Moist Heat Sterilization Process Decision Tree
Decision logic adapted from PDA Technical Report No. 1

Glossary:
Liquid Load: A load where the primary item being sterilized contains a free-flowing aqueous or non-aqueous liquid (e.g., filled vials, syringes, bulk solution bags).
Porous Load: A load that may trap air due to internal channels, packaging, or material structure (e.g., filters, stopper bags, textiles, coiled tubing, nested devices).
Hard Goods: Non-flexible, non-porous items such as stainless steel instruments or glassware that do not retain moisture post-sterilization.
Ballast: Inert material (e.g., water-filled vials) used during cycle development or routine runs to simulate thermal mass and ensure representative worst-case conditions

Key Principle; F_o is a time–temperature integral calculated from a measured temperature profile. It is meaningful only when the measured temperature accurately represents the lethal conditions experienced by microorganisms. This condition is met when lethality is governed by heat transfer into the product, rather than by air removal, steam penetration, or chamber conditions.
Accordingly, F_o is appropriate for liquid sterilization processes, including liquid cycles operated with air overpressure (AOP), as well as for steam–air mixture (SAM) processes and superheated water shower systems. It is not appropriate as a primary lethality metric for gravity displacement or pre-vacuum porous load steam cycles, unless effective steam penetration has already been unequivocally demonstrated

Validation is demonstrated by product temperature mapping and exposure time at the coldest point of the liquid load. Where applicable, biological indicators may be used as supportive evidence but are not the primary measure of lethality. Routine release of liquid cycles is based on confirmation that each cycle meets the validated product temperature profile and exposure time at the coldest point of the load, along with compliance with the validated cycle parameters (e.g. chamber conditions, controlled exhaust). F_o calculated at the product level may be used as the primary lethality metric for routine release, provided the cycle is executed within the validated state and no deviations are observed

Start: Un-Sterile Load

Is it a "Liquid Load" or does it contain a liquid?
- **YES** → **Are you planning to perform terminal sterilization (Sealed Product) as part of your process?**
  - **YES** → **Is there a risk of container damage due to over pressure during the heating phase**
    - **YES** → **Is the product–container system compatible with direct hot-water contact, effective drainage, acceptance of a wet load, and uniform heat transfer?**
      - **YES** → **Superheated Water shower Process**
      - **NO** → **Non-Saturated Steam Process; SteamPlus non-condensable gas (Air) Mixture Process (SAM) by a Load Probe uses air during both the Sterilization and exhaust phases**
    - **NO** → **Is there a risk of container damage due to over pressure during cooling phase**
      - **No** → **Saturated Steam Process; Use Liquid Cycle (with Slow/Ramped Exhaust)**
        - **NO** → **Does the liquid have a tendency to boil over easilyor if there is concern of significant evaporation (up to 5% volume loss) during the liquid cycle?**
          - **YES** → **Saturated Steam Process; Use Liquid Cycle with the Air Over Pressure (AOP) by a Load Probe in the cooling cycle**
          - **NO** → **Saturated Steam Process; Use Liquid Cycle (with Slow/Ramped Exhaust)**
      - **YES** → **Saturated Steam Process; Use Liquid Cycle with the Air Over Pressure (AOP) by a Load Probe in the cooling cycle**
  - **NO** → **Is there a risk of container damage due to over pressure during cooling phase**
    - **No** → **Saturated Steam Process; Use Liquid Cycle (with Slow/Ramped Exhaust)**
      - **NO** → **Does the liquid have a tendency to boil over easilyor if there is concern of significant evaporation (up to 5% volume loss) during the liquid cycle?**
        - **YES** → **Saturated Steam Process; Use Liquid Cycle with the Air Over Pressure (AOP) by a Load Probe in the cooling cycle**
        - **NO** → **Saturated Steam Process; Use Liquid Cycle (with Slow/Ramped Exhaust)**
    - **YES** → **Saturated Steam Process; Use Liquid Cycle with the Air Over Pressure (AOP) by a Load Probe in the cooling cycle**
- **NO (Solid; EQUIPMENT / COMPONENTS / MATERIALS or Semi Solid Lab. Waste)** → **Is it a porous /Hard goods load?**
  - **YES** → **Porous solids (hollow loads), including filters, stopper bags, textiles and gowning, long tubing with bends or narrow internal diameters, as well as any hard goods presenting a risk of air entrapment.**
    - **Is an existing pre-vacuum sterilizer available?**
      - **YES** → **Saturated Steam Process; Use Fractional Pre-Vacuum/Post vaccum Porous Load Cycle**
      - **NO** → **Stop/Warning! Gravity cycle not suitable for porous loads. Upgrade to Pre-Vacuum strongly recommended**
  - **NO** → **Non- Porous Solid e.g. Stainless Steel parts, polymer-based components (PTFE, silicone, PVC, or EPDM) hosing, Loose solid waste in single biohazard bag.**
    - **Is effective air removal and steam penetration achievable?**
      - **YES** → **Saturated Steam Process; Use Pre-Vacuum/post vacuum Load Cycle**
      - **NO** → **Simple geometry, fully non-porous e.g. Unwrapped solid Instruments, glassware, Waste ,Utensils ,Red-bags** → **Saturated Steam Process; Use Gravity Displacement Cycle**

End:
Cycle selection, qualification (URS, FAT, SAT, IQ, OQ, PQ), and routine control must comply with EU GMP Annex 1 (2022), ISO 17665-1, ISO 11138-8 PIC/S PI 014-3, PDA TR1, & compendium requirements (USP <1229>, Ph. Eur. 5.1.2).All cycle data (temperature, pressure, time, F_o, vacuum levels, etc.) must be recorded using qualified systems that comply with ALCOA+ principles, as required by EU GMP Annex 11

Ideal for terminal sterilization of sealed aqueous products in rigid or flexible containers when dry load is required. When properly designed and validated , it provides controlled and uniform heating through forced "steam–air" circulation, with counter-pressure protection to maintain container integrity. Rapid cooling and effective drying make it suitable for PFS, IV bags, and mixed formats where there are risks like forcing out the rubber plunger movement, drug loss, or incomplete sterilization exist. For PFS products, It is recommended that plunger movement, orientation, and worst-case nesting must be evaluated during PQ study.
Per EU GMP where lethality is demonstrated using F_o, the F_o value shall be scientifically justified and derived from measured product temperature. Reduced F_o approaches (e.g. $F_o \geq 8$ minutes) may be acceptable only when supported by enhanced bioburden control and regulatory justification.

ISO 17665-1 permits the use of moist heat sterilization processes that do not rely solely on saturated steam—such as steam–air mixtures (SAM) and superheated water shower processes—provided they are scientifically justified and fully validated.
Per EU GMP Annex 1, alternative moist heat sterilization processes (e.g. steam–air mixture and superheated water shower systems) must deliver a sterility assurance level (SAL) equivalent to that of saturated steam sterilization (i.e. SAL ≤ 10^{-6}). This equivalence must be demonstrated through comprehensive validation, including worst-case load configuration, product temperature mapping at the coldest point, and verification of reproducible heat transfer and process control. Where lethality is demonstrated using F_o, the F_o value shall be derived from measured product temperature and justified based on the intended sterilization approach.
- Temperature uniformity (thermal mapping),
- Characterization of heat distribution, penetration, & heat transfer between the load and heating medium
- Drainage efficiency confirmation
- Worst-case microbial challenge studies, and
Biological indicator (BI) penetration studies, with BI placement at worst-case locations identified during heat mapping (per ISO 11138).

Cycle validation and routine batch release rely primarily on F_o at the product level, supported by validated process controls and physical parameter monitoring. BIs are supportive, not primary, for sterility assurance but must still be integrated into the validation strategy.
These cycles are not suitable for all load types, particularly those with poor water drainage, air entrapment risks, or container integrity vulnerabilities

ideal for Liquid waste in open flasks or vented bottles

Ideal for terminal sterilization of aqueous products in rigid LVP containers or ampoules where load configuration allows effective water contact and drainage. This process provides controlled and uniform heat transfer with superheated water shower, counterpressure protection for container integrity, and rapid cooling for heat-sensitive formulations (ideal process when low-temperature sterilization e.g., 100–105°C is required). A wet load is acceptable for this process because products are terminally sealed. However, the cooling profile (rate, pressure, duration) and closure integrity must still be verified as part of cycle validation and periodic monitoring to ensure no compromise during sterilization or cooling.

Air overpressure (AOP) is applied solely for container integrity and pressure balance during cooling and does not affect the lethality mechanism, which remains governed by heat transfer into the liquid product.

Note I: Effective air removal is the critical challenge. For such loads, cycle validation and routine release are based primarily on physical parameters (e.g. chamber pressure, temperature plateau, and exposure time) supported by biological indicator (Bi) performance. F_o (lethality-calculation-based) measurement at product level cannot be used as the primary acceptance criterion until effective air removal and adequate steam penetration have been demonstrated."

Note II; Steam penetration and air removal should validated by:
-Bowie–Dick test (applicable to pre-vacuum porous load steam sterilizers only; performed daily per EU-GMP Annex 1 §8.61). Failure to pass this test prohibits sterilization of porous loads until the issue is resolved and re-validated.)
-Vacuum leak test (weekly)
-Load pattern validation (Min / Max / worst-case)
-BI penetration studies

Note: Validation approach depends on load characteristics (Loading different configuration including Min. & Max & worst -case loading.)

It usually exist in two types;
1- Simple
2- can be equipped with post-cycle vacuum feature the pressure in the autoclave chamber is released too quickly during the exhaust phase

Note; Meets basic sterilization needs; often Class N autoclaves. Proper design of steam traps (location, type & size should be considered. At the end of a Gravity Cycle, the load can still be hot and possibly wet.

As per EU GMP Annex 1, Parametric release based on physical parameters (e.g., temperature, time, F_o) is only acceptable when:
(a) the sterilization process has been fully validated (IQ/OQ/PQ completed),
(b) the cycle is executed within the validated design space,
(c) no critical deviations occurred, and
(d) the monitoring system is qualified and compliant with data integrity principles (ALCOA+).
Biological indicators may be used for periodic verification but are not required for batch release under a validated parametric release program.