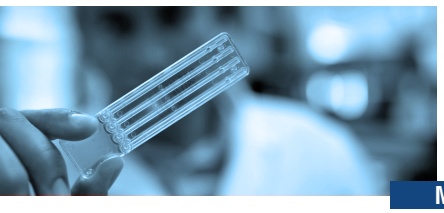
charles river



MICROBIAL SOLUTIONS

USP Chapter 1085 Frequently Asked Questions

The newest United States Pharmacopeia (USP) Chapter <1085> Guidelines on Endotoxins Testing fills in the regulatory gaps that have resulted following the withdrawal of the 1987 FDA Guideline and implementation of its replacement Q&A. Below are answers to some of the most frequently asked questions pertaining to USP Chapter <1085>. For more information, watch our webinar, What You Need to Know About USP <1085>, where the most important points of the chapter and the potential impact on our daily lab work are analyzed. We discuss:

- Preparatory requirements on RSE/CSE, consumables, analyst qualification, calibration, and the laboratory environment
- Method suitability for endotoxin limits, MVD, suitability testing, and common test interferences
- Routine testing for sampling, pooling, out of specification results, and retesting considerations and standard curve control
- Alternate test methods

Is USP Chapter <1085> applicable globally, or is it only for the US?

Every country will have their own regulatory bodies to refer to for guidelines and recommendations. USP is written for, and enforced in, the United States, but other countries reference USP material as an official regulatory source. USP should not take precedent over individual countries' written guidelines and requirements, but some countries do not have official compendia, so it is a reputable regulatory reference. USP should be used by any manufacturers outside of the US who are exporting product to the US, to ensure that they meet all the necessary requirements for importing to the country.

Does Charles River have any other documentation or input on how to best conduct an Out of Specification (OOS) investigation?

While the FDA guideline and USP Chapter <1085> provide very helpful information, each company will ultimately need to decide for themselves how to do their own investigations. Investigation processes can range from a simple, small checklist to a robust, 10+ page document. However, investigations should be thorough as the goal of the assay is ensuring product quality and patient safety. But that doesn't mean your Standard Operating Procedures (SOPs)/procedures can't be written to streamline that process.

Why isn't the Maximum Valid Dilution (MVD) adjusted for medical devices?

Here is an example for the US. Humans can tolerate 5 EU/kg of our body weight. If an average US person weighs 70 kg, that is 5 x 70 = 350 EU total. A medical device limit is 20 EU/device, so if 1 device is being implanted, and it can have a max of 20 EU, it is well under that 350 EU that humans can tolerate. Even if 10 devices are pooled and had 20 EU on each device, that would only be 200 EU, which again is still below the 350 EU that humans can tolerate. Medical devices already have safety factors built into them, so there is no need to add more safety factors through an adjusted MVD. The formula K*N/V is used to convert the limit from EU/device to EU/mL, with K being the endotoxin limit, N the number of devices, and V the total extract volume used. This formula considers the volume of extract and adjusts the endotoxin limit, in EU/mL, accordingly. The MVD can then be calculated from this limit, should further dilution of the extract be necessary.

Should analysts go through an endotoxin assay qualification periodically, or is the initial qualification good enough?

At the very least, analysts should do an initial qualification to show they can perform the assay, although some labs set schedules for re-qualifying their analysts. Charles River offers an endotoxin Proficiency Testing Program with 4 sessions per year that could be used for re-qualifying analysts. Also, if there is an analyst who is doing endotoxin testing daily, or even weekly, normal testing can be used as proof that they are qualified to run the assay. This is ultimately up to each company to decide.

What should be done if only a single lot is available for sample suitability/validation?

Some lots of products are manufactured once every few months or years. Of course, this is not typical, but in this case, it would not be acceptable to wait to validate on subsequent lots, or the validation could take many years. In a situation like this, and with appropriate risk assessments in place, you could validate the one lot on three separate days, with three different analysts performing the testing. After that, every time this product is tested, you are basically gathering more data that give you more confidence in the validation over time. While you are performing validation testing on one lot of product, keep in mind that other testing is involved, such as in-process or raw material testing. As noted in USP Chapter <1085>, it does suggest that the historical three lots may or may not be enough depending on certain factors.

When calculating the endotoxin limit according to the package insert of the product, but it is lower than the pharmacopoeia monograph, how is the endotoxin limit for raw materials of the product determined?

One should always choose the limit that is the most stringent when given multiple options. The goal of the assay is ensuring product quality and patient safety, so the safest option from a patient point of view is the lower limit. When determining the limit for a raw material that goes into that final product, it is recommended by both Charles River and USP Chapter <1085> to perform a risk assessment. Look at the source of the raw material, historical endotoxin in that raw material, historical endotoxin in the final product, % makeup of the final product, and any process that may increase or decrease the endotoxin levels. For example, putting a liquid raw material in an oven with higher temperature prior to going into the final product; that process would decrease endotoxin so you could set a lower limit for it. The risk assessment will help you determine what limit to apply. Before deciding on what limit to set, make sure you understand the interference properties of the raw material to ensure that you can test to that limit. There's nothing worse than setting a limit and then finding out that the raw material interferes with the test, so much so that you cannot get valid recovery within the MVD/MVC.

In the treatment of the results, is it better to use a linear regression or a polynomial curve?

Linear and polynomial both have their pros and cons, but one isn't necessarily better than the other. Regulations require that the linearity of the standard curve (the R-Value) be ≥ 0.980|, so if using polynomial, Charles River's advice is to first ensure the curve is valid with a linear regression. The Endosafe® EndoScan-V™ software has that built-in capacity to not let the user use polynomial if the R-Value is not linear to begin with. Linear regression is generally more stringent, i.e., less forgiving of errors than polynomial, which is why a valid linear regression is needed before using polynomial. Linear regression is normally sufficient for curves with a two or three log range, when used with quality reagents and accessories, by a proficient analyst.

In the case of using FDA-licensed Endosafe® cartridge technology, how do we qualify the analyst?

When utilizing cartridges, analyst qualification procedures are lab determined. At Charles River, we have the analyst test LAL Reagent Water (LRW) on a cartridge to ensure the analyst obtains a valid assay. Testing a known sample that has interference and/or endotoxin could also be a way to further qualify analysts. Our Charles River Proficiency Testing Program will ship a vial of unknown endotoxin for the analyst to test. After uploading the results, Charles River will contact the customer as to if the value is accurate or not. This is a good "blind" way to qualify your analysts, but every lab has to decide for themselves how to qualify their analysts when using Endosafe® cartridge technology.