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INDOOR MICROBIAL AIR CONTAMINATION IN SOME COLLEGE ROOMS AND HOSPITALS IN BASRA CITY

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ABSTRACT: Total of 120 samples have been collected from indoor air of hospitals and the College of Science, University of Basra, during the period from Dec. 2018 to Jan. 2019, in Basra governorate, Republic of Iraq. The study included samples from indoor air of Basra Teaching Hospital and Ibn Ghazwan Hospital for women and children. 96 bacterial isolated, 46 of which were in the College of Science, 25 in Ibn Ghazwan hospital, and 21 in Basra Teaching hospital. The results of the current study showed the differentiation of the presence of bacterial isolates that are Gm+ bacteria outperformed the number of isolates on Gm- bacteria, as the number of Gm+ bacteria reached 69, while the number of Gm- bacteria reached 21 bacterial isolates. The results showed the presence of *Staphylococcus* in all study sites, rare bacterial species have also been isolated for the first time in the indoor air of hospitals in Iraq (*Staph. hominis, Staph. haemolyticus, Granulicatella elegans* and *Pantoea* sp.) in Ibn Ghazwan hospital and (*Staph. warneri, Staph. vitulinus, Kocuria kristinae, Kocuria rhizophila* and *Pseudomonas fluorescens*) in Basra Teaching hospital, many of them cause hospital acquired infections. The sensitivity of bacterial isolates to some common antibiotics has been tested as streptomycin, vancomycin, Gentamycin, tetracycline and erythromycin. *Staph. vitulinus* has been shown to be resistant to all antibiotic used, while other bacterial isolates showed a different variation in their sensitivity to antibiotic.

Key words: Indoor air, microbial contamination, hospitals, college of science.

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INTRODUCTION

Most people spend 90% of their lives inside buildings in homes, offices, schools, universities and hospitals etc. (Abd EL-Hameed and Farag, 1999). So, the Environmental protection Agency (EPA) considered that indoor air quality is one of the most important environmental health problems, these problems are generally caused by two types of conditions, the first being the lack or inadequate ventilation inside the building, and the second is the exposure to one or more sources of pollution in the buildings (MS Hospital Consulting, 2001; Halli *et al*, 2015).

Air inside buildings can be polluted by a number of pollutants such as bacteria and other microorganisms (Amarasekera *et al*, 2010). Human activities inside indoor areas are the main factors that increase air pollution by microbes. Other sources are the presence of animals, plants m cooling system, dust and other factors that come from outside air (Jones, 1999). Microorganisms

that pollute the air inside buildings can cause allergies, respiratory and immune diseases (Douwes *et al*, 2003).

The complex environment of hospitals requires special attention to ensure air quality within hospitals, the protection of patients, as well as health care personnel from hospital infections (Verde *et al*, 2015). The presence of high levels of microorganisms in the air of the internal environment of hospitals is a worrying factor with regard of infection caused by these microorganisms (Lugauska and Krikstaponis, 2004), it also gives an indication of the degree of cleanliness of the internal environment of the hospitals that contains different types of microorganisms (Saad, 2003).

Hospitals may be the source of infection for many diseases such as tuberculosis, diarrhea and other serious diseases, as well as problems of increasing disease resistance of common antibiotics (Emuren and Ordinioho, 2015), in the United States, for example, there are about 2 million patients infected with hospitals due to air pollution,

where about 90 thousand people die yearly from these infections (Shokri *et al*, 2016).

Air pollution in hospital buildings, universities and buildings with a large number of people is affected by the weather and season, as well as the design and operation of the ventilation system, humidity rate, visitors, human activities and the age of the building (Verde *et al*, 2015).

This study was conducted to evaluate and determine type and number of bacteria in two hospitals in Basra governorate south of Iraq: Ibn Ghazwan hospital for women and children and Basra Republic hospital. This topic is of great importance globally because the infection that affects some patients is an infection acquired by them during their hospitalization, such infection has serious consequences in terms of increasing the number of mortality and infections, length of stay in hospital in addition to increase the total cost. Some bacterial species have also been isolated from air of the internal environment of University of Basra, College of Science, because large numbers of students spend most of their time in university buildings and they need safety and the susceptibility of isolated bacterial species to resistance to some commonly used antibiotics was also studied.

MATERIALS AND METHODS

Sampling sites

Samples were collected from indoor air for offices, scientific laboratories of the College of Sciences, University of Basra, Iraq, the collection on samples has been done two times at 9: AM and at 1: PM.

Samples were collected from indoor air for the children's, pre – mature infants and gynecological halls at Ibn Ghazwan hospitals for women and children, samples also collected from the esoteric and women's lounge and surgery halls of Basra Teaching hospital. Samples have been collected during winter in December 2018 and January 2019, air temperature was measured at the collection sites.

Passive air sampling

Samples were collected by using passive air sampling method. Petri dishes with different media [Nutrient agar, Blood agar, MacConky agar, Manitol salt agar and MeRes agar base (for Staphylococcus, Methicillin resistance)], were placed at a height of one meter in the center of the room and left open for 1 hour at the sites where samples are collected. The plates the transferred directly to the lab., where they were placed in an incubator at 37° C for 48 hours.

After the incubation period, the number of colonies

for all isolates is calculated as (CFU) Colony Forming Unit (Awosika *et al*, 2012).

Diagnosis of bacterial isolates

Bacterial isolates were transported separately and cultured on Petri dishes, then incubated at 37°C for 48 hours. The cultivar properties of the pure bacterial colonies were studied in terms of phenotypic characteristics, which included the shape, color, edges, diameter and texture of the colonies. The microscopic characteristics of the bacterial cells staining with Gram stain were studied in terms of the shape of cells, their regularity with each other and the type of interaction with Gram stain (Macffaddine, 2000).

 $VitEK_2$ device (at the private Mowasat hospital in Basra) identification system that provides highly accurant and reproducible results (Al-Mijalli, 2016).

Antibiotic susceptibility test

The test was preformed according to (CLSI) Clinical and Laboratory Standard Institute method, using disk diffusion method, the ability of bacterial isolates to resist a range of commonly used antibiotics was tested: Vancomycin 30mcg, Streptomycin 10 Mg, Gentamycin 10 mcg, Tetracycline 10 mcg and Erythromycin 10mcg from Bio-analyses company, Meullar Hinton agar was used from (Himedia, India).

Five tablets of antibiotic were distributed in each plate grown with bacterial isolates, which placed in the incubator at 37°C for 24 hours, inhibition zones were measured using a ruler inserted in mm (CLSI, 2011).

RESULTS AND DISCUSSION

The results of the current study, which collected 120 of indoor air for buildings, were distributed as follows: 40 samples from Basra Teaching Hospital, 40 samples from Ibn Ghazwan Hospital for women and children and 40 samples from College of Science, University of Basra in Basra province southern of Iraq. The ratio of the presence of bacteria in the College of Sciences were the most.

Also 95 bacterial isolates from indoor air isolated and diagnosed in the buildings under study. The temperature ranged between $26 - 28^{\circ}$ C in winter, as the isolated of bacteria were (Mesophillic aerobic bacteria), the low temperature and increase of humidity lead to an increase in the presence of bacteria in the air, this is consistent with (Valeria *et al*, 2003).

The results of the current study showed that the number of bacterial isolates in hospital indoor air is less than their ratio in college indoor air, 25 bacterial isolates were recorded in the indoor air of Ibn-Ghazwan hospital building, while the number of bacterial isolates within



Fig. 1: Initial isolation of bacteria from the air.



Fig. 2: Granulicatella elegans, isolated for the first time in Iraq.

Basra Teaching Hospital were 21, as for the College of Sciences, the number of bacterial isolates was higher than the other stations, especially at lecture hours at noon, 49 bacterial isolates were recorded.

The results are in agreement with Stryjakowska - Sekulska *et al* (2007), where he confirmed that the air pollution increased in the afternoon compared to the morning in the various rooms in universities, especially during the lectures, also Karwowska (2003) found that the presence of large numbers of bacterial species increases in the places where people crowd in workplaces, especially in rooms and hallways. As for the reason for the presence of bacteria inside the hospital, it may be due to the constant violation of medical staff inside and outside the halls and the inadequate cleaning and sterilization, or when the patient carries the bacteria from the contaminated operating bed to the hallway and this is consistent with Bengtsson *et al* (1979).

Hospitals from which bacteria have been isolated are among the hospitals with old buildings in the governorate where the old buildings have old – style ventilation systems and need repair, restoration and maintenance work as the spores of bacteria can enter the buildings through ventilation systems (Dhanasekaran *et al*, 2009), likewise microorganisms can reproduce in the water that collects

drainage tubes from ventilation or moisture formed from the secondary roofs of buildings and this is part of the problems in old buildings (Quaaylac, 1997). The ventilation air conditioning systems are considered one of the most important factors affecting the air quality in buildings (Wallace *et al*, 2004).

In general, the presence of microorganisms in buildings may be affected by several variables such as humidity, temperature, ventilation, number of inmates, human activity, concentration of particles such as dust, as well as air quality outside the building (Verde *et al*, 2015).

It became clear from the results of the current study after Gram staining that the number of Gm+ bacteria is more than Gm-bacteria at all study stations, as the number of Gm+ bacteria isolates reached 69, while the number of Gm- bacteria was 26 isolates and thes results are consistent with the results of Abed-raboo (2015), which found that Gm+ bacteria are more frequent than Gmbacteria in the air in hospitals buildings in Palestine. The results also agreed with a study of Yassin and Almoug – atea (2010), where the bacteria were isolated from Kuwait University, they found that the percentage of Gm+ bacteria was greater than the Gm- bacteria, while, Al -Mijalli (2016) shown in his study, were the bacteria isolated from the air inside the school buildings in KSA, that the percentage of Gm-bacteria was more than Gm+, as well (Badri et al, 2016) study that isolates bacteria from the indoor air of school buildings in Baghdad, Pseudomonas Gm- bacteria was common in this study.

The difference in the results of this study from the others is due to the differences in the geographical locations in the isolation areas and in environmental conditions.

The results of this study showed that Gm- bacteria were less than of Gm+ bacteria, because it has the ability to survive in the presence of biological materials and it ends with the evaporation of water and the removal of these substances (Saa,a, 1999), also (Jarvis and Martine, 1992) confirmed that the source of Gm- bacteria is not as the results of contamination, but rather it is spread in places with high humidity

Bacterial species

Bacterial species in the indoor air of College of Science

Staphylococcus: was the common species in the current study at all locations, where air sample were taken inside the college, the number of *Staph. aureus* was 22 isolates, in addition to other species. Staph. is Gm+ cocci bacteria, normal in human skin and mucous membrane, it

can spread and from skin peeling and remain in the air (Styiak *et al*, 2007).

Bacillus sp.: The number of Bacillus sp. Were 14 isolates, it is spore forming bacteria, can survive for an extended period of time and cause many medical problems (Emuren and Ordinioha, 2016).

E. coli: Nine isolates of *E. coli* were isolates from college's lab. and corridors, it is enteric Gm-bacteria, this is consistent with Karwowska (2003), who isolated *E. coli* from lab. rooms and hallways in schools and Stryjakowska – Sekulska *et al* (2007), who isolates from toilets inside University.

Pseudomonas aeroginosa: Four isolates from labs and office rooms were isolated, it is obligate aerobic non capsule forming Gm- bacteria (Kiska and Gilligan, 2003). It largely isolated from the air inside school buildings, according to Badri *et al* (2016).

Bacterial species in the indoor air of hospitals

Staphylococcus sp.: 22 isolates were isolated of Staph. aureas in Ibn Ghazwan hospitals, also 4 isolates of Staph. haemolyticus from premature babies Dept. and 2 isolates of Staph. hominis from women,s section. In aforementioned hospital has been isolated. Staph. hominis is uncommon bacteria has been isolated for the first time from hospitals indoor air.

Staph. aureas and Staph. haemoluticus were isolated by Alla – Werdi and Mehdi (2015) of blood poisoning cases in newborn children in Kirkuk city were children acquire it from the hospital environment during or after birth and its isolation was not recorded in the air. Ikhtiar et al (2017) found that Staph. aureas at hospitals in Indonesia was the most frequent bacterial isolates, they cause various types of infections such as urinary tract infection, skin infections, respiratory infections and food poisoning.

There are also disease called nosocomial infection resulting from contact with carrier of the disease, directly or indirectly, through the air or other methods (Verde *et al*, 2015). *Staph.* bacteria is considered as one of the opportunistic pathogenic bacteria which acquired the urinary tract in hospital (Nimmo and Coombs, 2008), it's illnessis attributed to its ability to hemolysis (Mims *et al*, 2004). Possessing the blood – analyzing enzyme is an important factor of ferocity and for supplying bacteria with iron (Dhakal and Mulvey, 2012), it is also produce capsule which is one of ferocity factors as it plays an important role in inhibiting the phagocytosis process (Ahmed *et al*, 2007), in addition to the presence of pili which the adhesion factors, bacterial invasion is embedded in the host cells (Carrol *et al*, 2016); therefore,

Staph. are considered to be very dangerous bacteria, in indoor hospitals air, because of the patient's low immunity and easily infected.

Eight isolates of *Staph. vitulinus* and two of *Staph. warneri* were isolated from Basra teaching hospital, two types are uncommon and haven't been isolated from the air previously. They cause acquired infections in hospitals and may have been present in the internal air of the hallways, which were isolated from them due to droplets of urine from patients with urinary tract infections in the internal medicine Dept., were microorganisms can remain as bio aerosol and spread, as a results infection occurs (Moletta – Denat *et al*, 2010).

Granulicatella elegans: Two isolates from the women's lounge at Ibn Ghazwan hospital have been isolated for the first time in Iraq, it's a Gm+ rare bacteria, it was first known in 1960 from a heart condition (endocarditis) and classified as separate genus depending on 16s rRNA (Collins and Lawson, 2000), it occurs in mouth as normal flora (Aas et al, 2005), however, it can cause a variety of infections, which are not common in medical isolates, and are often isolated from blood, abscesses and bone marrow (Christensen and Facklam, 2001).

E. coli: Seven isolates have been isolated in children's lounge at Ibn Ghazwan hospital, it's Gm-coliform bacteria, it is believed that water, food, the patient's intestine in hospitals are the most important source of pollution with *E. coli* (Haley *et al*, 1985).

Pantoea sp.: Gm- rare bacteria isolates from children lounge at Ibn Ghazwan hospitual, it is an opportunistic that causes acquired infections and wound injuries in hospitals (Jain *et al*, 2012). In other studies, it was isolated from children's hospitals from the blood stream, urinary tract and joint abscesses, as well as causing lung infections and organs transplants (Shubov *et al*, 2011). In 1970 – 1971, it caused 378 cause of septicemia in a hospital in USA (Maki *et al*, 1974), it has the ability to resist antibiotic and minerals and pathogenic for plant, animals and humans (DeMaayer *et al*, 2012). In Iraq Pantoea has been isolated from urine sample from a child with urinary tract infection at Kikuk teaching hospital and samara hospital (Al – Douri and Maaroof, 2018).

Kocuria sp.: Four isolates of Kocuria kristinae from the surgical department and two isolates of Kocuria rhizophila from the internal medicine Dept. at Basra teaching hospital have been isolated, it,s cocci Gm+ uncommon bacteria, occur as normal flora on the skin and in oral cavity, causing respiratory tract infections

(Mashouf *et al*, 2009). In a study by Al – Dori and Maaroof (2018), two types *Kocuria kristinae* and *Kocuria rosea* have been isolated from urine samples of patient with urinary tract infection of Kikuk and Samara Teaching hospital were among the uncommon isolates.

Pseudomonas fluorescense: Two isolates of this type were obtained from women's gynecology at Basra teaching hospital, which is bacilli Gm+ bacteria, it has been isolated from sputum in a study by Al –Dori and Maaroof (2018), it also has been isolated from wounds by Abas and Salih (2014) in Dhi – Qar governorates and also from skin by a patient, who had been in an Iranian hospital by (Ghana and Azimi, 2014). It wasn't isolated from the air inside any hospital buildings before except in the current study. Pseudomonas is considered as one of the most opportunistic infection agent, as it causes wound infections post operation, burns and middle ears infections and also bacteremia (Mims et al, 2004). It's a pathogenic bacteria in case of weakened body immunity in cases of wounds, burns and people with diabetes (Reese et al, 2000). It can reproduce in humid environments without the need for complex nutrients (Lennette et al, 1985). It has the ability to spread and stay in hospital environment because it has may virulence factors in addition to it's resistance to many antibiotics and antiseptics, which makes it a threat to the lives of patients in hospitals (Pena et al, 2003).

As the researchers, Amitoni and Wilson (1991) indicated, the importance of the cilia that the bacterium has in it's adhesion to the epithelial cells that line the gut and respiratory tract

Antibiotic sensitivity

The results of the current study showed that bacterial isolates were somewhat resistant to the antibiotics used, the reason may be that the bacteria have been isolated from the hospital environment and their control is so difficult. *Staphylococcus vitulinus* showed resistance to all the antibiotics used in the current study, the reason may be attributed to taking the antibiotics in large dozes without referring to the doctor and repeated infection with the same pathogen, in addition to staying for a long time in hospitals, as well as the lack of resort to the sensitivity test to determine the appropriate antibiotic in the treatment of various conditions.

Deikema *et al* (2004) explained that the resistant of Vancomycin to microorganisms are Staphylococcus vitulinus, Pantoea sp. and Pseudomonas flurescens are resistant to above antibiotic, the results of this study are consistent with the results obtained by Alla – werdi and Mehdi (2015), they found that *Staphylococcus warni*

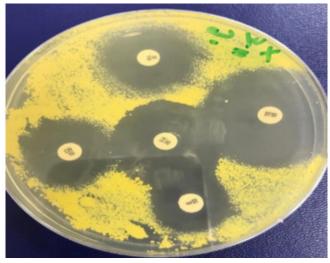


Fig. 3: Antibiotics sensitivity test.

are sensitive to all antibiotics and Staph. hemolyticus are resistance to Erythromycin, this agree with the results of the current study.

CONCLUSION

There is pollution in the University,s indoor air with aerobic bacteria, Types of microbes change and different during the day in college, as the afternoon is more because of the presence of student activity. The presence of microbes contamination in the indoor air of hospitals, which increases the risk of acquired diseases.

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