



Designation: F3274 – 21

Standard Guide for Testing and Characterization of Alginate Foam Scaffolds Used in Tissue-Engineered Medical Products (TEMPs)¹

This standard is issued under the fixed designation F3274; 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 Consistent functionality of scaffolds used in TEMPs can be made more predictable by monitoring relevant characteristics related to physical and biological properties. This guide may be used in the selection of suitable test methods of dried ionically gelled alginate foam scaffolds that may be a component of a medical device or considered a medical device itself.

1.2 This guide provides information relevant for the physical testing of alginate foam scaffolds such as mechanical properties, hydration properties, pore structure, and scaffold degradation. In addition, issues related to biological properties such as elemental impurities, bacterial bioburden, bacterial endotoxins, sterility, and biocompatibility are outlined.

1.3 This guide is intended to be used in conjunction with appropriate characterization and evaluation of any raw or starting materials utilized for fabrication of the alginate foam, such as described in Guides [F2027](#) and [F2064](#).

1.4 This guide addresses alginate foam scaffolds with and without bioactive agents or biological activity. This guide does not address the characterization or release profiles of any biomolecules, cells, drugs, or bioactive agents that are used in combination with the scaffold.

1.5 Only SI units are used in this standard.

1.6 *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, health, and environmental practices and determine the applicability of regulatory limitations prior to use.*

1.7 *This international standard was developed in accordance with internationally recognized principles on standardization established in the Decision on Principles for the Development of International Standards, Guides and Recommendations issued by the World Trade Organization Technical Barriers to Trade (TBT) Committee.*

2. Referenced Documents

2.1 ASTM Standards:²

- [D638](#) Test Method for Tensile Properties of Plastics
- [D1621](#) Test Method for Compressive Properties of Rigid Cellular Plastics
- [F748](#) Practice for Selecting Generic Biological Test Methods for Materials and Devices
- [F1635](#) Test Method for *in vitro* Degradation Testing of Hydrolytically Degradable Polymer Resins and Fabricated Forms for Surgical Implants
- [F2025](#) Practice for Gravimetric Measurement of Polymeric Components for Wear Assessment
- [F2027](#) Guide for Characterization and Testing of Raw or Starting Materials for Tissue-Engineered Medical Products
- [F2064](#) Guide for Characterization and Testing of Alginates as Starting Materials Intended for Use in Biomedical and Tissue Engineered Medical Product Applications
- [F2150](#) Guide for Characterization and Testing of Biomaterial Scaffolds Used in Regenerative Medicine and Tissue-Engineered Medical Products
- [F2312](#) Terminology Relating to Tissue Engineered Medical Products
- [F2450](#) Guide for Assessing Microstructure of Polymeric Scaffolds for Use in Tissue-Engineered Medical Products
- [F2739](#) Guide for Quantifying Cell Viability and Related Attributes within Biomaterial Scaffolds

2.2 ISO Standards:³

- [ISO 527](#) Plastics—Determination of tensile properties—Part 1: General principles
- [ISO 10993](#) Biological evaluation of medical devices
- [ISO 11135](#) Sterilization of health-care products—Ethylene oxide—Requirements for the development, validation and routine control of a sterilization process for medical devices

¹ This guide is under the jurisdiction of ASTM Committee [F04](#) on Medical and Surgical Materials and Devices and is the direct responsibility of Subcommittee [F04.42](#) on Biomaterials and Biomolecules for TEMPs.

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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 Available from American National Standards Institute (ANSI), 25 W. 43rd St., 4th Floor, New York, NY 10036, <http://www.ansi.org>.

2.3 Other Referenced Document:

AAMI ST72 Bacterial endotoxins—Test methods, routine monitoring, and alternatives to batch testing⁴

3. Terminology

3.1 Definitions:

3.1.1 *foam, n*—a solid structure that contains a large number of inherent or induced channels and open spaces (that is, pores; see Terminology F2312).

4. Summary of Guide

4.1 Alginate foam scaffolds are being used in the formulation of tissue-engineered medical products. Such scaffolds are often intended to be implanted in animals and humans. Factors such as foam integrity, strength, adhesion, biocompatibility, pore structure, and degradation will be critical for scaffold performance.

4.2 Several methods are available to produce alginate foam scaffolds, including porogen leaching, gas-foaming, freeze-drying, and phase-separation techniques. For example, pores in alginate foam may be introduced by the incorporation of air into a liquid alginate solution (wet foam) that is subsequently dried (dry foam), or pores in alginate foam may be introduced by the removal of ice crystals from a frozen solution of alginate through the use of freeze-drying techniques.

4.3 This guide summarizes physical, mechanical, and biological properties that are important for use of alginate foam scaffolds in TEMP.

5. Significance and Use

5.1 This guide is a guideline for the characterizing and testing of alginate foam scaffolds used in tissue-engineered medical products. Alginate foam scaffolds can be used in a number of tissue engineering and regenerative medicine applications such as anti-adhesion, internal wound healing, and guided tissue regeneration. In addition, alginate foam can be used as a matrix for cell-based cell therapies and for the release of bioactive agents.

6. Physical Properties

6.1 Physical properties of TEMP in the format of an ionically gelled alginate foam can be tuned and optimized to meet the requirements for its intended use. The major controllable characteristics are related to mechanical properties, hydration properties, pore structure, and degradation and will be presented in this section together with suggestions for test methods. The foam can be manufactured to be cut to size or in a size appropriate for surgical implantation. The dimensions and mechanical properties should allow for easy implantation and positioning and have an appropriate degradation profile. There may be additional relevant characterization procedures not covered by this guide that are specific to the designated end use of the foam.

6.2 Mechanical Properties:

6.2.1 Controlled mechanical properties of a foam scaffold are crucial for most applications. The foam must withstand forces exerted during insertion related to the selected method without rupture. When using a trochar, the foams typically must tolerate higher tensile stress and strain compared to foams implanted during open surgery.

6.2.2 *Tensile Properties*—Tensile properties of dry foams and rehydrated foams can be characterized as specified in Test Method D638 and ISO 527. The dumbbell-shaped test specimens are recommended to reduce the likelihood of rupture at the grips/clamps. Determination of tensile force at break (N), stress at break (Pa), strain at break (% elongation), and elasticity (Pa) will be relevant measures for these foams. If the intended use of the implanted foam is to provide mechanical support of a tissue or device, the foam must be hydrated before characterization in a model physiological solution. The mechanical properties of ionically gelled alginate structures are influenced by ions in the surrounding solution. To ensure relevant measures, it is important that the solution contains the biologically relevant concentration of non-gelling monovalent ions such as sodium ions (Na^+), gelling calcium ions (Ca^{2+}), and calcium-chelating phosphate ions (PO_4^{3-}) of approximately 154 mM, 1.3 mM, and 0.8 mM, respectively. Beware that simulated body fluids (SBF), cell culture media, phosphate buffered salt solutions (PBS), and water vary considerably in composition. During a defined time period the foam shall be added to a known amount of liquid versus volume or weight, or alternatively soaked in excess amounts. The selected strain rate may influence the results when characterizing these viscoelastic materials. Also monitor the temperature of the solution and environment.

6.2.3 *Compressive Properties*—The bulk elasticity of dry and rehydrated foams can be determined as specified in Test Method D1621. This may be relevant when aiming to match the compliance of the tissue at the implantation site. This elasticity will, however, not be comparable with, for example, local elasticities sensed by infiltrating cells.

6.2.4 *Suture Pull-Out Strength*—There are currently no recognized standard methods for determination of suture pull-out strength. Development of a method is recommended based on the specific application related to suture pattern, type, and depth, and expected forces exerted at the implantation site.

6.3 Hydration Properties:

6.3.1 Different aspects of the hydration properties of alginate foams can be considered and are presented below. The hydration properties depend on the swelling properties of the alginate in the pore wall and capillary bound liquid filling the pores of the foam. The swelling properties of alginate are dependent on the concentration of ions (as discussed in 6.2.2), in addition to pH, time, and temperature which should be monitored.

6.3.2 *Hydration Rate*—The interconnectivity of the pores of the foam is the most important characteristic that controls the hydration rate of the foam. Fast hydration rates are typically required for foams used when the aim is to absorb and retain body fluids such as blood during surgery. Slower hydration rates may, for example, be preferred for foams used as delivery vehicles for sustained release of bioactive agents. Different

⁴ AAMI ST72 available at <https://standards.aami.org/higherlogic/ws/public/download/12530/AAMI%20CDV1%20ST72%20public%20review%20draft.pdf>.