SIEMENS Healthineers

Dade® Innovin®

Revision bar indicates update to previous version.

Intended Use

For use in prothrombin time (PT) determinations and prothrombin time-based assays in citrated human plasma.

Summary and Explanation

Dade® Innovin® Reagent is prepared from purified recombinant human tissue factor produced in *E. coli,* combined with synthetic phospholipids (thromboplastin)[1], calcium, buffers and stabilizers. The reagent initiates clotting via the extrinsic and common pathways in a global screening test, the prothrombin time (PT). Dade® Innovin® Reagent has three major applications based upon the PT[2]:

1. as a rapid screening test to detect single or combined deficiencies of the extrinsic coagulation system indicative of hereditary and acquired coagulation disorders, liver disease or vitamin K deficiency;
2. as a sensitive monitoring test for oral anticoagulant therapy with vitamin K antagonists; and
3. as an assay for specific coagulation factors.

Additionally, various photo-optical coagulation analyzers are able to derive the fibrinogen value from the determination of the prothrombin time.

Dade® Innovin® Reagent is manufactured using recombinant human tissue factor and synthetic phospholipids which do not contain any other clotting factors such as prothrombin, F VII and F X. Therefore, it is highly sensitive to factor deficiencies and oral anticoagulant-treated patient plasma samples. The sensitivity of Dade® Innovin® Reagent is very similar to the WHO human brain reference thromboplastin[3]. Dade® Innovin® Reagent is insensitive to therapeutic levels of heparin. The high sensitivity of Dade® Innovin® Reagent to coagulation factors and its insensitivity to therapeutic heparin make it beneficial in monitoring oral anticoagulant therapy with vitamin K antagonists[3]. In addition, its high sensitivity (i.e. the responsiveness of the reagent to moderately depleted factor activity) allows differentiation of abnormal plasmas, even in the mildly pathological range.

Principle of the Method

The coagulation cascade is activated by incubating plasma with the optimal amount of thromboplastin and calcium; the clotting time is then measured.

Reagents

Note

Dade® Innovin® Reagent can be used manually or on automated coagulation analyzers.
Siemens Healthineers provides Reference Guides (Application Sheets) for several coagulation analyzers. The Reference Guides (Application Sheets) contain analyzer/assay-specific handling and performance information which may differ from that provided in these Instructions for Use. In such a case, the information contained in the Reference Guides (Application Sheets) supersedes the information in these Instructions for Use. In addition, please also consult the instruction manual of the instrument manufacturer!

Materials provided

10 x → 4 mL, REF B4212-41,
10 x → 10 mL, REF B4212-51,
12 x → 20 mL, REF B4212-101

Composition

Dade® Innovin® Reagent: Lyophilized reagent consisting of recombinant human tissue factor and synthetic phospholipids (thromboplastin), calcium ions, a heparin-neutralizing compound, buffers and stabilizers (bovine serum albumin).

Warnings and Precautions

For *in-vitro* diagnostic use only.

For laboratory professional use.

Safety data sheets (MSDS/SDS) available on siemens-healthineers.com/sds.

CAUTION!
Federal (USA) law restricts this device to sale by or on the order of licensed healthcare professionals.

Caution
This device contains material of animal origin and should be handled as a potential carrier and transmitter of disease.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with all government requirements.

Reagent Preparation

Reconstitute a vial of lyophilized Dade® Innovin® Reagent with distilled or deionized water using the volume stated on the vial label.

To ensure complete reconstitution, thoroughly mix the contents of the vial immediately after adding the water. If left to stand, Dade® Innovin® Reagent should be re-mixed before use in order to ensure a homogeneous solution. Store at 2 to 8 °C. Continuous mixing is not necessary.

Note: Do not use water that contains preservatives.

Storage and Stability

Store at 2 to 8 °C. At this temperature, the unopened reagent can be used until its expiry date (see vial label).

Stability after reconstitution:	2 to 8 °C	10 days (closed vial)
	15 to 25 °C	5 days (closed vial)
	37 °C	24 hours (closed vial)

Information about on-board stability is specified in the Reference Guides (Application Sheets) for the different coagulation analyzers.

Do not freeze!

Signs of expiry: Absence of vacuum when opening the vial; reagent is difficult to reconstitute; results are not reproducible.

Materials required but not provided

Control Plasma N or Dade® Ci-Trol® Level 1

Control Plasma P, Dade® Ci-Trol® Level 2 or Dade® Ci-Trol® Level 3

Standard Human Plasma or fresh normal plasma[4] for determining the mean normal PT (MNPT)[a]

Sodium citrate solution (0.11 mol/L or 0.13 mol/L / 3.2 % or 3.8 %) for blood collection

Distilled or deionized water without preservatives

Plastic tubes

Plastic transfer pipettes

Pipettes for precise measurement of 20.0 mL, 10.0 mL, 1.0 mL, 0.50 mL, 0.20 mL and 0.10 mL

Coagulation analyzer

[a] The mean normal PT (MNPT) is defined as the mean value of the normal range. It must be determined specifically for each thromboplastin lot using the method used to analyze the patient samples and, where appropriate, using the coagulation analyzer used for the analysis. Follow appropriate laboratory guidelines for establishing an MNPT, if applicable. For US customers the appropriate CLSI guideline is recommended.

Specimen Collection and Preparation

Mix nine parts of freshly collected patient blood with one part of 0.11 or 0.13 mol/L (3.2 % or 3.8 %) sodium citrate solution. An evacuated tube system or syringe may be used.

Centrifuge the blood specimen at 1500 x g for no less than 15 minutes at room temperature.

Store in an unopened tube at room temperature. Do not store on ice or at 2 to 8 °C as cold activation of F VII may alter results. Plasma should be tested within 24 hours of blood collection. Samples should not stand at 37 °C for more than 5 minutes. If the patient is on both heparin and coumarin-based anticoagulant therapy, the results may vary with time of storage.

Please refer to CLSI document H21-A5[5] for detailed information on sample preparation and storage.

Procedure

Manual Testing:

Prewarm Dade® Innovin® Reagent to 37 °C.

Pipet into coagulation tubes as follows:

	Test Sample	Control
Plasma	0.1 mL	-
Control	-	0.1 mL
Incubate tubes and samples for 1–2 minutes (max. 5 minutes) at 37 °C		
Add prewarmed Dade® Innovin® Reagent	0.2 mL	0.2 mL

Start stopwatch simultaneously with addition of Dade® Innovin® Reagent. Observe time of clot formation.

Internal Quality Control

Normal range: Dade® Ci-Trol® Level 1 or Control Plasma N

Pathological range: Dade® Ci-Trol® Level 2, Dade® Ci-Trol® Level 3 or Control Plasma P

Two levels of quality control material (normal and pathological range) must be measured at start of the test run, with each calibration, upon reagent vial changes and at least every eight hours on each day of testing.

The control material should be prepared and processed in the same manner as the samples.

A range of allowable variation should be established for controls in each laboratory. New control ranges should be established for each lot of reagent or control material. This range is usually based on ± 2.5 standard deviations (SD) from the control mean.

If the control values are outside of the established range, check the coagulation analyzer, controls and reagents. Do not release patient results until the cause of deviation has been identified and corrected.

Results

Currently, various methods are used for reporting PT results. ISI (International Sensitivity Index) values for Dade® Innovin® Reagent are provided for the particular reagent/instrument combination; these enable the results to be reported in INR (International Normalized Ratio)[4]. Computation and use of the INR are described below. Monitoring of oral anticoagulant therapy with vitamin K antagonists should only be reported with PT results expressed as INR as recommended in official guidelines and in the literature[4]. Alternatively, the patient's PT (in seconds) together with the reference range (in seconds) can be used to report results.

Example: patient result of 18 seconds; reference range 9.9 to 11.8 seconds.

Determination of INR (International Normalized Ratio)

Values using Dade® Innovin® Reagent

International PT (Prothrombin Time) Standardization for Oral Anticoagulant Therapy Monitoring

1. According to the joint recommendations of the World Health Organization (WHO)[4] and the International Committee on Thrombosis and Haemostasis, the PT results for patients on vitamin K antagonist oral anticoagulants should be reported as INR values. Reported INR results are independent of the reagents and methods used, and are specifically intended for assessing patients stabilized on longterm oral anticoagulant therapy.

   The INR is determined[4] according to the following equation:

   $$INR = R^{ISI}, \text{ where } R = \frac{\text{Patient PT}}{\text{MNPT}}$$

   ISI is the International Sensitivity Index of the reagent/instrument combination.

   The ISI values for Siemens Healthineers thromboplastin reagents are determined in accordance with WHO recommendations.

2. Methods of INR calculation:

   a. Calculators with exponential functions:

   These instructions refer specifically to the Texas Instruments TI-55 II calculator. Other calculators, e.g. Hewlett-Packard models, may require different key stroke sequences. Consult the calculator reference manual and check example problems against the Conversion Table to assure mastery of conversion procedures.

   Enter Patient PT in seconds, press "÷", enter MNPT, press "=". The display will now show R, the Patient Ratio. Now press the "y^x" key, then enter the specific ISI value of the thromboplastin/ instrument combination used and press "=". The result displayed is the patient's INR value.

Press	**Notes**
Example: 24	patient PT
÷	divided by
11.0	MNPT
=	patient ratio (display shows 2.1818)
y^x	exponential function key
1.1	**example** ISI value
=	result: INR (display shows 2.3588)
	Report as INR = 2.4

   b. Conversion Table:

   First, calculate the patient PT/MNPT ratio, R. The INR value can then be read from the enclosed INR Conversion Table by looking in the column under the appropriate ISI value, in the row that corresponds to the patient's PT ratio (R).

   c. Automatic:

   INR values can be computed automatically by various coagulometers. For details, consult the relevant instruction manual. Siemens Healthineers provides Reference Guides (Application Sheets) for several coagulation analyzers.

Derived Fibrinogen

Using Dade® Innovin® Reagent and the appropriate assay on Siemens Healthineers photometric coagulation analyzers or SYSMEX coagulation analyzers, the fibrinogen concentration may be derived by analyzing the change in optical signal during prothrombin time determinations, using a derived fibrinogen calibration curve. This calibration curve (master curve) is provided by Siemens Healthineers in the lot-dependent Table of Assigned Values.

Limitations of the Procedure

There are no other clotting factors in recombinant human tissue factor. Factor assay curves using Dade® Innovin® Reagent may therefore give longer clotting times at the lowest levels of the deficient factor than with other reagents. This may result in clotting times greater than 100 seconds for low factor levels in factor assay curves.

Derived fibrinogen results within the reference range can be directly reported. Results outside the reference range should be re-measured by a standard fibrinogen determination method, e.g. Fibrinogen method with Dade® Thrombin Reagent or with Multifibren® U Reagent. Derived fibrinogen testing is not suitable in patients with dysfibrinogenemia[6] or patients with prolonged PT, e.g. under oral anticoagulation[7,8].

Lipoglycopeptide antibacterial drugs (such as oritavancin or telavancin) may interfere with PT based assays. Consult Instructions for Use of respective drugs.

Siemens Healthineers has validated use of these reagents on various analyzers to optimize product performance and meet product specifications. User defined modifications are not supported by Siemens Healthineers as they may affect performance of the system and assay results. It is the responsibility of the user to validate modifications to these instructions or use of the reagents on analyzers other than those included in Siemens Healthineers Application Sheets or these Instructions for Use.

Results of this test should always be interpreted in conjunction with the patient's medical history, clinical presentation and other findings.

Interfering Substances

Many commonly administered drugs may affect the results obtained in prothrombin time testing[4]. This should be kept in mind especially when unusual or unexpected abnormal results are obtained. Unexpected abnormal results should be followed by additional coagulation studies to determine the source of the abnormality.

Turbidity of lipemic samples, e.g. with parenteral feeding, may preclude accuracy in the derived fibrinogen determination.

Dade® Innovin® Reagent is insensitive to concentrations of unfractionated heparin up to approximately 2.0 units per mL. The heparin sensitivity study was conducted using spiked normal pooled plasma and the sensitivity to heparin was defined by the concentration of heparin in the specimen that prolonged the PT results exceeding the upper limit of the reference range.

Inhibitors such as lupus anticoagulant may interfere with the prothrombin time and result for example in INRs that do not reflect the exact degree of anticoagulation[9].

Hirudin or other direct thrombin inhibitors in therapeutic dose result in prolonged prothrombin times[10,11].

Some blood collection tubes may contain Mg^{2+} ions, which have been shown to interfere with recombinant thromboplastins[12].

Blood plasma substitutes that contain hydroxyethyl starch (HES) may interfere with the analysis. Therefore, it is advised that plasma samples that contain such substitutes should not be analyzed with the PT derived fibrinogen method.

Expected Values

Values for healthy individuals vary from laboratory to laboratory depending on the technique used. Therefore, each laboratory should establish its own reference intervals based on the procedure and coagulation analyzers used.

In studies on the SYSMEX CA-7000 System with ostensibly healthy subjects the following reference intervals were determined:

Analyte	Samples n =	2.5^{th} to 97.5^{th} percentile
PT	158	9.9 to 11.8 seconds
Derived fibrinogen	124	1.8 to 3.5 g/L

Therapeutic Ranges

Therapeutic ranges for INR may vary depending on the indication of oral anticoagulant therapy[13].

Specific Performance Characteristics

Precision

Precision of prothrombin time results is generally limited by the method used. Therefore, within a single lot, the reagent should yield results which are reproducible within the quality control of the laboratory.

The precision of Dade® Innovin® Reagent on the SYSMEX CA-6000 system was estimated with quality control material from a total of six (6) separate runs over three (3) testing days (two runs per day) and four (4) replicates per control level, per run with the following results:

				Coefficient of Variation		
Assay	**Control Level**	**n**	**Mean**	**Intra-Assay**	**Inter-Assay**	**Total**
Prothrombin Time	1	24	12.4 s	1.3 %	2.0 %	2.4 %
	2	24	33.0 s	0.7 %	3.4 %	3.5 %
Derived Fibrinogen	1	24	2.37 g/L	4.0 %	2.8 %	4.9 %
	2	24	3.37 g/L	4.1 %	3.0 %	5.1 %

Technical Assistance

For customer support, contact your local technical support provider or distributor.

siemens-healthineers.com

Current Version of Application Sheets

Dade® Innovin® can be used in combination with various automated coagulation analyzers. Siemens Healthineers provides Reference Guides/Application Sheets for coagulation analyzers under the dedicated link below:

siemens-healthineers.com/rg

As Siemens Healthineers continuously monitors the product performance and safety, the users are required to ensure that they work with the correct revision of the instructions for the product lots in use. Please periodically review the availability of new electronic labeling revisions to ensure safe use of the product.

The IFU version number is visible on each product box label. Siemens Healthineers ensures that all products lots bearing the same IFU version number are compatible with the electronic labeling provided via siemens-healthineers.com/eIFU.

Bibliography

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Definition of Symbols

The following symbols may appear on the product labeling:

	Do not reuse		Use By
LOT	Batch Code	REF	Catalogue Number
	Caution		Manufacturer
EC REP	Authorized representative in the European Community		Contains sufficient for <n> tests
	Biological Risks	IVD	*In Vitro* Diagnostic Medical Device
	Temperature Limitation		Consult instruction for Use
	Non-sterile	CE	CE marking of conformity
CE 0197	CE marking of conformity with notified body ID number. Notified body ID number can vary.	CONTENTS	Contents
	Reconstitution volume	LEVEL	Level
	Keep away from sunlight and heat	WARNING	Warning
DANGER	Danger	RxOnly	Prescription device (US only)
UDI	Device Identification (UDI) barcode	REACH xx/xx/xx	REACH Authorization Number

Legal Information

Purchase of the Dade® Innovin® Reagent provides the purchaser with a limited license. For certain uses, in particular to make, use, offer for sale or sell products comprising the Dade® Innovin® Reagent or parts thereof in the USA and Canada, a separate license under US Patent No. 7,084,251 and Canadian Patent No. 1,341,589 is needed. Contact Siemens Healthcare Diagnostics Inc., Tarrytown, NY 10591, USA, for terms of such license.

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