BP803T · B.Pharm Semester VIII · Theory Notes

Industrial Pharmacy and Facility Design

Unit IV — Facility Design: Sterile Manufacturing (9 hours)

Unit IV (9 hours)

Pills & Python Notes

From-zero teaching standard: every technical term is defined before use.

Syllabus coverage: Unit IV (9 hours).

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CHAPTER 5: Sterile Manufacturing Facilities

5.1 Learning Objectives

After studying this chapter, the student will be able to:

1 Define sterile, aseptic, terminal sterilization, pyrogen and endotoxin in simple words, and explain why each concept matters in sterile manufacturing.

2 Describe the general layout principles of a sterile manufacturing facility as per Schedule M and current Good Manufacturing Practice (cGMP).

3 Explain what a cleanroom is, why it is needed, and how Sterilize-in-Place (SIP) works and where it is used.

4 Describe material flow and personnel flow in a sterile facility, including entry/exit procedures, gowning, airlocks and unidirectional flow.

5 Explain zoning, segregation and the Grade concept used to protect sterile products from contamination.

6 Compare cleanroom classifications and guidelines given by FDA, EMA, WHO and ISO, and explain "at rest" vs "in operation" conditions.

7 Explain the significance of HVAC in a sterile facility, name its components, and describe HEPA filters and the concept of DOP/PAO integrity testing.

8 Apply the qualification/validation approach (DQ, IQ, OQ, PQ) to HVAC and clean areas, and list the routine monitoring parameters of clean areas with reasons.

5.2 Definitions

Read this section first. Every technical word used in this chapter is defined here in the simplest possible way.

- **Sterile:** A product or surface is sterile when it is completely free from all living microorganisms (bacteria, fungi, viruses) and their spores. Think of "sterile" as "zero germs, guaranteed."
- **Aseptic:** "Aseptic" means "made and handled in a way that keeps germs away." An aseptic process is a process designed so that the product never meets contamination in the first place. Aseptic processing is used when the product cannot be sterilized at the end (for example, some heat-sensitive drugs).
- **Terminal sterilization:** Sterilization of the product AFTER it is already sealed in its final container (for example, a filled and sealed vial heated in an autoclave). The word "terminal" means "at the end."
- **Sterilization:** Any process that kills or removes ALL microorganisms, including spores. Examples: steam under pressure (autoclaving), dry heat, radiation, filtration through bacteria-removing filters.
- **Pyrogen:** Any substance that causes fever (a rise in body temperature) when it enters the blood. The word comes from "pyro" (fire/heat) + "gen" (producer) — a "fever producer."
- **Endotoxin:** A toxic part of the outer wall of certain bacteria (Gram-negative bacteria). When these bacteria die and break apart, endotoxins are released. Endotoxins are heat-stable (hard to destroy)

and are the most common type of pyrogen in injectable products. So: all endotoxins are pyrogens, but not all pyrogens are endotoxins.

- **Schedule M:** The part of the Drugs and Cosmetics Rules (India) that lists the Good Manufacturing Practices and the requirements for factory premises, plant and equipment. It is the Indian rulebook for how a pharmaceutical factory must be built and run.
- **cGMP (current Good Manufacturing Practice):** The set of rules, systems and practices that make sure medicines are consistently produced and controlled to quality standards. The word "current" means the company must follow today's best practices, not yesterday's. In India, Schedule M is the national expression of GMP.
- **Cleanroom:** A specially designed and controlled room in which the concentration of airborne particles (dust, germs) is kept below defined limits, and other conditions (temperature, humidity, air pressure) are controlled. Medicines that go directly into the blood (injectables) are made here.
- **Particle:** A tiny solid piece floating in air — dust, skin flakes, clothing fibres, droplets carrying microbes. Particles are the enemy of sterile manufacturing because microbes ride on them.
- **HVAC (Heating, Ventilation and Air Conditioning):** The system of machines and ducts that supplies clean, temperature- and humidity-controlled air to the facility. In a sterile plant, HVAC is the "lungs" of the building.
- **AHU (Air Handling Unit):** The big machine that takes air, filters it, cools or heats it, adjusts its humidity, and pushes it through ducts into the rooms.
- **HEPA filter (High-Efficiency Particulate Air filter):** A very fine filter that removes an extremely high proportion of tiny particles from air. It is the heart of cleanroom air supply. (Exact efficiency ratings are given in the filter specification and current standards — confirm the latest edition.)
- **Filter integrity test:** A test that proves a filter has no holes, cracks or leaks. The DOP/PAO test is the standard concept for this — it challenges the filter with a fine mist of test aerosol on one side and checks whether any particles leak through to the other side.
- **SIP (Sterilize-in-Place):** A method of sterilizing equipment (tanks, pipes, filling lines) with steam WITHOUT taking the equipment apart. The sterilization happens "in place" — right where the equipment stands.
- **CIP (Clean-in-Place):** The partner of SIP: cleaning equipment without dismantling it, by circulating cleaning solutions through pipes and tanks. Usually CIP comes first, then SIP.
- **Airlock:** A small buffer room with two doors (only one door opens at a time) between areas of different cleanliness. It "locks" the air so contamination cannot flow directly from a dirtier area to a cleaner one.
- **Gowning:** The special procedure of wearing sterile protective clothing (coveralls, hood, mask, gloves, boots) before entering a cleanroom. Gowning is a trained skill, not just "wearing clothes."
- **Personnel flow:** The designed path that people follow when they enter, move inside, and exit the facility — arranged so people (the biggest source of contamination) cannot carry dirt into clean areas.
- **Material flow:** The designed path that raw materials, packaging components and finished goods follow through the facility — arranged so materials move in one direction and never cross back.
- **Unidirectional flow:** A layout rule that says people and materials must move in ONE direction — from less-clean areas toward more-clean areas, never backwards. Like a one-way street.

- **Zoning:** Dividing the facility into zones of different cleanliness levels, with the strictest zone where the sterile product is exposed to air.
- **Segregation:** Physically separating operations, materials or people so that mix-ups and cross-contamination cannot happen. For example, keeping different products' components in separate, labelled areas.
- **Grade:** The name given to a cleanliness level of a zone (the EU/WHO GMP system uses grades such as Grade A, B, C, D, with Grade A being the most critical — the zone where the sterile product is directly exposed). Exact particle and microbial limits for each grade are given in the current regulatory guideline — always confirm the latest edition.
- **At rest:** The condition of a cleanroom when production equipment is installed and running as agreed, but NO people are working and NO product is being made. It measures what the room itself can achieve.
- **In operation:** The condition of a cleanroom during actual production — equipment running, people working, product being made. This is the real, hardest test of the cleanroom.
- **Qualification:** Documented proof that equipment or a facility works correctly and is fit for its purpose. The four stages are DQ, IQ, OQ and PQ (defined below).
- **DQ (Design Qualification):** Documented proof that the DESIGN of the facility/equipment is correct and meets the user's needs — checked on paper before anything is built.
- **IQ (Installation Qualification):** Documented proof that the equipment/facility was INSTALLED correctly — the right machine, connected the right way, in the right place.
- **OQ (Operational Qualification):** Documented proof that the equipment/facility OPERATES correctly across its expected range — it does what it should when switched on and run.
- **PQ (Performance Qualification):** Documented proof that the equipment/facility consistently PERFORMS correctly under real, routine production conditions.
- **Validation:** The overall documented program proving that a process consistently produces a product meeting its specifications. Qualification of facilities/equipment is one part of validation.
- **Pressure differential:** The difference in air pressure between two adjacent rooms. Cleanrooms are kept at HIGHER pressure than surrounding areas so that when a door opens, clean air flows OUT and dirty air cannot flow IN.
- **Air changes:** The number of times per hour the total volume of air in a room is replaced with fresh filtered air. More air changes = cleaner air (but also higher cost).
- **Monitoring:** Regular, planned checking and recording of cleanroom conditions (particles, microbes, pressure, temperature, humidity) to prove the area stays in control.

5.3 Terminology Table

Term	Simple meaning
Sterile	Completely free from all living germs
Aseptic	Made/handled so germs never get in
Terminal sterilization	Sterilizing the sealed final container

Term	Simple meaning
Pyrogen	Substance that causes fever if injected
Endotoxin	Fever-causing toxin from dead Gram-negative bacteria
Schedule M	India's GMP rulebook for factory premises and equipment
cGMP	Current best practices to make quality medicines consistently
Cleanroom	Room with controlled, very clean air
HVAC	System that supplies clean, conditioned air
AHU	Machine that filters, heats/cools and moves air
HEPA filter	Extremely fine air filter that traps tiny particles
DOP/PAO test	Test proving a HEPA filter has no leaks
SIP	Sterilizing equipment with steam without dismantling it
CIP	Cleaning equipment without dismantling it
Airlock	Small buffer room with interlocked doors between zones
Gowning	Wearing sterile protective clothing in the correct way
Unidirectional flow	One-way movement of people and materials
Grade	Named cleanliness level of a zone (A strictest, D least)
At rest	Room tested with equipment on, but no people or production
In operation	Room tested during actual production with people working
DQ / IQ / OQ / PQ	Prove design, installation, operation, performance — in that order
Pressure differential	Air-pressure difference that pushes clean air outward
Air changes	How many times per hour room air is replaced

5.4 Detailed Explanation (From Absolute Zero)

5.4.1 Sterile manufacturing — the big picture

Technical concept: Sterile manufacturing is the production of medicines that must be free from microorganisms and pyrogens, such as injections, eye drops and surgical implants.

Simple explanation: Most medicines we swallow (tablets, syrups) do not need to be sterile — our stomach acid kills most germs. But an injection goes DIRECTLY into the blood. Blood has no defences waiting at the door. If even a few germs or fever-causing toxins ride along with the injection, the patient can get a serious infection or a dangerous fever reaction. That is why injectable products must be **sterile** (no living germs) and **pyrogen-free** (no fever-causing substances).

Example: A 100 mL IV fluid bag for a dengue patient. If that bag contains even a small number of bacteria, the bacteria multiply in the warm fluid and the patient receives a "soup of germs" straight into the veins. This can cause septicemia (blood infection), which can be fatal. Hence every sterile product batch is tested for sterility and for endotoxins before release.

Text diagram — the two routes to a sterile product:

```
Route 1: TERMINAL STERILIZATION (preferred when possible)
  Make the drug solution → Fill into vials → SEAL the vials
      → Sterilize the SEALED vials (steam/radiation) → Done
  (The container protects the product during sterilization.)

Route 2: ASEPTIC PROCESSING (when the drug can't take heat)
  Sterilize the solution by FILTRATION → Sterilize the empty
  containers and closures separately
      → Fill + seal inside a highly controlled cleanroom
        with almost no human touch → Done
  (Here the PROCESS must keep germs away at every step.)
```

Key takeaway: Sterile products are made either by sterilizing the sealed container at the end (terminal sterilization) or by assembling everything under germ-free conditions (aseptic processing). Both routes need special facilities — this chapter is about those facilities.

5.4.2 Why pyrogens and endotoxins matter

Technical concept: Pyrogens, especially bacterial endotoxins, must be controlled in sterile products because they survive many sterilization processes.

Simple explanation: Killing germs is only half the battle. When Gram-negative bacteria die, their broken cell walls release **endotoxins** — chemical pieces that are very stable. Normal steam sterilization kills the bacteria but does NOT reliably destroy endotoxins. If these enter the blood, the patient gets fever, chills, and in severe cases shock.

Example: Water for Injection (WFI) — the special water used to make injections — is tested for endotoxins with the LAL/BET test (a test using horseshoe-crab blood extract that clots in the presence of endotoxin). If endotoxins are found above the allowed limit, the water cannot be used. This is why "sterile" alone is not enough — the product must also be "pyrogen-free."

Key takeaway: A sterile product can still harm a patient if it contains endotoxins. Facility design must therefore control both microbes AND their toxins — through proper water systems, validated depyrogenation (e.g., dry heat for glassware) and clean processes.

5.4.3 Schedule M and cGMP layout requirements for sterile facilities

Technical concept: Schedule M (Drugs and Cosmetics Rules, India) and cGMP principles require that sterile manufacturing premises be designed, laid out and maintained to minimize contamination risks and permit effective cleaning.

Simple explanation: The law does not just say "make good medicines" — it says "build your factory in a way that MAKES contamination almost impossible." Think of it like a hospital operation theatre: the building itself helps keep things safe.

General layout principles (cautious, principle-based):

1. **Logical, orderly layout:** Rooms are arranged in the sequence of production steps so materials flow forward smoothly. No zig-zagging, no backtracking.
2. **Adequate space:** Enough room for equipment, people and materials so nothing is cramped. Cramped rooms = accidental mix-ups and poor cleaning.

3 **Smooth, cleanable surfaces:** Walls, floors and ceilings must be smooth, without cracks or crevices where dust and germs can hide, and must withstand repeated cleaning and disinfection.

4 **Separation of operations:** Different operations that could contaminate each other are kept in separate areas or at separate times. No open product in a corridor where people walk with street dust.

5 **Controlled entry:** Entry of people and materials into clean areas is through defined, controlled points (change rooms, airlocks, pass boxes) — never through a casual door.

6 **Services and utilities placed sensibly:** Pipes, drains and ducts are designed so they cannot become sources of contamination (e.g., drains must not allow back-flow into clean areas).

7 **Maintenance without contamination:** The design allows maintenance work without bringing dirt into production areas.

Example: In a well-designed sterile plant, the raw-material store is at one end of the building, the clean manufacturing core is in the centre, and finished goods leave from the other end. Workers change clothes and wash before entering the core, and nobody walks from the packing hall back into the filling room without re-gowning.

Key takeaway: Schedule M/cGMP layout is "prevention by design" — the building's shape, surfaces and flows are the first line of defence against contamination. (For exact clause/section numbers, refer to the current edition of Schedule M — confirm the latest revision.)

5.4.4 The cleanroom concept

Technical concept: A cleanroom is an enclosed area where airborne particle concentration, temperature, humidity and pressure are controlled to defined levels.

Simple explanation: A normal room has millions of dust particles floating in every cubic metre of air, plus germs riding on them. A cleanroom is a room that has been "tamed" — special air systems constantly sweep particles away, and strict rules control what (and who) enters. It is like creating a small island of super-clean air inside a normal factory.

Why it is needed: Sterile products are filled into open vials and bottles. During those few seconds when the container mouth is open, the product is exposed to room air. If that air is dirty, the product becomes dirty. The cleanroom makes sure the air touching the open product is as clean as technically possible.

Example: Filling an eye-drop bottle. The bottle mouth stays open for a few seconds while the dropper liquid goes in. In a cleanroom, the air blowing over that open mouth has been HEPA-filtered and flows in one smooth direction (laminar/unidirectional flow), pushing any stray particle away from the product.

Key takeaway: The cleanroom protects the product during the moments it is exposed to air. It does this with filtered air, controlled pressure, controlled climate and strict discipline about entry.

5.4.5 SIP — Sterilize-in-Place

Technical concept: SIP is the sterilization of fixed process equipment (vessels, pipelines, filling manifolds) using saturated steam, without dismantling the equipment.

Simple explanation: Imagine a long pipeline carrying sterile drug solution from a mixing tank to the filling machine. You cannot put a 50-metre pipeline into an autoclave. SIP solves this: you close the system,

pump in steam at sterilizing temperature, hold it for the validated time, then cool it down — all without opening a single joint. The equipment is sterilized exactly where it stands.

Where SIP is used: Product pipelines, holding tanks, filtration housings, filling machine product-contact paths, WFI distribution loops, and bioreactor/fermenter vessels.

Typical sequence (concept):

```
1. CIP — clean the system with cleaning solutions (remove residues)
2. Drain and prepare — remove water, close vents, check the system is sealed
3. Heat-up — introduce clean steam, vent air (air pockets block sterilization)
4. Sterilizing hold — maintain sterilizing temperature for the validated time
5. Cool-down — cool with sterile air or sterile water, keeping the system closed
6. Use — the system is now sterile and ready for the sterile product
```

Key takeaway: SIP = steam sterilization without dismantling. It is essential for any fixed piping that carries sterile fluids, and it always follows CIP (you must clean before you sterilize — steam cannot sterilize through dirt).

5.4.6 Material flow and personnel flow

Technical concept: Material flow and personnel flow are the designed, documented, one-way paths for materials and people through the facility, arranged to prevent contamination and mix-ups.

Simple explanation: In a sterile factory, WHERE you walk and WHERE materials travel is as important as WHAT you do. People are the biggest contamination source — one person sheds thousands of skin particles per minute. So the factory is designed like a fortress with controlled gates.

Personnel flow — entry procedure (step by step):

```
Street clothes area
      ↓ (remove outdoor shoes, outer garments)
First change room — put on factory uniform + dedicated shoes
      ↓
Hand wash
      ↓
Second change room (cleaner side) — GOWNING:
  sterile coverall → hood → mask → sterile gloves → boots
      ↓ (through an AIRLOCK with interlocked doors)
Cleanroom — work
      ↓
Exit: reverse route — remove gowning in the change room,
  never carry cleanroom garments outside
```

Gowning concept: Gowning is not "putting on clothes" — it is a validated procedure. The order of dressing, the way gloves are put on without touching the outer surface, and the rule that gowning happens in progressively cleaner change rooms are all defined and trained. Operators are even tested (gowning qualification) to prove they can dress without contaminating the garments.

Material flow — entry procedure:

```
Receiving → sampling/dispensing (weighed in a controlled area)
      ↓
De-cartoning / wipe-down (outer packaging removed OUTSIDE the clean area)
      ↓
Pass-through chamber or pass box (materials passed into the
   cleaner zone WITHOUT people walking in)
      ↓
Clean staging → production use
```

Finished goods flow the opposite way — from the clean core outward to packing and dispatch — again without backtracking.

Airlocks — define simply: An airlock is a small room between two areas of different cleanliness with a door on each side. The two doors are **interlocked** — both cannot be open at the same time. You enter, close the first door, and only then open the second. This traps a pocket of air and stops dirty air from rushing into the clean zone.

Unidirectional flow: People move from the dirtiest allowed point toward the cleanest point and never go backwards. Materials move from receiving → production → packing in one direction. Think of a river: it flows one way, and you never swim upstream with dirty water.

Key takeaway: Design the paths so that people and materials always move from "less clean" to "more clean," cross through controlled gates (airlocks, pass boxes, change rooms), and never retrace their steps.

5.4.7 Zoning and segregation — the Grade concept

Technical concept: The facility is divided into zones (grades) of increasing cleanliness, with the strictest controls where the product is exposed.

Simple explanation: Not every square metre of the factory needs to be equally clean — that would be impossibly expensive. So the factory is zoned like an onion: the outermost areas are normal factory areas, and as you move inward toward the filling machine, each zone is cleaner than the last. The very centre — where open vials meet the air — is the cleanest zone of all.

The Grade concept (introduction): International GMP (EU/WHO style) names these zones **Grades A, B, C and D**:

- **Grade A** — the critical zone: the point where the sterile product is directly exposed to air (e.g., the filling zone under unidirectional airflow). The strictest controls.
- **Grade B** — the background zone surrounding Grade A in aseptic processing (the room in which the Grade A zone sits).
- **Grades C and D** — progressively less critical zones for earlier steps (preparation, washing, component handling) where the product is not yet exposed or is in closed systems.

(Exact particle and microbial limits for each grade are defined in the current regulatory guideline — confirm the latest edition; do not use old numbers from memory.)

Segregation to prevent mix-ups and contamination:

- **Physical segregation:** Walls and doors separate different products, different strengths, and different stages. Labels and status tags ("QUARANTINE", "APPROVED", "REJECTED") segregate materials by quality status.
- **Time segregation:** When the same room must be used for different products, they are made at different times with full cleaning and line clearance in between.
- **Air segregation:** Pressure cascades and airlocks keep air (and the particles in it) from moving from dirty zones to clean zones.

Example: A facility makes two injectable products. Product X's raw materials are stored in a segregated, labelled bay; Product Y's in another. They are never weighed on the same day in the same room without documented cleaning and clearance between them. This segregation prevents a mix-up that could put the wrong drug in a vial.

Key takeaway: Zone the factory so cleanliness increases toward the product; segregate everything (space, time, air, labels) so neither contamination nor mix-ups can travel.

5.4.8 Cleanroom classifications and guidelines — FDA, EMA, WHO, ISO

Technical concept: Different regulatory bodies classify cleanrooms by how many particles (and microbes) are permitted in the air, but they use different systems and test conditions.

Simple explanation: Just as different countries use different grading systems for exams, different regulators describe cleanroom cleanliness differently. What matters is the IDEA behind all of them: count the particles in the air, and the fewer the particles, the cleaner the room.

- **ISO (International Organization for Standardization):** ISO 14644 is the international standard for cleanroom classification. It defines **ISO Classes** based on the concentration of airborne particles — a lower class number means a cleaner room. (Class numbers and their particle-concentration limits are given in the latest ISO 14644 edition — confirm the current edition; do not quote numbers from memory.)
- **EU/EMA (European Union):** Uses the **Grade A/B/C/D** system described above, with requirements stated for both "at rest" and "in operation" conditions.
- **FDA (USA):** The FDA's aseptic-processing guidance describes clean areas and their controls, historically referencing ISO classes for air cleanliness, with emphasis on the critical zones where sterile product is exposed.
- **WHO (World Health Organization):** WHO GMP gives cleanroom requirements aligned with the Grade concept, widely followed by manufacturers supplying global markets and referenced in many national regulations.

"At rest" vs "in operation" — the concept: A cleanroom is tested in two states. "At rest" (equipment installed and running, but no people, no production) shows what the room and its HVAC can achieve on their own. "In operation" (real production, people working) shows what happens in real life — and real life is always dirtier, because people shed particles. Both states must meet their respective requirements. A room that passes "at rest" but fails "in operation" has a people-discipline or airflow problem, not a machine problem.

Key takeaway: FDA, EMA, WHO and ISO all classify cleanrooms by permitted particle levels but use different naming systems. Always test and qualify the room both "at rest" and "in operation," and always check the CURRENT edition of the guideline — limits are revised over time.

5.4.9 HVAC — significance, components, HEPA filters

Technical concept: HVAC supplies cleanrooms with filtered, temperature- and humidity-controlled air at the right pressure, and is the primary engineering control against airborne contamination.

Simple explanation: If the cleanroom is the island of clean air, HVAC is the machine that CREATES the island. It constantly pumps in fresh, filtered, cool, dry air and pushes old air out. Without HVAC, a cleanroom is just an expensive ordinary room.

Why HVAC is the heart of a sterile facility:

1. **Removes particles:** Continuous supply of filtered air dilutes and sweeps away particles shed by people and equipment.
2. **Controls pressure:** Keeps cleaner rooms at higher pressure than dirtier neighbours (pressure cascade), so air always flows outward from clean to less-clean.
3. **Controls temperature and humidity:** Operators in full gowns get hot; many products and microbes are sensitive to heat and moisture. Cool, dry air keeps both people and product safe.
4. **Provides unidirectional airflow:** In the most critical zones, air flows in one smooth direction (like a gentle waterfall of clean air) over the exposed product.

Components — defined simply:

- **AHU (Air Handling Unit):** The big box (usually on the roof or in a plant room) containing fans, cooling/heating coils and filter sections. It "handles" the air: filters it, cools/heats it, dries or humidifies it, then blows it into the ducts.
- **Filters (pre-filters and fine filters):** A series of filters of increasing fineness. Coarse pre-filters catch big dust and protect the expensive fine filters downstream; fine filters polish the air before the final HEPA stage.
- **HEPA filters (terminal filters):** The final, extremely fine filters mounted at the room ceiling or in the AHU, which remove the tiniest particles from the supply air.
- **Ducts:** The metal "pipes" (usually rectangular) that carry conditioned air from the AHU to each room and carry return air back.
- **Dampers:** Adjustable flaps inside the ducts that control how much air goes to each room — like taps controlling water flow. Used to balance the system and set pressure differentials.
- **Controls (sensors and controllers):** Temperature, humidity and pressure sensors in the rooms, connected to controllers that automatically adjust the AHU (more cooling, more airflow) to hold conditions steady. Often monitored through a BMS (Building Management System — see Chapter 6).

HEPA filters — efficiency concept: A HEPA filter is a dense mat of extremely fine fibres. Air is forced through it, and particles get trapped by the fibres through several physical mechanisms (they crash into fibres, get stuck by interception, or wander into fibres by diffusion). The result: the air coming out is dramatically cleaner than the air going in. Filters are rated by their efficiency at capturing the most

penetrating particle size — the exact ratings are in the filter's specification and current standards (confirm the latest edition).

Integrity testing — the DOP/PAO test concept: A HEPA filter is useless if it has a pinhole leak — dirty air would stream straight through the hole. The integrity test checks for leaks WITHOUT quoting pass values here:

```
Concept of the DOP/PAO test:
1. Generate a fine mist of test aerosol (DOP or PAO — oily liquids
   that form uniform tiny droplets) UPSTREAM of the filter.
2. Scan the DOWNSTREAM side (the clean side) with a detector probe,
   moving slowly across the whole filter face and its seals/frame.
3. If the detector finds aerosol downstream, there is a LEAK —
   the filter or its gasket must be repaired or replaced.
```

The test proves the filter is intact and properly seated. It is done at installation and then periodically, and after any work that could disturb the filter.

Key takeaway: HVAC creates and protects the cleanroom environment through filtered air supply, pressure control and climate control. HEPA filters do the final cleaning of air, and the DOP/PAO test proves they have no leaks.

5.4.10 Qualification and validation of clean areas (DQ/IQ/OQ/PQ)

Technical concept: Before a cleanroom is used for production, documented evidence must prove it was correctly designed, installed, operates correctly and performs consistently.

Simple explanation: You would not trust a new bridge without an engineer's certificate. Similarly, a company cannot simply build a cleanroom and start filling injections — it must PROVE, on paper, that the room does what it claims. This proof comes in four stages, in order:

Applied to HVAC/cleanroom:

1. **DQ — Design Qualification:** "Is the design right?" Before construction, engineers check the drawings and specifications against what production needs: Is the room big enough? Is the AHU capacity correct? Are the grades and flows logical? Approved on paper first.
2. **IQ — Installation Qualification:** "Was it built right?" After construction, verify: Is this the specified AHU model? Are the HEPA filters the specified type and correctly mounted? Are ducts, dampers and sensors installed as per drawing? Calibration certificates collected.
3. **OQ — Operational Qualification:** "Does it work right?" With the system running (but no production), test across the operating range: Does the room reach the required temperature and humidity? Do pressure differentials hold when doors open and close? Do alarms work? HEPA integrity (DOP/PAO) testing is typically part of OQ.
4. **PQ — Performance Qualification:** "Does it keep working right, in real life?" Run the facility under actual production conditions over a period of time and prove that particle counts, microbial counts, pressure, temperature and humidity stay within requirements — both "at rest" and "in operation."

Example: A new sterile block is built. DQ approves the design on paper. IQ confirms the AHU, filters and ducts match the design. OQ shows the rooms achieve the required conditions with the system running. PQ runs three simulated production batches with operators in gowns and proves the environment stays in control throughout. Only then is the block released for commercial production.

Key takeaway: DQ → IQ → OQ → PQ is the logical chain: right design, right build, right operation, right performance in real use. No stage is skipped.

5.4.11 Routine monitoring parameters of clean areas

Technical concept: After qualification, clean areas are monitored routinely to prove they remain in a state of control during every production run.

Simple explanation: Qualification is like passing a driving test — monitoring is like following traffic rules every day afterwards. Conditions are checked continuously or at defined intervals, results are recorded, and any drift is investigated before product quality is affected.

What is monitored and why:

1. **Airborne particle counts:** Particles are counted in the air (with a particle counter) because particles carry microbes. Rising particle counts are the earliest warning that something is wrong — a torn gown, a faulty filter, too many people in the room.
2. **Microbial counts — air:** Air is sampled (with air samplers that grow any captured microbes on plates) to check for living organisms floating in the room air.
3. **Microbial counts — surfaces:** Swabs or contact plates pressed onto floors, walls and equipment surfaces check whether surfaces are picking up contamination.
4. **Personnel monitoring:** Glove prints and gown plates from operators check whether gowning and working discipline are effective — people are the main source, so people are monitored.
5. **Pressure differentials:** Checked (often continuously with gauges/alarms) to confirm clean rooms stay at higher pressure than adjacent areas. A lost pressure difference means dirty air can flow in.
6. **Temperature and relative humidity:** Checked because heat and moisture encourage microbial growth, affect operator comfort (sweating operators shed more particles) and can affect some products.
7. **Air changes / airflow:** Verified to confirm the HVAC is still delivering the designed air volume — a drop suggests a blocked filter or fan problem.

(Specific alert/action limits for each parameter are defined in the company's procedures based on the current regulatory guideline — confirm the latest edition; this chapter does not quote limits.)

Key takeaway: Monitor particles, microbes (air, surface, personnel), pressure, temperature/humidity and airflow — because each one is an early-warning signal, and catching drift early protects the batch.

5.5 Classifications (With Tables)

5.5.1 Classification of sterile manufacturing approaches

Approach	When used	Facility demand
Terminal sterilization	Product can withstand heat/radiation in sealed container	Standard clean areas; final sterilizer (autoclave) needed

Approach	When used	Facility demand
Aseptic processing	Product is heat-sensitive (proteins, some antibiotics)	Highest-grade cleanrooms, strictest controls, maximum discipline

5.5.2 Classification of cleanroom zones (Grade concept)

Grade	Cleanliness level	Typical use (concept)
Grade A	Most critical — product directly exposed	Filling zone, under unidirectional airflow
Grade B	Background to Grade A	Room surrounding the Grade A zone in aseptic processing
Grade C	Less critical	Preparation areas, washing of components
Grade D	Least critical of the classified zones	Initial handling, gowning support areas

(Note: exact limits per grade are in the current regulatory guideline — confirm the latest edition.)

5.5.3 Classification of cleanroom guidelines by issuing body

Body	System used	Key idea
ISO (ISO 14644)	ISO Classes by particle concentration	International, purely particle-based classification
EMA/EU GMP	Grades A–D, at rest and in operation	European regulatory system
FDA	Guidance on aseptic processing; references ISO classes	US regulatory expectations
WHO GMP	Grade-based, aligned with EU approach	Global reference, widely adopted

5.5.4 Classification of airlocks

Type	How it works	Used for
Personnel airlock (PAL)	People pass through; often with change benches	Entry/exit of operators
Material airlock (MAL)	Materials pass through; people do not walk through	Transfer of goods between zones
Pass-through chamber / pass box	Small chamber in the wall between zones	Small items, tools, samples

5.5.5 Classification of HEPA filter tests

Test	What it checks
Integrity (DOP/PAO) test	No leaks through filter media or seals
Efficiency rating (manufacturer)	Filtration performance of the filter type
Periodic re-test	Filter still intact after time and maintenance work

5.5.6 Classification of monitoring methods

Parameter	Typical method (concept)	Frequency idea
Airborne particles	Particle counter	During operation (critical zones, continuous or frequent)
Airborne microbes	Air sampler onto culture plates	Routine, as per schedule
Surface microbes	Swabs / contact plates	Routine, as per schedule
Personnel	Glove prints, gown contact plates	Each shift or per entry (critical zones)
Pressure differential	Gauges, often with alarms	Continuous
Temperature / humidity	Sensors, recorded	Continuous

5.6 Mechanisms / Process Flows (Step-by-Step)

5.6.1 How a cleanroom stays clean — the mechanism

1 The AHU draws in air and passes it through pre-filters, fine filters and HEPA filters.

2 Clean, conditioned air is blown into the room, often through ceiling HEPA filters.

3 The room is kept at higher pressure than surrounding areas, so air leaks OUTWARD, never inward.

4 Air sweeps particles toward low-level return grilles and back to the AHU for re-filtration.

5 In Grade A zones, air moves in one smooth direction (unidirectional flow) over the exposed product, carrying particles away.

6 People enter only through change rooms and airlocks, in full gowns, so they add minimum particles.

7 Continuous monitoring watches every parameter; alarms warn of any drift.

5.6.2 Personnel entry flow — step by step

1 Enter the outer change room in street clothes; remove outdoor footwear and outer garments.

2 Put on dedicated factory uniform and cleanroom shoes; cross the step-over bench (dirty side / clean side).

3 Wash and disinfect hands as per procedure.

4 Enter the next-stage change room; put on sterile gown in the defined order: coverall, hood, mask, boots, then sterile gloves (without touching outer glove surfaces).

5 Check appearance in the mirror — no skin or hair exposed.

6 Pass through the airlock (wait for one door to close before opening the next).

7 Enter the cleanroom; sanitize gloves at defined points during work.

8 On exit: leave through the airlock, remove gowning in the change room (never wear it outside), dispose or send for laundering as per procedure.

5.6.3 Material entry flow — step by step

1 Materials arrive at the receiving dock; outer cartons are removed OUTSIDE the clean zone.

2 Materials are wiped down or decontaminated as per procedure.

3 Items are placed into the pass-through chamber from the less-clean side; the door is closed.

4 Personnel on the cleaner side open the other door and collect the materials.

5 Materials move to clean staging, then to production — always forward, never backward.

5.6.4 SIP cycle — step by step

1 **CIP first:** Circulate cleaning solutions through the equipment to remove all product residues.

2 **Drain:** Remove rinse water; confirm the system is sealed and ready.

3 **Air removal:** Introduce steam and vent out air — trapped air prevents sterilization.

4 **Heat-up:** Bring all points of the system to sterilizing temperature.

5 **Hold:** Maintain temperature for the validated holding time (the coldest point must stay at temperature throughout).

6 **Cool-down:** Cool the system with sterile air or sterile cooling water while keeping it closed.

7 **Use or hold sterile:** Either use immediately for sterile product or maintain sterile condition until use.

5.6.5 HVAC air path — step by step

1 Fresh air + recirculated return air enter the AHU.

2 Pre-filters remove coarse dust; fine filters remove smaller particles.

3 Cooling/heating coils adjust temperature; humidifier/dehumidifier adjusts moisture.

4 The supply fan pushes conditioned air through ducts.

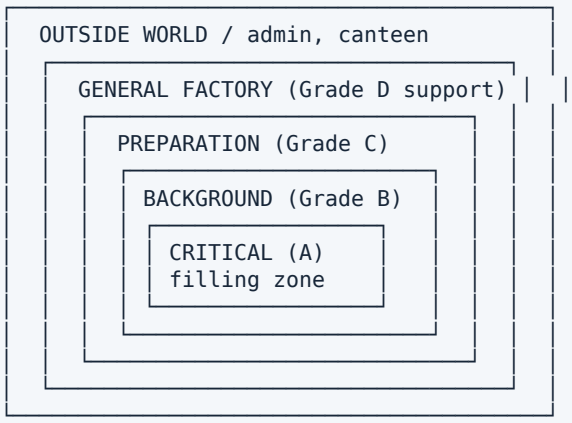
5 Dampers balance the airflow to each room.

6 Terminal HEPA filters give the final cleaning as air enters the room.

7 Room air, now carrying shed particles, exits through low-level return grilles back to the AHU — and the cycle repeats.

5.7 Text Diagrams / Flowcharts

5.7.1 Facility onion — zoning diagram



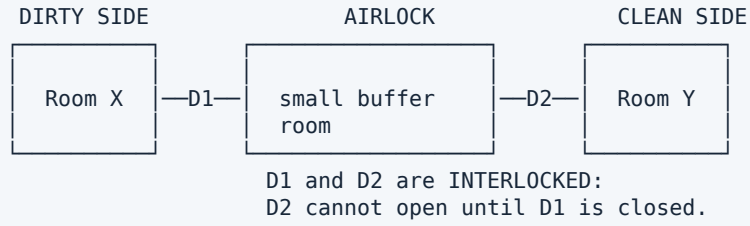
Cleanliness INCREASES toward the centre.
Pressure is HIGHEST at the centre, falling outward.

5.7.2 Pressure cascade diagram

```
Grade A (highest pressure)
   │  air flows out →
   ▼
Grade B (slightly lower)
   │  air flows out →
   ▼
Grade C (lower)
   │  air flows out →
   ▼
Grade D / corridor (lowest of classified areas)
   │  air flows out →
   ▼
Unclassified area

RULE: Air always flows from MORE clean to LESS clean.
Dirty air can never swim upstream against the pressure.
```

5.7.3 Airlock working



D1 and D2 are INTERLOCKED:
D2 cannot open until D1 is closed.

5.7.4 Qualification sequence

```
DQ (design right on paper)
        ↓
IQ (installed as designed)
        ↓
OQ (operates correctly across range)
        ↓
PQ (performs consistently in real production)
        ↓
ROUTINE MONITORING (stays in control every day)
```

5.8 Infographic Concepts

(Describe each as a visual idea for a designer or for the student's own sketch.)

1. **"The Onion Facility":** Concentric coloured rings from grey (outside) through light blue (D), medium blue (C), dark blue (B) to deep navy centre (A), with arrows showing people/materials flowing inward and air pressure arrows pointing outward.
2. **"One-Way Streets":** A floor-plan-style graphic with green one-way arrows for personnel flow and orange one-way arrows for material flow, with red "NO ENTRY" marks on reverse paths.
3. **"The Invisible River":** Unidirectional airflow shown as smooth parallel blue streamlines flowing down over an open vial, carrying red particle dots away from the product.
4. **"SIP Without Dismantling":** A pipeline with steam (orange wavy arrows) entering one end and condensate leaving the other, with a crossed-out wrench symbol meaning "no dismantling needed."
5. **"The Four Qualifications":** Four connected badges — DQ (blueprint icon), IQ (wrench icon), OQ (gauge icon), PQ (factory-in-action icon) — in a left-to-right chain.
6. **"Pressure Cascade Staircase":** Rooms drawn as descending steps from highest (Grade A, top step) to lowest, with air flowing down the steps like water over a waterfall.

5.9 Summary Tables

5.9.1 Chapter 5 at a glance

Topic	One-line essence
Sterile vs aseptic	Sterile = no germs; aseptic = process that keeps germs out
Terminal sterilization	Sterilize the sealed container at the end
Pyrogen / endotoxin	Fever-causers; endotoxins survive sterilization, must be controlled separately
Schedule M / cGMP	Design the building to prevent contamination
Cleanroom	Controlled island of clean air for exposed product
SIP	Steam-sterilize fixed equipment without dismantling
Personnel/material flow	One-way, gated paths; people are the biggest risk
Airlock	Interlocked double-door buffer between zones
Grades A–D	Onion zones; A = most critical
At rest vs in operation	Room alone vs room with people producing
HVAC	Creates and protects the clean environment
HEPA + DOP/PAO	Final air cleaning; test proves no leaks
DQ/IQ/OQ/PQ	Design → install → operate → perform, then monitor
Monitoring	Particles, microbes, pressure, climate, airflow — early warnings

5.9.2 Contamination sources and controls

Source	Control
People (skin, breath, clothes)	Gowning, change rooms, airlocks, training, personnel monitoring
Air (dust, outdoor air)	HVAC, HEPA filters, pressure cascade, airlocks
Materials/packaging	De-cartoning outside, wipe-down, pass boxes
Equipment	CIP/SIP, validated sterilization, qualification
Water	WFI system, endotoxin control, validated distribution
Pests	Sealed building, pest-control program, no inward back-flow from drains

5.10 Worked Examples and Calculations

All numbers below are **illustrative examples** invented only to show HOW a calculation is done. They are NOT regulatory requirements. Real acceptance values come from the current guideline and the company's validated design — confirm the latest edition.

Worked Example 1 — Air-change rate (illustrative)

Question: A cleanroom has a volume of 120 cubic metres. The AHU supplies 3,600 cubic metres of air per hour to this room. What is the air-change rate?

Step-by-step:

1 Formula: Air changes per hour (ACH) = Air supplied per hour ÷ Room volume.

2 Air supplied = 3,600 m^3/h. Room volume = 120 m^3.

3 ACH = 3,600 ÷ 120 = 30 air changes per hour.

4 Meaning: the entire air volume of the room is replaced 30 times every hour — once every 2 minutes.

Interpretation: A higher ACH means particles are swept away faster. (This number is only an arithmetic illustration — actual ACH requirements for each grade are set by the current guideline and validated design.)

Worked Example 2 — Pressure cascade check (illustrative)

Question: Three adjacent rooms show these pressure readings on their gauges (illustrative values): Filling room = +15 units, Background room = +10 units, Corridor = +5 units (all relative to the outside). Is the cascade correct?

Step-by-step:

1 Rule: pressure must FALL as we move from the cleanest room to the least clean area.

2 Filling room (+15) > Background room (+10) > Corridor (+5). The sequence falls correctly.

3 Air will flow: Filling room → Background room → Corridor → outside. Clean air always pushes outward.

4 If the background room gauge ever read HIGHER than the filling room, the cascade would be broken and dirty air could enter the filling room — production must stop and the cause investigated.

Interpretation: You do not need to memorize numbers — you need to check the DIRECTION of the cascade. (Units and limits are illustrative; follow the site's validated values.)

Worked Example 3 — Particle-count trend (illustrative)

Question: A Grade B room's particle counts (illustrative) over three days: Day 1 = 200 units, Day 2 = 220 units, Day 3 = 900 units. What does this tell you?

Step-by-step:

1 Days 1–2 are stable and similar — the room is in its normal state.

2 Day 3 shows a sudden jump to roughly 4 times the normal value.

3 A sudden jump is an early warning: possible causes — a damaged HEPA filter, a door left open, extra people in the room, a torn gown, construction dust nearby.

4 Action: investigate immediately, check pressure readings and filter status, review who entered the room — BEFORE the next batch is filled.

Interpretation: Monitoring is about TRENDS, not single numbers. A sudden change is the alarm bell.

5.11 Analogies

1 **Cleanroom = operation theatre of the factory.** Just as a surgeon operates in a special germ-free theatre — not in a general ward — sterile products are filled in a special germ-free room, not in an ordinary hall.

2 **Pressure cascade = water flowing downhill.** Water never flows uphill on its own; similarly, air pushed by higher pressure in the clean room always flows "downhill" toward lower-pressure dirty areas, and dirty air can never climb back up.

3 **Airlock = the air-lock of a submarine.** A submarine has two hatches so the sea never rushes straight in. An airlock's two interlocked doors do the same job against dirty air.

4 **Gowning = a spacesuit for germs.** An astronaut's suit keeps space out; a sterile gown keeps the person's particles in. Both are worn in a strict order and checked before "the mission."

5 **HEPA filter = a mosquito net for dust.** A mosquito net's tiny holes stop mosquitoes but let air through. A HEPA filter's fibres stop microscopic particles but let clean air through — and the DOP/PAO test is like checking the net for holes.

6 **SIP = steaming a pressure cooker without opening it.** You do not dismantle a pressure cooker to sterilize it — steam does the job inside the closed vessel. SIP does the same for factory pipelines.

7 **DQ/IQ/OQ/PQ = building a house.** DQ: the architect's plan is approved. IQ: the house is built exactly as per the plan. OQ: all taps, switches and doors work when tested. PQ: the family lives in it for a month and everything still works fine.

8 **Monitoring = daily health check-up.** Qualification proved you were healthy once; monitoring is the daily check that you STAY healthy.

5.12 Pharmacy Relevance

- **Patient safety:** Every concept in this chapter exists for one reason — an injection or eye drop contaminated with germs or endotoxins can kill. The sterile facility is the pharmacist's promise of safety made concrete in bricks, steel and air.
- **Pharmacist's role:** Pharmacists in sterile manufacturing work as production officers, QA officers and validation executives. They approve gowning procedures, review monitoring trends, investigate deviations (e.g., a pressure alarm during filling), and decide whether a batch can be released.
- **Batch release decision:** If routine monitoring shows the cleanroom went out of control during filling (say, pressure cascade lost for 10 minutes), the pharmacist/QA must assess whether the batch is still safe — sometimes the whole batch is rejected. Understanding this chapter is understanding WHY that decision is made.
- **Aseptic technique in practice:** Hospital pharmacists preparing IV admixtures or chemotherapy doses use the same principles on a small scale — laminar airflow cabinets, gowning, disinfection, unidirectional work flow. This chapter is the large-scale version of what a clinical pharmacist does daily.
- **Regulatory inspections:** Drug inspectors (CDSCO/state FDA in India, US FDA, EU auditors) walk through sterile facilities checking exactly these topics — flows, grades, HVAC qualification, monitoring records. A pharmacist must be able to explain and defend each one.

5.13 India Context

- **Schedule M** of the Drugs and Cosmetics Rules is the Indian legal backbone for sterile facility design. Any Indian manufacturer of injectables must comply with it, and state drug control departments inspect against it.
- **Indian injectable industry:** India is one of the world's largest producers of vaccines and injectable medicines ("pharmacy of the world"). Large sterile facilities operate in and around Hyderabad, Ahmedabad, Mumbai, Bengaluru and the Delhi-NCR region, supplying both domestic hospitals and export markets.
- **Export expectations:** Indian sterile plants that export to the US or Europe must ALSO satisfy FDA/EMA expectations — which is why this chapter covers FDA, EMA, WHO and ISO systems together. A single Indian facility often works to all of them at once.
- **CDSCO and WHO prequalification:** Vaccines and injectables supplied to global programs often pass WHO prequalification, where facility design, HVAC qualification and monitoring are inspected in depth.
- **Practical note for students:** If you visit or train in an Indian sterile plant, you will see change rooms with step-over benches, Grade B/C/D signboards on doors, pressure gauges with green "normal" zones, and pass boxes between corridors and production rooms — every item in this chapter, in real life.

5.14 Key Points

1 Sterile products (injections, eye drops) must be free from microorganisms AND pyrogens/endotoxins.

2 Two manufacturing routes: terminal sterilization (sterilize the sealed container) and aseptic processing (keep germs out throughout).

3 Schedule M and cGMP require the facility itself to be designed to prevent contamination — layout, surfaces, flows and segregation.

4 A cleanroom is a controlled island of clean air; it protects the product when containers are open.

5 SIP sterilizes fixed equipment with steam without dismantling; CIP (cleaning) always comes first.

6 Personnel and material flows are one-way (unidirectional), gated by change rooms, airlocks and pass boxes; people are the biggest contamination source.

7 Zones are graded (A strictest → D least); cleanliness and pressure increase toward the product.

8 ISO, FDA, EMA and WHO classify cleanrooms differently but all control particle levels; rooms are qualified both "at rest" and "in operation."

9 HVAC is the heart of the sterile facility: AHU, filters (pre/fine/HEPA), ducts, dampers and controls supply clean, conditioned, pressurized air.

10 HEPA filter integrity is proven by the DOP/PAO test, which detects leaks.

11 Clean areas are qualified in sequence: DQ → IQ → OQ → PQ, then kept in control by routine monitoring.

12 Routine monitoring covers particles, microbes (air/surface/personnel), pressure differentials, temperature/humidity and airflow — trends are early warnings.

5.15 Common Confusions

1 **Sterile vs aseptic:** "Sterile" describes the STATE of the product (no germs). "Aseptic" describes the PROCESS (made without letting germs in). An aseptically produced vial should BE sterile — the words describe different things.

2 **Sterilization vs disinfection:** Sterilization kills ALL microbes including spores; disinfection only reduces them. A disinfected surface is NOT sterile.

3 **Pyrogen vs endotoxin:** All endotoxins are pyrogens, but not all pyrogens are endotoxins. Endotoxin = the specific bacterial-wall toxin; pyrogen = anything causing fever.

4 **SIP vs CIP:** CIP = CLEAN in place (remove dirt with chemicals). SIP = STERILIZE in place (kill germs with steam). CIP always comes before SIP.

5 **At rest vs in operation:** At rest = equipment on, no people, no production (tests the room). In operation = real production with people (tests real life). Both must pass.

6 **Grade A vs ISO Class:** Grade A/B/C/D is the EU/WHO GMP zone system; ISO Classes are the ISO 14644 particle-based system. Different naming systems for related ideas — do not mix them up.

7 **Qualification vs validation:** Qualification proves FACILITIES/EQUIPMENT are fit (DQ/IQ/OQ/PQ). Validation proves the whole PROCESS consistently makes good product. Qualification is part of validation.

8 **Airlock vs pass box:** An airlock is a walk-through buffer room for people (or large materials); a pass box is a small wall chamber for small items. Both have interlocked doors.

5.16 Common Mistakes

1. **Quoting limits from memory:** Writing a specific particle count, pressure value or air-change rate as "the requirement" without checking the current guideline edition. Limits change — always write "as per the latest guideline."
2. **Thinking sterilization destroys endotoxins:** Standard steam sterilization kills bacteria but does not reliably destroy endotoxins. Depyrogenation needs separate validated methods (e.g., dry heat for glassware).
3. **Skipping CIP before SIP:** Trying to sterilize dirty equipment. Steam cannot sterilize through layers of residue — clean first, then sterilize.
4. **Opening both airlock doors together:** Defeats the entire purpose of the airlock. Interlocks exist to prevent this — never bypass them.
5. **Walking backwards in flow:** Entering the cleanroom and then walking back out through the entry change room into a dirtier area and re-entering without re-gowning. Flow is one-way, always.
6. **Confusing DQ with PQ:** DQ is checked on PAPER before building; PQ is proven in REAL PRODUCTION after building. They are opposite ends of the qualification chain.
7. **Ignoring trends:** Seeing particle counts slowly rising week after week and doing nothing because "today's value is still okay." Trends are warnings — act early.
8. **Poor gowning discipline:** Touching the outer surface of sterile gloves, exposing wrists or hair, rushing through change rooms. The gown is a contamination barrier only if worn correctly.

5.17 Mnemonics

- **Grades A–B–C–D: "A Big Clean Door"** — A = most critical (filling), B = background, C = cleaner prep, D = door/entry support. Cleanliness DECREASES from A to D.
- **Qualification order DQ–IQ–OQ–PQ: "Design It, Qualify Operations Properly"** — Design → Installation → Operation → Performance. Or simply: **"Did I Order Pizza?"**
- **SIP before use: "Clean, Seal, Steam, Stay shut"** — CIP clean → seal the system → steam sterilize → keep it shut until use.
- **Monitoring parameters: "PMP-PTA"** — Particles, Microbes, Pressure — Plus Temperature, Airflow. ("Papa Makes Pure Products Through Testing Air.")
- **Contamination sources: "PEMAW"** — People, Equipment, Materials, Air, Water. People first — they are the biggest source.
- **Airlock rule: "One door at a time, every time."**
- **Flow rule: "Forward only, like a river."**

5.18 Exam Summary

Sterile manufacturing produces medicines (injections, eye drops) that must be free from microorganisms and pyrogens/endotoxins, because they enter the blood directly. Two routes exist: terminal sterilization (sterilize the sealed container) and aseptic processing (exclude germs throughout). Schedule M and cGMP demand prevention-by-design: logical layout, cleanable surfaces, segregation, and controlled one-way flows for people and materials. The cleanroom is the controlled island of clean air where exposed product

is protected. Fixed equipment is sterilized without dismantling by SIP (after CIP). People enter through change rooms with strict gowning; materials enter through pass boxes/chambers; airlocks with interlocked doors separate zones. Zones are graded A (most critical) to D, with cleanliness and pressure increasing toward the product. ISO, FDA, EMA and WHO classify cleanrooms by particle levels using different systems; rooms are qualified "at rest" and "in operation." HVAC (AHU, pre/fine/HEPA filters, ducts, dampers, controls) is the heart of the facility, creating clean, conditioned, pressurized air; HEPA integrity is proven by the DOP/PAO leak test. Clean areas are qualified DQ → IQ → OQ → PQ and then kept in control by routine monitoring of particles, microbes (air, surface, personnel), pressure differentials, temperature/humidity and airflow, watching trends as early warnings. (Exact numerical limits: as per the latest guideline edition — confirm current requirements.)

5.19 One-Minute Revision

- Sterile = no germs; aseptic = germ-free process; terminal sterilization = sterilize sealed container; endotoxins = fever toxins surviving sterilization.
- Schedule M/cGMP = design the building to prevent contamination.
- Cleanroom = controlled clean-air island; SIP = steam-sterilize without dismantling (CIP first).
- Flows are one-way: change rooms + gowning for people; pass boxes for materials; airlocks = interlocked double doors.
- Grades A→D: cleanliness increases toward the product; pressure cascades outward.
- At rest = room alone; in operation = real production. Both must pass.
- HVAC = AHU + filters + ducts + dampers + controls; HEPA = final filter; DOP/PAO = leak test.
- DQ→IQ→OQ→PQ, then routine monitoring: particles, microbes, pressure, temperature/humidity, airflow.
- People are the biggest contamination source. Trends are early warnings.

5.20 MCQs

Q1. A product is sterile when it is: A. Free from visible dirt B. Completely free from all living microorganisms C. Treated with a disinfectant D. Filtered once

Q2. Terminal sterilization means: A. Sterilizing raw materials before manufacturing B. Sterilizing the product in its sealed final container C. Sterilizing the cleanroom air D. Sterilizing operators' hands

Q3. Endotoxins are: A. Toxins produced by all bacteria that cause allergy B. Heat-stable fever-causing parts of Gram-negative bacterial walls C. Chemicals used to sterilize equipment D. Preservatives added to injections

Q4. In a sterile facility, an airlock is: A. A lock on the main gate B. A buffer room with two interlocked doors between zones of different cleanliness C. A type of HEPA filter D. A sterilizer for garments

Q5. Unidirectional flow means: A. Air flows in all directions equally B. People and materials move in one direction, from less-clean to more-clean areas C. Only one person may enter at a time D. Materials move in circles

Q6. In the Grade A–D system, the most critical zone (where sterile product is directly exposed) is: A. Grade D B. Grade C C. Grade B D. Grade A

Q7. "In operation" condition of a cleanroom means: A. Equipment installed but switched off B. Testing during actual production with people working C. Testing with equipment on but no people and no production D. The room under maintenance

Q8. The DOP/PAO test is used to: A. Measure room temperature B. Check HEPA filters for leaks C. Count microbes on surfaces D. Test water for endotoxins

Q9. The correct order of qualification stages is: A. PQ → OQ → IQ → DQ B. DQ → IQ → OQ → PQ C. IQ → DQ → PQ → OQ D. OQ → PQ → DQ → IQ

Q10. SIP stands for: A. Sterilize-in-Place — steam sterilization without dismantling equipment B. Standard Inspection Procedure C. Single-use Injection Pack D. Sterile Ingredient Powder

Q11. The biggest source of contamination in a cleanroom is: A. The AHU B. People C. The walls D. The lighting

Q12. Pressure differentials are maintained so that: A. Dirty air flows into clean rooms B. Clean rooms stay at higher pressure, so air flows outward to less-clean areas C. All rooms have equal pressure D. Air stops moving completely

Answer Key: Q1–B, Q2–B, Q3–B, Q4–B, Q5–B, Q6–D, Q7–B, Q8–B, Q9–B, Q10–A, Q11–B, Q12–B

5.21 Viva / Application Questions

1 Why must injectable products be free from pyrogens in addition to being sterile? Explain with the example of an IV fluid.

2 A new operator asks why she cannot simply walk from the packing hall into the filling room. Explain personnel flow, gowning and unidirectional flow to her in simple words.

3 During filling, the pressure gauge shows the Grade B room at higher pressure than the Grade A zone. What is wrong, and what should be done?

4 Explain why CIP must always come before SIP, using a real equipment example (e.g., a product pipeline).

5 What is the difference between "at rest" and "in operation" qualification? Why are both required?

6 Particle counts in a filling room show a steady rise over two weeks but are still "within limits." As QA officer, what is your response and why?

7 Compare terminal sterilization and aseptic processing: when would you choose each, and how do their facility requirements differ?

8 An inspector asks you to prove that the facility's HEPA filters are intact. Which test do you describe, and what does it check?

5.22 References

- ISO 14644 cleanroom standards (latest edition — confirm current requirements)
- WHO GMP guidelines for sterile pharmaceutical products
- FDA guidance for industry on sterile drug products produced by aseptic processing
- EU GMP Annex on manufacture of sterile medicinal products (latest revision)

- Drugs and Cosmetics Rules, India — Schedule M (current edition)