Clean Room Basics & Rules （A1043）

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Clean room is a clean environment designed to reduce the contamination of processes and materials. This is accomplished by removing or reducing contamination sources.

Principles of the Clean Environment:

(i) Air is highly HEPA- (High Efficiency Particulate Air) filtered (99.99% @ 0.3m);
(ii) Layout should minimize particle sources in filtered air stream;
(iii) Air flow should remove most particles generated by process.

Clean Room Classifications | Standards (*ISO 14644-1*):

Class	Maximum Particles/m³						FED STD 209E equivalent
	≥0.1 μm	≥0.2 μm	≥0.3 μm	≥0.5 μm	≥1 μm	≥5 μm	
ISO 1	10	2					
ISO 2	100	24	10	4			
ISO 3	1,000	237	102	35	8		Class 1
ISO 4	10,000	2,370	1,020	352	83		Class 10
ISO 5	100,000	23,700	10,200	3,520	832	29	Class 100
ISO 6	1,000,000	237,000	102,000	35,200	8,320	293	Class 1,000
ISO 7				352,000	83,200	2,930	Class 10,000
ISO 8				3,520,000	832,000	29,300	Class 100,000
ISO 9				35,200,000	8,320,000	293,000	Room Air

☞**Room A1043 is Class 10,000 cleanroom compliant with Class 1000 localized clean areas for the optical benches. Additionally, it features a precisely controlled environment with temperature maintained at 22.8°C ± 0.3°C, humidity at 46% ± 2%, and noise levels below 50 dB. Since the clean room is designed for optics and microscopy, *clean room* and *microscope room* will be used interchangeably hereafter.**

Types of Contamination:

- Particulate
  Dust, skin, hair, makeup…
- Chemical
  Oil, grease, metal ions, perfume…
- Biological
  Bacteria, fungi, cockroach???
- Radiation
  Ultraviolet light…

Particle Characteristics:

- 50 μm particles are visible
- Average human hair is about 100 μm
- Time to fall 1 meter in still air
  - 33 seconds for 10 μm particle
  - 48 minutes for 1 μm particle
- Humans generate $>1\times10^5$ particles per minute when motionless (fully gowned)
- Humans can generate $>1\times10^6$ particles per minute when walking in the cleanroom

Contamination Sources:

- People ~75%
- Ventilation ~15%
- Room Structure ~5%
- Equipment ~5%

Contamination Control:

- *Personnel Control*
  - Dress code
  - Personal Hygiene
  - Gowning
- *Environmental Control*
  - Entrance and exit
  - Materials and supplies
  - Cleaning and maintenance
  - Atmospheric

Dress code:

- No sleeveless shirts
- No shorts or skirts
- No slippers or sandals
- No jewelry allowed especially those that can reflect laser light
- Avoid clothing that sheds

Personal Hygiene:

- Shower each day before entry
- Control Dermatitis & Dandruff
- Do not smoke before entry
- No chewing gum or tobacco
- No Cosmetics should be worn
- Facial hair will need to be covered

Gowning:

Anti-static clean room suit is provided. Dress properly, do not expose your hair. Don't reach inside your garment while in the cleanroom.

Entry and Exit (*via* Air Shower Room):

Only people signed up in the scheduling system are allowed to enter the microscope room. Arrange experiments such that number of entrances should be minimized. The logbook should record each and every entrance and the reason. Unauthorized entry or unreasonable frequent re-entrances during experiments will be warned or incur temporary access ban.

Air Shower Room Logic: wait until the shower is done and inner door deadbolt pops open to enter the microscope room; wait (you may have to move around a bit) until the pressure sensor senses your weight and hear deadbolt locks up to exit the air shower area.

Materials and Supplies:

- Do Not carry non-cleanroom items into the cleanroom;
- Do not carry cleanroom items out of the cleanroom;
- Do not use pencils or erasers;
- Only one clean lab notebook is allowed during experiments;
- Printed materials or scratch papers are prohibited unless authorized;
- Do Not cut the cleanroom wipers;
- Clean everything you carry into the cleanroom;
- Trashcan should be emptied after each experiment;
- Do not use clean room as sample storage, the only exception may be your trusted calibration sample(s). Any samples left stored in the clean room should be clean with no cracks/glass debris or oil residues after each use.

Chemicals:

- Do not take new chemicals into the cleanroom without permission
- Currently the only chemicals allowed for storage in the microscope room are index matching oil for oil immersion objective and isopropanol in special small bottles.
- Allowed Chemicals for Experiments [Pay attention to volume restrictions]:
  (i) Neutral aqueous buffers: **< 50 mL**
  (ii) Acidic/Basic buffers (pH < 6.0 or pH > 8.4): **< 15 mL**
  (iii) Any buffer containing DMEM/phenol: **< 15 mL**
  (iv) Alcohols (Methanol/Ethanol/Isopropanol): **< 15 mL**
  (iv) Thiols (BME, MEA etc.), triplet state quenchers (COT etc.): **< 2 mL** (pure stock not recommended)
  (v) Nanoparticles, proteins (enzymes, antibodies), DNAs, antibiotics, dyes: **< 2 mL**

Clean Room On-Duty Checklist:

Empty trashcans: Please ensure trashcans are emptied after each experiment to prevent overflows. Use fresh trash bag each time. To minimize contamination, please avoid trash bags that shed or have strong odors. On-duty person should also make sure there is enough trash bag supply for at least another week.
Maintain a clean workspace: Keep tissues, parafilm, and pipette tips off the floor.
Cleanliness of optics: Inspect objectives and the optical table for oil residues and clean them promptly.
Dust control: Use packing tape to remove human hair, dandruff, or visible dust from any surface.
Maintain a professional environment: Kindly remind colleagues who violate clean room regulations. Repeated violations by others will not excuse the On-Duty Person's responsibility.
Documentation: Document any violations and report issues after each group meeting.