AutoGenomics

INFINITI® Warfarin Assay

(2C9 & VKORC1 Multiplex Assay)

Directional Package Insert (DPI)

For *In Vitro* Diagnostic Use

CE

FOR EXPORT ONLY

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INTENDED USE

The INFINITI Warfarin Assay is an *in vitro* diagnostic test for the detection and genotyping of the *2 and *3 CYP2C9 genetic variants and the VKORC1 3673 (-1639) intronic variant in genomic deoxyribonucleic acid (DNA) obtained from EDTA-anticoagulated whole blood samples. The INFINITI Warfarin Assay is a qualitative assay for use in clinical laboratories upon prescription by the attending physician.

The INFINITI Warfarin Assay is indicated for use to identify individuals at risk for sensitivity to Warfarin.

BACKGROUND INFORMATION

Warfarin is a widely used anticoagulant prescribed for patients with venous thrombosis and pulmonary embolism, chronic atrial fibrillation and prosthetic heart valves. FDA estimates that two million persons start taking Warfarin in the United States every year to prevent blood clot[2].

Individual differences such as the intake of vitamin K, illness, age, gender, concurrent medication and body surface area, and genetic differences may affect a patient's response to warfarin.

Warfarin acts by interfering with the recycling of vitamin K, which leads to reduced activation of several clotting factors. Two genes affecting the pharmacokinetic and pharmacodynamic parameters of Warfarin are CYP2C9 (cytochrome P_{450} 2C9) and *VKORC1* (vitamin K epoxide reductase complex subunit 1). These two genes, together with other factors such as age, body surface area, and gender, partly explain the inter-individual variation in warfarin response[1]. Research suggests that some of the inter-patient variability in response to warfarin may depend on a patient's variants of the genes CYP2C9 and VKORC1[2].

CYP2C9

Warfarin is supplied as a racemic mixture of *R*- and *S*-isomers; the *S*-isomer has 3-5 times the potency of the *R*-form. After oral administration, free Warfarin is taken up by the liver where it is metabolized by cytochrome $P_{450.}$ The *S*- isomer is metabolized by cytochrome $P_{450,}$ subfamily IIc, polypeptide 9 protein (CYP2C9) and eventually excreted in the bile[3].

CYP2C9*2 and CYP2C9*3 are among the CYP2C9 variants that have been shown to exhibit decreased enzymatic activity[5,9]. The *2/*2 homozygous variant leads to a reduction to approximately 12% of wild-type CYP2C9 activity and the *3/*3 homozygous variant has a <5% of wild type CYP2C9 activity[4]. In a retrospective association study, individuals with one or both genetic variants were reported to require a lower maintenance dose of Warfarin compared with patients without these variations[5,7,8].

VKORC1

Warfarin's anticoagulant activity results from inhibition of vitamin K epoxide reductase important for the activation of various coagulation factors. Studies suggest that single nucleotide polymorphisms (SNPs) in the C1subunit of the vitamin K 2,3 epoxide reductase complex (VKORC1) may influence Warfarin sensitivity.

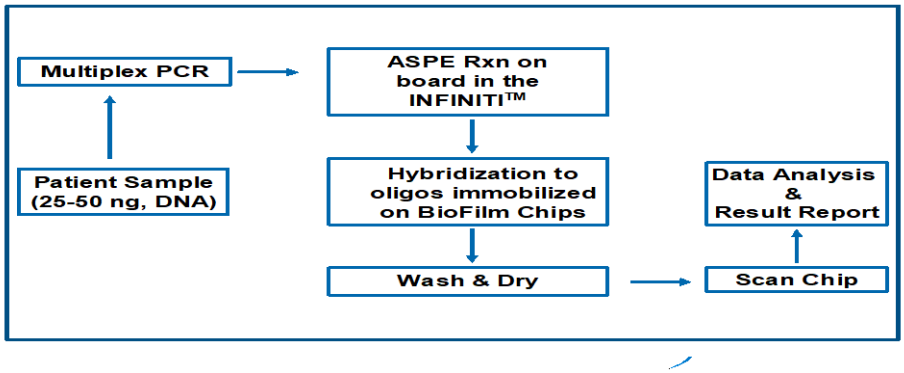
Polymorphisms of the VKORC1 gene tend to occur in haplotype blocks with particular combinations of polymorphisms that exhibit strong linkage disequilibrium. The identification of one member of a haplotype block is often predictive for the overall haplotype.[7] The identification of the VKORC1 3673 (-1639) polymorphism, which is common to the H1 and H2 haplotypes, could be predictive of an individual's response to Warfarin.

TEST PRINCIPLE / ASSAY OVERVIEW

The INFINITI Warfarin Assay is designed to simultaneously detect the *2 and *3 CYP4502C9 genetic variants and the VKORC1 3673 (-1639) intronic variant. The assay protocol is based on the following major processes:

(a) DNA extraction.
(b) PCR amplification of purified DNA from human genomic DNA.
(c) Labeling of the amplified product (allele specific primer extension).
(d) Hybridization of the labeled amplified product to a microarray by signature Tag/Capture probe hybridization under isothermal conditions.
(e) Scanning of the microarray.
(f) Signal detection and analysis [determination of the 2C9*2, 2C9*3 and VKORC1 3673 (-1639) genotypes].

Steps (c) through (f) are automated by the INFINITI PLUS Analyzer. A schematic overview of the assay is shown below.



DEVICE DESCRIPTION

The INFINITI Warfarin Assay, an *in vitro* diagnostic device, utilizes AutoGenomics' proprietary film-based microarray technology combined with process automation, reagent management and software technology for the detection and genotyping of the 2C9*2, 2C9*3, and VKORC1 3673 (-1639) mutations from human whole peripheral blood samples.

The INFINITI Warfarin Assay is comprised of the BioFilmChip® Microarray, and the Intellipac® Reagent Module.

The **BioFilmChip Microarray** consists of a polyester film coated with proprietary multi-layer components designed for DNA analysis. The layers have been designed to provide a versatile surface to enhance test performance. The microarrays are designed to be assay specific. The INFINITI Warfarin Assay uses a microarray chip (R-Chip) which contains unused Capture Probes which could potentially be used for certain specific assays. Therefore, multiple assays can be developed using the same microarray.

The **Intellipac Reagent Module** which acts as a communication link contains four reservoirs that house the test reagents and has an integrated memory chip. Information on the reagent such as lot number, expiration date and volume usage is archived in the memory chip.

The INFINITI Warfarin Assay should be run using the AutoGenomics **INFINITI PLUS Analyzer**. The Analyzer is an instrument used for clinical multiplex systems intended to measure and sort multiple signals from a clinical sample. The Analyzer is designed to measure fluorescence signals of labeled DNA target hybridized to BioFilmChip microarrays. The Analyzer automates the 2C9 and VKORC1 assays and integrates all the discrete processes of sample (PCR amplicon) handling, reagent management, hybridization, detection, and results analysis. The assays are

processed automatically and the spots are read by the built-in confocal microscope. Results are analyzed and presented as genotype calls.

Instructions on how to use the Analyzer is provided in the Operator's Manuals.

The INFINITI PLUS Analyzer is CE marked.

WARNINGS AND PRECAUTIONS

Handling Requirements

- **For *in vitro* diagnostic use. To be used by qualified laboratory personnel.**
- This test is to be used only with whole blood collected in EDTA. Do not freeze / thaw blood samples. Specimens should be assayed as soon as possible.
- All patient specimens are potentially hazardous and care should be taken when handling materials of human origin. No test method can offer complete assurance that HCV, HIV or other infectious agents are absent.
  Follow the CLSI Guidelines (Molecular Diagnostics Methods for Infectious Diseases; Approved Guidelines; MM3-A).
- Upon receipt of samples, visually inspect sample condition. Specifically, look for abnormal signs that indicate that sample integrity has been compromised (e.g., evaporation, decrease in volume, precipitation, spills, discoloration, sedimentation, separation, turbidity, etc.). If you observe or suspect any sample abnormality, do not perform any test.
- Samples should be handled with extreme caution to prevent contamination, spillage, sample mix-up. Sample containers should be labeled clearly to prevent mix-up.
- Store samples at the specified conditions.
- To minimize the risk of cross contamination, sample preparation, PCR reaction set up and PCR product analysis should be performed according to approved guidelines such as CLSI (Molecular Diagnostic Methods for Genetic Diseases: Approved Guideline).
- Do not pool / mix reagents from different lots.
- Do not use a kit or reagent past its expiration date.
- Store kits and reagents according to the product label.

Laboratory Procedures

- Follow normal precautions for handling laboratory reagents.
- Follow safe laboratory procedures: do not pipette by mouth; wear protective clothing (e.g., disposable gloves laboratory coats) and eye protection; do not eat, drink or smoke in the laboratory work areas; wash hands thoroughly after handling samples and reagents.

Waste Handling

- Dispose of unused reagents, specimens and waste according to applicable country, federal, state and local regulations.
- Safety data Sheets (SDS) are available upon request from AutoGenomics Customer Service.

Sample Preparation

- Refer to the safety instructions in the package insert provided with the DNA extraction kit used.
- The PCR product cannot be stored prior to loading it onto the microarray. Use immediately.

INFINITI PLUS Analyzer

- **Read the Operator's Manuals before operating the instrument.** Pay particular attention to "Notes".
- Follow the Caution and Safety Warning in the Operator's Manual.
- Refer to the Installation Requirements Section when installing the instrument.

- Refer to the Errors Section when errors are encountered while operating the instrument.
- Refer to the Help Section when problems are encountered.

STORAGE / STABILITY

BioFilmChip Microarray: 24 months Refrigerated (2 to 8°C)
Intellipac Reagent: 18 months Refrigerated (2 to 8°C)
Note: Remove the Intellipac from the Analyzer and store refrigerated as soon as possible. Do not use after Intellipac has been opened for four weeks.
Amplification Mix: 18 months Frozen (-30 to -15ºC)

Note: Specific product expiration date is printed on the product label.

SPECIMEN COLLECTION AND STABILITY

- Peripheral blood drawn in an EDTA (purple-top) tube.
- Do not freeze / thaw blood samples. Specimens should be assayed as soon as possible.

MATERIALS PROVIDED (EACH PACKAGE IS SUFFICIENT FOR 48 TESTS)

- AutoGenomics Product Number 03-1140-02 INFINITI Warfarin Assay BioFilmChip® Microarray Magazine: 4 magazines per package
- AutoGenomics Product Number 03-2140-02 INFINITI Warfarin Assay Intellipac® Reagent Management Module: 2 modules per package; 24 tests per module. Each Intellipac module contains:
  - 1.1ml ASPE Master Mix:
    - dNTPs
    - Labeled –dCTP
    - Allele Specific Primers
    - PCR Extension Buffer
  - 2.6ml Hybridization Buffer
    - SSC
    - EDTA
- AutoGenomics Product Number 03-3140-02 INFINITI Warfarin Assay Amplification Mix. Each package contains 2 x 500µl vials of AMP Mix containing:
  - dNTPs
  - dCTP
  - PCR Primer Mix
  - $MgCl_2$
  - PCR Reaction Buffer
- Product Number 12-0330-00: Wash buffer

REAGENTS REQUIRED BUT NOT PROVIDED BY AUTOGENOMICS

- DNA Extraction Kits - The INFINITI Warfarin Assay can detect 2C9*2, 2C9*3, and VKORC1 3673 (-1639) genetic variations using genomic DNA isolated from blood with sufficient purity, i.e., with the ratio of absorbance at 260nm to absorbance at 280nm of 1.7 to 2.0. Any DNA extraction method that meets this specification may be used. The INFINITI Warfarin Assay has been tested with several commercially available kits. The user can contact AutoGenomics for further information.
- Distilled Water (DNAse and RNAse free)
- Titanium Taq DNA Polymerase (see AutoGenomics Product catalog for recommended Titanium Taq DNA Polymerase and supplier)

EQUIPMENT
The following equipment is required but not provided with the assay reagents

- Pipettors
- Mini Centrifuge
- Pipette tips
- Microfuge tube Racks
- Thermocycler
- Vortex
- 0.2 ml thin wall tubes for PCR
- 1.5 ml microcentrifuge tubes
- 8-well Flat Strip Caps (Genesee Scientific, Catalog No. 22-623)
- AutoGenomics Product Number 11-0060-00: INFINITI Waste tray Stir Bars
- AutoGenomics Product Number 11-0020-00: INFINITI Waste Tray Liners
- AutoGenomics Product Number 11-0080-00: INFINITI Pipette Tips

- **FOR INFINITI PLUS Analyzer***:*
    - AutoGenomics Product Number 10-0020-99: INFINITI PLUS Analyzer
    - AutoGenomics Product Number 11-0100-00: 48-Well Plates
    - AutoGenomics Product Number 11-0110-00: 48 Well Plate Lid (reusable)
    - AutoGenomics Product Number 11-0070-00: Tip Barrel Plugs

ASSAY PROCEDURE
DNA Extraction
Follow the instructions provided with the DNA extraction kit used.
PCR Reaction
Note:

- Keep Taq DNA polymerase on ice.
- Completely thaw reagents on ice.
- Vortex the amplification mix tube for 2 to 5 seconds then centrifuge briefly to bring the contents to the bottom of the tube.
- To avoid contamination, a separate area is recommended for assembly of the PCR reaction. Decontaminate pipettes and all work surfaces with freshly prepared 10% bleach.
- Filter tips and gloves must be used when handling specimens and controls.
- The PCR product cannot be stored prior to testing. Use immediately.

Note*:*

- For the INFINITI PLUS Analyzer use the 48WP.

1. Prepare the PCR master mix.

Amplification mix	17.75 µl
Titanium Taq DNA polymerase	0.25 µl
Total	18.0 µl

Note: Calculate the amount of each reagent needed based on the number of reactions.

2. Gently vortex the PCR master mix then dispense 18 µl of master mix into wells of the well plate.

3. Add 2 µl of sample DNA (25 ng/µl) to each well.

PCR master mix	18.0 µl
Sample DNA	2.0 µl
Total volume of amplification reaction	20.0 µl

4. Place the well plate, sealed with 8-well flat strip caps, in a thermocycler and immediately commence the amplification reaction using the following program:

Multiplex 2 Tm PCR Conditions

Step No.	Temperature °C	Time	No. of Cycles
1	94	2 minutes	1
2a 2b 2c	94 60 - 54 (-0.5 °C/cycle) 72	20 seconds 30 seconds 30 seconds	12x
3	94 54 72	20 seconds 30 seconds 30 seconds	30x
4	72	2 minutes	1
5	4	Hold	

Note:

- After each cycle in step 2b the temperature is decreased by 0.5°C. When using an Eppendorf Mastercycler EP was used with the ramp rate set at 75%, the total cycling time was 1 hour and 40 minutes (± 10 min). If using other thermocycler models we recommend adjusting the ramp rate in order to obtain an equivalent total cycling time.
- Make sure there is no abnormal evaporation of the PCR product. After PCR is complete, visually inspect for any volume change. All PCR reaction volumes should be about 20 µl. Otherwise, do not proceed with the assay.

Sample Loading

1) Carefully remove the 8-well flat strip caps to avoid splashing.
2) Load the well plate in the appropriate orientation (with well A1 in the back left corner) into the Analyzer
   - **INFINITI PLUS Analyzer:** Load the assembled 48WP with a clean 48WP lid (see instructions in the INFINITI PLUS Analyzer Operator's Manual) (Catalog # 11-0110-00, reusable).
3) Load the following: assay specific magazines, Intellipac, INFINITI Static Free Pipette tips, and buffer.
   - **FOR INFINITI PLUS Analyzer***:*

     Follow the INFINITI PLUS Analyzer Operator's Manual for checking and replacing Wash buffer.

Operation of the Analyzers

Follow the instructions in the Operator's Manuals

INFINITI PLUS Analyzer Operator's Manual (Part Number EM-34041)

QUALITY CONTROL

- Maintain calibration of thermocycler according to manufacturer's specifications.
- Maintain calibration of INFINITI PLUS Analyzer according to AutoGenomics' specifications.
- Maintain calibration of pipettes according to manufacturer's specifications.

It is recommended that a positive (heterozygous and/or homozygous for the three genotypes) sample for each mutation, a negative control (a sample that does not contain the mutation of interest, i.e., a wild type sample); and a "Non Template Control" (Molecular Grade water 1XTE) be included with each test run. Please contact AutoGenomics for recommendations.

All quality control requirements and testing should be performed in conformance with local, state and/or federal regulations or accreditation requirements.

Note: The thermal cycler used should be regularly maintained and calibrated with an external temperature standard, according to the laboratory's regulatory and QC requirements.

LIMITATIONS

The results obtained from the INFINITI Warfarin Assay should be used and interpreted only in the context of the overall clinical diagnosis. AutoGenomics is not responsible for any clinical decisions that are taken.

In the VKORC1 gene, additional polymorphisms other than 3673G>A have been observed. Similarly, 2C9 variants other than the *2 and *3 have been observed. This test does not report those variations.

INTERPRETATION OF RESULTS

Results from the INFINITI Warfarin Assay are reported to the user as a genotype "call", indicating which genotype was detected in the sample, i.e., Wild Type, Homozygous, or Heterozygous for the 2C9*2, 2C9*3 and VKORC1 3673 (-1639) variants.

Important: All results with an "Indeterminate" or no call require a repeat assay.

A blank page with a message is displayed when the assay is not completed, and no genotype call is made. The assay has to be repeated. The message will indicate the reason why no genotype call was made. If errors occur during the assay, "Test Error" message (e.g., "low DNA") is shown. An Error Log is generated which identifies the problem. When an error message occurs, please refer to the Message Display section of the INFINITI PLUS Analyzer Operator's Manual. Depending on the error message/problem, the assay may have to be repeated only after the problem/error is corrected.

PERFORMANCE CHARACTERISTICS

Analytical Specificity

Studies related to specificity were conducted during assay development. PCR primer specificity was determined by amplicon size on a gel and sequencing the amplicon. ASP primer specificity was determined by the correct calls made by the assay using known genomic samples. Capture probe specificity was determined by hybridizing different oligos and demonstrating that only the correct oligo hybridizes to the known spot.

Limit of Detection (analytical sensitivity)

Serial dilutions (200, 100, 50, 25, 10, 1, 0.1ng DNA) were prepared from a known purified DNA sample. Each serial dilution was assayed three times using the INFINITI Warfarin Assay. The study established the minimum DNA concentration for the INFINITI System Assay for 2C9-VKORC1 to be 1ng DNA. The recommended DNA concentration for the INFINITI Warfarin Assay is 25ng/µl. The assay requires 2µl of DNA sample or the equivalent of 50ng per test.

In addition, the same study demonstrated that DNA concentrations of 100ng and 200ng, which were in excess of the recommended concentration (50ng), did not interfere with the INFINITI Warfarin Assay.

Percent Agreement vs. Bi-directional Sequencing
The results of the comparison studies conducted in three clinical sites comparing the INFINITI Warfarin Assay to bi-directional sequencing demonstrated:

98.0% agreement for 2C9*2 as compared with bi-directional sequencing on 1st run, 100% final
97.3% agreement for 2C9*3 as compared with bi-directional sequencing on 1st run, 99.3% final
98.0% agreement for VKORC1 3673 (-1639) as compared with bi-directional sequencing on 1st run, 100% final

A summary of the method comparison results is provided in Table1.

Table 1: Agreement between INFINITI Warfarin Assay and Bi-Directional DNA Sequencing

Genotype	Number Tested	Number of Correct Calls on First Run	Number of Incorrect Calls on First Run	Number of No Calls on First Run	Agreement First Run	Number of Correct Calls After Repeat Run	Number of Incorrect After repeat Run	Number of No Calls After Repeat Run	Agreement After repeat Run (Final)
2C9*2 *1/*2	35	34	0	1	97.1%	35	0	0	100%
2C9*2 *2/*2	2	2	0	0	100.0%	2	0	0	100%
2C9*2 *1/*1	113	111	0	2	98.2%	113	0	0	100%
Total for 2C9*2	150	147	0	3	98.0%	150	0	0	100%
2C9*3 *1/*3	19	19	0	0	100.0%	19	0	0	100%
2C9*3 *3/*3	1	1	0	0	100.0%	1	0	0	100%
2C9*3 *1/*1	130	126	1*	3	96.9%	129	1*	0	99.2%
Total for 2C9*3	150	146	1	3	97.3%	149	1	0	99.3%
VKORC1 3673 (-1639) GA	63	62	0	1	98.4%	63	0	0	100%
VKORC1 3673(-1639) AA	27	25	0	2	92.6%	27	0	0	100%
VKORC1 3673 (-1639) GG	60	60	0	0	100.0%	60	0	0	100%
Total for VKORC1 3673 (-1639)	150	147	0	3	98.0%	150	0	0	100%
Total for Assay	450	440	1*	9	97.8%	449	1*	0	99.8%

* Initial INFINITI results (*1/*1 for 2C9*2, ***1/*3 for 2C9*3*** and GG for VKORC1 3673) did not match bi-directional sequence results (*1/*1 for 2C9*2, ***1/*1 for 2C9*3*** and GG for VKORC1 3673). The same INFINITI results were obtained on repeat run. Reason unknown.

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Assay Inter-Laboratory Reproducibility

A three-site study was conducted to demonstrate the reproducibility of the INFINITI Warfarin Assay. The study involved three lots of the INFINITI Warfarin Assay. The sites ran identical samples comprised of seven genomic DNA samples and five whole blood samples. The sites were blinded to sample identity. Each site used a different DNA extraction method. At each site, each sample was run in duplicate per day / operator for six days. Three operators were required for each site. Results of the inter-laboratory reproducibility study are summarized in Table 2.

Table 2: Inter-Laboratory Reproducibility of the INFINITI Warfarin Assay by Genotype calls

Genotype	Samples Tested	Tests per Site	Site	# Genotype calls	First Run				Final (After Repeat Run)			
					Correct Calls	Incorrect Calls	No Calls	% Correct Calls	Correct Calls	Incorrect Calls	No Calls	% Correct Calls
2C9*2 *1/*2	3	36	1	36	36	0	0	100	36	0	0	100
			2	36	34	0	2[b]	94.4	36	0	0	100
			3	36	36	0	0	100	36	0	0	100
			total	108	106	0	2	98.15	108	0	0	100
2C9*2 *2/*2	1	12	1	12	12	0	0	100	12	0	0	100
			2	12	11	0	1[b]	91.7	12	0	0	100
			3	12	12	0	0	100	12	0	0	100
			total	36	35	0	1	97.22	36	0	0	100
2C9*2 *1/*1	8	96	1	96	95	0	1[a]	99.0	96	0	0	100
			2	96	96	0	0	100	96	0	0	100
			3	96	96	0	0	100	96	0	0	100
			total	288	287	0	1	99..65	288	0	0	100
Total for 2C9*2				432	428	0	4	99.07	432	0	0	100
2C9*3 *1/*3	2	24	1	24	24	0	0	100	24	0	0	100
			2	24	22	0	2[b]	91.7	24	0	0	100
			3	24	24	0	0	100	24	0	0	100
			total	72	70	0	2	97.22	72	0	0	100
2C9*3 *3/*3	2	24	1	24	24	0	0	100	24	0	0	100
			2	24	24	0	0	100	24	0	0	100
			3	24	23	0	1[c]	95.8	24	0	0	100
			total	72	71	0	1	98.61	72	0	0	100
2C9*3 *1/*1	8	96	1	96	95	0	1[a]	99.0	96	0	0	100
			2	96	95	0	1[b]	99.0	96	0	0	100
			3	96	96	0	0	100	96	0	0	100
			total	288	286	0	2	99.31	288	0	0	100
Total for 2C9*2				432	427	0	5	98.84	432	0	0	100
VKORC1 3673 (-1639) GA	5	60	1	60	60	0	0	100	60	0	0	100
			2	60	59	0	1[b]	98.3	60	0	0	100
			3	60	60	0	0	100	60	0	0	100
			total	180	179	0	1	99.44	180	0	0	100
VKORC1 3673 (-1639) AA	2	24	1	24	24	0	0	100	24	0	0	100
			2	24	22	0	2[b]	91.7	24	0	0	100
			3	24	24	0	0	100	24	0	0	100
			total	72	70	0	2	97.22	72	0	0	100
VKORC1 3673 (-1639) GG	5	60	1	60	59	0	1[a]	98.3	60	0	0	100
			2	60	60	0	0	100	60	0	0	100
			3	60	60	0	0	100	60	0	0	100
			total	180	179	0	1	99.44	180	0	0	100
Total for VKORC1 3673 (-1639)				432	428	0	4	99.07	432	0	0	100

* Results after one repeat of samples.

Drug Interference
Evaluation of potential interference from bilirubin, cholesterol, and heparin demonstrated that presence of these compounds in concentrations of 8mg/dl bilirubin, 70mg/dl cholesterol and 133v/dl heparin does not interfere with the INFINITI Warfarin Assay.

Sample Carry-Over
No sample carry-over was detected when 300ng of a positive sample was followed by 10ng of a second positive sample, and when 300ng of a positive sample was followed by a "No Template Control" or water. All genotype calls were 100% correct.

Assay Interference
Running the INFINITI Warfarin Assay and the INFINITI Assay for Factor II & Factor V on the same instrument did not affect the results of the assays, i.e., the INFINITI Warfarin Assay did not affect the results of the INFINITI Assay for Factor II & Factor V, and vice versa.

REFERENCES

1 Wadelius M et al "Pharmacogenetics of warfarin: current status and future challenges" The Pharmacogenomics Journal (2007) 7, 99-111

2 "FDA Approves Updated Warfarin (Coumadin) prescribing Information" FDA News, August 16, 2007

3 Gage B "Pharmacogenetics-Based Coumarin Therapy" Hematology 2006

4 Caldwell M et al "Evaluation of Genetic factors for Warfarin Dose prediction" Clinical medicine & Research (2007) Volume 5, Number 1:8-16

5 Cho H et al "Factors affecting the interindividual variability of warfarin dose requirement in adult Korean patients" Pharmacogenomics (2007) 8(40, 329-337

6 Lee CR, Goldstein JA, Peiper JA "Cytochrome P450 2C9 polymorphisms: a comprehensive review of the in-vitro and human data" Pharmacogenetics 2002; 12: 251-261-263

7 Rieder M et al "Effect of VKORC1 Haplotypes on Transcriptional Regulation and Warfarin Dose" The New England Journal of Medicine (June 2, 2005) 352;22

8 Takahashi H et al "Pharmacogenetics of CYP2C9 and interindividual variability in anticoagulant response to warfarin" The Pharmacogenomics Journal (2003) 3, 202-214

9 Takahashi H et al "Different contributions of polymorphisms in VKORC1 and CYP2C9 to intra- and inter-population differences in maintenance dose of warfarin in Japanese, Caucasians and African-Americans" Pharmacogenetics and Genomics 2006, Vol 16 No 2