This guidance was written prior to the February 27, 1997 implementation of FDA's Good Guidance Practices, GGP's. It does not create or confer rights for or on any person and does not operate to bind FDA or the public. An alternative approach may be used if such approach satisfies the requirements of the applicable statute, regulations, or both. This guidance will be updated in the next revision to include the standard elements of GGP's.

# REVIEW CRITERIA FOR IN VITRO DIAGNOSTIC DEVICES FOR DETECTION OF IGM ANTIBODIES TO VIRAL AGENTS

This is a flexible document representing the current major concerns and suggestions regarding in vitro diagnostic devices employing immunochemical or other methodologies for detection of IgM antibodies to specific virus in human serum or plasma specimens. It is based on 1) current basic science, 2) clinical experience, and 3) the Safe Medical Devices Act of 1990 (SMDA) and FDA regulations in the Code of Federal Regulations (CFR). As advances are made in science and medicine, these review criteria will be re-evaluated and revised as necessary to accommodate new knowledge.

**PURPOSE:** The purpose of this document is to provide guidance on information to present to the Food and Drug Administration (FDA) before a device to detect IgM antibodies to specific viral agents in serum or plasma specimens may be approved/cleared for marketing. This document is an adjunct to the CFR and the FDA 87-4214 Premarket Approval (PMA) Manual.

**DEFINITION:** This generic type device is intended for use in clinical laboratories as an in vitro diagnostic test for qualitative or semi-quantitative measurement of IgM class antibodies to specific viruses in human serum or plasma by immunochemical or other methodologies.

In addition to the guidance presented here, a review of the National Committee for Clinical Laboratory Standards (NCCLS), "Specifications for Immunological Testing for Infectious Diseases"[1] is suggested. The NCCLS document may be used for definitions of terms used in this guidance document.

This guidance document is to be used as a general guideline for manufacturers to consider when submitting serological testing devices for virus-specific human immunoglobulin class M (IgM). It is written with the assumption that these assays are qualitative or, in special instances, semi-quantitative. The term qualitative refers to the ability to determine the presence or absence of detectable antibody, i.e., "a positive test implies only that the assay signal exceeds the analytical threshold or a cut-off point which may have been set to give an arbitrary combination of sensitivity and specificity."[1] The term semi-quantitative refers to the ability to determine an increase and or decrease in detectable antibody, ie., "essentially qualitative assays with an additional option for a response range"[1] providing information on relative amounts of IgM antibody in single assays compared to standard levels or a dilution of the patient sample.

Quantitative assays are rare in virological serologic testing when the true definition of quantitation is employed: "Assays which generate a spectrum of signal responses which correlate with the concentration of the analyte of interest. If analyte preparations with known concentrations are available for calibration, the actual concentration of analyte can be determined."[1] Assays which use an accepted standard, such as World Health Organization (WHO) standards measured in international units per milliliter (IU/mL), could be considered quantitative if it is proven that the secondary calibrator and controls are linear with the WHO standard and results obtained from patient sera correlate between the new device and one of the methods used to establish the standard

across the full range of the assay. The method for reporting these results should be expressed in and correlate with the international unit terminology.

FDA has concerns regarding safety and effectiveness when IgM testing is utilized in the clinical laboratory. Laboratories which do not have a virology unit capable of isolating and identifying viruses, may depend on IgM testing for the diagnosis of viral infections. If an IgM test is not capable of distinguishing between a true positive and a false positive in the specific population tested, it could have adverse consequences for the patient. With the increased usage of antiviral therapy, false positive results could expose the patient to unnecessary therapy and false negative results may delay therapy or cause therapy to be withheld. Missed or falsely diagnosed infections could have adverse implications for pregnant women and the fetus. With these factors in mind, the manufacturers must test the device in such a manner to prove the device's safety and effectiveness in the population for which the device is to be used. The device must be labeled with these populations well defined.

**PRODUCT CODE(S):** According to specific virus

**CLASSIFICATION:** I/II/III (according to specific virus)

**PANEL:** MICROBIOLOGY (83)

**REVIEW REQUIRED:** Premarket Notification [510(k)] or; Premarket Approval (PMA) if there are issues of safety and effectiveness

## I. CLINICAL INDICATION / SIGNIFICANCE / INTENDED USE

Provide a concise discussion to include the following as appropriate. Support the discussion with key literature citations.

A. Background description of the virus, infectious process(es), and pathology.

B. Description of disease syndrome(s) associated with infection by the specific viral agent.

C. Salient concerns of the medical community, including relevant medical/societal issues that may impact on the review process or possibly the development of public policy.

D. Significance and clinical impact of false positive and false negative results.

E. Historical summary of all test methodologies used to detect IgM antibodies to the specific virus.

F. Description of epidemiology of the disease, prevalence rates within the population, and which population group(s) is(are) at risk for acquiring infection.

## II. DEVICE DESCRIPTION

Key issues in the review of a new device are the specific intended use (the analyte detected, the clinical utility, and the indications for use), the type of specimen collected, and the technology utilized. The following descriptive information must be included to adequately characterize the new in vitro device. Appropriate literature references that have been subjected to peer review and package insert(s) for (an)other similar commercial test assay(s) must be attached.

A. Intended Use.

Describe the intended use based on the technology/methodology employed in the device. The following questions should be addressed:

1. What patient populations should be tested?

2. What are the conditions and limitations for use of the device when used to diagnose or manage a specific syndrome?

3. What is the clinical utility of the device?

B. Detailed Principle of the Test Methodology.

Discuss the principle of the test methodology(ies). For new types of technologies, provide information to substantiate application of the methodology to the detection of specific antibodies. Cite literature references where appropriate. If available, furnish copies of appropriate scientific references for any recombinant nucleic acid, synthetic protein, or monoclonal antibody utilized. If scientific references are not available, discuss how the device antigen was determined to be representative of the native antigen. Include a complete description of the following device or procedural components when appropriate:

1. Any specimen pretreatment procedure.

2. Antigen utilized in the assay.

   a. If a native antigen, from what source was the antigen obtained.

   b. If a recombinant antigen or synthetic peptide (oligonucleotide), from what source was the native antigen derived and what nucleic acid or protein sequence was used to prepare the antigen.

   c. Provide a justification for the selection of the antigen.

3. Antisera used in the assay.

   a. Specify the species in which antisera were produced.

b. If a native antigen was used as the immunogen, identify the source from which the antigen was obtained.

c. If a recombinant or other artificial antigen was used as the immunogen, furnish the source of the native nucleic acid and provide the sequence for the derived recombinant.

d. Explain and provide documentation on how the specificity of the antisera was determined.

4. Enzymatic, fluorescent, or other substrate used to detect the antigen-antibody complexes.

5. Determination of the cut-off value(s) or endpoint(s) for the assay. Provide validation data as described in III.A.1.

6. Controls/calibrators included in the assay kit and what aspects of the procedure are verified.

   Quality control material should be representative of and correlate with the intended use and clinical utility of the device. In addition to the requirements listed below, the manufacturer should refer to the current CLIA '88 Quality Control Guidance.

   Qualitative assay:

   a. At a minimum, include in the device, make available, or recommend the use of two controls (positive and negative) in the same matrix as test specimens indicated for use with the assay. Controls should be within a statistically significant range of the cut-off. This range should be appropriate for the statistical method used to determine the cut-off. Provide documentation and justification for the value(s) selected for the control.

   b. Make recommendations for, and justify the frequency of, testing control material.

   Semi-quantitative assay:

   In addition to the controls and recommendations listed for a qualitative assay, include a control at the upper end of the linear range with a known expected value.

7. Any additional reagents or methods which contribute to the effectiveness of the device.

8. Collection and transport materials provided in the kit or recommended for use.

9. Software elements and dedicated instrumentation that are responsible for specimen handling and/or that are used to calculate assay results. See requirements for Minor Level of concern in Reviewer Guidance for Computer Controlled Medical Devices Undergoing 510(k) Review

(available from the Division of Small Manufacturers Assistance). Furnish the following for dedicated instrumentation and software elements:

a. Reference premarket notification [510(k)] submission number for any dedicated instrument.

b. Algorithms used to calculate results in either dedicated or non-dedicated instruments.

c. Mathematical curve-fitting method(s) used when results are calculated by instrument-related software.

C. Merits and Limitations of the Methodology(ies).

Discuss the merits and limitations/advantages/disadvantages of the test methodology(ies) of the new device.

D. Specimen Type(s).

List all specimen types (matrices) indicated for use with the device.

E. Collection, Transport, and Storage of Specimens.

If appropriate, discuss any relevant issue related to the appropriate collection transport, and storage of specimens to be tested with the device.

## III. SPECIFIC PERFORMANCE CHARACTERISTICS

~~The~~ FDA requests different types and amounts of data and statistical analyses to market in vitro diagnostic devices. The amount and types of data requested depend on the intended use and the technological characteristics of the new device. The data and statistical evaluation should be sufficient to determine if the device is safe and effective for all claimed specimen type(s). Additional data may be necessary to substantiate certain claims of intended use or clinical significance, and to validate use of a new technology.

Since viral IgM serologies are primarily used to replace isolation methods and for rapid diagnosis of acute infections, it must be shown the device will detect a **true disease** state (infection). Appropriate clinical studies must be done to show the relationship of results to the disease state (qualitative = **DISEASE PRESENCE** or **DISEASE ABSENCE**, semi-quantitative = early, acute, waning infection).

Clearly document all protocols for in vitro testing. Present test data with analyses and conclusions. Summarize results and include explanations for unexpected results and any additional testing performed. When appropriate, charts (scattergrams, histograms, receiver operator curves (ROC), etc.) may be used as part of analyses and conclusions. Furnish raw laboratory data with quality control results for each day performance testing was done from each site of testing.

Submission of the following data is required to determine the device's ability to detect viral-specific IgM antibodies:

A. Analytical Laboratory Studies.

1. Validation of Cut-off and/or Calibration Curve.

   Describe the rationale for determination of the assay CO(s). Furnish descriptive information and laboratory data to show how the cut-off (CO) (distinction between positivity and negativity) was determined for the assay. The number of specimens included in each disease syndrome should be statistically significant in relation to the intended use and proposed clinical utility of the device. Furnish documentation as to how the specimens were characterized.

   If a vaccine exists for the target virus, documented vaccine recipients may be used for this study. If plasma, rather than serum, is used in any of the testing, these specimens should be identified.

   a. Define the population(s) used, including the following information:

      (1) geographical area(s) from which the population was derived;

      (2) number of samples comprising the population, with samples summarized according to gender and age groups in decades;

      (3) graphical (e.g., histogram, scattergram, etc.) representation of population characteristics.

   b. Define the statistical method used to determine the CO.

   c. Present an ROC analysis of CO selection and other graphical representations as appropriate.

   d. Define the basis for the equivocal zone (if applicable).

   e. If the assay is semi-quantitative, perform appropriate studies to show the relationship of results to the stage of infection (e.g., early, acute, waning infection) for which value ranges have been established. For each stage of infection, present results from a minimum of 10 patients.

   f. For devices which determine interpolated values (e.g., ELISAs), verify the accuracy and working range of the calibration curve used by serially diluting patient samples.

2. Establish the prevalence of the analyte in a normal population (healthy individuals without symptoms) using the specified CO.

a. With the new device assay a statistically significant number of specimens which are representative of the intended use, clinical utility, and matrix of the specimens.

b. Furnish results from the new device and results from a pre-existing device or a published study from the same general population performed with a device of the same methodology.

c. Summarize the distribution of the population according to age groups (in decades), geographical area, and the number of positive, negative, and equivocal results.

3. Validation of Assay Specificity.

a. Perform cross-reactivity studies with sera containing relatively high levels of IgM antibody to other viruses which are in the same class and also sera containing IgM antibodies to members of the herpesvirus family which are known to cause infection in humans (other than monkey B virus), influenza, and paramyxovirus families. Furnish the levels (e.g.,"titers") tested and describe the methods (i.e., legally marketed devices) used to determine the amount of viral-specific IgM.

b. If the antigen utilized in the device is a recombinant, test sera containing antibodies against the organism in which the vectors were induced.

c. If the antisera utilized in the device were produced by using recombinant(s) as the immunogen(s), test sera containing antibodies against the organism in which the vectors were induced.

4. Interference Studies.

Any potentially cross-reacting or interfering substances or conditions potentially encountered in specific specimen types or conditions should be tested using the assay system, e.g., storage conditions, hemolysis, lipemia, freeze-thawing, etc.

a. Verify that recommended specimen storage conditions are compatible with the assay. State the optimal conditions based on specimen storage stability studies. Both the possibility of false positivity and false negativity due to storage conditions should be evaluated. Can the specimen be frozen and thawed one or more times without affecting the qualitative or semi-quantitative detection of the analyte?

b. If the use of plasma is claimed, a study with each anticoagulant must be performed to show the anticoagulant does not interfere with the assay.

(1) For each anticoagulant, test 10 concurrently obtained serum and plasma specimens which have positive reactivity near the CO. These specimens may be artificially prepared as long as the matrix is maintained and it is noted in the submission that the samples were artificially created.

(2) For each anticoagulant, test 10 concurrently obtained non-reactive serum and plasma specimens.

c. If heating of the specimen is claimed not to interfere with the assay:

(1) Test 10 specimens with positive reactivity near the CO, for each matrix type claimed, heated and not heated. These specimens may be artificially prepared as long as the matrix is maintained and it is noted in the submission that the samples were artificially created.

(2) Test 10 specimens which are non-reactive for each matrix claimed, heated and not heated.

d. Determine the lack of interference/cross-reactivity from rheumatoid factor (RF), anti-nuclear antibodies (ANA), viral-specific IgG, heterophilic antibodies, etc. The following testing is recommended:

(1) If an absorbent is used to remove interference of RF, ANA, or viral-specific IgG, document the amount of total IgG removed in mg/dL or mg/sample volume. State this value in the LIMITATIONS section of the package insert.

(2) Assay five specimens, in all matrices claimed, containing various levels of viral-specific IgG in viral-specific IgM samples at or near the CO (these samples may be artificial). Test these samples before and after removal of total IgG. Furnish data for both studies.

(3) Assay five specimens, in all matrices claimed, containing high levels of RF and ANA with no viral-specific IgM to show the lack of or the amount of interference.

(4) Furnish the levels tested and describe the methods used to determine the amount of viral-specific IgG, RF, and ANA present. These methods should be legally marketed devices.

(5) Prove IgM class antibody is detected by assaying a minimum of 10 sera containing viral-specific IgM antibody. Destroy the IgM antibody (e.g. 2-mercaptoethanol or dithiothreitol (Cleland's reagent)), then retest to show specimens are now non-reactive.

5. If semi-quantitation is claimed, the linearity of the device must be proven over the claimed range. For each matrix claimed, assay in triplicate a minimum of five patient samples and three to five controls, diluted independently in duplicate in the appropriate matrix [5 patients specimens + 5 controls x 2 independent dilutions x 3 separate runs]. Perform regression coefficient analysis on this data and furnish the y-intercept, slope, and $r^2$ obtained.

Present a scattergram of the mean results with 95% confidence intervals delineated in the Performance Characteristics section of the package insert.

6. Reproducibility

The National Committee for Clinical Laboratory Standards (NCCLS) recommends an analysis of variance experiment that permits estimation of within-run and total standard deviations (SD).[2] See the NCCLS Guideline for recommended data collection formats and calculations. Perform separate calculations for each specimen tested for within-run and total precision.

Test six to ten patient blinded sera with varying degrees of reactivity plus controls supplied with device in triplicate on three different days at three laboratory sites (6-10 sera tested X 3 X 3 days X 3 sites). One testing site may be in-house. If additional matrices may be used, include three additional samples (non-reactive, low positive, and positive) for each matrix.

For calculated endpoint tests (e.g., ELISAs), present coefficients of variation for each set of values for with-in run and total precision, using absorbance values **and reporting units defined in the test procedure.**

For single endpoint assays, provide percentage of results negative, borderline/equivocal, or positive for each set of tests.

If dedicated instrumentation is used in specimen handling, or reading and interpreting results, use a different instrument (different serial number) at each site. If non-dedicated instruments are used, state specifications of instrument(s) used at each site.

B. Clinical Studies.

Clinical studies provide data on the ability of the system to accurately detect viral-specific IgM antibodies. It should be demonstrated that the performance of the device is safe and effective when used as an aid in the diagnosis of specific viral infections.

Provide the names and telephone numbers of principal investigators and sites at which testing was performed. Clinical testing should be performed by at least two (three for a PMA submission) independent investigators at separate independent locations not affiliated with the manufacturer. Identify the clinical laboratory sites by institutional name and address; include the name, title, and phone number of the responsible investigators at each site.

1. Demonstrate the clinical utility of the device.

Testing should be performed on an adequate number of positive and negative clinical specimens (following collection, storage, and testing procedure recommended in the package

insert) from a population consistent with the intended use of the device. Perform the following studies at the site of specimen collection:

a. Test specimens from individual patients who are in the acute stage of infection and from whom the virus has been isolated. Cell culture isolation and identification should be used, if possible, to identify these patients. If cell culture isolation for a particular virus is not available, other well-accepted methodologies may be used. Provide a justification and description of the method(s) used.

b. Test specimens from individual patients in the acute phase of infection and from whom other viruses have been isolated. List the virus isolated for each patient and date of isolation.

c. Test specimens from individual patients with disease syndromes from other than viral infections but with otherwise similar symptomatology or diagnostic criteria. List the disease for each patient and state whether the disease was defined by serologic results, clinical diagnosis, or isolation of an etiologic agent.

Calculate clinical sensitivity and specificity values from the above information.

Provide information on the prevalence of the other viruses from III.B.1.a. in the test population.

2. For 510(k) submissions, perform a comparison of the device to a legally marketed device. Ideally this study should be done at independent clinical laboratory site(s). All testing, with both devices, must be performed using testing, storage, collection procedures, and interpretative criteria as specified in the package inserts.

   a. Test sera, concurrently, with the new device and a device which is legally marketed in the U.S. Specimens in this group should be exclusively from patients submitted for the diagnosis of infection with the virus. Fifty percent (50%) of this group should be specimens that have not been frozen and are tested within limits established by the manufacturer in the labeling.

   b. Discrepant results between the new device and comparison device may be clarified by re-testing at the site, by consensus testing with a second legally marketed device (at the manufacturer's site), or documentation of virus presence. If consensus testing is performed, it is recommended that a percentage of specimens, for which the results with the new device and the legally marketed device are in agreement, also be tested with the third assay.

   Calculate agreement, relative sensitivity, and relative specificity from both a. and b. above. Include these data in the Performance Characteristics Section of the package insert.

3. As an option, results obtained from a well-documented panel of sera from CDC or another source may also be tested. This study may be conducted at the manufacturer's facility or at a clinical laboratory site. If a manufacturer's sera panel is used, furnish the criteria for defining the diagnosis and/or disease stage of the patients from whom samples were obtained.

## IV. LABELING CONSIDERATIONS

The following are additional details for some of the points in the statute [502(f)(1)] and regulations [21 CFR § 809.10(b)].

A. The Intended Use Statement

The intended use statement should be a concise description of the essential information about the product. It should communicate the following information:

1. Test methodology.

2. That the assay detects viral-specific IgM class antibody.

3. Indications for use.

   These conditions for use may be addressed further in either the Summary and Explanation, Limitations, or Performance Characteristics section of the package insert.

4. What specimen source(s) may be tested.

5. Whether the assay is to be used only with special instrumentation.

   Example: The ____ Assay is for the qualitative determination of IgM antibodies to XXXXX in human serum using ____(methodology) as an aid in the diagnosis of recent XXXXX infection (primary or reactivation/reinfection)

B. Warnings and Precautions

Previously frozen specimens should be thoroughly mixed after thawing prior to testing.

C. Specimen Collection and Handling

1. State the type of specimen to be collected, and the types of collection devices which may be used.

2. State the conditions for patient preparation, e.g., timing of collection, order of collection, etc.

3. Provide adequate directions for sample collection and/or references for appropriate collection procedures, e.g., textbooks, journals, etc.

4. Identify interfering substances or conditions.

5. State the specimen storage conditions and stability periods.

D. Quality Control

Information provided in a Quality Control section should include the following information:

1. Provide recommendations for frequency of quality control.

2. Provide directions for interpretation of the results of quality control samples.

3. The Quality Control section should conclude with a statement similar to the following: "If controls do not behave as expected, results are invalid and patient results should not be reported".

4. Refer to current CLIA '88 FDA Quality Control Guidance for additional information.

E. Expected Values

1. Reference expected prevalence of viral-specific IgM antibodies in different populations.

2. Indicate that prevalence may vary depending on geographical location, age, gender of population studied, type of test employed, specimen collection and handling procedures, clinical and epidemiological history of individual patients, etc.

F. Results

1. Include appropriate explanation for interpretation of results.

2. Include terminology recommended for reporting patient results (not expressed simply as positive or negative). The methods for reporting non-reactive results could be "no antibody detected", "non-reactive at less than X dilution". The term "negative", used alone, should be avoided. Use a similar approach for results above the CO.

3. Provide recommended follow-up for equivocal/borderline results (e.g. "If specimen is equivocal on repeat redraw patient within one week and re-assay" or "If specimen is equivocal on repeat assay specimen by a different methodology").

G. Limitations of the Procedure

List important test limitations and all known contraindications, with references when appropriate. The following are examples of statements which may apply:

a. The test results should be used in conjunction with information available from the patient clinical evaluation and other available diagnostic procedures.

b. Drying of whole blood or serum onto filter paper inactivates, to varying extents, IgM class antibodies.[3]

c. Samples obtained too early during infection may not contain detectable levels of IgM antibody. If a viral infection is suspected, a second sample should be obtained 7-14 days later and tested concurrently with the first specimen to look for seroconversion or a significant rise in titer of viral-specific IgM or viral-specific IgG which is indicative of primary infection.

d. Falsely low or negative IgM results may occur due to competition by high titers of virus-specific IgG antibodies for antigen binding sites.[4] "This is a particular problem with sera from newborns with congenital viral infections, since their sera contain high levels of virus-specific IgG of maternal origin and relatively low levels of virus-specific IgM produced by the fetus."[5]

e. Because of the possibility of contamination of cord blood with maternal IgM, it is prudent to confirm positive viral IgM antibody results on cord blood samples by testing a follow-up specimen from the infant[5], preferably within the first five days of life.

f. "Since laboratory tests in pregnant women cannot reliably identify fetuses at risk of disease, screening for asymptomatic maternal infection coupled with termination of pregnancy cannot be recommended."[6]

g. Test results of specimens from immunosuppressed patients may be difficult to interpret.

h. Positive test results may not be valid in persons who have received blood transfusions or other blood products within the past several months.

i. Anti target-specific viral IgG antibodies may compete with the less avid target specific viral IgM antibodies, which would decrease the sensitivity of the assay.[3]

j. Anti target-specific viral IgM responses do not occur only in primary infections. It is recognized that an IgM response may occur in secondary and reactivated infections.[3]

k. The predictive value of a positive test decreases when prevalence decreases. Interpretation of positive results in a low risk patient population should be made with caution. Usefulness of

this test has only been established in testing sera from patients with _____ (state patient population(s)).

1. Specific IgM antibodies are usually detected in patients with recent primary infection, but they may be found in patients with reactivated or secondary infections, and they are sometimes found in patients with no other detectable evidence of recent infection.

H. Performance Characteristics:

Summarize the data upon which the performance characteristics are based, e.g., clinical sensitivity and specificity compared to clinical diagnosis; also include summary of reproducibility studies. Positive and negative predictive values should be based on specific populations sampled for each disease syndrome. State the prevalence at each testing site. Also show the effect of prevalence on positive and negative predictive values in different test populations.

1. Present cross-reactivity studies in a tabular form, indicating negative, positive, and borderline/equivocal/indeterminate results for each condition/disease.

2. Summarize within-run and total reproducibility.

3. Present data from clinical studies, using separate categories for different patient categories. Clearly display all borderline/equivocal/indeterminate results. Discrepancies between test and clinical diagnosis may be discussed and presented as footnotes.

4. Present data from comparison studies. Clearly display all borderline/equivocal/indeterminate results. Discrepancies between test and reference method may be resolved and presented as footnotes or in a separate table.

## V. BIBLIOGRAPHY

1. National Committee for Clinical Laboratory Standards. Specifications for immunological testing for infectious diseases, proposed guideline. 1991. Order code I/LA 18-P.

2. National Committee for Clinical Laboratory Standards. Evaluation of precision performance of clinical chemistry devices, tentative guideline. 1991. Order Code EP5-T2.

3. Schmidt NJ, Emmons RW. General principles of laboratory diagnostic methods for viral, rickettsial and chlamydial infections. In: Schmidt NJ, Emmons, RW, eds, Diagnostic procedures for viral, rickettsial, and chlamydial infections, 6th edition. Washington, DC: American Public Health Assoc. 1989:1-35.

4. Chernesky MA, Ray CG, Smith TF. Laboratory diagnosis of viral infections. Cumitech 15, pg. 10. Cumulative Techniques and Procedures in Clinical Microbiology. ASM. Washington. March 1982.

5. Schmidt NJ. Update on class-specific viral antibody assays. Clinical Immunology Newsletter. June 1984;5(6):81-85.

6. Griffiths PD, Baboonian C, Butler D, Peckham C. Congenital and maternal cytomegalovirus infections in a London population. Br J Obstet Gynaecol. 1991;98(2):135-140.