Guidance for Industry and FDA Staff

Class II Special Controls Guidance Document: Factor V Leiden DNA Mutation Detection Systems

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Center for Devices and Radiological Health

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Preface

Public Comment

Written comments and suggestions may be submitted at any time for Agency consideration to the Division of Dockets Management, Food and Drug Administration, 5630 Fishers Lane, Room 1061, (HFA-305), Rockville, MD, 20852. Alternatively, electronic comments may be submitted to http://www.fda.gov/dockets/ecomments. Please identify your comments with the docket number listed in the notice of availability that publishes in the *Federal Register* announcing the availability of this guidance document. Comments may not be acted upon by the Agency until the document is next revised or updated.

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This guidance represents the Food and Drug Administration's (FDA's) current thinking on this topic. It does not create or confer any rights for or on any person and does not operate to bind FDA or the public. You can use an alternative approach if the approach satisfies the requirements of the applicable statutes and regulations. If you want to discuss an alternative approach, contact the FDA staff responsible for implementing this guidance. If you cannot identify the appropriate FDA staff, call the appropriate number listed on the title page of this guidance.

1. Introduction

This document was developed as a special control to support the classification of Factor V Leiden (FVL) DNA mutation detection systems into class II (special controls). This document does not address submissions for genetic tests that are class III (high risk), although many of the analytical principles elaborated here may apply to those submissions. Genetic tests for the FVL mutation are devices that may consist of different reagents and instruments depending on the final configuration of the device, which may include polymerase chain reaction (PCR) primers, hybridization matrices, thermal cyclers, imagers, and software packages. The devices are intended as an aid in diagnosis in individuals with suspected thrombophilia. This document does not address tests intended for stand-alone diagnostic purposes, prenatal screening, or mass screening of populations.

This guidance is issued in conjunction with a *Federal Register* notice announcing the classification of Factor V Leiden mutation detection systems. Following the effective date of the final rule classifying the device, any firm submitting a 510(k) premarket notification for a Factor V Leiden mutation detection system will need to address the issues covered in the special controls guidance document. However, the firm need only show that its device meets the recommendations of the guidance or in some other way provides equivalent assurances of safety and effectiveness.

FDA's guidance documents, including this guidance, do not establish legally enforceable responsibilities. Instead, guidance documents describe the Agency's current thinking on a topic and should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in Agency guidances means that something is suggested or recommended, but not required.

The Least Burdensome Approach

The issues identified in this guidance document represent those that we believe should be addressed before your device can be marketed. In developing the guidance, we carefully considered the relevant statutory criteria for Agency decision-making. We also considered the burden that may be incurred in your attempt to comply with the guidance and address the issues we have identified. We believe that we have considered the least burdensome approach to addressing the issues presented in the guidance document. If, however, you believe that there is a less burdensome way to address the issues, you should follow the procedures outlined in the "A Suggested Approach to Resolving Least Burdensome Issues" document. It is available on our Center web page at http://www.fda.gov/cdrh/modact/leastburdensome.html.

2. Background

FDA believes that special controls, when combined with the general controls, will be sufficient to provide a reasonable assurance of the safety and effectiveness of Factor V Leiden DNA mutation detection systems. A manufacturer who intends to market a device of this generic type should (1) conform to the general controls of the Federal Food, Drug, and Cosmetic Act (the Act), including the premarket notification requirements described in 21 CFR 807, Subpart E, (2) address the specific risks to health associated with Factor V Leiden DNA mutation detection systems identified in this guidance and, (3) obtain a substantial equivalence determination from FDA before marketing the device.

This guidance document identifies the classification regulation and product code for Factor V Leiden DNA mutation detection systems. (Refer to Section 4 – **Scope)**. In addition, other sections of this guidance document list the risks to health identified by FDA and describe measures that, if followed by manufacturers and combined with the general controls, will generally address the risks associated with these assays and lead to timely premarket notification [510(k)] review and clearance. This document supplements other FDA documents regarding the specific content requirements of a premarket notification submission. You should also refer to 21 CFR 807.87 and other FDA documents on this topic, such as the **510(k) Manual - Premarket Notification: 510(k) - Regulatory Requirements for Medical Devices**, http://www.fda.gov/cdrh/manual/510kprt1.html.

Under "**The New 510(k) Paradigm - Alternate Approaches to Demonstrating Substantial Equivalence in Premarket Notifications; Final Guidance**[1]," a manufacturer may submit a Traditional 510(k) or has the option of submitting either an Abbreviated 510(k) or a Special 510(k). FDA believes an Abbreviated 510(k) provides the least burdensome means of demonstrating substantial equivalence for a new device, particularly once FDA has issued a guidance document. Manufacturers considering modifications to their own cleared devices may lessen the regulatory burden by submitting a Special 510(k).

3. The Content and Format of an Abbreviated 510(k) Submission

An Abbreviated 510(k) submission must include the required elements identified in 21 CFR 807.87, including the proposed labeling for the device sufficient to describe the device, its intended use, and the directions for its use (21 CFR 807.87(e)). In an Abbreviated 510(k), FDA may consider the contents of a summary report to be appropriate supporting data within the meaning of 21 CFR 807.87(f) or (g); therefore, we recommend that you include a summary report. The report should describe how this guidance document was used during device development and testing and should briefly describe the methods or tests used and a summary of the test data or description of the acceptance criteria applied to address the risks identified in this document, as well as any additional risks specific to your device. This section suggests information to fulfill some of the requirements of 807.87, as well as some other items that we recommend you include in an Abbreviated 510(k).

Coversheet

The coversheet should prominently identify the submission as an Abbreviated 510(k) and cite the title of this guidance document.

Proposed labeling

Proposed labeling should be sufficient to describe the device, its intended use, and the directions for its use. (Refer to Section 8 for specific information that should be included in the proposed labeling for devices of the types covered by this guidance document.)

Summary report

We recommend that the summary report contain a:

- Description of the device and its intended use. We recommend that the description include a complete discussion of the performance specifications and, when appropriate, detailed, labeled drawings of the device. You should also submit an "indications for use" enclosure.[2]

[1] http://www.fda.gov/cdrh/ode/parad510.html

[2] Refer to http://www.fda.gov/cdrh/ode/indicate.html for the recommended format.

- Description of device design requirements.

- Identification of the Risk Analysis method(s) used to assess the risk profile in general, as well as the specific device's design and the results of this analysis. (Refer to Section 5 for the risks to health generally associated with the use of this device.)

- Discussion of the device characteristics that address the risks identified in this guidance document, as well as any additional risks identified in your risk analysis.

- Brief description of the test method(s) you have used or intend to use to address each performance aspect identified in Sections 6 and 7 of this guidance document. If you follow a suggested test method, you may cite the method rather than describing it. If you modify a suggested test method, you may cite the method but should provide sufficient information to explain the nature of and reason for the modification. For each test, you may either (1) briefly present the data resulting from the test in clear and concise form, such as a table, **or** (2) describe the acceptance criteria that you will apply to your test results.[3] (See also 21 CFR 820.30, Subpart C - Design Controls for the Quality System Regulation.)

- If any part of the device design or testing relies on a recognized standard, (1) a statement that testing will be conducted and meet specified acceptance criteria before the product is marketed, or (2) a declaration of conformity to the standard.[4] Please note that testing must be completed before submitting a declaration of conformity to a recognized standard. (Section 514(c)(1)(B), 21 U.S.C. 360d(c)(1)(B).) For more information, refer to the FDA guidance, **Use of Standards in Substantial Equivalence Determinations; Final Guidance for Industry and FDA**, http://www.fda.gov/cdrh/ode/guidance/1131.html.

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[3] If FDA makes a substantial equivalence determination based on acceptance criteria, the subject device should be tested and shown to meet these acceptance criteria before being introduced into interstate commerce. If the finished device does not meet the acceptance criteria and, thus, differs from the device described in the cleared 510(k), FDA recommends that submitters apply the same criteria used to assess modifications to legally marketed devices (21 CFR 807.81(a)(3)) to determine whether the finished device requires clearance of a new 510(k).

[4] See Required Elements for a Declaration of Conformity to a Recognized Standard (Screening Checklist for All Premarket Notification [510(K)] Submissions), http://www.fda.gov/cdrh/ode/reqrecstand.html.

If it is not clear how you have addressed the risks identified by FDA or additional risks identified through your risk analysis, we may request additional information about aspects of the device's performance characteristics. We may also request additional information if we need it to assess the adequacy of your acceptance criteria. (Under 21 CFR 807.87(l), we may request any additional information that is necessary to reach a determination regarding substantial equivalence.)

As an alternative to submitting an Abbreviated 510(k), you can submit a Traditional 510(k) that provides all of the information and data required under 21 CFR 807.87 and described in this guidance. A Traditional 510(k) should include all of your methods, data, acceptance criteria, and conclusions. Manufacturers considering modifications to their own cleared devices should consider submitting Special 510(k)s.

4. Scope

The scope of this document is limited to the following devices as described in 21 CFR 864.7280 (product code: NPQ).

21 CFR 864.7280 – Factor V Leiden DNA mutation detection systems Factor V Leiden mutation detection systems are devices that consist of different reagents and instruments, which include polymerase chain reaction (PCR) primers, hybridization matrices, thermal cyclers, imagers, and software packages. The detection of the Factor V Leiden mutation is intended as an aid in the diagnosis of patients with suspected thrombophilia.

5. Risks to Health

There are no known *direct* risks to patient health when tests are used as an aid to diagnosis. However, failure of the test to perform as indicated or error in interpretation of results may lead to improper medical management of patients with clotting disorders. A false negative interpretation could lead to under-management of the patient, with increased risk of future thrombotic events. A false positive result could lead to inappropriate treatment and alteration of present and future drug selection and treatment.

In the table below, FDA has identified the risks to health generally associated with the use of FVL tests as addressed in this document. The measures recommended to mitigate these identified risks are given in this guidance document, as shown in the table below. We recommend that you conduct a risk analysis, before submitting your premarket notification, to identify any other risks specific to your device. The premarket notification should describe the risk analysis method. If you elect to use an alternative approach to address a particular risk identified in this document, or have identified risks additional to those in this document, you should provide sufficient detail to support the approach you have used to address that risk.

Identified risk	**Recommended mitigation measures**
Improper medical management	Sections 6, 7, and 8
Misdiagnosis, improper treatment and drug selection and dosing	Sections 6, 7, and 8

6. Performance Characteristics

Genetic testing for the FVL mutation has been performed for a number of years, but has chiefly been confined to in-house developed testing. New technological developments and increased demand for genetic tests have led to the demand for marketing of devices capable of detecting the FVL mutation and associated disease-specific polymorphisms. This guidance document makes recommendations for manufacturers who are preparing submissions for tests to be marketed in interstate commerce, without specific regard to the technology used to detect the mutations or polymorphisms (see Section 7).

The following are areas that we believe should be addressed in the preparation of a submission for a device incorporating technology designed to detect the FVL mutation.

A. Intended Use and Indications for Use of a Test or Device

The intended use should specify what the test is intended to measure, why it is measured, and should specify populations to which the test is targeted, where appropriate. In this case, the intended use should clearly specify the FVL mutation. References to professional society recommendations are acceptable.

Some tests may have multiple intended uses. FDA recommends a separate application for each intended use that requires unique and separate supporting studies. You should consult the appropriate review divisions in FDA for advice on submitting tests with multiple intended uses.

B. Preanalytical Factors

Consideration of preanalytical factors is essential for high-quality genetic tests. Manufacturers intending to provide reagents for extraction and preparation of DNA for testing should validate each step in the preanalytical process for its effects on reproducibility, robustness, and stability of product. Manufacturers who do not intend to provide these

reagents in their kits should provide specifications for assessing the quality of the assay input DNA and other required reagents, so that the user can select appropriate reagents. Justification for specifications given should also be provided in the submission.

Other preanalytical factors to be considered are the source of DNA (blood, PBMC, buccal swab, etc.), validation of its suitability for extraction at an acceptable quality level (e.g., heparin-preserved vs. EDTA-preserved blood, stored vs. fresh sample), and validation of acceptable storage conditions and stability for the sample and the extracted product.

C. Analytical Factors

Samples

You should perform analytical studies that demonstrate that the device detects the mutations it claims to detect, and does not detect mutations when none are present. The sample type used to perform analytical studies should include patient samples to show that the device will perform as claimed when patients are tested and that the entire process is controlled. FDA recognizes that in some situations, the FVL mutation may not be present in the sampled population at high enough rate to ensure significance of test results. In these cases, you may use archived and/or retrospective samples in addition to prospective patient samples in order to expand the number and proportion of mutations available. In certain cases, you may use "artificial" samples in which DNA containing the mutation has been added at a level simulating that which would be found in a natural sample. You should choose the sample number to achieve a stated statistical confidence that the test performs as expected. For all samples, you should give starting material, extraction method, concentration, and purity. Both heterozygous and homozygous mutant samples are acceptable; homozygous samples may decrease the overall number of samples needed for testing if chromosome count is used as a metric.

Mutations

The known FVL mutation is G1691A. Additional rare mutations in Factor V are known at A1692C, G1689A and A1696G. We recommend that you assess the possibility of the rare Factor V mutations to give false FVL results and report this as a limitation, if applicable.

Test Methodology

FDA recommends that you describe in detail the methodology that will be used to detect mutations. If sample extraction matrices are provided, you should describe this methodology as well. Illustrations or photographs of non-standard equipment or methods can be helpful in understanding novel methodologies.

Controls and Calibrators

FVL molecular mutation testing should include both positive and negative controls. For different technologies, these controls may differ, but the user should be able to determine if critical reactions have proceeded properly and without contamination or cross-hybridization. Controls should approximate sample DNA concentration in order to adequately exercise the system.

We recommend that you implement the calibration of systems where it can aid in generation and interpretation of results. Depending on the technology selected, calibration may or may not be critical for proper use of the test.

Analytical Sensitivity

You should validate the analytical sensitivity of your test, i.e., what the minimum amount of input nucleic acid is, and approximate the amount of sample required to generate this minimum input. If the assay has an upper, or saturation, limit, you should also validate and state this in labeling.

Interference

You should assess interference in FVL mutation detection from any input into the system. Some common known interferences may occur from extraction technique, original sample matrix, excess or inadequate Mg^{2+} concentration, etc. You should investigate other potential interferences and describe them in labeling, if necessary.

Reproducibility/Robustness

FDA recommends manufacturers fully examine the reproducibility and robustness of their device. NCCLS EP-5A describes an acceptable reproducibility testing plan. FDA also recommends that 3 or more sites with varying molecular experience be used to test reproducibility of panels of mutations and wild-type sequences, using the procedure that you will describe in final device labeling. If extraction reagents are not included in the test kit, each site should use and validate their own extraction procedures and demonstrate that the resulting input material (e.g., DNA) meets manufacturer-supplied specifications. Preferably, you should include multiple operators, multiple product lots, and instruments, if these are part of the device, to adequately test the expected performance of the system. If training will be necessary for users to perform the test once it is marketed, you should provide information on operator training. If such training is not expected to be necessary for users, you should not provide additional training (other than the package insert) at the testing sites.

Instrumentation

Instrumentation that is specific and dedicated to the device should be analyzed according to the "*Guidance for FDA Reviewers and Industry: Guidance for the Content of Premarket Submissions for Software Contained in Medical Devices"* document available at http://www.fda.gov/cdrh/ode/software.pdf. You should include a copy of instrumentation manuals for specified instrumentation. If general purpose instrumentation is to be used, you should provide specifications for the required instruments to be used in labeling.

Clinical Testing

Prospective clinical testing to determine clinical validity and utility will generally not be necessary for the FVL mutation and polymorphisms. You should use clinical samples, however, as often as practicable in analytical testing, in order to demonstrate that correct results can be obtained from clinical material. Because the mutation is well-known, and high quality clinical literature and professional society recommendations supporting clinical utility of mutations and polymorphisms tested exists, these may be substituted for prospective clinical studies, providing that there is some bridge in this literature to an accepted reference method. You should succinctly summarize these materials and provide references to the agency in the submission. If clinical testing is performed, you should select patients prospectively in order to maximize the number of mutations detected, e.g., from patients referred for thromboses, etc. You should describe inclusion and exclusion criteria, and these should conform to the intended use population.

Reporting

FDA recommends that examples of test reports as would be supplied to the ordering personnel be provided to the agency. Reports should be consistent with current recommendations of genetics professional societies and should contain adequate interpretation guidelines for the use of the ordering physician/counselor. FDA recommends that FVL mutations be reported as "present" or "absent" rather than "normal," "wild-type," or "mutant."

7. Method comparison

The abundance of technologies that could be used to detect the FVL mutation raises the possibility that assays may vary significantly in terms of methodology, instrumentation, and sample source, and make direct comparability difficult to assess. In order to facilitate product review, you should consider comparing the new assay to a reference method or "gold standard," for example,, bidirectional DNA sequencing, to define performance. We recommend that you validate and document the accuracy of the reference method used (e.g., percent correct sequence calls or "phred score"). You may then claim the sensitivity and specificity of your device. We recommend that you

describe the inclusion and exclusion criteria for all samples tested. Actual comparison of the new device performance to that of a previously cleared device is optional. However, if you wish to include this comparison in the new product labeling, you should also include a description of the study in the submission. You may perform discrepancy resolution for comparison studies, but you should use original unresolved results for all performance calculations unless the resolver is applied to all samples, to avoid bias.

8. Labeling

The premarket notification should include labeling in sufficient detail to satisfy the requirements of 21 CFR 807.87(e). The following suggestions are aimed at assisting you in preparing proposed labeling that satisfies the requirements of 21 CFR 807.87(e) and final labeling.[5]

Directions for use

You should provide clear and concise instructions that delineate the technological features of the specific device and how the device is to be used on patients. Instructions should encourage local/institutional training programs designed to familiarize users with the features of the device and how to use it in a safe and effective manner. Devices incorporating nucleic acid amplification should provide work-flow recommendations in labeling.

Quality Control

We recommend that you provide a description of quality control recommendations in the labeling.

Precautions for interpretations

We recommend that you address the limitations of your assay with statements in the labeling, such as:

- The presence of rare mutations in the Factor V gene may result in false positive reports for FVL.
- This test should not be used alone to diagnose thrombophilias. It is recommended that activated protein C (APC) testing be done alongside the genetic tests.

[5] Although final labeling is not required for 510(k) clearance, final labeling must comply with the requirements of 21 CFR 809.10 before an in vitro diagnostic device is introduced into interstate commerce.

Stability

We recommend that you assess the stability of your reagents and recommended samples and DNA inputs.

Performance

You should provide device performance in comparison to an accepted reference method or gold standard in the form of 2 X 2 tables, sensitivity and specificity percentages, or other illustrative examples. You should calculate sensitivity and specificity from all tested samples. Failed assays (e.g., inability to sequence sample) should be considered disagreements for the purposes of reporting performance characteristics.