PhEn-602

Notes # 4

J. Manfredi

Clean Rooms and Controlled Environments

Basic definitions

- Clean Room: A room in which the concentration of airborne particles is controlled and contains one or more clean zones
- Clean Zone: A defined space in which the concentration of airborne particles is controlled to meet a specified airborne particulate class.

Clean Rooms and Controlled Environments

- Federal Standard 209E (FS-209E) provides Clean Room Classes
- Rooms classified based on number of particles > 0.5 micron per cubic foot
- Class descriptions still in use today.
- FS 209E is concerned about the following particle sizes, in microns:

  0.1, 0.2, 0.3, 0.5, 5.0

Clean Rooms and Controlled Environments

- Types of Contaminants
  - Viable Particulates
  - Non-Viable Particulates

Controlled Environments - Types of Contaminants

- Non-viable Particulates
  - Metal specks, fiber from clothing
  - Obtained from: Equipment, people, tools
- Viable (micro-organisms)
  - Bacteria
  - Yeast, molds
  - Obtained from: People, outside air, water, equipment, tools, excipients, active ingredients

Clean Rooms and Controlled Environments

Sources of particulate generation

- Internal:
  - Personnel
    - Normally the highest source of contamination
  - Process
  - Air conditioning system
  - Introduction of raw materials
  - Introduction of equipment and materials
- External
  - Outside air

Clean Rooms and Controlled Environments

Some interesting facts:

- Visible indoor air particles constitute only about 10% of particles present in indoor air.
- It may be possible to see particles as small as 10 microns
  - It may be possible to see particles as small as 10 microns under favorable conditions.
- The majority of harmful particles are 3 microns or less in size.
- Particles of 1 micron or less adhere to surfaces by molecular adhesion. Scrubbing is generally the only way to remove them.
- Larger particles tend to settle out of the atmosphere due to weight.

Clean Rooms and Controlled Environments

Some more interesting facts:

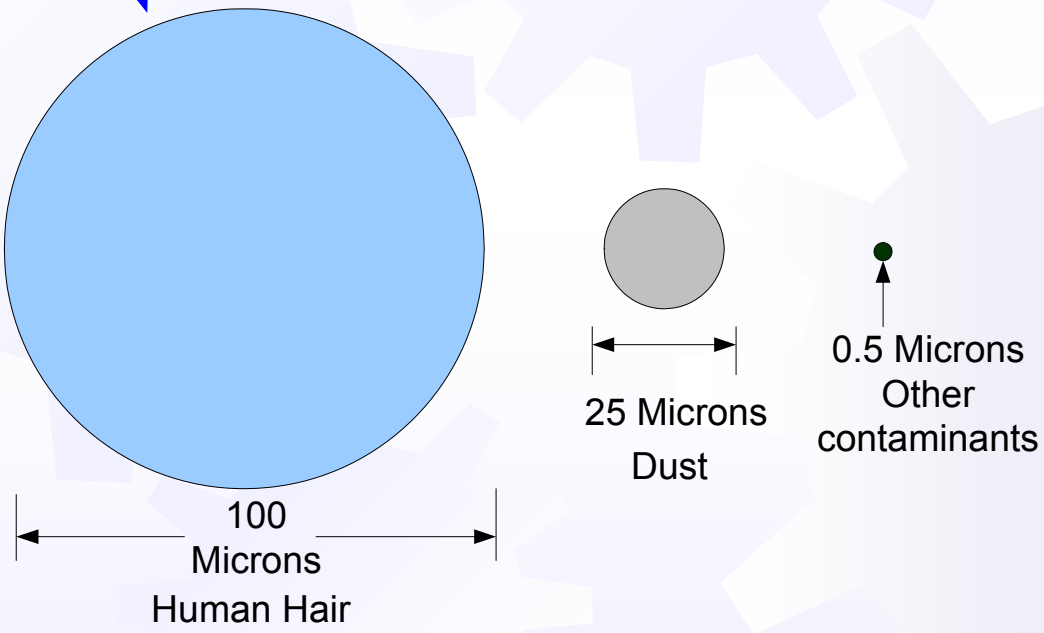
- Smaller, "respirable" particles remain virtually suspended in the air until breathed in.
- Approximately 98-99% of all particles by count are in the size range of 5 microns or less. These particles tend to remain in suspension or settle out so slowly.

From: www.peakpureair.com/particlesize.htm

Clean Rooms and Controlled Environments

Common items and their respective particle sizes:	Microns
Eye of a needle	1,230
Human Hair	40 – 300
Oil Smoke	0.03 to 1
Bacteria	0.3 to 20
Pollens	10 to 1,000
Coal Dust	1 to 100
Beach sand	100 to 2,000
Mold spores	0.5 to 20
Tobacco smoke	0.01 to 1
Typical Atmospheric Dust	0.001 to 30

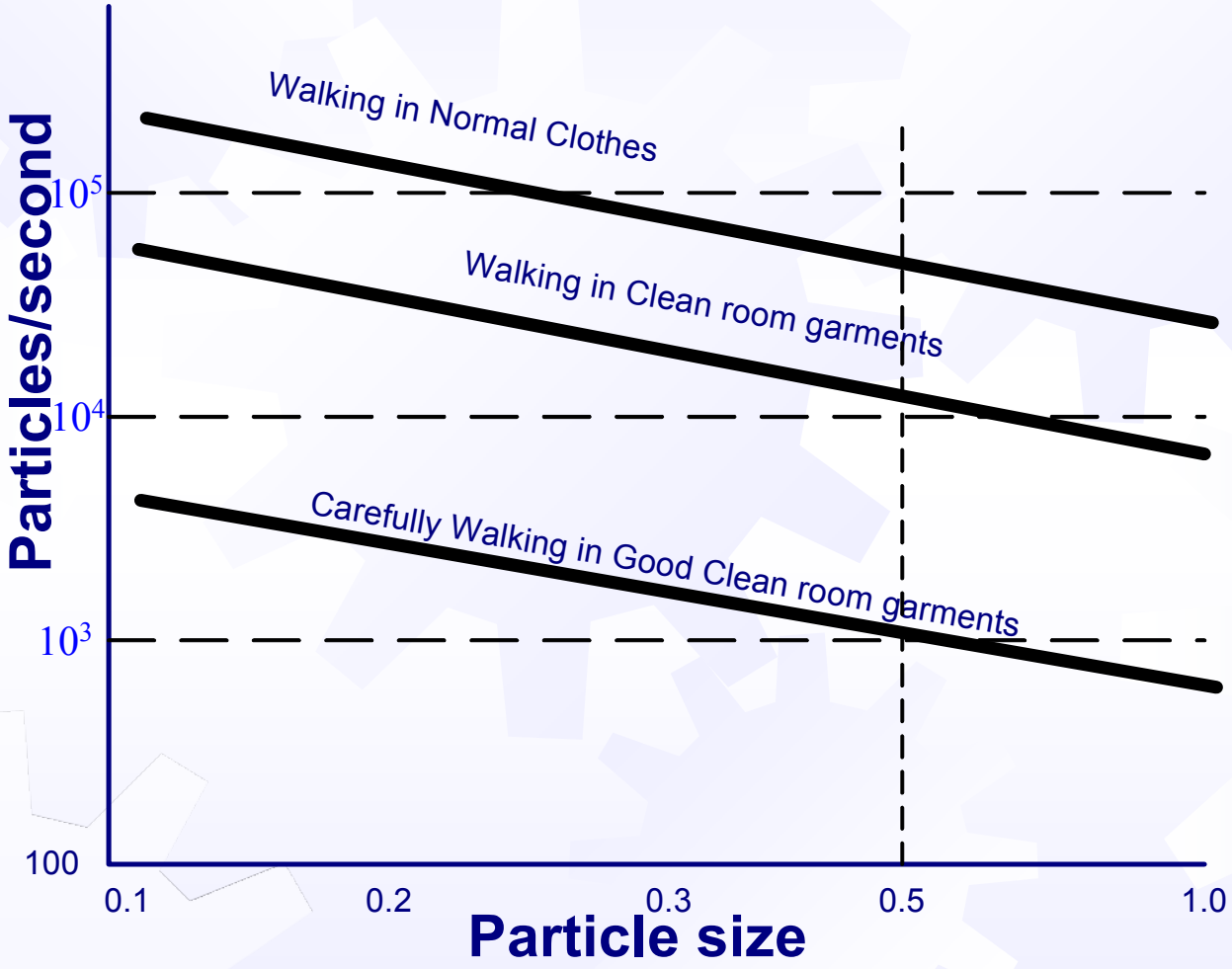
Clean Rooms and Controlled Environments



Personnel – Largest Source of Contamination

- People are huge sources of contamination - *the biggest source of viable and non-viable contamination*
- Each adult loses about 6 - 14 grams of dead skin material every day
- Each person loses a complete layer of skin about every four days - equivalent to 10,000,000 particles per day!
- Ordinary walking movements emit about 10,000 particles per minute.

Personnel – Largest Source of Contamination – Particle Generation Rate



Clean Rooms and Controlled Environments

- Occupancy state of the cleanroom:
  - As-Built: As constructed, with no equipment or personnel in room
  - At-Rest: Equipment in room, but no personnel
  - Operational: (also called “In-Operation”) Personnel and equipment in room, under normal operations

Clean Rooms and Controlled Environments

FDA Aseptic Guidelines – 1987

Requires pharmaceutical companies to use the FS-209E classifications for aseptic manufacturing.

Non-viable particle levels must meet the FS 209E classes.

Concerned only with the “in-operation” condition.

Concerned only with particles greater than or equal to 0.5 microns.

Contains limits for viable particles also – to be discussed later.

Website: www.fda.gov/cder/guidance /old027fn.pdf

Clean Rooms and Controlled Environments

FDA Aseptic Guidelines – 1987

Controlled Area:

"A controlled area is one in which unsterilized drug product, in-process materials or containers/closures are prepared….. acceptable air quality if it has a per-cubic-foot particle count of not more than 100,000 in a size range of 0.5 micron and larger (Class 100,000) when measured in the vicinity of the exposed articles during periods of activity.

Clean Rooms and Controlled Environments

FDA Aseptic Guidelines – 1987

Critical Area:

"A critical area is one in which the **sterilized drug product, containers, and closures are exposed** to environmental conditions….**acceptable air quality of Class 100 in a size range of 0.5 micron or larger when measured no more than 1 ft away from work site…**"

Clean Rooms and Controlled Environments

- **FDA Aseptic guidelines:** section 4, Buildings and Facilities: "Clean area control parameters should be supported by microbiological and particle data obtained during qualification studies. Initial cleanroom qualification includes, in part, an assessment of air quality under as-built, static conditions. It is important for area qualification and classification to place most emphasis on data generated under dynamic conditions (i.e., with personnel present, equipment in place, and operations ongoing). An adequate aseptic processing facility monitoring program also will assess conformance with specified clean area classifications under dynamic conditions on a routine basis".

FDA Aseptic Guide – Class of Clean Room

- **Supporting Clean Areas**
- Supporting clean areas can have various classifications and functions. Many support areas function as zones in which nonsterile components, formulated products, in-process materials, equipment, and container/closures are prepared, held, or transferred. These environments are soundly designed when they minimize the level of particle contaminants in the final product and control the microbiological content (bioburden) of articles and components that are subsequently sterilized.

FDA Aseptic Guide – Class of Clean Room

- **Supporting Clean Areas**
- The nature of the activities conducted in a supporting clean area determines its classification. ***FDA recommends that the area immediately adjacent to the aseptic processing line meet, at a minimum, Class 10,000 (ISO 7) standards under dynamic conditions***. Manufacturers can also classify this area as Class 1,000 (ISO 6) or maintain the entire aseptic filling room at Class 100 (ISO 5). An area classified at a Class 100,000 (ISO 8) air cleanliness level is appropriate for less critical activities (e.g., equipment cleaning).

Clean Rooms and Controlled Environments

FDA Aseptic Guidelines – as mentioned:

Originally published in 1987

Revised - September 2004

"Guidance for Industry – Sterile Drug Products Produced by Aseptic Processing"

http://www.fda.gov/cder/guidance/5882fnl.htm

These are Guidelines – not regulations

(21 CFR 210 and 211 are regulations/law).

However, as far as manufacturers are concerned, they are as important as the law, since it represents FDA's current thinking and expectations.

Section IV: Buildings and Facilities

*important guidelines re: aseptic facility design

Clean Rooms and Controlled Environments

- Federal Standard 209E Cleanroom Classes

Class (English)	Max. # Particles/ft³ of a size ≥ 0.5 m
1	1
10	10
100	100
1,000	1,000
10,000	10,000
100,000	100,000

Clean Rooms and Controlled Environments

The development of ISO standards:

- Before 2000 each country developed its own cleanroom standards
- US followed Federal Standard 209E
- Several European Countries assembled as part of CEN - Committee for European Normalization - to standardize cleanroom standards – CEN standards were initiated
- Prior agreement between ISO and CEN
  - ISO standards would take precedence

Clean Rooms and Controlled Environments

Cleanroom Standards:

- FS 209E - The traditional cleanroom standard for all industries.
- **Obsolete as of November 2001, replaced by ISO standards**
- FS 209E still being used by many…..
- Industry currently uses both 209E and ISO classifications in the design of today's cleanroom, but 209E will eventually fade-away

Cancellation of FED-STD-209E

The U.S. General Services Administration (GSA) released a Notice of Cancellation for FED-STD-209E, Airborne Particulate Cleanliness Classes in Cleanrooms and Clean Zones, on November 29, 2001. (Please note that the IEST address changed after the issuance of the notice. The new address is 5005 Newport Drive, Suite 506, Rolling Meadows, IL 60008-3841.)

The Notice of Cancellation is based on the recommendation by IEST Working Group CC100 that FED-STD-209E no longer be maintained. The IEST, assigned by the GSA as the Preparing Activity organization for FED-STD-209E, has recommended that International Standard ISO 14644, Cleanrooms and controlled environments- Part 1: Classification of air cleanliness, and Part 2: Specifications for testing and monitoring to prove continued compliance with ISO 14644-1, supersede FED-STD-209E.

ISO 14644-1 and 14644-2 are two parts of a multi-part group of ISO Standards developed by ISO Technical Committee 209 (ISO/TC 209). All ISO/TC 209 Standards may be ordered online through IEST. Click here to enter the IEST Publications Store.

NOTICE OF CANCELLATION FED-STD-209 NOTICE 1
November 29, 2001

FEDERAL STANDARD

AIRBORNE PARTICULATE CLEANLINESS CLASSES
IN CLEANROOMS AND CLEAN ZONES

Federal Standard 209E dated September 11, 1992 is hereby canceled and superseded by International Organization for Standardization (ISO) Standards. International Standards for Cleanrooms and associated controlled environments, ISO 14644-1 Part 1: Classification of air cleanliness; and ISO 14644-2 Part 2: Specifications for testing and monitoring to prove continued compliance with ISO 14644-1.
Application for copies of ISO Standards 14644-1 Part 1, and 14644-2 Part 2; may be addressed to the Institute of Environmental Sciences and Technology (IEST), 940 East Northwest Highway, Mount Prospect, IL 60056-3444. Phone: 847-255-1561, Fax: 847-255-1699, Web site: www.iest.org, E-mail: publicationsales@iest.org.

Preparing Activity: GSA-FSS
FSC 3694

Clean Rooms and Controlled Environments

- ISO Technical Committee 209 was formed (no relation to FS 209 which was to be superceded)
- Each country sent their delegates to the committee meetings
- First meeting in 1993
- Scope: To develop a set of international standards which standardize equipment, facilities and operational methods for cleanrooms and associated controlled environments

Clean Rooms and Controlled Environments

- Scope included defining operational limits, procedural limits and testing attributes to minimize micro-contamination.
- Over 36 countries were involved
- US Team consisted of representatives from industry, consultants, cleanroom manufacturers, IEST, FDA
- IEST: Institute of Environmental Sciences and Testing

Clean Rooms and Controlled Environments

Eight ISO standards originally conceived

- 14644-1 "Classification of Air Cleanliness" *Approved*
- 14644-2 "Specifications for Testing and Monitoring to Ensure Continued Compliance with ISO 14644-1" *Approved*
- 14644-3 "Metrology and Test Methods"
- 14644-4 "Design, Construction and Start-up"
- 14644-5 "Operation"
- 14644-6 "Terms and Definitions" (dictionary) 14644-7 "Separative enclosures (clean air hoods, gloveboxes, isolators, mini environments)"
- 14644-8 "Molecular Contamination"

Clean Rooms and Controlled Environments

ISO Standards: 14644-1 and 14644-2 most important of all standards

Part 1: Classification of air cleanliness

Gives the airborne particle limits for different standards of cleanrooms. It also gives the methods that should be used to measure the airborne particles when testing a cleanroom to determine its class.

Part 2: Specifications for testing and monitoring to prove continued compliance with ISO 14644-1

Gives information, including time intervals, for testing a cleanroom to show that it still complies with the ISO 14644-1 standard.

Clean Rooms and Controlled Environments

ISO Standards:

Part 3: Metrology and test methods

Gives a description of the test methods that should be used to test the cleanroom to show that it is working correctly.

Part 4: Design, construction, and startup

Gives general guidance as to how a cleanroom should be designed, constructed and made ready for handing over to the user.

Part 5: Operation

Gives general advice on how to run a cleanroom.

Part 6: Terms and definitions

A collection of all the definitions of terms used in the ISO cleanroom standards.

Part 7: Separative enclosures (clean air hoods, gloveboxes, isolator, mini environments)

Gives information on clean air devices such as isolators and mini-environments

Part 8: Molecular contamination

Gives information on gaseous contamination in cleanrooms.

Clean Rooms and Controlled Environments

- **Must classify each clean room in terms of:**
  - ISO class
  - Occupancy state
  - Particle Size

e.g.: ISO Class 5 “as-built” at 0.2 and 0.5 micrometers

Clean Rooms and Controlled Environments

ISO 14644-1 gives a method to classify cleanrooms. The classification is based on the following equation:

$$C_n = 10^N (0.1/D)^{2.08}$$

- Cn is the maximum permitted concentration (in particles/m³ of air) of airborne particles that are equal to, or larger, than the considered particle size.
- Cn is rounded to the nearest whole number, using no more than three significant figures.
- N is the ISO classification number, which shall not exceed the value of 9. Intermediate ISO classification numbers may be specified, with 0.1 the smallest permitted increment of N.
- D is the considered particle size in mm.
- 0.1 is a constant with a dimension of mm.

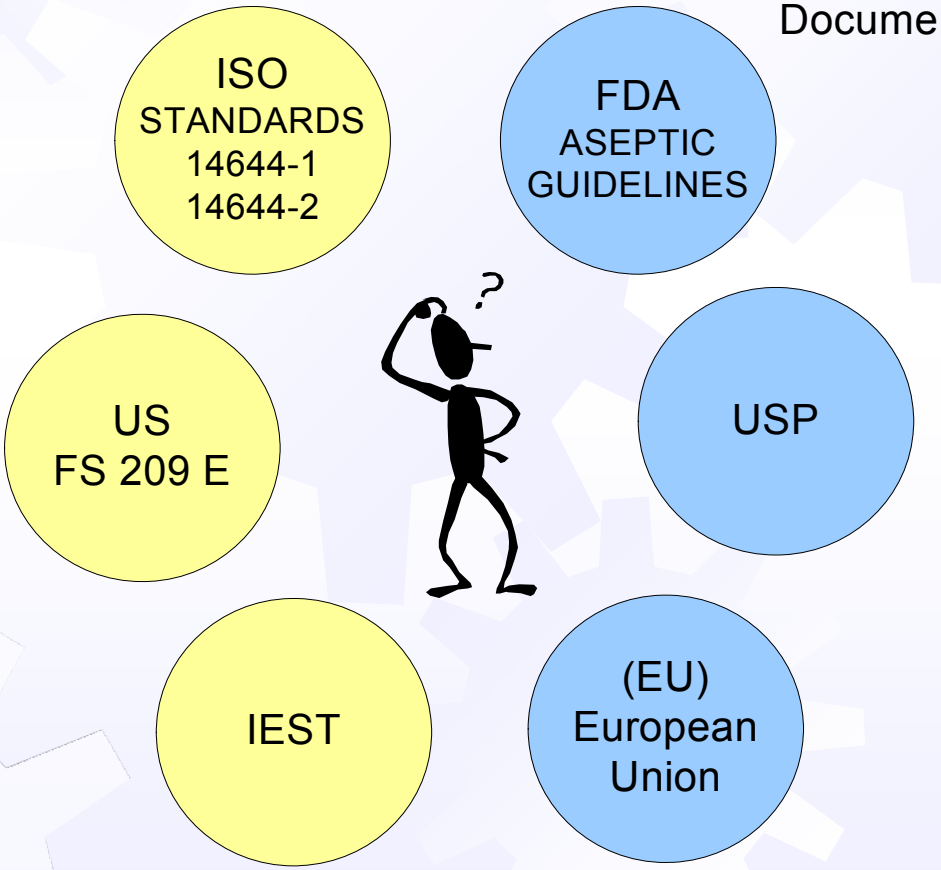
Clean Rooms and Controlled Environments – Cleanroom Standards

- FS 209E - US Federal Standard 209E
- ISO - International Standards Organization – the "NEW" standards –very important
- IEST- "Institute of Environmental Sciences and Testing"
- FDA Aseptic Guidelines
- EU - European Union GMP's
- USP - United States Pharmacopeia

Clean Rooms and Controlled Environments – Cleanroom Standards

General Standards

Pharmaceutical Regulatory/Guidance Documents



Clean Rooms and Controlled Environments

Major issues addressed in the next few weeks:

Understand differences between FDA Aseptic Guidelines and EU Requirements

Understand differences between FS 209E and ISO 14644-1

Perform basic particle distribution calculations to verify class

Be able to relate manufacturing process to the Class of Clean Room, both for FDA and EU requirements.

Clean Rooms and Controlled Environments

- Unit of measure for particle size:
- Metric: Micron….same as micrometer, 1 micron=10^{-6} meters.
- Denoted as: Micron: μm
- 1inch=25,400 microns
- 1/16 of an inch=1,587 microns

Clean Rooms and Controlled Environments
FDA Aseptic Guidelines

Important Notes:

- **FDA Aseptic guidelines do not allow averaging at a sampling site!**
- **Each discrete sample must be below the class limit.**
- **This is important, since you can pass ISO and FS 209E, and not meet FDA requirement.**
- **FDA aseptic guidelines still reference the FS 209E classes, as well as the ISO classes. They are allowing manufacturers too use either system for cleanroom certification.**
- **The Pharmaceutical Industry does not typically use Class 1 or Class 10 rooms. These designations are commonly used in the semiconductor industry.**

Clean Rooms and Controlled Environments

- Federal Standard 209E Cleanroom Classes – with metric designations

SI Class	Class (English)	# Particles/ft^3 ≥ 0.5 m
M 1.5	1	1
M 2.5	10	10
M 3.5	100	100
M 4.5	1,000	1,000
M 5.5	10,000	10,000
M 6.5	100,000	100,000

FS 209E and other particle size limits

AIRBORNE PARTICULATE CLEANLINESS CLASSES

Class limits are given for each class name. The limits designate specific concentrations (particles per unit volume) of airborne particles with sizes equal to and larger than the particle sizes shown*

Class Name**		Class limits									
		0.1 µm		0.2 µm		0.3 µm		0.5 µm		5 µm	
		Volume units		Volume units		Volume units		Volume units		Volume units	
SI	English***	(m^3)	(ft^3)	(m^3)	(ft^3)	(m^3)	(ft^3)	(m^3)	(ft^3)	(m^3)	(ft^3)
M 1		350	9.91	75.7	2.14	30.9	0.875	10.0	0.283	–	–
M 1.5	1	1 240	35.0	265	7.50	106	3.00	35.3	1.00	–	–
M 2		3 500	99.1	757	21.4	309	8.75	100	2.83	–	–
M 2.5	10	12 400	350	2 650	75.0	1 060	30.0	353	10.0	–	–
M 3		35 000	991	7 570	214	3 090	87.5	1 000	28.3	–	–
M 3.5	100	–	–	26 500	750	10 600	300	3 530	100	–	–
M 4		–	–	75 700	2140	30 900	875	10 000	283	–	–
M 4.5	1000	–	–	–	–	–	–	35 300	1000	247	7.00
M 5		–	–	–	–	–	–	100 000	2 830	618	17.5
M 5.5	10 000	–	–	–	–	–	–	353 000	10 000	2 470	70.0
M 6		–	–	–	–	–	–	1 000 000	28 300	6 180	175
M 6.5	100 000	–	–	–	–	–	–	3 530 000	100 000	24 700	700

Clean Rooms and Controlled Environments

ISO Classes- Particles per Cubic Meter

CLASS	PARTICLE SIZE					
	0.1µm	0.2µm	0.3µm	0.5µm	1µm	5µm
ISO 1	10	2				
ISO 2	100	24	10	4		
ISO 3	1,000	237	102	35	8	
ISO 4	10,000	2,370	1,020	352	83	
ISO 5	100,000	23,700	10,200	3,520	832	29
ISO 6	1,000,000	237,000	102,000	35,200	8,320	293
ISO 7				352,000	83,200	2,930
ISO 8				3,520,000	832,000	29,300
ISO 9				35,200,000	8,320,000	293,000

Clean Rooms and Controlled Environments

- **Pharmaceutical:**
  - A controlled* area where personnel are required to be in a minimal amount of gowning. (e.g.: Packing hall). Typical gowning consists of coat, hat and shoe covers

*controlled term is used generically. It is different from the controlled area referenced in the 1987 Aseptic guidelines.

- **Pharmaceutical with local monitoring:**
  - A pharmaceutical area that has at least some portions designed as Class 100,000 at rest.

Clean Rooms and Controlled Environments

- **<u>Pharmaceutical also called</u> “Controlled Not Classified”**
- Often designed as EU Grade D

Clean Rooms and Controlled Environments

Unclassified area:

- self explanatory….typical of general access areas, mechanical rooms,…etc.

Clean Rooms and Controlled Environments

US Pharmacopeia (USP) Considerations:

Classification of clean rooms and other controlled environments is based on Federal Standard 209E.

Pharmaceutical and Medical Device manufacturers have adopted the classifications:

Class 100, Class 10,000 and Class 100,000

- These classes are now represented by M3.5, M5.5, M6.5.

Clean Rooms and Controlled Environments

- USP Considerations
- Pharmaceutical industry deals with class M3.5 and above.
- Higher cleanliness levels are typically used in the electronics industry.
- Classes are based on “in-operation” or dynamic mode.

Clean Rooms and Controlled Environments

USP Considerations

- "Although there is no direct relationship established between the 209E controlled environment, it is generally accepted by scientists that airborne microorganisms in controlled environments can influence the microbiological quality of the intermediate or final products manufactured….."

Clean Rooms and Controlled Environments

USP Considerations

- “Monitoring of total particulate count in controlled environments, even with the use of electronic instrumentation on a continuous basis, does not provide information on the microbiological content of the environment…... While airborne microorganisms are not free-floating or single cells, they frequently associate with particles of 10 to 20 µm. Particulate counts as well as microbial counts within controlled environments vary with the sampling location and the activities being conducted during sampling”.

Clean Rooms and Controlled Environments

- For the Pharmaceutical Industry in the US, the primary document to follow is the *FDA Aseptic Guidelines*
- For the European community, the EU GMP's provide requirements for cleanrooms…we'll discuss this in more detail later……
- Both the FDA and EU GMP's requirements are consistent with the classes represented in FS 209E and ISO standards
- For the US, the USP also contains requirements for controlled environments/clean rooms
- Latest USP publication: #31

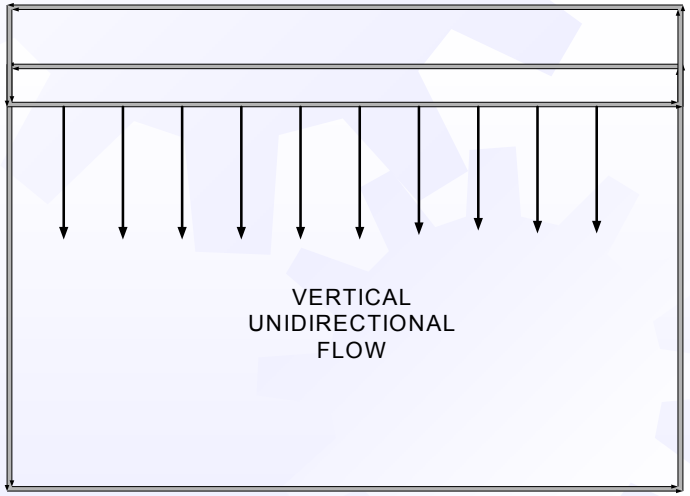
Controlled Environments

How do we reduce the level of contaminants?

- Airborne particles are HEPA filtered
- Contact parts are cleaned and sterilized
- Use of steam sterilization or irradiation of components
- Water purification systems are installed
- Limit aseptic core interventions
- Sterile filter the bulk solution (product)
- Wear clean room garments - limit shedding..follow proper aseptic techniques….very important.

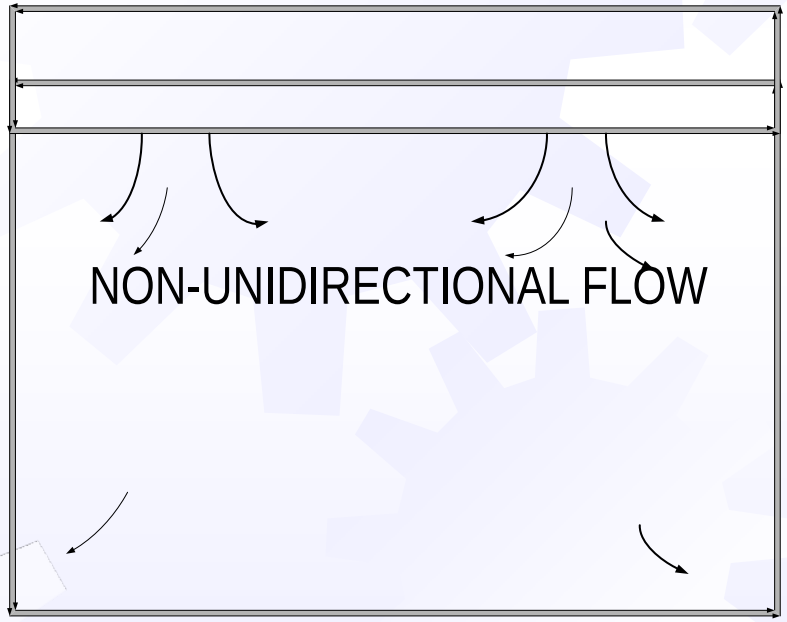
Clean Rooms and Controlled Environments

- Unidirectional Flow: Also called *Laminar Flow* – airflow having generally parallel streamlines, operating in a single direction, and with uniform velocity over its cross section. For example:



Clean Rooms and Controlled Environments

- Non-unidirectional Flow: Also called “mixed-flow” or “turbulent flow” – airflow which is not unidirectional. For example:



Clean Rooms and Controlled Environments

ISO vs FS209E: key differences

- Three new classes were introduced; ISO Class 1 and Class 2, both of which are cleaner than FS 209E Class 1, & ISO Class 9.
- For the most part, ISO Class 3 through 8 are very similar to FS 209E Class 1 through 100,000.
- ISO added the 1.0 micron particle size
- ISO generally requires fewer sampling locations than FS 209E
- With ISO, number of sample locations is based on clean room area, whereas FS 209E it is based on Class, size of clean room, and whether or not unidirectional flow is present
- ISO has a minimum 1 minute sample time, FS 209E does not.

Clean Rooms and Controlled Environments

Common items: ISO Standard 14644-1 & 2 and FS 209E

- **No fewer than two sample locations**
- **If less than 10 samples are taken, *then use statistical methods*.**
- **Minimum sample volume and time is also dictated in each standard**
- **Averaging particle readings at a site is allowed. Two rules:**
  - **Acceptable as longs as average at each site is below the class limit.**
  - **Average of all sites should not exceed class limit, adjusted to 95% upper confidence interval…normal distribution assumed.**

Clean Rooms and Controlled Environments

FS 209E & ISO Standard 14644-1 & 2

- **Note that the number of sampling locations is the minimum required.**
- **It's often easier to sample a room at 10 or more locations, rather than going through the statistical analysis**

Clean Rooms and Controlled Environments

Common items - ISO Standard 14644-1 & 2 and FS 209E

- **If less than 10 samples are taken, *then use of statistical methods is required*.**
- **Averaging particle readings at a site is allowed.**

  Two rules:

  - **Acceptable as longs as average at each site is below the class limit.**
  - **The 95% UCL (Upper Confidence Level) of the averages of all sites should not exceed the class limit.**

Clean Rooms and Controlled Environments

FS 209E & ISO Standard 14644-1 & 2

- **Sampling height is within 1 foot of equipment work surface area**
- **If no equipment present, typically 40” aff. is used**

  (aff.= above finished floor)
- **Note that the number of sampling locations is the minimum required.**
- **It’s often easier to sample a room at 10 or more locations, rather than going through the statistical analysis**

Clean Rooms and Controlled Environments

- European Union Guide to Good Manufacturing Practices (EU cGMP)
- Formal title: The Rules Governing Medicinal Products in the European Union. Volume 4. Good Manufacturing Practices – Medicinal Products for Human and Veterinary Use"
- Establishes four grades: Grade A, B, C, D
- Each grade has limits for viable and non-viable particulates