Reducing Manufacturing Risk with Rapid Microbiological Methods (RMMs)

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azbil

BioVigilant®
Instantaneous Microbial Detection

Agenda

General overview of RMMs

Compare to agar plates

RMM example--IMD-A system

TR33

Examples of RMM reducing risk

Why adopt an RMM?

Faster, Better

and/or

Cheaper

(and accepted by regulators)

Sample FMEA – Agar Plates

Function	Potential Failure Mode	Potential Effects of Failure	Potential Causes of Failure	Current Process Control	Occurrence	Severity	Detection	RPN	Risk
enter gowning room, don gown	contaminated gowns	contamination allowed into cleanroom	gown supplier: inadequate cleaning or sterilization of gowns	Supplier audited routinely for conformance to best practice standards	2	5	4	40	High
	incorrect gowning technique	contamination allowed into cleanroom	procedure not validated	Quality system requires validation of cleanroom materials. Compliance to USP 1116.	1	5	4	20	Medium
			validation inadaquate	compliance to USP 1116	1	5	4	20	Medium
			operator error	training demonstrated to be effective	3	5	4	60	High

RMM Detection Taxonomy

Microbial Detection

Growth Dependent

Spectroscopy

Chemical Byproduct

BioLumix (BioLumix)
Bactometer (BioMerieux)
BacT Alert 3D (BioMerieux)
Neogen (Soleris)

Intrinsic Fluorescence

Growth Direct (Rapid Micro Biosystems)

Extrinsic Fluorescence

Advance (Celsis)
Milliflex Rapid (Millipore)
Milliflex Quantum (Millipore)
Pallchek (Pall)

Growth Independent

Spectroscopy

Extrinsic Fluorescence

FACS MicroCount (BD)
Biomaytector (Hitachi)
Endosafe PTS (Charles River)
ScanRDI (Chemunex)
D-Count/BactiFlow (Chemunex)

Intrinsic Fluorescence

IMD-A (ABV)
BioLaz (PMS)
BioTrak (TSI)

Intrinsic Raman

REBS (Battelle)
BPE (rap.ID)

Mie Scatter

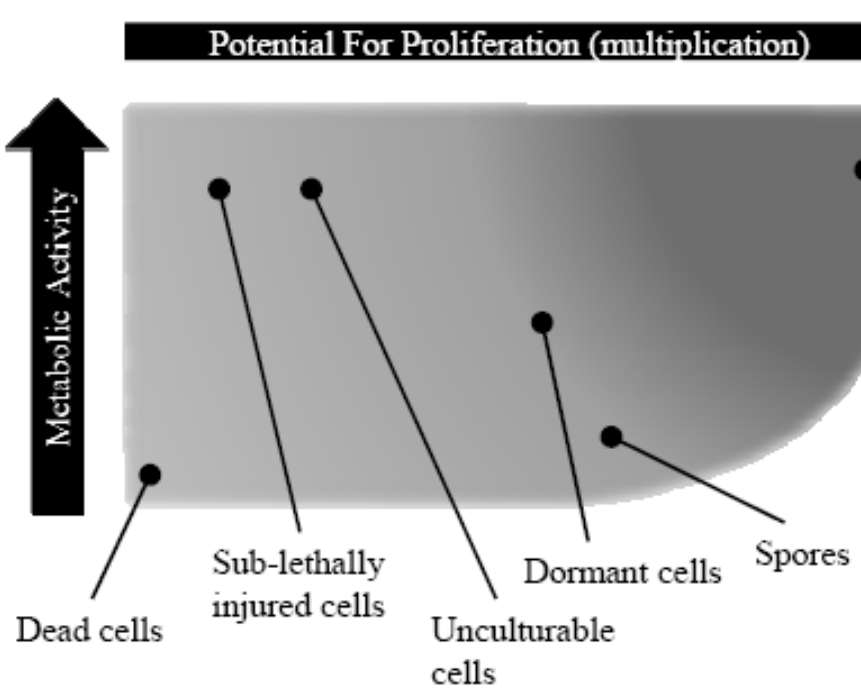
BioSentry (JMAR)

Nucleic Acid (PCR)

MicroSEQ (AB)
HSS (Biotecon)
ceeramTools (ceeram)
BAX (Dupont Qualicon)
MicroCompass II (Lonza)
GeneDisc (Pall)

The Microbial Environment

Microorganisms Defined By Physiology & Culturability



"Sterile and Non-Sterile Product Risks From Viable But Non-Culturable Bacteria"

Edward C. Tidswell PhD
Senior Director
Baxter Healthcare Corporation

October 2009, PDA 5th Annual Global Conference on Pharmaceutical Microbiology, Washington, DC

Typical Human Upper Respiratory Flora

BACTERIUM	Nose	Pharynx	Mouth
Staphylococcus epidermidis	++	++	++
*Staphylococcus aureus**	+	+	+
Streptococcus mitis		+	++
Streptococcussalivarius		++	++
*Streptococcus mutans**		+	++
*Streptococcus pneumoniae**	+/-	+	+
Streptococcus pyogenes		+	+
Neisseria sp.	+	++	+
Neisseria meningitidis	+	++	+
Proteus sp.	+	+	+
*Haemophilus influenzae**	+	+	+
Lactobacillus sp.		+	++
Corynebacteria	++	+	+
Actinomycetes		+	+
Spirochetes		+	++
Mycoplasmas		+	+

Green: Will grow on TSA/TSB

Red: Will NOT grow* on TSA/TSB

Orange: May Grow on TSA/TSB

Under typical incubation conditions.

"*Regulatory Expectations for Aseptically Produced Parenterals*"

Ian Symonds, Director Aseptic Quality Assurance , GlaxoSmithKline

December 2009 PDA Meeting, Milan

Microbial Culturability and Detectability

Most bacteria need other bacteria to grow[1]

- Need growth factors called siderophores that bind iron
- 1% of bacteria make growth factors for the other 99%

1. Kim Lewis (2010) Siderophores from neighboring organisms promote the growth of uncultured bacteria. Chemistry & Biology. 17(3):256-264

Microbial Culturability and Detectability

Open Question:

- Have agar plates worked so well because they grow up the microbes that are most dangerous (make growth factors)?

False negatives and non-biological positives

Microbial Detection

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Comparing RMMs

- False Negatives (Plates)
  - Can be viable microbes that don't grow under conditions used
  - Can be from inhibitors in testing material
  - Cannot use scientific methods to discriminate between real and false negatives
  - Less likely to raise questions with regulators
- "False" Positives (RMMs)
  - Can be instrument noise
  - Can be non-biological positives
  - Use scientific methods to discriminate between real and non-biological or false positives
  - Might raise questions with regulators

Episodic Sampling

- Air samplers do not measure most of the time
- Some RMMs can do continuous sampling to detect fluctuations in microbes over time

Sample FMEA – Agar Plates

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FMEA Example – Growth-Independent RMM

FMEA – Risk Assessment and Characterization

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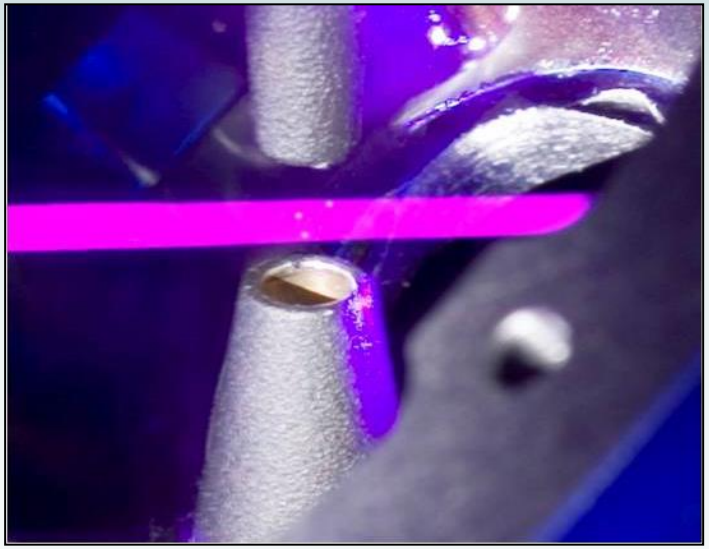
	incorrect gowning technique	contamination allowed into cleanroom	procedure not validated	Quality system requires validation of cleanroom materials. Compliance to USP 1116.	1	5	1	5	Low
			validation inadaquate	compliance to USP 1116	1	5	1	5	Low
			operator error	training demonstrated to be effective	3	5	1	15	Low

Example RMM Technology

How the IMD-A works

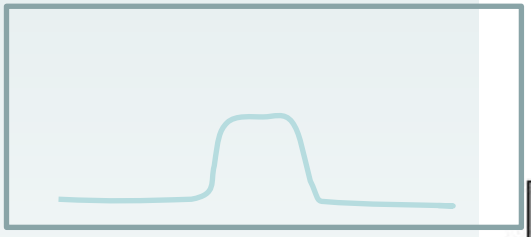
- Laser shines on microbes
- Detects intrinsic fluorescence
- Primary markers: NADH, Riboflavin, and DPA



Small Inert Particle

Fluorescence Detector

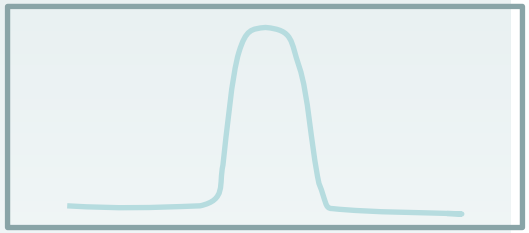
Scattering Detector



Large Inert Particle

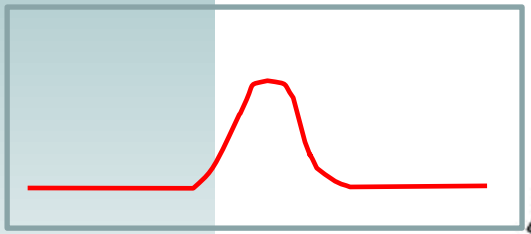
Fluorescence Detector

Scattering Detector

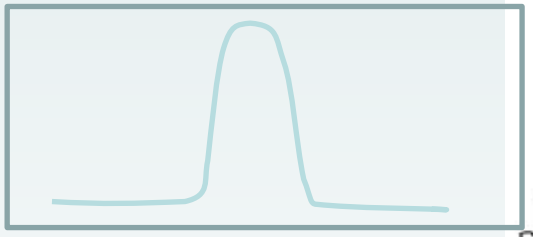


Large Biologic Particle

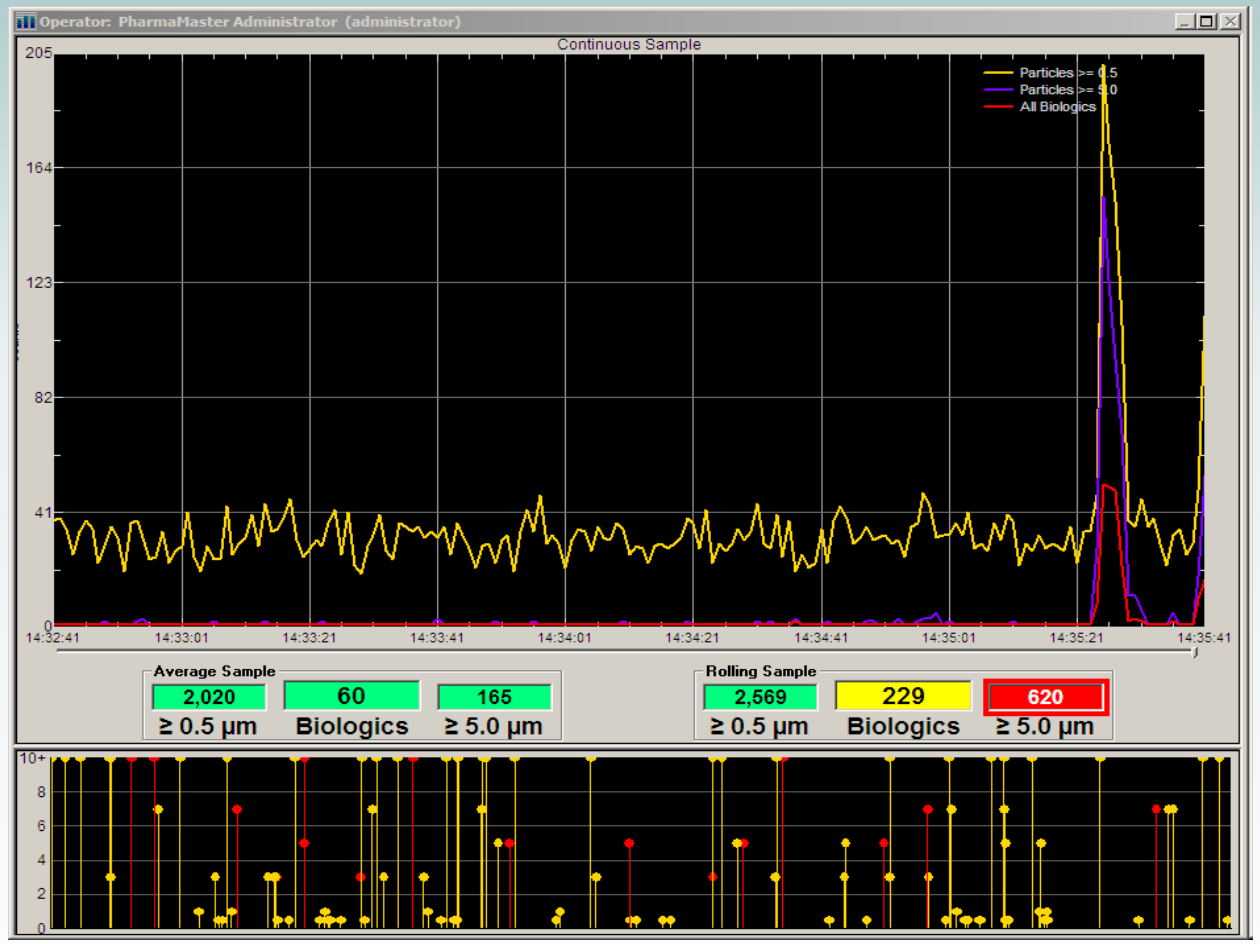
Fluorescence Detector



Scattering Detector



Real-Time Data Acquisition

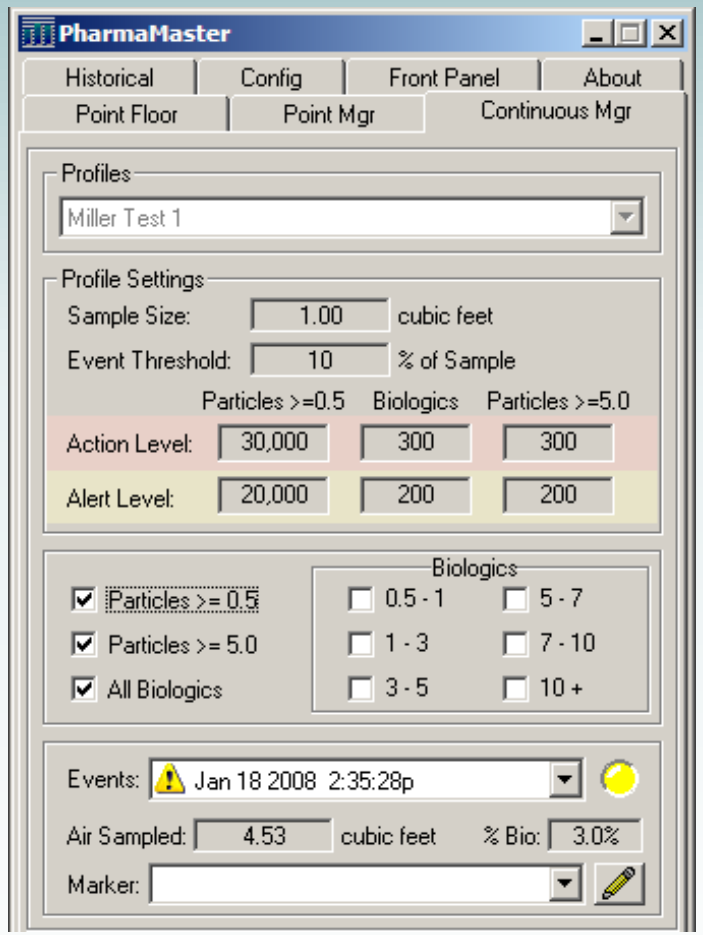


Data is acquired and displayed every 1 second (bottom screen). Yellow bars are non-viable particles and red bars are viable particles.

The top screen shows continuous monitoring data. This is refreshed every 1 second. Note the spike in counts at the far right.

The rolling sample is the particle count for the last sample size collected. The average sample is the average particle count per sample size over the entire monitoring run.

User Interface



Video is synced with monitoring data and can be used to support a root cause analysis of a contamination event

TR33

Technical Report No. 33 (Revised 2013)

Evaluation, Validation and Implementation of Alternative and Rapid Microbiological Methods

TR33

- Much longer (56 pages) than previous TR33, EP 5.1.6 or USP <1223>
- What needs to be done (not how to do it)
- Written for liquid suspensions, different approaches may be needed for air systems
- Not likely to be harmonized with EP 5.1.6 and USP <1223>

Reducing Risk with RMM

Real life examples

Reducing Risk with RMM

HEPA filter ajar

- RMM showed the air was clean, only the frame was askew

RMM showed high counts in class B room

- Indicated that air from HEPA filter was not aseptic

Reducing Risk with RMM

- Air samplers detected high CFU in some rooms of a class D aseptic suite
  - Did not indicate the source
- Long samples with RMM did not go down to zero at rest in rooms with high CFU
- RMM detected high counts in air coming in through light switches and electric plugs
- These rooms had negative pressure, should have been positive

Reducing Risk with RMM

- Air samplers detected mold in room
  - Do not show where the mold was
- RMM “sniffer” detected crack in plaster with high counts
- Behind the plaster was a large mold colony

Reducing Risk with RMM

Overnight sample showed peak around midnight

- Review of video showed presence of cleaning crew

Multi-day sample showed high counts in aseptic suite corresponding to construction activity next door

- Door between areas was not allowing construction dust in (working as it should)
- Sniffer function detected previously unknown conduits between the aseptic suite and construction area

Reducing Risk with RMM

Fill line restarted after cleaning and upgrades

- Fill line itself was well controlled, very low counts
- Operator loading vials generated many particles

Opportunities with an RMM

Faster,

Better

and/or

Cheaper…

And less risk

Opportunities with an RMM (assuming you want to improve)

Yes, it's old technology, but it's validated.

PharmaEvolution.com cartoon contest, June/July 2013

Improvement Opportunities (assuming you want to improve)

Contact Information

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