Consistent Product Robustness, the Science and the Technologies for **Proven Single-Use System Integrity (SUSI)**

Case Study: Helium Integrity Test

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Leak vs. Integrity Testing & Definitions

Leak test – a test used to identify leaks of certain sizes in a SUS.

Integrity Test – a test used to confirm the defined barrier properties of a SUS

Maximum Allowable Leakage Limit –the greatest leakage rate (or leak size) tolerable for a given product package that poses no risk to product safety and no or inconsequential impact on product quality.

Integrity Assurance – a holistic approach of risk analysis and mitigation by means of product and process robustness, quality and process control and integrity testing.

Non-destructive test method – a test method that maintains the tested SUS in a condition for further use, without impacting its quality attributes.

Destructive test method – a test method that may destroy the tested SUS during the test and not allow further use.

Single Use Process Integrity Is a Key Industry & Regulatory Challenge

Validated single-use system integrity required to:

- Strengthen regulatory compliance
- Improve patient safety

Single-use system integrity testing (SUSIT) is meaningful when it correlates to liquid leaks & microbial ingress

- Lack of understanding of defect size causing liquid leaks and microbial ingress under real life conditions
- Current testing is not correlated to leaks & microbial ingress

> Bag failures cost ~$100K to $1M per bag
>
> R. Wong, Bayer

> Leaks have been reportedly responsible for up to $20M worth of products per year at some larger facilities.
>
> Bioplan

Users cannot show real proof of process integrity to regulatory organizations

Single Use Process Integrity Is a Key Industry & Regulatory Challenge

SUS Used in Drug Substance & Drug Product Process Steps Require Higher Assurance of Integrity

> We're very much concerned about the integrity of SUS used in final filling applications.
>
> FDA Inspector 02|2018

Regulatory Bodies Are Pushing for More Testing

European Commission

Annex1 - Manufacture of Sterile Medicinal Products *(Revision – Not yet published)*

8.115 Appropriate systems should be in place to assure the integrity of those components used. The manner in which this is conducted should be determined based on QRM principles. Appropriate system integrity tests should be considered when there is a risk of compromising product sterility.

8.117 Single-use systems (SUS) are those technologies used in manufacture of sterile medical products [...].

8.118 There are some specific risks associated with SUS which include, but are not limited to:

a) [...]
g) Pinhole and leakage
h) The potential for compromising the system at the point of opening the outer packaging

FDA-ASTM Workshops – Oct 2016[(1)] & Apr 2018[(2)]

Validation for SU fill finish assemblies for sterile product manufacturing

- **Physical integrity test correlated to microbial ingress**
- Packaging integrity at the supplier, post shipping & post-installation

Microbiological challenge testing

- Identification of defect size that would allow ingress of bacteria **under process conditions**

SUS fill finish assemblies for sterile products must meet requirements to ensure flow-path sterility and integrity

- Supporting sterilization validation summary data and information on the gamma irradiation process
- **Integrity tests are also reviewed during the review of the BLA[(3)] and on inspection**

[(1)] SUS: A Microbiology Product Quality Perspective - Patricia F. Hughes, Ph.D.., Branch Chief (Acting), CDER/OPQ/OPF/DMA
[(2)] Sterile drug product, New developments - Patricia F. Hughes, Ph.D.., Branch Chief (Acting), CDER/OPQ/OPF/DMA
[(3)] Biologics License Application

Current Industry Guidance and Standards Initiatives

USP<1207> 2016 Package Integrity Evaluation – Sterile Products

ASTM E55 WK64337 Standard Practice for Integrity Assurance & Testing of Single-use Systems *(passed the E55 main-committee ballot in 12|2019)*

ASTM E55 WK64975 Test Method for Microbial Ingress Testing on Single-use Systems *(rejected during E55.04 sub-committee ballot in 12|2019)*

NEW PDA TR on Pharmaceutical Package Integrity (to replace existing TR27)

BPSA 2017 Design, Control, and Monitoring of SUS for Integrity Assurance

The Challenge Is to Ensure That No Product Leakage or Microbial Ingress Can Occur During the Process

- QbD & validation to reach consistent robustness
- Process control, quality control & integrity testing

» SU Product Robustness

- Understand liquid leakage & bacteria ingress mechanisms
- Correlate integrity testing detection limits to liquid leaks & microbial ingress under process conditions

» SU System Integrity Science

- Implement 100% physical supplier integrity testing
- Provide point of use testing

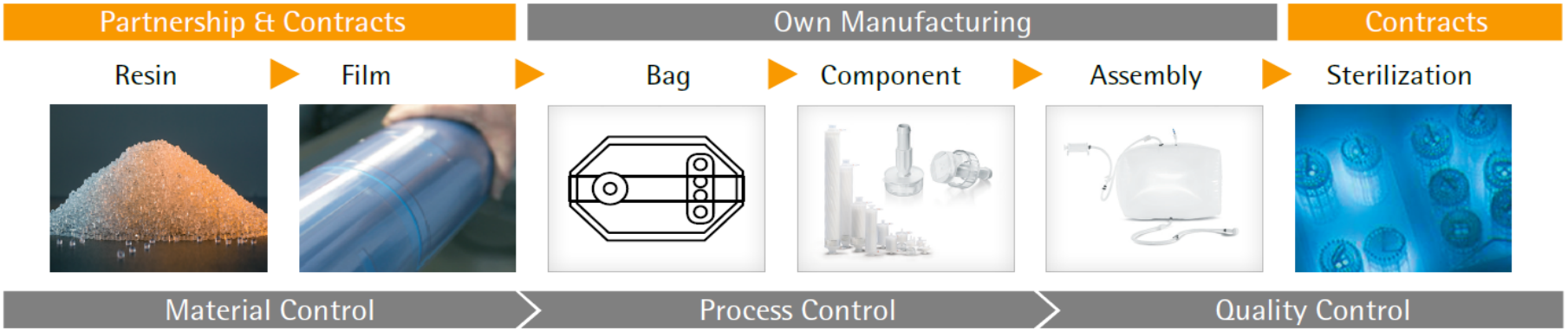
» Integrity Testing Technology

Combined with the robustness of our self-deploying bags, our integrity tests ensure that no product leakage or microbial ingress can occur during the process

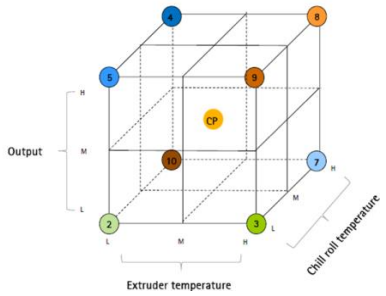
Process Control & Quality Control Ensure Integrity Along Our Entire Production Cycle

Process control reduces the risks of defects on film, seals, welds & connections
- Extrusion temperature & speed,
- Weld & seal temperature & time

Quality controls confirm the absence of leaks
- Film & seal strength integrity,
- Microbial Ingress Testing – ISO15747 - Immersion in B. Diminuta
- 100% Bag Chamber Leak Test (BC-LT)



Process Control & Quality Control Ensure Integrity Along Our Entire Production Cycle



~ 3000 robustness tests

Tensile Strength: 930

Elongation at break: 930

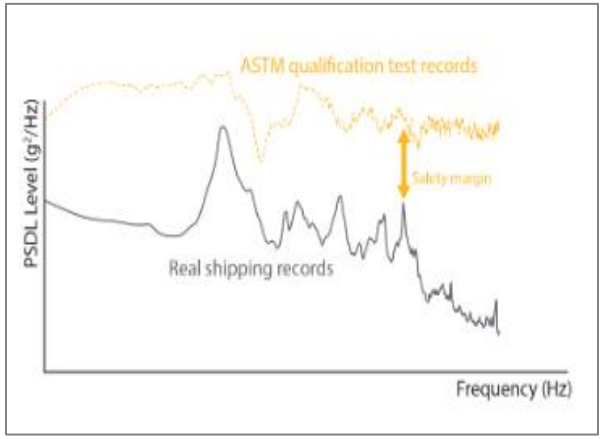
Puncture tests: 930

Flex durability: 186

Water burst tests : 26

Most stringent standards:

ASTM D4169
validation package with
real & lab. tests

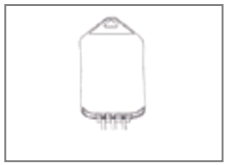


Passed ASTM Shipping Test

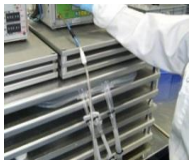
Self-deploying bag
during installation

Avoids mishandling
&
reduces bag failures

Process Control & Quality Control Ensure Integrity Along Our Entire Production Cycle



Bag chamber leak test 40 – 90 µm



Discard defects from manufacturing operations

Assembly Helium SIT 2 µm

Discard additional defects from manufacturing operations

But, why 2µm?

PoU-IT 10 µm FlexAct® BT

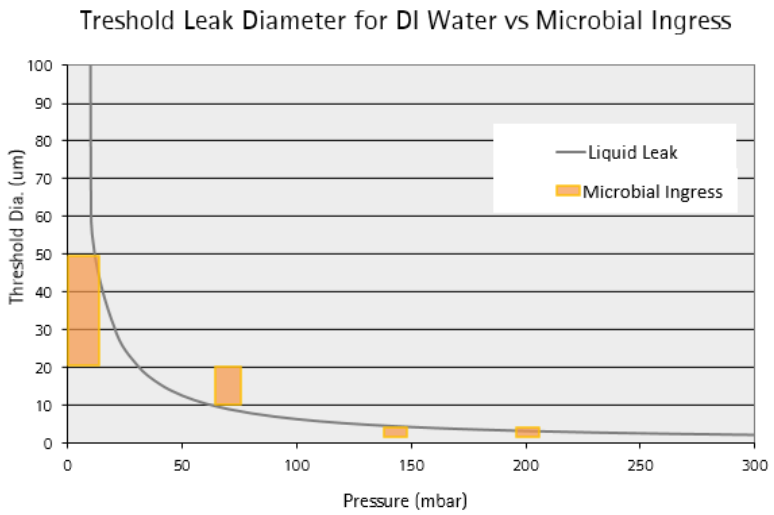
Discard additional defects from transportation & handling

Existing Data in Peer-reviewed Publications on Critical Leak Size for Sterile Package Using Micro Tubes

Gibney, 2000 $P_o > P_{atm} + \left[\left(\frac{4\sigma}{D_h} - \rho g L\right) \times 0.390\right]$

Liquid leak & bacterial ingress depend on
- Process & applications conditions
- Liquid properties
- Size of defect

Correlation between liquid leak & bacterial Ingress
- Microbial ingress cannot occur without liquid flow *(Kirsch & Morton, 1989)*



2µm is the maximum allowable leakage limit that never produce liquid leak or microbial ingress in any process conditions using micro tubes

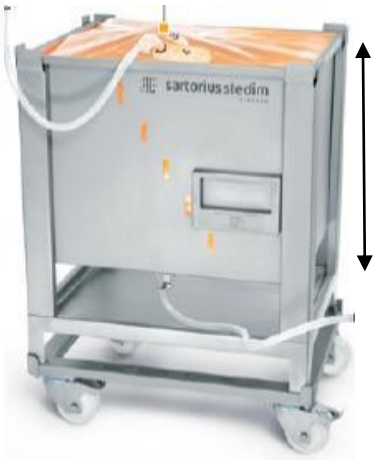
Understand Liquid Leakage and Microbial Ingress Mechanisms on Film Materials Used in Single-use Biomanufacturing

- Demonstrate the relation between microbial ingress and liquid leak
- Establish a predictive model to determine the Maximum Allowable Leakage Limit (MALL*) under any process conditions
- Establish a correlation between liquid leak/microbial ingress and physical integrity testing
- Develop and validate the physical test methods with detection limits that guarantee the absence of liquid leak/microbial ingress in SUS

* MALL: is the greatest leak size tolerable that poses no risk to product safety, USP<1207>

Process Conditions May Impact Liquid Leak & Microbial Ingress

Hydrostatic pressure in storage



~0.7m / 2'

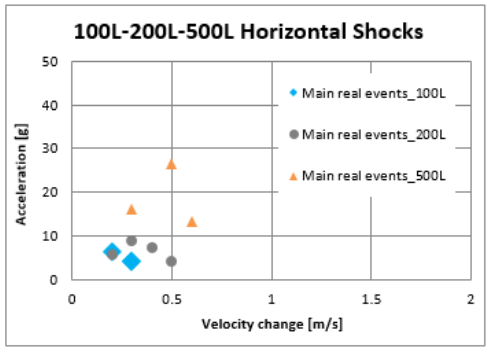
Max pressure 70mbar

Flexboy® 20 L

Flexsafe® 500 L

Application	Pressure range
Storage	~10 – 100 mbar
Shipping	~100 – 250 mbar

Shipping can generate up to 20g acceleration



Liquid Tests on Film Show No Leak on 2µm Defects at **300mbar** Pressure

Liquid leak results for PE film at 300mbar imposed pressure

	Hole size µm	2 µm	3 µm	5 µm	10 µm
Water	Number of samples	3	3	3	3
	Number of leaks	0	2	2	3
TSB*	Number of samples	3	3	3	N/A
	Number of leaks	0	1	1	N/A

* Trypticase soy broth (growth media for BCT trials)

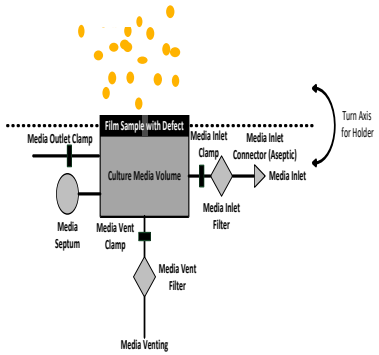
Multiple module setup:

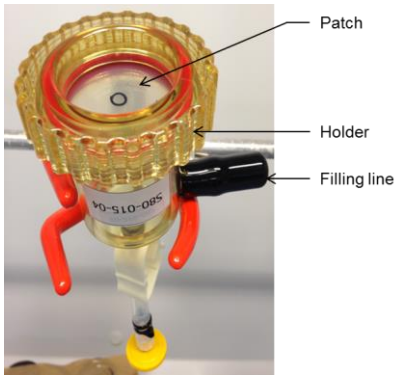
- Holders with different defect size patches
- 100ml model solution filled into each holder
- Holders connected to constant pressure source
- Indicator paper and stopwatch for leak observation

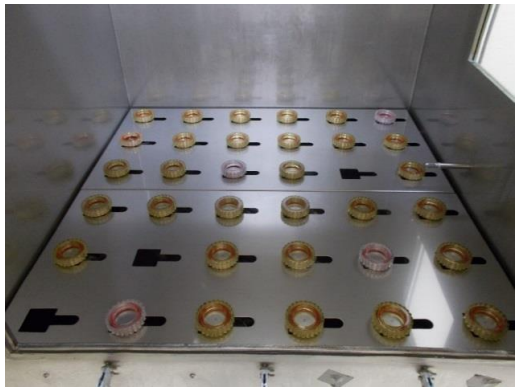
Microbial Aerosol Tests on 300 Film Samples Show No Ingress for 1µm Defects at **300mbar** Pressure

BCT results at 300mbar imposed pressure

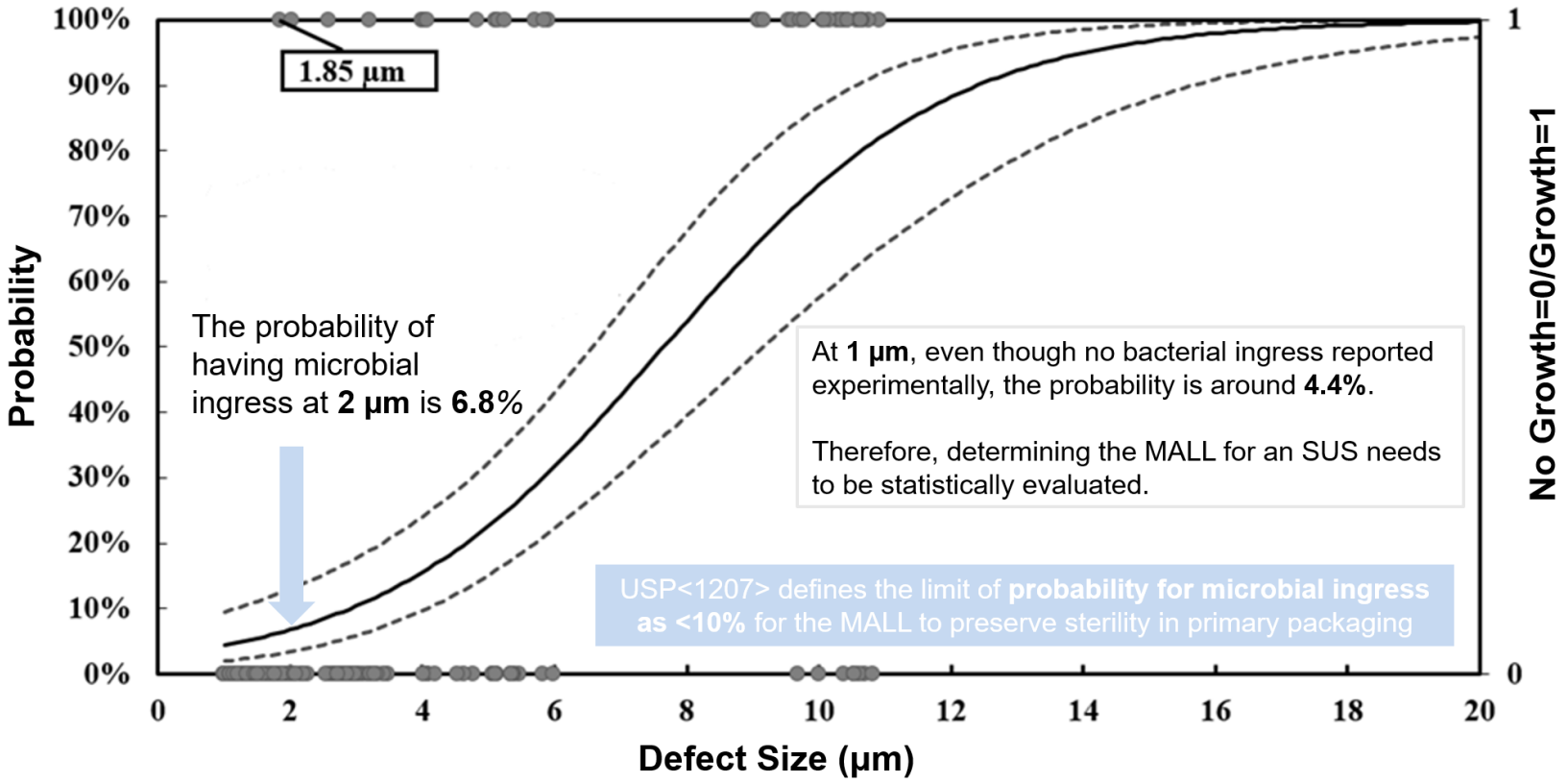
	Hole size µm	1µm	2µm	3µm	5µm	10µm
PE film	Number of samples	30	30	30	30	30
	Number of ingress	0	2	2	10	22
EVA film	Number of samples	30	30	30	30	30
	Number of ingress	0	0	9	17	13





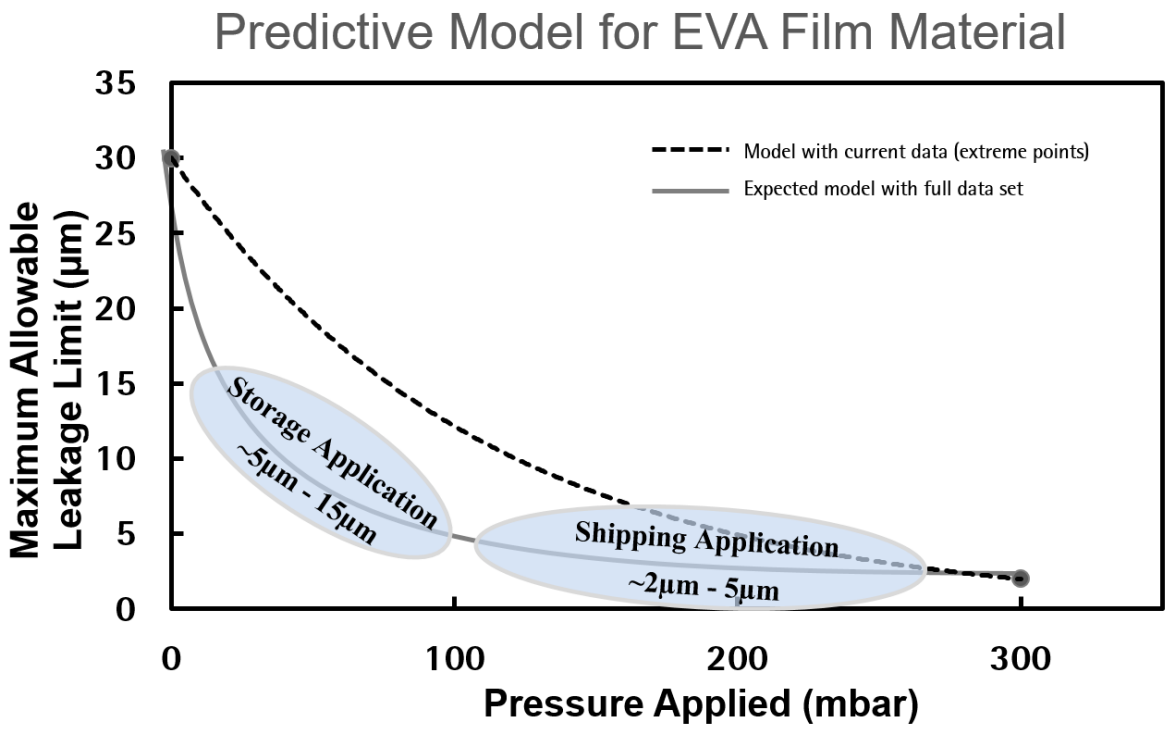


Probability for Microbial Ingress at **300 mbar** Pressure on PE Film



Predictive Model to Define Application-specific MALL

- Establishes the MALL for any process condition
- Based on observed data for liquid leaks and microbial ingress at various pressure conditions
- Each MALL is associated with a certain probability of leak/ingress



Interim Conclusion from Results Obtained on Liquid Leak & Microbial Ingress Studies on SUS

- Results obtained on film materials fall into the existing models for sterile package using micro tubes
- Both studies, liquid leak and microbial ingress ones, tend to confirm that leak sizes for liquid flow are not significantly different from leak sizes for sterility loss
- The conditions of use (pressure) does significantly impact the Maximum Allowable Leakage Limit (MALL)
- 2 µm is the MALL under any conditions and both, for liquid leak and microbial ingress

Case Study: Helium Integrity Testing

Case Study: Helium Integrity Testing

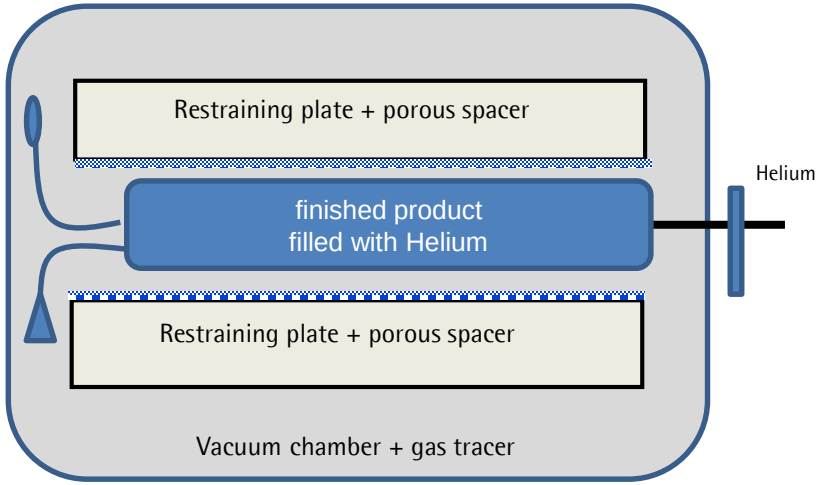
Main Requirements:

- Test of finished product (bag chamber incl. lines & connectors)
- 100% non-destructive testing (deterministic approach)
- Sensitivity that can be correlated to bacterial ingress
- Capable to be implemented in-line in routine production

Case Study: Helium Integrity Testing

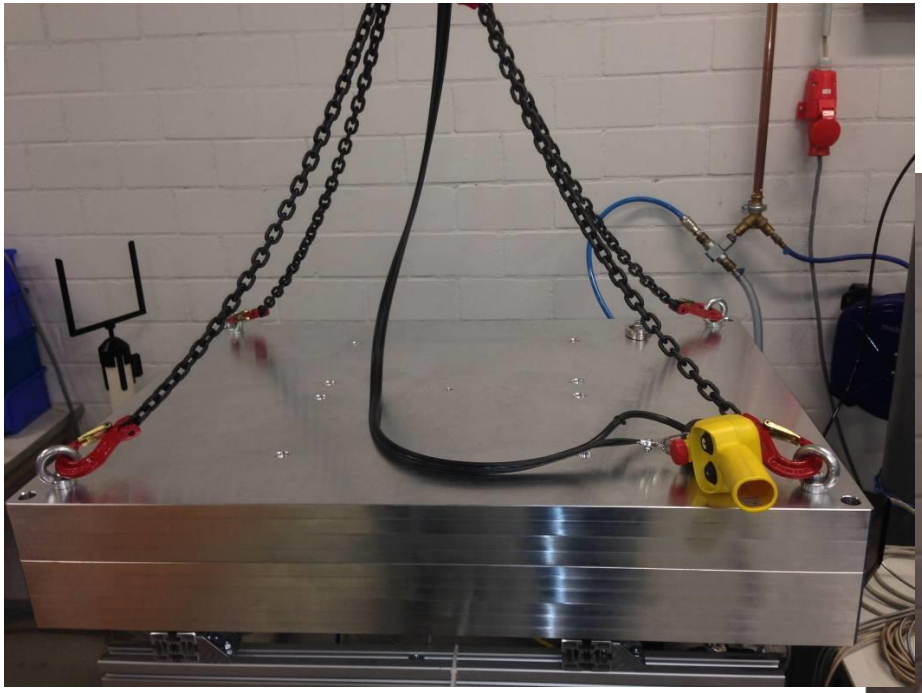
Test setup:

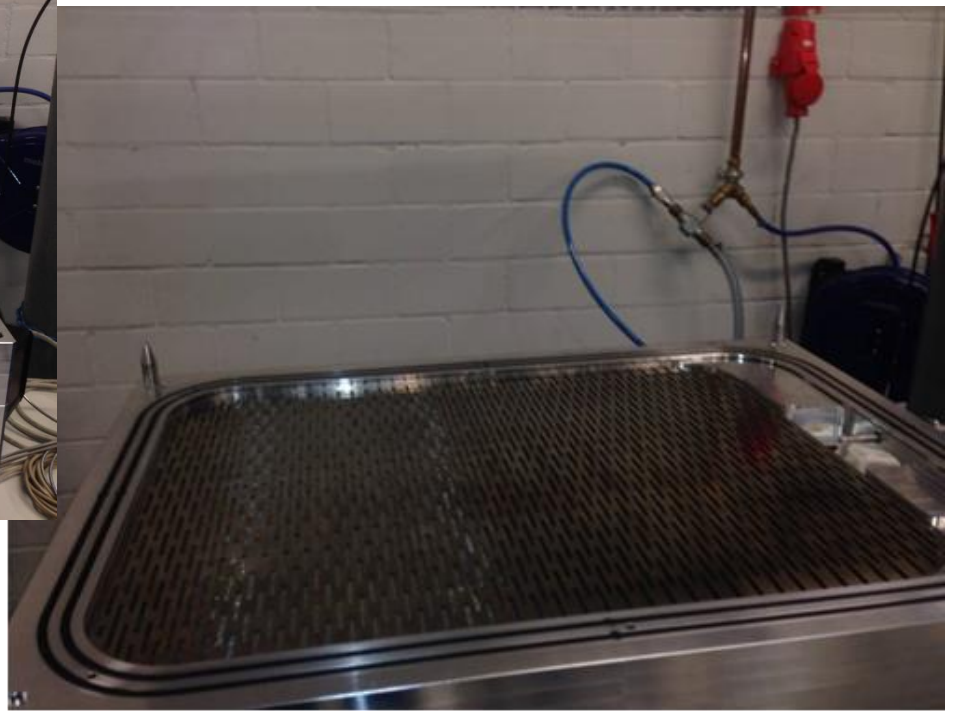
- Porous spacers avoid masking effect
- Restraining plates reduce stress on bag
- Test can be used as “Integrity Test” covering the whole SUS
- Gas leak rate can be correlated to a leak size of 2µm



small inflation volume + high test pressure = best sensitivity

Case Study: Helium Integrity Testing



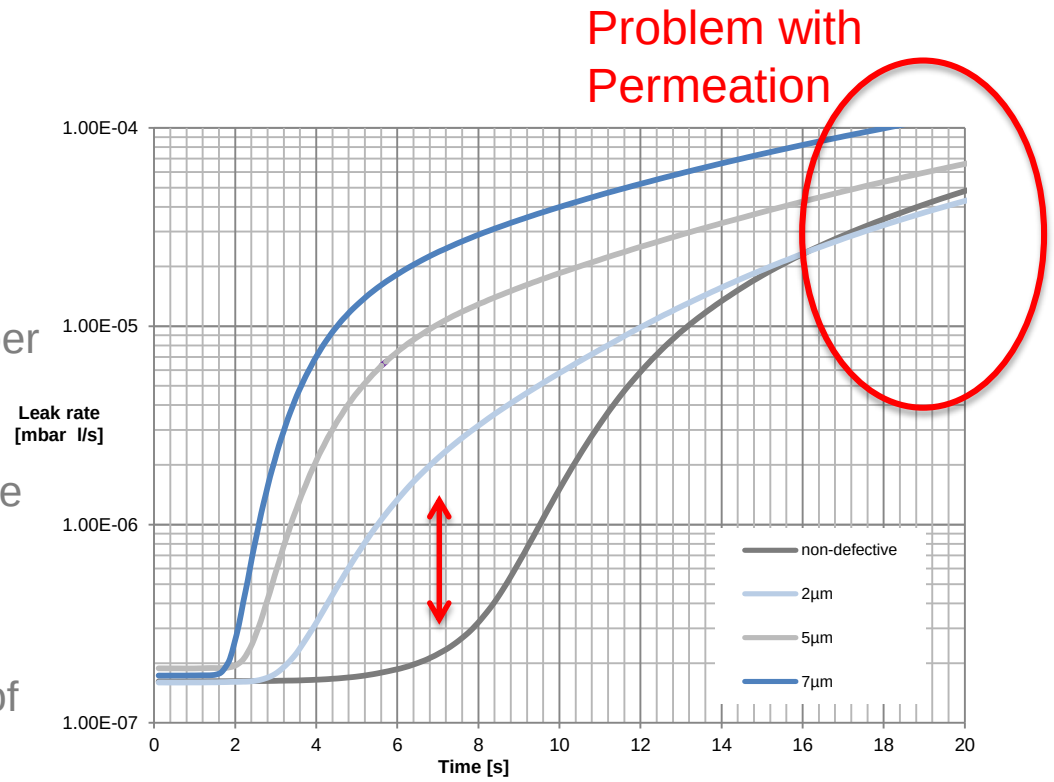


Feasibility testing on 50l test chamber

Case Study: Helium Integrity Testing

Test testing results:

- Feasibility tests done with three different test gases
- Results based on 20 samples per leak size (50l bags)
- One decade differentiation in the time frame between 4-10 s
- Helium gas tracer was the best test method to achieve a level of detection down to 2 µm



Case Study: Helium Integrity Testing

Challenges for industrialization:

- High permeability of helium through the thin bag film
- Outgassing of volatile components
- Vacuum level vs. tact time
- Helium background concentration & absorption

Case Study: Helium Integrity Testing

Solutions for industrialization:

- Machine concept with fast helium filling to measure before the start of permeation
  => dynamic leak rate evaluation
- High vacuum to get lowest background level
- High vacuum pump capacity to reach vacuum in a reasonable time frame
- 2 chamber design for alternating operation to reach target tact time
- Flexible chamber design to cover different product families

Case Study: Helium Integrity Testing

Helium Gas Tracer in Vacuum Chamber

- Restraining plates
  - Reduce stress on bag
  - Provide small inflation volume
  - Allow high test pressure: 300 mbar
- Porous spacer avoid masking effect of potential leaks

With low background noise and dynamic leak rate measurement helium gas tracer is the best method to achieve reliably highest sensitivity

➢ Detection limit: **2 µm correlated** to microbial ingress and liquid leaks at shipping pressure conditions

Case Study: Helium Integrity Testing

Validation Approach:

- A **reliable** test method using samples from multiple production lots
- A **reproducible**, **accurate** and **robust** test method using a 6 sigma confidence interval for the differentiation between defective and non-defective samples
- A **sensitive** test method using defective samples, calibrated on a pinhole equivalent
- A **One-for-all** validation procedure, whatever the test method is

Case Study: Helium Integrity Testing

Definition of defect:

- Aerosol microbial ingress & liquid leak studies performed on different bag film materials
- Identification of MALL for microbial ingress & liquid leak **under different process conditions**
- **2µm estimated as MALL** for worst-case (shipping) conditions -> target for the helium test validation
- Deliberate defects manufactured and confirmed with flow calibration

Case Study: Helium Integrity Testing

Definition of defect:

- Different types of defects tested: patches and capillaries
- Defects placed at worst-case positions

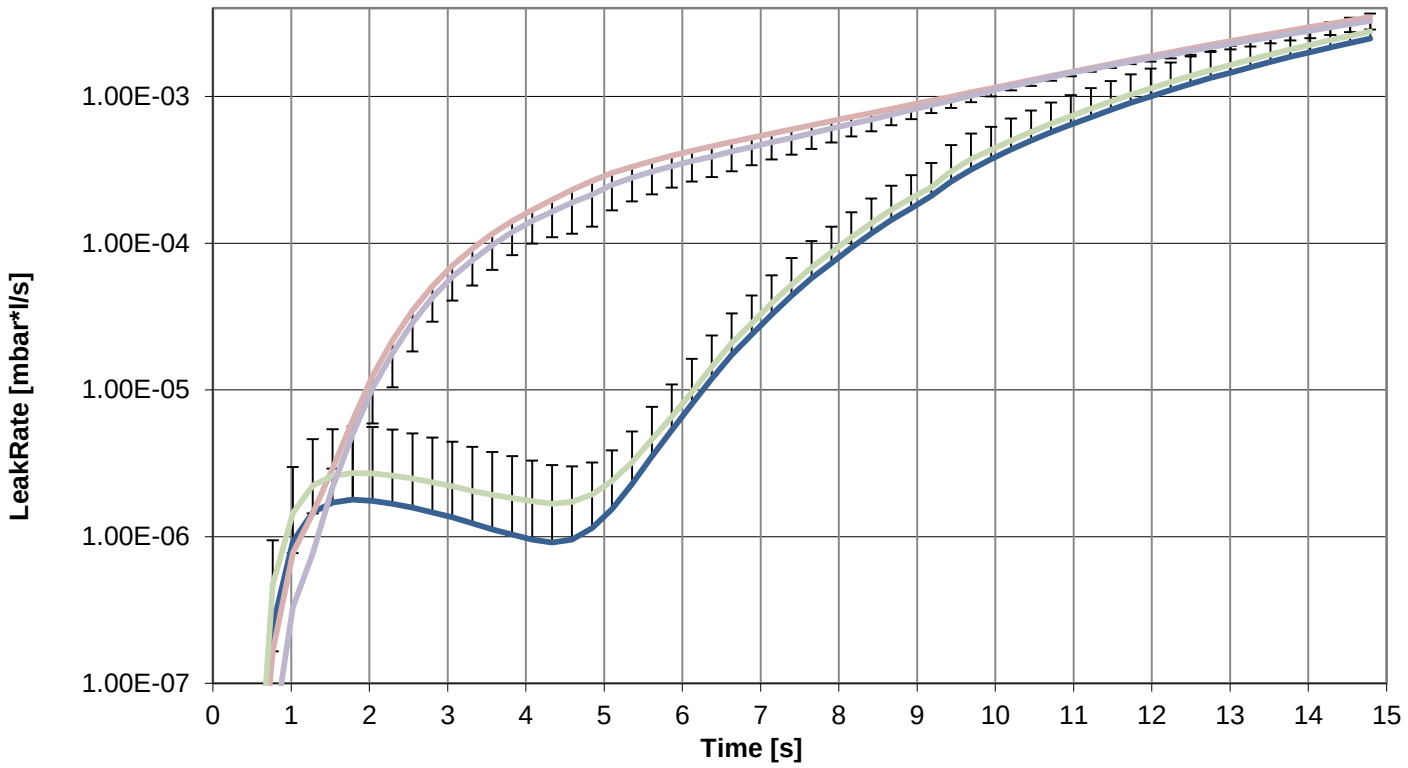
Case Study: Helium Integrity Testing

Parameter Study:

- Evaluate impact of different product designs
  - Volume
  - Components
  - Material
- Determine optimum test parameter set(s)
- Tests on a limited number of test samples

Case Study: Helium Integrity Testing

Parameter study: Results of 50l bags



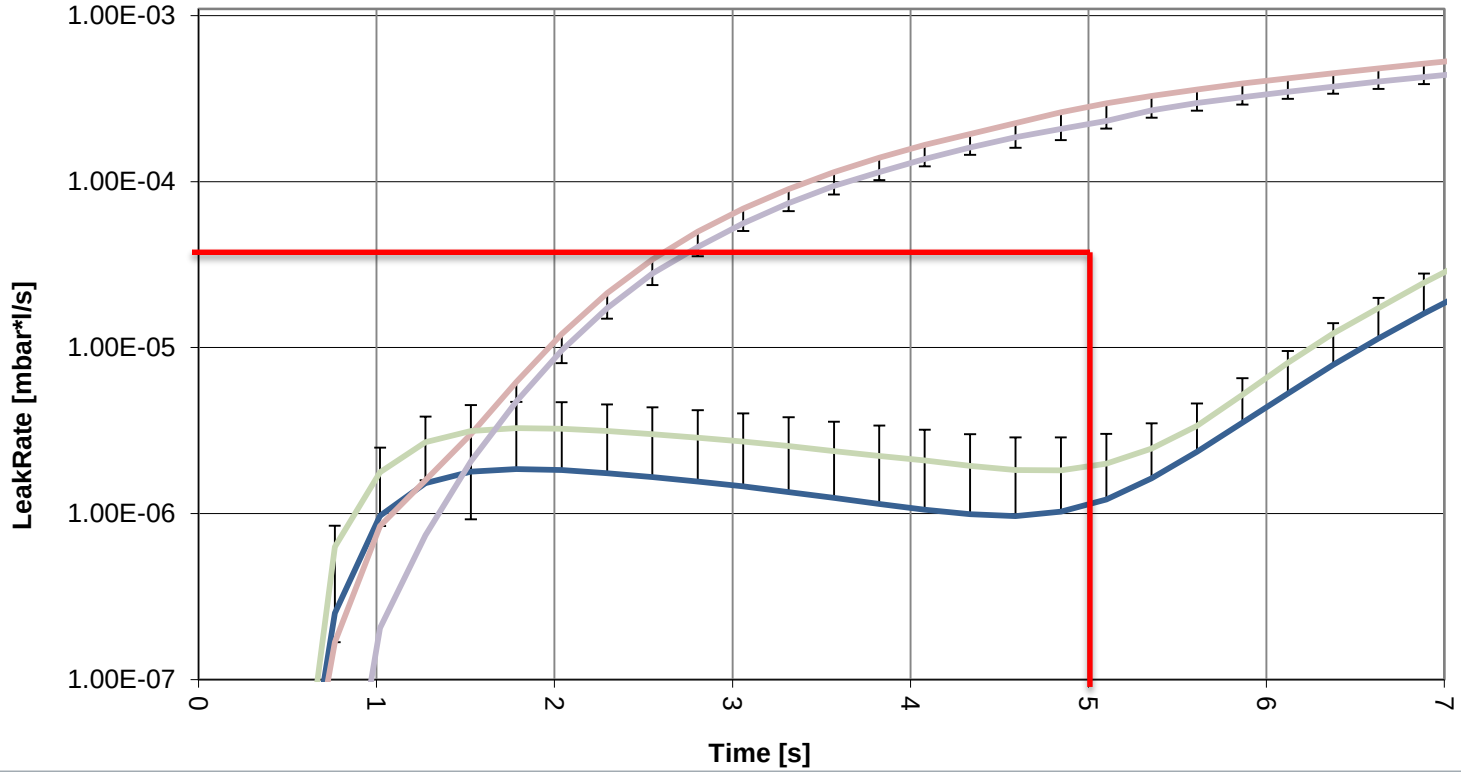
Case Study: Helium Integrity Testing

Validation Study:

- Verification of selected test parameter set(s)
- Determine pass/fail criteria (time & leak rate limit) => dynamic leak rate
- Tests on a statistically significant number of test samples

Case Study: Helium Integrity Testing

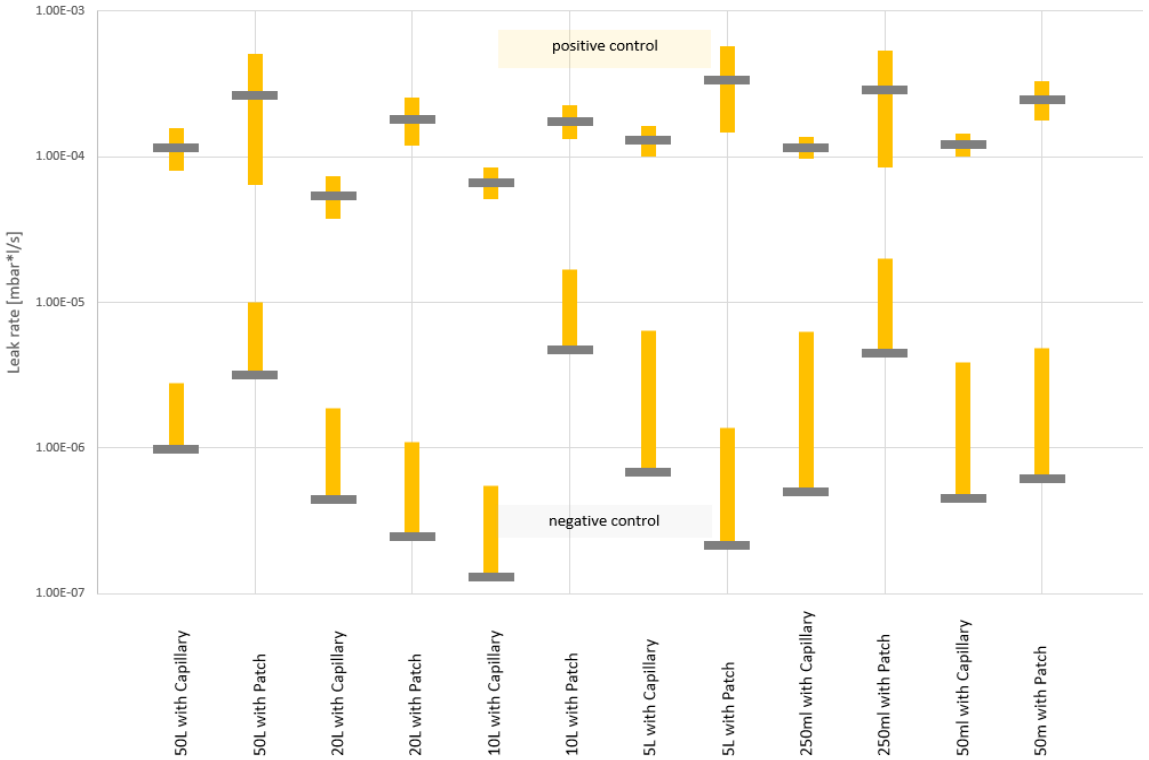
Validation study: Results of 50l bags



Case Study: Helium Integrity Testing

Validation Summary:

- Measurement of defective & non-defective samples
- Use of different types of artificial defect, like film patches & capillaries
- Defects deliberately calibrated
- Reliability proven using samples from multiple batches
- **2µm** detection validated applying a **6 sigma** confidence interval



Validation study with >380 tests to establish test specifications

Proven Integrity of Your Process Improves Patient Safety, Regulatory Compliance, Production Costs and Drug Availability

- Enhances patient and operator safety
- Prevents any risk of high value product loss at commercial phase
- Meets cGMP regulatory expectations for SU container closure integrity
- Speeds up drug product manufacturing capacity and market availability with SU proven process integrity

Thank you for your attention.
Questions?