Minireview

Validation Study and Routine Control Monitoring of Moist Heat Sterilization Procedures

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The proposed approach to validation of steam sterilization in autoclaves follows the basic life cycle concepts applicable to all validation programs. Understand the function of sterilization process, develop and understand the cycles to carry out the process, and define a suitable test or series of tests to confirm that the function of the process is suitably ensured by the structure provided. Sterilization of product and components and parts that come in direct contact with sterilized product is the most critical of pharmaceutical processes. Consequently, this process requires a most rigorous and detailed approach to validation. An understanding of the process requires a basic understanding of microbial death, the parameters that facilitate that death, the accepted definition of sterility, and the relationship between the definition and sterilization parameters. Autoclaves and support systems need to be designed, installed, and qualified in a manner that ensures their continued reliability. Lastly, the test program must be complete and definitive. In this paper, in addition to validation study, documentation of IQ, OQ and PQ concretely were described.

Key words: Moist heat sterilization / Validation study / IQ / OQ / PQ.

INTRODUCTION

Installation qualification (IQ), operational qualification (OQ) and performance qualification (PQ) tests are completed during the validation process to demonstrate that the sterilizer has been installed and provided with utilities as directed by the specifications. Testing performed during IQ/OQ/PQ is conducted to demonstrate that the system is capable of delivering a sterilization process that is consistently able to meet predetermined criteria and that it is efficacious and in a high level state of control (Agalloco and Carleton, 2008; Block, 2001; Carlton, 1998; Clotz, 2009; Denyer and Baird, 2007; Fraisse, et al., 2004; Gardner and Peel, 1998; Hallis, 1994; ISO 14161, 2010; McDonnell, 2007; Nash and Wachter, 2003; Saghee, et al., 2011; Shintani, 2011). The following should be completed prior to the initiation of the validation: process definition of the product and associated loading patterns, identification of the biological indicator (BI) organism and associated performance attributes, identification of the process challenge devices (Gardner and Peel, 1998) or master solutions, demonstration of sterilization process compatibility for the products or items to be sterilized, the development of the sterilization process and optimal parameter operating ranges, identification of key and critical parameters, and preliminary biological lethality assessments. If it is more practical to complete the elements of validation in a sequence that is different than the traditional IQ/OQ/PQ, and this is considered acceptable as long as all validation activities are completed for the final product, final equipment configuration, and final process prior to the manufacture of commercial product.

Installation Qualification (IQ)

The procedures for installation qualification (IQ) must be defined. A requirements definition should be developed to document the proposed design requirements for the sterilizer, associated instrumentation and utility interfaces. The IQ is completed by
reviewing the installed state of the equipment and documenting that the equipment has been provided and installed in accordance with its specifications. The installed chamber and associated utilities should be compared to the requirements, definition and associated drawings to ensure that these match.

The utilities for the sterilizer should also meet following requirements:

**Steam**
Various types of steam are used for moist heat sterilization applications including plant steam, process steam and pure steam.

**Sterilizer Fill/Cooling Water**
Water quality must be consistent with steam requirements for each of the applications. The level of microorganisms in the equipment fill and cooling water should be controlled and monitored. The sterilizer fill and/or cooling water should not offer an opportunity for the recontamination of the sterilized product. This is not an issue with sealed liquid containers. For products with the potential to be exposed to fill/cooling water, a closed system approach is employed for the sterilizer in which the fill/cooling water is sterilized with and receives the same heat exposure as the product.

**Compressed Air**
Process air should be dry and free from oil for use with air overpressure sterilization processes.

**Air Used for Vacuum Break for Porous/Hard Goods**
For porous/hard goods loads sterilized in the pre-vacuum process, the air must be filtered since it comes in contact with sterilized goods. In cases where packaging is not an absolute microbial barrier, a comprehensive filtration program must be employed that requires a microbial-retentive air filter and associated integrity testing.

**Operational Qualification (OQ)**
The procedures for operational qualification must be defined. The OQ phase of the validation process includes verification that all installed equipment is operating properly and within predetermined limits when used by trained personnel in accordance with operating procedures. The OQ includes formal test procedures or inspection steps that should be performed at this stage of the validation. However, many of these evaluations should be adopted for subsequent use during routine monitoring and also used to demonstrate the continuous high-level control of the sterilization process during routine operations. This part summarizes tests that are specific to pre-vacuum and/or saturated steam sterilization processes followed by general considerations in support of OQ that are applicable to all sterilization process types.

**Pre-vacuum Sterilizer Vacuum Leak Rate Test**
This test is performed only on pre-vacuum sterilizers to demonstrate the integrity of the sterilizer chamber is integral and that there is no risk of air leakage that could affect the sterilization process during the phases of the cycle that operate below atmospheric pressure. The use of positive pulses can be used to partially mitigate this risk to the efficiencies of the sterilization process. The vacuum leak test is performed on an empty chamber by creating vacuum conditions in the chamber to a level that is consistent with the levels utilized during the pre-vacuum conditioning phases of the cycle. This is followed by a five to ten minute stabilization phase that allows any water present to vaporize and the chamber conditions to reach equilibrium. The hold phase is then initiated for the required five or ten minute hold time. The difference in chamber pressure from the beginning of the hold is subtracted from the chamber pressure at the end of the hold with the absolute value of that difference divided by the time of the hold phase in minutes to determine the leak rate. The leak rate from the chamber is compared to a defined limit. The leak rate test is performed regularly (weekly or monthly) based on historical data but also must be performed any time that the chamber integrity has been potentially breached (e.g., door gasket replacement, wired thermocouple installation, replacement of check valves on exhaust piping, etc.). In addition to the examples just presented, vacuum leak failures have also been attributed to leaky piping connections and check valve malfunctions.

**Steam Penetration or the Bowie Dick Type Test**
The steam penetration test is conducted in an empty chamber and is applicable only to pre-vacuum sterilization chambers since it measures air removal and steam penetration. This test can be performed on a daily basis or on a weekly basis usually at the start of the week, if applicable, as a minimum requirement. The standard test configuration contains porous filler materials such as linen or cellulose-based paper with a Chemical indicator (CI) in the hardest to penetrate section of the test system (Gardner and Peel, 1998). The filler materials contain air which must be removed by the sterilization process in order for effective steam penetration to occur. Disposable and reusable items of this test system are available from
most sterilization indicator manufacturers. Evidence of effective steam penetration is indicated based on CI color change (Fig. 1, Gardner and Peel, 1998). If an incomplete color change occurs, the most probable reason for the inadequate steam penetration will be the incomplete air removal from the porous materials in the test system. The most common causes for this condition is an inadequate number of pre-vacuum pulses, vacuum evacuation levels that are not deep enough or performance in a chamber that has a leak or that has not been properly "warmed up". Addition of pulses or evacuation to deeper levels should increase the effectiveness of the air removal and resulting steam penetration. Performing a test cycle each time after startup or the elimination of leaks of the sterilizer may alleviate this condition. However, if a test cycle is performed as a prerequisite for conducting the steam penetration test, the test cycle must also be performed prior to sterilizing the product or items on days that the steam penetration test is not performed.

**Steam Quality Tests**

Steam quality tests are conducted only for saturated steam sterilization processes and are based on the physical and not the chemical attributes of steam. These tests include procedures for determination of non-condensable gasses in steam, the dryness fraction of steam, and presence of superheat in steam. Steam quality tests may be completed on an annual basis if supported by a successful history. General description and guidance will be provided for these tests. More detailed information especially on test procedures and limits for steam quality testing is reported by Shuttleworth (2000).

Non-condensable gasses that are entrained in the steam supply can act like air in the sterilization chamber by insulating the contents of the load from sterilizing conditions and lead to inadequate physical and biological lethality. In addition to testing the steam supply near the use point at the sterilizer, the presence of non-condensable gasses may also be detected by the steam penetration test. The most common cause of non-condensable gasses in the steam supply is insufficient deaeration of the boiler feed water. Boiler feed water should be deaerated using the highest temperature possible since this minimizes the amount of dissolved gasses in the feed water.

The dryness fraction of steam is a measurement that describes the ratio of dry saturated steam to water in a steam supply. The greatest amount of latent
heat and associated sterilization efficiency is contained in 100% saturated steam. As the amount of entrained water in the steam increases, the amount of latent heat decreases and the likelihood of having a wet product at the end of the processing is also increased.

Superheated steam is steam that is present at a temperature that is significantly greater than values listed for specific saturated steam pressures on steam tables. The implications of superheated steam are confined to saturated steam processes. Superheated steam will not condense and provide energy and moisture at the same rate as saturated steam. Superheat can be detected by comparing simultaneous temperature and pressure readings to the steam tables. Additionally, superheat may also be the cause for unexpected survival levels of BIs that are detected in a sterilization process run that delivers adequate physical lethality. The behavior of superheated steam more closely resembles dry heat. Common causes of superheat include a jacket temperature set higher than the exposure temperature, steam supply to the chamber that is stepped down at a ratio of greater than 2:1, and the processing of excessively dried linens or cellulosic materials that contain low levels of residual moisture.

The OQ should also include the following general tests, procedures and evaluations:

1) Calibration of sterilizer operating and test instruments, using instruments traceable to a recognized national or international standards laboratories
2) Software validation
3) Initiation of a preventive maintenance program
4) Confirmation that safety systems operate as designed
5) Establishment of maintenance requirements and operating procedures
6) Evaluation to demonstrate that the requirements of each the following are met:
   6)-1 Absence of leakage for utility inputs
   6)-2 Sterilizer water purity limits (applicable if there is potential contact with the product)
   6)-3 Steam chemical limits (applicable if there is potential contact with the product)
   6)-4 Compressed air limits (physical and microbiological)
   6)-5 Verification of compliance with temperature and pressure relationships for steam within defined limits based on steam tables to support the presence of saturated steam (saturated steam processes only)

Empty chamber temperature distribution studies should also be conducted to demonstrate uniformity of the sterilant. If temperature distribution studies indicate that the distribution of the sterilant is non-uniform, corrective action must be taken or the presence of cold spots must be addressed in the design of development and qualification studies. The results from the empty chamber studies may also be used to demonstrate that the sterilizer is capable of operating within parameter limits although additional testing with a loaded chamber is still necessary. Although not required, it is recommended that temperature distribution studies be conducted during OQ or early in PQ using the range of chamber loading that will be utilized in routine production to demonstrate that the presence of a load in the chamber does not affect the distribution of sterilizing medium in the chamber.

**Performance Qualification (PQ)**

The PQ phase includes studies to verify that the sterilization equipment can reproducibly deliver a specified process that achieves the desired lethality and SAL for the entire load of the product. The qualification of the moist heat sterilization process is accomplished through the use of combination studies (minimum of 3 consecutive studies per ISO 17665-1, 2006) that utilize both physical and biological measurements simultaneously to confirm that the process is reproducible and can reliably meet efficacy requirements.

Physical measurements include sterilizer instrumentation used to demonstrate the achievement of process parameters within required ranges, temperature distribution probes to demonstrate uniform distribution of the heating medium, and heat penetration probes to generate temperature data that can be used to assess the heat history and physical lethality delivered to the load. BIs are also placed into the product and items, and used to assess the biological lethality delivered to the load. Inoculated BI test articles are positioned immediately adjacent to test articles containing temperature penetration probes whenever possible to provide for the ability to confirm agreement between physical and biological lethality. Penetration probes and BIs are placed in the hardest-to-sterilize locations (cold spots) of products or items that represent the greatest challenge to the sterilization process. Multiple BI test articles may be positioned next to each penetration temperature probe to provide for increased detection sensitivity and more accurate determination of survivorship levels determination in support of biological lethality. In some cases where demonstration of sterility is required but heat penetration probes cannot be used due to size, BIs must be utilized exclusively to
support that the sterilization process delivers a SAL $\leq 10^6$.

If no cold spots are identified during OQ, temperature distribution probes should be positioned randomly and/or geometrically in the load. If cold spots are detected and not corrected, temperature distribution probes should be positioned to focus on monitoring any cold spots. Any available probes that are not positioned in cold-spots should be randomly and/or geometrically positioned in the sterilizer. Limits must be specified for the individual temperature measurements and temperature ranges at key time points or cycle phases during the performance of the sterilization process. Since the exposure phase is the most highly controlled phase of the sterilization process, it is common to closely monitor temperature distribution measurements during this phase.

Temperature penetration probes are positioned in or on the product or items to measure the penetration temperature that is used to calculate the physical lethality delivered. The hardest-to-sterilize locations (cold spots) for a process challenge device or for a solution container is used as the placement location for temperature penetration probes. Care must be taken during the placement of penetration probes to ensure that the sensing element remains in the defined cold spot for the duration of the sterilization process. Requirements for minimum and/or maximum penetration temperature, physical lethality, equilibration time (for porous hard products only), and time at a specific temperature are typically specified for penetration temperature probes. Additionally, the physical lethality data for the sterilization process should be used to demonstrate that the SAL is $\leq 10^6$.

Similar to the use of temperature distribution probes, the measurements from temperature penetration probes are closely evaluated during the exposure phase and also during the heating and or cooling phases for physical lethality data from these phases are used to demonstrate the achievement of minimum requirements.

Bls should be placed in the same test article as temperature penetration probes. In addition, Bl organisms are inoculated into master solutions or onto carriers (including commercially available paper strips) that are placed in the hardest-to-sterilize areas (cold spots) in a process challenge device for porous/hard goods or into closure systems of liquid product container systems. In the case of solutions in containers, it is common to inject a suspension of the Bl into the container through an open port prior to sealing the port. In this instance, it is not possible to locate the entire population of the Bl in the defined cold spot for the container. However, this inoculation approach is valid because the dispersion of the Bl in the solution mimics the dispersion of natural solution bioburden in the solution. Typical specified limits for Bl test articles include maximum survivorship levels, minimum spore log reduction levels, and the requirement that a SAL of $\leq 10^6$ is achieved based on biological lethality determination.

**Use of Physical Lethality in the Qualification of Moist Heat Sterilization Processes**

Physical lethality data gathered from penetration test articles are used to evaluate whether the sterilization process has met predefined requirements. In addition to demonstrating that physical lethality requirements have been met, physical lethality data can also be used to determine the SAL of the sterilization process. The SAL is calculated using the semilog survivor curve along with physical lethality data from studies and assumptions for the product bioburden population and resistance that were made during cycle design.

For example, if a moist heat sterilization process delivered a minimum physical lethality of $F_0 = 12$ minutes (using the coldest or slowest to heat probe in a qualification study run) to the center of a tubing open at both ends in a porous/hard good load, the SAL for this overkill process can be calculated using the attributes of an assumed product bioburden of $1 \times 10^6$ CFU microorganisms/carrier with a $D_{121.1^\circ C}$ value of 1.0 minute and a z-value of $10^\circ C$ (ISO 17665, 2006).

It is important to reiterate that physical data should be used to qualify the efficacy of a sterilization process since heat penetration probes are capable of being employed in all locations required to be sterile and to confirm that saturated steam conditions are present. Physical lethality data should be used to complement biological lethality data and vice versa in the support of the qualification of moist heat sterilization processes.

**Use of Biological lethality in the Qualification of Moist Heat Sterilization Processes**

Biological lethality data should be used along with physical lethality data to support process efficacy for moist heat sterilization processes. Qualification is performed to demonstrate that the biological lethality delivered by the sterilization process is able to meet the minimum lethality requirements set during the cycle development.

In order for a sterilization process to be considered an overkill process, the $F_0$ and $F_{bio}$ must both be greater than or equal to 12 minutes. In the example provided below, a $121.1^\circ C$ ($250^\circ F$) overkill process achieved an exposure $F_0$ of 12 minutes that
inactivated *Geobacillus stearothermophilus* ATCC 7953, BI organisms (D$_{121\,\text{°C}}$ = 1.5-2.5 minutes, z = 10 °C, ISO 11138-3, 2006) from an initial population of 1.0 X 10$^6$ CFU/carrier to SAL of 10$^6$ or more. The combination of physical and biological data gathered during the performance of this study can be evaluated to determine if this process meets the requirements of an overkill process.

**ROUTINE CONTROL OF THE MOIST HEAT STERILIZATION PROCEDURE**

After successful development and validation of the moist heat sterilization process along with proper regulatory approval, routine sterilization of items or products can be initiated in the support of the manufacture of sterile products. In addition to ensuring that critical parameters are met to support the sterile product release, additional elements of the sterilization program are employed including preventive maintenance, calibration, sterilizer functionality testing, re-qualification of the sterilization process and product and process change control.

**Preventive and Unplanned Maintenance**

A program for application of preventive maintenance is initiated during OQ to ensure that the sterilizer and associated utility systems are capable of consistently providing a reliable and high level of process control for the sterilization process. The manufacturer of the sterilizer should be consulted about the frequency at which parts that are expected to be adversely affected by wear over time should be replaced. The deviation patterns and rate for critical and key sterilization process parameters should also be utilized to supplement the guidance provided by the sterilizer manufacturer.

Unplanned maintenance of the sterilizer and utility systems occurs whenever there is a malfunction or breakdown of these systems. Thorough investigation is critical and can be instigated by the sterilizer’s inability to meet key and critical parameters, detection of unfavorable trends in the performance of the sterilization process or due to the failure or near failure of re-qualification studies. When the assignable cause is identified, it is immediately remedied to prevent further reoccurrence of the situation.

Upon completion of the preventive or unplanned maintenance, demonstration that the sterilizer and associated utilities have been returned to the previously validated state is essential. A change control system for the validated system must be employed that identifies the reason for the maintenance and summarizes the work performed or corrective action applied to resolve the situation. Testing procedures must be developed and results documented to demonstrate that the system has been returned to the previously validated state upon completion of maintenance. In cases where modification of the sterilizer or utilities is necessary, some or all of the original moist heat process validation tests may need to be re-performed. In general, replacement of parts by those that resemble may not require full validation repeatedly and may only require the completion of a successful test cycle that includes demonstration that parameter tolerances for all phases of the routine moist heat sterilization process can be met.

**Calibration Program**

A program for the calibration of the sterilizer and utility instrumentation is required to ensure the performance of a consistent and efficacious sterilization process. The calibration program must include the following for each Instrument:

1) Unique reference number
2) Calibration procedure
3) Precision and accuracy requirements
4) Frequency that calibration is performed
5) Calibration tolerance limits
6) Actions to be taken when tolerance limits are not met

**Ongoing Sterilizer and Utility System Functionality Tests**

There should be a comprehensive program that regularly confirms the sterilizer and utility system functionality to support routine sterilization operations. This program is initiated during the OQ stage of validation but is also designed to provide ongoing assurance that the sterilizer is properly functioning at a high state of control and serves as a supplement to re-qualification studies. Results from ongoing monitoring should also serve as an input into the support for the sterile product release.

**PRODUCT AND PROCESS CHANGE CONTROL**

In addition to the change control considerations discussed for the sterilizer and associated utility systems, the development and deployment of a change control system for the product and sterilization process is also required to support routine sterilization operations. The change control system for products and processes must be administered by individual(s) with demonstrated competency in sterilization engineering and sterilization microbiology in concurrence with organizations that set quality standards.
Changes to the validated product may include changes to the configuration size or geometry, chemical formulation, materials of construction, container/closure systems, or packaging systems. The change control system must evaluate the effect of the change on the validated state and sterilization efficacy achieved by the process for the affected products. In instances where the change results in a sterilization challenge that is unknown or greater than the current validation tests can support, some or all of the previous validation studies must be performed to support adoption of the change. In cases where there has been investment in a very robust development effort to support anticipated changes as with the process challenge device approach for porous/hard goods products or Master Solution Approach for liquid products, adoption of the change can be supported through the drafting of a scientifically sound rationale with minimal, if any, sterilization studies required to support the change.

**STERILE PRODUCT RELEASE PROCESS**

The sterile product release process is developed to support the release of sterile products in the commercial marketplace or the release of sterile items for use as inputs for aseptic processing. Sterile product release of terminally sterilized finished products is based on the achievement of critical and required key sterilization cycle parameters and the successful results from a sterility test. A successful sterility test is not required for the release of items as intermediates for the aseptic process. Sterile product release is the result of the culmination of outputs from all of the previous descriptions that comprise the comprehensive and science-based moist heat sterilization program.

The following must be considered in the sterile disposition of a product that was processed with a moist heat sterilization process:

1. Product bioburden meets limits (where applicable)
2. The sterilizer was operating properly as designed
3. All sterilization cycle parameters required to achieve sterility were met
4. Required results for chemical and/or BIs were achieved
5. Sterility test results, if applicable, were acceptable
6. Appropriate SAL (i.e., 10⁶) is achieved

**Bioburden Testing**

Depending upon the cycle design approach utilized and amount of bioburden history gathered, it may be a requirement that bioburden levels be considered as part of the sterile product release process. When the overkill cycle design approach is used, bioburden test process may not always be required to support the sterile product release. Conversely, when the product specific design approach is used, bioburden test results may be required to support the sterile product release. However, bioburden testing results may not be required for release with the product specific design approach when there is a large safety margin with the sterilization process along with a substantial history in all seasons that supports that the bioburden is consistently low in number with a low occurrence of heat resistant spores with resistance levels that are always much lower than that of BI used to qualify the moist heat sterilization process. Since the identical pre-sterilization portion of the manufacturing process is often used for a family of products or items, it is possible to leverage the successful existing bioburden information from an existing product family to cover new products or items that will be added to the family and manufactured under the existing process.

The bioburden test procedure should be validated in a similar way to the sterility test by demonstrating that the recovery of targeted microorganisms, mainly bacteria spores, is not significantly affected by the solution or item being tested. Since moist heat resistant spores are the focus of this testing effort, a heat shock to screen out vegetative organisms is incorporated prior to plating using an adaptation of the total count testing procedure. To provide for the selective recovery of moist heat resistant spores, heat shock temperatures may range from 80 to 100 °C with associated exposure times of 10 to 30 minutes. The resistance of any spores recovered from heat shock testing is further characterized with additional heat screening procedures and D_{217°C} value determination. Finally, the population level and resistance of spores recovered from the bioburden is compared to that for the BI used to validate the sterilization process. The load must be rejected if the sterilization challenge for the bioburden exceeds that for the BI. Normally the BI utilized is *Geobacillus stearothermophilus* ATCC 7953 with 1x10⁶ CFU inoculated onto the paper (ISO 11138-1,3, 2006).

**Sterilizer Functionality**

Preventive maintenance, calibration, and sterilizer functionality tests must be conducted according to procedure and schedule to support sterile release. Some of the tests that were performed during OQ should also be performed at periodic intervals to support routine operations. Verification for compliance to this can be provided with a confirmation at the time of
release or through the use of systems designed to reliably assure compliance.

Sterilization Cycle Parameters
A thorough manual review performed independently by multiple numbers of trained individuals, a validated electronic system review or a combination of these may be used to assess compliance of a sterilization cycle run with critical and key sterilization cycle parameters. Similar to change control, individual(s) with competency in sterilization engineering and sterilization microbiology should provide confirmation regarding sterility when sterilization cycle parameter deviations occur. In order to support the sterile product release, all critical sterilization cycle parameters must be met. Although it is not required that all key sterilization cycle parameters be met for release, a scientific rationale that supports the sterile product release must be developed for situations in which key parameters have not been met. In either case, an investigation should be performed to identify the assignable cause when critical or key parameters are not met. The investigation should include immediate corrective actions taken, a scientific rationale for product disposition, and actions taken to prevent reoccurrence.

CI and BI Results
Depending on requirements, acceptable CI and/or BI results may be required to support the sterile product release. The use of Class I of CI results is not approved in validation studies, but that of class 4 to 5 of CI may or may not be considered in validation studies as well as in routine control procedures. Although CIs cannot provide absolute confirmation of sterility, CIs can also be used as an effective segregation tool that provides visual evidence of whether a load of the product has been exposed to the sterilization process. BIIs of 10⁶ CFU/carryer must be used in validation studies to attain a SAL of 10⁶. In a routine control process to support the release of sterilized products, when there are significant benefits due to segregation and control in a product load and there is complete BI inactivation. BI does always provide demonstration of the required SAL of ≤10⁶.

The Sterility Test
Sterility tests must be validated according to local pharmacopoeial requirements. A successful sterility test is not always required to support the release of sterile items used for aseptic processing. Although significantly flawed in terms of sensitivity in the accurate support of the sterility for a load of the product, completion of a successful sterility test is a common regulatory requirement to support the release of moist heat terminally sterilized pharmaceutical products. Based on calculations performed by the author, the sterility test will give positive sterility test results only 18% of the time when testing a sample set of 20 samples that have a contamination rate of 1/10 (SAL = 10⁵). In summary, the sterility test provides little value in assessing product sterility even when the bioburden survivorship level is four orders of magnitude greater (SAL=10⁻⁴ vs. required SAL= 10⁻⁶) than the requirement for sterility. Therefore, the sterility of a load of products can only be supported by the comprehensive science-based sterilization program from development through validation, qualification and routine operations. This last statement serves as the foundation for a parametric release moist heat sterilization program.

Parametric Release
Parametric release is a sterility release program based on effective control of a validated sterile product manufacturing process where the product release is based on demonstrated achievement of parameters required to achieve sterility in lieu of end-product sterility testing. Although regulatory approval is required for parametric release without reliance on the sterility test, it is apparent that parametric release is practiced than indicated by the number of approved submissions for parametric release. It can be noted that parametric release is practiced as the exclusive release mechanism for items sterilized as intermediates for use in an aseptic process. With the realization above regarding the shortcomings of the sterility test in assessment of sterility, it would be most accurate to state that the development of a moist heat sterilization program based on scientifically sound principles provides the best possible approach for assurance of sterility. It is on that basis that the author considers the importance of the development of parametric release sterilization programs over programs emphasizing the conventional sterility test for moist heat sterilization processes.

Various guides, pharmacopoeial references, and regulatory requirement documents (PIC/S, 2007) describe approaches that are used for moist heat parametric release programs. Many of concepts and practices highlighted in the references (PIC/S, 2007) have also been included.

CONCLUSION
A robust moist heat sterilization program can only be developed based on a solid foundation of sterilization science. Sterilization engineering and sterilization
microbiological principles must be utilized in a complementary fashion to form the backbone and framework for the design, qualification, and routine operation of the moist heat sterilization program. Specific product attributes serve as inputs to ensure the proper selection of the sterilization cycle design approach, sterilization cycle type, and sterilization cycle parameters. The output of this effort is a moist heat sterilization process that is efficacious and capable of reliably producing products that are sterilized to a SAL of $\leq 10^{-6}$ with the assurance that product stability, product functionality and microbial barrier properties are maintained over the shelf-life of the product. The true value of this endeavor is ultimately realized with the use of sterile products to save and sustain the lives of the patients that we serve.

**Example of an outline for an Installation Qualification protocol**

1. Purpose: The purpose for which the protocol was written should be stated.
2. Scope: Describe the equipment and support equipment and utilities that have been installed and will be tested.
3. References: Provide a list of all regulatory standards, equipment drawings, process and instrument drawings, process specifications and operating procedures that were used to develop the protocol.
4. Responsibilities: Define what portion of the installation testing for which the vendor of the sterilizer will be responsible and what the owner will be responsible to conduct.
5. Equipment and Materials: Provide a list of equipment and utilities that are to be tested and a list of all equipment that will be used to perform the testing.
6. Procedure: This section should address the following;
   a) Verification that the all components that were received are in agreement with the purchase specifications and the bill of materials.
   b) The P&IDs reflect the sterilizer as it has been installed; the drawings should be updated to reflect the installed sterilizer.
   c) Verification that the electrical components are correctly installed in accordance with all electrical drawings.
   d) Verification that all utilities are properly installed.
   e) Calibration of all controlling and monitoring equipment.
   f) Verification of the version of software that is being used to control the sterilizer. A backup copy of the software should be archived.
   g) Verification of all control input and output and testing of the human machine interface.
   h) Testing of all alarms
7. Discrepancies: All discrepancies should be documented along with any design changes or corrections that were made as a result of the discrepancy.
8. Completion Activities: Define the completion activities such as completing IQ, updating equipment drawings and approval of IQ report.

**Example of an outline for Operational and Performance Qualification protocols.**

The contents should be modified as necessary for the type of sterilizer being used, the sterilization process being assessed and whether OQ or PQ is to be conducted.

1. Purpose: The purpose for which the protocol was written should be stated.
2. Scope: Describe the equipment and/or products will be qualified for sterilization after completion of the testing.
3. References: Provide a list of all regulatory standards, validation documents, process specifications and operating procedures that were used to develop the protocol or to operate the sterilization equipment.
4. Responsibilities: Define who is responsible to assure compliance with the protocol, who is responsible to perform the test runs, who is responsible for calibration of the test equipment, who is responsible for testing of the microbial challenge samples, who will write the completion report and who will review and approve the report.
5. Equipment and Materials: List the equipment that will be tested, the equipment that will be used to obtain thermal, pressure, speed or other test data, the product/load that will be tested, the microbial challenge to be used and other incidental materials.
6. Calibration: Describe how the equipment that will be used in the qualification runs will be calibrated in routine in-house calibration, external laboratory certification or calibration at time of use. Provide the calibration temperatures for the thermal monitoring equipment as well as any software setup requirements to calculate instantaneous minimum, maximum, average and differential temperatures as well as calculation of any $F_0$ values.
7. Strategy/Rationale: This section should address the rationale used for selecting the master product that will be used in the qualification runs, the rationale behind selection of the product load
configuration(s) that will be used for the qualification runs and the method that will be used to demonstrate that the production cycle will be capable of providing the necessary SAL.

8. Procedure: This section should describe the procedures for preparation, storage and testing of the biological test samples. There should be diagrams to show the placement of the test samples and the temperature sensing devices in each of the load configurations that are to be tested. Any special test sample handling procedures should be documented.

The process parameters, such as times, temperatures, pressures, belt speed that are to be used in the qualification runs should be documented. References should be made to the equipment operation procedure and to settings of all other process settings that are not affected. All alarm settings that need to be changed as a result of the parameters used for the qualification runs should be specified. If $F_0$ is to be calculated from the temperature data, then the reference temperature, the $Z$-value and the temperature interval for which it is to be calculated should be specified.

This section should address the number and types of qualification runs that would be performed. During OQ the runs would consist of temperature distribution runs, minimum load and maximum temperature distribution runs for batch ovens, and beginning, middle and end of load temperature distribution runs for continuous process sterilizers. During PQ the runs would consist of minimum and maximum load heat penetration runs for batch ovens and beginning, middle and end of load heat penetration runs for continuous process sterilizers. The heat penetration runs would be conducted at minimum process temperatures and faster belt speeds or shorter times to minimize process lethality if the runs are conducted in conjunction with the microbial challenge test runs.

Qualification of sterilization processes should include a run to show that the biological challenge device is more resistant to the sterilization process than the product bioburden. A product functionality run should be performed at the maximum process temperature, a longer exposure time or minimum belt speed, and with a belt stoppage that represents the maximum allowable in the process specification to provide the most severe thermal stress on the product. If re-sterilization of the product is allowed, then product should be sterilized multiple times to support the number of re-sterilizations that will be allowed.

If the load is to be reused, then there should be a procedure for reconditioning of the product prior to reuse.

9. Acceptance Criteria: The protocol should address the temperatures and allowable ranges of those temperatures that will be used for pre- and post-calibration of the temperature monitoring devices. The allowable uncorrected and corrected deviations of the temperature devices from the references shall be established for the pre-run calibration. The maximum post-run deviation of the temperature devices shall be established; additionally, the number of temperature devices that must meet the post-run calibration requirement shall be established.

The requirements for microbial growth for the bioburden/biological challenge device run and the microbiological performance runs shall be established. The acceptance criteria for any positive and negative controls that are performed in conjunction with the microbial testing will be defined.

Requirements for $F_0$, if calculated should be defined. It should be noted that the microbial criteria are the critical acceptance criteria and take precedent of this requirement.

10. Training: Provide a list of personnel and their training that is required prior to conducting the OQ or PQ.

11. Equipment/Product Disposition: Define the disposition of any product that will be used for the qualification runs and who will be responsible for the disposition.

12. Completion Activities: Define completion activities such as completing the Qualification report, preparing operational specifications preparing the validation report and certification of the validation process.

REFERENCES


Anonymous (2010) ISO 14161 Sterilization of Health Care Products - Biological Indicators - Guidance for the Selection, Use and Interpretation of Results


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Halls, N.A. (1994) *Achieving Sterility in Medical and Pharmaceutical Products*, Marcell Dekker, New York,
Shintani, H. (2011) *Guide for Sterilization of Pharmaceutical and Medical Devices*, Jyohokiko (Information organization), Tokyo, Japan,