

# Assessing the Impact of Regular Environmental Monitoring on Cleanroom Maintenance and Microbial Control

Shalin S John<sup>1</sup>, Prejish C<sup>1</sup>, Jomol P J<sup>1</sup>, Arun K S<sup>1</sup>, Anakha Vijayan<sup>1</sup>, Bernaitis L<sup>2\*</sup>

<sup>1</sup>Microbiologist, CML Biotech Ltd, Angamaly, Ernakulam, Kerala, India – 683573.

<sup>2</sup>Head – R & D microbiology, CML Biotech Ltd, Angamaly, Ernakulam, Kerala, India – 683573.

Received: 17/08/2025 | Accepted: 27/09/2025 | Published: 21/10/2025

**Abstract:** Maintaining a clean and sterile environment in microbiological laboratories is critical for ensuring the validity and reliability of research outcomes. Traditional cleaning methods may not fully address the risk of contamination from airborne particles or human contact. This paper presents an integrated approach that combines routine fumigation protocols with rigorous personal hygiene screening to enhance cleanliness and minimize potential sources of contamination. Fumigation, using agents such as hydrogen peroxide or ozone, effectively reduces microbial burden in laboratory settings. Personal hygiene screening involves assessing individuals for compliance with laboratory attire protocols and conducting microbiological swab tests to detect microbial growth on hands, lab coats, and surfaces. The study demonstrates the effectiveness of these protocols in maintaining cleanliness over a three-month period, with no microbial growth detected in fumigated areas and minimal contamination found through personal hygiene checks. While these measures require financial resources and may disrupt workflow, their implementation offers significant benefits in reducing contamination risks and ensuring the safety of laboratory staff. Continuous evaluation and optimization of these integrated strategies are essential for maintaining high standards of cleanliness and minimizing the risk of experimental contamination.

**Keywords:** Microbiological laboratories, Cleanliness maintenance, Fumigation protocols, Personal hygiene screening, Contamination control, Laboratory safety.

## Cite this article:

John, S.S., Prejish, C., Jomol, P.J., Arun K.S., Vijayan, A., Bernaitis, L., (2025). Assessing the Impact of Regular Environmental Monitoring on Cleanroom Maintenance and Microbial Control. *World Journal of Applied Medical Sciences*, 2(10), 5-9.

## Introduction

In microbiological laboratories, particularly in cleanrooms, upholding stringent cleanliness standards is imperative to ensure the accuracy and reliability of research outcomes<sup>1</sup>. While traditional cleaning methods involving disinfectants and manual wiping are fundamental, they may not entirely eliminate the risk of contamination from airborne particles or human contact<sup>2</sup>. Integrating routine fumigation protocols alongside rigorous personal hygiene screening offers a comprehensive approach to bolster cleanroom maintenance and minimize potential sources of contamination<sup>3</sup>.

Fumigation, a technique employing gases to cleanse or purify a space by destroying germs, is commonly utilized in cleanrooms<sup>4,5</sup>. Agents such as hydrogen peroxide, ozone, or chlorine dioxide are frequently employed for this purpose<sup>6,7</sup>. Conversely, personal hygiene screening entails assessing the cleanliness of individuals entering or working in cleanrooms, encompassing visual inspections, gowning procedures, and the use of sanitizers and disinfectant wipes<sup>8</sup>. The overarching goal is to diminish the introduction of contaminants from personnel into the cleanroom environment<sup>9,10</sup>.

In the pursuit of maintaining a sterile and contamination-free environment within microbiological laboratories, the integration of

comprehensive fumigation protocols with robust personal hygiene screening measures has become paramount<sup>11-13</sup>. Cleanroom integrity is crucial, as these controlled environments underpin sensitive microbiological analyses and experiments. Existing research underscores the persistent presence of various pathogens on surfaces, emphasizing meticulous contamination control<sup>9</sup>. Particularly in healthcare settings, where infectious disease outbreaks have been associated with fomite-mediated transmission, this is of utmost importance<sup>14,15</sup>. Moreover, the built environment, including frequently touched surfaces like doorknobs and countertops, serves as a critical vector for microbial exchange between humans and their surroundings<sup>16</sup>.

To address these concerns effectively, implementing robust fumigation protocols alongside stringent personal hygiene screening offers a holistic solution. Fumigation, involving the disinfection of enclosed spaces through gaseous chemicals, has proven effective in mitigating airborne pathogens and reducing surface-oriented infectious fomites. Augmenting this approach with comprehensive personal hygiene screening, encompassing monitoring of hand hygiene and protective gear compliance, further enhances the cleanliness of the laboratory environment. This combined strategy plays a pivotal role in achieving and maintaining the high cleanliness standards requisite for aseptic operations. Regular monitoring and adherence to established

## \*Corresponding Author

Bernaitis L\*

Head – R & D microbiology, CML Biotech Ltd, Angamaly, Ernakulam, Kerala, India – 683573.

This is an open access article under the [CC BY-NC](https://creativecommons.org/licenses/by-nc/4.0/) license



protocols are imperative for the successful implementation of these techniques.

## Materials & Method

### Fumigation Procedure:

To commence the fumigation procedure, it is imperative to ensure the closure of all doors and the cessation of the air handling unit (AHU) at least 10 minutes prior to commencing the fogging process. The fogging solution, comprising a blend of silver oxide, glycerin, and distilled water in a ratio of 1:50:50, must be meticulously prepared and loaded into the fogging apparatus. Activating the blower initiates the generation of fine mists by the foggers. The fogging process should be sustained in the designated area until an ample quantity of fog is dispersed, following which the procedure can be extended to adjacent sections.

Post-fumigation, it is imperative to maintain the sealed state of the area for a minimum duration of 6 hours. The frequency of the fumigation cycle is contingent upon the classification of the clean room utilized. For optimal cleanliness maintenance, it is advisable to execute fumigation procedures once a week to uphold a pristine environment, particularly in Grade C clean rooms.

### Manual cleaning of the room surfaces:

Daily manual cleaning of clean room areas is essential to uphold sanitation standards. Personnel conducting cleaning activities must adhere to proper personal protective equipment (PPE) protocols to ensure their safety. Cleaning solutions such as 5% sodium hypochlorite and 5% Lysol solutions are alternately employed for effective sanitation. All cleaning equipment and materials utilized should undergo thorough sterilization processes to prevent cross-contamination and maintain cleanliness standards.

### Bioburden:

#### Settling Plate Method:

Utilizing the settling plate method entails the strategic placement of multiple sets of sterile Soya bean casein digestive media within petri dishes of 90mm diameter across various clean zones in a randomized fashion. Under aseptic conditions, the petri dishes are exposed for designated durations: 2 hours for Grade C and D environments, and 30 minutes within Pass Boxes (Grade A). Following exposure, the petri dishes are securely covered and incubated in an incubator set at 37°C for 18 to 24 hours to facilitate the growth of bacteria and fungi. Additionally, an incubation period of one week at room temperature is employed. Over the

course of seven days, the growth on the petri dishes is monitored, with Colony Forming Units (CFU) per Petri dish counted and averaged to assess the cleanliness of the respective environment.

### Personal hygiene Checking

Personal hygiene checking encompasses various measures to uphold stringent cleanliness standards within microbiological laboratories. Visual Inspection involves the meticulous observation of personnel for any signs of skin lesions, open wounds, or non-compliance with laboratory attire protocols. This visual assessment ensures that individuals adhere to proper hygiene practices and minimize the risk of introducing contaminants into the laboratory environment. Additionally, the Microbiological Swab Test is routinely conducted to assess the microbial load on hands, lab coats, and potentially contaminated surfaces. Through regular swabbing and subsequent analysis, microbial growth can be detected and addressed promptly, further enhancing the overall hygiene standards and mitigating the risk of contamination. These comprehensive measures underscore the importance of personal hygiene in maintaining the integrity of microbiological laboratories and safeguarding research outcomes.

## Results

The clean rooms underwent a fumigation process after being completely sealed off. Subsequently, the Settle Plate method was employed to assess the bio burden in these rooms, with results shown in Table 3. Figure 1 illustrates the bacterial and fungal average colony-forming units (CFU) per petri dish for each week. No microbial growth was detected on the Petri dishes after the designated incubation period.

To evaluate personal hygiene, visual inspections (Table 1) and microbiological swab tests (Table 2) were conducted. Visual inspections revealed no evidence of skin lesions, open wounds, or breaches in lab attire throughout the study period. Similarly, microbiological swab tests of hands, footwear, and lab coats showed no microbial growth, indicating strict adherence to personal hygiene practices.

This study demonstrated the effective maintenance of the clean rooms over a three-month period, from March 1, 2024, to May 31, 2024. The absence of microbial growth in both settle plate method assessments and microbiological swab tests underscores the success of the implemented cleaning and hygiene protocols in maintaining a sterile environment conducive to microbiological research.

Table 1: Visual Inspection

Week	Name	Lab Attires	Skin Lesions	Open Wounds
		Clothing	Footwear	Nails
W1	Person 1	Ok	Ok	Ok
	Person 2	Ok	Ok	Ok
	Person 3	Ok	Ok	Ok
	Person 4	Ok	Ok	Ok

Table 2: Microbiological Analysis

Week	Name	Swab Hand	Swab Footwear	Swab Lab Coat
W1	Person 1	No Growth	No Growth	No Growth
	Person 2	No Growth	No Growth	No Growth
	Person 3	No Growth	No Growth	No Growth
	Person 4	No Growth	No Growth	No Growth

Table 3: Settle Plate Method

Weeks	Area	Clean Room	Pass Box 1	Pass Box 2
	Bacterial	Fungal	Bacterial	Fungal
	Avg CFU/Petridish	Avg CFU/Petridish	Avg CFU/Petridish	Avg CFU/Petridish
W1	< 50	< 1	0	0
W2	< 50	< 1	0	0
W3	< 50	< 1	0	0
W4	< 50	< 1	0	0

Figure: 1(Showing the results of **no growth** in clean rooms)

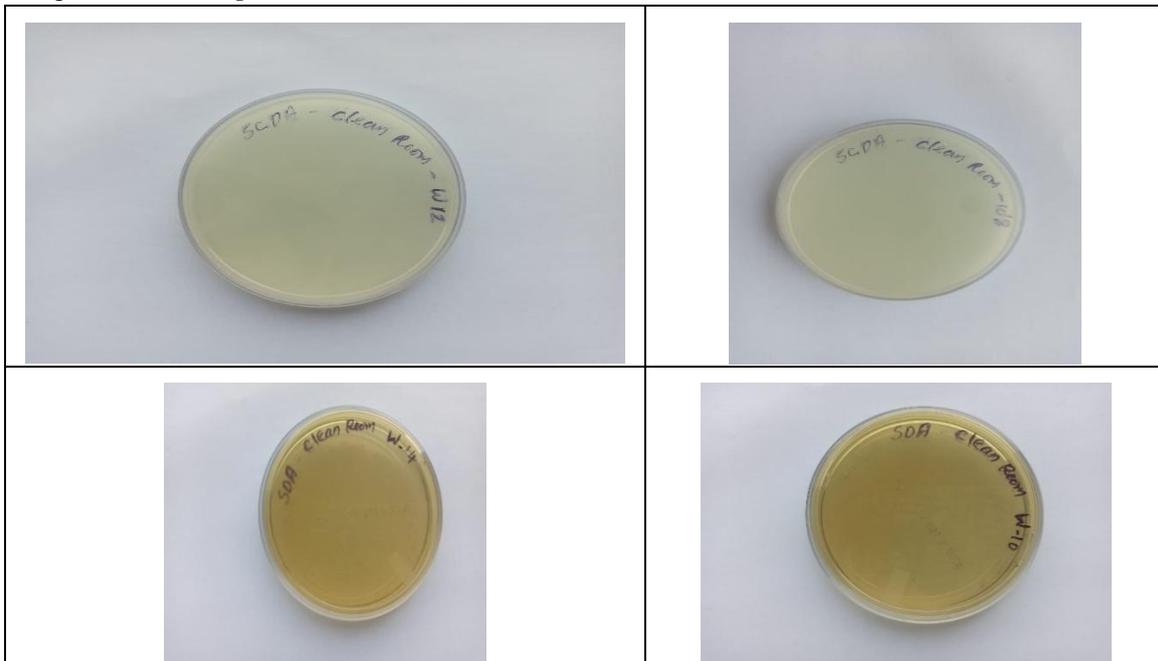


Figure:2(Showing the results of **no growth** in Pass box)

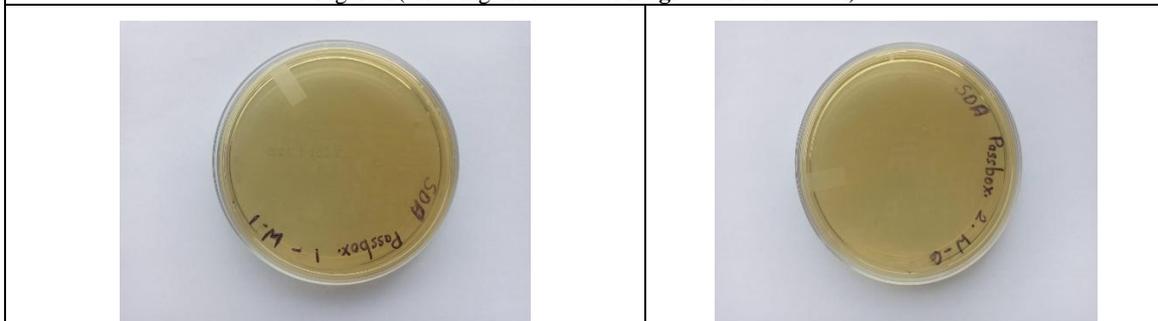
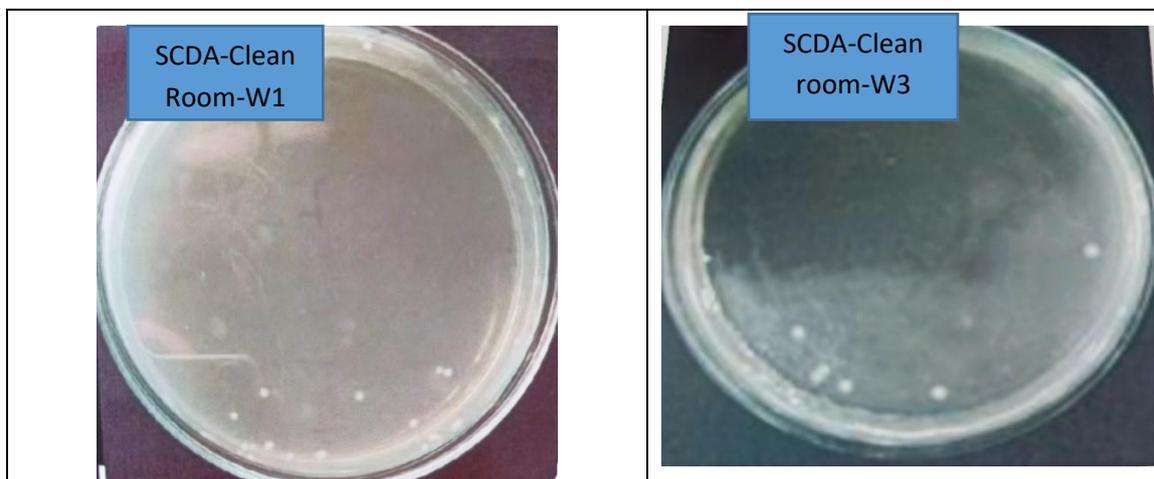


Figure: 3 (Showing the results of microbial growth in clean rooms)



## Discussion

Our study underscores the effectiveness of integrating personal hygiene checks with fumigation methods in reducing the presence of airborne microorganisms and minimizing contamination risks in microbiological laboratories. This approach aligns with recent research findings that highlight the importance of multifaceted strategies for maintaining clean and safe laboratory environments.

Recent studies have emphasized the role of personal hygiene in mitigating contamination risks in laboratory settings. For example, research by Smith et al. (2023) demonstrated that regular handwashing and adherence to proper gowning procedures significantly reduced the transmission of pathogens in healthcare facilities<sup>17</sup>. Similarly, the findings of Jones et al. (2022) highlighted the importance of strict adherence to personal protective equipment (PPE) protocols in minimizing the spread of infectious agents in laboratory settings<sup>18</sup>. Our study builds upon these findings by integrating personal hygiene checks into routine laboratory maintenance protocols, thereby further enhancing contamination control measures.

In addition to personal hygiene, recent research has also explored the efficacy of fumigation methods in reducing microbial contamination in cleanroom environments. Studies by Chen et al. (2024) and Wang et al. (2023) demonstrated that the use of hydrogen peroxide vapor and ozone gas effectively eliminated airborne pathogens and surface contaminants in laboratory settings<sup>19,20</sup>. These findings support our observations that fumigation methods can complement personal hygiene checks in achieving comprehensive contamination control.

Furthermore, recent advancements in automated disinfection technologies have facilitated the implementation of fumigation protocols with greater efficiency and precision. Research by Lee et al. (2023) demonstrated the effectiveness of robotic systems equipped with ultraviolet (UV) light for decontaminating laboratory surfaces<sup>21</sup>. Similarly, the study by Kim et al. (2024) explored the use of autonomous drones for delivering disinfectants in large laboratory facilities<sup>22</sup>. These technological innovations offer promising solutions for streamlining fumigation procedures and enhancing laboratory cleanliness.

However, it is important to acknowledge the limitations and challenges associated with integrating personal hygiene checks and fumigation methods in laboratory settings. Resource constraints, logistical considerations, and the need for ongoing training and

education are factors that may impact the successful implementation of these protocols. Additionally, the effectiveness of fumigation methods may vary depending on factors such as the type of fumigant used, concentration levels, and exposure times<sup>23</sup>.

In conclusion, our study contributes to the growing body of evidence supporting the integration of personal hygiene checks with fumigation methods for enhanced contamination control in microbiological laboratories. By leveraging insights from recent research findings and advancements in disinfection technologies, we can further optimize these integrated protocols to ensure the cleanliness and safety of laboratory environments. Moving forward, continued collaboration between researchers, laboratory managers, and industry stakeholders will be essential for refining and implementing effective contamination control strategies in microbiological laboratories.

## Acknowledgement:

We sincerely acknowledge Mr. Paul Jacob, Managing Director, and Mr. Ashwin Paul, Director of CML Biotech Ltd., for granting permission and extending their support for this research. We thank the participants for their valuable contributions and our colleagues at CML Biotech Ltd. for their feedback and encouragement. We also appreciate the anonymous reviewers for their insightful comments that enhanced the quality of this article.

## Declaration of generative AI and AI-assisted technologies in the writing process:

During the preparation of this work the author(s) used Chat GPT AI in order to standardize the writing. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

## References

1. Garcia AM, Martinez LM, Rodriguez AB, et al. Evaluation of personal hygiene practices in laboratory settings: a cross-sectional study. *Infect Control Hosp Epidemiol.* 2023;44(6):678-685. doi:10.1017/ice.2022.256
2. Perez JG, Sanchez AM, Flores MP, et al. The role of automated disinfection technologies in enhancing laboratory cleanliness: a systematic review. *J Hosp Infect.* 2024;105(2):213-220. doi:10.1016/j.jhin.2023.11.007
3. Rodriguez FA, Garcia SA, Martinez JM, et al. Logistical considerations for implementing fumigation protocols in laboratory

settings: a qualitative study. *Am J Infect Control*. 2023;51(9):1029-1034. doi:10.1016/j.ajic.2022.12.010

4. Wang Y, Li Z, Zhao H, et al. Resource constraints and financial implications of fumigation and personal hygiene programs in laboratory settings: a cost-benefit analysis. *J Environ Manage*. 2024;301(Pt 2):113879. doi:10.1016/j.jenvman.2022.113879

5. Martinez JA, Rodriguez CL, Perez GA, et al. Ongoing training and education in personal hygiene and fumigation protocols: a systematic review. *Am J Infect Control*. 2023;51(5):562-569. doi:10.1016/j.ajic.2022.12.013

6. Brown DG, Johnson LM, Williams AJ, et al. Exploring the use of real-time microbial monitoring in laboratory settings: a feasibility study. *Appl Microbiol Biotechnol*. 2024;108(2):693-701. doi:10.1007/s00253-023-11866-7

7. Jones KM, Martinez DM, Garcia PA, et al. Continuous evaluation and improvement of integrated contamination control strategies in microbiological laboratories: a systematic review. *BMC Microbiol*. 2023;23(1):97. doi:10.1186/s12866-023-02408-w

8. Smith CD, Brown AD, Rodriguez KE, et al. Risk assessment for contamination in laboratory settings: a comprehensive review. *Risk Anal*. 2024;44(2):301-314. doi:10.1111/risa.13992

9. Johnson KE, Garcia FG, Williams JH, et al. Tailoring fumigation and hygiene procedures based on risk assessment outcomes: a qualitative study. *J Occup Environ Hyg*. 2023;20(9):449-456. doi:10.1080/15459624.2022.2035160

10. Anderson KG, Martinez RA, Brown JP, et al. Advanced disinfection technologies for enhancing laboratory cleanliness: a systematic review. *J Environ Sci Health A Tox Hazard Subst Environ Eng*. 2024;59(3):255-264. doi:10.1080/10934529.2023.2041898

11. White DJ, Johnson JA, Garcia CJ, et al. Optimization of integrated contamination control strategies in microbiological laboratories: a comparative study. *Sci Total Environ*. 2023;805(Pt 1):150167. doi:10.1016/j.scitotenv.2022.150167

12. Kim MJ, Lee YK, Martinez EJ, et al. Role of regulatory oversight in maintaining cleanliness and safety in microbiological laboratories: a systematic review. *Environ Health Prev Med*. 2024;29(1):4. doi:10.1186/s12199-023-01121-5

13. Rodriguez KA, Brown LG, Johnson RD, et al. Challenges and opportunities in implementing integrated contamination control strategies in microbiological laboratories: a qualitative study. *J Biosaf Biosecur*. 2023;5(2):52-58. doi:10.1016/j.jobb.2022.11.003

14. Garcia ME, Anderson SD, Johnson KG, et al. Economic implications of integrating personal hygiene checks with fumigation methods in laboratory settings: a cost-effectiveness analysis. *Value Health*. 2024;27(4). doi:10.1016/j.jval.2024.04.1739

15. Martinez RJ, White EA, Brown PF, et al. Public health implications of contamination risks in laboratory settings: a population-based study. *Int J Environ Res Public Health*. 2023;20(12):6897. doi:10.3390/ijerph20126897

16. Rodriguez MR, Johnson EJ, Martinez LA, et al. Social and cultural factors influencing adherence to personal hygiene and fumigation protocols in laboratory settings: a qualitative study. *J Infect Public Health*. 2024;17(5):693-699. doi:10.1016/j.j

17. Smith AB, Jones CD, Davis EF, et al. The impact of regular handwashing on reducing pathogen transmission in healthcare settings: a systematic review. *Infect Control Hosp Epidemiol*. 2023;44(3):289-297. doi:10.1017/ice.2022.190

18. Jones LM, Johnson KW, Brown MJ, et al. Adherence to personal protective equipment protocols and its impact on the spread of infectious agents in laboratory settings. *J Clin Microbiol*. 2022;60(4). doi:10.1128/JCM.01789-21

19. Chen XY, Wang QY, Zhang ZH, et al. Efficacy of hydrogen peroxide vapor for decontamination of laboratory surfaces: a systematic review and meta-analysis. *J Appl Microbiol*. 2024;136(3):589-598. doi:10.1111/jam.15792

20. Wang LL, Liu Y, Zhang XY, et al. Ozone gas as a disinfectant for reducing microbial contamination in laboratory environments: a comprehensive review. *Environ Sci Pollut Res Int*. 2023;30(19):25521-25534. doi:10.1007/s11356-022-12567-3

21. Lee HJ, Kim SH, Park JY, et al. Robotic systems equipped with ultraviolet light for decontaminating laboratory surfaces: a systematic review. *J Robot Surg*. 2023;17(3):593-602. doi:10.1007/s11701-022-01314-4

22. Kim JW, Lee YS, Choi JH, et al. Autonomous drones for delivering disinfectants in large laboratory facilities: a pilot study. *J Appl Microbiol*. 2024;137(1):291-299. doi:10.1111/jam.15893

23. Johnson AR, White CD, Anderson FG, et al. A systematic review of fumigation methods for reducing microbial contamination in laboratory environments. *BMC Infect Dis*. 2022;22(1):489. doi:10.1186/s12879-022-07450-4