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Microbiological Test Method Validation?

It is a given in pharmaceutical cleaning validation that analytical methods for chemical residues, such as residues of the drug active or of the cleaning agent, will be validated for those specific residues. The principles of ICH Q2 are usually a starting point for such analytical method validation. For chemical residues, the analytical methods are demonstrated to appropriately analyze the specific chemical residue, usually in the test solution (the desorbed swab sample or the rinse sample). Performing recovery studies is also required (although this may or may not be part of the formal analytical method validation protocol).

How do we deal with microbiological testing methods for cleaning validation purposes? Can we do the same things for bioburden that we do for chemical residues? The general answer to that is "No". We certainly can't officially apply the principles of ICH Q2, because those are specifically for chemical residues. The additional question to ask is, "How would we validate the procedure?" This brings up the question of whether microbiological test methods, particularly those for enumeration, can be "validated". There are all sorts of questions about microbiological enumeration techniques, such as "What organism or organisms do I use to validate the method?" and "How can I be assured that the results with my lab organisms are truly appropriate for environmental (that is, on equipment surfaces) flora?" Furthermore, "If I do sampling recovery studies (recoveries from surfaces and not the USP <1227> recovery), what organisms do I use and how can I determine acceptable recovery?"

What is done for microbiological techniques, if they are not validated? My usual recommendation is to write in a cleaning validation policy/master plan that microbiological techniques do not require validation if they are "approved microbiology laboratory procedures". In other words, if my microbiology lab has an approved technique for enumeration in a membrane filtration procedure, no additional work is required for that procedure to be used for testing samples in a cleaning validation protocol. In one sense, that begs the question since I can then ask "How can a procedure be approved?" There is a truth to that, but the important thing is that I am now talking about "approved" procedures as opposed to "validated" procedures. Perhaps the issue now is similar to qualifying a pharmacopeial method in my lab. For example, I am more concerned about making sure that I am using the right media, that is prepared correctly, that cultures (for controls) are maintained appropriately, that the incubation conditions are appropriate, and that my operators are trained to successfully carry out the procedure. I will certainly set up controls (positive and negative) to check performance of a testing event (of course, I would want to do that for a validated chemical analytical procedure). I might also have to do some testing to demonstrate that residues of the active or cleaning agent are adequately neutralized and don't interfere with the results of the procedure by inhibiting growth. Each microbiology lab should have procedures to make sure specific test procedures are appropriately qualified. A good analogy is to treat it much like system suitability for pharmacopeial methods.

One difference between microbiological enumeration techniques and quantitative chemical analytical tests is that, in general, chemical procedures are specific for a chemical species, but microbiological techniques are designed to measure any of a variety of species. One might object that there are non-specific procedures like TOC that are not for measuring a certain chemical species, but rather any molecule containing organic carbon. However, when I use TOC in a cleaning validation protocol, typically I am using it to try to quantitate a certain chemical residue, and I will want to confirm (validate) that the TOC method can measure that specific organic

molecule. With a bioburden enumeration procedure, I don't know (usually) what organisms I might find (except that I might expect to find aerobes rather than anaerobes). Note that I don't want to take this analogy or contrast too far, because at some point it will fall apart and no longer be helpful.

The main point is that for cleaning validation purposes, microbiological enumeration methods should be handled differently than chemical quantitation methods in regards to any requirement for method validation. As I suggested before, the best way to handle this is to specify in a high level cleaning validation document that microbiological methods may be used if they are approved microbiological lab methods. Certainly, more work may be done, but one has to ask the value of that additional work. One example is that of performing quantitative surface recovery studies using the sampling techniques and microbiological methods. We all realize that microbiological sampling techniques give low recoveries from surfaces, but it adds little value (and is a frustrating exercise) to try to qualify sampling procedures using a recovery procedure (much like a recovery study for a chemical residue). Problems with recovery studies for microbial sampling were discussed in my July 2002 Cleaning Memo.

There will be some exceptions. For example, if one is developing an entirely new method, certainly a lot more work would be expected. However, the same criteria might apply: working to make the new procedure an "approved microbiology lab procedure", and then utilizing it (as appropriate) for a cleaning validation protocol. Another exception might be instrumental methods for identification of organisms. Certainly it is an expectation that those procedures be validated. However, the formal validation support package can be expected to be supplied by the instrument manufacturer, and something similar to a system suitability is all that is required.

I must confess I consider myself a "subject matter expert" in cleaning validation, but not in microbiology. I am also not a degreed microbiologist, but I do understand the arguments about what it means to validate a measurement procedure. Also, doing extensive work on microbiological method validation just for cleaning validation purposes is probably not going to get you into trouble from a compliance perspective. However, there may be other areas or tasks where you can more appropriately spend your time and money to improve quality and compliance for your cleaning validation program.