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Evaluation of Microbial Contamination of Floor Mats Installed on A University Campus

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ABSTRACT

Floor mats have excellent dust-absorbing properties and durability and are often installed at the entrance to various facilities to prevent contamination by soil, moisture, dust, and microorganisms. Carpets and floor mats are made from similar materials, and there are no clear differences between the products themselves; however, they are generally distinguished by their size and location. In a previous study, we confirmed that carpeted floors in university computer rooms are prone to dust collection and are highly contaminated. We also confirmed that microorganisms can enter from outside and that the number of users can increase. Floor mats are typically installed in localized areas, which may lead to a concentration of dirt and debris and higher levels of contamination compared to carpets. In addition, the difficulty of effectively removing dust through cleaning suggest that contamination levels are likely to increase over time.In this study, we surveyed floor mats installed on university campuses and examined the actual state of microbial contamination with general bacteria, fungi, *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli*, and coliform bacteria, considering factors such as the environment at the installation location and foot traffic.

Keywords: Floor mats, Microbial contamination, Foot traffic, University campus

1. INTRODUCTION

Floor mats are highly absorbent and durable and often installed at the entrances of various facilities to prevent contamination by soil, moisture, dust, and microorganisms. Factors that affect the effectiveness of blocking soil and moisture include weather conditions and foot traffic; however, floor mats have been suggested to be more effective in removing soil and moisture[1)]. Carpets and floor mats are made of similar materials, and there are no clear differences between these products; however, they are generally distinguished by their size and location[2)]. Floor mats are often small and are installed in specific areas. The surface material is complex, and the pile (made of yarn-woven loops) is structured to trap dust easily. Because of this complexity, it is difficult to completely remove the trapped dust by cleaning, and any remaining dust will likely accumulate within the pile[3)]. If dust that cannot be removed through repeated cleaning becomes noticeable, the mat should be replaced. However, carpets are often laid across an entire room. They are often installed to maintain room temperature and reduce noise. Although it is less effective than a floor mat in removing dust and other particles, it is easier to return it to the original clean state. They are usually replaced less frequently than floor mats.

In a previous study, we investigated the microbial contamination levels on the floors of university computer rooms and restrooms. The results confirmed that the carpeted floor of the information seminar room was more prone to dust accumulation and contamination than the tiled floor of the toilet[4)]. In another study, we investigated microbial contamination levels on carpeted floors and found that microorganisms can enter from the outside and increase with foot traffic[5)]. These studies investigated the microbial contamination of indoor carpeted floors; however, few studies have investigated floor mats installed outdoors or close to the outdoors. Furthermore, no previous studies have examined the microbial contamination levels of floor mats at educational institutions such as universities. Because floor mats are often installed locally, they may be more contaminated than carpets. The difficulty of removing dust through cleaning is also an indirect basis for the idea that contamination levels are likely to increase.

In this study, we conducted a survey of floor mats installed on university campuses and examined the state of microbial contamination with general bacteria, fungi, *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli*, and coliform bacteria, considering factors such as the environment at the installation location and foot traffic.

2. METHODS

2-1 Survey target overview

Table 1 provides an overview of the survey targets. In this experiment, a survey was conducted on floor mats installed near the entrances of Buildings A, B, C, and D, and in indoor corridors and facility entrances of K University. The floor mats surveyed were in the entrance on the first floor of Building A, the shop entrance on the first floor of Building B, cafeteria on the first floor of Building B, entrance on the first floor of Building B, under the stairs leading from the first floor to the basement of Building C, entrance on the first basement floor of Building C, staff-only entrance of Building D, and entrance of the corridor leading from Buildings B to D; each floor mat sampled is hereafter denoted as **I** to **VIII**. In addition, the susceptibility of the mats to the intrusion of dust and other particles from the outside was estimated by comparing them with information about the environment of the installation locations (e.g., whether they were near a door facing outside or wind direction).

Floor mats **I**, **III**, **IV**, and **VI** were Yamazaki Sangyo's (Osaka) CONDOR Lonstep absorbent mats; **V**, **VII**, and **VIII** were Teramoto (Osaka, Japan) Hyperon 900 × 1800 mats; and no description was given for mat **II**. Mats **I**, **II**, **III**, **IV**, **V**, and **VI** were purchased and installed in 1998, whereas mats **VII** and **VIII** were purchased and installed in 2011. Both the CONDOR and Hyperon mats are touted for their excellent dust absorption and durability, and both have nylon surfaces, but the former has an approximately 0.5 mm shorter pile length[6),7)].

The floor mats were cleaned once every 1–2 weeks, and in areas where they were visibly dirty, outdoor mats were dusted off, whereas indoor floor mats were vacuumed. Cleaning personnel are assigned to each building, and it is believed that the detailed cleaning methods and time required vary among personnel. Cleaning work was conducted from Monday to Friday, excluding public holidays, and continued during summer vacation.

Because it was difficult to determine the total foot traffic, we used the online university guide and interviews with administrative staff to survey the number of faculty members and students in each faculty for reference. As of May 1, 2024, 1,038 faculty members and students were enrolled on campus (Table 2). Building B contains a store, cafeteria, and library, and which are likely to be used by faculty members and students from all faculties. Therefore, we predict that Building B would have the highest foot traffic. Renovation work on the plaza in front of Building B was conducted from July 28th to the end of the study period, and only those involved in the construction work were allowed to pass over mat **IV**. August 7 to September 8 corresponded to the summer vacation period, thus, the expected student foot traffic was expected to be lower than usual. Based on this, we semi-quantitatively estimated the foot traffic (Table 1).

2-2 Equipment, devices, culture medium, and water used

The sterilized water used in the experiment was prepared using the Autostill WS200 (Yamato Scientific, Tokyo, Japan) pure water production system, RFU414BA (Advantech Toyo's, Tokyo, Japan) ultrapure water production system, and the ST201 laboratory automatic high-pressure steam sterilizer (Yamato Scientific). Microorganisms were collected using sterile cotton swabs (code 06526; Shimadzu Diagnostics, Tokyo, Japan). For ultrasonic treatment, a DG-1 ultrasonic cleaner (Iuchi Seieido, Osaka, Japan) with an oscillation frequency of 43 kHz was used. An incubator (FCI-280G; AS ONE, Osaka, Japan) was used for microbial culture. Aseptic procedures were performed using a biohazard cabinet (HME-S1300A2-PJ; Panasonic, Osaka, Japan). Temperature and humidity were measured using a CO_2 Manager (CO2MG-001; Toa Sangyo, Tokyo, Japan).

The Shimadzu Diagnostics' Compact Dry Simple Culture Media was used to measure bacterial counts, including those for general bacteria, yeast, mold (rapid type), *E. coli* and coliform counts, *S. aureus*, and *B. cereus*. After checking with the manufacturer, a fungal count medium from a yeast and mold count kit was used.

Pure water was obtained from the pure water production system installed at the university, and then purified using an ultrapure water production system to produce ultrapure water with a resistivity of ≥ 18 MΩ・cm, and sterilized in the high-pressure steam sterilizer at 121°C for 15 min to create sterilized water. Subsequently, using aseptic procedures in a biohazard cabinet, 7 mL of sterilized water was dispensed into sterilized 15 mL centrifuge tubes using a micropipette and used for measurement.

2-3 Sampling and measurement methods

The target floor mats were swabbed between 7:00 and 8:00 a.m. on July 22, August 13, September 2, September 23, and October 14, 2024, and microbial counts were measured as follows: the CO_2 Manager was placed on each floor mat to measure the temperature

and humidity at the time of swabbing. Each measurement point was sampled using a sterile cotton swab moistened with 7 mL of sterile water and placed in a centrifuge tube. During swabbing, a homemade cardboard frame was placed at the center of the floor mat and the area inside the frame was wiped such that swabbing covered an area of 30 × 30 cm. After wiping each floor mat, the frame was wiped with alcohol disinfectant and stored in a plastic bag to prevent contact with dust until its next use. Swabbing was performed in the vertical, horizontal, and diagonal directions for 1 min each (total of 3 min). To prevent bias in the results due to the order of measurements, samples were taken in the order of **I** → **VIII** on the first, third, and fifth sampling days, and **VIII** → **I** on the second and fourth sampling days (Table 1). Cotton swabs were carefully placed in the centrifuge tubes containing the remaining sterile water, while ensuring not to touch it with the bare hands (assuming that the amount of sterile water remained unchanged at 7 mL). Centrifuge tubes containing sterilized cotton swabs were sonicated for 5 min, and 1 mL of the resulting microbial suspension was homogenized by stirring using a touch mixer in the biohazard cabinet. One milliliter of the suspension was seeded onto a medium for cultivation. To minimize the contamination of components between media, they were seeded in the following order: *B. cereus* → *S. aureus* → *E. coli*/coliform bacteria → fungi → general bacteria.

We referred to the instruction manual for the culture conditions, and based on our preliminary tests, we set the incubation conditions at 35°C for two days for general bacteria, 25°C for three days for fungi, 35°C for four days for *E. coli*, coliforms, and *S. aureus*, and 25°C for four days for *B. cereus*. Microbial counts were measured visually as colony counts. To increase accuracy, two individuals observed both the front and back of each culture medium. The average of the two measurements was multiplied by seven to account for the volume of sterilized water and was used as the microbial count for each floor mat surface. We referred to the instruction manual for the colony colors used for measurement: red for general bacteria, all colors for fungi; blue to bluish-purple for *E. coli*; pink to reddish-purple for coliforms; light blue to blue for *S. aureus*; and blue to light blue for *B. cereus*.

In addition to the weather, the average temperature and humidity on the university campus during sampling days were recorded using meteorological data from Kochi City (data not shown). The mean temperature, humidity, and precipitation on and between sampling days were also recorded (data not shown). This was done to determine whether changes in the average temperature and humidity during the study period affected microorganism growth.

3. RESULTS AND DISCUSSION

3-1 Comparison based on mat location

Figure 1 shows the number of bacteria detected in each survey subject. General bacteria were detected in high numbers in mats **II** and **VI** throughout the study period. Mat **II** is generally used by students and staff members from all faculties and, therefore, has very high levels of foot traffic, whereas mat **VI** is at an entrance used by students and staff from the faculty with the highest enrollment numbers at K University. It is believed that when foot traffic is high, dirt and foreign objects from the soles of shoes are more likely to accumulate on floor mats compared to areas with lower foot traffic, which increases the level of microbial contamination on the floor mats and may have affected the detection results. General bacteria were not detected in large numbers on mats **III** and **VII**. Mat **III** was located in front of a completely closed and unused door, while mat **VII** is thought to be primarily used by office staff and visitors; therefore, it was assumed that the amount of foot traffic was lower here than in other locations, which may have affected the detection results.

Figure 2 shows the number of fungi detected in each area. Fungi were abundant in mats **I** and **VIII** throughout the study period. Soil with high organic matter content is a primary factor for fungal habitation[8]. Mat **I** was located outdoors, and we visually confirmed that a large amount of grass had fallen onto this floor mat on sampling days (data not shown). Mat **VIII** was located relatively close to the outdoors, and we visually confirmed that hair, grass, and dust had fallen onto the floor mat on sampling days (results not shown); therefore, these mats were likely contaminated by microorganisms that adhered to the dust particles and grass.

Figure 3 shows the number of *S. aureus* isolates detected in each area. *S. aureus* detected frequently on mats **II** and **VI** throughout the study period. *S. aureus* is a normal skin flora distributed in the nasal cavity, skin, and intestinal tract[9]. On mats **II** and **VI**, the incidence was thought to be primarily influenced by the number of people passing by, similar to that of general bacteria. As shown in Table 1, the foot traffic on mat **VIII** was predicted to be high but was not detected very often. This was likely due to the area having high foot traffic, and frequent opening and closing of doors, which creates strong air currents that might move microorganisms to other areas.

Figure 4 shows the number of *B. cereus* detected in each area. *B. cereus* was frequently detected on mats **I** and **VIII** throughout the study period. *B. cereus* is widely distributed in natural environments such as soil and plants[10]. Because mat **I** was installed outdoors and mat **VIII** was installed outside double doors, they may have been contaminated by soil, plant materials, and other outdoor

elements.

Figures 5 and 6 show the number of *E. coli* and coliform bacteria detected in each area. Both *E. coli* and coliform bacteria were highly abundant in the first sample collected on mat **V** and the third sample collected on mat **VI**. *E. coli* is present in the intestines of humans and animals and is considered an indicator of fecal contamination, whereas coliform bacteria include not only *E. coli* but also microorganisms commonly found in the environment, such as those present in plants, soil, and water[9]. Because they was detected at high concentrations in specific locations and only on certain sample collection days, their presence may not have been influenced by weather or the environment, but rather by the emergence of a temporary contamination source, such as human or animal fecal matter carried on shoe soles or clothing.

3-2 Comparison by sampling date

For each microorganism, we compared the fluctuations in microbial counts across the five sampling dates. The general bacterial count decreased from the first to second sampling periods and from the third to fourth sampling periods but increased significantly from the fourth to fifth sampling periods (Figure 1). Because the average humidity fluctuated on each measurement date and in the three weeks preceding sampling (data not shown), we speculate that humidity may have influenced the results. We also believe that the decrease from the third to fourth sampling periods and the increase from the fourth to fifth sampling periods may have been due to the influence of foot traffic and the individual circumstances of each passersby at the end of the summer vacation.

Fungi grow optimally at temperatures between 20°C and 30°C under high humidity[11], but in this experiment, no clear trend related to temperature or humidity was observed, suggesting that the fungi were influenced by factors other than temperature and humidity (Figure 2).

Although no clear trends in *S. aureus* counts were observed at each location, the rate of increase in counts from the fourth to fifth sampling periods was higher than that in the other periods (Figure 3). This increase was likely due to an increase in the number of people entering the university campus following the end of summer vacation.

The *B. cereus* count decreased from the second to third sampling periods and from the third to fourth sampling periods, along with temperature and humidity. However, no correlation with temperature or humidity was observed between the first and second sampling periods, or between the fourth and fifth sampling periods (Figure 4).

E. coli and coliform counts were likely affected by temperature and humidity, as both the temperature and humidity were higher during the first, second, and third sampling periods than in subsequent periods (Figures 5 and 6). Since *E. coli* is found in feces, and coliform bacteria include not only *E. coli* but also bacteria commonly found in the environment, they are thought to be influenced by foot traffic and the materials attached to each individual's footwear.

This study had one limitation. The number of floor mats surveyed was limited, making it difficult to compare the time of use and mat type under standardized conditions; therefore, the results cannot be generalized. Furthermore, more reliable data could be collected by conducting surveys that include newly purchased floor mats.

4. CONCLUSION

In this study, we evaluated the differences in the level of microbial contamination of floor mats installed at various locations within a university, based on factors such as the installation environment, foot traffic, and time since installation.

The results of this study suggest that bacteria and *S. aureus* are mainly affected by foot traffic, whereas fungi and *B. cereus* are affected by the installation environment. In this study, the increase or decrease in *E. coli* and coliform bacteria may have been the result of temporary contamination, as no clear trend in their counts were observed. When comparing the sampling dates, the percentage of microbial counts that increased was highest between the fourth and fifth sampling periods for general bacteria, *S. aureus*, and *B. cereus*. This may be due to the increase in the number of people entering the university campus as the summer vacation period ended. We speculate that *E. coli* and coliform bacteria may be affected by temperature and humidity; however, further studies are required to confirm this.

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Table 1 Overview of surveyed locations

Sampling location	Mat installation location				Susceptibility to external influences[*1]	Estimated Number of Passengers[*2]	Sampling order	
	Building	Floor	Location	Environment			July 22, September 2, October 14	August 13, September 23
I	A	1	Entrance	Outdoors	Susceptible	Medium	1	8
II	B	1	Shop entrance	Indoors	Not susceptible	Medium	2	7
III		1	Cafeteria entrance	Inside a single door	Susceptible	Low	3	6
IV		1	Main entrance	Inside a double door	Susceptible	Low[*3]	4	5
V	C	B1	Under the stairs of the gymnasium	Indoors (Hallway)	Not susceptible	Medium	5	4
VI		B1	Staff entrance	Inside a single door	Susceptible	High	6	3
VII	D	1	Staff entrance	Inside a single door	Susceptible	Low	7	2
VIII		2	Entrance to Building B	Outside a double door (Near outdoors)	Susceptible	High	8	1

[*1] Regarding susceptibility to external influences, floor mats installed outdoors or in places where dust and grass are likely to be blown in were classified as “Susceptible,” whereas those considered less exposed were classified as “Not susceptible.”

[*2] The foot traffic at each sampling location was categorized into three levels: low, medium, and high.

[*3] At the front entrance of Building B, the foot traffic is typically "High," but during this study period, renovation work was being carried out on the plaza in front of Building B, restricting access to people other than construction workers, thus, the area had low foot traffic levels.

Table 2 Number of staff members and students enrolled at K University

Number by Type	Building type		
	A	C	D
Undergraduate students	296	334	167
Graduate students	0	59	24
University faculty	22	51	13
Graduate faculty	4		
Office staff	0	0	68
General Information Center staff	1		
Health and Longevity Center staff	1		
Total	1038		

Based on the university website and interviews with administrative staff, the numbers of students, faculty, and staff as of May 1, 2024, are shown in the table.

Building B was not included in the table as it contains a shop, cafeteria, and library, but no faculty rooms.

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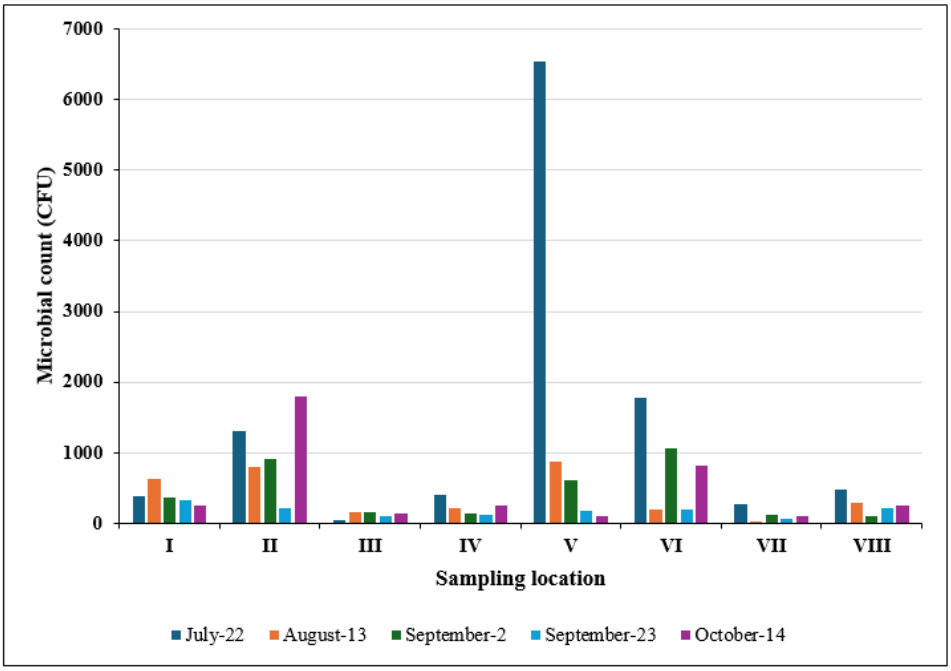

Figure 1 Number of general bacteria detected at each location

Numbers **I**–**VIII** represent floor mats at the various installation locations (shown in Table 1).

The experiment was conducted over the same area regardless of the floor mat size.

Each mat was swabbed the same number of times to measure the microbial count.

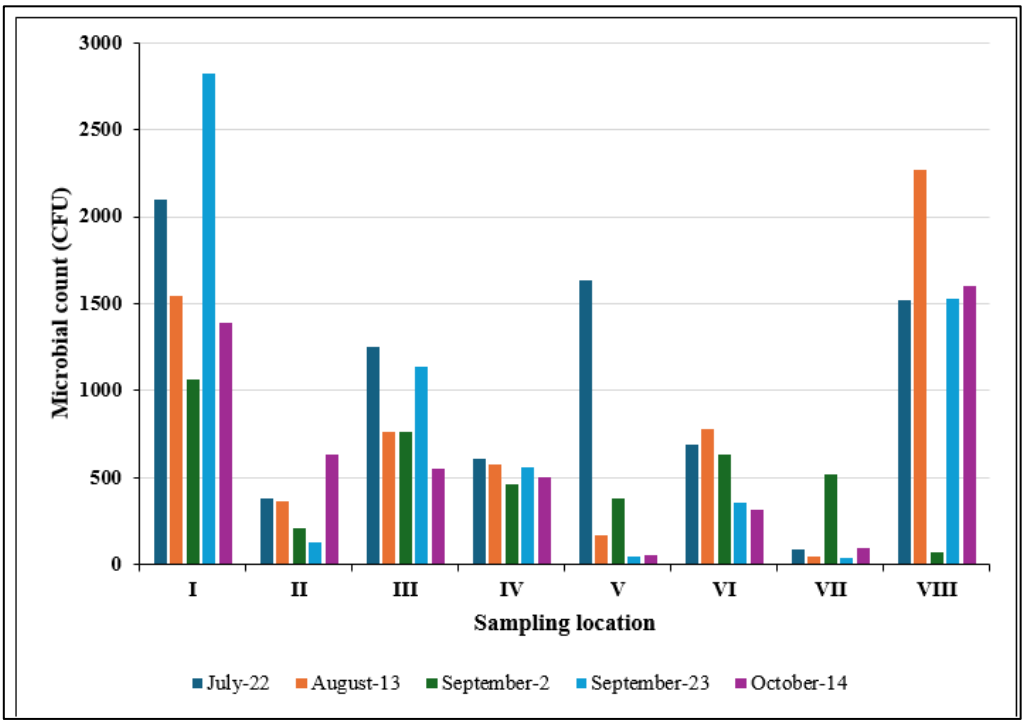

Figure 2 Number of fungi detected at each location

Numbers **I**–**VIII** represent floor mats at the various installation locations (shown in Table 1).

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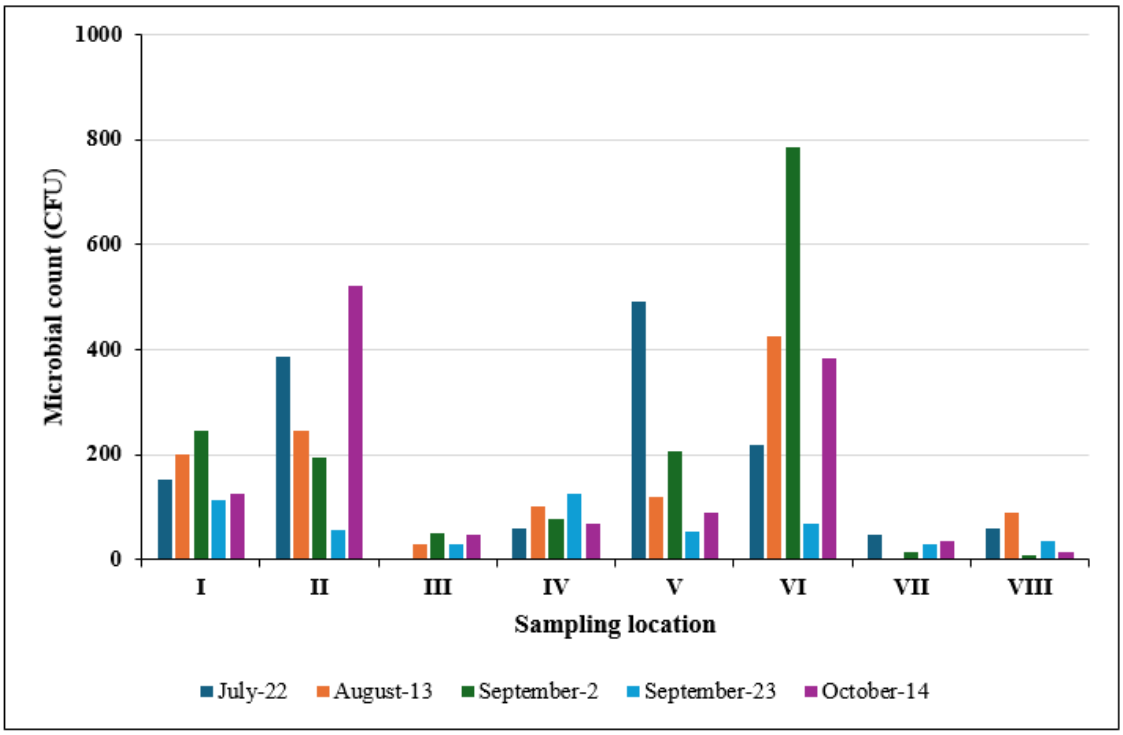

Figure 3 Number of *Staphylococcus aureus* detected at each location

Numbers **I–VIII** represent floor mats at the various installation locations (shown in Table 1).

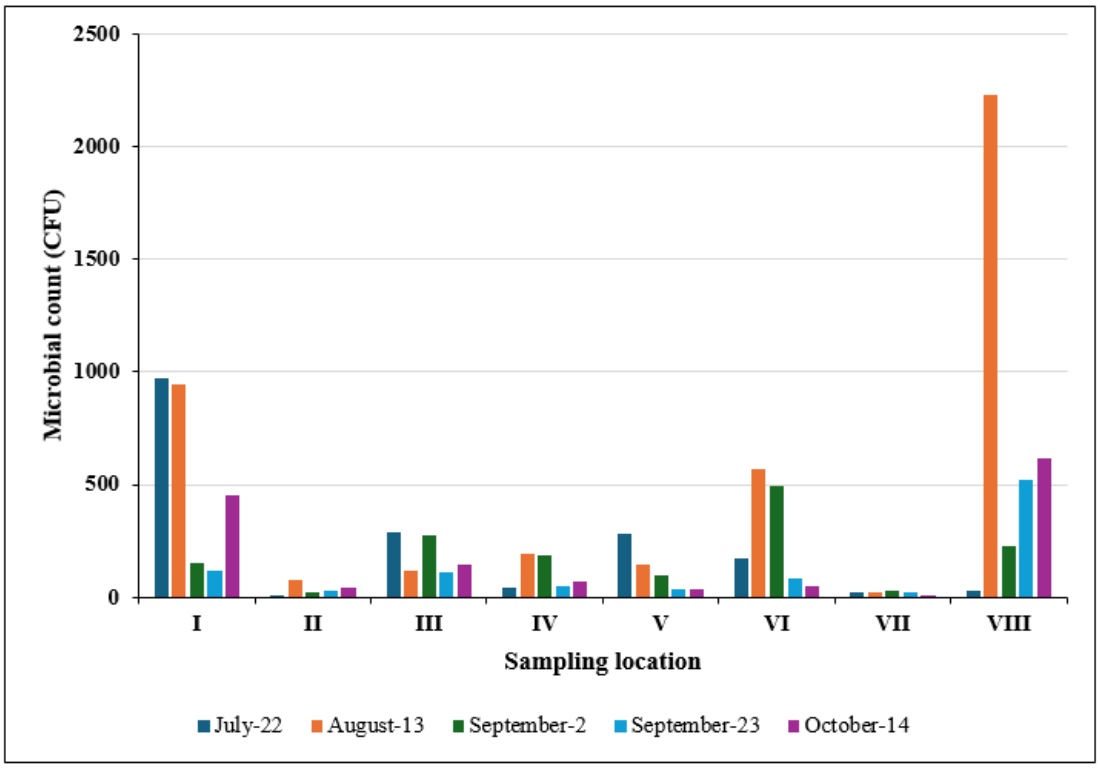

Figure 4 Number of *Bacillus cereus* detected at each location

Numbers **I–VIII** represent floor mats at the various installation locations (shown in Table 1).

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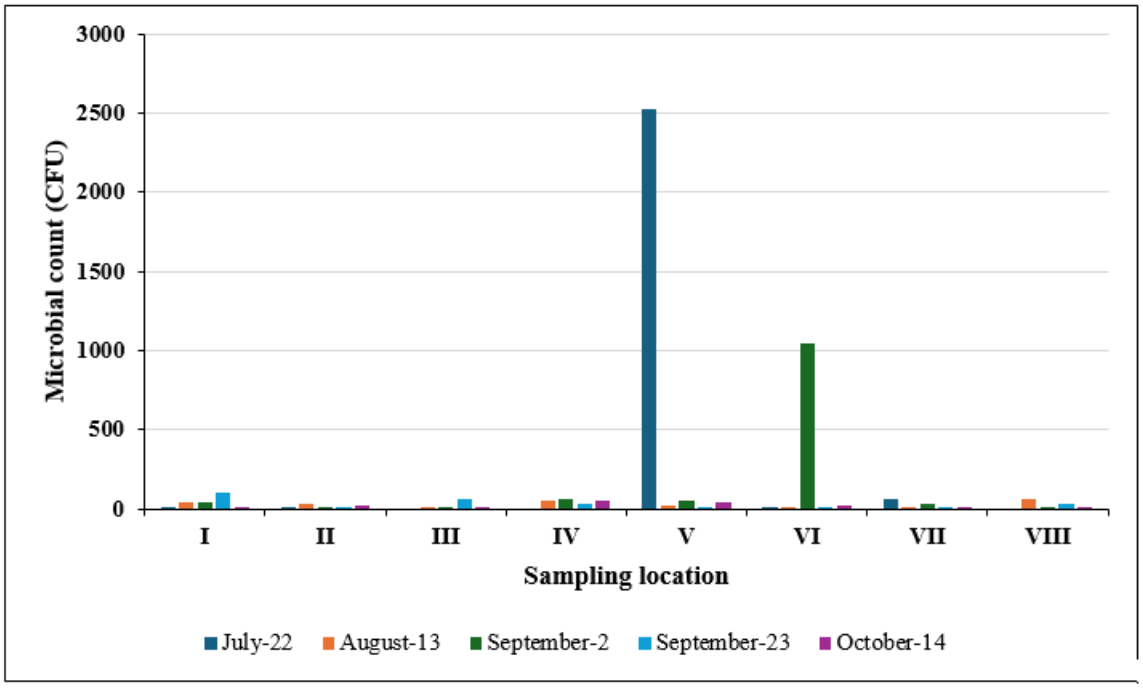

Figure 5 Number of *Escherichia coli* bacteria detected at each location

Numbers **I–VIII** represent floor mats at the various installation locations (shown in Table 1).

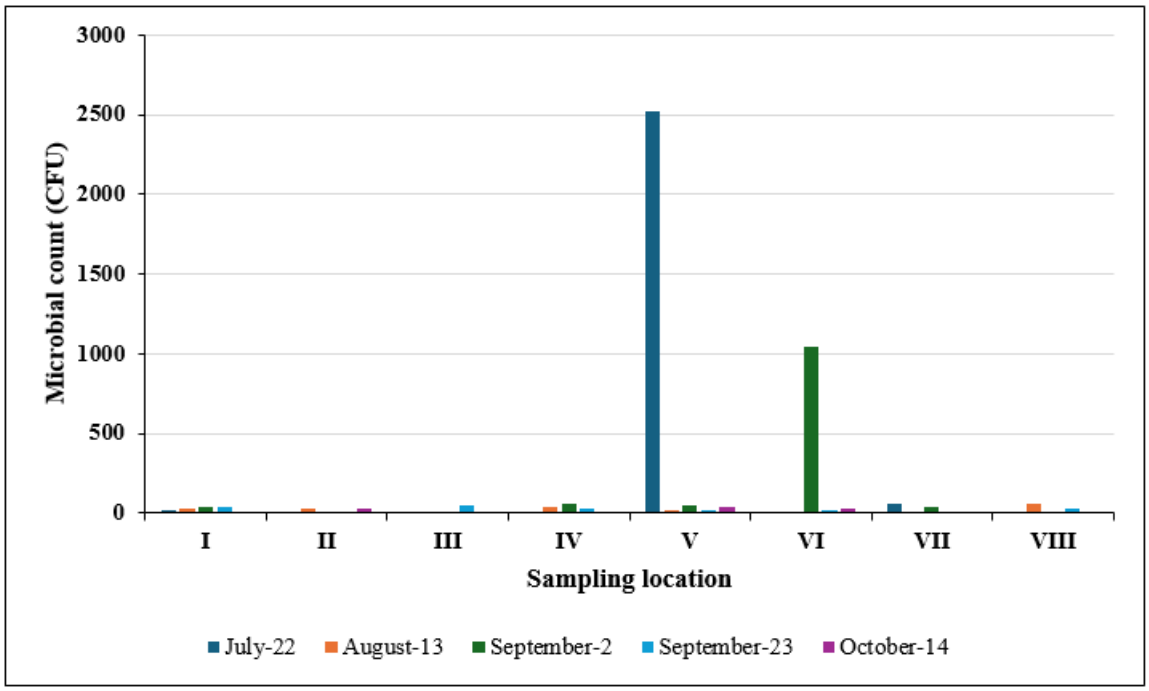

Figure 6 Number of coliform bacteria detected at each location

Numbers **I–VIII** represent floor mats at the various installation locations (shown in Table 1).

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