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Filtration & Testing Products

Application Note

Integrity Test Troubleshooting – Beyond Rewet and Retest

Integrity testing is a critical operation, especially for sterilizing grade filters used in biopharmaceutical processing. When performed correctly, an integrity test is a fast, definitive, non-destructive way to assure filter retention performance. Fortunately, there are few ways a non-integral filter will pass the integrity test, eliminating the possibility a non-retentive filter is used undetected. Unfortunately, there are a lot of ways an integral filter can fail the integrity test, resulting in retests, lost time, lost productivity and potentially lost product.

Filter integrity tests are primarily based on capillary forces that hold liquid in the pores of wet membranes. The smaller the pores, the stronger the capillary forces. The “bubble point” test measures the force in gas pressure required to overcome the capillary forces, and therefore provide an assessment of pore size. The “Diffusion” type tests measure gas flow across the wet membrane at a pressure below the bubble point. If gas flow is below an established specification the assumption is capillary forces have not been exceeded and therefore, all the pores are small enough to meet retention requirements. Test errors come from any phenomena impacting capillary forces, gas diffusion, or gas flow or pressure measurement accuracy.

It is a common assumption that false integrity failures are the result of incomplete membrane wetting. Incomplete wetting is certainly a common problem, but it is not the only potential problem. Simply rewetting and retesting may or may not produce a passing result and may not reveal the root cause of the problem. In this Application Note, we will consider all the potential sources of test error and apply a logical approach to resolution and retesting. The goals are to strengthen confidence in the result, provide justification for retests, and ultimately, to understand specific challenges and eliminate them to assure the integrity test can be performed correctly the first time.

General Integrity Test Result Categories

It is helpful to consider the general magnitude of test results. These can be categorized into four ranges:

- **Pass**. Bubble point and/or diffusion are in specification and in typical range. For example, a filter with a minimum bubble point of 50 psi might have actual results in the range of 52 to 58 psi. Or a maximum diffusion rate of 13.3 mL/min and typical results range 8 to 12 mL/min. When passing results in the typical range are achieved we have the highest confidence in filter integrity.
- **Gross Failure**. For example, high gas flow at low pressure is observed with either bubble point or diffusion tests. Gross failure is typical of truly non-integral filters. If a filter is damaged due to high differential pressure, physical impact, or excessive heat, the resulting defect will be orders of magnitude larger than the pore size of the integral filter. The result is very low capillary forces and high gas flow at low pressure. When high gas flow at low pressure is observed, troubleshooting and retest procedures should be applied. But there would be low expectation that retests will show the filter to be truly integral.
- **Marginal Failure**. For example, bubble point specification is 50 psi and actual result is 48 psi. Or diffusion spec is 13.3 mL/min and actual result is 20 mL/min. Typically marginal results are not due to oversized pores, but due to phenomena impacting capillary forces or gas diffusion (ie. low surface tension, poor wetting) or test error. When marginal results are observed, troubleshooting and retest procedures should be applied and there would be a high expectation that retests will show the filter to be truly integral.

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- **Invalid Test**. Results are in spec but out of typical range. For example, bubble point spec is >/= 50 psi and actual result is 80 psi. Or diffusion spec is </= 13.3 mL/min and actual result is 0 mL/min. This is an indication of a problem with the test execution, most commonly a valve closed that should be open. While instances of this category of result are very rare, people responsible for executing tests or reviewing results should be trained to recognize when a test is invalid and initiate a retest.

Potential Causes of Integrity Test Failure

To think beyond 'poor wetting' as the root cause of all integrity test failures, it is valuable to consider all the potential causes of failure. The following list is not intended to be exhaustive. A specific application or installation could eliminate some of these causes, or create others. A process specific list should be created when developing a troubleshooting procedure for a specific site.

Table 1

Filter Related Failure Modes	Test Method Failure Modes
Membrane damage	Wrong test selected
O-ring damage	Wrong test gas used
Device damage	Leaks
Surface tension suppression	Instrument/gauges out of calibration
Poor wetting	Temperature change
Air lock	Valves improperly open or closed
Wrong membrane	Untrained operator
	Wrong wetting fluid

Develop an SOP

A well-constructed SOP is essential for efficient troubleshooting. The SOP should answer two fundamental questions –

- Is the filter integral or not? Answering this question accurately is critical, especially in post-use integrity testing situations where batch disposition depends on filter integrity.
- Why did it fail? Identifying a root cause of failure will allow that root cause to be addressed and corrective actions taken to minimize or eliminate future false failures. Knowing why a test failed is also valuable for justifying a re-test. There is a common misconception that two re-tests are allowed and then the filter must be considered non-integral. Blindly performing two re-tests without consideration for root cause is inefficient and a potential compliance issue. On the other hand, any number of re-tests might be considered if the root cause of the previous test result can be clearly identified and documented.

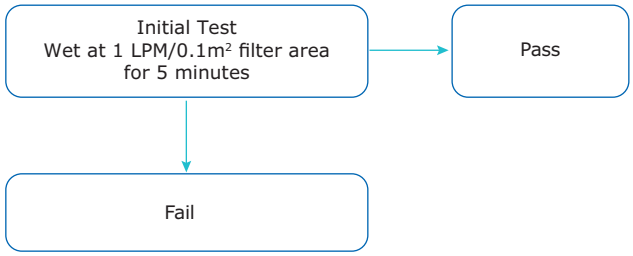
Develop a Troubleshooting Flow Chart

A flow chart based on an understanding of integrity testing principles and applies clear logic is a central component to a good integrity test troubleshooting SOP. There are many good examples, including in MilliporeSigma lit. no. P35515 (Wetting Guide) and PDA Technical Report 26. Specific site or application conditions and constraints need to be considered for each end user. But here we provide a general troubleshooting flow chart and the logic used to create it.

For the initial test, it is always important to follow as closely as possible the filter manufacturers wetting and testing recommendations. For Durapore® or Millipore Express® hydrophilic membrane devices, the recommendations include

- Fill system slowly
- Vent completely
- A one minute static high pressure hold
- 5 minutes of flow at 1 LPM/0.1m^2 filter area

These initial wetting conditions have been proven to be robust and capable of resulting in First Time/Every Time integrity test success.



If the integrity test fails after the initial wetting procedure, often the typical reaction is simply re-wet and re-test. If the reason for initial test failure is poor wetting, simply re-wetting may fix the problem and result in a passing result. But, as we saw in Table 1, poor wetting is only one of many reasons for test failure. Ignoring the possibilities beyond poor wetting can result in repeated failures, frustration, lost time or lost product.

Prior to a re-test, many of the failure modes in Tables 1 can be addressed by examining the system, the test and the filter. In **Table 2** the potential failure modes that can be identified without re-test are highlighted.

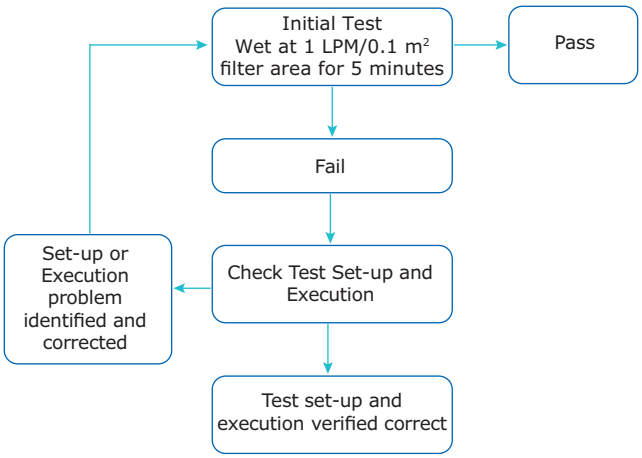
Table 2

Filter Related Failure Modes	Test Method Failure Modes
Membrane damage	Wrong test selected
O-ring damage	Wrong test gas used
Device damage	Leaks
Surface tension suppression	Instrument/gauges out of calibration
Poor wetting	Temperature change
Air lock	Valves improperly open or closed
Wrong membrane	Untrained operator
	Wrong wetting fluid

Some of the failure causes in Table 2, such as the wrong filter or test, are obvious in a visual check. Others, such as temperature changes require training to understand

the gas flow curve generated during the test, and being able to recognize the impact of various faults on the curve trend. For example, a plot of flow rate v. time during the diffusion test is expected to be a straight line. If the slope of the line changes during the test, this is an indication of a temperature change. Recognizing this and addressing the cause of the temperature change is important prior to running a re-test.

Step 2 in the troubleshooting flow chart is to check the test set-up and execution.



When the test is verified to be run correctly, the cause of failure must be due to one of the non-highlighted items in **Table 2**.

- Filter, seal or device damage
- Surface tension suppression
- Poor wetting
- Air lock

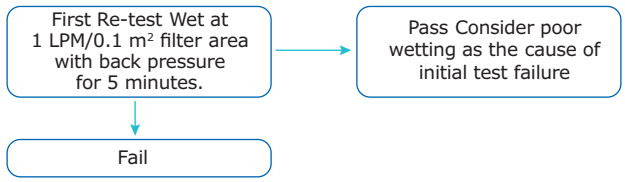
Enhanced wetting and re-testing are now needed. There are several options for enhanced wetting. First we must consider the options for enhanced wetting.

Table 3

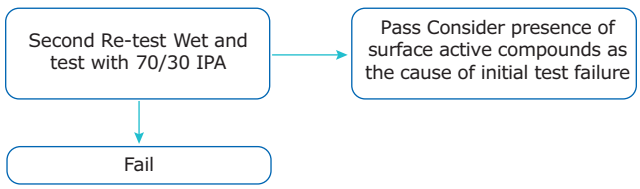
Wetting Option	Ease of Implementation	Failure Modes Addressed	Failure Modes Not Addressed
Longer Time	Easy	Poor wetting, non-adsorbed surface active residuals	Damage, Air Lock, adsorbed surface active residuals
Warm Water Flush	Easy if facilities are available	Poor wetting, non-adsorbed water soluble surface active residuals	Damage, Air lock, adsorbed surface active residuals
Higher Flow Rate	Easy depending on system capabilities	Poor wetting, non-adsorbed surface active residuals	Damage, Air Lock, adsorbed surface active residuals
Higher System Pressure	Easy	Poor Wetting, non-adsorbed surface active residuals, air lock	Damage, adsorbed surface active residuals
Alcohol	Complex when alcohol contamination in the process is a risk	Poor wetting, non-adsorbed surface active residuals, adsorbed surface active residuals, air lock	Damage

Of the 4 options in **Table 3** the most effective for demonstrating filer integrity is alcohol wetting. Because an alcohol solution such as 70/30 IPA/water has a low surface tension it will wet the membrane very thoroughly, overcoming problems of poor water wetting. In addition, surface active compounds that lower the surface tension of water are unlikely to impact the surface tension of alcohol solutions. Generally, specifications are provided for 70/30 IPA. Therefore alcohol wetting and testing will definitively answer the question about membrane integrity. But alcohol testing will not differentiate between wetting issues and surface tension issues. An initial re-test with alcohol therefore will not help answer the question “why did the first test fail?”

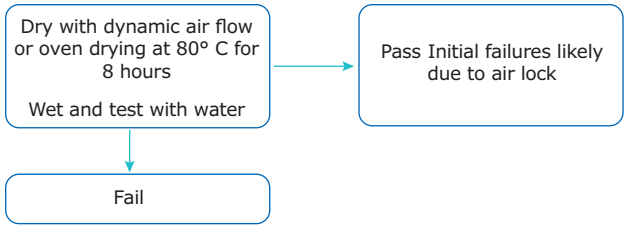
The second best option in **Table 2** is high pressure wetting. High pressure wetting means restricting downstream flow in order to create system pressure. Typically a system pressure of 40 psi is targeted but the enhanced wetting can be performed at whatever system capabilities are available and compatible with the pressure limit specifications of the filter device. Ideally, the flow rate of 1 LPM/0.1 m² filter area is maintained. With this technique air trapped in the membrane may be forced into solution and flushed out, fully wetting and solving the problem of poor membrane wetting. If the cause of failure is adsorbed surface active compounds, pressure wetting may be ineffective at flushing these compounds from the membrane surface.



If the filter fails again after high pressure wetting, it’s time for alcohol wetting and testing.



If the filter continues to fail after multiple wetting attempts, the problem may be air lock. Air lock is a phenomana where the upstream and downstream surfaces of the membrane are wet simultaneously, trapping a pocket of air within the thickness of the membrane. Air lock can be very difficult to remove with flushing. The best option for eliminating air lock is complete drying by dynamic air flow for 2 hours or static drying using a cross flow oven at 80°C for 8 hours. Filter should then pass after standard water wetting.



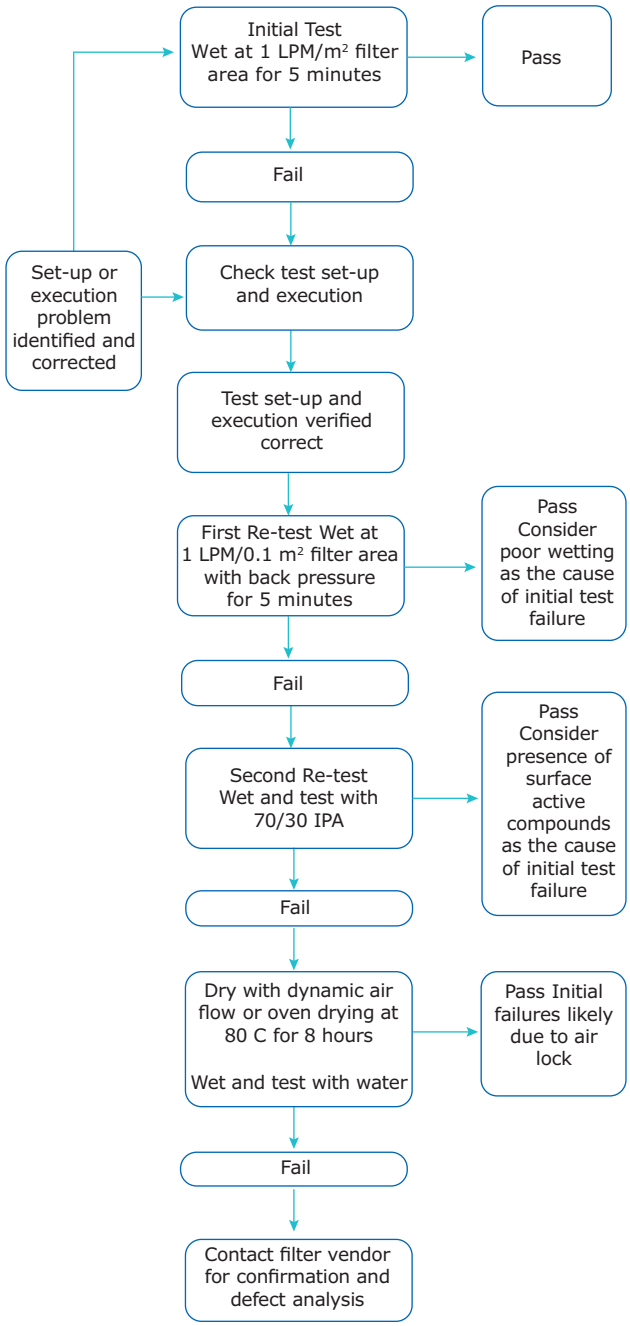
In the SOP it may be wise to allow a provision for a re-test at any step when a clear assignable cause for test inaccuracy is identified.

When all tests fail, it is recommended to return the filter to the vendor for confirmation and defect analysis. Defect analysis should be able to determine if loss of integrity is the result of physical impact, excessive pressure and/or temperature, a filter manufacturing defect, or another cause.

Summary

The complete troubleshooting flowchart is shown in the figure to the right. The procedure may be considered as is, or could be modified to incorporate process/product/site specific failure causes or testing constraints.

It is not always possible to have a definitive failure mode. For example, re-wetting with high pressure water may remove surface active compounds simply because it is additional flushing. But a procedure based on logic and typical known integrity test failure modes, should provide an efficient process for resolving failures and direction for identifying and applying corrective actions.



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