

# Normal and dusty days comparison of culturable indoor airborne bacteria in Ahvaz, Iran

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**Abstract** Important sources of chemical and biological indoor pollutants include outdoor air, the human body and human activities, emission from materials, furnishings, appliances and use of commodities. The main purpose of this study was to identify culturable indoor airborne bacteria in normal and dust event days in indoor environments of a school, a hospital and a university in Ahvaz city, which individuals such as children, teenagers, adolescences and old people had activity there. Samples were collected using the biostage sampler, an

Andersen-based method, with a flow rate of 28.3 l/min, from July 2010 to March 2011. Temperature and humidity were measured and registered in each time of sampling. The identification of bacteria was performed to genus level by using appropriate methods and standard biochemical tests. Gram-positive bacteria in both normal and dust event days with more than 90 % had the highest concentration and frequency. Predominant bacteria in normal and dust event days were *Staphylococcus* spp. (72.9, 87.9 %), *Streptomyces* spp. (60.9, 62.1 %), *Bacillus* spp. (94, 89 %) and

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*Micrococcus* spp. (65.4, 71.2 %), respectively. The highest concentrations of bacteria in normal and dust event days were in winter. The range of bacteria in normal and dust event days were 0–4,800 and 210–10,000 cfu/m<sup>3</sup>, respectively. There was a significant difference between the concentration of bacteria in normal and dust event days ( $p = 0.001$ ) and also a significant association was found between the concentration of total bacteria with temperature and humidity ( $p < 0.05$ ). The concentration of bacteria in dust event days was 1.8 times higher than normal days. Consequently, the concentrations of bacteria in all three sampling sites were higher in dust event days than normal days indicating the impact of dust storms on increased bacterial concentration in indoor environment.

**Keywords** Biostage sampler · Backward trajectory · Khuzestan province · Indoor air · HYSPLIT

## 1 Introduction

Indoor air pollutants can be classified in different ways. One approach is to divide them into chemical, physical and biological agents. Another approach is to classify them on the basis of origin. The origin of a particle has an important impact on its composition, which may include chemical and biological agents besides the physical nature of the particle itself. Indoor air quality has been influenced by numerous factors such as type of sources, ambient conditions, building structure and materials, occupant behavior and activities, ventilation and air exchange speed. For indoor environments where there are no apparent indoor sources, occupant-related activities may become a major source of particles (Tippayawong et al. 2009). It is mostly believed one of the major factors influencing indoor particulate levels is the outdoor source (Abt et al. 2000). Many factors have effect on the concentration of bacteria in indoor environment such as ventilation, concentration of particulate matter, activities in environments, population and air quality of outdoor environment (Jo and Seo 2005; Fang et al. 2007).

Therefore, indoor to outdoor relationships in view of bioaerosol can be investigated to enable authorities to act against biological sources, especially when indoor environment is not properly maintained. Biological

activities in the atmosphere are complicated which are related to climate and weather changes, local biological sources, and artificial and natural pollutant sources such as dust storms (Ho et al. 2005). Dust storm cause increasing in the level of particulate matter concentration as well as fraction size such as PM<sub>10</sub>, PM<sub>2.5</sub> and PM<sub>1</sub> in indoor and outdoor environments.

Some clinical and model studies have shown that airborne particle and gaseous pollutants have become a growing concern for the public after proofing a relationship between exposure and adverse health effects (Oberdorster et al. 1995; Pope et al. 1995; Goudarzi et al. 2012, 2013).

It is identified that man inhales nearly 10 m<sup>3</sup> of air every day (Adhikari et al. 2004). Youngsters of school ages are more liable to some environmental contaminants than adults because they inhale greater volumes of air to their body weights, and their tissues and organs are growing (Tippayawong et al. 2009; Mendell and Health. 2005).

Bacteria are in all places of our existing environment, and they have natural and anthropogenic sources. Dust storm whether it is originated from natural or anthropogenic sources plays an important role in carrying pathogens or increasing the biogeographically range of various bacteria by helping long-distance spreading events (Kellogg and Griffin 2006; Soleimani et al. 2013). Dustborne microbes in particular can directly affect human health by pathogenesis, contact of those sensitive to cellular components, pollen and fungal allergens, lipopolysaccharide, etc., and the increase of sensitivities (i.e., asthma through long exposure) (Griffin 2007). Epidemiological studies have shown that diseases attributed to indoor environments such as humidifier fever, asthma and Sick building syndrome (SBS) are related to presence of pollutants particularly bioaerosol in those places. Bacteria are commonly identified as causes of infectious diseases (Pastuszka et al. 2000; ACGIH 1989; Hwang et al. 2010).

In the last decade, dust storm has added to other environmental problems particularly in the Middle East region. Since 2002, the Middle Eastern dust storm frequencies and intensities have increased significantly in Khuzestan province in which dust storm frequencies in Ahvaz as the capital city of this province were 29, 33 and 55 in 2005, 2006 and 2007, respectively. In some periods, the residence time was 48–72 h. Hospital admission rate increasing, cancellation of flights, governmental and private

sectors closure, crops loss and migration (brain drain) can be detrimental effects and social outcomes of this phenomenon (Derakhshandeh et al. 2014; Goudarzi et al. 2014; Soleimani et al. 2013; Shamsavani et al. 2012a, 2012b).

A large number of studies have conducted to examine microorganisms in ambient air during dust events in the world (Griffin et al. 2006; Kellogg et al. 2004; Kwaasi et al. 1998; Lyles et al. 2005; Prospero et al. 2005; Christina et al. 2006; Schlesinger et al. 2006; Soleimani et al. 2013). Many investigations related to ambient air quality in Ahvaz city have been conducted recently (Goudarzi et al. 2014; Heidari-Farsani et al. 2014; Dehdari Rad et al. 2014), but microbial quality especially bacteria have not investigated at indoor air environments during dust event days in Ahvaz yet. Therefore, the aim of this study was to investigate the bacteriological aspect of incoming dust storm at indoor environments of educational and therapeutic places in Ahvaz city.

## 2 Material and method

### 2.1 Sampling approach

Samples were collected from indoor air of school, university and hospital, where individuals had activity there, with the biostage sampler (SKC Inc., PA, USA), an Andersen-based method (Andersen 1958). Samplings were performed in school and university classrooms, but in hospital, internal wards were selected. The flow rate of biostage sampler was 28.3 l/min and sampling times were 2 and 5 min. Based on preliminary experiments, the sampling time in normal days was 5 min, and it was 2 min for dust event days. Sampling devices were put in 1.5–2 m above the ground and with more than a meter from any walls and other obstacles (Kim et al. 2009; Fang et al. 2007; Jo and Seo 2005). Sampling was performed once every 6 days in three seasons and in different places of three stations, 266 times during normal days and 66 times in dust event days. In each place, sampling was done twice a day in the morning (10–12 am) and in the afternoon (4–5 pm). The average of temperature and relative humidity was measured by using humidity and thermometer devices (Barometer Model: PHB-318) at each point of sampling. Geographic view of Khuzestan province, Ahvaz urban area, sampling stations and wind rose are shown in Fig. 1.

Before starting, the device was calibrated and the biostage part was sterilized with 70 % alcohol. Trypticase Soy Agar (TSA, Merck, Germany) was used as a culture media to isolate bacteria. Cycloheximide (500 µg/l) as a fungal growth inhibitor was added to TSA (Kim et al. 2009; Fang et al. 2005; Jo and Seo 2005). After autoclaving, these culture mediums were distributed on the plates, in the sterile condition beside of flame. Then, the culture medium was placed inside biostage, and sampling was done for 5 and 2 min during normal and dust event days, respectively.

### 2.2 Experimental procedures

Bacteria-containing medium was placed for 48–72 h into incubator with a temperature of 35–37 °C (Kim et al. 2009; Fang et al. 2007). The numbers of colonies were reported as colony-forming units per air volume (cfu/m<sup>3</sup>). Colony-forming units per cubic meter of air was calculated as follows:

$$\frac{[\text{Number of colony of bacteria (cfu)}]}{[\text{sampling time (min)} \times \text{velocity of air flow (l/min)} \times 0.001 (\text{m}^3/\text{l})]}$$

Bacterial genus identification was conducted using macroscopic and microscopic examinations of Gram-stained prepared smears and standard biochemical tests (Augustowska and Dutkiewicz 2006). Besides, the spore staining and partial acid-fast staining were used to identify *Bacillus* and *Nocardia* genera, respectively. *Enterobacteriaceae* was identified using API kit (Fang et al. 2007).

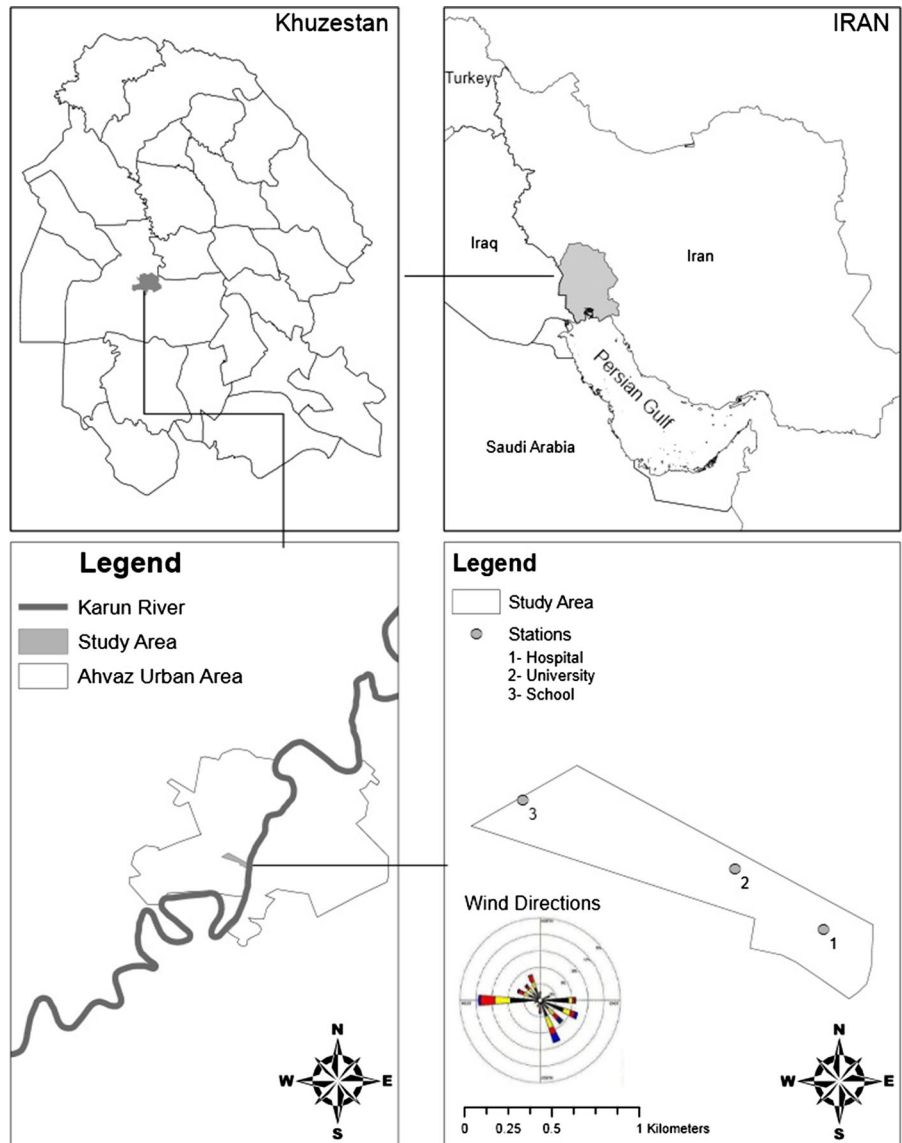
### 2.3 Indoor air quality guideline

Many criteria and guidelines associated with indoor and outdoor air quality have been reported in the world to enable researchers and scientists to provide a better judgment about their findings. A guideline for comparing our result with good indoor air quality is presented in Table 1.

### 2.4 Statistical analysis

Statistical analysis was conducted by using SPSS version 17. Analysis of variance was used to compare means at different places, conditions and seasons. The

**Fig. 1** Geographic illustration of Khuzestan province, Ahvaz urban area, sampling stations and wind rose



**Table 1** Recommended maximum concentrations for specific classes of contaminants (NEA 1996)

Parameter	Limit for acceptable indoor air quality (cfu/m <sup>3</sup> )
Total bacterial counts	500
Total fungal counts	500

one Sample Kolmogorov–Smirnov test was employed to evaluate the normality of the data. If the data were not distributed normally, Kruskal–Wallis test was used for comparing samples in different places or seasons. Mann–Whitney *U* test was employed to indicate

significance of bacteria concentration differences in normal and dust event days. Spearman’s correlation coefficient as nonparametric measure was used to realize statistical dependence between airborne bacteria concentration and some of meteorological data such as temperature and humidity during two circumstances, respectively.  $p < 0.05$  was considered as significant.

### 2.5 Backward trajectories

An internet- or web-based hybrid single-particle lagrangian integrated trajectory model (HYSPLIT)

**Table 2** Concentration of airborne bacteria, temperature and relative humidity measured during normal and dust event days in three stations

Parameter	Weather condition	University <sup>a</sup>			Hospital <sup>b</sup>			School <sup>c</sup>					
		Min	Max	Mean	Min	Max	Mean	Min	Max	Mean	Median		
Total bacteria (cfu/m <sup>3</sup> )	Normal	14.0	2,964	306.5	171.0	8	1,350	315.9	234.0	132	4,800	600.8	526.5
	Dust	280.0	3,798	702.5	658.0	210	822.5	520.6	507.5	315.0	10,000.0	1,047.6	533.0
Temperature (°C)	Normal	18.5	36	25.9	24.3	20	27	24.8	24.0	21.5	32.0	25.0	24.6
	Dust	16.0	33	22.3	20.3	20	26	21.3	21.0	19	27.0	21.7	22.5
Relative humidity (RH %)	Normal	24.0	72	37.5	34.0	29	65	42.7	42.0	18	65.0	40.9	40.8
	Dust	25.0	58	32.2	31.4	26	38	33.0	33.2	32.0	55.0	42.3	42.3

Number of samples for normal days: <sup>a</sup>  $n = 88$ , <sup>b</sup>  $n = 90$ , <sup>c</sup>  $n = 88$

Number of samples for dust event days: <sup>a</sup>  $n = 22$ , <sup>b</sup>  $n = 22$ , <sup>c</sup>  $n = 22$

was used to realize backward air mass trajectory of incoming dust storm. Coordination of 48°0.68' and 31°0.32' was introduced to the model as longitude and latitude of Ahvaz city, respectively. It was used to investigate the origin, route and direction of dust storms.

### 3 Results

#### 3.1 Concentrations of bacteria in different sites

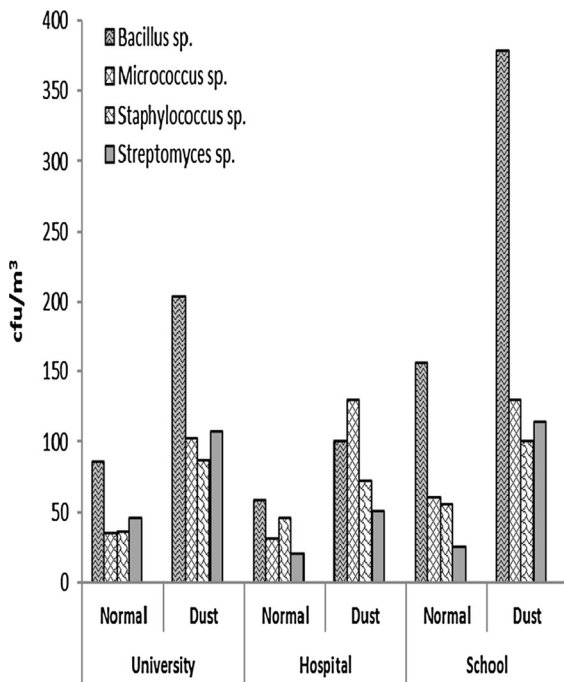
Mean, median, minimum and maximum of bacteria concentration as cfu/m<sup>3</sup>, temperature and humidity in three sampling points in normal and dust event days from July 2010 to March 2011 are shown in Table 2. The concentrations of extracted bacteria in sampling points in normal and dust event days were between 8–4,800 and 210–10,000 cfu/m<sup>3</sup>, respectively.

Mean concentration of bacteria in normal and dust event days in school was higher than two other points. Also mean concentration of bacteria at school (600.8 cfu/m<sup>3</sup>) was higher than two other points during normal days. Although total concentration of bacteria at school was significantly different from university and hospital ( $p < 0.001$ ), there was significant difference for total concentration of bacteria between university and hospital ( $p = 0.041$ ) during normal days.

Predominant bacteria in normal and dust event days were *Bacillus* spp., *Micrococcus* spp., *Streptomyces* spp. and *Staphylococcus* spp. (Fig. 2). In normal situation in school, the concentrations of *Bacillus* spp., *Staphylococci* spp. and *Micrococcus* spp. were 156.3, 55.7 and 60.8 cfu/m<sup>3</sup>, respectively, which were higher than the university and hospital. Mean concentration of *Streptomyces* spp. in university (45.5 cfu/m<sup>3</sup>) was higher than the other two sampling points.

In normal situation, mean concentrations of *Bacillus* spp. in university, hospital and school were 85.4, 58 and 156.3 cfu/m<sup>3</sup>, respectively, also there was a statistically significant difference ( $p = 0.005$ ) in the concentrations of *Bacillus* between school and hospital (Table 3).

During dust event days, there were no significant differences ( $p = 0.114$ ) in concentration of *Bacillus* spp. between hospital with university, but there were significant differences ( $p < 0.001$ ) in concentration of *Bacillus* spp. between school and two other points



**Fig. 2** Concentration of culturable dominant bacteria in three sampling sites during normal and dust event days

(Table 3). There was no significant difference in the total concentrations of bacteria between hospital with school and university ( $p > 0.05$ ), also concentrations of *Bacillus* spp., *Staphylococcus* spp. and *Streptomyces* spp. in school were 378.3, 100 and 114.2 cfu/m<sup>3</sup>, respectively, which were higher than other places.

### 3.2 Bacteria concentrations in different seasons

Mean, median, minimum and maximum of the total bacteria concentration, temperature and humidity in normal and dust event days of three seasons (summer, autumn and winter) are presented in Table 4. The highest mean concentrations of bacteria genera during normal and dust event days were 537.7 and 1,353.8 cfu/m<sup>3</sup>, respectively, which occurred in winter. In normal and dust event days, there was a significant difference in total concentrations of bacteria in summer with these concentrations in autumn as well in winter ( $p < 0.05$ ).

The concentrations of predominant bacteria such as *Bacillus* spp., *Micrococcus* spp., *Streptomyces* spp. and *Staphylococci* spp. in different seasons were shown in Fig. 3. In winter and normal situation, the

**Table 3** Average concentrations (cfu/m<sup>3</sup>) of predominant bacteria in normal and dust event days and evaluation of differences between stations

Parameter		Mean concentration (cfu/m <sup>3</sup> )		Evaluation of significant differences (comparisons between stations)			
		Normal days	Dust event days	Normal day		Dust event days	
				Hospital	School	Hospital	School
<i>Bacillus</i> spp	Hospital <sup>a</sup>	58.0	100.4	–	–	–	–
	School <sup>b</sup>	156.3	378.3	$p = 0.005$	–	$p < 0.0001$	–
	University <sup>c</sup>	85.4	203.5	$p = 0.593$	$p = 0.078$	$p = 0.144$	$p = 0.005$
<i>Micrococcus</i> spp	Hospital	30.7	129.8	–	–	–	–
	School	60.8	129.8	$p < 0.0001$	–	$p = 0.996$	–
	University	35.4	102.1	$p = 0.613$	$p = 0.009$	$p = 0.974$	$p = 0.949$
Staphylococci spp	Hospital	46.4	72.2	–	–	–	–
	School	55.7	100.0	$p = 0.651$	–	$p = 0.554$	–
	University	35.2	86.5	$p = 0.133$	$p = 0.537$	$p = 0.991$	$p = 0.467$
<i>Streptomyces</i> spp	Hospital	20.0	50.1	–	–	–	–
	School	25.7	114.2	$p = 0.281$	–	$p = 0.042$	–
	University	45.5	106.7	$p < 0.0001$	$p = 0.003$	$p = 0.078$	$p = 0.958$

Number of samples for normal days: <sup>a</sup>  $n = 90$ , <sup>b</sup>  $n = 88$ , <sup>c</sup>  $n = 88$

Number of samples for dust event days: <sup>a</sup>  $n = 22$ , <sup>b</sup>  $n = 22$ , <sup>c</sup>  $n = 22$

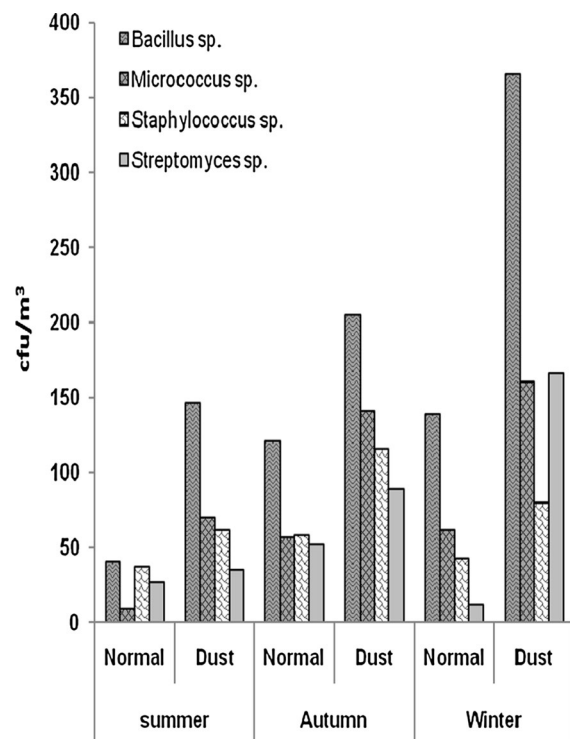
**Table 4** Concentration of airborne bacteria, temperature and relative humidity measured during normal and dust event days

Parameter	Weather condition	Summer <sup>a</sup>				Autumn <sup>b</sup>				Winter <sup>c</sup>			
		Min	Max	Mean	Median	Min	Max	Mean	Median	Min	Max	Mean	Median
Total bacteria (cfu/m <sup>3</sup> )	Normal	46.0	295.0	180	120.5	10.0	4,800.0	500.2	439.5	8.0	1,872.0	537.7	428.6
	Dust	210.0	998.0	484.9	455.0	420.0	998.0	581.3	525.0	472.5	10,000	1,353.8	673.5
Temperature (°C)	Normal	28.0	36.0	29.0	32.0	18.5	33.0	24.2	24.2	20.0	27.0	23.1	23.7
	Dust	25.0	33.0	28.2	29.4	17.0	25.6	18.5	22.0	16.0	24.0	17.5	19.4
Relative humidity (RH %)	Normal	24.0	52.0	33.5	33.0	29.0	72.0	42.5	40.4	18.0	65.0	44.9	42.5
	Dust	32.0	43.0	36.2	36.5	25.0	58.0	39.6	38.0	26.0	48.0	30.4	29.0

Number of sample for normal days: <sup>a</sup>  $n = 88$ , <sup>b</sup>  $n = 90$ , <sup>c</sup>  $n = 88$

Number of sample for dust event days: <sup>a</sup>  $n = 24$ , <sup>b</sup>  $n = 24$ , <sup>c</sup>  $n = 18$

mean concentrations of *Bacillus* spp. and *Micrococcus* spp. were 138.4 and 61.5 cfu/m<sup>3</sup>, respectively, which were higher than other seasons. The mean concentrations of *Staphylococcus* spp. and *Streptomyces* spp. in autumn were 58.3 and 52 cfu/m<sup>3</sup>, respectively, which were higher than summer and winter. In normal and dust event days, there was a significant difference in concentration of *Staphylococcus* spp. between summer with winter and autumn ( $p < 0.05$ ).



**Fig. 3** Concentration of culturable dominant bacteria in three sampling seasons during normal and dust event days

During dust event days, the mean concentrations of *Bacillus* spp., *Micrococcus* spp. and *Streptomyces* spp. in winter were 365.4, 160.4 and 166.2 cfu/m<sup>3</sup>, respectively, which were higher than summer and autumn, whereas the mean concentration of *staphylococci* in autumn (115.6 cfu/m<sup>3</sup>) was higher than the other seasons.

### 3.3 Comparison of bacteria concentration in normal and dust event days

Table 5 shows the frequencies of identified bacteria in three sampling points. As it is shown in this table, 19 genera of bacteria and two species were identified. In normal situation, frequencies of *Bacillus* spp., *Micrococcus* spp., *Staphylococcus* spp. and *Streptomyces* spp. in university, hospital and school were 67.0–94.3, 52.2–91.1 and 61.4–96.6 %, respectively. In dust event days, they were 68.2–86.4, 54.5–100 and 63.6–100 %, respectively. The frequencies of other Gram-positive bacteria in normal situation at university, hospital and school were 0–50.0, 0–44.4 and 0–46.6 %, whereas they were 0–45.5, 0–45.4 and 0–54.5 % during dust event days, respectively (Table 5). The frequencies of Gram-negative bacteria in normal and dust event days were 0–33 and 0–36.4 %, respectively.

According to Fig. 4, concentrations of bacteria in dust event days were 1.2–17 times more than normal days in which the most increasing ratio was related to *Bacillus cereus* (17) and the least was associated with *Enterobacter* spp. (1.2). This ratio for total bacteria was 1.8. There was a significant difference in mean concentration of total bacteria between normal and dust event days ( $p = 0.001$ ).

**Table 5** Overall frequencies of airborne bacteria, in three sampling stations during normal and dust event days

Gram-positive	Weather condition	University <sup>a</sup>	Hospital <sup>b</sup>	School <sup>c</sup>
<i>Frequency (%)</i>				
<i>Bacillus</i> spp.	Normal	94.3	91.1	96.6
	Dust	86.4	81.8	100
<i>Micrococcus</i> spp.	Normal	73.9	58.9	63.6
	Dust	72.7	63.6	77.3
<i>Staphylococcus</i> spp.	Normal	67.0	72.2	79.5
	Dust	81.8	100	81.8
<i>Streptomyces</i> spp.	Normal	69.3	52.2	61.4
	Dust	68.2	54.5	63.6
<i>Corynebacterium</i> spp.	Normal	8.0	34.4	4.5
	Dust	22.7	45.4	50.0
<i>Microbacterium</i> spp.	Normal	10.2	8.9	14.8
	Dust	9.1	45.4	22.7
<i>Stomatococcus</i> spp.	Normal	0	6.7	6.8
	Dust	31.8	45.4	40.9
<i>Arcanobacterium</i> spp.	Normal	4.5	0	0
	Dust	0	0	4.5
<i>Dermabacter</i> spp.	Normal	0	2.2	3.4
	Dust	31.8	0	9.1
<i>Rhodococcus</i> spp.	Normal	2.3	3.3	3.4
	Dust	4.5	0	22.7
<i>Brevibacterium</i> spp.	Normal	2.3	2.2	0
	Dust	4.5	0	9.1
<i>Deinococcus</i> spp.	Normal	4.5	18.8	3.4
	Dust	22.7	0	18.2
<i>Arthrobacter</i> spp.	Normal	4.5	3.3	4.5
	Dust	31.8	22.7	18.2
<i>Bacillus cereus</i> .	Normal	0	18.8	0
	Dust	18.2	4.5	9.1
<i>Cellulomonas</i> spp.	Normal	0	2.2	2.3
	Dust	0	4.5	9.1
<i>Nocardia</i> spp.	Normal	17.0	3.3	0
	Dust	45.5	27.2	40.9
No identification	Normal	50	44.4	46.6
	Dust	36.4	45.4	54.5
Gram-negative	Weather condition	University <sup>a</sup>	Hospital <sup>b</sup>	School <sup>c</sup>
<i>Frequency (%)</i>				
<i>Achromobacter</i> spp.	Normal	11.4	8.9	4.5
	Dust	0	18.2	9.1
<i>Pseudomonas</i> spp.	Normal	4.5	4.4	3.4
	Dust	18.2	13.6	9.1
<i>Enterobacter</i> spp.	Normal	4.5	0	0
	Dust	9.1	0	13.6
<i>Serratia</i> spp.	Normal	2.3	0	11.4
	Dust	18.2	0	13.6
<i>Klebsiella pneumoniae</i>	Normal	5.7	0	3.4
	Dust	9.1	13.6	0
No identification	Normal	33.0	24.4	17.0
	Dust	31.8	22.7	36.4

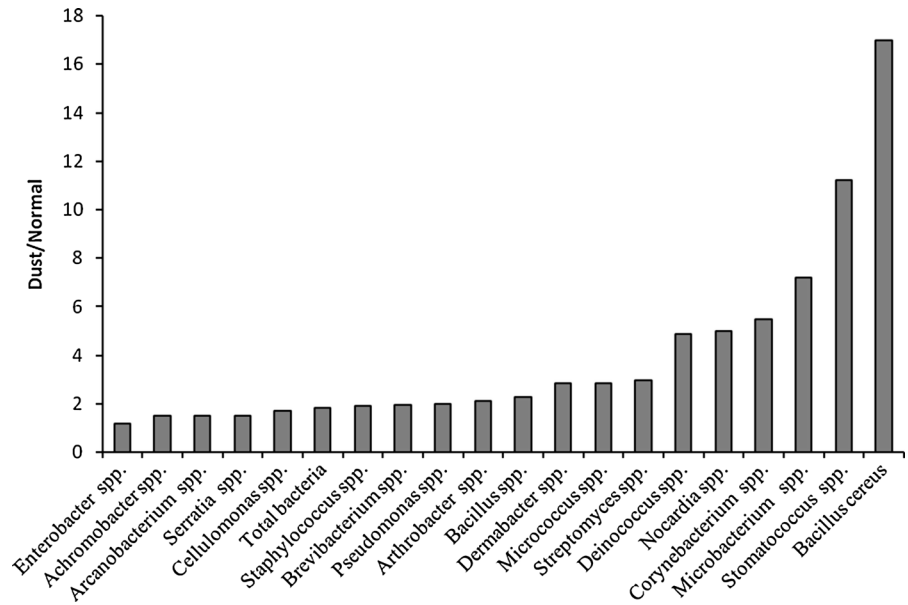
Number of samples for normal days: <sup>a</sup>  $n = 88$ , <sup>b</sup>  $n = 90$ , <sup>c</sup>  $n = 88$

Number of samples for dust event days: <sup>a</sup>  $n = 22$ , <sup>b</sup>  $n = 22$ , <sup>c</sup>  $n = 22$

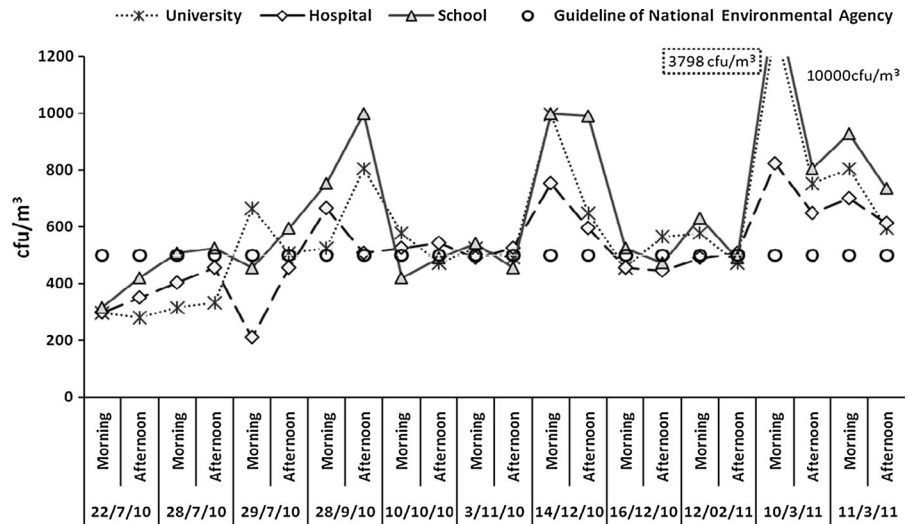
Frequency = number of positive samples/total number of samples



**Fig. 4** The ratios of indoor airborne bacteria in dusty days to normal days



**Fig. 5** Airborne bacteria concentration in comparison with NEA guideline

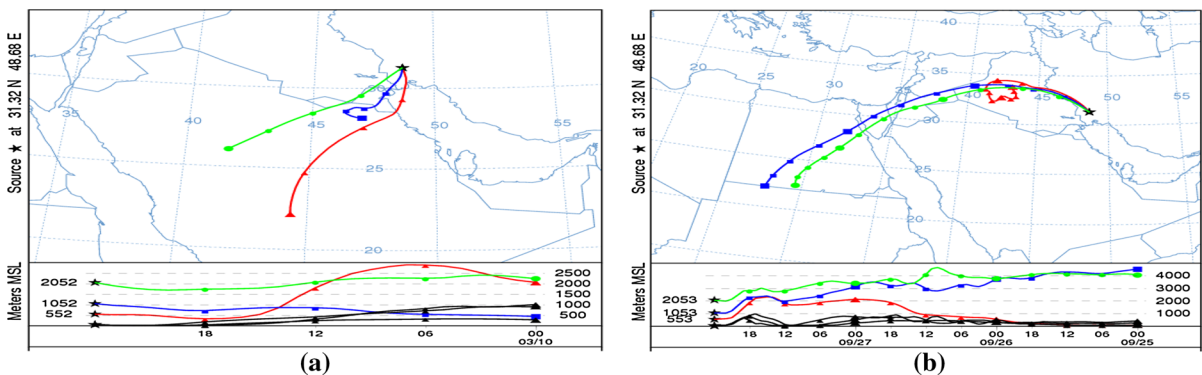
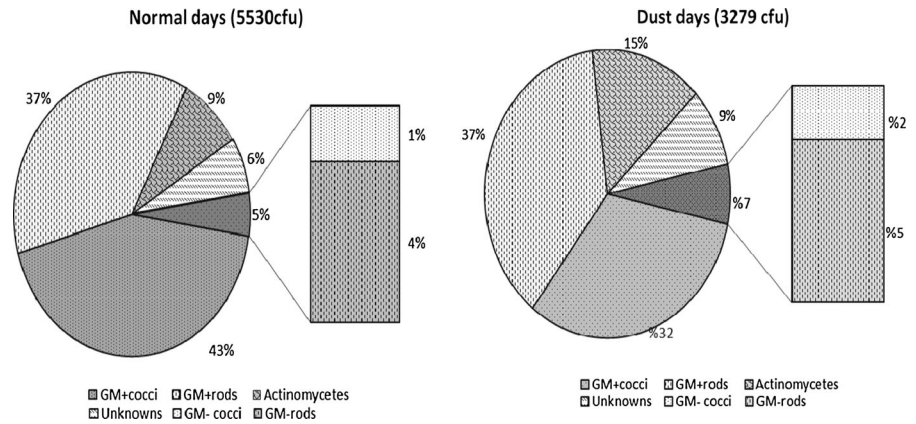


There were significant differences in concentration of *Streptomyces* spp. and *Bacillus* spp. between normal and dust event days ( $p < 0.001$ ) and also significant differences in concentration of *Micrococcus*, *Microbacterium*, *Stomatococcus* and *Corynebacterium* genera between normal and dust event days were observed ( $p = 0.03$ ). Figure 5 shows comparison of bacteria concentration in three sampling points during normal and dust event days with National Environmental Agency (NEA) guidelines. More than 58 % of total number of samples at three sampling

points during normal and dust event days were higher than the guidelines ( $500 \text{ cfu/m}^3$ ). The most pollutant days were September 28, 2010, December 14, 2010, March 10 and 11 2011.

The numbers of identified colonies in the normal and dusty days are presented in Fig. 6. Results indicated that Gram-positive bacteria had the highest concentration in both normal and dusty days. Gram-positive cocci with 2,378 cfu (43 %) and Gram-positive rods with 2,046 cfu (37 %) had the highest concentration in normal days. There were 1,213 cfu

**Fig. 6** Composition of culturable airborne bacteria during normal and dust event days at indoor air



**Fig. 7** Backward trajectories of dust storms (a September 28, 2010, b March 11, 2011)

(37 %) and 1,049 cfu (32 %) during dust event days for Gram-positive rod and Gram-positive cocci, respectively.

### 3.4 The relation between concentration of bacteria and environmental factors

As it is shown in Table 2, the overall mean of temperature and relative humidity in summer during normal and dust event days were 29.0, 28.2 °C and 33.5, 36.2 %, respectively. In the case of temperature, there was a significant difference between selected seasons during normal days ( $p < 0.001$ ). The same significant difference was also found during dust event days ( $p < 0.05$ ).

Although there were significant differences in the amount of relative humidity between summer with autumn and winter ( $p = 0.001$  and  $p < 0.0001$ , respectively), there was not a significant difference for this parameter between autumn and winter ( $p > 0.05$ ) during normal days, and it was also the

same during dust event days ( $p > 0.05$ ). In normal situation, there were low but significant linear correlations between relative humidity and the concentrations of *Streptomyces* spp., *Micrococcus* spp. and *Corynebacterium* spp. ( $p = 0.001$  and  $r_{\text{spearman}} = -0.27$ ,  $p = 0.002$  and  $r_{\text{spearman}} = 0.24$ ,  $p = 0.021$  and  $r_{\text{spearman}} = 0.19$ ). In dust event days, there were linear correlations between the total concentrations of bacteria and *Bacillus* spp. with temperature ( $p = 0.018$  and  $r_{\text{spearman}} = -0.37$ ,  $p = 0.004$  and  $r_{\text{spearman}} = -0.45$ ). Also there were moderate linear correlation between the concentration of *Streptomyces* spp. and *Bacillus cereus* with relative humidity ( $p = 0.002$  and  $r_{\text{spearman}} = -0.465$ ,  $p = 0.013$  and  $r_{\text{spearman}} = -0.39$ ).

### 3.5 Backward trajectory analyzing

According to previous studies, wind blows from west and south to east most of the time in Ahvaz city (Soleimani et al. 2013; Shahsavani et al. 2012b).

Therefore, dust had come to Ahvaz mostly from western and southern neighborhoods. Normal backward trajectories of two dust storms during this study are presented in Fig. 7. Backward trajectory in March 11, 2011, showed that dust storm was originated from Saudi Arabia (Fig. 7a). The total run time was 24 h at different altitude (500, 1,000 and 2,000 m). In Fig. 7b (September 28, 2010), the backward air mass trajectories of Ahvaz at the same altitudes within three days as total run time was simulated. As it has been shown, dust was transferred from North Africa particularly Egypt and Sudan to Ahvaz. Also a small circle of dust storm was formed in dried lands of Iraq which was depicted in red circle in Fig. 7b.

#### 4 Discussions

Particulate matter can transfer biological agent from outdoor to indoor environments. In present study, the investigation of airborne bacteria in indoor air environments of educational and therapeutic places that they did not have natural ventilation system and many people had activities in these places had done.

The concentrations of extracted bacteria in sampling points in normal situation were between 8 and 4,800 cfu/m<sup>3</sup> which were higher than other studies (Salonen et al. 2007; Naddafi et al. 2009). For example Kim et al. (2009), reported airborne bacteria concentrations have ranged from 256 to 716 cfu/m<sup>3</sup> in 10 hospitals, 206–408 cfu/m<sup>3</sup> in 10 elderly welfare facilities and 334–1,555 cfu/m<sup>3</sup> in 10 childcare centers. In Korea, Salonen et al. (2007) observed 14–1,550 cfu/m<sup>3</sup> as airborne bacteria in mold-damaged building. An average of 696 cfu/m<sup>3</sup> was also recorded by Naddafi et al. (2009) as airborne bacteria concentration in a children hospital in Tehran.

The most important result of this study was increasing in concentration of bacteria in indoor environments during dust event days. The main cause of this result might be dust storm occurrence during this study which impressed Ahvaz weather conditions.

There were significant differences in concentrations of bacteria between sampling points during normal and dust event days ( $p < 0.05$ ). The most levels of microbial load were related to dust storms in winter. The maximum residence time of dust storm in this study was 48 h which was related to March. In time of dust storms, the concentrations of particulate

matter in indoor and outdoor environments have been increasing. Although these places had natural ventilation and they were off in time of dust storms, the concentrations of bacteria in indoor environment also have been increasing. Based on other studies, each Gram of desert soil has been ranging from 0 to 10<sup>7</sup> colonies of bacteria (Maier et al. 2004), so the mean concentration of bacteria in time of dust storms in Saudi Arabia was  $1,892 \pm 325$  cfu/m<sup>3</sup> (Kwaasi et al. 1998), but the mean concentrations of bacteria in the present study for three stations were between 520.6 and 1,047.6 cfu/m<sup>3</sup>.

According to these results, in present study, the genera of bacteria which extracted in dust storms situation were like other studies in other parts of the world (Kwaasi et al. 1998; Lyles et al. 2005). Predominant genera of bacteria in dust storms were *Bacillus*, *Streptomyces*, *Micrococcus* and *Staphylococcus*. The concentration of *Streptomyces* spp. in March (dust event days) was higher than other months; also the concentration of *staphylococcus* spp. in December (during dust event days) was higher than other months. The concentrations of *Bacillus* spp. in all sampling seasons as well as all sampling points were high. Based on studies which had been conducted throughout the world, *Bacillus* (Kwaasi et al. 1998; Lyles et al. 2005; Prospero et al. 2005; Kellogg et al. 2004) and *Microbacterium* (Griffin et al. 2007; Kellogg et al. 2004) were predominant genera. *Bacillus* genus was observed in long distance from dust storm origin in comparison with other bacteria. Many bacteria are destroyed by solar radiation, drought and shortage of nutrient. The main cause for increasing of mentioned genera concentration during dust storms in comparison with other bacteria was their resistance due to spores (Kellogg and Griffin 2006). Frequency and types of microbial agent in air are varied, and they are related to suspended organic and inorganic matter, temperature, humidity, vegetation, UV incoming, precipitation, etc.

Totally, concentration of *Bacillus*, *Nocardia*, *Corynebacterium* and *Stomatococcus* in all three sampling sites were higher in dust event days than normal days indicating impact of dust storms on increased bacterial concentration in indoor environment. We also observed incremental concentration of total and unknown bacteria during dust storms compared to normal days. According to studies, many of these bacteria were extracted from Saudi Arabia and

Turkey's dust storms (Kwaasi et al. 1998; Griffin et al. 2007).

There is no doubt that temperature and humidity play an important role in good indoor air quality. Moon et al. (2014) reported that concentration of culturable indoor airborne bacteria was significantly higher in summer than in winter. Statistically, they reported a significant association between higher culturable indoor airborne bacteria with increases in both relative humidity and temperature. In terms of temperature or season and its relation to bacteria concentration, our findings did not comply with results of other researcher and scientists around the world. We found that concentration of bacteria were higher in autumn and winter than in summer. It was supposed that the fluctuation of bacteria concentration and its relationships with humidity and temperature in the present study should be similar to other investigations. Apparently, dust was a confounding agent, in which the balance of indoor air quality was disrupted especially during two peaks in winter time (Fig. 5), so that a high load of bacterial population was injected into the indoor by such peaks. There was another peak in autumn (Fig. 5) and that was why bacteria concentration in autumn was higher than summer. In some studies (Jo and Seo 2005; Chan et al. 2009), the concentration of bacteria were higher in summer and autumn compared to the other seasons. It should be noted that elevated relative humidity particularly in winter can promote the growth of mold, bacteria as well as fungi. Moon et al. (2014) also found a correlation between total and Gram-positive bacteria concentration with indoor humidity. In present study, predominant extracted bacteria in both normal and dust event days were Gram positive. Based on a study which had been done in Iran (Massoum et al. 1998) and also around the world (Chan et al. 2009; Jo and Seo 2005; Hwang et al. 2010), the concentrations of Gram-positive bacteria were more than Gram-negative bacteria. Air dryness is a cause of bacteria death. Bacterial active-growing cells are sensitive to dryness and are dying soon. Gram-negative bacteria are much more sensitive than Gram-positive bacteria. Meantime Gram-positive bacteria have thick peptidoglycan large cell wall so that they are resistant to dryness or undesirable situation, and this is why their concentration is high in extracted bacteria from air (Kellogg and Griffin 2006; Chan et al. 2009).

Bioaerosols such as viruses, bacteria, fungi and pollen can be pathogenic for human, animals and plants (Schlesinger et al. 2006). Inhalable bioaerosols with diameter equal or less than 10  $\mu\text{m}$  can have adverse effects on human health. With respect to the results of this study, different genera of bacteria which were extracted from air can have undesirable effects on human health. The concentrations of *Bacillus* spp. and *Streptomyces* spp. in time of dust storms were so high. Bear in mind that in dominant infections caused by different genres of bacteria, the path that they enter the body is through respiratory ways (Leger et al. 2009; Mehta et al. 2011; Mcneil and Brown 1994). Therefore, hospital admissions for respiratory and cardiovascular diseases attributed to particles were increased during dust storm (Tam et al. 2012a, b). Other extracted bacteria genera were *Micrococcus*, *Stomatococcus*, *Corynebacterium*, *Arthobacter*, *Staphylococcus*, *Nocardia*, *Gram-positive cocci* and *Stomatococcus* which may cause infection in patients with immune deficiencies and patients with bronchiectasis (Yuan et al. 2013).

Although *Actinomycetes* in normal situation hardly cause disorders in human and animals, *Streptomyces* and different species of *Nocardia* can cause large types of diseases such as pulmonary, eye, skin and wound infections. They may cause some fatal infectious diseases such as acute pulmonary infections, septicemia and brain abscess in patients suffering from HIV and cancers (Hua et al. 2007; Leger et al. 2009; Mcneil and Brown 1994). By increasing in prevalence of immune system deficiencies in human societies, the role of these bacteria becomes so important (Yildis and Doganay 2006; Brust et al. 2004; Verma et al. 2006). *Actinomycetes* have large variety which extracted are from east of Asia and in some cases from South Korea (Hua et al. 2007).

In HYSPLIT model, the dispersion of dust storms with a specified source may be calculated by considering either puff or particle dispersion. In the puff module, dust plume extend in three directions ( $x$ - $y$ - $z$ ), in which the standard deviation for  $x$  and  $y$  directions is the same, while the behavior of plume in  $z$  direction is absolutely different from downwind and crosswind of puff. In the particle module of HYSPLIT, that is very convenient to simulate dust storm dispersion into the atmosphere, a certain number of particulates are advected about the model domain by the mean wind field and spread by a turbulent component. In the

present study, based on westerly wind (Fig. 1), we used particle dispersion in a three-dimensional particulate distribution (horizontal and vertical). Trajectories of model in Fig. 7 showed that dust storms are originated mostly from Africa and southwestern neighborhoods of Iran where Khuzestan province is located. Longer run time of model revealed north of Africa as the substantial source of dust storm due to massive area of desert.

## 5 Conclusions

Present study showed that the concentrations of bioaerosols particles and soil-originated bacteria in indoor environments of educational centers particularly school and university increased during dust storms, although these places were off in time of this phenomenon. Clearly, outdoor air has an impact on indoor air quality. We can therefore conclude that during dust storm occurrence, bacteria concentration in indoor air was higher than normal days. Individual activities and age of buildings can be added to parameters which had impact on the concentration of bacteria in these places. Our study showed that culturable airborne bacteria concentrations in 58 % of samples were higher than the standard (500 cfu/m<sup>3</sup>). It means that indoor air quality in view of bacteria was not acceptable so that preventive measures such as artificial ventilation and indoor air cleaners particularly for hospital were recommended.

The HYSPLIT model showed that there were different locations as origin of dust during dust event days. According to this model, dust can be mounted from Africa, dried lands in Iraq as well as Saudi Arabia desert. It can be transferred from Iraq, Saudi Arabia, Syria, Turkey and Jordan and rarely from high latitudes. Therefore, population of airborne bacteria and its diversity can be changed from time to time.

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