International Journal of PharmTech Research
CODEN (USA): IJPRIF, ISSN: 0974-4304
Vol.9, No.4, pp 220-226, 2016

Qualification of Autoclave

N. Vishal Gupta*, Shukshith K.S.

Pharmaceutical Quality Assurance group, Department of Pharmaceutics, JSS College of Pharmacy, JSS University, Sri Shivarathreeshwara Nagara, Mysuru - 570015, Karnataka, India

Abstract: In accordance with GMP, each pharmaceutical company should identify what qualification work is required to prove that the critical aspects of their particular operation are controlled. The key elements of a qualification and validation programme of a company should be clearly defined and documented. Qualification is the integral part of GMP and there is no effective QA program without qualification. Now-a-days it is mandatory to incorporate qualification activity for any system in the manufacturing premises for all pharmaceutical industries.

The purpose of this study is to initially develop the sterilization process parameter for the porous load articles then implement the sterilization process for the porous articles. The process development included qualification of equipment and the articles. The auto clave cum bung processor which is used for the cleaning and sterilizing rubber stoppers, garments and machine parts. This was followed by performing the qualification of the equipment which describes the entire test right from Vacuum leak test, Bowie dick test, heat distribution test and heat penetration test were equipment passes all test and the equipment is suitable for sterilization purpose which meeting its predetermined specification and quality attributes.

Keywords: Autoclave qualification, Sterilization, Vacuum leak test, Bowie-dick test, Heat distribution test, Heat penetration test.

Introduction

Definition of Autoclave

An autoclave is a pressure chamber used to carry out industrial processes requiring elevated temperature and pressure different to ambient air pressure. Autoclave are used in medical application to perform sterilization, and in the chemical industry to cure coatings, vulcanize rubber and for hydrothermal synthesis[1].

Definition of Sterilization

Sterilization can be defined as any process that effectively kills or eliminates transmissible agents (such as fungi, bacteria, viruses) from surface, equipment, foods, medications or biological culture medium[2]. A sterility assurance level (SAL) of 10-6 means that there is less than or equal to one chance in million that particular item is contaminated or unsterile following sterilization process[3].

Autoclave was invented by Charles Chamberland in 1879[4]. The name comes from greek auto- self, and latin clavis- key, a self- locking device[5].

The standard temperature and pressure of an autoclave Processes conducted at high temperatures for short time periods are preferred over lower temperatures for longer times. Some standard temperatures/ pressures employed are 115 °C/10 psi, 121 °C/ 15 psi, and 132 °C/27psi. (psi=pounds per square inch). In university autoclave, autoclaving generally involves heating in saturated steam under a pressure of approximately 15 psi, to achieve a chamber temperature of a least 121°C (250°F) but in other applications in industry, for example, other combinations of time and temperature are sometimes used. Please note that after loading and starting the autoclave, the processing time is measured after the autoclave reaches normal operating conditions of 121°C (250°F) and 15 psi pressure, NOT simply from the time you push the "on" button[6].

Need and importance

1. The proper sterilization of medical devices, surgical instruments, supplies and equipment utilized in direct patient care and surgery is a critical aspect of the modern health care delivery system and directly impacts patient safety
2. Sterilization is very important in the medical industry. Without sterilization, infections would fly around and thousands of lives would be lost. Sterilization helps to prevent the development and spread of infection.
3. In the pharmaceutical industry it used for surgical dressings, sheets, surgical and diagnostic equipment, containers, closures, aqueous injections, ophthalmic preparations and irrigation fluids etc.
4. It is generally accepted that sterility assurance level (SAL) of 10-6 is appropriate for items intended to come into contact with compromised tissue, which has lost the integrity of natural body barriers[7].

Basic Qualification approach

User requirement specification (URS)

User or customer of equipment has certain expectation about the equipment which wants to use. These expectations are generally in the form of his requirements. It is called as user requirement specifications.

Design qualification (DQ)

Design qualification may verify that design of equipment, system/facility is according to requirement of user and current good manufacturing practices.

Installation qualification (IQ)

Installation qualification is conducted to prove that equipment/system has been installed as per user and manufacturer recommendation and verifying that all required utilities have provided safe operation of equipment/system.

- Utilities specification
- Drawing specification (electrical, mechanical)
- Construction material in product contact
- Operating and maintenance manual

Operational qualification (OQ)

The operational qualification process is intended to demonstrate that the components are operating properly and ready for performance or load testing. Operational qualification shall be done “without load”

Performance qualification (PQ)

Performance qualification is documented evidence to prove that equipment/system is performing under specified condition. It involve in taking trial under “loaded condition”[8].

Qualification of equipment

Steam sterilizer sterile the article using saturated steam and equipped specifically for application in the pharmaceutical field. Sterilizer is an effective equipment to ensure the sterilization of the garment, cleaning aids

filter, utensil, vial filling, machine parts, rubber stoppers etc. The set of main activities for treating the processed materials in order to sterilize them is termed as sterilization process/cycle.

Performance of autoclave

It involve in taking trial under "loaded condition". The calibration of instruments, apparatus carried out at suitable intervals in accordance with an established written program containing specific directions, schedules, limits for accuracy and precision. Operating range, approval standard operating procedure is used for verification. Performance qualification is integrate, procedure, personal, system and material verify pharmaceutical grade utility, and environment, equipment, and system produce required output[9, 10].

Risk management

- Autoclaves are inspected every 3 months and certified annually by spore strips. The inspection, service and repair records are maintained in the lab. The name of the person responsible for the autoclave shall be posted near the autoclave.
- An authorized training session must be successfully completed by users prior to use of autoclaves.
- It is the supervisor's responsibility to ensure employees are trained before operating any autoclave unit.
- Procedural and instructional documents must be followed.
- Personal protective clothing and equipment must be worn when loading and unloading the autoclave.

Equipment to protect against scalds and burns:

- Heat-insulating gloves that provide complete coverage of hands and forearms.
- Closed-toed footwear.

Packaging and Loading

- use approved autoclave bags
- prepare and load material to ensure steam penetration
- ensure all containers including bags are vented
- do not overfill containers (prevent spill and boil over)
- ensure sufficient water in load to allow steam penetration
- use secondary containers
- label all material (name, contents)
- ensure material is permitted to be autoclave
- do not mix clean and contaminated material in the same load
- complete "Daily Autoclave Log"
- do not allow bags to touch or strap sides of autoclave

Operating an Autoclave

- ensure the autoclave is operating properly before commencing
- determine the appropriate exposure time for the load, consider the many factors effecting exposure time
- ensure the autoclave attains the desired temperature (normally 121oC) and pressure (minimum 15 psi) for the desired time (minimum 30 min.)
- record information in "Daily Autoclave Use Log"
- undertake weekly testing using a biological indicator (B. Stearothermophilus. Record results on "Biological Test Indicator Results" form.

Unloading the Autoclave

- wait until the chamber pressure gauge reads zero before opening
- wait 10 minutes for the contents of the autoclave to cool.
- remove the waste in manner reduce the risk of spillage, use a trolley
- verify temperature and duration of exposure has been met.

The different tests are follows for qualification of autoclave are

- Vacuum leak test
- Bowie-dick test
- Heat distribution study
- Heat penetration study

Vacuum leak test:

Objective:

To verify the leakage in sterilization chamber during vacuum hold time when the sterilizing chamber is empty.

Principle:

These tests are designed to show that the sterilizer chamber does not leak in empty chamber. Leakage of air into the chamber is not acceptable for two reasons:

1. The presence of air inhibits penetration of the load by the sterilant (Steam) and prevents sterilization.
2. Air leaking into the chamber during the sterilization and drying cycle are not passed through the bacteria retentive filter, and therefore there is a risk of contamination of the load.

The test is performed by measuring the change in vacuum in the chamber when all the valves leading to it have been closed and the vacuum source is isolated.

Procedure:

Ensure that the chamber temperature is stable at ambient and compressed air is on with high pressure and ensure that gasket lubrication is proper and switch provided on panel board. Start the vacuum leak rate test cycle and observe the pressure in the pressure gauge of steam sterilizer and Cycle allow the pressure to drop down. Machine will close all the valves connected to the chamber and stop the vacuum pump and note the time and pressure (P1). Wait for stabilization period of 5 minute (±10 second) and note down the pressure again (P2) and Wait for another 10 minute (± 10 second) and note down the pressure third time (P3). Return to atmospheric pressure and continue to run for next cycle where vacuum leak rate should not be more than acceptance criteria.

Acceptance criteria:

Vacuum leak rate should be NMT 0.013 bar / 10 minutes.

Frequency:

Run the test for 3 consecutive cycles at the time of Initial Qualification.

Bowie- Dick test:

Objective:

To ensure that the vacuum pulses applied before the sterilization hold period are sufficient to remove the entrapped air or non-condensable gases so as to facilitate the event and rapid steam penetration into all parts of load and maintaining this condition during sterilization holding time.

Principle:

Sterilization is achieved by the rapid and even penetration of steam into all parts of the load and the maintenance of these conditions for the specified holding time. To ensure this, it is essential to remove air from the chamber and load, and to provide a steam supply which contains a minimal volume of Non-condensable gases .The Bowie Dick test shows whether or not steam penetration is taking place by testing the presence of

Non condensable gases in the chamber, but it does not confirm that the sterilization condition in the load is achieved or not.

Procedure:

Place the Bowie Dick test paper on the bottom shelf of the sterilizer just above drain point (100mm over the drain) Air removal study shall be performed in empty chamber by placing the Bowie Dick test paper. it consist of standard paper pack and indicator sheet Start the cycle by pressing enter key After the cycle is over open the door from control area side and take the sterilized test paper from the autoclave and check the indicator paper for uniform colour change As Bowie Dick test paper is designed to simulate the garment pack, it used to test the efficiency of the air removal from the steam sterilizer Three cycle of air removal study shall be performed (initially) by using fresh indicator paper This test shall be performed by using Bowie Dick test cycle. To fulfil the maximum exposure requirement, the sterilization cycle shall have 17 minutes at 121°C to 123°C sterilization period.

Set Parameters:

17 minutes cycle at a temperature of 121°C

Place of keeping Bowie -Dick Indicator:

Place the Bowie dick indicator approximately 100 mm to 200 mm above the sterilization chamber base.

Acceptance criteria:

The Bowie dick indicator should show uniform color change (Yellow to Brown / black) after the cycle. No change or no uniform change or air entrapment (bubbles) spot on the test sheet indicates inadequate air removal from the sterilization base chamber.

Frequency:

Run the test for 3 consecutive cycles at the time of Initial Qualification.

Heat distribution study (empty chamber)

Objective:

To verify the temperature uniformity throughout the chamber and to locate the cold spot in Empty Chamber.

The sterilizer is capable attaining a temperature of 121° C throughout the sterilizing hold period in Empty Chamber.

Procedure:

Insert 16 no of temperature sensors inside the chamber through the validation port of sterilizer. Seal the port with silicon sealant to ensure that no steam leakage during operations of sterilizer. Fix all the probe at different location in the sterilizer so that sensors do not touch the metallic surface of the chamber. Connect the temperature sensors to the data logger, which can scan and print the actual temperature and pressure at different locations.

After completion of sterilization cycle check the thermograph in the data logger for attaining set temperature and pressure during the sterilizing hold period. If any deviation observed repeat the cycle after taking necessary correction.

Acceptance criteria:

Temperature distribution within the chamber must be between 121°C to 123°C at all location during the sterilization period (dwell time)

There should not be any slowest heating point (cold spot) in the autoclave chamber and equilibrium time should not be more than 30 second.

Heat distribution study (loaded chamber)

Objective:

To verify the temperature uniformity throughout the chamber and to locate the cold spot in loaded Chamber.

The sterilizer is capable attaining a temperature of 121°C throughout the sterilizing hold period in loaded Chamber.

Procedure:

Insert 16 no of temperature sensors inside the chamber through the validation port of sterilizer. Seal the port with silicon sealant to ensure that no steam leakage during operations of sterilizer. Fix all the probe at different location in the sterilizer so that sensors do not touch the metallic surface of the chamber. Load the article as per loading pattern in the autoclave chamber. Loaded chamber heat distribution study shall be performed separately for all loading patterns. One cycle shall be performed for each loading type for loaded chamber heat distribution study Connect the temperature sensors to the data logger, which can scan and print the actual temperature and pressure at different locations.

After completion of sterilization cycle check the thermograph in the data logger for attaining set temperature and pressure during the sterilizing hold period. If any deviation observed repeat the cycle after taking necessary correction.

Acceptance criteria:

Temperature distribution within the chamber must be between 121°C to 123°C at all location during the sterilization period (dwell time)

There should not be any slowest heating point (cold spot) in the autoclave chamber and equilibrium time should not be more than 30 second.

Heat penetration study

Objective

In order to verify sterilizing temperature has been reached in each load subjected to moist heat sterilization, it is necessary to conduct heat penetration studies. This study is conducted to ensure that the coolest unit within a pre-defined loading pattern (including minimum and maximum loads) will consistently be exposed to sufficient heat lethality (minimum "F0").

Procedure:

Insert 16 no of temperature sensors inside the chamber through the validation port of sterilizer. Seal the port with silicon sealant to ensure that no steam leakage during operations of sterilizer. Fix all the probe at different location in the sterilizer so that sensors do not touch the metallic surface of the chamber. Load the article as per loading pattern in the autoclave chamber. Loaded chamber heat distribution study shall be performed separately for all loading patterns. Arrange the load as specified keep at least 15 biological indicators and 10 thermo chemical indicators shall be used for each cycle. Load placed at the identified cold points must have indicator and temperature sensor in all three runs. One biological indicator and thermo chemical indicator along with external temperature sensor shall be placed at drain point in all three cycle. Perform the sterilization by operating the program specified for each load type as per standard operating procedure and start the data logger and steam sterilizer simultaneously. After the sterilization cycle is completed, stop data logger and open the sterilizer and take out the biological indicator and thermo chemical indicator from load and send to microbiological lab for testing. The biological indicator shall aseptically inoculate into sterile soybean casein digest media (SCDM) and incubated at 55 - 60^{o}C and liquid load at 35 - 39^{o}C for 7 days and check the thermo

chemical indicator for the compliance as per manufacturer recommendation for colour change (i.e. Brown). Take out temperature chart/data logger and inbuilt temperature recorder, report of biological indicator. Check against acceptance criteria for compliance and determine the *Fo* value and compare against acceptance criteria. Take out external temperature sensor from the chamber and perform vacuum leak rate test.

Acceptance criteria

Temperature distribution within the chamber must be between 121°C to 123°C at all location during the sterilization period (dwell time)

Sterilization temperature should be maintained for NLT 15 minute for minimum 10 thermo couple during hold time. Biological indicator (Geobacillus Stearothermophillus) should show complete sterilization (i.e. no growth after incubation) [11-14].

Conclusion

Qualification is a fundamental concept of cGMP. Where autoclave is used for sterilization of the garments, cleaning aids filters, utensil, vial filling machine parts, rubber stopper etc. This was followed by performing the qualification of the equipment which describes the entire test right from vacuum leak test, bowie-dick test, heat distribution study (empty chamber, loaded chamber) and heat penetration test. All the parameters and processes which are described were found within the acceptance criteria. Hence autoclave is considered to be qualified and can be routinely used.

References

1. Jacquelyn G B, Microbiology: principles and applications, Englewood cliffe, N. J: prentice hall 1993.
2. Sultana Y. Pharmaceutical Microbiology and Biotechnology, Sterilization Method and Principle, Jamia Hamdard, Hamdard Nagar, New Delhi, 2007.
3. American Society for Healthcare Central Service Professionals, Training Manual for Central Service Technicians, 2006.fifth ed., pp.25-27.
4. Fernbach E, Joubert, Roux E, Pasteur E, Straurs, Charles chamberland, En collaboration arec, 1851-1908.
5. Online etymology dictionary, available at www.etymonline.com
6. Seymour S B, Disinfection, Sterilization and preservation, Lippincott Williams and wilkins, ISBN 978-0-683-30740-5, 2001.
7. Thomas C., John P.J., Principles and Methods of Sterilization in Health Sciences, second ed., 1983.
8. Fabritz H, Autoclave Qualification and Validation, Expert reff 2007-14
9. Validation of Steam Sterilization Cycles, Technical Monograph No.1, Parenteral Drug Association, Inc., Philadelphia, pp.5-6.
10. European Committee for Standardization, Sterilization of Medical-Devices Validation and Control of Sterilization by Moist Heat Sterilization, EN 554:1994. pp. 264-269.
11. Guy D., Hodges N., and Hanlon G., 2003. Endotoxins and Depyrogenation in Industrial Pharmaceutical Microbiology, Standards and Controls, Euromed, pp.12.1 - 12.15
12. Akers M.J., 1994. Parenteral Quality Control, Sterility, Pyrogen, Particulate Matter and Package Integrity Testing, second ed. Marcel Dekker, New York, pp.1-4
13. Sigwart V., Stark A., 2003. Effect of Carrier Materials on the Resistance of Spores of Bacillus stearothermophilus. PDA Journal of Pharmaceutical Science and Technology,
14. E.U. Guidelines to Good Manufacturing Practice, 2008. Manufacture of Sterile Medicinal Products, pp.1-16.

*****