Volatile Chemicals and Class II Type A2 “Recirculated” BSCs: How Much is Safe?

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Presentation agenda

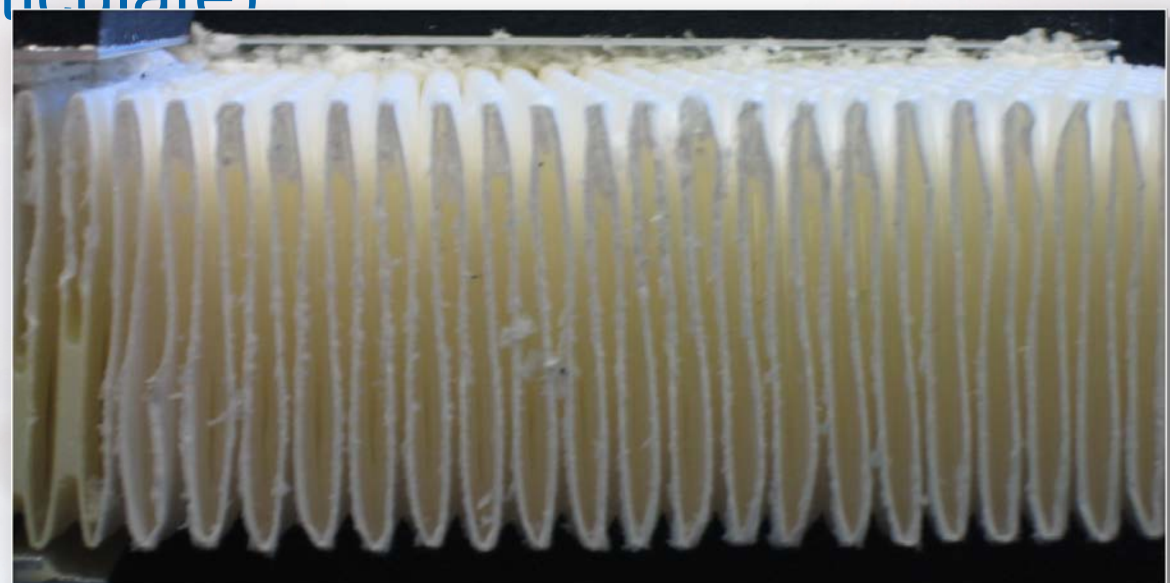
- Types of BSCs and HEPA filters
- Volatile Chemical use in Biology
- Calculations of Airflow and Chemical Concentrations
- How much can I use?
- Recirculated A2 vs. 100% exhausted B2
- NSF safety standards – “minute” amounts

Biosafety Cabinets (BSCs)

- A ventilated enclosure for work with biohazard agents assigned to biosafety levels 1 through 4, as per the BMBL.
- Provides levels of **CONTAINMENT**
  - Personnel protection
  - Product protection
  - Environmental protection

HEPA filtration

- **H**igh **E**fficiency **P**articulate **A**ir
  - Filter out 99.97% (or better) of 0.3 µm mass median particles of DOP (Dispersed Oil Particulate)



  - **CANNOT filter out gasses and vapors!**

Biosafety Cabinet Classification

Class I
- Personnel and Environmental Protection

Class II
- Personnel, Product, and Environmental Protection

Type A1
- Recirculated
- Minimum 75 fpm intake velocity

Type A2
- Recirculated
- Minimum 100 fpm intake

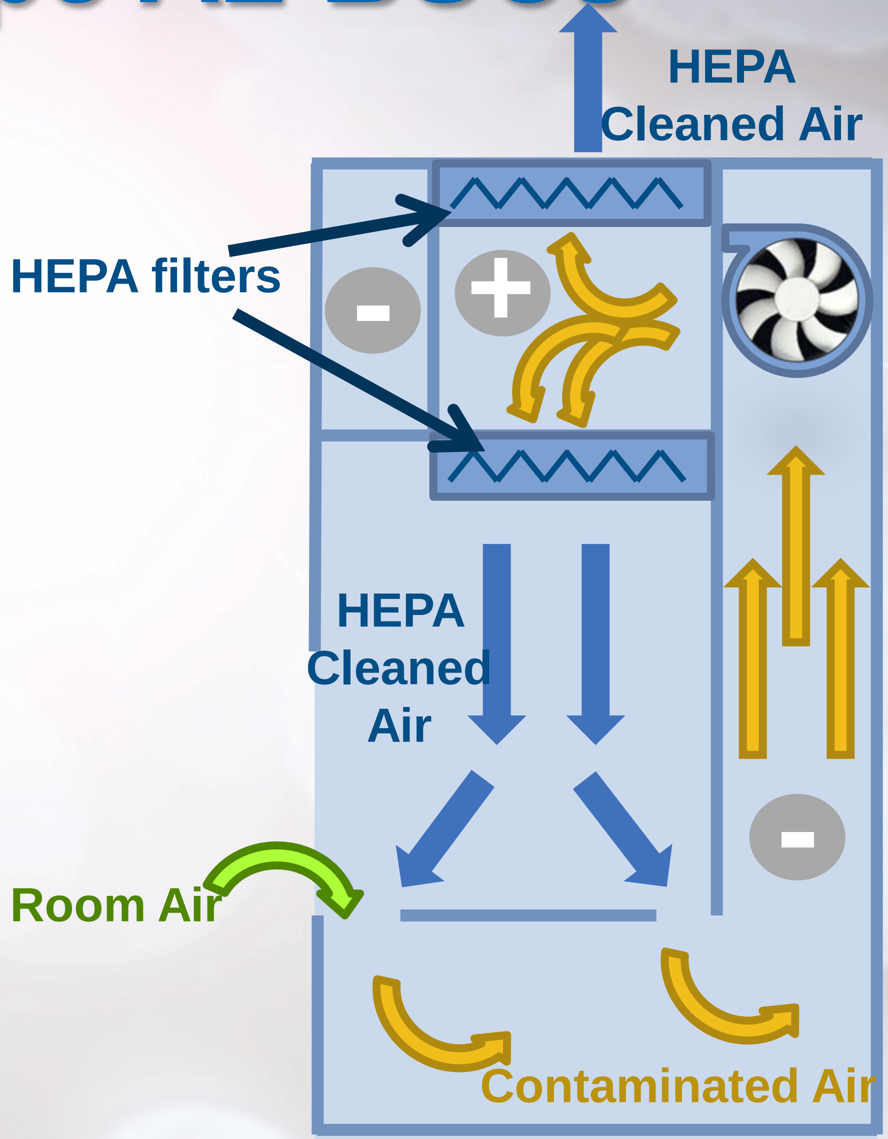
Type B1
- Partial recirculation

Type B2
- 100% Exhaust

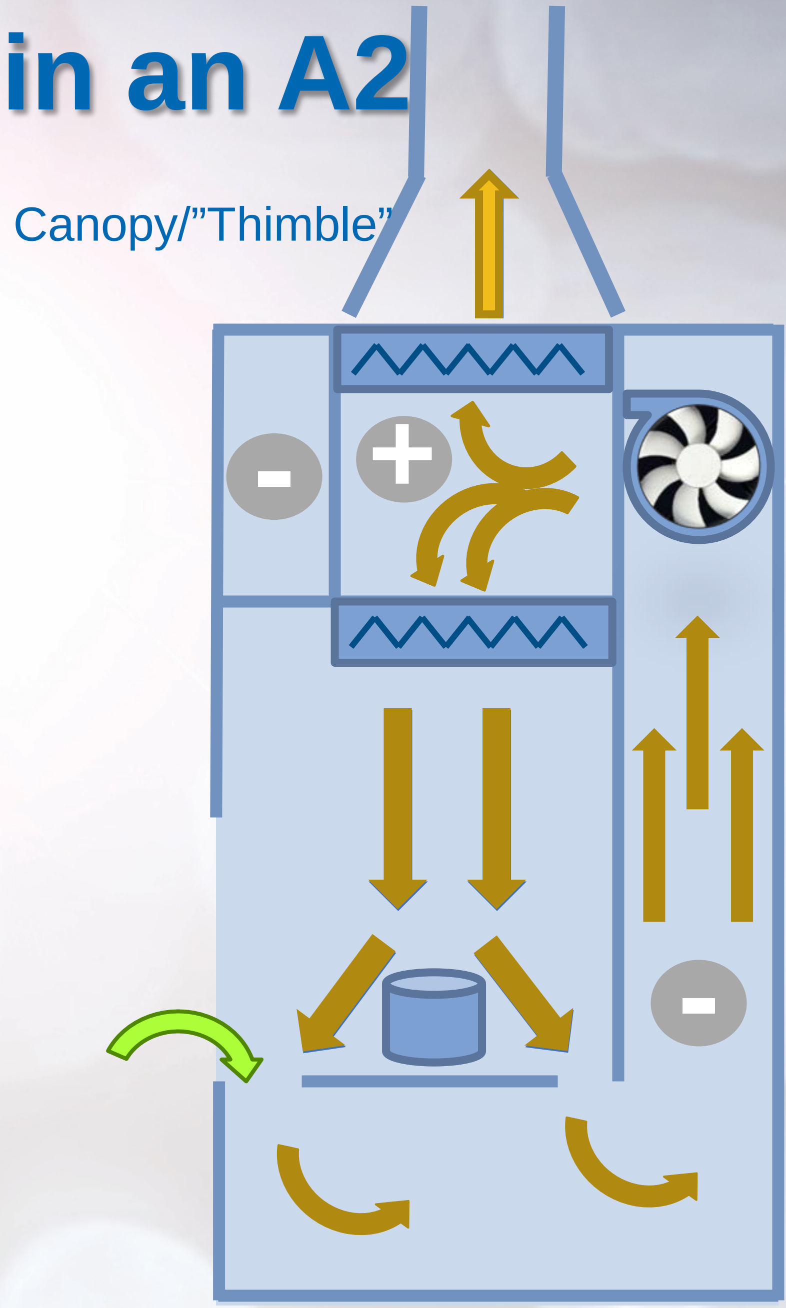
Class III
- For highly infectious agents
- Gas-tight, physical barrier

Class II Type A2 BSCs

- Personnel, Product and, Environmental Protection
- Minimum 100 fpm intake air
- Partial recirculation



Volatile Chemicals in an A2



- If a volatile chemical is introduced in the work area, all air becomes contaminated.
- How much is safe?
  - Vent to room vs. Canopy
  - Explosion limits for BSC
  - Chemical exposure limits
  - "Minute" amounts per NSF Standard 49

Why can't we just assign a value for a maximum amount of chemical allowed?

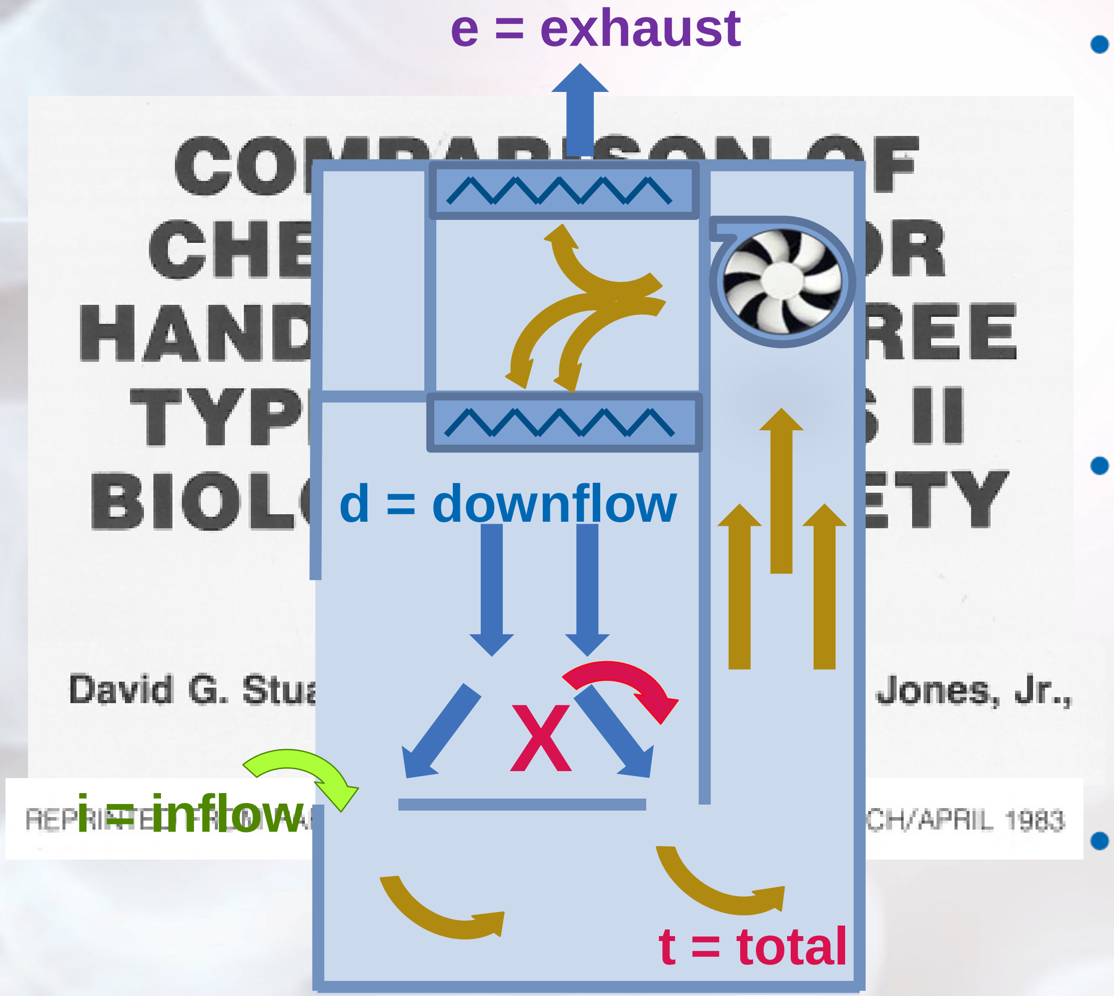
- NSF wanted to put a 250 mL limit on volatile chemicals for A2s.
- Not all chemicals are created equal!
  - Different harmful levels
  - Different evaporation rates
- Depends on the surface area of the container used
  - Spill vs. using a capped bottle?
- What about new chemicals not yet evaluated?
- Depends on the amount of air being diluted into

Calculating Airflow

- Airflow is designated by “Q”
- Airflow is the product of velocity and area
  - Q=V*A
    - V = 100 fpm intake velocity,
    - A = front access opening
      (e.g. 8”sash, 4’ cabinet = 2.67 ft^2)
    - Q = 267 cfm

*Round numbers are used for simplicity. Actual values may vary depending on manufacturer.

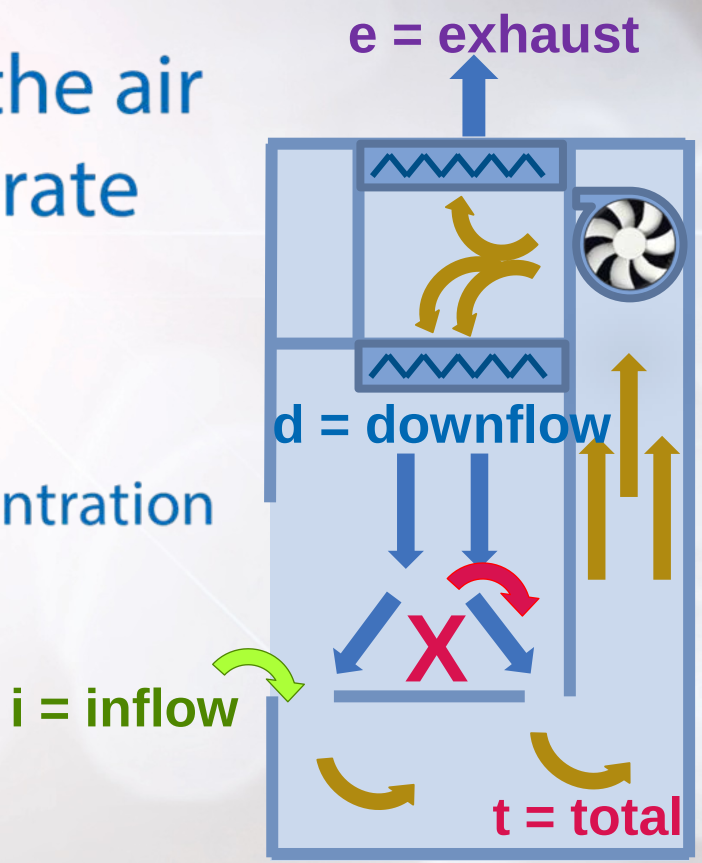
Mathematically Derived and Empirically Proven Equations



- Class II Type A cabinets
  - $C_d = C_i + \frac{X}{Q_i}$
  - If C_i in lab = 0, then $C_d = \frac{X}{Q_i}$
- Example:
  - $C_d = \frac{1558\ mg/min}{7.1\ m^3/min}$
  - $C_d = \mathbf{219\ mg/m^3}$
- Confirmed with toluene empirically

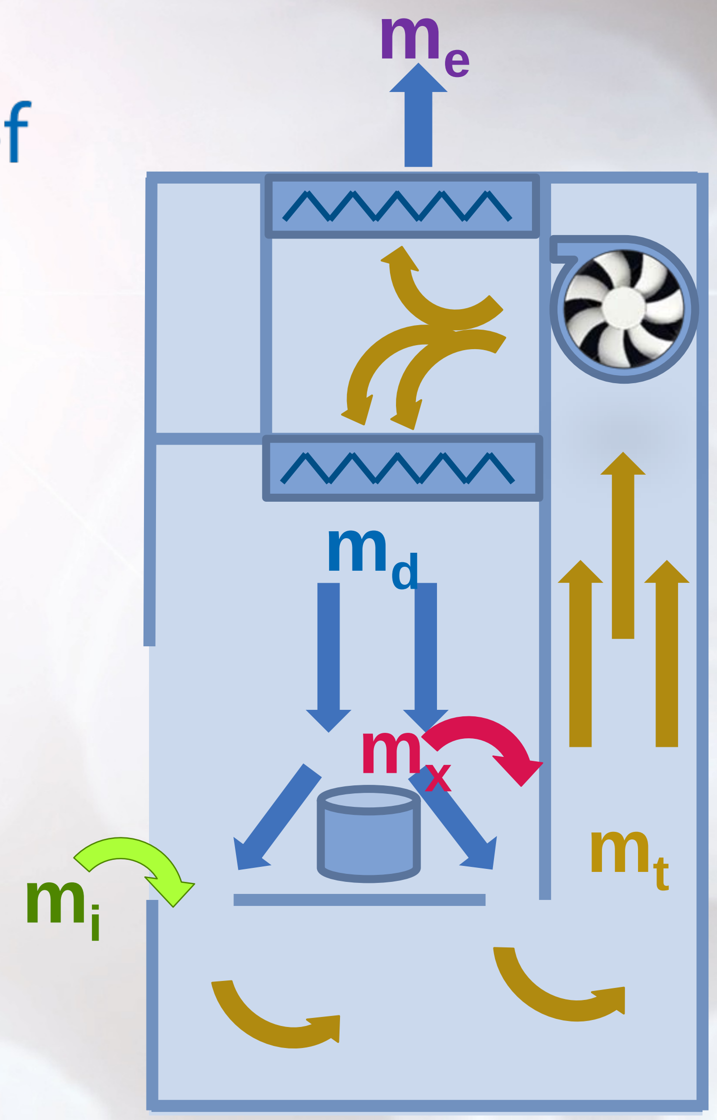
Calculating airflows and volumes

- The amount of chemical in the air is determined by mass flow rate ($\dot{m}$)
  - $\dot{m} = Q*C$
    - Mass flow rate = airflow*concentration
    - Example: $\dot{m}_i = Q_i C_i$



Mass Flow Rates

- Solved as an infinite series of recirculations:
  - $\dot{m}_t = \dot{m}_d + \dot{m}_i + \dot{m}_x$
  - Where $\dot{m}_{d_n} = R\dot{m}_{t_n}$
  - Thus:
    - $\dot{m}_{d_\infty} = \left(\frac{R}{1-R}\right)(\dot{m}_i + \dot{m}_x)$



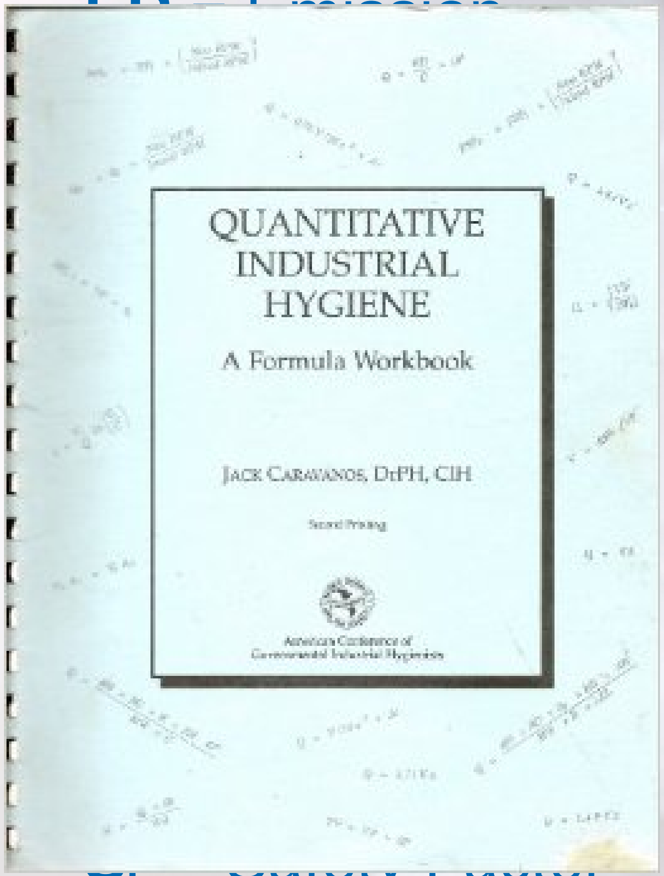
Mass Flow Rate Solution

- Using the empirical data from the toluene example:
- $\dot{m}_{d_\infty} = \left(\frac{R}{1-R}\right)(\dot{m}_i + \dot{m}_x)$
- Toluene was released at 1.8 mL/min =1558 mg/min; 70% recirculation
- $Q_d C_d = 2.33(Q_i C_i + \dot{m}_x)$
- $16.0\ m^3/min(C_d) = 2.33(0 + 1558\ mg/min)$
- $C_d = 226\ mg/m^3$

219 mg/m^3 (empirically confirmed) ≈ **226** mg/m^3 (calculated)

How MUCH can I use?

- Dilution Ventilation equation:
- $ER = \frac{Q_i * MW * LEL * 473}{403 * SG * S_F * 100}$
- S_F generally = 4 for 25% of LEL, more cautious users S_F = 10 (10% of LEL)



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How much can I use?

- Dilution Ventilation equation:

- $ER = \frac{Q_i * MW * LEL * 473}{403 * SG * S_F * 100}$

- *Example: isopropanol in 4ft A2 BSC*
- $ER = \frac{267cfm * 60g/mol * 2.0\% * 473}{403 * 0.786 * 4 * 100}$

- $ER = \mathbf{120\ mL/min}$

Calculating a spill

100 mL of 70% isopropanol spilled on the work surface of an 6’ A2 BSC.

- Covered 0.313 m^2 area
- Took 1 hour to evaporate

250 mL of 70% isopropanol covering the entire the work surface of a 6’ A2.

- Covers 0.79 m^2 area
- Will take 1 hour to evaporate

Empirical study done by The Baker Company

Am I safe?

As Calculated

120 mL/min 100% isopropanol

-or-

171 mL/min 70% isopropanol

As Spilled

250 mL/hr 70% isopropanol

-or-

4.2 mL/min 70% isopropanol

40-fold less than the safe limits!

How long does it take to clear?

- Reduce contaminated air to a nominal or background amount.
- One recirculation takes ~4 seconds*

- Helium injected to 73,000 ppm
- Measured to background = **60 to 90 seconds**

- $C_X * R^n = C_{X,low}$

- $73{,}000\ ppm * (0.7)^n = 10\ ppm$
- $n = 25\ recirculations$
- $25 * 4s = \mathbf{100\ seconds}$

*Round numbers are used for simplicity. Actual values may vary depending on manufacturer.
Empirical study done by The Baker Company

Is a 100% exhausted Type B2 cabinet really needed for volatile chemical use?

- B2 has greater chance of **failure**, because it is reliant on the building's exhaust system.
  - A2 can vent to room or through canopy connection
- B2 has a greater **operating** and **installing cost** relative to A2
- B2 is more **complex** to operate properly
  - Room pressurization, room size, alarms, filter loading lessening static pressure
- Do a risk assessment to determine which is right for your needs!

Determination of "Minute"

- The least amount of chemical needed for your experiment currently being conducted.
- Below explosion and exposure limits
- In appropriate containers, preferably capped.
- ALWAYS perform a Risk Assessment

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Questions?