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Sterilization And Disinfectant In Pharmaceutical Industry

Zahid Aziz Ul Hussain[1], Mrs. Khadijah Al Khadir[2], Abdur Rafay Muzzamil[3]
[1]PG Student, Department of Microbiology, Mumtaz Degree and PG College. Hyderabad, India.
[2]Head, Department of Microbiology, Mumtaz Degree and PG College. Hyderabad, India.
[3]Director, NSQ Pharma Pvt Ltd, Hyderabad, India.
zauh313@gmail.com
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Abstract:
Grasping the principles of disinfection, sterilization, cleaning, and asepsis is crucial for preventing infection transmission to patients and safeguarding healthcare workers (HCWs). The proper disinfection of surfaces after cleaning, a crucial requirement at all times, has gained particular importance during the ongoing epidemic. Cleaning, the first phase of sterilization, is crucial for minimizing bioburden. The required disinfection method is contingent upon the characteristics of the equipment and its intended use. For instance, important goods need extensive disinfection. This narrative review elucidates the techniques of decontamination and sterilizing. A variety of chemicals may serve both sterilizing and disinfection purposes, with the distinction depending on the concentration of the chemical and the duration of exposure. Sterilization is the process by which all forms of life are eradicated, eliminated, or permanently inactivated. While this definition posits sterility as an absolute ideal, in industrial practice, sterilization is often described as the technique that reduces the likelihood of survival of unwanted organisms to an arbitrarily low level. This level is generally so low that the probability of even a single organism surviving the sterilizing procedure is considered minimal. This stringent criterion is warranted, particularly in scenarios where the possible health implications (e.g., in the formulation of parenteral medications) or the integrity of the process (e.g., in antibiotic fermentation) render any degree of microbial contamination unacceptable. This probabilistic method of sterility does not suggest that the sterility standards of the procedure may be reduced. Rather, it indicates that particularly extensive industrial sterilization procedures are influenced by numerous variables (e.g., the initial organism count, the inherent variability in each microorganism's heat resistance, and the challenge of precisely regulating temperature throughout all sections of specific equipment) that cannot be entirely controlled. Disinfectants exhibit varying efficacy against distinct bacteria, a discrepancy attributed to both the inherent resistance of various microbes and the diversity of disinfectant kinds and formulations. Moreover, several disinfectants function differently based on their active components. The article concentrates on the legislation and procedural criteria, as well as the qualifications necessary for the introduction of a disinfectant into use.
Keywords: Sterlization, Disinfectant, Decontamination, Sanitization, Antiseptics, Fumigation, biological indicator

1-Introduction

Sterilization is described as any procedure that efficiently eradicates transmissible agents, including fungus, bacteria, viruses, and prions, from surfaces, equipment, food, pharmaceuticals, or biological culture media. Sterility is attained by exposing the thing to be sterilized to a chemical or physical substance for a designated duration. Agents used as steriliants include excessive temperature, ionizing radiation, and chemical liquids or gasses. The efficacy of the procedure is contingent upon the selected sterilizing technique.

Sterilization is the procedure of eliminating all kinds of microbial life present on or inside a specific item or preparation. It is the logarithm of the decrease in bacterial burden. Microbiologically, sterile material is defined as containing no live organisms, and the word sterile indicates the absence of viable organisms. The sterilizer (Autoclave) has a completely automated microprocessor controller that manages and monitors the sterilizing process. This autoclave utilizes saturated steam (or pure steam) as the sterilizing agent. The sterilizing apparatus mitigates the influence of cold air on temperature using vacuum technology. Vacuum dehumidification coupled with jacket heating for item desiccation. Ultimately, the sterile air is evacuated to attain pressure equilibrium.

Pharmaceutical Importance of Sterilization

- **Moist heat sterilization** is the most efficient biocidal agent. In the pharmaceutical industry it is used for: Surgical dressings, Sheets, Surgical and diagnostic equipment, Containers, Closures, Aqueous injections, Ophthalmic preparations , Irrigation fluids and **microbiological media and testing articles** etc.
- **Dry heat sterilization** can only be used for thermostable, moisture sensitive or moisture impermeable pharmaceutical and medicinal. These

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include products like; Dry powdered drugs,
Suspensions of drug in nonaqueous solvents, Oils, fats, waxes, soft hard paraffin, silicone, Oily injections, implants, ophthalmic ointments and ointment bases etc.

2 Disinfectant In Pharmaceutical

Disinfectant

A robust cleaning and sanitization protocol is essential for controlled conditions used in the production of Pharmacopeia goods to avert microbiological contamination of these products. Sterile drug products may be contaminated by their pharmaceutical ingredients, process water, packaging components, manufacturing environment, processing equipment, and manufacturing personnel. Current Good Manufacturing Practices (cGMPs) underscore the dimensions, design, structure, and positioning of facilities and materials, as well as the suitable material flow to enhance cleaning, maintenance, and efficient operations in medicinal product manufacturing. In a manufacturing setting, it is essential to exercise caution while using disinfectants to avoid contaminating the drug product with chemical disinfectants due to their intrinsic toxicity. Aseptic processing requires easily cleanable floors, walls, and ceilings with smooth, nonporous surfaces; controls for particulates, temperature, and humidity; and established cleaning and disinfection protocols to establish and sustain aseptic conditions. The cleaning and sanitization program must meet established cleanliness standards, manage microbial contamination of products, and be structured to avert chemical contamination of pharmaceutical ingredients, product-contact surfaces, equipment, packaging materials, and, ultimately, the drug products. These principles are applicable to nonsterile dosage forms, where microbial contamination is managed through the selection of appropriate pharmaceutical ingredients, utilities, manufacturing environments, effective equipment cleaning protocols, specially formulated products to regulate water activity, incorporation of suitable preservatives, and thoughtful product packaging design. Besides disinfectants, antiseptics are used to sanitize human skin and exposed tissue, and may be utilized by staff before entering the industrial area. Chemical sterilants may be used to disinfect surfaces in industrial and sterility testing environments. Additionally, sterilants may be used for the sterilization of Pharmacopeial items, while UV irradiation may serve as a surface sanitizer. This chapter on general information will address the selection of appropriate chemical disinfectants and antiseptics, the demonstration of their bactericidal, fungicidal, and sporicidal effectiveness, the application of disinfectants in sterile pharmaceutical manufacturing, and regulatory and safety considerations. The production of biofilms and its association with disinfectants are not addressed in this chapter. Further material not addressed in the chapter may be found from authoritative books on disinfectants and antiseptics.

Antiseptic— A substance that prevents or eliminates the growth of microbes on live tissue, such as skin, oral cavities, and wounds.

Chemical Disinfectant— A chemical treatment used to inanimate surfaces and objects to eradicate pathogenic fungus, viruses, and bacteria, albeit not always their spores. Sporicidal and antiviral agents may be classified as a distinct category of disinfectants. Disinfectants are often classified as high-level, intermediate-level, and low-level by medical organizations based on their effectiveness against different pathogens.

Cleaning Agent— An agent designed to eliminate product residues from facility and equipment surfaces that might deactivate sanitizing agents or harbor bacteria.

Decontamination—The removal of microorganisms by disinfection or sterilization.

Disinfectant—A chemical or physical agent that destroys or removes vegetative forms of harmful microorganisms when applied to a surface

Sanitizing Agent—An agent for reducing, on inanimate surfaces, the number of all forms of microbial life including fungi, viruses, and bacteria.

Sporicidal Agent—An agent that destroys bacterial and fungal spores when used in sufficient concentration for a specified contact time. It is expected to kill all vegetative microorganisms.

Sterilant— An agent that eradicates all types of microbial life, including fungus, viruses, and all varieties of bacteria and their spores. Sterilants are agents in liquid or vapor form. Microorganisms exhibit significant variability in their resistance to disinfectants. The hierarchy of resistance among clinically relevant microbes to chemical disinfectants, arranged from most resistant to least resistant.

Chemical disinfectants are categorized according to their chemical composition. This encompasses aldehydes, alcohols, halogens, peroxides, quaternary ammonium compounds, and phenolic compounds.

General Classification of Antiseptics, Disinfectants, and Sporicidal Agents.

Chemical entity	Classification	Examples
Aldehydes	Sporicidal agent	2% Glutaraldehyde
Alcohols	General purpose disinfectant, antiseptic, antiviral agent	70% Isopropyl alcohol, 70% alcohol

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Hydrogen peroxide	Vapor phase sterilant, liquid sporicidal agent, antiseptic	4 µg per g H_2O_2 vapor, 10%–25% solution, 3% solution
Uaternary ammonium compounds	General purpose disinfectant, antiseptic	200 µg per g Benzalkonium chloride

Selection Of A° Disinfectant For Use In A Pharmaceutical Manufacturing Environment

When selecting a disinfectant for use in a pharmaceutical manufacturing area, the following points should be considered: the number and types of microorganisms to be controlled; the spectrum of activity of commercially available disinfectants; the reputation of the disinfectant supplier; the claims as a sterilant; the disinfectant or sanitizer supported by the EPA registrations; the concentration, application method, and contact time of the disinfectant; the nature of the surface material being disinfected and its compatibility with the disinfectant; the amount of organic compounds on the surface that may inactivate a disinfectant; the possible need to maintain a residual bactericidal activity of the disinfectant on the surface; the corrosiveness of the disinfectant to equipment with repeated application; the safety considerations for operators applying the disinfectant; the compatibility of the disinfectant with cleaning agents and other disinfectants; the planned disinfectant rotation; and the steps that need to be taken to avoid the contamination of pharmaceutical products by a disinfectant

Theoretical Discussion Of Disinfectant Activity

Graphs depicting the logarithm of the microbial count per mL remaining in a disinfectant solution suggest that first-order kinetics serve as a rough estimate for the decline in microbial population over time. The graphs exhibit a more sigmoid curve, characterized by a gradual initial decline in numbers, then followed by an accelerating increase over time. The rate constant, K, for the disinfection process may be determined using the following formula:

$(1 / t)(\log NO / N)$

in which t is the time, in minutes, for the microbial count to be reduced from NO to N; NO is the initial number of organisms, in cfu per mL; and N is the final number, in cfu per mL, of organisms.

As with a first-order chemical reaction, the same concentration of disinfectant reduces the number of organisms more rapidly at elevated temperatures. This can be expressed as a

temperature, T, coefficient per 10° rise in temperature, $Q10$, calculated by the formula:

Time to decontamination at T° / Time to decontamination at T

in which T is $T^{\circ} - 10$.

Additional evidence indicating that a first-order reaction inadequately characterizes disinfection is that the Q10 values for chemical and enzymatic reactions range from 2 to 3, yet the prevalent disinfectants phenol and alcohol exhibit Q10 values of 4 and 45, respectively.

An knowledge of the impact of disinfectant concentration on microbial decrease is essential for the effective use of disinfectants. A graph depicting the logarithm of the time required to diminish the microbial population in a typical inoculum to zero vs the logarithm of the disinfectant concentration is linear, with the slope referred to as the concentration exponent, n. The connection may be articulated as follows:

n = (log of the kill time at concentration $C2$) – (log of the kill time at concentration $C1$) / (log $C1$ – log $C2$)

in which $C1$ and $C2$ are the higher and lower disinfectant concentrations, respectively.

The significant variations in concentration exponents, n, have practical implications for selecting the use dilution of various disinfectants and for using dilution to neutralize a disinfectant in effectiveness testing and regular microbiological monitoring of the industrial environment. For instance, mercuric chloride with a concentration exponent of 1, so a 3-fold dilution would diminish its disinfection efficacy by 31 (or one-third), but phenol, with a concentration exponent of 6, will experience a 36 (or 729-fold) drop in disinfecting activity. Disinfectants having a higher concentration exponent or dilution coefficient quickly diminish in efficacy upon dilution.

Microbial Resistance To Disinfectants

The emergence of microorganism resistance to antibiotics is a well-documented occurrence. The emergence of microbial resistance to disinfectants is less probable, as disinfectants are more potent biocidal agents than antibiotics and are utilized in high concentrations against low populations of microorganisms that are typically not actively proliferating, resulting in diminished selective pressure for resistance development. The bacteria most often identified from an environmental monitoring program may regularly undergo use dilution testing with the chemicals used in the disinfection program to verify their susceptibility.

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3-Materials And Methods

Heat Penetration Cycle # 1 Outcome:

S.No.	Test Parameters	Acceptance Criteria	Actual Observations	
1.	Heat Penetration	➢ All temperature sensors should be within (121-124) °C for a period of 30 minutes during sterilization.	Complies	
2.	Lethality F0 Value	➢ Lethality will be calculated for each sensor and should be equal to or more than the set sterilization hold time duration.	Complies	
3.	Biological indicator	➢ All biological indicators should comply (no growth should be observed after incubation of 7 days)	Complies	
Conclusion of Cycle: All the PQ test results comply with acceptance criteria			☑ Yes	❑ No

Heat Penetration Study -Decontamination with Maximum Load -Cycle # 2

Sterilization Set Temperature		121°C (Sterilization band: 121°C -124°C)					
Time		30 minutes					
PQ Cycle No.		2					
Loaded Pattern Details		Load Description	Quantity		Tray		
		Microbiology Waste	2 POLY BAG/25 bottles (250 ml)		01		
Attachment # 22		Decontamination Heat Penetration Study results and graphs for cycle #2- Maxiumum load					
Attachment # 24		Biological Indicators' Results					
Serial No.	Sensor No.	Study Summary					
		Average Value °C	Minimum Value °C	Maximu m Value °C	Delta=Max - Min	F0 Value	BI Results
1.	R8522	122.478	122.05	122.58	0.53	41.33	Comply

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2.	R8495	122.512	122.43	122.58	0.15	41.64	N/A
3.	R8510	122.539	122.44	122.63	0.19	41.91	Comply
4.	R8500	122.433	121.80	122.58	0.78	40.92	N/A
5.	R8523	122.408	121.79	122.56	0.77	40.68	Comply
6.	R8519	122.341	121.79	122.50	0.71	40.06	N/A
7.	R8501	122.507	122.17	122.60	0.43	41.60	Comply
8.	R8491	122.446	121.67	122.60	0.93	41.05	N/A
9.	R8507	122.401	121.75	122.53	0.78	40.61	Comply
10.	R8493	122.285	121.59	122.47	0.88	39.56	N/A
11.	R8518	122.537	122.21	122.63	0.42	41.89	Comply
12.	R8521	122.478	121.93	122.60	0.67	41.34	N/A
13.	R8515	122.388	121.88	122.50	0.62	40.49	Comply
14.	R8481	122.222	121.48	122.45	0.97	39.00	N/A
15.	R8494	122.523	122.44	122.58	0.14	41.75	N/A
16.	R8508	122.504	121.96	122.61	0.65	41.58	Comply
17.	R8307P	2.1318	2.1047	2.1511	0.0464		

Summary of Study Locations					
Min. Temp		Max. Temp.		Min. F0	
Sensor -R8481	121.48°C	Sensor-R8510	122.63°C	Sensor - R8481	39.00
Min. Average Temp		Max. Average Temp.		Max. F0	
Sensor-R8481	122.222°C	Sensor-R8510	122.539°C	Sensor - R8510	41.91
Temp. Overall Mean		122.435°C	Delta=Max(max)-Min(min)		1.150°C
Max Single-Point Negative Deviation		-0.96	Max Single-Point Positive Deviation		0.19
Max Average Negative Deviation		-0.21	Max Average Positive Deviation		0.10
Sterilization Hold time		30 minutes			

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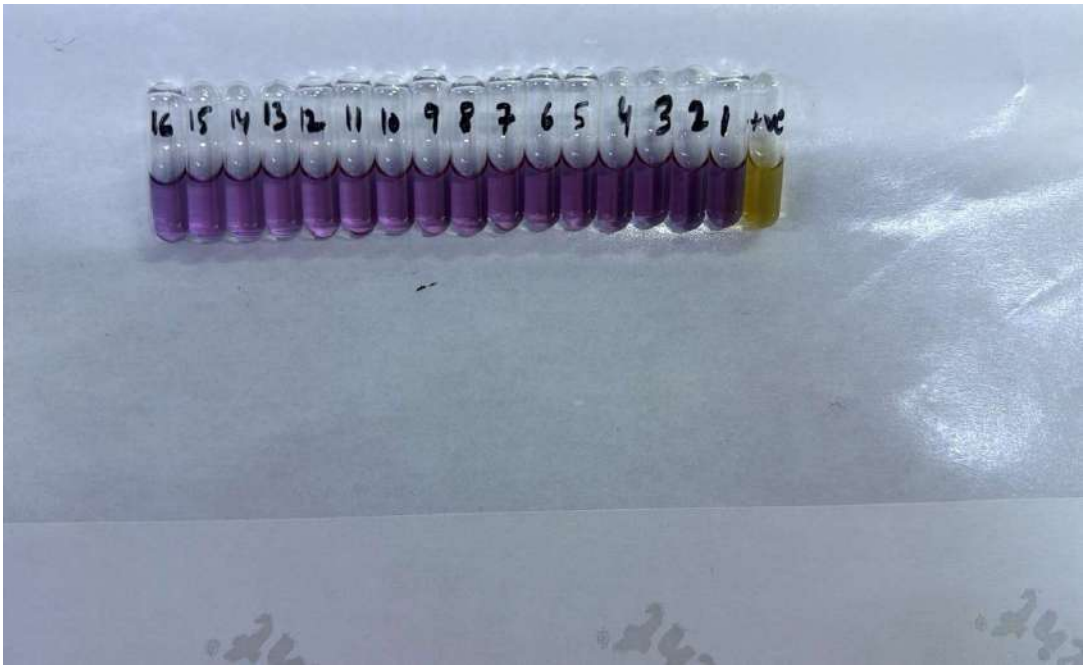
4-Results



After incubation the positive colour changes to yellow while other BI ampules remains purple after sterilization

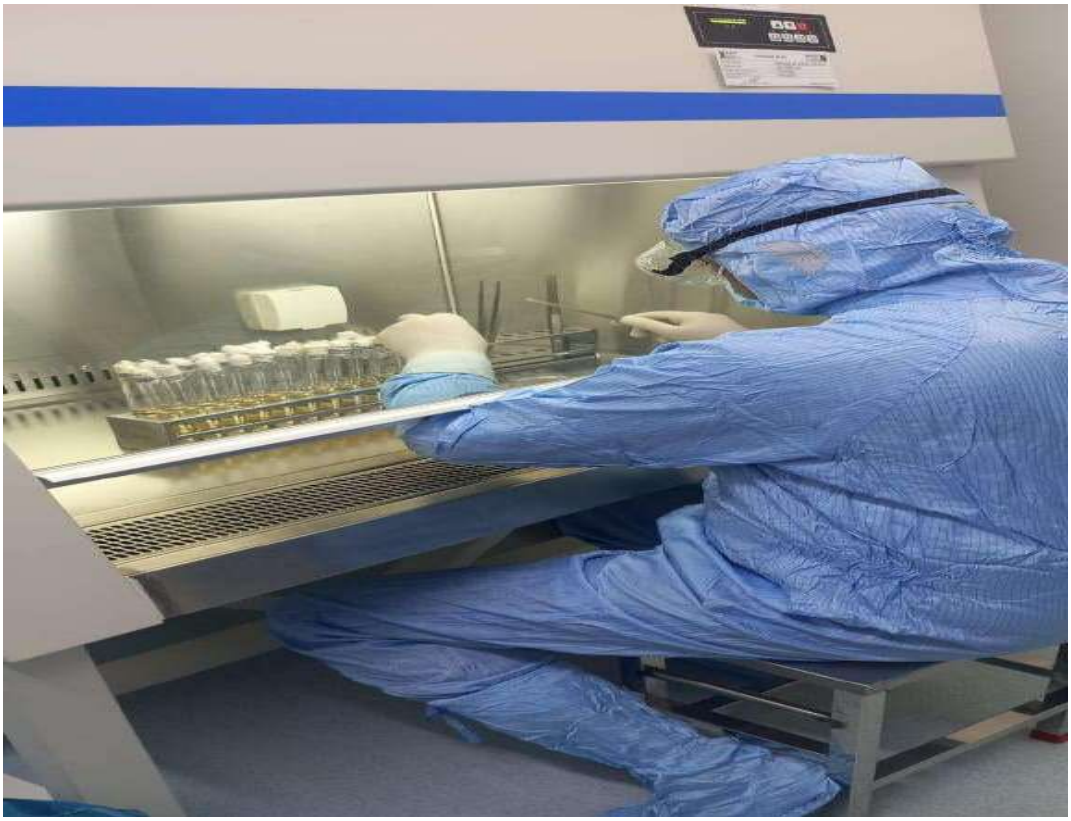


Fig: Testing of Biological Indicators after fogging

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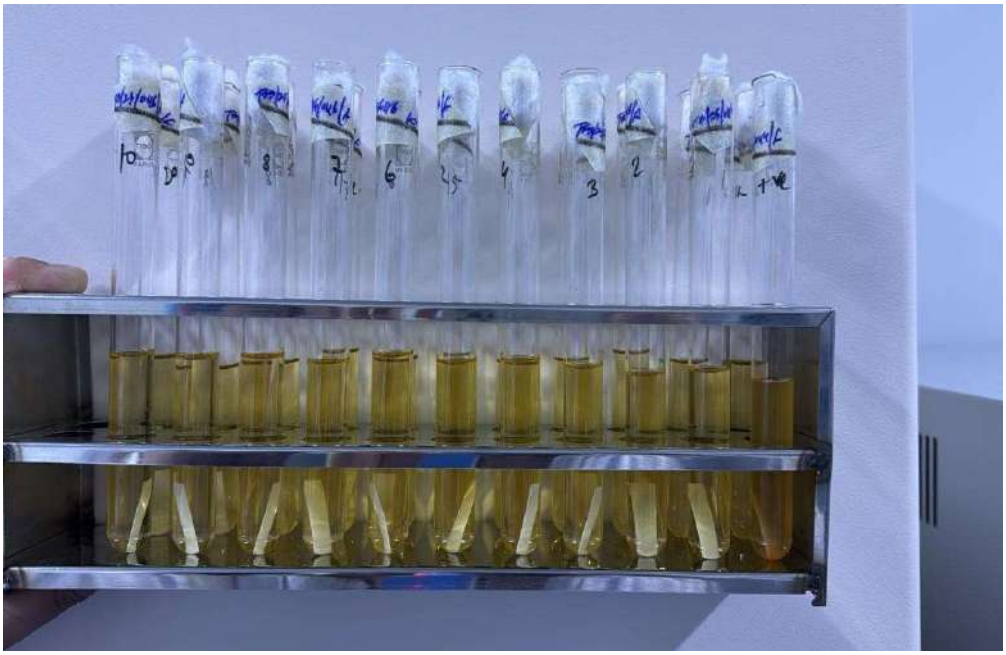
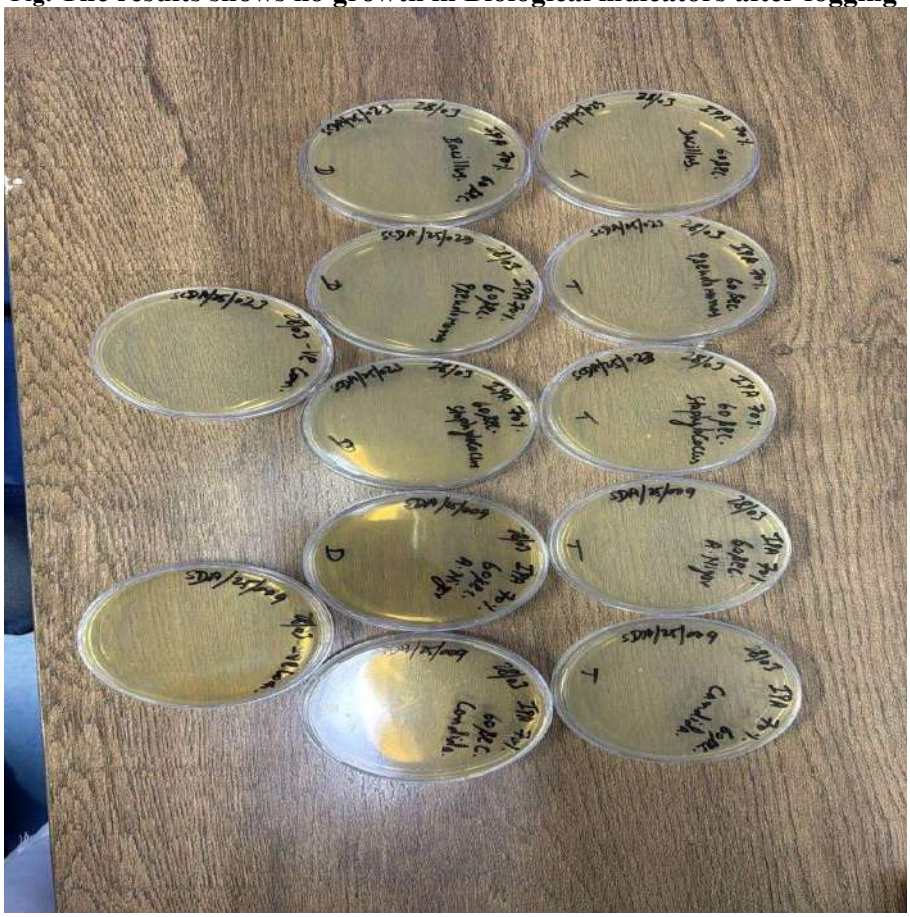


Fig: The results shows no growth in Biological indicators after fogging



After challenge with disinfectant shows the 3.log reduction for bacteria and 2 log reduction for fungi

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Fig: Fumigation in Clean Room

5-DISCUSSION

Location	Room Code	Result after 7 days	Result
Filling room fix to AHU-1	224	-ve	Pass
Filling room left side corner	224	-ve	Pass
Filling room fix to AHU-2	224	-ve	Pass
Filling room fix to Filling M/C	224	-ve	Pass
Fix to Air Lock Room AHU	221	-ve	Pass
Fix to Air Lock Room AHU	222	-ve	Pass
Preparation room fix to AHU	223	-ve	Pass
Preparation room fix to Tank	223	-ve	Pass
Preparation room Right side corner	223	-ve	Pass
Fix to center of the corridor	Cord-1	-ve	Pass
Right side corner of corridor	Cord-1	-ve	Pass
Left side corner of corridor	Cord-1	-ve	Pass
Fix to Raw Material Storage room AHU	211	-ve	Pass
Fix to change room AHU	202	-ve	Pass
Fix to Buffer room	204	-ve	Pass

Fix to change room AHU	201	-ve	Pass
Fix to Buffer room	203	-ve	Pass
Fix to Film Pipe Temporary Storage room	213	-ve	Pass
Fix to Dispensing room AHU	220	-ve	Pass
Fix to change room AHU	541	-ve	Pass
Fix to change room AHU	542	-ve	Pass
Fix to change room AHU	543	-ve	Pass
Fix to Sterility Test Room AHU	544	-ve	Pass

6-Conclusion

- After the fogging with 10% H2O2 the area was hold for 24 hrs.
- After 12hrs the Bi has collected and tested.
- After testing BI, it Concludes that all the location placements are complies.
- After testing BI's incubated for 7 days the test BI's does not show any growth and positive control shows growth.
- The fogging had shown efficacy with 10% H2O2
- 10% H_2O_2 is recommended for fogging.

References

[1] Adapted from University of Iowa

[2] Adapted from University of Iowa.

[3] APIC (Association for Professionals in Infection Control and Epidemiology) "Guidelines for Selection and Use of Disinfectants" reprint from American Journal of Infection Control; Vol. 24, No. 4, pp. 313-342, August 1996.

[4] Ashley, M.H.J. (1982). Continuous sterilization of media. *Chem. Eng. (London)* **1** (2): 54–58.

[5] Block, S.S.; Disinfection, Sterilization and Preservation; 5th Edition (2000) Lippincott Williams & Wilkins; Philadelphia

[6] Davis, B.D. Dulbecco, R., Eisen, H.N. et al; Microbiology, Third Edition (1980); Harper and Row; Hagerstown, MD

[7] **Google Scholar**

[8] Joklik W.K., Willet, H.P.; Zinsser Microbiology; 18th Edition 1984; Appleton Century-Crofts/ New York

[9] Kruse, R.H.; Puckett, William H.; "Biological Safety Cabinetry"; Clinical Microbiological Reviews; Vol 4 (2), pp 207-241; (1991) American Society for Microbiology, Washington, D.C.

[10] Lin, S.H. (1979). Residence time distribution of flow in a continuous sterilization process. *Process Biochem.* **14** (7): 23–27.

[11] Manufacturing Chemists Association: Laboratory Waste Disposal Manual. Revised August 1975 MCA 1825 Connecticut Avenue, Washington DC 20009

[12] Pelczar, M.J. Reid, R.D., Chan, E.C.S.; Microbiology, 4th Edition (1977); McGraw Hill Book Company, New York

Richards, J.W. (1968). *Introduction to Industrial Sterilization*. New York: Academic Press.

[13] Rutala W.A., Waber D.J. New disinfection and sterilization methods. Emerg Infect Dis. 2001;7(2):348–353. doi: 10.3201/eid0702.010241. [DOI] [PMC free article] [PubMed] [Google Scholar]

[14] U.S. DHEW/P.H.S/NIH/NCI; Effective use of the Laminar Flow Biological Safety Cabinet; NCI, Bethesda, MD

[15] United States Pharmacopeia (USP) (2017). *National Formulary, USP40-NF35. Microbiological Control and Monitoring of Aseptic Processing Environments ⟨1116⟩; Sterilization and sterility assurance of compendial articles ⟨1211⟩ and Sterilization of compendial articles ⟨1229⟩*. Rockville, MD: The USP Convention, Inc.

[16] USP ⟨1072⟩ DISINFECTANTS AND ANTISEPTICS.

[17] Wallhäusser, K.H. (1985). Sterilization in biotechnology. *J. Chem. Technol. Biotech.* **2**: 699–723.

[18] **Web of Science®Google Scholar**

[19] WHO good manufacturing practices for sterile pharmaceutical products

[20] WHO Infection Control Guidelines for Transmissible Spongiform Encephalopathies, Geneva, Switzerland, 23-26 March 1999.