Designation: D 6500 – 00

Standard Test Method for Diameter of Wool and Other Animal Fibers Using an Optical Fiber Diameter Analyser[1]

This standard is issued under the fixed designation D 6500; 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.

INTRODUCTION

Instruments based on image analysis have been designed to reduce the level of operator skill required and to speed up the measurement process while concurrently maintaining acceptable levels of precision and accuracy. An additional advantage of image analysis instruments is the ability of the operator to see the measurement points and to audit the process, if required, though this is not carried out during routine measurement. As with projection microscope measurements, and the Sirolan-Laserscan, the Optical Fiber Diameter Analyser (OFDA) system covered by this test method provides a count of readings grouped into diameter classes. Because the fiber snippets are measured automatically by an optical and image processing system, controls are provided in the image processing software to minimize the inclusion of multiple measurements on the same fiber and false diameter readings that arise from non-fiber material.

1. Scope

1.1 This test method covers a procedure that uses an Optical Fiber Diameter Analyser (OFDA) for the determination of the average fiber diameter and the fiber diameter variation in wool and other animal fibers in their various forms.

NOTE 1—This test method may also be applied to other fibers having a round cross section such as some polyamides, polyesters, and glass; it may also be applied to a limited number of polyacrylics and regenerated cellulose-type fibers.

NOTE 2—In subsequent sections of this test method, the term "wool" also signifies other animal fibers where applicable.

NOTE 3—For fineness specifications of wool, wool top, mohair, mohair top, alpaca, and cashmere, refer to Specifications D 3991, D 3992, D 2252, and Test Method D 2816, respectively.

1.2 The OFDA reports average fiber diameter and standard deviation of fiber diameter in micrometer units (μm). The coefficient of variation of fiber diameter is reported as a percentage.

1.3 *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:*

D 123 Terminology Relating to Textiles[2]

D 584 Test Method for Wool Content of Raw Wool—Laboratory Scale[2]

D 1060 Practice for Core Sampling of Raw Wool in Packages for Determination of Percentage of Clean Wool Fiber Present[2]

D 1776 Practice for Conditioning Textiles for Testing[2]

D 2130 Test Method for Diameter of Wool and Other Animal Fibers by Microprojection[2]

D 2252 Specification for Fineness of Types of Alpaca[2]

D 2816 Test Method for Cashmere Coarse–Hair Content in Cashmere[2]

D 3510 Test Method for Diameter of Wool and Other Animal Fibers by Image Analyzer[3]

D 3991 Specifications for Fineness of Wool or Mohair and Assignment of Grade[4]

D 3992 Specifications for Fineness of Wool Top or Mohair Top and Assignment of Grade[4]

D 4845 Terminology Relating to Wool[4]

2.2 *Federal Standards:*

Official Standards of the United States for Grades of Wool, Section 31.0[5]

Measurement Method for Determining Grade of Wool, Section 31.204[5]

Official Standards of the United States for Grades of Wool Top, Section 31.1[6]

Measurement Method for Determining Grade of Wool Top, Section 31.301[6]

[1] This test method is under the jurisdiction of ASTM Committee D13 on Textiles, and is the direct responsibility of Subcommittee D13.13 on Wool and Wool Felt.

Current edition approved March 10, 2000. Published June 2000.

[2] *Annual Book of ASTM Standards*, Vol 07.01.

[3] Discontinued. See 1986 *Annual Book of ASTM Standards*, Vol 07.02.

[4] *Annual Book of ASTM Standards*, Vol 07.02.

[5] *Federal Register*, Vol 30, No. 161, August 20, 1965, pp. 10829-10833.

[6] *Federal Register*, Vol 33, No. 248, December 21, 1968, pp. 19073-19076.

USDA Grade Standards for Grease Mohair and Mohair Top[7]

2.3 *IWTO Standards:*[8]

IWTO-8-66 Method of Determining Wool Fiber Diameter by the Projection Microscope

IWTO-12-93 Measurement of the Mean and Distribution of Fibre Diameter Using a Sirolan-Laserscan Fibre Diameter Analyser

IWTO-19-98 Determination of Wool Base and Vegetable Matter Base of Core Samples of Raw Wool

IWTO-47-98 Measurement of the Mean and Distribution of Fibre Diameter of Wool Using an Optical Fibre Diameter Analyser (OFDA)

3. Terminology

3.1 *Definitions*—For definitions of "wool" and other textile terms used in this test method, refer to Terminologies D 4845 and D 123.

3.1.1 *average fiber diameter*, *n*—the arithmetic mean width of a group of fibers.

3.1.1.1 *Discussion*—In wool and other animal fibers, all animal fibers, regardless of species, can be measured using the OFDA to determine average fiber diameter.

3.1.2 *grade*, *n—in wool and mohair*, a numerical designation used in classification of fibers in their raw, semi-processed, and processed forms based on average fiber diameter and variation of fiber diameter.

3.1.3 *snippet*, *n*—a wool or other animal fiber that has been cut to a specified length.

4. Summary of Test Method

4.1 This test method describes procedures for sampling wool in various physical forms, the reduction of the sample to small test specimens, and measurement of the diameter of a number of fibers from the test specimens using the OFDA. Snippets comprising a test specimen cut from the various forms of wool are cleaned where required, conditioned, and spread uniformly over the surface of a microscope slide. A cover slide is placed over the specimen and the slide placed on a microscope stage, that is moved under computer control. The slide is stepped through the field of view of a low-power microscope objective. At each step, the video system is instructed to capture and analyze a fiber image frame. Each diameter measurement is allocated to a diameter class and, at the completion of the slide, the class contents are statistically analysed to produce the mean and standard deviation of the fiber diameter for the specimen. Full distribution data are also available in the form of a printed histogram.

5. Significance and Use

5.1 This test method specifies sampling and testing procedures for the measurement of average fiber diameter and variation in diameter of animal fibers.

5.2 This test method is considered satisfactory for acceptance testing of commercial shipments of wool and other animal fibers in raw and sliver form because current estimates of between-laboratory precision are acceptable. In cases of disagreement arising from differences in values reported by two or more laboratories when using this test method for acceptance testing, the statistical bias, if any, between the laboratories should be determined with each comparison being based on the testing of specimens randomly drawn from one sample of material of the type being evaluated. Test Method D 2130 shall be used as a referee test method.

5.3 This test method may be used for determining compliance with average fiber diameter and diameter variation to assign grades when determining conformance of shipments to material specifications given in Specifications D 2252, D 3991, and D 3992, and Test Method D 2816.

5.4 The procedures for determining mean fiber diameter and standard deviation of fiber diameter provided in this test method and in IWTO Method 47-98 are in essential agreement.

6. Apparatus, Materials, and Reagents

6.1 *Optical Fiber Diameter Analyser*[9], consisting of a transmission light microscope, fitted with a stage (motor-driven and controlled by a computer), stroboscopic illumination that is synchronised with the stage movement, and a CCD camera; an image acquisition and analysis hardware system; a means for controlling the interaction between the camera, stage motors and illumination unit; a data acquisition and processing computer, with optionally, control and reporting software; and, a video monitor, capable of displaying each image frame in real time, for audit purposes. See Fig. 1.

6.2 *Glass Microscope Slides*[9], of float glass, sufficiently robust to withstand repeated handling having dimensions 70 by 70 by 2 mm. Two identical slides are taped together so that one supports the fiber samples with the other serving as a cover slide. Slides that are scratched on their inside surfaces are unsuitable as they may lend to erroneous measurements.

6.3 *Cleaning and Conditioning Apparatus and Facilities*, suitable for cleaning and drying the subsamples in accordance with Test Method D 584 and conditioning them as described in Practice D 1776.

6.4 *Apparatus for Snippet Preparation*, having either two parallel cutting edges between 1.8 and 2.0 mm apart (for example, guillotine[9,10] or snippeter[11]; see Figs. 2 and 3), or a cutting diameter of between 1.9 and 2.1 mm (for example, minicore[9,12]; see Fig. 4). A minicore consists of a cylindrical sample holder, designed for relatively large samples, in which a sample is manually packed, then compressed, and a coring head is driven pneumatically into the sample. The sample is compacted by a spring-loaded platen. Six or more minicore tubes with 2-mm diameter tips pass through perforations in the

[7] *Federal Register*, Vol 36, No. 129, July 3, 1971, pp. 12681-12658.

[8] Available from the International Wool Textile Organization, International Wool Secretariat, Commercial Development Department, Valley Drive, Ilkley, Yorkshire LS29, 8PB, England, UK.

[9] Available from BSC Electronics Pty, Ltd., 1A Thurso Rd., Myaree, Western Australia, 6154.

[10] Available from Symtech Systems and Technology, I-85 and Bryant Rd., PO Box 2627, Spartanburg, SC 29304.

[11] Available from CSIRO, Division of Wool Technology, PO Box 21, Belmont, VIC 3216, Australia.

[12] Available from the South African Wool Testing Bureau, Gomery Ave., Summerstrand, PO Box 1867, Port Elizabeth 6000, South Africa.

FIG. 1 The Optical Fiber Diameter Analyser

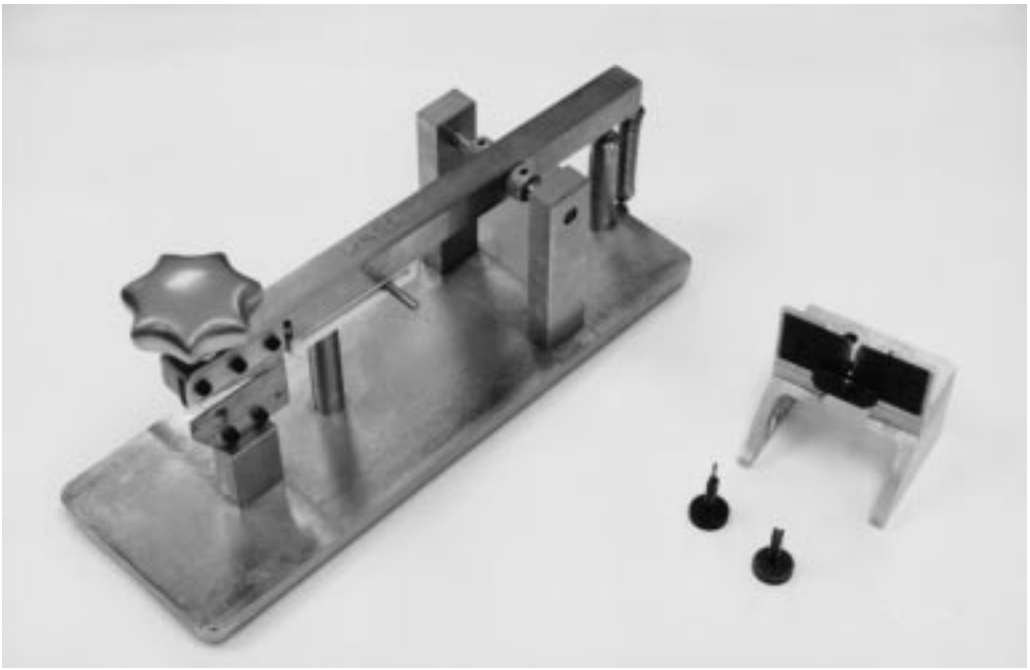

FIG. 2 Guillotine and Snippeter

platen when the force supplied by the pneumatic cylinder exceeds the force from the preloaded spring. At the end of the stroke, the cutting tips have penetrated to within 0.5 mm of the base of the sample holder. The sample collected by the minicore tubes is automatically expelled into a collection device upon retraction of the coring head.

6.5 *Heavy-Duty Sectioning Device*[13], comprised of a metal plate with slot and compressing key and equipped with a propulsion mechanism by which the fiber bundle may be extruded for sectioning. The instrument is designed to hold a sliver or top or equivalent bulk of fibers, yarn, or fabric (see Fig. 1 of Test Method D 2130). Alternatively, this instrument can be used to generate the snippets.

[13] Available from MICO Instruments, 1944 Main St., PO Box 451, Marshfield Hills, MA 02051-0451.

FIG. 3 Guillotine

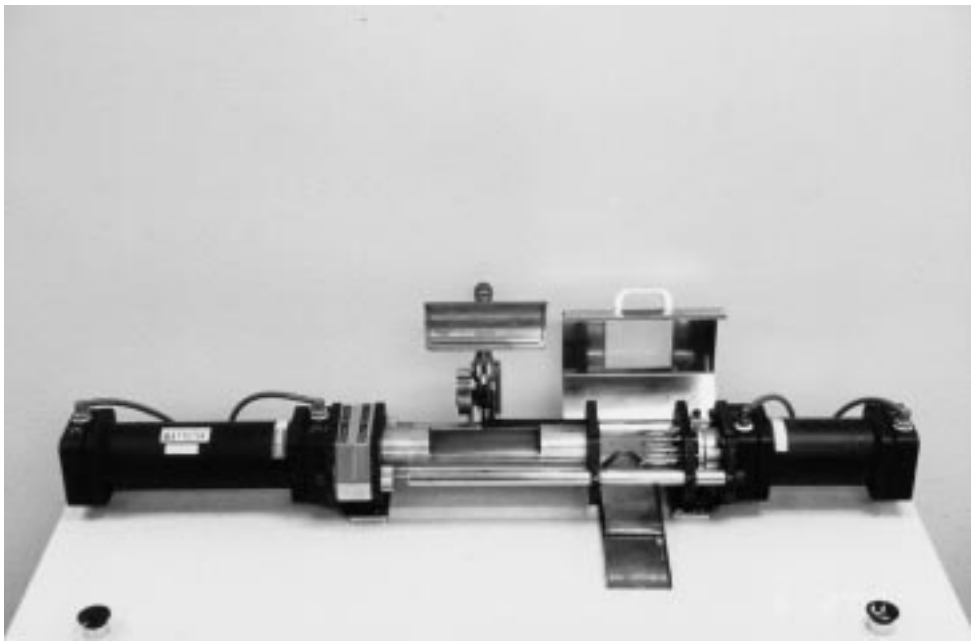

FIG. 4 Minicoring Device

6.6 *Safety Razor Blades*, single-edge or double-edge (if used with blade holder).

6.7 *Slide Preparer*[9] capable of uniformly spreading a portion of the cleaned, conditioned snippet sample over the surface of a clean glass slide at a predetermined, controlled density. For the OFDA, the optimum obscured areas, that is, the ratio of fiber to the total field area, is between 15 and 25 %. There are different versions of slide preparers (spreaders) available and it must be ensured that the same slide preparer is used for both calibration and routine OFDA measurements. See Fig. 5.

6.8 *Box for Compressing Loose Fibers*, 300 by 150 by 375 mm deep, inside dimensions, equipped with a floating top that has 16 randomly spaced holes 20 mm in diameter over its area. The sample may be firmly compressed by applying pressure on the top. The top is held in place by two rods extending through holes in the side of the box and over the top. The coring tube is thrust through the holes in the top to sample the wool.

6.9 *Pressure Coring Tube*, 13-mm inside-diameter metal tube, approximately 760 mm long, reamed and tapped on one end to hold a sharp 10 or 13-mm cutting tip. The tube is fitted with a "T" cross bar about 500 mm long.

6.10 *Core Extruder*, 6-mm wood dowel or aluminum rod slightly longer than the coring tube to push the sample from tube.

6.11 *Solvents*—Petroleum spirit (boiling range 40 to 70°C) and 1,1,1, trichloroethane. When the preparation method calls for the cleaning of sliver subsamples, one of these two solvents shall be used. **Warning**—Both solvents have associated hazards in terms of volatility, toxicity, and, in the case of petroleum spirit, flammability. In both cases, care should be taken in storage, handling, use, and disposal in accordance with

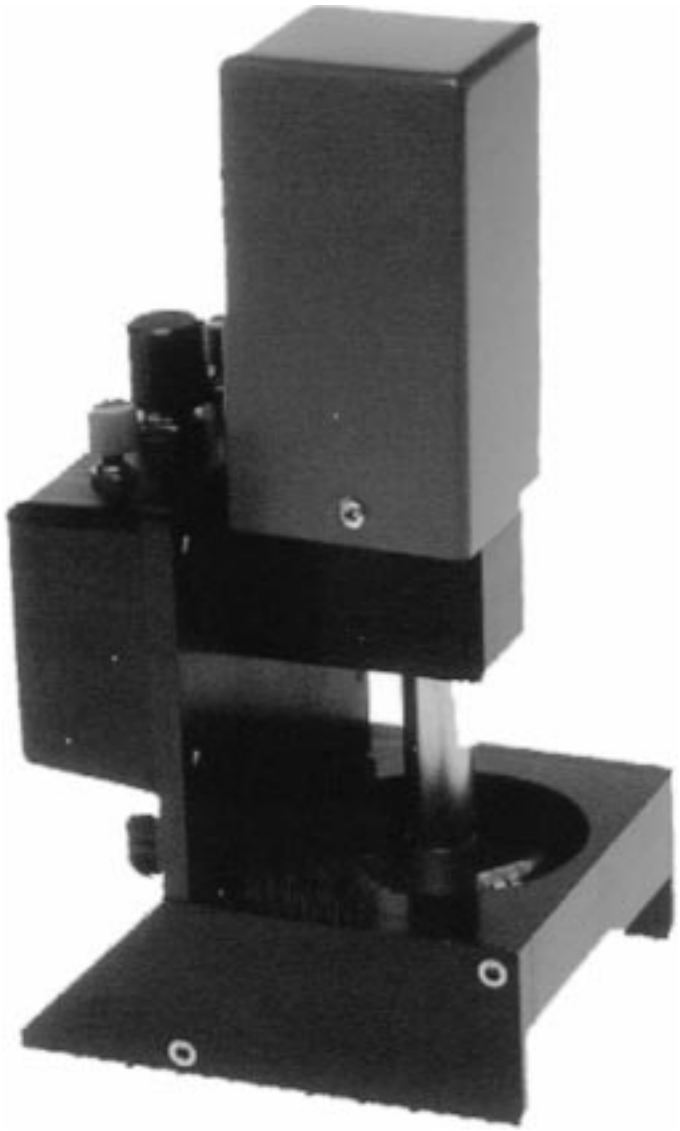

FIG. 5 Slide Spreader

the appropriate safety procedures. Refer to manufacturers' material safety data sheets (MSDS).

6.12 *Calibration Standards*—Used for instrument calibration. For wool, use current Interwoollabs IH Standard Tops[14] and for mohair, use current International Mohair Association Standard Tops[15].

7. Sampling

7.1 *Loose Fibers*—The method of obtaining a representative sample of wool differs according to circumstances. The sampling procedures and major circumstances encountered are as follows:

7.1.1 *Lots of Packaged, Grease, Pulled, or Scoured Wool*—Take core samples as directed in Practice D 1060. Clean or scour the raw wool sample as directed in Test Method D 584. If a representative portion of the scoured wool core sample resulting from the test for clean wool fiber present is available, it may be used for fiber diameter determination. If core sampling is not feasible, take at random, by hand, at least 50 handfuls of wool from not less than 10 % of the packages. The aggregate mass of the sample shall be at least 1.5 kg.

7.1.2 *Major Sort*—For packaged grease wool in fleece form for which a diameter test is needed for only the major sort of the fleece, hand sample by drawing one or more handfuls of wool from the major sort portions of at least 50 fleeces taken at random from the lot. The aggregate mass of the sample shall be at least 1.5 kg.

7.1.3 *Piles of Graded or Sorted Wool*—Sample piles of graded or sorted wool by taking from random locations in the pile at least 50 handfuls of wool, the aggregate mass of which shall be at least 1.5 kg. If the wool is in fleece form and a test is needed for only the major sort, take the sample as directed in 7.1.2.

[14] Available from Interwoollabs Secretariat, Boite 14 Rue de Luxembourg 19/21, 1040 Brussels, Belgium.

[15] Available from International Mohair Association, Mohair House, 68 The Grove, Ilkley, West Yorkshire, LS29 9PA, England, UK.

7.1.4 *Card Sliver*—Sample the wool card sliver by drawing 10 600-mm lengths at random from the lot, preferably during the carding operation.

7.1.5 *Top*—Sample the top by drawing from each 9000 kg or fraction thereof, 4 sections of sliver, each of which shall be at least 1 m in length and taken from different balls of top selected at random. Take only one ball from any one bale or carton. For broken top, take an equivalent aggregate length of sliver at random.

8. Test Samples and Test Specimens, Number and Preparation

8.1 *Test Samples (One from Each Lab Sampling Unit)*:

8.1.1 *Grease Wool, Pulled Wool, Scoured Wool*:

8.1.1.1 *Sub-Coring*—Randomly pack the core or hand sample (see 7.1.1, 7.1.2, and 7.1.3) into a suitable container (see 6.8) and compress to approximately 14 kPa by loading a weight of 667 N on the floating top. By means of a 10- or 13-mm tipped pressure coring tube, extract at least 5 cores to provide a test specimen of at least 20 g of scoured wool. Scour or otherwise clean the test specimen if it is grease wool or pulled wool as directed in Test Method D 584.

8.1.1.2 *Gridding, Core Test Residue*—If the sample comprises an adequate amount of scoured wool resulting from core testing a lot for clean wool fiber present, divide the sample into 40 portions of approximately equal size. From each portion, draw at random at least 0.5 g. Mix or blend these 40 portions to form the test specimen.

8.1.1.3 *Gridding and Machine Blending*—For samples other than those specified in 8.1.1.1 and 8.1.1.2, divide the sample into 40 portions of approximately equal size. From each portion draw at random a sufficient quantity of fiber to provide a clean test specimen of 20 g. Scour or otherwise clean the test specimen of grease or pulled wool.

8.1.2 *Card Sliver*—Strip off portions of each of the 10 600-mm lengths of sliver (see 7.1.4). Combine these portions to form a composite sliver about 600 mm in length. This constitutes the test specimen.

8.1.3 *Top*—Each of the 4 sections of sliver comprising the sample (see 7.1.5) constitutes a test specimen.

8.2 *Test Specimens*:

8.2.1 Test 1 test specimen from each bulk subsample and 2 specimens from each sliver and top subsample. Prepare approximately 25-mg test specimens by cutting enough fiber snippets to measure the diameters of at least 2000 fiber segments for each test specimen measured. Obtain snippets using a minicore (8.2.1.1) or guillotine (8.2.1.2). When required to achieve the necessary quantity of snippets, combine snippets from one sliver subsample or bulk subsample to form the test specimen.

8.2.1.1 *Minicore (Applicable to Raw Wool, Card Sliver, or Top)*—Minicore each sliver subsample or each bulk subsample, as appropriate, using cutting tips between 1.8 and 2.0 mm in diameter. If the whole sliver subsample or bulk subsample cannot fit into the minicore, divide the coring sample into approximately equal portions of a size to produce at least 2000 individual fiber measurements. Where appropriate, samples of greasy wool shall be scoured by the procedures outlined in Practice D 584 before minicoring. Snippets from tops, aqueous scoured, or carbonized wool should be solvent washed, dried, and conditioned before measuring.

8.2.1.2 *Guillotine (Applicable to Staples, Card Sliver and Top)*—Cut snippets from the subsample with a guillotine or microtome set to a length between 1.8 and 2.0 mm. Make the same number of cuts from each subsample. Do not cut snippets within 100 mm of either end of the sliver or make sequential cuts within the length of the longest fibers.

8.2.2 Remove any large pieces of vegetable matter and excessively long fibers from the test specimens. During removal of large pieces of vegetable matter and excessively long fibers, handling of the specimen must be kept to a minimum to avoid preferential separation of fibers of differing diameter.

9. Calibration of OFDA

9.1 A complete calibration and validation of the analyzer will be necessary following any of the following conditions: every 3 to 6 months, dependant upon measurement performance monitoring; after a change of Interwoollabs standard top series or a change of International Mohair Association standard top series; whenever any significant instrument hardware or software changes or adjustments are made; and, after moving the instrument.

9.2 Calibration and verification tests for the OFDA are described in Annex A1.

10. Conditioning

10.1 Condition the cleaned, dried samples for at least 4 h in the standard atmosphere for testing textiles, 65 ± 2 % relative humidity and 21 ± 1°C, as directed in Practice D 1776.

NOTE 4—Experience to date has involved only sub-sampling and test specimen preparation from conditioned samples and subsamples. Subsequently, it is only necessary to store and measure the test specimens under standard conditions. Theoretically, it seems reasonable that small amounts of snippets could be cleaned and dried such that only the test specimen would need to be conditioned. This approach has not yet been fully investigated and can not, therefore, be recommended.

11. Procedure

11.1 A single operator is sufficient for OFDA testing.

11.2 *Pre-Measurement Checks*—At the start of each measurement session, ensure that the instrument is set up according to the operating manual. Measure at least one test specimen from a fine top of known diameter and one from a coarse top of known diameter. During the pre-measurement check, check the microscope focus and adjust if necessary according to the operator manual. If at any time during measurement, the software indicates the microscope is out of focus, manually adjust the microscope back into focus. If either mean fiber diameter result varies by more than 0.3 µm from the known value, or from the value determined by at least 10 measurements carried out immediately after calibration of the instrument, check, adjust or recalibrate the instrument until satisfactory performance is obtained.

11.3 *Preparation of Subsamples*—See Fig. 6 for the procedure for preparing subsamples from greasy wool cores that should be used in conjunction with ASTM Test Method D 584. Fig. 7 is applicable to sliver subsamples.

11.4 *Preparation of Snippet Samples*—Prepare snippet samples from subsamples that have been properly cleaned and

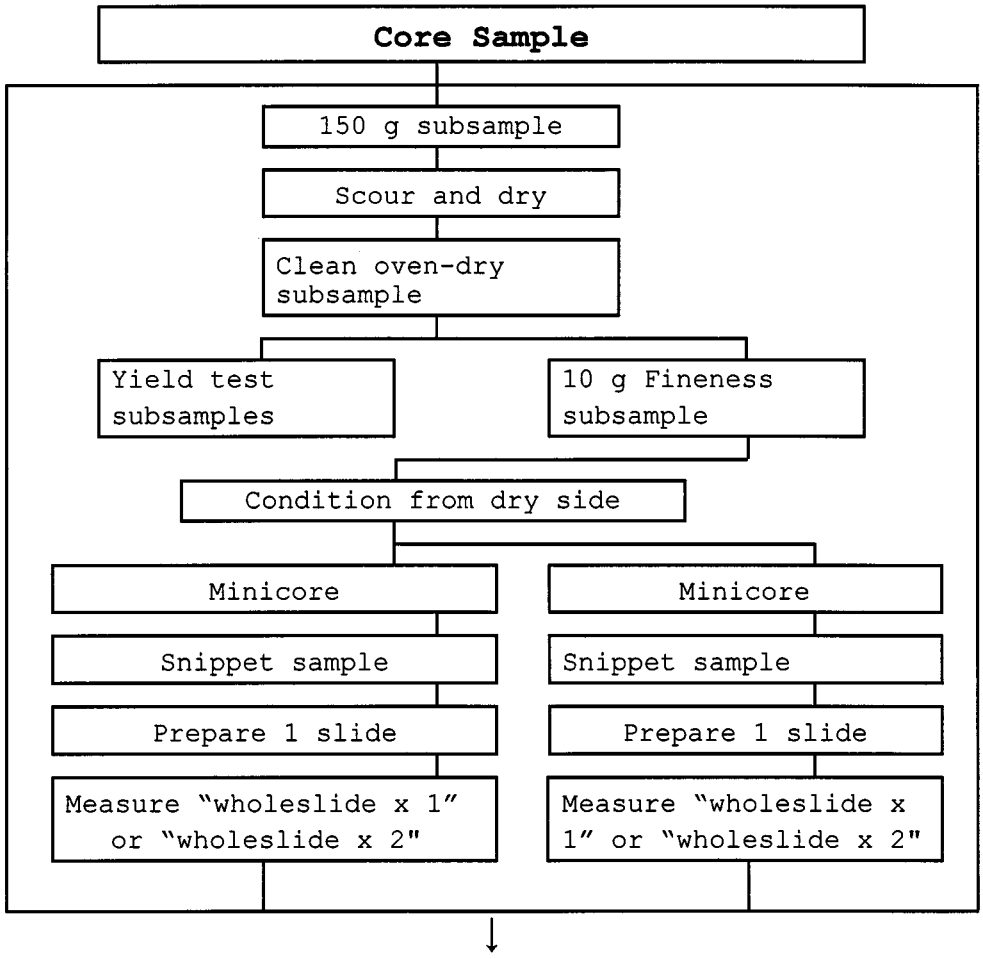

FIG. 6 Preparation of Test Specimens from Core Samples

conditioned using minicoring, guillotining, or microtoming. Obtain enough fiber snippets from each subsample to allow, on each test specimen, at least 2000 snippet measurements. Table 1 gives the average fiber diameter tolerance ranges obtained when the procedures and minimum number of acceptable measurements are made for slivers and greasy wool cores. For further details on snippet sample preparation, refer to A1.3.1.

11.5 *Preparation of the test specimens*—Refer to A1.3. Upon completion of the snippet spreading, carefully lower the clean cover slide over the fibers without disturbing the snippets and secure in place with a small piece of tape. Prepare only one slide from each snippet sample. Measure any slides that appear to be scratched on the inside surfaces without fibers, and, if readings are observed, discard the slides.

11.6 *Measurement of Fiber Diameter Distribution*—Ensure that the instrument settings under menu items "setup" and "calibration" are correct and that the optical system and stage remain clean and free of extraneous fibers. Ensure that the test specimen slide has no scratches or grease spots and place it securely in the stage clamps. Measure the whole slide using the "wholeslide× 1" or "wholeslide × 2" options to ensure that at least 2000 snippets are measured.

11.6.1 *Range Checks*—Calculate the range checks using the following:

$$\text{Range between subsamples} = \text{Studentised range}^{*}\sqrt{\sigma^2_{\text{within laboratory}}} \quad (1)$$

where the sigma squared term is the component of variance for between subsamples (for example, within laboratory) calculated from the 1995 round trials conducted under the auspices of IWTO.

11.6.1.1 Determine the range between 2 slides from the same subsample. Where this exceeds the value shown in Table 1 for the appropriate mean fiber diameter, prepare a new slide. If the range between 2 of the 3 results falls within the allowable range, discard the third result. If the range criterion cannot be satisfied, take a new snippet sample and prepare and measure 2 more slides. If the range between the 2 slides satisfies the criterion, discard the original measurements. If the range is still not satisfied, combine all 5 measurements. Calculate the mean fiber diameter for the subsample.

11.6.1.2 When all the subsamples required (either greasy cores or sliver) have been measured and the mean fiber

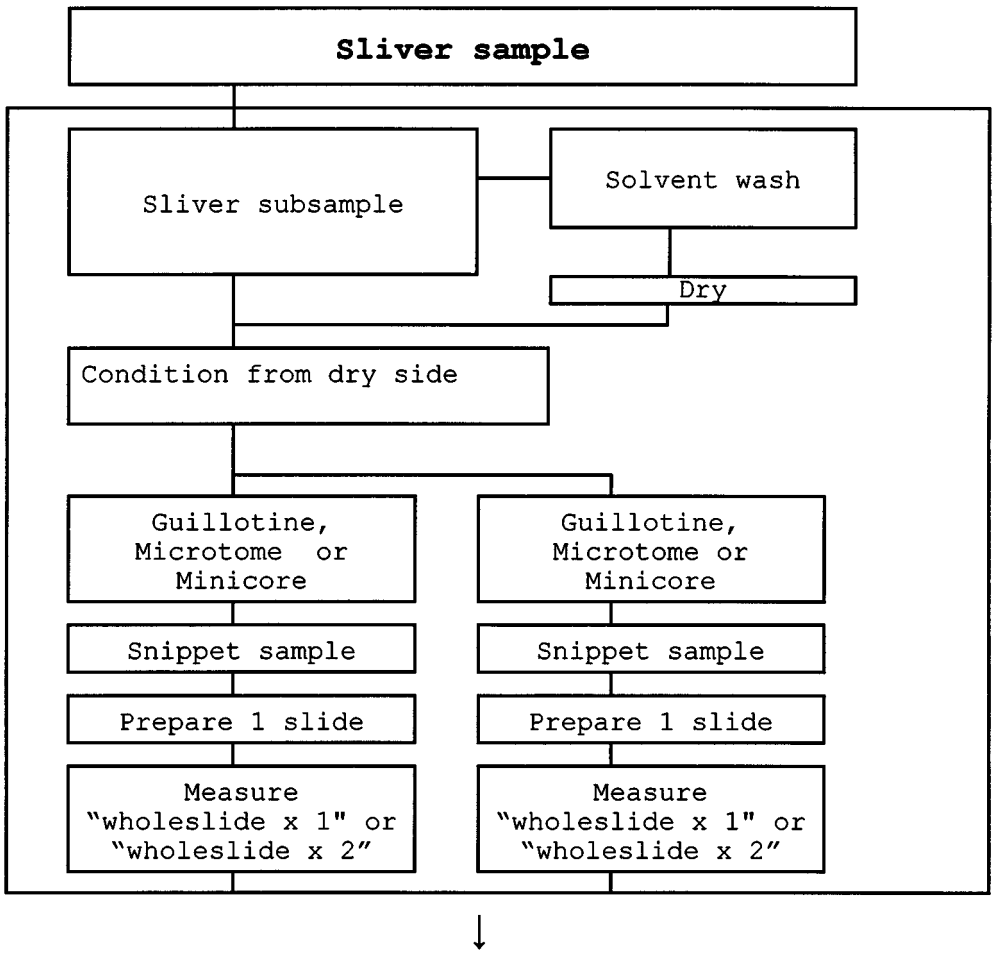

FIG. 7 Preparation of Test Specimens from Sliver Samples

TABLE 1 Average Fiber Diameter Tolerance Ranges for Various Numbers of Subsamples from Slivers and Greasy Cores

	Mean Fiber Diameter, µm			
	Sliver		Greasy Wool Cores	
	Less than 26.0	26.0 or more	Less than 26.0	26.0 or more
Range Between Two Slides, µm	0.4	0.7	0.5	0.9
Range Among Subsamples, µm:				
Number of Subsamples:				
2	0.3	0.5	0.4	0.7
3	0.3	0.6	0.4	0.8
4	0.3	0.6	0.5	0.9
5	0.4	0.6		
6	0.4	0.7		
7	0.4	0.7		
8	0.4	0.7		

diameters calculated for each subsample, calculate the range between subsamples.

11.6.1.3 In the case of greasy cores, if the appropriate range for 2 subsamples is exceeded, prepare and measure 2 more subsamples by scouring and drying 2 greasy core subsamples from the keeper material. If the appropriate range for 4 subsamples is exceeded, examine the results and determine whether one of the subsamples can be considered an outlier. If this is the case, remove that result and determine whether the range now complies with the appropriate range for 3 subsamples. If so, discard the outlier. If the range still cannot comply, combine the results from all 4 subsamples.

11.6.1.4 In the case of sliver subsamples, if the range exceeds the appropriate allowable range, test 2 further subsamples. If the appropriate range is still exceeded, examine the results and determine whether one of the subsamples can be considered an outlier. If this is the case, remove that result and determine whether the range now complies with the appropriate range for one less subsample. If so, discard the outlier. If the range still cannot comply, combine the results from all subsamples.

12. Calculation

12.1 For the sample, calculate the average of the mean fiber diameter, standard deviation and the coefficient of variation from the results on each slide.

13. Report

13.1 State that the specimens were tested as directed in ASTM Test Method D 6500 and state the type and number of samples taken and the kind of material that was tested.

13.2 Report the following information:

13.2.1 The average fiber diameter, µm, to one decimal place,

13.2.2 The number of accepted counts (individual measurements made on fiber snippets),

13.2.3 The standard deviation of fiber diameter, µm, to one decimal place,

13.2.4 The coefficient of variation of fiber diameter, to the nearest whole number,

13.2.5 Optionally, the distribution of fiber diameter as a frequency table or histogram with data grouped into appropriate classes,

13.2.6 Optionally, the 95 % confidence limits for the sample mean.

14. Precision and Bias

14.1 *Precision*—An international interlaboratory study was conducted in 1995 under the auspices of IWTO. This led to the acceptance of IWTO-47 in 1995. In Part I of the study, 40 greasy core samples were selected to cover broad ranges of average fiber diameter, vegetable matter base, and wool type. Each sample was supplied to the 4 participating laboratories as 2 150-g subsamples for scouring using the procedures defined in IWTO-19. Thus, each lab was required to test 80 individual subsamples using OFDA (also Laserscan and airflow). Each subsample was tested twice with at least 2000 fibers measured on every slide (that is, a total of 4 slides and greater than 8000 fibers measured per sample). In Part 2 of the study, 30 Interwoollabs IH Standard Tops were selected to cover a broad range of average fiber diameter. For all tops, airflow mean fiber diameter and projection microscope mean fiber diameters and standard deviations were known. Each sample was supplied to 11 participating laboratories in duplicate, coded to represent different samples. Each coded sample was measured only one time with at least 2000 fibers being measured on each slide.

14.1.1 Estimates of the components of variance and the 95 % confidence limits for wool tops and greasy wool cores are shown in Table 2. Table 3 provides the 95 % confidence limits in 5-µm increments for guillotined tops and aqueous scoured core samples. Similar information for other animal fibers (mohair, alpaca, cashmere) is being generated by members of ASTM Subcommittee D13.13.

14.2 *Bias*—The procedure described in this test method produces values of average fiber diameter that are not different than those produced when the same samples are measured by microprojection (see Test Method D 2130). Hence, this test method is considered to have no bias for the measurement of average fiber diameter of wool. In contrast, OFDA measures of standard deviation tend to be greater than microprojector measurements, hence the test method is biased in terms of measurements of standard deviation compared to the referee method.

TABLE 2 Components of Variance and the 95 % Confidence Limits for OFDA Measurements

NOTE 1—The 95 % confidence limits in Table 2 were calculated as follows: 95 % confidence limits = 1.96

$$\sqrt{\sigma^2\ within\ laboratory + \sigma^2\ between\ laboratory}.$$

Type of Sample and Fiber Diameter Range	Variance Between Laboratories, µm²	Variance Within Laboratory, µm²	Variance Total, µm²	95 % Confidence Limit, µm, ±
Guillotined Tops, 17.1–37.4 µm	0.0484	0.0354	0.0838	0.53
Aqueous Scoured Core Samples,16.0–39.0 µm	0.0443	0.0380	0.0823	0.51

TABLE 3 95 % Confidence Limits for OFDA Measurements in 5-µm Increments

Mean Fiber Diameter, µm	OFDA 95 % Confidence Limits, µm, ±	
	Guillotined Tops	Aqueous Scoured Core Samples
15.0	0.18	0.25
20.0	0.30	0.37
25.0	0.42	0.48
30.0	0.54	0.60
35.0	0.66	0.70
40.0	0.78	0.82
45.0	0.90	0.92

15. Keywords

15.1 animal fibers (except wool); diameter; diameter distribution; wool

ANNEX

(Mandatory Information)

A1. CALIBRATION OF THE OFDA

A1.1 *Principle*—The OFDA is calibrated for measuring wool snippets using test specimens prepared from all eight of the current Interwoollabs IH Standard Tops that have known mean fiber diameters as determined in round trials using projection microscopes (a similar set of mohair tops is also available). The eight mean values obtained from OFDA measurements are converted to non-dimensional" W" numbers, that are linearly regressed against the average mean fiber diameter values supplied by Interwoollabs, so as to allow determination of the coefficients *A* and *B* in the following equation:

$$IH_{mean} = A + B\ (OFDA\ "W") \qquad (A1.1)$$

where:

IH_{mean} = average mean fiber diameter value from Interwoollabs projection microscope interlaboratory trials, and

OFDA "W" = non-dimensional fiber diameter number generated by the OFDA.

This equation is used to calculate the calibration table that allows the individual OFDA fiber measurements to be assigned to classes of 1 µm width.

A1.2 *Calibration Considerations*:

A1.2.1 When a calibration is being established for measurement of greasy core samples, the calibration shall be established by cutting the calibration tops into 12- to 15-mm lengths, that shall then be scoured and dried using the procedures outlines in Test Method D 584. The scoured tops are then brought to equilibrium with the standard atmosphere from the dry side and subsequently minicored.

A1.2.2 A calibration being established strictly for measuring snippets guillotined or microtomed directly from conditioned sliver can be used to establish a calibration strictly for measuring slivers. If the slivers to be routinely measured are expected to contain more than 1.5 % ethanol extract (or 1.0 % dichloromethane extract), the slivers shall be cleaned to remove greasy compounds using petroleum spirit or 1,1,1 trichloroethane. Solvent extracted sliver shall then be dried and conditioned prior to cutting snippets. If solvent cleaning is routinely required on slivers to be measured, it should also be done on the calibration materials.

A1.3 *Calibration Procedure*:

A1.3.1 *Preparation of Snippet Sample*—Prepare snippet samples from subsamples of top that have been brought into equilibrium with the standard atmosphere from the dry side. Cut enough fiber snippets from each subsample to allow, on each test specimen, at least 2000 snippet measurements. A mass of between 15 mg at a mean fiber diameter of 20 µm and 25 mg at 35 µm is usually sufficient. Two methods may be used to obtain snippets.

A1.3.1.1 *Minicoring (Applicable to Sliver and Raw Wool)*—Minicore the subsample, or, where the whole subsample cannot be accommodated in the minicorer, select a representative portion of about 10 g mass and minicore this portion. Minicore the subsample or the selected 10-g portion a sufficient number of times to provide adequate snippet sample for each subsample.

A1.3.1.2 *Guillotining or Microtoming (Applicable Only to Sliver)*—Cut snippets from each subsample with a fiber cutting instrument. Make the same number of cuts from each subsample. In the case of sliver, do not cut snippets from within 100 mm of either end of the piece. Cut a sufficient number of times to provide adequate snippet sample for each subsample.

A1.3.2 *Storage of snippets*—Whatever method is used to obtain snippets, they must be collected and, if necessary, stored, in clean vessels of glass or metal, in order to avoid any segregation that may result from the effects of static electricity. Snippet samples must be protected against contamination and drafts that may cause loss of finer fibers.

A1.3.3 *Preparation of the Test Specimen(s)*—Clean all surfaces of each glass slide pair by wiping with a lintless swab dampened with alcohol. Take care at all times to avoid leaving fingerprints or other deposits on the glass slides, since this may lead to focusing difficulties and measurement inaccuracy. Do not allow alcohol to come into contact with the adhesive holding the hinge between the pair of slides. Insert a clean glass slide in the slide preparer and, using a pair of tweezers or a miniature sampling spoon, select about five equally representative portions of the snippet sample and introduce them to the slide preparer. It is preferable to use the entire snippet sample, or, where this is not feasible, ensure that snippets are representatively selected from the entire depth of the sample to avoid the effects of snippet segregation. Also, ensure that the entire part of the sample placed in the spreader is allowed to pass through onto the slide, since in some samples the coarse fibers fall first through the spreader. Where the snippet samples have been prepared from a sample containing high levels of vegetable matter, it may be necessary to remove residual non-fibrous particles from the snippet samples using a pair of tweezers in order to avoid focusing problems caused by inability to fully close the slide pair. Upon completion of the snippet spreading, carefully place the clean cover slide over the fibers without disturbing the snippets and secure in place. Prepare one slide from each snippet sample.

A1.3.3.1 *Number of Test Specimens*—In order to maintain equal precision of the mean for each top, the number of slides necessary shall be determined by the between-slides variance. The variance shall be determined by measuring a suitable number of slides prepared for each top. Calculate the number of slides, *m*, in order to achieve a standard error of the mean of 0.1 µm. As guidance, for the 10th Interwoollabs IH series (1994), the following minimum numbers of slides are recommended: 17.0 and 19.1 µm—2 each; 20.5 and 23.6 µm—4 each; 27.1 and 29.5 µm—6 each; 34.3 and 37.7 µm—8 each.

A1.3.4 *Measurement of Fiber Diameter Distribution*—Operate the instrument in a clean, substantially dust-free environment maintained at standard atmosphere. Where the electricity supply is "noisy" or subject to disturbance, the equipment supply shall be protected by an uninterruptable power supply and conditioning unit.

A1.3.4.1 *Premeasurement Check*—Ensure that the instrument is set up in accordance with the instructions in the operating manual. Check the microscope focus and adjust if necessary. If at any time during measurement the software indicates the microscope is out of focus, manually bring the microscope back into focus.

A1.3.4.2 *Measurement of Calibration Slides*—Measure each slide with the instrument set on "Wholeslide× 1" or "Wholeslide × 2." In each case, ensure that the total count size is at least 2000. Print the mean values. Record the offset and slope for the calibration used in the measurements.

A1.4 *Calibration Calculations*—The following calculations may be conveniently carried out on any spreadsheet program that allows calculation of linear regression coefficients. The normal precautions should be taken to ensure that the program gives the same results as the manual procedure below. Rounding should not be carried out.

A1.4.1 *Calculation of Means for Each Top*—For each of the *m* slides prepared, calculate the average mean fiber diameter

value, d_i from the mean fiber diameters reported for each of the slides, d_n:

$$d_i = (\sum_{n=1}^{m} d_n)/m \quad \text{(A1.2)}$$

From the OFDA calibration menu, determine the offset and slope of the current calibration, that may be the factory calibration. For each top, calculate the mean "W" value:

$$W_i = (d_i - \text{current offset})/\text{current slope} \quad \text{(A1.3)}$$

A1.4.2 *Calculation of Coefficients*:

A1.4.2.1 Calculate the following means:

$$\alpha = (\sum_{i=1}^{8} W_i)/8 \quad \text{(A1.4)}$$

$$\beta = (\sum_{i=1}^{8} \Phi_i)/8 \quad \text{(A1.5)}$$

where Φ_i is the mean fiber diameter value supplied by Interwoollabs for the *i*th top.

A1.4.2.2 Calculate coefficient *B* from:

$$B = \frac{\sum_{i=1}^{8} (W_i - \alpha)(\Phi_i - \beta)}{\sum_{i=1}^{8} (W_i - \alpha)^2} \quad \text{(A1.6)}$$

and coefficient *A* from:

$$A = \beta - B\alpha \quad \text{(A1.7)}$$

On completion of the calibration calculations, select "Calibration" on the instrument menu and input the new calibration using the value of *A* as the new "Offset" and the value of *B* as the new "Slope."

A1.5 *Verification of Calibration*:

A1.5.1 *Principle*—Test samples of wools with known mean fiber diameters and distribution characteristics are measured using the new calibration. Preferably, these are different tops than those used for calibration. The results are compared to the known values and the calibration is accepted if the differences fall within acceptable limits outlined in A1.5.3. If the criteria are not satisfied, corrective action (that is, recalibration) must be taken.

A1.5.2 *Procedure*—With the new calibration installed, prepare and measure (using identical sample preparation, cutting, conditioning, and spreading methods as were used for calibration) a new set of slides comprising at least 2 from each of 8 tops with known characteristics. More slides may be necessary for the coarser tops, depending on the level of between-slide variance. For the mean of each pair of slides, calculate the difference (Γ_i) between the measured mean fiber diameter (d_i) and the known value (Φ_i).

A1.5.3 *Criteria*—Each of the following criteria must be satisfied:

A1.5.3.1 *Criterion 1*—Calculate the mean, *D*, variance S_D^2, and *t*-value, *t*, for the differences using Eq A1.8-A1.10 respectively:

$$D = (\sum_{i=1}^{8} \Gamma_i)/8 \quad \text{(A1.8)}$$

$$S_D^2 = \{\sum_{i=1}^{8} \Gamma_i^2 - [(\sum_{i=1}^{8} \Gamma_i)^2/8]\}/7 \quad \text{(A1.9)}$$

$$t = (D\sqrt{8})/S_D \quad \text{(A1.10)}$$

The *t*-value must not exceed 2.365, this being the Student's t-value for 7 (that is, *n*-1) degrees of freedom at the 0.05 level.

A1.5.3.2 *Criterion 2*—Calculate the gradient, *g*, variance, S_g^2, and *t*-value for the regression of the differences against the given Interwoollabs mean fiber diameters using Eq A1.11-A1.13 respectively:

$$g = \frac{8\sum_{i=1}^{8} \Gamma_i^2 - (\sum_{i=1}^{8} \Gamma_i)(\sum_{i=1}^{8} \Phi_i)}{8\sum_{i=1}^{8} \Phi_i^2 - (\sum_{i=1}^{8} \Phi_i)^2} \quad \text{(A1.11)}$$

$$S_g^2 = \left| \frac{8\sum_{i=1}^{8} \Gamma_i^2 - (\sum_{i=1}^{8} \Gamma_i)^2}{8\sum_{i=1}^{8} \Phi_i^2 - (\sum_{i=1}^{8} \Phi_i)^2} - g^2 \right| /6 \quad \text{(A1.12)}$$

$$t = g/S_g \quad \text{(A1.13)}$$

the *t*-value must not exceed 2.447, this being the Student's t-value for 6 (that is, *n*-2) degrees of freedom at the 0.05 level.

A1.5.3.3 *Criterion 3*—To calculate the mean square error of the calibration, use the mean measured diameters obtained from the eight sets of verification slides ($OFDA_i$) and the eight projection microscope values (PM) supplied by Interwoollabs for the tops (Φ_i). Calculate the regression between the assigned PM values and the average OFDA values as follows:

$$U = \sum_{i=1}^{8} \text{OFDA}_i)/8 \quad \text{(A1.14)}$$

$$V = (\sum_{i=1}^{8} \Phi_i)/8 \quad \text{(A1.15)}$$

where Φ_i is the mean fiber diameter value supplied by Interwoollabs for the *i*th top. Calculate coefficients *S* and *T* using the following equations:

$$S = V - TU \quad \text{(A1.16)}$$

$$T = \frac{\sum_{i=1}^{8} (\text{OFDA}_i - U)(\Phi_i - V)}{\sum_{i=1}^{8} (\text{OFDA}_i - U)^2} \quad \text{(A1.17)}$$

Calculate the equivalent regressed PM values for each top using the following equation:

$$\text{PM}_i = S + T \times \text{OFDA}_i \quad \text{(A1.18)}$$

Calculate the mean square error as follows:

$$MSE = \frac{\sum_{i=1}^{8} (\Phi_i - \text{PM}_i)^2}{6} \quad \text{(A1.19)}$$

The MSE must not exceed 0.10 μm^2.

A1.5.4 *Verification*—If any of the criteria cannot be satisfied, the instrument may need adjustment, or one of the slides may not be representative. An anomaly usually may be isolated by examining plots of the calibration and verification data against the regression line. The instrument must be recalibrated if it needs to be adjusted. Immediately after a calibration has been concluded and verified, it is often convenient to remeasure the mean fiber diameter and standard deviation of all or a selection of the slides used in the calibration (and any others that may be considered helpful to cover an extended range of fiber characteristics), and to record the results on labels affixed to the slides. Such slides, as long as they are securely taped around the edges, stored horizontally, kept at the standard conditions, handled carefully, and kept free from grease or other surface contamination, may be used for routine premeasurement checks on the instrument.