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INDOOR MICROBIAL CONTAMINATION THROUGH WATER MIST AEROSOL AT PUBLIC RESTAURANTS

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Abstract

In Malaysia and many other Asian countries, countless number of restaurants are made of open or semi-open air buildings. These restaurants are commonly located beside roads, factories, and construction sites. The foods are often exposed to the air, increasing their tendency to be contaminated due to poor indoor and outdoor air quality. There are very few studies characterising and comparing the levels of indoor air pollutants in restaurants. Other than that, scarce data are available on dining establishments especially in the presence and absence of water mist application system. Due to these reasons, no best practices or guidelines can be developed. Hence, this present study aimed (1) to assess and compare the physical indoor air quality (IAQ) characteristics and airborne pollutants between different types of restaurant settings; (2) to identify microbes isolated in the presence and absence of water mist system; (3) to analyse bacterial counting within and between the different restaurant settings; and (4) to determine the relationship between physical IAQ characteristics and airborne microbial contaminants. Instruments known as Dustmate and VelociCalc® Multi-Function Ventilation Meter 9565 were used to measure the physical IAQ characteristics and airborne particulate matters. On the other hand, Surface Air System Indoor Air Quality (SAS IAQ) was use to seize the microbial contaminants. All the data obtained were compared with the standard reference known as the Industrial Code of Practice on Indoor Air Quality (2010) constructed by the Department of Occupational Safety and Health (DOSH). This study later indicated that the level of indoor PM_{10} concentrations was influenced by changes of physical IAQ parameters at the two restaurants investigated. As the PM_{10} increased, the colony forming unit (CFU) counting also increased. Although microbial contaminations were found during both periods of exposure (i.e., in the presence and absence of water mist) at both restaurants, significant relationship between the parameters measured cannot be determined. The bacterial species obtained during the presence and absence of water mist application system for both of the restaurants were also identified. *Gemella morbillorum* was found as the most dominant species, followed by other species such as *Aerococcus viridans*, *Globicatella sanguinis*, *Leuconostoc* spp., and *Proteus penneri*.

Keywords: Indoor air quality (IAQ), restaurants, water mist aerosols, airborne microbes and PM_{10}

1.0 INTRODUCTION

In Malaysia, restaurants and cafeterias can be found near the roads, adjacent to construction sites, and close to the industrial areas. The foods served at the open and semi-open restaurants are exposed to the indoor or outdoor air. Without a proper handling and because of the location of the restaurants, these foods may be contaminated easily [1]. Due to the locations of the restaurants, the dining areas are usually hot, and water mist fan is used to cool down the temperature inside the restaurants. The water mist fan is also installed to maintain and elevate the humidity in an open-compound restaurant [2].

Previous study reported that the misting fan can reduce the temperature and increase the relative humidity [3]. However, this water mist fan resulted in a higher number of bacteria from the mist produced. The study found that the number of airborne bacteria and fungi was higher when the water mist fan was used in the restaurants.

There is a limited number of studies in Malaysia examining the association between the presence of water mist on restaurant environment and microbial contamination due to the water mist fan in food that are being served. In fact, the number of studies on indoor air quality (IAQ) particularly in dining establishments is also inadequate in this country, thus no regulations, guidelines, or best practices can be developed. Therefore, the Industrial Code of Practice (ICOP) 2010 established by DOSH was used as a standard in this study.

2.0 EXPERIMENTAL

2.1 Study Sites

2.1.1 Restaurant #1 (X): Open-air Restaurant Near a Road

This restaurant chain or locally known as a Kopitiam had an indoor and open-air dining area, and the open air section was located about 10 m away from a one-way road. The floor was made of woods, and the restaurant was open 24 h daily. The kitchen was separated from the dining area by a wall. The foods were served on a countertop, and the style of cooking was deep frying. A filtered water mist system and four ceiling fans were installed at the open-air dining area.

2.1.2 Restaurant #2 (Y): Open-air Restaurant Beside a Road

This food-joint restaurant had an indoor and open-air dining area, and the open-air section was located about 5 m away from the traffic road. The floor was made of cement, and the restaurant was open 24 h daily. The kitchen was openly connected to the dining area, and the foods were cooked using open-wok frying and deep-frying styles. A waste mist system was installed at the open-air dining area besides the four ceiling fans available.

2.2 Measurement of Physical IAQ Parameters and Airborne Pollutants

Dustmate from Turnkey Instruments, UK was used to determine the concentration of airborne particles and dust in this study. Light scattering is the technique used by Dustmate to determine the concentration of airborne particles and dust of the diameter ranging from about 0.4 µm to about 20 µm. It works by continuously drawing the air sample into the instrument by a pump with a flow rate set by the microprocessor at 10 cc/ sec (600 cc/min).

The physical parameters of both locations were measured using a specialised instrument called the VelociCalc® Multi-Function Ventilation Meter 9565 by TSI. This 9565 model comes with a probe that has the ability to detect various types of physical IAQ parameters including air flow rate, CO_2 concentration, air velocity, temperature, relative humidity, and CO concentration.

Readings for both instruments were taken at an interval of 1 min for 15 min sampling time at both restaurants.

2.2.1 Monitoring and Sampling of Microbes

Surface Air System (SAS) IAQ by PBI International is considered the international standard for portable air microbiological samplers. This instrument is suitable for the IAQ study and was used in this study to perform microbiological evaluation of the air at both sampling locations. The sampling time for this instrument is 60 s.

Preparation of petri dishes with nutrient agar medium was done beforehand. The drilled head of the SAS IAQ was first unscrewed, and a plate containing medium was placed into the housing. The nominal air flow was later set to 1.5 L of air per min before the environmental air was aspirated over the agar surface of the plate. Airborne particles were captured on the agar by impaction. Then, the plate was removed and incubated at 37 °C for 24 h.

Besides, depending on SAS IAQ method, gravitational air sampling method was also used to perform microbiological evaluation of air at the sampling locations. In this study, the nutrient agar (NA) plates were placed at the same level as SAS IAQ in order to assess if there was a correlation between the results of the two sampling methods. The number of CFU was calculated after 24 h incubation of plates of both methods.

All the three instruments namely Dustmate, VelociCalc® Multi-Function Ventilation Meter 9565, and SAS IAQ were stationed at the human breathing zone approximately 1.5 m from the floor. The samples were collected in duplicates during two time periods, morning and afternoon, at the indoor dining environment. Air from each dining area was sampled concurrently with both SAS IAQ and gravitational methods in the presence and absence of the water mist.

2.2.2 Bacterial Identification Using API System

Analytical Profile Index (API) strips are commercially prepared identification system of bacteria. They are readily available and used in conjunction with extensive databases that characterise biochemical reactions of microorganisms. On one hand, API 20E System is a standardised, miniaturised microtube system consisting of 21 conventional biochemical tests designed for the identification of *Enterobacteriaceae* and other nonfastidious Gram-negative bacteria. Some complementary tests needed to be conducted upon identifying the bacteria such as oxidase test, oxidation fermentation test, motility test, and MacConkey test. On the other hand, API Strep system is used to identify Gram-positive bacteria. This system is very significant in the medical bacteriology. It is able to identify most of the streptococci bacteria that are mostly pathogens. For this study, the bacterial identification procedure was by using the API system as in the manufacturer's manual.

3.0 RESULTS AND DISCUSSION

3.1 Air Quality Assessment

Table 1 The comparison on daily average of physical IAQ parameters and airborne pollutants between the two restaurants with standard reference ICOP 2010

Parameters		Acceptable limit set by DOSH	Restaurants					
			With water mist			Without water mist		
			X	Y	P Value	X	Y	P Value
Physical Indoor Air Quality	Temperature (°C)	23–26	**31.95 ± 2.90**	**32.05 ± 0.21**	0.97	**33.75 ± 1.06**	**33.70 ± 0.28**	0.97
	Velocity (m/s)	0.15-0.50	0.26 ± 0.01	**0.80 ± 0.18**	0.16	0.27 ± 0.13	**0.64 ± 0.16**	0.03
Airborne Pollutants	Inhalable (mg/m³)	NA	1.58 ± 1.22	1.12 ± 0.23	0.63	0.12 ± 0.06	0.52 ± 0.46	0.40
	Thoracic (mg/m³)	NA	1.32 ± 1.05	0.66 ± 0.17	0.49	0.10 ± 0.06	0.36 ± 0.26	0.31
	Respirable (mg/m³)	0.15	**0.27 ± 0.08**	**0.26 ± 0.04**	0.89	0.06 ± 0.05	**0.18 ± 0.08**	0.11
	CO_2 (ppm)	1000	395 ± 0.00	402 ± 1.41	0.09	401.50 ± 20.51	404.50 ± 26.16	0.94
	CO (ppm)	10	1.10 ± 0.28	1.25 ± 0.35	0.21	0.85 ± 0.21	1.30 ± 0.99	0.56
CFU (CFU/m³)	SAS IAQ	500	446.67 ± 188.56	**1133.34 ± 377.12**	0.13	**933.33 ± 0.00**	**1133.33 ± 565.69**	0.71
	Gravitational	NA	66.67 ± 0.00	166.67 ± 47.14	0.21	33.34 ± 47.14	200 ± 282.84	0.61

significant at $p<0.05$ ****Bold* *readings indicate the values exceeding the range established by DOSH.*

3.2 Air Quality Assessment based on Physical IAQ Parameters Monitoring

Temperatures at the investigated restaurants, X and Y (in the presence and absence of water mist application), were found to exceed the standards established by the DOSH, as shown in Table 1. Both restaurants showed slightly higher temperatures in the absence of water mist environment (33.75 °C for restaurant X and 33.70 °C for restaurant Y). These figures are believed to be influenced by the locations of the open-air dining eating compounds situated near or beside the busy roads. Furthermore, in agreement with the previous study by the American Society for Heating, Refrigeration, and Air Conditioning Engineers (ASHRAE), this present study also found that the average temperature in the restaurants did not meet the guidelines by ASHRAE [4].

This study found higher values of air velocity at the restaurant Y, which exceeded the standards established by DOSH. As suggested by a previous study, air-conditioned buildings should have lower pollutant levels as compared to naturally ventilated buildings [5]. Nevertheless, the naturally ventilated buildings do not hold external air pollutants for long periods due to their high air-change rates.

3.3 Air Quality Assessment based on Airborne Pollutants Monitoring

Airborne pollutants monitoring was also conducted in this study to determine the criteria pollutants such as particulate matters and carbon monoxide (CO). These pollutants are crucial indicators for both indoor and outdoor air quality, following their detrimental effects on human health [1]. This study measured four types of particulate matters namely inhalable, thoracic, respirable, and ultrafine particulates. However, no standards are available to assess air quality for three of the particulate matters namely inhalable, thoracic, and ultrafine particulates except using the standard as established by DOSH in ICOP 2010. Hence, this paper only discusses the respirable particulate matters.

Respirable particulate matters can also be recognised as indoors PM_{10}, consisting of particles with a diameter of 2.5–10 µm [6]. Most of the studies on particulate matters in houses and restaurants have taken indoors PM_{10} or $PM_{2.5}$ into consideration [7, 8, 9]. Based on the airborne pollutants monitored during the presence of water mist, the concentrations of total respirable particulates within both restaurant X and Y were found to be 0.27 and 0.26 mg/m^3, respectively. These concentrations, however, were much higher than the permitted value specified in the ICOP 2010, i.e., 0.15 mg/m^3.

In the absence of water mist, the concentration of total respirable particulate for restaurant X was 0.06 mg/m^3, which was lower than the permitted value. However, for restaurant Y, the concentration of respirable particulate was 0.18 mg/m^3, which slightly exceeded the acceptable limit. See Table 1. Therefore, in general, these results show that whether in the presence or in the absence of water mist, the concentrations of respirable particulate at the restaurants were high, exceeding the standard given in ICOP 2010. The heating and cooking activities [10] at the restaurants probably contributed to the high concentration of total respirable particles, particularly PM_{10} and $PM_{2.5}$ [7].

Although not a criteria pollutant, airborne microorganisms or bioaerosols are also considered an important air quality parameter [11] especially in the restaurants. Airborne microorganisms or bioaerosols may originate from occupants, organic waste, and microbial growth and have the tendency to increase the risk of food contamination [12]. This present study found that most of the CFUs for the airborne bacteria collected at both restaurants (during the usage of SAS IAQ and gravitational methods) were higher than the standard (500 CFU/m^3). Only the CFU of bacteria during the application of water mist at the restaurant X was observed to be within the acceptable limit, 446.67 CFU/m^3.

The significantly high figures obtained were expected as the restaurants were consistently busy and occupied during the periods of investigation. Moreover, distributions of bioaerosols very much depended on the human activities at the restaurants [13]. These activities could cause the dust particulate to suspend and form airborne microorganisms. Apart from that, this study discovered that the SAS IAQ accumulated greater amount of bacteria than gravitational method at both restaurants. This trend was in line with the previous study where active method (SAS IAQ) was more preferable than gravitational method to be used in obtaining viable particles [14]. Since there was no standard as a reference for gravitational method, only CFU counting during the usage of SAS IAQ is discussed next.

In relation to the parameters measured, the total colony count was likely to increase with the increment of PM_{10} due to the large surface area of course particles aiding the microorganisms' attachment [1] plus that water in the mist is an essential reservoir for the growth of bacteria [15]. Nevertheless, evaporative cooling such as the water mist fan can lead to improved comfort [3] other that reservoir for bacterial growth.

3.5 Bacterial Identification

Table 2 Assessment of Gram positive and Gram negative bacteria by using API System

API database code	Strain	Total group of colonies	Percentage (%)	Note	Gram (+/−)	Location(s)	Water mist application
4140014	*Aerococcus viridans 2*	4	96.7	Good identification	+	X	×
5002053	*Aerococcus viridans 1*	3	92.9	Good identification	+	Y	✓ and ×
0142013	*Gemella morbillorum*	9	92.6	Good identification	+	X and Y	✓ and ×
4016753	*Globicatella sanguinis*	3	94.7	Low discrimination	+	X and Y	✓ and ×
5300000	*Leuconostoc spp*	4	79.7	Low discrimination	+	Y	✓
5200051	*Leuconostoc spp*	3	86.2	Acceptable identification	+	X	✓
001402043	*Proteus penneri*	2	83.9	Doubtful profile	−	X	×

✓ *- In the presence of water mist* × *- In the absence of water mist*

Further microbiological analyses showed that Gram-positive bacteria were more abundant compared to Gram-negative bacteria. Gram-positive cocci (micrococci, streptococci, staphylococci, and diplococci) were the dominant microbial morphologies, followed by Gram-positive bacilli and pathogenic Gram-negative enterobacteriaceae. These trends were relevant with other studies that reported the domination of Gram-positive bacteria in both indoor and outdoor environments [16]. Dispersion and spreading of these microorganisms are through direct skin-to-skin contact and are dismissed via respiration [17]. The existence of cocci in the air of the restaurants indicates overcrowding affiliated with insufficient ventilation [18]. As reported in the previous study [16], Gram-positive bacilli are often linked with outdoor sources such as soil emissions, water, dust, air, faeces, vegetation, wounds, and abscesses. Although most of these species are considered harmless to human health, infections can still be caused by certain species especially to immunocompromised individuals [19]. Enterobacteriaceae is related to *E. coli* and *Salmonella*, hence directly associated with food poisoning [20]. Possible origins of these species include inappropriate handling, serving, and preparing of food. On top of that, the presence of these pathogens in restaurants is likely to give a rise to a risk towards human health.

4.0 CONCLUSION

Two typical open-air restaurants were surveyed and the concentrations for relative humidity, CO_2, and CO were observed to abide the standards set by the DOSH, while most of the rest significantly exceeded the acceptable limits. Other parameters like temperature, velocity, and flow rate had correlations with the level of indoor PM_{10} concentrations. As PM_{10} increased, CFU also increased. There are pros and cons of utilising water mist application. Although microbial contaminations were found during both periods of exposure (in the presence and absence of water mist) at both restaurants, the significant relationship between these parameters cannot be established due to the limitation of time during the samplings between the presence and absence of water mist application. Furthermore, the bacterial communities found at both restaurants were *Aerococcus viridans 1* and *2*, *Gemella morbillorum*, *Globicatella sanguinis*, *Leuconostoc* spp., and *Proteus penneri*. It was found that the bacterial distributions were influenced by the changes of the physical IAQ parameters at the investigated restaurants. Hence, there was a significant relationship between physical characteristics, airborne pollutants, and microbial contaminants between different types of restaurants settings.

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References

[1] Yusup, Y., Ahmad, M. A., and Ismail, N. 2014. Indoor Air Quality of Typical Malaysian Open-Air Restaurants. *Environment and Pollution*. 3(4): 10-23.

[2] Raihan, S. M., Ullah, M. H., Muhida, R. 2011. A Prototype Design to Maintain Temperature and Humidity in an Open Compound Restaurant. *European Journal of Scientific Research*. 63(2): 164-171.

[3] Wong, N. H. and Chong, A. Z. M. 2010. Performance Evaluation of Misting Fans in Hot and Humid Climate. *Building and Environment*. 45(12): 2666-2678.

[4] ASHRAE [American Society for Heating, Refrigerating and Air-Conditioning Engineers]. ASHRAE Standard [ANSI/ASHRAE Standard 62.1 -2007]: Ventilation for Acceptable Indoor Air Quality in Low-Rise Residential Buildings. ISSN 1041 -2336. ASHRAE, Inc. U.S. 2007.

[5] Kukadia, V., and Palmer, J. 1998. The Effect of External Atmospheric Pollution on Indoor Air Quality: A Pilot Study. *Energy and Buildings*. 27(3): 223-230.

[6] Wallace, L. 1996. Indoor Particles: A Review. *Journal of the Air and Waste Management Association.* 46(2): 98-126.

[7] Lee, S. C., Li, W.-M., and Yin Chan, L. 2001. Indoor Air Quality at Restaurants with Different Styles of Cooking in Metropolitan Hong Kong. *Science of the Total Environment*. 279(1-3): 181-193.

[8] Monkkonen, P., Pai, P., Maynard, A., Lehtinen, K. E. J., Hameri, K., and Rechkemmer, P. 2005. Fine Particle Number and Mass Concentration Measurements in Urban Indian Households. *Science of the Total Environment*. 347: 131-147.

[9] Fortmann, R., Kariher, P., and Clayton, R. 2001. Indoor Air Quality: Residential Cooking Exposure. *Report of State of California Air Resources Board*.

[10] Baek, S. O., Kim, Y. S., and Perry, R. 1997. Indoor Air Quality in Homes, Offices and Restaurants in Korean Urban Areas Indoor/Outdoor Relationships. *Atmospheric Environment*. 31(4): 529-544.

[11] Reanprayoon, P., and Yoonaiwong, W. 2012. Airborne Concentrations of Bacteria and Fungi in Thailand Border Market. *Aerobiologia*. 28(1): 49-60.

[12] Fabian, M. P., Miller, S. L., Reponen, T., and Hernandez, M. T. 2005. Ambient Bioaerosol Indices for Indoor Air Quality Assessments of Flood Reclamation. *Journal of Aerosol Science*. 36(5-6): 763-783.

[13] Rajasekar, A., and Balasubramaniam, R. 2011. Assessment of Airborne Bacteria and Fungi in Food Courts. *Buildings and Environmen.* 46(10): 2081-2087.

[14] Napoli, C., Marcotrigiano, V., and Montagna M. T. 2012. Air Sampling Procedures to Evaluate Microbial Contamination: A Comparison Between Active and Passive Methods in Operating Theatres. *BMC Public Health*. 12: 594.

[15] Tsao, Y. C., and Hwang, Y. H. 2013. Impact of a Water-Damaged Indoor Environment on Kindergarten Student Absences Due to Upper Respiratory Infection. *Building and Environment.* 64: 1-6.

[16] Aydogdu, H., Asan, A., and Tatman Otkun, M. 2010. Indoor and Outdoor Airborne Bacteria in Child Day-Care Centers in Edirne City (Turkey), Seasonal Distribution and Influence of Meteorological Factors. *Environmental Monitoring and Assessment*. 164(1): 53-66.

[17] Chikere, C. B., Omoni, V. T., and Chikere, B. O. 2008. Distribution of Potential Nosocomial Pathogens in a Hospital Environment. *African Journal of Biotechnology*. 7(20): 3535-3539.

[18] Awad, A. H. A. 2007. Airborne Dust, Bacteria, Actinomycetes and Fungi at a Flourmill. *Aerobiologia*. 23(1): 59-69.

[19] Mckernan, L. T., Wallingford, K. M., Hein, M. J., Burge, H., Rogers, C. A., and Herrick, R. 2008. Monitoring Microbial Populations on Wide-Body Commercial Passenger Aircraft. *Annals of Occupational Hygiene*. 52(2): 139-149.

[20] Northcutt, J. K., Jones, D. R., and Musgrove, M. T. 2004. Airborne Microorganisms during the Commercial Production and Processing of Japanese Quail. *Poultry Science*. 83(10): 1812-1812.