PDA Global Headquarters
Bethesda Towers,
Suite 600
4350 East West Highway
Bethesda, MD 20814 USA
TEL: +1 (301) 656-5900
FAX: +1 (301) 986-0296

PDA Europe gGmbH
Am Borsigturm 60
13507 Berlin
Germany

22 March 2024

Leslie Furr, Associate Scientific Liaison
USP Compendial Science
12601 Twinbrook Parkway
Rockville, MD 20852

Reference: USP Chapter <1119.1> Bioburden Test

Dear Madam or Sir,

PDA appreciates the opportunity to provide feedback to the USP Microbiology Expert Committee on the proposed addition of the new chapter for Bioburden Test <1119.1>. In our attached comments, PDA offers specific comments and feedback that we believe will be helpful in the further development of this important Chapter.

PDA is a non-profit international professional association of more than 10,000 individual members comprising scientists, industry professionals and consultants having an interest in fields of pharmaceutical, biological, device manufacturing, and quality. Our comments have been prepared by a committee of PDA members with expertise in the areas covered in the Public Docket on behalf of PDA's Science Advisory Board.

If you have any questions, please do not hesitate to contact me via email at wright@pda.org.

Sincerely,

Glenn E. Wright
President and CEO

cc. Josh Eaton,PDA; Carrie Horton,PDA; Jessie Lindner, PDA; Danielle Bretz, PDA

USP <1119.1> Bioburden Test

General Comments		
Comment to Text	**Proposed Change**	**Rationale for Change**
Throughout the document, the terms "limit", "specification" and "acceptance criteria" are used. PDA recommends avoiding the use of these terms due to variations in reader interpretation and these terms are typically interpreted with release testing which is not covered within the scope of this Chapter. Therefore, the use of these terms is inaccurate.	PDA suggests the use of the term "level(s)" in place of the currently used terms of "limit(s)" "acceptance criteria" or "specification".	The scope of this chapter is for bioburden monitoring and not release testing; therefore, the term "level" more accurately reflects the scope of this Chapter.

Section: Growth Promotion Test, Negative Controls, and Suitability of the Counting Method – *Preparation of Test Strains*			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
Table 1. Preparation and Use of Test Microorganisms "Suitability of Counting Method in the Test Matrix and Bioburden Testing"	PDA recommends updating column heading to remove reference to Bioburden Testing.	*Table 1. Preparation and Use of Test Microorganisms* "Suitability of Counting Method in the **Presence of Test Matrix**"	Bioburden testing incubation requirements are included in the Testing for Bioburden Section of this Chapter. Also, the table is specific to incubation conditions associated with growth promotion and method suitability testing. By making this change, this Chapter verbiage would align with that found in Chapter⟨61⟩.

Section: Growth Promotion Test, Negative Controls, and Suitability of the Counting Method – *Growth Promotion of the Nutrient Culture Media*			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
"Inoculate portions or plates of the nutrient culture media with a small number [not more than 100 colony-forming units (CFU)] of the microorganisms indicated in Table 1, using a separate portion or plate of nutrient culture medium for each. Incubate according to the conditions described in Table 1."	PDA recommends updating guidance to remove references to "portions" so the reader is not directed to prepare multiple microorganism strains on a single plate.	"**Inoculate plates** of the nutrient culture media with a small number [not more than 100 colony-forming units (CFU)] of the microorganisms indicated in Table 1, **using an individual** plate of nutrient culture medium for each. Incubate according to the conditions described in Table 1."	With an acceptable recovery percentage (recovery ratio) of 50-200% of the inoculum, there is a risk to create too numerous to count conditions if the upper limit of 100 colony-forming units is approached and multiple organisms are inoculated on a single plate. For this reason, it is advised to use one plate per inoculum.
"Liquid nutrient culture media are deemed suitable when clearly visible microbial growth is comparable to that from a previously qualified batch of nutrient culture medium."	PDA recommends deleting this text.	**Delete the stated text.**	In the Chapter Section, Testing for Bioburden, it describes Membrane Filtration, Pour Plate and Surface Spread Methods. Liquid nutrient culture media is not used for any of these methods.

Section: Growth Promotion Test, Negative Controls, and Suitability of the Counting Method – *Suitability of the Counting Method in the Presence of Product:*			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
Suitability of the Counting Method in the Presence of Product	PDA encourages the addition of a section/sub-section addressing the amount of product to be used.	**Recommend adding similar guidance as in <61> as per below along with a reference to Chapter <61> for additional guidance:** • **Water-Soluble Products** • **Nonfatty Products Insoluble in Water** • **Fatty Products**	By adding this section/sub-section, it will provide additional guidance for the reader on sample quantity. Adding reference to Chapter <61> for additional guidance will give readers awareness of which Chapter provides detailed guidance regarding this topic. While not all of Chapter ⟨61⟩ is applicable to the scope of the products covered in this Chapter, guidance would be applicable to some products (e.g., powders).
"Add a sufficient volume of microbial suspension (not more than 100 CFU) to the prepared sample and a control (with no test material included). The inoculum volume should not exceed 1% of the volume of the prepared sample."	PDA recommends adding clarification to the term "prepared sample" so readers can link back to the test material.	"Add a sufficient volume of microbial suspension (not more than 100 CFU) to the prepared sample **(test material)** and a control (with no test material included). The inoculum volume should not exceed 1% of the volume of the prepared sample **(test material)**."	Provides the reader with additional clarity of the prepared solution versus the control which already contains clarification.
"Prepare a test sample as described under *Test Sample Preparation*, *Inoculation and Dilution*, and *Neutralization*. For each of the microorganisms listed, separate tests should be performed in duplicate."	PDA recommends removing reference to "in duplicate" from this paragraph.	"Prepare a test sample as described under *Test Sample Preparation*, *Inoculation and Dilution*, and *Neutralization*. For each of the microorganisms listed, separate tests should be performed."	Requirements for performing in singular or duplicate is detailed within the subsequent sections based on the method. For membrane filtration, the relevant section states to perform in singular so the edit will ensure alignment throughout the section.

Section: Growth Promotion Test, Negative Controls, and Suitability of the Counting Method – *Suitability of the Counting Method in the Presence of Product:*			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
"Transfer a suitable quantity (ensuring adequate test method sensitivity) of the test sample to the membrane filter, filter immediately, and rinse the membrane filter with an appropriate volume of diluent."	PDA recommends clarifying statement so readers do not misinterpret that a rinsing step is a requirement in all cases as this should be driven based on method suitability. Additionally, this Chapter's scope includes water samples.	"Transfer a suitable quantity (ensuring adequate test method sensitivity) of the test sample to the membrane filter **and, filter immediately. Where determined necessary via method suitability**, rinse the membrane filter with an appropriate volume of diluent."	This update clarifies for the reader that a rinse step is not needed in all circumstances (e.g., standard solution products) and directs the reader back to method suitability to determine if a rinsing step is appropriate and if appropriate, the amount to rinse the filter.
"Surface-spread method: For Petri dishes 9 cm in diameter, add 15–20 mL of soybean–casein digest agar held at not more than 45° and allow to solidify. If larger Petri dishes are used, increase the volume of the agar accordingly. Dry the plates, for example, in a laminar-air flow cabinet or an incubator."	PDA suggests updating this statement to clarify that the recommended guidance is not applicable to ready-to-use media.	**"Surface-spread method: For media plates prepared from dehydrated media**, add 15-20 mL of soybean–casein digest agar held at not more than 45**° to Petri dishes 9 cm in diameter** and allow to solidify. If larger Petri dishes are used, increase the volume of the agar accordingly. Dry the plates, for example, in a laminar-air flow cabinet or an incubator. (**NOTE: Not applicable for ready to use media**)."	This update clarifies for the reader that the steps discussed in this statement of the guidance are not needed/appropriate for ready-to-use media.

Section: Testing for Bioburden			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
"The amount of sample tested must be sufficient to determine compliance with the established specification for bioburden."	PDA recommends alternate wording to clarify the intent behind this statement.	**"The sensitivity of the method (e.g., sample quantity tested and/or dilutions) must be sufficient to determine compliance with the established bioburden level.**"	Revised wording will provide additional clarity to the reader for ensuring the test parameters are suitable for supporting the respective bioburden level.

Section: Testing for Bioburden			
Current Text	**Comment to Text**	**Proposed Change**	**Rationale for Change**
"Transfer the appropriate amount to a membrane filter, filter immediately, and wash the filter."	PDA recommends updating the current text to provide additional guidance and clarification on when it is suitable to "wash the filter".	"Transfer the appropriate **test** amount to a membrane filter, filter immediately **and where required, rinse with the appropriate volume of diluent based on method suitability."**	This update clarifies for the reader that a rinse step is not needed in all circumstances and directs the reader back to method suitability to determine if a rinsing step is appropriate and if appropriate, the amount to rinse the filter.
"Transfer the membrane filters to the surface of soybean-casein digest agar and incubate at30°–35° for 3–5 days. Calculate the number of CFU per quantity (e.g., CFU/mL, CFU/g, or CFU/unit) of sample."	PDA recommends changing the membrane filters from plural to singular format to align with the rest of the details around membrane filtration which were stated in singular.	"Transfer the membrane **filter** to the surface of soybean-casein digest agar and incubate at30°–35° for 3–5 days. Calculate the number of CFU per quantity (e.g., CFU/mL, CFU/g, or CFU/unit) of sample."	Based on the entire section for membrane filtration, the intention was for the testing to be performed in singular which aligns with the use of singular terminology throughout with exception of this one reference in plural. This also aligns with the use of one filter under the method suitability section for membrane filtration.