INVESTIGATION OF INDOOR AND OUTDOOR AIR BACTERIAL DENSITY IN TEHRAN SUBWAY SYSTEM

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ABSTRACT

The wide use of subway system by citizens underlines the importance of hygienic issues including indoor air pollution in these public places especially in metro stations. The aim of this study was to investigate the bacterial contamination in indoor and outdoor air of two metro stations (Imam Khomeini and Sadeghiyeh stations) in Tehran subway system. In this cross sectional study, three sampling locations were selected in each station. Also, sampling was conducted in indoor air of two types (old and new) of trains. The range of bacterial colony count was 35-1501 CFU/m^3. Maximum and minimum bacterial contamination levels in Imam Khomeini and Sadeghiyeh platform stations were averagely 1073 CFU/m^3 and 242 CFU/m^3, respectively. 14 bacterial species and genera were isolated; among them the dominant species were *Staphylococcus epidermidis*, *Micrococcus luteus* and *Bacillus spp*. Results showed that bacterial concentrations in indoor air were higher than the outdoor air; also the bacterial counts correlated significantly with number of the passengers ($p<0.001$) and air temperature ($p<0.001$).

Key words: Tehran Subway; bacterial density; bioburden; indoor air; out door air

INTRODUCTION

Nowadays, subsurface transport systems - or common words subway systems- are commonly being used in major cities of the world in order to improve the quality of transport, reduce traffic and air pollution. This solution has a very important role as a management index in the planning of large cities (Vasconcellos, 2001). Operation of subways began in the mid-nineteenth century. Millions of passengers spend part of their time in the subway systems and a large number of employees are working in these systems (Nieuwenhuijsena *et al.,* 2007).Based on some features such as being trapped, often limited ventilation, special sources of pollutant release and specific environmental conditions, subway systems are considered as a special environments (Salma *et al.,* 2007). Although one of the most important purposes of subway systems are reducing ambient air pollution, but extension of subway systems have their own consequences, so that it can be said that, an under ground city with new challenges and issues such as air pollution in closed spaces of stations and trains has emerged new challenges which can be more severe than ambient air pollution problems.

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The primary studies related to effects of pollutants on subway passengers were done in the late 1980s in the state of Boston (Chan *et al.,* 1991; Chillrud *et al.,* 2005). Thereafter, more studies followed assessing levels of air pollution in metro systems. In more than a decade of researches, various pollutants such as suspended particles (PM) (Furuya *et al.,* 2001), polycyclic aromatic hydrocarbons (PAHs) (velasco *et al.,* 2004), volatile organic compounds (VOCs) (Shiohara *et al.,* 2005), nitrogen oxide and carbon monoxide gases (Dor *et al.,* 1995; Fernandez *et al.,* 1995; Chan *et al.,* 1999), metals (Seaton *et al.,* 2005) have been measured. Many qualitative and quantitative studies have been done with airborne biological pollutants in various indoor environments such as hospitals (Jeffrey *et al.,* 2003), schools (Liu *et al.,* 2000), houses (Pastuszka *et al.,* 2000) and other residential areas; however, there are limited studies and information related to measurement of these air pollutants in indoor subway stations.
Airborne biological contaminants include bacteria, fungi, viruses and Pollens (Colls, 2002). Between these biological contaminants, those bacteria with ability for spore formation and tolerance to adverse environmental conditions and longer survivals can create different health problems including respiratory diseases (Colls, 2002). Considering such hazards and lack of any study related to airborne bacteria in Tehran's subway stations, this study was designed to specify bacterial bioburden of indoor air in Tehran subway.

MATERIALS AND METHODS

The research field in this study was metro stations in Tehran. Within the stations of four operating lines two stations with different location, structure and crowding rate were selected. The first station, "Imam Khomeini", is an underground station located at cross point of two most busy lines (lines 1 and 2). It is one of the most crowded metro stations in Tehran, and is equipped by mechanical ventilation systems and maintained under positive pressure to remove pollution. The second station, "Sadeghiye", is a surface station except for its metro office area which is underground. It is characterized by good natural ventilation and fewer passengers' density. Two sampling points were considered within each station was two points: platform and hall of station office area. Besides, indoor air of trains (both old and new ones) and also adjacent outdoor air of the stations were studied. Air sampling for presence of bacteria was conducted in respiratory height (about 1.5 m) for 2 min using a microbial air sampler (Quick Take-30, SKC, USA) operating at a flow rate of 28.3 L / min, once every 6 days for 4 months from December to March (Jeffrey *et al.,* 2003; Kaoruko *et al.,* 2005). Sampling time was 9-11 am. During sampling period, number of passengers in 2 m radius, relative humidity and temperature were measured and recorded. Samples were transferred through transport media to Tryptic Soy agar. After incubation for 48 h at 35 ± 5° C, colonies on each plate were counted and reported as colony forming unit (CFU/ m^3). Species of bacteria were determined using diagnostic tests such as Gram staining and biochemical tests including catalase, oxidase, DNAse, Bile esculin, urease, resistance to Bacitracin and novobiocin (NB) disc, sugar and other diagnostic tests.

RESULTS

During four months, 19 samples were taken from each of the eight sampling sites. The bacterial count ranged from 35 CFU/m^3 in outdoor air of Imam Khomeini station to 1501 CFU/m^3 in air samples of platform of this station. Table 1 shows the density of bacteria in different sampling locations. The number of passengers of the sampling points varied from 4 passengers at the outside of the station of Imam Khomeini to 120 passengers in old trains.
During the study, relative humidity and temperature ranged from 21% to 38% and 6^{oC} to 27 ^{oC}, respectively. Table 2 shows Pearson's correlation coefficients of the bacterial counts and other variables. Bacterial density in term of CFU/m^3 had significant

Table1: Statistical indices of bacterial colony in different sampling locations

Statistical Index	Density of bacteria (CFU/m^3)							
	Imam Khomeini station			Sadegiye station			Trains	
	Outdoor air	office area	Platform	Outdoor air	office area	Platform	New	Old
Min	35	212	530	53	530	70	53	123
Max	565	1519	1501	618	1325	494	724	919
Mean	268	790	1073	245	828	242	376	492
S.D	155.37	293.55	246.25	131.27	208.86	120.34	201.23	221.20

Table 2: Correlation coefficients[a] between counts of bacteria and number of passengers, temperature andrelative humidity during sampling

CFU/m^3 and statistical index	Number of passenger (2-meter radius)	Temperature (ºC)	Relative humidity (%)
Density of bacteria (CFU/m^3)	0.518*	0.247*	0.068
Range	4-120	6-27	21-38
Mean	64	17	27

* p< 0.01 (2- tailed).
a. Pearson's correlation coefficients

correlation with number of passengers in the 2 m radius and temperature ($p<0.001$). No significant correlation was found between bacterial density and relative humidity ($p>0.01$). Levels of bacterial contamination in different parts of stations and in new and old trains were compared separately with one-way ANOVA test. Between the two stations, according to obtained $p<0.01$, a significant difference between the average concentration of bacteria in term of CFU/m^3 in the air of these two stations was observed. Mean bacterial concentrations of new and old train shows no significant difference between two types of trains. Based on diagnostic tests, totally 14 different genera and species of bacteria were isolated. Types of isolated bacteria and their average (in term of CFU/m^3) during the period of sampling in different points are given in Table 3. *Bacillus spp* (22.44% of identified genera), *Staphylococcus epidermidis* (14.37% of identified genera) and *Micrococcus luteus* (13.38 of identified genera) were predominant bacteria (Fig.1).

DISCUSSION

According to the results, maximum bacterial contamination was in Imam Khomeini station with an average of 1073.16 CFU/m^3 and minimum was in sadeghiye station with an average of 242.58 CFU/m^3. High concentration of bacterial contamination in Imam Khomeini station platform can be attributed to the high population density at this sampling site and the fact that the station platform is located at greater depth below the ground, which results in poor ventilation and little air current in this station. The low

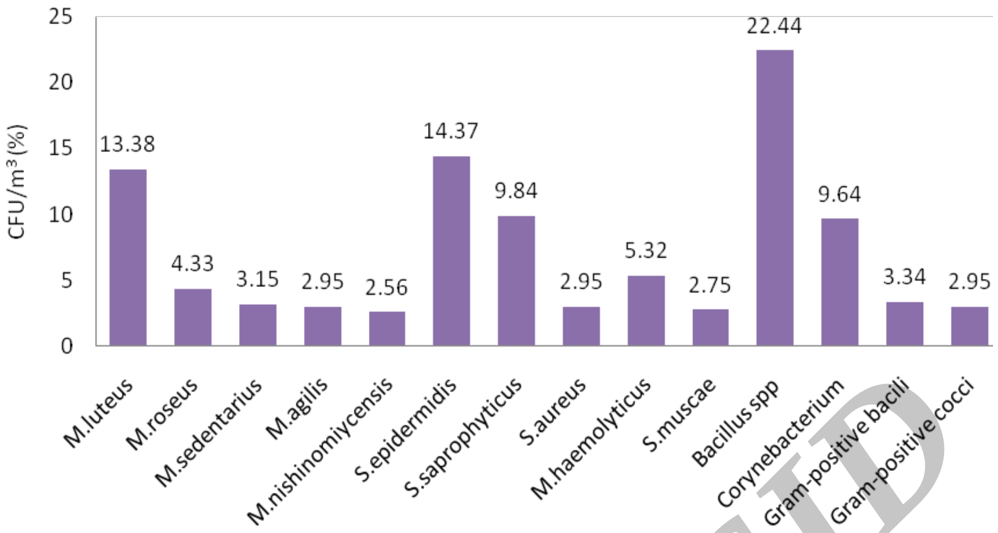

Fig 1: Comparison of percentage of average count of different bacteria in all sampling points

Table 3: Types of isolated bacteria and their average, in term of CFU/m3, during the period of sampling in differing points

Bacteria	Mean density of bacteria (CFU/m3)							
	Imam Khomeini station			Sadegiye station			Trains	
	outdoor air	office area	platform	outdoor air	office area	platform	new	old
M. luteus	41	86	119	31	103	38	44	88
M. roseus	5	46	25	17	35	2	15	6
M. sedentarius	1	23	46	0	41	9	6	3
M. agilis	0	28	48	0	22	6	1	15
M. nishinomiycensis	3	23	51	6	18	6	3	6
S. epidermidis	43	114	140	35	121	34	40	96
S. saprophyticus	30	62	110	37	81	35	31	41
S. aureus	1	24	46	0	35	0	2	19
S. haemolyticus	22	53	70	5	40	6	12	25
S.muscae	0	15	55	5	22	0	1	6
Bacillus spp.	96	167	133	79	182	81	162	124
Corynebacteriums	26	107	96	25	88	16	49	55
Other G^+ bacilli	0	23	61	0	23	6	4	4
Other G^+ cocci	0	19	53	5	17	3	6	4
Total	268	790	1073	245	828	242	376	492

concentration of pollution in the Sadeghiyeh station platform can be ascribed to the fact that this station is located on the surface.

Regarding the trains, the bacterial contamination in the old trains was higher than new ones (mean 491 CFU/m^3 versus mean 375 CFU/m^3; $p < 0.01$). This can be attributed to the fact that the interior space of old trains is separated and confined, while in the new trains this space is interconnected along all parts of the trains, so the passengers move relatively more evenly in different parts of the train and air movement is better achieved . Also, due to new technologies in these trains, their ventilation performances are better.

In both stations, the concentration of bacteria for the outdoor air was less than that of indoor air. Few studies have been done to measuring bacterial contamination in metro stations. In a study performed at two metro stations in Cairo, Egypt, the average concentration of bacterial contamination ranged from 2.9410^3× CFU/m^3 in the tunnel station to 2.8110^3× CFU/m^3 in surface station (Awad, 2002,(this result was150–1380 CFU/m^3 in an underground concourse in Tokyo (Kaoruko *et al.,* 2005). Climate and environmental conditions including ventilation would influence the variation of bacterial counts. In closed spaces, the number of people affects on concentration of air- transmitted bacteria (CEC, 1993). Since human activities affects air temperature in closed environments, so in this study, in addition to the correlation between population and bacterial concentration, significant correlation was also observed between temperature and concentration of bacteria. Comparison of this analysis with results of other studies on air quality in confined spaces shows acceptable consistence. In a study in Tokyo, there was a significant correlation between population and concentration of bacteria (Kaoruko *et al.,* 2005). Also the results of this study are consistent to the results obtained in indoor air of a hospital in Singapore (Jeffrey et al., 2003). The Gram-positive bacteria dominance in the air has been reported in many confined environments including classrooms (Liu *et al.,* 2000), residential rooms (Pastuszka *et al.,* 2000) and health care centers (Sarica *et al.,* 2002; Shintani *et al.,* 2004). In our study, however, *Bacillus spp*, *Staphylococcus epidermidis* and *Micrococcus luteus* were dominant captured bacteria. The dominance of Bacillus spp. can be attributed to the ability of these bacteria to form spores and to be resistant against adverse environmental conditions. Since *Micrococcus spp*. and *Staphylococcus epidermidis* are constituents of the normal human skin flora (CEC, 1993), it is likely that the airborne bacteria in the stations are originated from passengers skin flora.

Due to high concentrations of airborne bacteria in confined spaces like metro stations, it is necessary to perform cross sectional surveys and monitor ventilation systems and air conditioners at these stations. This study has provided an evidential example of bacterial bioburden in the indoor and outdoor airs of two metro stations in Tehran subway system.

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