAS-100), previously seated at air flow volume of 50 liters, was used for atmospheric air sample collection. The altitude of the sampler was kept at 180cm above the ground level in a horizontal position. The time of sample collection was at 10pm. To find the volume of the classes, width, length, and height of the classes were measured and counted in cubic meter. Class ventilation was classified into three groups, i.e. non-ventilated (i.e. with out windows), semi-ventilated (i.e. with small windows), and well-ventilated (i.e. with large windows). Number of students per unit area of the school class reveals nearly an approximate values at all examined sites, i.e. $0.64m^2$.

Types of the examined bacteria

The following table illustrates the types of the examined bacteria, the medium used for each type, and the symbol represents the bacteria:

Bacteria	Bacteriological media	(*) Symbol
total bacteria grown on blood agar	blood agar	blood agar
total bacteria grown on nutrient agar	nutrient agar	nutrient agar
haemolytic bacterium	blood agar	haemolytic bacteria
lactose fermenting bacteria	MacConkey agar	lactose fermenter
non-lactose fermenting bacteria	MacConkey agar	non-lactose fermenter

(*) These symbolic terms were applied in the figures.

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Bacteriological analysis

After samples collection, they were subjected to the bacteriological analysis according to the method described by Collee *et al*, 1996 [7]. The time between sample collection and bacterial plating never exceeded one hour. From each site, three replicates were performed.

Preparation of the figures

To consolidate comprehension the original data were processed and expressed either as average or as a percentage and then fed to Microsoft Excel computer program to display the figures.

Results and Discussion

Figure 1. shows that, in the school classes, the average total bacteria grown on blood agar, nutrient agar, blood agar (haemolytic), MacConkey agar (lactose fermenter), and MacConkey agar (non-lactose fermenter) were 523, 441, 36, 32, and 10 cfu50L⁻¹ respectively. However, Collee, *et al*, 1996 [7] have reported in studies concerned with airborne infection of man, that the particles counted are those that carry bacteria capable growing on blood agar during aerobic incubation for 24 or 48 hours at 37^oC. This may explain the result of the present study that illustrates bacteria grown on blood agar formed the highest count, even higher than those grown on nutrient agar. This figure also reveals that blood-haemolytic bacteria formed a countable (i.e. 36 cfu 50L⁻¹) value. This result is expected since the collected samples taken from inside the school classes harboring students with variable health standards. The increased values of lactose to non-lactose fermenting bacteria, that illustrated by this figure, could be explained to the fact that strains of bacteria that ferment lactose are more widely spread than those of non-lactose fermenting bacteria [8 and 9].

The relationship between the average concentration of blood agar grown bacteria and class room ventilation is illustrated in Fig. 2. It shows that the levels of these bacteria in non-ventilated, semi-ventilated, and well-ventilated class rooms were 492, 269, and 246 cfu/50 liters respectively. It was clear that ventilation has played a role in the reduction of bacterial concentration inside the class room and vice versa. It has been reported that conventionally ventilated rooms commonly show contamination levels between 150 and 4000 cfu/m³



Bacteria in the school classes

Fig. 1. Comparison of the average numbers of bacteria in the primary and secondary school classes using the following media (*):

(*) blood agar = average bacteria grown on blood agar nutrient agar = average bacteria grown on nutrient agar haemolytic bacteria = average haemolytic bacteria grown on blood agar lactose fermenter = average lactose fermenting bacteria grown on MacConkey agar non-lactose fermenter = average non-lactose fermenting bacteria grown on MacConkey agar.



Fig. 2. The relationships between the concentration of blood agar grown bacteria and classroom ventilation in well, semi, and non-ventilated school classes.

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Figure 3. reveals that the average concentration of blood agar bacteria in the school classes and school squares in the primary and secondary schools. It illustrates that, in both school levels, the concentration of bacteria was higher inside the classes, compared to school squares. This may be explained by the body movements, inside the classes, that acted in increasing of both humidity and temperature. These two variables are known to be important bacterial growth parameters (10). This figure also shows that primary schools contained higher count of bacteria inside school classes and school squares, i.e. 370 and 356 cfu/50 liters respectively. In secondary schools these values were 279 and 224 cfu/50 liters respectively. The increased values in the primary schools may be explained by the fact that the students with younger ages in primary schools, are more vulnerable to infectious diseases, compared to elder students of secondary school.



Fig. 3. The average concentration of the blood agar grown bacteria in school classes and school squares of the primary and secondary schools.

The concentration of the average blood agar grown bacteria at two different groups of class sizes is illustrated in Fig. 4. This figure shows that, in both groups of school class sizes, in spite of a constant value of student per unit class area (i.e. $0.64m^2$), the counts of the bacteria was higher in group I (with class size of 92.5 m³). It gives a bacterial concentration of 106400 cfu/50L (i.e. 5320 cfu/m³), compared to group II (with class size of 212 m³), that gives a lower average value of 68400 cfu/50L (i.e. 3420 cfu/m³). In other word, at a constant student per class area, the class size was inversely proportional to the concentration of the bacteria inside the school class. This observation howed that the bacterial concentration of a well, ventiated classroom was 246 cfu/20 may be explained by the higher capacity of large classes to replace the ambient air of the school classes, with tresh air. This situation may be less efficient in smaller school (lasses). Moreover, such results confirm the needs for a well ventilation in the indoor areas especially at the crowded sites. It has been reported that most of the contaminants are harmless saprophytes and commensals, and even when carriers or infected students are present, usually less than

commensals, and even when carriers or infected students are present, usually less than 1% and commonly only 0.01-0.1%, of the airborne bacteria are pathogens (11). In class



Fig. 4. Comparison for the distribution of the average blood agar grown bacterial number in two different class sizes (size $I = 92.5m^3$ and size $II = 212m^3$).

rooms occupied by patient students with tonsillitis or infected wounds, *Streptococcus pyogenes* may be present between $0.1-50/m^3$ (7). Such level of contamination may seen small, it must remembered that normal person inhales about 15 m³ of air/24hr. The probability of a person becoming infected will be greatest if he is exposed to high concentration of airborne pathogens, but no level of contamination, however low, can be regarded as certainly safe. Infection may usually initiated by the deposition of a single infected particle at a favorable site in the respiratory tract, although the probability of any one such particle initiating infection is likely to be low for common pathogen, e.g. 10^{-2} to 10^{-5} for acquisition of *S. aureus* in the nose (12). It may be high for some, e.g. for acquisition of *M. tuberculosis* in the lung (13).

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تحليل التوزيع البكتيري في الهواء المحيط بداخل فصول المدارس الابتدائية والثانوية وحولها

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ملخص البحث. تم در اسة تحليل التوزيع البكتيري للهواء المحيط داخل الصفوف لستة عشرة مدرسة ثانوية وأربعة عشرة مدرسة ابتدائية في مدينة إب باليمن للفترة ما بين فبر اير إلى مايو لسنة ٢٠٠٢م.

كان معدل أعداد البكتيريا النامية على وسط الدم الصلب في الفصول بدون تهوية، وذات التهوية المتوسطة، والتهوية الجيدة ٢٦٩، ٢٦٩ و٢٤٦ وحدة مكونة للمستعمرة/٥٠ لتر من الهواء. أظهرت المدارس الابتدائية تراكيز أعلى للبكتيريا مقارنة بالمدارس الثانوية. كانت أعداد البكتيريا داخل الفصل الدراسي أعلى مما هو عليه في باحة المدرسة. على الرغم من تقارب نسبة الطالب إلى مساحة الفصل، لوحظ أن أعداد البكتيريا تناسبت عكسيا مع حجم الفصل.