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Standard Guide for Determining the Impact of Extractables from Non-Metallic Materials on the Safety of Biotechnology Products[1]

This standard is issued under the fixed designation E2097; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This guide covers procedures and test methods for process component qualification by the end user. The goal is to assess the safety impact of extractables from non-metallic process components used in contact with bioprocessing solutions. This encompasses the impact of extractables on the safety of the final product as it passes through the various stages of the manufacturing process. This guide is not designed for evaluation of metallic materials, final product container/closures or those components intentionally added to the product or production streams during the manufacturing process. Testing of solids and extracts is specified in other ASTM standards. Materials must be qualified by specific use.

1.2 The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.

1.3 There is no companion guide available.

1.4 *Safety/Fire Hazards*: Extractions with organic solvents will be infrequent under this guide, but, when used must be treated as potential fire/explosion hazards.

1.5 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 *ASTM Standards:*[2]

D4951 Test Method for Determination of Additive Elements in Lubricating Oils by Inductively Coupled Plasma Atomic Emission Spectrometry

F619 Practice for Extraction of Medical Plastics

3. Terminology

3.1 See the *Dictionary of Engineering Science and Technology*. Review with Subcommittee E48.91 on Terminology. See Section A7 on Terminology and Part E on Terminology in ASTM Standards in *Form and Style for ASTM Standards* for details.

3.2 *Definitions:*

3.2.1 *biopharmaceutical*—any drug product produced from living organisms.

3.2.2 *biotechnology solution*—a solution containing or producing products from living microbial, animal or plant cells or by the enzymes from those cells.

3.2.3 *biotechnology product*—a discrete chemical entity produced by growing single cell organisms with unique genetic information.

3.2.4 *elution cytotoxicity*—see USP.

3.2.5 *emission spectrographic analysis (ESA)*— an analytical technique for determining metals in a sample vaporized in a plasma arc.

3.2.6 *extractables*—residues from solid process components not intentionally part of the product process stream.

3.2.7 *fermentation*—the biochemical reaction process where microorganisms in a nutrient medium convert a feedstock to a product.

3.2.8 *inductively coupled plasma (ICP)*—an analytical technique designed to quantitate chemical elements.

3.2.9 *materials of construction*—high molecular weight or solid materials, used in biopharmaceutical process equipment which contact process solutions and can potentially release extractable residues.

3.2.10 *non-volatile residue (NVR)*—non-volatile material remaining after evaporating a solvent into which the residue has been extracted (see USP).

3.2.11 *oxidizable substances (OS)*—chemical compounds which may be oxidized by potassium permanganate under specified conditions (see USP).

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[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

3.2.12 *product contact material*—a material which physically contacts a solution containing the chemical entity designated the product.

3.2.13 *process materials*—high molecular weight or solid materials which contact process solutions potentially releasing extractable residues.

3.2.14 *purification*—the process by which the desired product is separated from the production process solution.

3.2.15 *residue on ignition (ROI)*—the residue remaining after ashing a material at high temperature.

3.2.16 *total organic carbon (TOC)*—an analytical technique for measuring the carbon associated with organic molecules in a solution.

4. Significance and Use

4.1 This guide applies to the determination of the safety of non-metallic materials used in contact with biotechnology product containing solutions. Process materials leach low level of residues into water, cell culture media, buffers, and other product containing solutions. This document offers guidance on determining the safety of these materials (process materials) for use. The goal is to prevent toxic extractables from entering process streams and ultimately contaminating the final product in unacceptable levels.

4.2 The purpose of this guide is to describe tests to qualify materials with respect to any extractable substances so as to prevent unintentional introduction of a potential source of objectionable substances. An extractable material is objectionable if it is toxic, interacts with product constituents, interferes with required assays, or otherwise affects the process stream so as to adversely affect critical quality parameters, for example, purity, safety, efficacy, identity, strength of the final product or its successful production. All organizations producing pharmaceutical products should consider the points in this guide when qualifying process materials for use in their production processes.

4.3 This guide outlines the application of the process material tests primarily in ASTM or USP. Typical process materials include high molecular weight polymers and solids such as hoses, filters, filter housings, containers, valve diaphragms, gaskets, o-rings, chromatography resins, and chromatographic columns.

4.4 The battery of tests described in this guide is intended to cover a wide variety of potential attributes of materials and to characterize possible extractables.

4.5 The material specification will vary depending on the impact on the final product and the point in the process that the product solution contacts the material. Tighter specifications should be considered for extractables for final product purification process materials than for fermentation media process materials.

5. Reagents

5.1 The quality of reagents used for the procedures indicated in this guide are specified in the test standards referenced (for example, ASTM and USP).

6. Procedure

6.1 During research and development to define the manufacturing process for a desired biotechnology product, select functional product contact materials predicted to be suitable based on manufacturer specifications. Choose materials which have specifications defined by pharmaceutical compendia to the extent possible. The goal is to find and use materials that will permit an acceptable level of extractables into the process solution. Materials should be approved by specific process use. A written protocol should be prepared outlining the tests to be done on each process material qualified. Qualified materials must be well defined and documented to assure equivalent replacements may be obtained. Vendor audits are necessary for all suppliers of product contact material with significant extractables.

6.2 When a high quality, functional material is identified, subject it to the following procedure as part of the validation of the process.

6.3 Choose the production function from Table 1. Use already validated ASTM or USP test methods wherever possible. If the product is to be licensed in a country with other compendial requirements, those will have to be considered as well. If test methods are the same but limits are different, use the more stringent limits.

NOTE 1—The cumulative effect of the ongoing removal of extractables can potentially affect the performance of plastics in certain applications.

6.4 Perform the tests designated in Table 1. Where extractions are done, follow Practice F619 – 79 (1991). Increase the time, temperature and concentration of the extraction several fold beyond production conditions to build in safety factors and insure worst case. Also it may be appropriate to exacerbate other factors affecting the extraction capability of the solvent such as organic concentration and pH. Demonstration of depletion of extractable material can be shown by repeated extraction and testing for non-volatile residue or oxidizable substances.

6.5 Characterize the product contact process material by through the film, pyrolysis, attenuated total reflectance or solution infrared methods. The infrared scan will become the reference for subsequent lots of the material unless a manufacturer or other valid scan is available.

6.6 Evaluate the product contact process material for heavy metals using Residue on Ignition followed by Emission Spectrographic or Inductively Coupled Plasma methodologies. In this case the amount of Residue on Ignition is not important except as it allows you to calculate the concentration of metals in the solid. If unacceptable levels of heavy metals are found, appropriate extracts should be tested by Atomic Absorption Spectroscopy to determine if the metals are extractable into the relevant process solution.

6.7 *Distilled Water Extract*—Follow Practice F619 – 79 (1991), Sections 6 through 12. When choosing a set of extraction conditions, choose a temperature similar to the worst case use conditions. Extend the extraction time as appropriate to create safety factors.

TABLE 1 Recommended Tests on Non-Soluble, Non-Metallic Process Components Used in Biotechnology Manufacturing

Test	Test Method	Fermentation	Purification	Final Product
Solid Component				
Characterize by IR	Thin film or ATRIR	A	B	C
Metals by ESA or ICP	Ash, and analyze residue	A	B	C
Dried Extract				
Characterize by IR	KBr pellet	A	B	C
Metals[A] by AA or ICP	Ash, and analyze residue	A	B	C
Water Extract Solution				
Appearance	Visual	A	B	C
Oxidizable Substances or TOC	USP 23 for Purified Water	A	B	C
Non-Volatile Residue	USP 23 for Plastic Extract	A	B	C
Elution Cytotoxicity[A]	USP 23 for Plastic Extract	A	B	C
UV Scan		A	B	C
Organic Solvent Extract[B]				
Appearance	Visual	A	B	C
Non-Volatile Residue		A	B	C
UV Scan		A	B	C

A = Specification for Fermentation Process Materials
B = Specification for Purification Process Materials
C = Specification for Final Product Process Materials (other than container/closure)

[A] If elution cytotoxicity fails, run United States Pharmacopeia 23: <87> Biological Reactivity Tests, In-Vitro, p. 1697. Failure of this latter test renders a material unacceptable for product contact.
[B] If Applicable

6.7.1 *Oxidizable Substances*—results will normally be significantly higher than USP Purified Water limits.

6.7.2 *Total Organic Carbon*—measurements are very sensitive and may be used in place of oxidizable substances in organic free solvents.

6.7.3 State of the art materials should be significantly below the current USP limit for non-volatile residue from plastic containers. Extraction solvents must be pure and used as controls.

6.7.4 The UV/VIS scan of water solutions should show only show end absorption as defined in the USP Monograph for Dehydrated Alcohol. Significant UV absorbing peaks may indicate reactive or toxic compounds. These compounds may need to be identified if any cytotoxicity is observed.

6.7.5 Extract solutions of state of the art materials should show no elution cytotoxicity. If a cytotoxic effect is observed, a toxicologist should be consulted.

6.7.6 Organic solvent extracts should be done if product is contained in same. Record the amount of residue into such solvents and look for the residue or a marker in the final product if levels are significant.

6.7.7 Further identification of extractables including chromatography, infrared, mass spectrometry, or other appropriate methods should be undertaken if the extractables level is such that it would impact final product quality. Identification of a well defined component of the extract allows use of that material as a marker in the final product for extract residues.

6.7.8 Consideration should be given to endotoxin content of extracts if applicable. It should be recognized that a number of materials can give false positives in the Limulus Amebocyte Lysate assay.

6.7.9 Repeat the extraction a second time. Test the extract for one or more critical parameters (for example, non-volatile residue). If the level of the extractable residue is not reduced, additional review is needed to determine the extent of the total amount of the contaminant that can be extracted.

6.7.10 It is appropriate to do stability testing on product contact materials which undergo various process cycles. This may be accomplished by running a number of process cycles in excess of that anticipated in production and testing extract for critical parameters such as non-volatile residue and elution cytotoxicity.

6.7.11 Use the following procedure to qualify a system containing a number of different material.

6.7.11.1 Obtain a list of all product contact materials in the system. This is necessary to assure replacement with identical materials to those you qualify.

6.7.11.2 Determine the outside parameters for the system use, for example, maximum temperature, maximum contact time, hold up volume, most effective extraction medium (typical order of increasing extraction capability is water then cell culture medium then serum solutions). lt should be recognized that cell culture and serum containing media will not be compatible with standard physicochemical tests such as Non-Volatile Residue, Oxidizable Substances, and so forth.

6.7.11.3 Take each parameter and exacerbate it to the extent practical to create safety factors. For example, if the maximum operating temperature is 37°C and the system will withstand steam, start with 80°C Purified Water and hold or let it ramp down to operating temperature. If a process run is eight hours extract for at least twice that amount of time. Use the minimum hold up volume of solution to obtain the maximum surface to volume ratio. Recirculate with Purified Water after taking a 2 liter control. Use the tests listed in Table 1 for Water Extract Solution or Organic Extract solution or Test Regimen A below for a process solution

Test Regimen A (Process solution)
pH change versus control
UV scan versus control
Metals as above versus control
Elution cytotoxicity (USP) versus control where possible

7. Test Methods

Non-Volatile Residue, USP 23, or most recent version, Physicochemical Tests—Plastics, p. 1783
Oxidizable Substances, USP 23, Purified Water, p. 1636
Inductively Coupled Plasma Analysis, ASTM D4951
Atomic Absorption Spectroscopy, American Chemical Society, Specifications, Reagent Chemicals (1993), pp. 35–42
Water Extraction, ASTM F619 – 79 (1991) or USP 23, Physicochemical Tests-Plastics, p. 1783
Elution Cytotoxicity, USP 23, <87>, Biological Reactivity Tests, In Vitro, pp. 1697–1699
UV Scan, USP 23, <197>, Spectrophotometric Identification Tests, p. 1724
Endotoxin test, USP 23, <85>, Bacterial Endotoxin Test, pp. 1696–1697
Residue on Ignition, USP 23, <281 >, Residue on Ignition, p. 1731
Identity by Infrared Spectroscopy, USP 23, < 197>, Spectrophotometric Identification Test, p. 1724
Total Organic Carbon, USP 23, Fifth Supplement, pp. 3464–3465

8. Keywords

8.1 biotechnology products; components; cytotoxicity; extractables; extraction; raw materials

APPENDIX

(Nonmandatory Information)

X1. RATIONALE

X1.1 Since potential effects of extractables will vary widely depending on the application, the end user must establish a rationale for applying the results to selection of materials. Acceptance limits for any single test specified above or for a combination may be set according to any of the following:

X1.2 Calculation from a quantitative result of the worst case amount of extractables per clinical dose followed by a toxicological assessment. Exaggerated tests are selected to ensure that a higher concentration of extractable material will be obtained over and above the level that could occur in the actual process.

X1.3 Comparison of the extractables profile from one or more tests with results from characterized reference materials known to have no adverse effects.

X1.4 Direct application of compendial limits. If a compendial test is being applied for a use other than the compendial intent, a rationale must be given.

X1.5 Correlation of test results with materials certification by supplier. It may be necessary to justify that the attribute certified is a rational ground for acceptance of the material based on its intended use.

X1.6 21 CFR 211.65a states that “Equipment shall be constructed so that surfaces that contact components, in-process materials, or drug products shall not be reactive, additive, or absorptive so as to alter the safety, identity, strength, quality, or purity of the drug product beyond the official or other established requirements.”

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