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Standard Specification for Biological Properties of Industry Reference Materials (IRM)[1]

This standard is issued under the fixed designation D7850; 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 specification covers the biological quality specifications or requirements, or both, for Industry Reference Materials (IRMs) as cited in Practice D4678 and other standards.

1.2 IRMs, as evaluated and referenced in Practice D4678, are vitally important to conduct product, specification, and development testing in the rubber and carbon black industries.

1.3 Before a new lot of material can be accepted as an IRM, it must comply with the specifications prescribed in this specification. However, these specifications are only part of the requirements. Other requirements as given in Practice D4678 shall be met before a candidate material can be formally accepted as a biological IRM.

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

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]

D4678 Practice for Rubber—Preparation, Testing, Acceptance, Documentation, and Use of Reference Materials

D5712 Test Method for Analysis of Aqueous Extractable Protein in Natural Rubber and Its Products Using the Modified Lowry Method

D6499 Test Method for The Immunological Measurement of Antigenic Protein in Natural Rubber and its Products

D7427 Test Method for Immunological Measurement of Four Principal Allergenic Proteins (Hev b 1, 3, 5 and 6.02) in Natural Rubber and Its Products Derived from Latex

3. Significance and Use

3.1 IRMs are vitally important in product and specification testing, in research and development work, in technical service work, and in quality control operations in the rubber and carbon black industries. They are especially valuable for referee purposes. Many ASTM rubber standards for the evaluation of natural or synthetic rubber require the use of specific IRMs for better laboratory repeatability and reproducibility.

3.2 New material lots that have been selected as candidates for IRM approval shall conform to the appropriate specifications given in this standard and meet requirements given in Practice D4678 before the lots may be accepted as IRMs.

3.3 The biological IRM specifications shown will ensure some consistency in IRM properties from one lot to the next. However, the specifications cannot ensure exact inter-lot consistency.

4. Specifications

4.1 The following are specifications for the Biological Industry Reference Materials:

4.1.1 *Specification for IRM 913*[3]—Ammoniated Latex Antigenic Protein:

4.1.1.1 Material description: Sealed vial for hypodermic extraction—403 test quantity.

4.1.1.2 Specifications are given in Table 1.

4.1.2 *Specification for IRM 914*[3]—Rabbit Anti AL Antisera:

4.1.2.1 Material description: Screw-cap vials with O-Ring seals—301 test quantity.

4.1.2.2 Specifications are given in Table 2.

4.1.3 *Specification for IRM 915*[4]—Mouse monoclonal antibody Hb1-5:

4.1.3.1 Material description: Vials with frozen material—up to 13 400 test quantity.

[1] This test method is under the jurisdiction of ASTM Committee D11 on Rubber and is the direct responsibility of Subcommittee D11.40 on Consumer Rubber Products.

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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] An approved lot has been reserved and is available from Akron Rubber Development Lab Inc., 2887 Gilchrist Rd., Akron, OH 44305, website: www.ardl-.com.

[4] An approved lot, IRM 915 to 924, has been reserved and is available from Scripps Laboratories, 6838 Flanders Drive, San Diego, CA 92121, website: www.scrippslabs.com.

TABLE 1 Specification for IRM 913—Ammoniated Latex Antigenic Protein

Property	ASTM Designation	Limit/Target
Source	D6499	Ammoniated NR Latex (high and low)
Protein Amount	D5712	Stock solution should be adjusted to 1 mg protein per mL
Allergen Concentration	D7427	Allergen Concentration of four allergens reported per ug total protein as determined by D5712 with and without background subtraction
SDS-PAGE	D6499	Full distribution of NR proteins (between 5 kDa and 200 kDa)
Western blot profile	D6499	Full distribution of NR antigens (between 5 kDa and 200 kDa)
Inhibition ELISA	D6499	Adjust antigen content based on D6499 determination.

TABLE 2 Specification for IRM 914—Rabbit Anti-AL Antisera

Property	ASTM Designation	Limit/Target
Animal	D6499	Rabbits
Antigen	D6499	Ammoniated NR reference protein
Adjuvant Titer	D6499	Freund's complete and incomplete Minimum 1/8000
Western blot profile[A]	D6499	Reactivity with entire spectrum of SDS-PAGE[B] visible AL protein
Inhibition ELISA reactivity[C]	D6499	Comparative analysis of extracts from a minimum of 5 NR products

[A] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[B] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[C] ELISA represents the enzyme linked immuno-sorbant assay.

4.1.3.2 Specifications are given in Table 3.

4.1.4 *Specification for IRM 916*[4]—Mouse monoclonal antibody Hb1-1:

4.1.4.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.4.2 Specifications are given in Table 4.

4.1.5 *Specification for IRM 917*[4]—Hev b 1 Reference Protein:

4.1.5.1 Material description: Vials with lyophilized material—up to 1000 test quantity.

4.1.5.2 Specifications are given in Table 5.

4.1.6 *Specification for IRM 918*[4]—Mouse monoclonal antibody Hb3-5:

4.1.6.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.6.2 Specifications are given in Table 6.

4.1.7 *Specification for IRM 919*[4]—Mouse monoclonal antibody Hb3-10:

4.1.7.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.7.2 Specifications are given in Table 7.

4.1.8 *Specification for IRM 920*[4]—Hev b 3 Reference Protein:

4.1.8.1 Material description: Vials with lyophilized material—up to 1000 test quantity.

4.1.8.2 Specifications are given in Table 8.

4.1.9 *Specification for IRM 921*[4]—Mouse monoclonal antibody Hb5-4:

4.1.9.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.9.2 Specifications are given in Table 9.

4.1.10 *Specification for IRM 922*[4]—Mouse monoclonal antibody Hb5-11:

4.1.10.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.10.2 Specifications are given in Table 10.

4.1.11 *Specification for IRM 923*[4]—Hev b 5 Reference Protein:

4.1.11.1 Material description: Vials with lyophilized material—up to 1000 test quantity.

4.1.11.2 Specifications are given in Table 11.

4.1.12 *Specification for IRM 924*[4]—Mouse monoclonal antibody Hb6.02-2:

TABLE 3 Specification for IRM 915—Mouse monoclonal antibody Hb1-5

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb1 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/4000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 1 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 4 Specification for IRM 916—Mouse monoclonal antibody Hb1-1

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb1 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 1 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 5 Specification for IRM 917—Hev b 1 Reference Protein

Property	ASTM Designation	Limit/Target
Source	D7427	r MBP-Hevb1
Protein Amount	D7427	Stock solution should be adjusted to 1000 ng/mL
RP chromatography profile	D7427	One major peak detectable
Western blot profile[A]	D7427	One band detectable at molecular weight approximately 10 to 15 kDa
Capture ELISA reactivity[B]	D7427	Adjust antigen content based on D7427 determination

[A] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[B] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 6 Specification for IRM 918—Mouse monoclonal antibody Hb3-5

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb3 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/4000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 3 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 7 Specification for IRM 919—Mouse monoclonal antibody Hb3-10

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb3 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 3 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

4.1.12.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.12.2 Specifications are given in Table 12.

4.1.13 *Specification for IRM 925—Mouse monoclonal antibody Hb6.02-N3:*

TABLE 8 Specification for IRM 920—Hev b 3 Reference Protein

Property	ASTM Designation	Limit/Target
Source	D7427	Field Latex (FL)[A]
Protein Amount	D7427	Stock solution should be adjusted to 1000 ng/mL
RP chromatography profile	D7427	One major peak detectable
Western blot profile[B]	D7427	One band detectable at molecular weight approximately 20 to 25 kDa
Capture ELISA reactivity[C]	D7427	Adjust antigen content based on D7427 determination

[A] Natural Rubber Latex designated as Field Latex.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 9 Specification for IRM 921—Mouse monoclonal antibody Hb5-4

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb5 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Capture ELISA reactivity[B]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 10 Specification for IRM 922—Mouse monoclonal antibody Hb5-11

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	rMBP-Hevb5 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Capture ELISA reactivity[B]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] ELISA represents the enzyme linked immuno-sorbant assay.

4.1.13.1 Material description: Vials with frozen material—up to 13 400 test quantity.

4.1.13.2 Specifications are given in Table 13.

4.1.14 *Specification for IRM 926—Hev b 6.02 Reference Protein:*

4.1.14.1 Material description: Vials with lyophilized material—up to 1000 test quantity.

4.1.14.2 Specifications are given in Table 14.

5. Keywords

5.1 allergens; biological properties; immunoassays; industry reference materials; IRM; monoclonal antibody; NR latexrabbit anti AL antisera

TABLE 11 Specification for IRM 923—Hev b 5 Reference Protein

Property	ASTM Designation	Limit/Target
Source	D7427	Natural Rubber Latex C-serum
Protein Amount	D7427	Stock solution should be adjusted to 100 ng/mL
RP chromatography profile	D7427	One major peak detectable
Mass spectrometry profile	D7427	One major peak detectable at molecular weight approximately 16 kDa
Capture ELISA reactivity[A]	D7427	Adjust antigen content based on D7427 determination

[A] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 12 Specification for IRM 924—Mouse monoclonal antibody Hb6.02-2

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	Recombinant avidin-Hevb6.02 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 6.02 or Hev b 6.01 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 13 Specification for IRM 925—Mouse monoclonal antibody Hb6.02-N3

Property	ASTM Designation	Limit/Target
Animal	D7427	Mouse
Antigen	D7427	Recombinant avidin-Hevb6.02 protein
Adjuvant	D7427	Freund's complete and incomplete
Titer	D7427	Minimum 1/3000
SDS PAGE Coomassie stain profile[A]	D7427	Two bands detectable
Western blot profile[B]	D7427	Reactivity with FL[C] and Hev b 6.02 or Hev b 6.01 protein
Capture ELISA reactivity[D]	D7427	Comparative analysis of extracts from a minimum of 5 NR products

[A] SDS-PAGE represents sodium dodecylsulfate polyacrylamide gel electrophoresis.
[B] Western blot represents the transfer of protein from SDS-Page to a membrane and subsequent antigen analysis using NR latex anti-sera.
[C] Natural Rubber Latex designated as Field Latex.
[D] ELISA represents the enzyme linked immuno-sorbant assay.

TABLE 14 Specification for IRM 926—Hev b 6.02 Reference Protein

Property	ASTM Designation	Limit/Target
Source	D7427	Natural Rubber Latex
Protein Amount	D7427	Stock solution should be adjusted to 200 ng/mL
RP chromatography profile	D7427	One major peak detectable
Mass spectrometry profile	D7427	One major peak detectable at molecular weight approximately 4 to 5 kDa
Capture ELISA reactivity[A]	D7427	Adjust antigen content based on D7427 determination

[A] ELISA represents the enzyme linked immuno-sorbant assay.