Date: December 20, 1994

From: Director, Center for Biologics Evaluation and Research

Subject: Recommendations to Users of Medical Devices That Test for Infectious Disease Markers by Enzyme Immunoassay (EIA) Test Systems

To: All Registered Blood Establishments

This memorandum provides recommendations to assist users of medical devices that test for infectious disease markers by enzyme immunoassay (EIA) in complying with the Food, Drug, and Cosmetic Act, the Public Health Service Act, and all applicable regulations, including those found in 21 CFR, Parts 200 and 600. Recommendations provided address the areas of test equipment documentation, i.e., performance data, installation instructions, validation procedures, calibration methodology, verification and recalibration, precision and accuracy determinations, problem detection and correction, troubleshooting, and preventive maintenance.

The attached recommendations are directed to all establishments engaged in the collection, processing, testing, storage, and distribution of blood and blood products, specifically including blood banks, laboratories engaged in blood product related testing, plasmapheresis centers, and transfusion services.

The attachments to this memorandum provide blood establishments with a brief listing of necessary information to successfully operate the equipment (Attachment I), typical problems they may encounter, and their possible solutions (Attachment II). The development of the recommendations on validation and the attachments into a checklist format adapted to your establishment's particular needs should assist you in applying the recently issued Quality Assurance Guidelines.

This memorandum does not supersede any previous memoranda but should be used in conjunction with the References/Resources listed to obtain the optimum benefit of the test system.

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Kathryn C. Zoon, Ph.D.

RECOMMENDATIONS TO USERS OF MEDICAL DEVICES THAT TEST FOR INFECTIOUS DISEASE MARKERS BY ENZYME IMMUNOASSAY (EIA) TEST SYSTEMS

TABLE OF CONTENTS

I. Introduction

With the increased emphasis of blood centers and transfusion services on preventing transfusion transmitted diseases, blood establishments need additional training, quality control, preventive maintenance checks, and troubleshooting and problem solving skills to ensure successful operation of enzyme immunoassay test systems.

Manufacturers of the equipment discussed in these recommendations are required to document specifications as well as installation, validation, calibration, calibration verification, quality control and preventive maintenance procedures as described in 21 CFR 809.10 (b)(6),(8)(v) and (vi), 21 CFR 820.100 (a), and 21 CFR 820.152. Frequently, the equipment manufacturer's Customer Service Representative (CSR) or Technical Service Representative (TSR) will provide much first-hand assistance. However, the **blood establishment** should ensure that the above documentation is provided in order that it can comply with 21 CFR 211.68 and 21 CFR 606.60.

The amount of testing to be done and performance data for the above procedures depends on the equipment used and the particular test(s) associated with the equipment. The resources listed in part IX may be helpful in determining acceptable amounts of testing for performing each of the above procedures.

II. Equipment (Device Definitions)

A. Sampler/Sample Identifier/Micropipettor/Dilutor - a multifunctional device that may identify, dilute and/or aspirate a predetermined amount of sample and/or dispense the necessary reagents.

B. Microplate washer - a device that adds a predetermined amount of wash solution and aspirates the solution a pre-set number of times. The manufacturer determines the amount of solution and the number of washes necessary to prepare the sample for the next stage of the testing procedure.

C. Incubators (dry or water) - a device used to bring the sample/reagent mixture to the proper temperature and maintain it at a pre-set temperature for the amount of time determined necessary for a reaction to take place.

D. Microplate readers (Spectrophotometer) - device used to measure the amount of color development in each well (reaction chamber) of the microplate as compared to a standard.

E. Refrigerators, Centrifuges, Rotators & Shakers, and Balances - definition of these instruments is self-explanatory. While not integral components of the EIA test system, these

instruments can have significant effects on the outcome of this testing if they are not functioning properly.

III. Installation

The blood establishment should ensure that the manufacturer installs the equipment or provides complete installation/setup instructions. The installation/setup instructions should include, when applicable, a linearity check, volume verification for any required volume measurements, comparison and confirmation of configuration/parameter settings, diagnostic communication checks for any interfaced instruments, testing of samples for each protocol, and verification of accuracy of readouts.

IV. Specifications

Specifications are the operating parameters of the instrument or test system. The device must be capable of delivering the level of performance (accuracy, precision) consistent with the requirements of the test kit package insert. The blood establishment should ensure that the manufacturer provides these performance specifications in the instrument Operator's Manual and the specifications should address random and systematic (both constant and proportional) error occurring in both intra-assay and inter-assay runs, the calibration methods used, a direct comparison of the method to an established reference method (with total error below that deemed clinically significant), the reportable range of test results, and, when applicable, results of linearity studies using, for example, National Committee for Clinical Laboratory Standards (NCCLS) listed in part IX or equivalent guidelines.

V. Validation

Validation establishes documented evidence which provides a high degree of assurance that a specific process will consistently produce a product meeting its predetermined specifications and quality attributes. The data should be collected using appropriate samples to support test performance characteristics of accuracy, precision, sensitivity, and specificity, and when applicable, reportable and reference range(s), and any other applicable performance characteristics. Process validation is performed to evaluate the performance of a system with regard to its effectiveness based on the intended purpose, such as meeting the test kit manufacturer's reagent specifications.

A. EQUIPMENT DOCUMENTATION

Infectious disease test products are medical devices. Firms that manufacture and distribute such devices must comply

with regulatory requirements applicable to these devices. The blood establishment should ensure that
manufacturers provide an operator/user manual that has accurate and complete instrument user instructions plus a listing of the required equipment to validate the instrument. The manual should contain these items:

1) a functional description of the microprocessor-controlled test system, i.e., its intended use, the applications and reagents that have been cleared for use with the device, and a mechanism to update the manual when new tests are available;

2) general operating instructions and system limitations with warnings/cautions;

3) a description of the hardware which includes the installation instructions (even if performed by the instrument manufacturer), all integrated functional components, configuration and linkage of integrated components, environmental and maintenance requirements, interface specifications, and all warning/error messages and their appropriate corrective action;

4) a description of the software that includes a file/record structure, field descriptions, user definable database options, warning/error messages and their appropriate corrective action, an assurance of data accuracy, and software performed calculations;

5) specimen specifications (which, although provided in each test kit package insert, should also be in the operator/user manual), such as serum or plasma (and type of anticoagulant used), maximum age at which sample can be tested, storage and preparation required, amount needed for testing, and a list of substances known to interfere with the test; and

6) change control and problem reporting procedures, document control process, and customer support services.

The blood establishment should provide adequate staff training and guidelines for the development of a test plan that addresses software control functions, calculations performed by the software, conditions that trigger alarms, error and warning messages, data entry methods, boundaries, field limits and ranges, security procedures and program overrides, and data storage, transfer, and retrieval. The software should identify the version and all default parameters used in running the test, both at the time of running the test and on the printout, and should be validated by the manufacturer for all calculations, "flags" for limits, and failures.

B. RECOMMENDATIONS FOR BLOOD ESTABLISHMENTS

The user should perform the necessary equipment validation to establish confidence that the device and its ancillary systems are capable of consistently operating within established limits and tolerances.

SAMPLER/SAMPLE IDENTIFIER/MICROPIPETTOR/DILUTOR - can be validated with calibrated pipet tips which hold the proper volume or by a gravimetric method if a balance sensitive enough to weigh very small volumes is available. Sample identification can be validated by testing with barcode controls of known definition.

MICROPLATE WASHER - can be validated by obtaining an exact measurement of the fluid being dispensed, either from each dispensing probe or from all probes combined into a calibrated volumetric flask.

INCUBATOR - can be validated with the use of a total immersion verification thermometer preferably with 0.2° temperature demarcations; however, 1.0° temperature demarcations are acceptable.

MICROPLATE READER (Spectrophotometer) - can be validated using the manufacturer's instructions, keeping in mind that an operational verification, an alignment verification, and a linearity test are necessary.

REFRIGERATORS, CENTRIFUGES, ROTATORS & SHAKERS, and BALANCES - can be validated by well documented procedures.

VI. Calibration and Calibration Verification

Calibration is the process of testing and adjusting an instrument, kit, or test system to provide a known relationship between the measurement response and the value of the substance that is being measured by the test procedure. Calibration verification is the process used to assure that the calibration of the instrument, kit, or test system remains stable throughout the laboratory's reportable range for test results. Calibration and recalibration of less complex instruments can be performed by following the instructions in the operator's manual. Some more complex instruments also have components that can be calibrated and recalibrated by the user, and many have troubleshooting guides which are helpful in correcting minor instrument problems. The device or test system manufacturer should provide the necessary calibrators or specify what calibrators may be used and information on the items needed to perform this process.

The blood establishment should ensure that the instrument meets the requirements described in 21 CFR 211.68 and 21 CFR 606.60 for calibration.

VII. Quality Assurance/Quality Control

Quality assurance consists of the actions, planned and taken, that provide confidence that all systems and elements that influence the quality of the product are working as expected (individually and collectively).

Quality control is a component of quality assurance that includes the activities and controls used to determine the accuracy of the establishments' personnel, equipment, and operations.

Quality control procedures recommended by the Center for Biologics Evaluation and Research (CBER) for blood establishments are set out in CBER's Draft Guideline for Quality Assurance in Blood Establishments, dated July 2, 1993.

VIII. Preventive Maintenance/Function Checks

Preventive maintenance and function checks performed on a regular basis will help to ensure that the instrument will perform correctly, will have less downtime, and will reduce other maintenance/repair costs. The blood establishment should ensure that an acceptable maintenance and service schedule is provided, which includes cleaning, adjusting, and testing assay equipment.

There should be protocols for maintenance and function checks with documentation that calibration verification and quality control will remain stable between the intervals recommended.

IX. References/Resources

The following resources may be obtained from the Center for Biologics Evaluation and Research, Division of Congressional and Public Affairs, HFM-12, 1401 Rockville Pike, Suite 200N, Rockville, MD 20852-1448, FAX (301) 594-1938:

1. Draft Guidelines for Quality Assurance in Blood Establishments, Docket No. 91N-0450, July 2, 1993.

2. FDA Memorandum, Recommendations for Implementation of Computerization in Blood Establishments, April 6, 1988.

3. FDA Memorandum, Requirements for Computerization of Blood Establishments, September 8, 1989.

4. FDA Memorandum, Changes in Equipment for Processing Blood Donor Samples, July 21, 1992.

5. Draft Guideline for the Validation of Blood Establishment Computer Systems, Docket No. 93N-0394, October 28, 1993.

The following resource may be obtained from the Superintendent of Documents, U.S. Government Printing Office, Mail Stop: SSOP, Washington, DC 20402-9328.

6. Title 21 Code of Federal Regulations, Parts 200-299, 600-799, and 800-1299, revised annually and available April 1 of each year.

The following resource may be obtained from the Center for Devices and Radiological Health, Division of Consumer Affairs, HFZ-210, 1901 Chapman Avenue, Rockville, MD 20857 (301) 443-4190.

7. Guideline on General Principles of Process Validation, May, 1987.

The following resource may be obtained from the American Association of Blood Banks, 8101 Glenbrook Road, Bethesda, MD 20814-2749, (301) 907-6977.

8. Blood Bank/Transfusion Service Computer Systems, prepared by AABB Information System Committee and HIMA ad hoc Computer Committee, August 27, 1993.
   a) User Validation Guidelines.
   b) Software Manufacturing Process Guidelines
   c) Validation Guidelines for Microprocessor Controlled Test Instruments.

OTHER USEFUL RESOURCES:

9. National Committee for Clinical Laboratory Standards. Villanova, PA.
   a) Internal quality control testing: principles and definitions. Approved Standard. 1990.
   b) User comparison of quantitative clinical laboratory methods using patient samples. Proposed Guideline. 1992.
   c) Preliminary Evaluation of Clinical Chemistry Methods. Tentative Guideline. 1989.
   d) Evaluation of precision performance of clinical chemistry devices - second edition. Tentative Guideline. 1991.
   e) How to define, determine and utilize reference intervals in the clinical laboratory. Proposed Guideline. 1991.
   f) Specifications for Immunological Testing for Infectious Diseases. Proposed Guideline. 1991.

Technical questions, information, and comments may be directed to the:

Division of Blood Applications, HFM-380
1401 Rockville Pike, Suite 200N
Bethesda, MD 20852-1448
Phone: (301) 594-6487 FAX: (301) 594-6431

ATTACHMENT I.

LABELING APPLICABLE TO MEDICAL DEVICE MANUFACTURERS
(not to be considered all inclusive)

A. All devices, either as individual products or as components in a total system, should contain the following in the labeling section (package insert) [see 21 CFR 809.10 (b)]:

1. Intended use or function.
2. Installation instructions and special requirements.
3. Introduction and Principles of Operation.
4. Performance characteristics and specifications.
5. Operator's Control Manual and operating instructions (which should include a Troubleshooting Guide).
6. Calibration procedures including materials and/or equipment to be used.
7. Operational precautions and limitations.
8. Hazards.
9. Preventive Maintenance Procedures and service information.
10. Interpretation of Data.
11. Quality Control Procedures and Schedules.

Information contained in the performance section should be from a statistically significant number of samples tested. Sample size depends on the particular application of the device being demonstrated.

B. <u>Sampler/Sample Identifier/Micropipettor/Dilutor</u>

1. Performance data should include test results that demonstrate:
   a) the sampler provides correct identification;
   b) the micropipettor processes the correct amount of sample;
   c) the dilutor makes the appropriate dilution;
   d) reproducibility of results; and
   e) stability and accuracy in volumes pipetted and diluted.

2. Specific labeling information should include the following:
   a) recommended pipet tips;
   b) instructions for repositioning and changing tips;
   c) instructions for verifying that pipet tips were properly changed;
   d) information on battery, shelf life, and effects of power outage;
   e) validation, calibration, and quality control procedures.

C. Microplate Washer

1. Performance data should include test results demonstrating the following:
   a) the washer adds the recommended amount of wash solution;
   b) the washer removes all wash solution each wash cycle;
   c) the effects of improper washing; and
   d) the washer has performed the appropriate number of soaks and the duration of each.

2. Specific labeling information should include the following:
   a) instructions for repositioning pipet tips;
   b) instructions for replacing or correcting a seal between the pipet tip and holder;
   c) instructions for priming the washer;
   d) schedules and instructions for changing tubing and connectors;
   e) disinfection procedures;
   f) instructions for pressure problems;
   g) instructions for determining proper fluid level heights and fluid volume settings;
   h) recommended microplates that will be compatible with the device; and
   i) validation, calibration, and quality control procedures.

D. Incubators

1. Performance data should include test results that demonstrate the following:
   a) the recommended temperature is that required by the test kit manufacturer;
   b) the effects on samples incubated outside of the recommended range; and
   c) the recommended time of incubation is that required by the test kit manufacturer.

2. Specific labeling information should include the following:
   a) procedures for making minor adjustments in temperature;
   b) procedures for detecting and correcting hot or cold spots; and
   c) validation, calibration, and quality control procedures.

E. Microplate readers (Spectrophotometers)

1. Performance data should include test results demonstrating the following:
   a) the effects of power surges;
   b) the interference effects, if any, of other equipment;
   c) reproducibility of results;
   d) stability in maintaining the manufacturer's recommended settings; and
   e) that wavelength used is optimal.

2. Specific labeling information should include the following:
   a) wavelength required for each test that can be performed;
   b) linear limits (absorbance range);
   c) linearity or accuracy;
   d) drift;
   e) repeatability;
   f) bandwidth;
   g) a method to prevent and detect the use of the wrong wavelength;
   h) a method to detect incorrect programming;
   i) information on battery - shelf life, effects of power outage and surges;
   j) recommended placement of the device to avoid environmental interferences;
   k) recommended procedures for realigning the light source;
   l) a procedure for daily quality control of the barcode reader; and
   m) validation, calibration, and quality control procedures.

ATTACHMENT II.

SAMPLE TROUBLESHOOTING GUIDE (not to be considered all inclusive)

The purpose of this guide is to assist the blood establishment in correcting minor problems encountered any time a device is in use, thus minimizing downtime. This guide should be included in the User's or Operator's manual. It should be prepared and provided by the device manufacturer with input from users. With the continuing advances in technology, this guide should be in a format that facilitates additions, deletions, and corrections.

A list of equipment, problems associated with each, and the reasons for their occurrence follows:

A. Sampler/Sample Identifier/Micropipettor/Dilutor

1. Incorrect amount or no sample or reagent added or aspirated
   a) the pipet tip is incorrectly positioned which in turn can cause air to be aspirated, thus reducing the amount of sample drawn or causing the incorrect amount of reagent to be added to the reaction chamber;
   b) the wrong pipet tips are used (not recommended by the device manufacturer or the device manufacturer has not specified which pipet tips have been found to be compatible with the device through parallel testing);
   c) the tips are not changed in accordance with the manufacturer's instructions (or the manufacturer has failed to provide adequate instructions);
   d) the tip threshold calibration has not been performed when the pipet tip nozzle, tubing syringe, or syringe plunger were changed;
   e) there are occasions when programmable dilutors do not hold the program and revert back to an unprogrammed state;
   f) the liquid level detector is not performing properly and does not signal the microprocessor to relay an error message;
   g) there are clogged lines or air bubbles in the system which have gone undetected; or
   h) the lines have been mixed and the wrong reagent has been used.

2. Misalignment of pipet tip
   a) the device picks up the pipet tips;
   b) the wrong tips are used;
   c) the wrong micropipettor is used;

d) there is a misalignment of either the pipet tip shaft of the device or the sample, dilution or reagent holding rack;
e) the user has failed to verify that the device is operating properly or has failed to follow SOPs; or
f) the manufacturer has failed to adequately validate this stage of the process or has not provided adequate instructions.

3. Incorrect sample identification
a) there are undetected errors with the built-in barcode reader;
b) the sample tube is incorrectly aligned;
c) the barcode has been damaged in some way;
d) label supplier has been changed without verifying that the labels are compatible with the device;
e) the wrong label is placed on tube;
f) the user fails to quality control the barcode reader each day of use;
g) the manufacturer fails to thoroughly test the barcode reader before distribution; or
h) the manufacturer fails to provide adequate quality control instructions to the user.

4. Missing samples
a) the micropipettor fails to pick up a pipet tip;
b) the micropipettor fails to aspirate a sample; or
c) no sample is put into the sample well of a reaction cell.

B. Microplate Washer

1. Residual wash solution in well
a) the pipet tip is not properly positioned and does not dip far enough into the well;
b) the seal between the pipet tip and the shaft is not complete;
c) the wrong microplates are used;
d) the washer not properly primed;
e) the user fails to follow the appropriate SOPs;
f) the manufacturer fails to properly test the device before distribution;
g) the manufacturer fails to provide adequate instructions for the user;
h) the device does not have an adequate vacuum source;
i) the vacuum tubing is not of the recommended diameter and wall thickness;
j) the lines have become clogged;
k) the waste container is full;
l) the line filter is wet or clogged; or

m) the reaction tray (microwell plate) is not positioned correctly onto washer.

2. Bacterial/fungal growth in wash reservoir
   a) the system is not used on a regular basis;
   b) the tubing and connectors are not changed on a regular basis;
   c) the regularly scheduled maintenance is not performed;
   d) the wash solution is contaminated by improper handling;
   e) the recommended disinfectant is not used; or
   f) the disinfectant procedure is not followed.

3. Inadequate washing cycle/uneven filling of wells
   a) the wash solution reservoir is not checked to determine that an adequate amount of solution is in the reservoir;
   b) the washer is not properly primed before each use;
   c) salt crystals or particulate matter clog the dispensing tip;
   d) the pressure is insufficient to deliver the appropriate amount of wash solution;
   e) there is a crimp or blockage in the fill line;
   f) the volume of wash solution is inadequate due to an incorrect volume setting or the plate is not properly seated in the washer;
   g) the proper working height is not used;
   h) the correct fluid levels are not maintained; or,
   i) the inlet tubing is not the recommended diameter and wall thickness;
   j) the plates are used with strip wells that are not properly seated; or
   k) plates are used that are not recommended by the manufacturer.

C. Incubators

1. Temperature fluctuations
   a) the chamber door or lid is opened too often;
   b) the temperature controller is not functioning properly;
   c) the device is not located properly, i.e., placed close to sunlight, heat or air conditioning vents);
   d) the water pan (found in some incubators) dries out; or
   e) the device is moved without recalibrating.

2. Hot spots/cold spots
   a) the circulating fan is not working properly; or,
   b) the device is moved and the heating elements are out of alignment.

D. Microplate Readers (Spectrophotometers)

1. Incorrect readings (falsely lowered or elevated)
   a) a particular reagent is omitted;
   b) the wrong reagent or the wrong amount of reagent is added;
   c) the microplate wells have scratches on the bottom;
   d) the wrong plates or dirty plates are used;
   e) the microplate wells have been allowed to dry out;
   f) the wrong wavelength is used;
   g) the device is programmed incorrectly;
   h) the microprocessor operating the system fails to use the correct wavelength;
   i) the plate is not seated correctly in the reader;
   j) the bottom exterior of plates contains fingerprints or moisture;
   k) the reader is exposed to excessive dust, vibration, strong magnetic fields, direct sunlight, draft, excessive moisture or large temperature fluctuations;
   l) the linearity verification and/or drift verification were not performed either at all or according to manufacturer's directions;
   m) residual amounts of diluted Plate Wash Buffer remain in test wells prior to the addition of Substrate;
   n) the Substrate tablets are not completely dissolved;
   o) the Substrate tablet has been contaminated by moisture or metal forceps, or is not intact (< a whole tablet);
   p) the temperature equilibration of the test components is incomplete;
   q) the reader is not properly warmed up;
   r) the position of the blank well may have been changed and the wrong value was subtracted from each reading; or
   s) results may not have been read within the allowable time.

2. Data not transferred
   a) the reader and the microprocessor have different character sets;
   b) the baud rates do not match;
   c) the transmit/receive pin configuration is not set properly;
   d) the communication interface software routines fail; or
   e) the connection cable is improperly connected.

3. Light beam misalignment
   a) the device is shipped or moved without the necessary precautions;

b) the installation or preventive maintenance is performed; or,
c) the light source (lamp) is changed and is not seated properly or aligned correctly.

4. Incorrect sample identification
   a) the plates are not loaded properly; or,
   b) the wrong sample identification has been entered into the reader.

E. Computer

1. Failure to give error messages
   a) software section controlling conditions that trigger alarms, errors, and warning messages has not been thoroughly tested and validated by the manufacturer.

2. Failure to detect malfunctions
   a) functional components of the system (liquid level detection system, fluid handling, etc.) or the interface routines have not been thoroughly tested.