



Standard Test Method for Use of a Centrifugation Method to Quantify/Study Cell- Material Adhesive Interactions¹

This standard is issued under the fixed designation F3088; 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 test method covers a centrifugation cell adhesion assay that can be used to detect changes in adhesive characteristics of cells with passage or treatments. This approach measures the force required to detach cells from a substrate. Adhesion, among many variables, may vary due to changes in the phenotype of the cells.

1.2 This test method does not cover methods to verify the uniformity of coating of surfaces, nor does it cover methods for characterizing surfaces.

1.3 The cells may include adult, progenitor or stem cells from any species. The types of cells may include chondrocytes, fibroblasts, osteoblast, islet cells, or other relevant adherent cell types.

1.4 This test method does not cover methods for isolating or harvesting of cells. This test method does not cover test methods to quantitate changes in gene expression, or changes in biomarker type or concentration, as identified by immunostaining. Nor does this test method cover quantitative image analysis techniques.

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

[F2603 Guide for Interpreting Images of Polymeric Tissue Scaffolds](#)

[F2664 Guide for Assessing the Attachment of Cells to Biomaterial Surfaces by Physical Methods](#)

¹ This test method is under the jurisdiction of ASTM Committee F04 on Medical and Surgical Materials and Devices and is the direct responsibility of Subcommittee F04.43 on Cells and Tissue Engineered Constructs 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.

[F2739 Guide for Quantitating Cell Viability Within Biomaterial Scaffolds](#)

[F2944 Test Method for Automated Colony Forming Unit \(CFU\) Assays—Image Acquisition and Analysis Method for Enumerating and Characterizing Cells and Colonies in Culture](#)

3. Summary of Test Method

3.1 *Centrifugation Cell Assay on Cell Populations*—A conventional centrifuge can be used to apply a normal or shear force to cells depending on the orientation of the cells with respect to the centrifugal force (**1-3**).³ The centrifugal force that the cells are subject to can be calculated according to the following formula assuming that the plates are 90° (normal) to the spindle of the centrifuge.

$$F_D = (p_{\text{cell}} - p_{\text{medium}})V_{\text{cell}}\text{RCF} \quad (1)$$

where:

F_D = detachment force applied per cell,

V_{cell} = cell volume,

p_{cell} = density of the cell,

p_{medium} = the density of the surrounding medium, and

RCF = Relative Centrifugal Forces = $r\omega^2$, where r = centrifugation radius and ω = centrifugation speed.

See [X1.6](#) for example of a calculation of “detachment force per cell.”

3.1.1 Such tests are easy to conduct and the results represent a population average. The method can be performed in a moderately outfitted cell biology lab with an inverted microscope or a fluorescence microplate reader and a centrifuge with a microtiter plate holder. Three factors need to be considered when using this methodology: the potential influences of forces applied during the period of spin up time, the maximum spin speed, and the test duration. The method is only able to correlate with cell detachment with the maximum force applied after the centrifuge has reached its set spin speed. It should also be considered that the maximum force that can be applied is limited by the centrifuge, sample configuration (that is, configuration and type of multiwell plate) and time of centrifugation. In some instances, the cell adhesion strength will be

³ The boldface numbers in parentheses refer to the list of references at the end of this standard.