CONSOLIDATED STERILIZER SYSTEMS

How to Validate an Autoclave

Introduction

There is an array of qualification tests that can be conducted to validate an autoclave. Many laboratories validate autoclaves by simply using biological indicators (BIs). For some labs, however, a simple validation with BIs is not enough and a more elaborate validation process must be followed. These labs typically require installation, operation, and performance qualifications (IQ/OQ/PQ) to help comply with the current USA and International Standard for steam sterilization as set forth in ISO 17665.

In this eBook, we will explain a variety of validation tests you may need to perform in order to properly and effectively validate your autoclave.

Table of Contents

1. Calibration

"Calibration" is a word that is frequently used in the steam sterilization industry. At its very basic premise, calibration is bringing the response of a sensor (e.g. transducer, thermocouple, etc.) to within a specified range relative to a primary reference standard. For example, an autoclave's chamber temperature probe's response can be compared to a previously calibrated device, or "standard", whose response is traceable to a national reference standard, maintained in the United States by the National Institute of Standards and Technology (NIST).

Why Calibrate?

The short answer is **calibration ensures consistent results from a process**. Steam sterilization efficacy is highly dependent upon actual temperature. For example, if a steam autoclave is running at 120°C for 15 minutes, the theoretical lethality of that cycle is only 82% of a cycle running at 122°C for the same amount of exposure time.

Since most laboratory autoclaves do not require temperature to be accurate to better than ±1°C, this variability can be more common than one would think. If your temperature transducers are calibrated, this problem will diminish.

Equipment Required to Calibrate

The proper way to calibrate an autoclave is with the use of a NIST-traceable device (standard) such as a dry block, oil bath, or temperature probe. If using a dry block or an oil bath (pricing starts at approximately $1,250) make sure it is designed to control to a constant temperature (±0.1°C). If the dry block or oil bath is not NIST-traceable, or its calibration has expired, then use a NIST-traceable temperature probe (cost is approximately $500) with ±0.1°C minimum accuracy.

Note: As a rule of thumb, the standard should be five times as accurate as the device being calibrated. Therefore, when calibrating a temperature probe with a desired accuracy of ±0.5°C, the calibration standard should be accurate to ±0.1°C.

If the above-recommended equipment is not available, you can use boiling water to help calibrate the autoclave's temperature sensors. Boiling water can act as a constant-temperature (i.e. 100°C/212°F) bath that is somewhat near sterilization temperature. However, if the facility isn't exactly at sea level then boiling water isn't necessarily going to be 100°C/212°F. Check the atmospheric pressure in your area (obtained online at www.weather.gov) or use an absolute pressure manometer (mercury column or electronic) to obtain the exact pressure reading, then calculate the actual "pressure corrected" boiling temperature of water by using this steam table: (http://www.efunda.com/materials/water/steamtable_sat.cfm). If you place your sensor into boiling water (not in contact with the bottom of the vessel holding the water) and it isn't within 1°C of the theoretical temperature then you will need to carry out a calibration.

Calibration Methods

Calibration instructions can vary by the number of calibration points measured (i.e. 1-point, 2-point, or 3-point). What is the difference?

Single Point Calibration

A single point calibration is valid only within the accuracy at that specific point. When sterilizing at only one temperature, say 121°C/250°F, this is not too much of a problem, although you have no idea what is happening in any process excursions to higher or lower temperatures.

Two Point Calibration

Some calibration instructions recommend taking two measurements and calculating the slope (gain) and y-intercept (zero offset). Using two points for calibration is relatively fast and convenient; however, two points define a straight line and reveal nothing about any non-linearity in the probe's reading. Also, any errors in the two measurements are not going to be evident.

Multipoint Calibration

A multipoint (more than two point) calibration will indicate if the probe is behaving in a nonlinear manner, which could be a good reason to replace it, and allows any measurement errors to be averaged out over the greater number of points. These instructions suggest taking three or more measurements and performing a linear regression to get the slope and y-intercept. Any multipoint calibration should be done with points both outside the working range of the sterilization cycle(s) you will use. For example, a lab running at 121°C (and only 121°C) should calibrate at 116, 121, and 126°C or at 116 and 126°C for a two-point calibration. This allows the response of the measurement and control system to be considered. If the lab is running cycles over a greater range, calibration should start and finish 5°C below the minimum temperature and 5°C above the maximum temperature used. A good rule of thumb is to calibrate against at least the number of degrees in °C plus one. So, if you run cycles at three different temperatures (e.g. 115°C, 121°C, and 134°C), you would make calibration measurements at four points (e.g. 110°C, 120°C, 130°C, and 140°C.

Sample Calibration Procedure for an Autoclave

1. Record the as-found calibration data (zero and gain) for each sensor to be calibrated.
2. Using appropriate caution (shut the steam off and wait for the pressure to go to zero!),remove the sensors to be calibrated from the sterilizer, leaving their cables connected to the control system.
3. Set the zero and gain to 0 and 1, respectively.
4. If using a NIST-traceable dry block or oil bath place the sensor in the dry block or oil bath.
5. If using a NIST-traceable temperature probe, place the temperature probe into a central position in the dry block or oil bath and the sensors as close to it as possible.
6. Measure at the selected temperatures and record the standard and transducer data You should wait for at least one minute at each temperature to allow the measurement to stabilize. Don't rush this step.
7. Do a linear regression of the data collected. This is straightforward in Microsoft Excel [intercept() and slope() functions] with the standard's data on the y-axis and the sensor's data on the x-axis.
8. Do a correlation as well [correl() function]. It should be at least 0.999999 (i.e. a very straight line).
9. Enter the zero and gain values to the sterilizer controller to enter the calibration.
10. Verify the calibration using at least one point like your process temperature. If more than one process temperature, then verify at each one. Then you will have an exact statement of the accuracy of the sensors.
11. Re-install the sensors in the sterilizer.

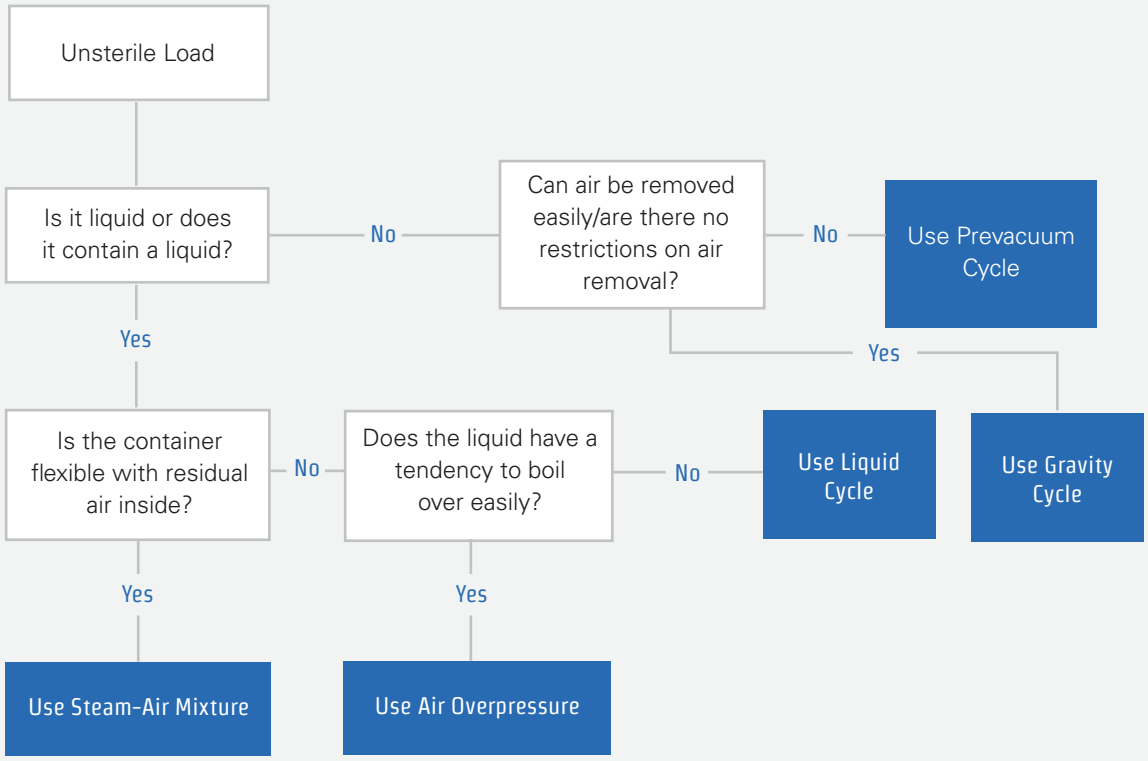
2. Cycle Development

Cycle development is performed to provide you with a sterilization process (i.e. cycle type, sterilization temperature, sterilization time, etc.) that can be validated. Ideally, cycle development should begin before the sterilizer is purchased, since the cycle types will affect the type of autoclave you need, and changes in the field after installation are generally difficult and expensive. At the very least, cycles should be developed prior to performing a full validation of the autoclave and, more specifically, before Performance Qualification (PQ) validation.

It's worth pointing out that in most laboratories cycle development isn't required or has already been performed by others in the lab. Cycle development is most commonly used for biotechnology, pharmaceutical, or medical device companies that are regularly validating their autoclaves and/or sterilizing new or unique loads.

Selecting a Sterilization Cycle

Refer to the flow-chart below to determine which cycle type is best for your load.



Once the cycle type is determined you must decide on the key cycle parameters: sterilization temperature and sterilizer time.

The vast majority of loads sterilized in the laboratory setting are sterilized at 121.1°C/250°F for 30-90 minutes. For loads that are sensitive to heat, contact your autoclave validation expert for advice on calculating your sterilization time using the F0 relationship.

Once you have determined your cycle type and parameter, you must validate the cycle to ensure the load will be sterile. These test cycles should be loaded in a way that is representative of the actual load to be sterilized. It is recommended to run your test cycles at half of the desired sterilization time (this is referred to as the "half-cycle" method).

Sterility testing is confirmed with the use of biological indicators (BIs). The most common test organism for steam sterilization is Geobacillus stearothermophilus. BIs are placed in intimate contact with the load and are placed throughout the chamber. Typically, at least 10 BIs are used per cycle. If all BIs show no growth (i.e. all the bacteria are destroyed) in three successive cycles, you have validated the cycle.

3. Installation Qualification (IQ)

Installation Qualification (IQ) provides documentary evidence that the equipment has been built and installed to specification, and that all supporting services (i.e., utilities such as electricity, water, and steam) are available and connected properly. The IQ process methodically documents all aspects of the installation, the machine components, and any testing equipment used to provide a complete, closed-loop assessment. In addition, IQ includes NIST traceable calibration (see above) verification of the unit's critical components.

Equipment Procurement

IQ begins before you issue the purchase order for the autoclave. Completing a successful IQ is the result of careful planning and there should be no surprises when the unit arrives at your facility. Naturally, what you ordered and what you receive should match.

IQ is typically carried out concurrently with installation or soon after installation at the user's facility. The IQ is performed by following a specific IQ protocol tailored for each piece of equipment.

Equipment Installation

Installation has requirements that are unique to each machine, or at least should be treated that way. As part of IQ, these requirements must be verified. They include:

Utilities

- ✓ Electrical: voltages, current, fusing, correct number of phases, and three-phase configuration
- ✓ Compressed air: pressure (static AND dynamic), flow
- ✓ Water: pressure (static AND dynamic), flow, temperature(s), purity(ies), dissolved gas content
- ✓ Steam: pressure, purity, pipe insulation, pipe size and peak flow rate
- ✓ Drain: capacity, temperature tolerance

Installation Area

- ✓ Dimensions: including space to allow service
- ✓ Free-standing or through-wall mounting
- ✓ Biosafety barrier
- ✓ Seismic anchoring
- ✓ Leveling: for sterilizer performance and to align with delivery cart for removable load carriages

An IQ should list the requirements and provide documentation of the presence and adequacy of each utility and feature listed above (if so furnished).

Calibration

Calibration requires standards that are traceable to the standards maintained by the United States National Institute of Standards and Technology (NIST). In other countries, traceability is to be to that country's national equivalent of NIST.

Transducers and Sensors

For autoclaves, this means that a pressure standard and a temperature standard should be available, as well as a means of providing stable pressure for calibrating the pressure transducer(s) and a dry block or oil bath for calibrating the temperature transducers. Optimally, a calibration standard should be five times as accurate as the device being calibrated needs to be. For a ±0.5°C accuracy on the sterilizer, the standard should have an accuracy of ±0.1°C. Details of the calibration depend upon the sterilizer's controls system and how it handles calibrations. Generally, at least a three-point calibration is required, with more points providing a better indication of the linearity of the system. The golden rule of calibration is, "If you do it fast, you get to do it twice." So waiting for a stable value is key in a successful calibration.

Time

Another aspect of calibration is that of the internal clock of the sterilizer. The standards related to steam sterilizers generally have accuracy requirements for the process control clock(s) in the controller. Testing is straightforward, since NIST has a dial-in phone number (303-499-7111) that tells you the national standard time on a one-minute interval, providing you with ±1 second accuracy. This level of accuracy is adequate for any laboratory autoclave. Measurements at either end of a 24-hour period would be acceptable to any audit.

A thorough IQ will enable you to have confidence in your autoclave, and more importantly, documented proof of its ability to carry out the task at hand, not just because the manufacturer tells you so, but also because it has been demonstrated in place.

IQ is a critical part of the process for fully commissioning an autoclave and putting it into its appointed use.

4. Operational Qualification (OQ)

Operational Qualification (OQ) verifies that the autoclave meets the desired and intended performance standards of the lab. OQ testing examines the autoclave's ability to run the sterilization process correctly and repeatedly while also responding appropriately to error conditions. OQ testing typically includes the following tests:

- ✓ Empty chamber temperature mapping
- ✓ Simulated load chamber temperature mapping and, if required, verification of sterilization efficacy using biological indicators (BI)
- ✓ Alarm conditions and expected results

Equipment Capabilities and Specifications

In general, the operational requirements for a sterilizer are detailed in a User Requirement Specification (URS). The URS defines the required performance, parameter limits, accuracy, ancillary functions, and standards to which the equipment's performance must meet. It is the basis of OQ and can be written by the customer or may be the manufacturer's specification that the user accepts.

Autoclaves have specific critical process parameters:

- ✓ Temperature set point range and control
- ✓ Temperature uniformity throughout the chamber and load
- ✓ Pressure range and control
- ✓ For prevacuum sterilizers: air evacuation capability, including depth and control of vacuum setpoints
- ✓ For any sterilizer: the capability to achieve saturated steam pressures given the temperature setpoint and control range
- ✓ It may also include evacuation and pressurization rate control
- ✓ Exposure time (and other dwell phase) control

Successful cycle execution requires that these parameters are achieved repeatedly and with predefined precision. If the machine must comply with a specific standard (ISO 17665, PDA Technical Report 1, PDA Technical Report 48, EN 285), the accuracy and precision of the process parameters will be specified in the standard.

Equipment Testing

OQ validation generally includes both empty chamber and loaded chamber temperature mapping for all cycle types that the facility runs (e.g., gravity, liquid, pre-vacuum, etc.). If an autoclave has the capability to run a cycle but it is not currently used, the facility does not have to test it since the validation for that cycle can always be run at a later date.

Empty Chamber Testing

Empty chamber temperature mapping is performed to verify and document that the temperature range delivered throughout the empty autoclave is within required specifications. Interestingly, temperature mapping an empty chamber can create more of a challenge than testing a fully-loaded chamber because the load's thermal mass aids in providing temperature homogeneity and limiting overshoot.

Empty-chamber testing is done with temperature datalogging probes. Temperatures should be measured at a minimum of five locations in the chamber (plus at a point adjacent to the controlling temperature sensor in chambers up to 800 liters). More sensors provide a more-detailed analysis of temperature profile.

Examples of acceptance criteria are:

- ✓ Chamber temperature -0/+3°C relative to setpoint
- ✓ Chamber temperature range over all dataloggers at any given time ≤2°C

Pressure datalogging should also be performed only if pressure is a controlled parameter.

Simulated Load Testing

OQ validation must also test simulated loads that mimic the real loads a laboratory plans to run. These loads may be solid, liquids, glassware, medical devices, in short, anything that would normally be sterilized. For simulated load testing, ensure that at least three of the items in each load are being monitored for temperature and biological indicator (i.e. place the probe and BI right next to or on top of the load).

Diverse items processed in a specific cycle type may be tested together, however, their temperature and indicator (see below) data should be evaluated independently. Therefore, if mixed loads are to be tested, at least three of each item type should be included, and arrayed throughout the sterilizer chamber, not all adjacent to one another.

Simulated load testing may also include biological and/or chemical indicator testing. For overkill cycles, half-cycle testing (the cycle has exposure duration of half the normal exposure time) should be carried out for these cases. Biological indicators must have a 106 population of G. stearothermophilus spores for validation testing of steam sterilization. If chemical indicators are used, a Class 4 or 5 indicator is required. Class 6 indicators are only suitable for full-cycle testing, since they are not expected to show a complete cycle in less exposure time.

Alarm Testing

Here is sample list of alarms that should be tested during an OQ validation:

Alarm/Interlock	Action	Expected Results	Method & Actual Results
Recycle Alarm	Simulate recycle alarm (Alarm occurs when temperature falls below sterilization temperature setpoint for 1 minute or more during the sterilize phase of a cycle)	Buzzer sounds, alarm message printed, and steam supply to chamber is shut off and chamber exhausts	
Over Temperature Alarm	Simulate over temperature alarm (Alarm occurs when chamber temperature exceeds setpoint by x°C)	Buzzer sounds, alarm message printed, steam supply to chamber is shut off and chamber exhausts	
Time Limit Alarm	Simulate time limit alarm (Alarm occurs when temperature is not achieved within 1 hour of cycle start)	Buzzer sounds, alarm message printed, steam supply to chamber is shut off and chamber exhausts	

The method used to activate the alarm must be documented and should simulate the process error that would cause it to take place. The actual result must be identical to the expected result in order for it to "pass" the OQ validation.

Ancillary Items to Test

Other unit-specific items that should be tested include, but are not limited, to:

- ✔ Switches to prevent steam from being admitted to the chamber with the door unsealed
- ✔ Interlocks to ensure that, in a double-door unit, both doors cannot be opened at the same time in normal operation
- ✔ For units with dual transducers, the transducers must read within a specified amount of one another during exposure phase

Operational Qualification provides documented proof that the autoclave can execute the required processes and that its safety and ancillary features work properly. These capabilities must be demonstrated in the unit's final location of

5. Performance Qualification (PQ)

Performance Qualification demonstrates that the autoclave not only runs the required cycles, but provides the required result: sterility. The PQ process involves testing the loads that must be successfully sterilized in the autoclave. The prerequisites for PQ are successful IQ and OQ tests.

Sample Test Protocol

PQ is comprised of at least three repeated tests for each defined load. The critical aspect of the tests is to validate sterility in each of the loads. Each load type will have its own challenges so it's important that each is specified with information such as: number and type of objects being sterilized, sterile barrier material used, and in certain cases, orientation of the objects being sterilized in the autoclave.

Here is a sample PQ test protocol:

1. Objective

The Objective is to prove sterilization of the specified load. The sterility assurance level may derive from a standard, from a client SOP or specification, or from a manufacturer's specification. Specific citations to any of these should be made in this section.

2. Procedure or Setup

The Procedure or Setup explains how to setup and execute the test. The following specifications should be noted here:

- The type of cycle (vacuum, gravity, liquids, air-overpressure, low temperature, etc.) including all critical parameters
- Any equipment needed for the test
- The number and location of each item in the load
- If a mixed load is to be run, the number of each item and the items' locations should be specified
- Locations of the indicators and datalogger probes – these should be placed in "worst-case" locations in the load to provide assurance that the entire load is sterilized
- Sterile barrier materials or vessel covers (like aluminum foil for flasks), as well as their application to the items closed by them
- A step-by-step listing of how to carry out the test

3. Acceptance Criteria

The Acceptance Criteria section lists the required result or range of results. Examples of acceptance criteria for Performance Qualification involve sterility and temperature. For example, if the "acceptable criteria" for the temperature range within the chamber during sterilization is -0/+3°C relative to the actual set point, then the chamber temperature at all measured points must be within that range for a successful outcome to be recorded.

4. Result Record

The Result Record is where all of the test data is recorded. At least three iterations are typically run for process tests. Any failure to comply with the acceptance criteria makes an "iteration" a failed test. The required number of iterations must be run successively with successful outcomes for the test to be considered a pass. The record must include:

- ✔ Sterilizer cycle tape or electronic record
- ✔ Biological (and chemical, if used) indicator results
- ✔ Datalogger results and their compliance with the Acceptance Criteria
- ✔ Analysis of the datalogger (thermocouple) records for accumulated lethality (Fo) and compliance with allowable load temperature ranges in the exposure phase

5. Verification

After the testing is done, the results are typically reviewed for compliance with the acceptance criteria. The review is finalized in this section with signatures from the person who ran the test and, preferably, with countersignature of a second person or manager.

Sterility Testing & Record Keeping

Sterilization cycles are to be run sequentially and the load setup should follow the Procedure or Setup section of the test. Indicators and data logger probes are to be placed as specified. Sterile barrier or closure materials are to be applied as specified. If sample loads are to be reused, they should be allowed to cool to a specified temperature (optimally room temperature) between cycles to provide a real-world thermal load for the sterilizer and a real-world exposure for the indicators.

In all tests, record keeping is essential to the integrity of the test. Records may include cycle tape printout from the sterilizer, data logger data files, photos, sketched diagrams, etc. These must be attached or provided with the report, with each identified by the section of the PQ protocol to which it applies. Any electronic data records should be provided in a non-volatile form with the final report, i.e., as a CDROM or DVDROM, or via cloud storage.

A more extensive list of the required records is listed here:

- ✓ A catalog of who carried out the testing, the verification of results, and approvals with initials and signatures
- ✓ A listing of reference or other test equipment, manufacturer, model, serial number, and calibration due date, if applicable
- ✓ A listing of biological indicators and chemical indicators (if used), including manufacturer model number, lot number, D-value, and nominal population
- ✓ If inoculum is used, a recovery study shall be done to ascertain the actual recoverable population of the indicators; also, a growth promotion study shall be done to ensure that the item upon which the inoculum is deposited does not have a negative effect on out growth of the indicator organism
- ✓ A calibration record for test equipment (e.g. thermocouples) if done at the work-site, both before and after a series of tests and/or after the entire test protocol is performed

A well-executed PQ will enable you to demonstrate and document that the autoclave can execute the desired processes and provide the sterility assurance level that is required for your application.

PQ is generally the final step in qualification. This testing creates an unassailable baseline level of proof of the autoclave's ability to do the specified job, and is the basis for requalification at periodic intervals.

6. Steam Quality Testing

The quality of the steam feeding an autoclave is an important factor in steam sterilization. Like time, temperature, and pressure, steam is a critical variable in the success and repeatability of the sterilization process. As such, steam quality should be part of the validation of any steam sterilizer.

Steam quality is defined as the measurable physical aspects of steam used for sterilization. These physical aspects include temperature (superheat), dryness (liquid water content), and non-condensable gas content. (Steam quality is not a measure of the impurity content of the steam.) Deviations from established ranges of these aspects of the steam can result in the following issues:

- ✔ Wet loads
- ✔ Damaged loads
- ✔ Unsterile loads
- ✔ Sterilization (biological and chemical) indicator failures
- ✔ Staining and corrosion of instruments and containers

Each of these issues has a specific cause or causes and can usually be remedied.

What to Know About Steam Quality

Almost every sterilizer manufacturer recommends "97% pure steam". In general, this is not defined, rarely measured, and, if discussed at all, is relegated to the mythology of sterilizer arcana. The good news is that essentially all laboratory autoclaves on the market today can provide sterile, dry, and intact sterilization loads if provided good quality steam from the steam supply. The bad news is that any autoclave can experience the above problems, and the cause is not always something that can be predicted.

With careful design, following well-established principles, and proper maintenance, the system (steam supply and sterilizer) can be engineered to provide a large margin of security against steam quality noncompliance. For a production or GMP environment, steam quality testing should be part of annual preventative maintenance and qualification testing.

When steam quality testing is performed, three parameters are measured:	
Steam Dryness	The amount of the steam by weight that is steam and not liquid water
Non-condensable gases	The amount of the steam by volume that is not steam or water, but is air or other gas that does not contribute meaningfully to sterility of the load
Superheat	The temperature of the steam above the temperature of saturated steam for a given moisture content

EN 285, the European Large Steam Sterilizer standard [1], is the world's baseline authority for steam quality acceptance criteria. It is referenced in most national standards and in ISO 17665 [2]. With the release of EN 285:2015, the bar has been raised. The acceptance criteria are shown in the following table.

Steam Dryness	Non-condensable gases	Superheat
>0.95 w/w*	≤3.5% v/v	≤25K

**For laboratory autoclaves, >0.90 w/w is considered acceptable.*

Steam Dryness

Steam dryness is calculated by measuring the temperature change in a known amount of water in relation to the mass of steam that is required to cause that temperature change. Ideally, the temperature rise is exactly proportional to the amount of steam delivered to the water to heat it, resulting in a dryness value of 1.0 (i.e. perfectly dry steam with no liquid water content.) Normally, the dryness value is less than 1.0, as there are thermal losses in any piping system even if it is well insulated. Because the dryness value of the steam at the chamber entry point can be quite a bit lower than the dryness value in the sterilizer, measurements of steam dryness should be made at both locations.

The acceptance criterion for steam dryness (the fraction of steam relative to water – 1.0 = all steam, no water) is at least 0.95, or 95% by weight. A dryness level down to 90% is considered acceptable for laboratory autoclaves, however, steam below this value is considered to be wet steam.

Wet steam does not deliver as much energy to the load as >90% saturated steam and can cause what is known as "wet packs". If the steam is wet, or if the saturation level has decreased since the last validation, the expected Sterility Assurance Level is probably not being achieved. This is especially important for bioburden-based validations, since overkill cycles have more of a safety margin by their very nature.

Non-condensable gases

Non-condensable gases are generally air and air is a poor sterilant compared to steam. As an example, a typical dry-heat sterilization exposure phase lasts upwards of two hours at a temperature of at least 160°C/320°F. Steam sterilization typically is done with exposure phases of 15 minutes at 121°C/250°F or 3.5 minutes at 134°C. The efficacy difference is notable. For a comparison, consider a contact lens manufacturer that must sterilize contact lens blisters to a 10-6 sterility assurance level. Sterilization can be performed using a "steam/air mix" cycle that runs at 122°C/252°F for 45 minutes with a steam/air mix of approximately 48% steam (using absolute pressures for the calculation). The same result can be achieved in 15 minutes with saturated steam alone.

In short, non-condensable gases decrease sterilization efficacy. As with wet steam, the Sterility Assurance Level will be less than expected if non-condensable gas content has increased since product sterility validation. The percentage of non-condensable gases in the steam should be less than or equal to 3.5% by volume.

Superheat

The steam is sampled in free expansion into ambient air. The maximum temperature measured at a precise location in the jet is the temperature upon which the superheat analysis is based. When the temperature and moisture content do not match up, two things can happen:

1. If the moisture content is higher than saturation for the temperature, wet loads occur, as discussed previously.

2. When the moisture content is lower than saturation for the temperature, the condition is called superheat. In superheat, the steam is too dry and its energy content is too high. When the steam condenses on the load, the energy released is enough to melt plastic packaging and actually char paper packaging. Neither is a good outcome.

The amount of superheat present in the steam should be no more than 25 degrees Celsius above the temperature in free expansion into atmosphere at the current atmospheric pressure. For all intents and purposes, this corresponds to an upper limit of 125°C in the measurement.

The amount of superheat present in the steam should be no more than 25 degrees Celsius.

What causes non-compliant results?

Wet steam

- ✓ Inadequate insulation around the sterilizer or steam piping, allowing energy loss and condensation
- ✓ Poorly controlled steam boiler chemistry (especially a deficiency of sulfites)
- ✓ Low sections of piping between the boiler and the sterilizer, allowing condensate to pool and be carried over with the steam entering the chamber
- ✓ Too great a pressure drop across a regulator or between the jacket and chamber, which causes the "extra" water in the steam at the higher pressure to fall out as condensate
- ✓ No/clogged steam filters, either letting condensate pass if no filter, or causing a pressure drop that causes condensate to fall out
- ✓ No/clogged steam traps/separators, in either case, condensate in the steam line is not removed
- ✓ Steam trap/filter too far from the sterilizer, allowing condensate to be generated between the trap or filter and the sterilizer
- ✓ Inadequate number of steam traps for the distance that steam must travel from its source to the sterilizer
- ✓ Bad steam system design (vertical drops of steam direct to the sterilizer, no traps, no header, etc.)
- ✓ Load too dense/too cold when placed in sterilizer
- ✓ Foaming of the water in the boiler

What causes non-compliant results, Cont.

Non-condensable gases

These are brought into the steam primarily via two sources:

- ✓ Leaks/cracks in the steam plumbing, filters, separators, etc.
- ✓ Inadequate deaeration of the boiler feed water

Superheat

Superheat can result from the following sources:

- ✓ Jacket temperature/pressure too high
- ✓ Steam pressure/temperature too high entering the sterilizer
- ✓ Steam flowing through a small orifice or tight-radiused direction change between its source and the chamber causing a large pressure reduction/steam velocity increase

The temperature shown on the sterilizer controls generally will not show superheat values, even if superheat is present. Since the temperature is measured in the drain of the sterilizer chamber, superheat will have been dissipated into the load, chamber wall, door and backhead long before it reaches that sensor.

Solutions

Each of the steam quality parameters can be measured and, if issues arise, addressed. The first step is to measure, even if there are no problems. This should be done on a regular basis — at initial installation, and after preventative maintenance to establish a baseline for the system. Measurements made when there are no problems can also provide an indication if the sterilizer is close to having a problem. Measurements should also be made when changes are made to supply plumbing.

Persons experienced in steam quality analysis can usually make cost-effective suggestions to fix the problems, and of course measure to see if the problem is, in fact, fixed.

7. Factory Acceptance Testing (FAT)

A steam sterilizer, whether used for pharmaceutical or laboratory sterilizing applications, is designed to use steam under pressure as the sterilizing agent. When constructed, installed and operated correctly, a steam sterilizer should function properly and sterilize your load. But, how does a sterilizer manufacturer ensure the unit they've built is assembled properly and is in good working condition prior to being shipped? Furthermore, how does the end-user know that the sterilizer they are buying is up to spec? The answer: Factory Acceptance Testing (FAT).

What is Factory Acceptance Testing?

Factory Acceptance Testing (FAT) is a process that evaluates the sterilizer during and after the assembly process by verifying that it is built and operating in accordance with design specifications. FAT ensures that the sterilizer's components and controls are working properly vis-a-vis the functionality of the sterilizer itself. As the name suggests, this testing is performed at the factory.

FAT is typically conducted in order to assess any discrepancies and non-conformities, as well as develop a process for how they are to be handled. Deviations or abnormalities observed during testing are documented in a problem report and corrected prior to shipment. While the end goal for a manufacturer is to ship a sterilizer as soon as possible, FAT must be conducted in a thorough and forthright manner. A poor or rushed FAT can lead to missed non conformities, which can only then be corrected after the equipment is installed—which in turn can wreak havoc on a project schedule.

In short, the purpose of FAT is to document the results of the sterilizer testing at the factory prior to shipment.

The Factory Acceptance Testing Process

There are several components that make up FAT. The product must be assembled, prepared, and connected to utilities for testing. Documentation such as drawings, ASME certificates, and test procedures must be gathered. And finally, testing must be conducted in the factory that follows specific test procedures.

Preparation

Before the FAT begins, the manufacturer should present the FAT procedure to the client for review and approval. The procedure should include testing of as much functionality as is practical in the factory and, where possible, should show pass/fail criteria or desired results for each item tested.

Once the procedure is approved, the manufacturer should test the equipment before the FAT begins. Keep in mind that some clients like to visit the factory during FAT, so this "pre-test" will confirm that the sterilizer is set up with the correct options, ensure a smoother FAT process and minimize down-time during the FAT.

Documentation

There is much documentation to be gathered and checked during FAT (see Table 1 below for a sample list of key documents). These documents are provided by the manufacturer before, during, and after the testing phase.

Table 1

Sample List of Required Documents for FAT
Dimensioned Outline Drawings
Assembly Drawings
Schematic Piping Drawings
Schematic Wiring Drawings
Installation, Operation and Maintenance Manuals
Piping and Instrumentation Diagram(s)
Maintenance Procedures
Detailed Parts List and Materials of Construction
Instrument Index including Tag, Location, Specifications, Ranges and Tolerances
NIST Traceable Calibration Data Sheets for Monitoring Instruments
Software and/or Controls Parameters, PID Constants
Pressure Test Reports
Manufacturer/Vendor Certificates (Materials, ASME, etc.)
Supplier SOPs for Setting up the Sterilizer with Adjustments using Manual Needle Valve

Testing

Next comes the critical testing involved in FAT. Each critical system of the sterilizer is tested and referred to as a Test Case (see Table 2). In order to confirm that the autoclave is operating correctly you must then set test objectives for each Test Case and develop acceptance criteria in accordance with your pre-test procedures. The actual results of each test will be documented and indicated as either passed or failed. The initials of the tester are provided for each test as well as a signature sheet. Some FAT forms also include an area for comments in case there are any discrepancies or non-conformities, as well as suggested remedies.

Table 2

Test Cases	Objective	Test Procedure	Acceptance Criteria
Dimensioned Outline Drawings			
Assembly Drawings			
Schematic Piping Drawings			
Schematic Wiring Drawings			
Installation, Operation and Maintenance Manuals			

FAT may be a necessary step in the validation process, especially for pharmaceutical clients where special one-off features may be ordered. In some instances, FAT can actually be conducted during Installation Qualification (IQ). It is best to review your project and validation requirements to determine if this is possible for your situation.

Interestingly, most laboratory clients and general purpose applications do not require FAT. Where FAT is not a requirement, any reputable manufacturer will still follow a testing process to ensure the autoclave is functioning properly prior to shipping.

8. Simple Validation Process

If the budget isn't available to perform periodic validation then the easiest and most inexpensive way to ensure the autoclave is sterilizing properly is to run cycles with biological indicators (BIs). See below for a step-by-step protocol for simple validation.

1. Calibrate sterilizer temperature probe
   - ✔ Use NIST traceable thermocouple and meter (or bath)
2. Run an empty chamber cycle (5-10min)
3. Load the autoclave with a typical load (e.g. bottles, cages, liquids, etc.)
   - ✔ Load should be worst case scenario
4. Place BIs in the load and near drain
   - ✔ Minimum of 3-5 BIs per test cycle
5. Run cycle
6. Repeat steps 3-5 for each autoclave load or cycle type
7. Check and record results after 48-hour incubation

The frequency at which you perform this type of simple validation is your choice: once per week, once per month, once every six months. Whatever it is that you decide, try to be consistent and if any tests fail (i.e. the BIs show growth) then contact your local service representative to investigate and resolve the problem.

The procedures mentioned throughout this eBook allow researchers, technicians, facility managers, and consultants to gain a better understanding of laboratory autoclave validation.

The validation requirements for each lab will be different, some requiring more documentation and testing than others. Whatever procedures that are executed will add a layer of comfort to the users that the autoclave is functioning properly.

Consolidated Sterilizer Systems manufactures top-quality steam sterilizers for leading universities, hospitals and biotechnology facilities worldwide. Our autoclave validation specialists are available to answer **all** your questions.

Reference: ANSI/AAMI/ISO 17665-1: 2006/(R)2013 Sterilization of health care products—Moist heat—Part 1: Requirements for the development, validation, and routine control of a sterilization process for medical devices, definition 3.17.

CONSOLIDATED
STERILIZER SYSTEMS

Please contact us for more information on
autoclave validation services.

www.consteril.com
617.782.6072