Storage and Stability: Before and during testing, the plasma sample should be maintained in the plastic tubes at 2 to 8°C to ensure stability of the factors. If testing is delayed for more than 2 hours, the plasma may be stored at -20°C for up to one month. Frozen samples should be thawed rapidly at 37°C before testing.

**PROCEDURE**

**Materials Provided:**
- Thromboplastin Li Reagent (10 x 5 mL vials)
- Thromboplastin Li Reagent (10 x 10 mL vials)

**MATERIALS AND EQUIPMENT REQUIRED:**
- Coagulation Instrument
- 37°C Water Bath
- Reaction Cups or plastic test tubes (12 x 75 mm)
- Pipette

**PROCEDURE NOTE:** Throughout the procedure, all test tubes, syringes, and pipettes should be plastic.

**AFFECTED INIONS:**
- Calcium ions
- Tissue thromboplastin

**STORAGE AND STABILITY:**
- Reagents should be stored at 2 to 8°C
- The plasma should be stored at 2 to 8°C before and during testing.

**SUMMARY AND PRINCIPLE**

The one-stage prothrombin time test (PT) has become the basic coagulation screening test for the diagnosis of congenital and acquired deficiencies of Factors II, VII, IX and X. Oral anticoagulant drugs inhibit hepatic synthesis of the vitamin K dependent clotting Factors II, VII, IX, and X. Therefore, the prothrombin time test is appropriate to monitor oral anticoagulant therapy since it is sensitive to three of the four factors involved. It can be modified to quantify factor levels using appropriate factor deficient plasma.

**INSTRUMENTS**

- Prothrombin time reagent
- Plasma collection tube
- Reaction cup

**SPECIMEN COLLECTION AND HANDLING**

**NOTE:** After initial whole blood collection, 3.2% sodium citrate, all test tubes, syringes and pipettes used during testing should be plastic.

**SPECIMEN:**
- Plasma obtained from whole blood anti-coagulated with 0.1 M (3.2%) sodium citrate.

**SPECIMEN COLLECTION:**
- Nine parts freshly collected whole blood should be immediately added to one part anticoagulant.

**SPECIMEN PREPARATION:**
- Specimen should be centrifuged at 1500 x g for 15 minutes and the plasma removed with a plastic pipette, placed in a plastic tube and capped. PT assays should be completed within 24 hours of collection. If testing is not completed within 24 hours, plasma should be removed from cells and frozen at -20°C for up to 2 weeks or -70°C for up to 6 months.

**REFERENCES:**


**COMPARISON**
- A control system was done using the Thromboplastin Li Reagent and another thromboplastin reagent. One hundred five (105) plasma specimens, both normal and abnormal clinical specimens were tested with both PT reagents. The linear regression equation and coefficient of determination (r²) of the INR values are reported.

**RESULTS**

The results of the Prothrombin Time Tests should be reported to the nearest tenth of a second. Results below the lower limits of the range may be considered abnormal and follow-up testing should be performed. PT values below the lower limits of the range may indicate a compromised sample, and a new sample should be collected.

**LIMITATIONS**

Expected values for the prothrombin time test will vary from one laboratory to another, depending on several variables. These include the method of clot detection, temperature, pH, sample collection technique, type of anticoagulant and time and method of plasma storage. Therefore, laboratories should establish their own expected values for patients, and well defined performance standards for control plasma. The use of icteric, lipemic, or hemolyzed sample results is avoided due to possible interference. Especially when using photo-optical instruments. The impact of other therapeutic drugs, in addition to oral anticoagulant therapy, can influence interpretation of PT test results. Obtaining an accurate patient history and noting specific drug therapies can help in the proper understanding of the potential impact on laboratory test results. The presence of heparin as a contaminant in the patient sample should always be considered when an abnormal result is obtained.

**PERFORMANCE CHARACTERISTICS**

**PRECISION**

Precision studies were performed to establish Within-Run and Between-Run CV’s for normal control plasma and abnormal control plasma. A single lot different thromboplastin reagent for these studies was used. Results are shown below.

**PHOTO-OPTICAL**

<table>
<thead>
<tr>
<th>Mean</th>
<th>SD</th>
<th>CV%</th>
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<tbody>
<tr>
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**REFERENCE**

- For Sales, Technical and Order Information and Service Assistance, call 800-231-5663 toll free.

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In no case will Helena Laboratories be liable for consequential damages even if Helena has been advised as to the possibility of such losses.

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**ECG**

European Community Authorized Representative
Helena Laboratories UK, Ltd.

Sunderland Enterprise Park
Sunderland SR5 3XB

England

**REFERENCES**


**THROMBOPLASTIN LI Reagent**

**Cat. No.**
- Thromboplastin Li Reagent (10 x 5 mL) 5248
- Thromboplastin Li Reagent (10 x 10 mL) 5249

**EQUIPMENT AND SUPPLIES**

- Coagulation S.A.R.P. (10 x 1.0 mL) 5185
- Norm-Trol Coagulation Control (5 x 1.0 mL) 5183
- Ab-Trol Coagulation Control (10 x 1.0 mL) 5183
- Cascade M 1710
- Cascade M-4 1711

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The foregoing warranties are in lieu of all warranties expressed or implied including, but not limited to, the implied warranties of merchantability and fitness for a particular purpose.
Prothrombin Time Reagent

**SUMMARY AND PRINCIPLE**

The prothrombin time test (PT) is the most commonly used coagulation test in clinical laboratories. It measures the time required for plasma to clot and is used to screen for and monitor the effectiveness of oral anticoagulant therapy. The test is based on the intrinsic pathway of coagulation and is sensitive to deficiencies in the extrinsic pathway of coagulation. It is used to monitor warfarin therapy, and the international normalized ratio (INR) is calculated based on the ratio of the patient’s PT to the mean of the reference range. The INR is used as a convenient way to compare the coagulant activity of plasma samples. The lower the ISI for a given reagent the more sensitive the reagent is to coagulation inhibitors. The lower the ISI the more sensitive the reagent is to coagulation inhibitors.

**EQUIPMENT AND SUPPLIES**

- Coagulation S.A.R.P. (10 x 1.0 mL) 5185
- Norm-Trol Coagulation Control (10 x 1.0 mL) 5186
- Ab-Trol 2 Coagulation Control (10 x 1.0 mL) 5183
- Cascade M
- Cascade M-4

**PERFORMANCE CHARACTERISTICS**

**PRECISION**

- Within Run: Mean 53.0 ± 0.2
- Between Run: Mean 53.0 ± 0.2

**REFERENCES**


**COMPARISON**

A combination of studies was done using the Thromboplastin LI Reagent and another thromboplastin reagent. One hundred five (105) plasma specimens, both normal and abnormal clinical samples were tested with both PT reagents. The linear regression equation and coefficient of determination (r) of the INR values are reported.

**RESULTS**

The results of the Prothrombin Time tests should be compared to the nearest tenth of a second. Results greater than 12 from the lower limits of the range should be considered abnormal and follow-up testing should be performed. PT values below the lower limits of the range may indicate a compromised sample, and a new sample should be collected.

**LIMITATIONS**

Expected values for the prothrombin time test will vary from one laboratory to another, depending on several variables. These include the method of clot detection, temperature, pH, sample collection technique, type of anticoagulant and method and time of plasma storage. Therefore, laboratories should establish their own expected values for patients and well defined performance standards for control plasma. The use of icteric, lipemic, or hemolyzed plasma is not recommended due to possible interference, especially when using photo-optical instruments. The impact of other therapeutic drugs, in addition to oral anticoagulant therapy, can influence interpretation of PT test results. Obtaining an accurate patient history and noting specific drug therapies can help in the proper understanding of the potential impact on laboratory test results. The presence of heparin as a contaminant in the patient sample should always be considered when an abnormal result is obtained.

**SPECIFIC COLLECTION AND HANDLING NOTE**

**STORAGE AND STABILITY:** Before and during testing, the plasma sample should be maintained in the plastic tubes at 2 to 8°C to ensure stability of the factors. If testing is delayed for more than 2 hours, the sample may be stored at 2 to 8°C for up to two weeks or at -70°C for up to one month. Frozen samples should be thawed rapidly at 37°C before testing.

**EQUIPMENT AND SUPPLIES**

- Cascade M-4
- Cascade M

**SPECIMEN COLLECTION AND HANDLING NOTE**

**STORAGE AND STABILITY:** Before and during testing, the plasma sample should be maintained in the plastic tubes at 2 to 8°C to ensure stability of the factors. If testing is delayed for more than 2 hours, the sample may be stored at 2 to 8°C for up to two weeks or at -70°C for up to one month. Frozen samples should be thawed rapidly at 37°C before testing.

**QUALITY CONTROL:** Each laboratory should establish a quality control program that includes normal and abnormal controls to evaluate instrument, reagents, and technical performance. The normal and abnormal controls should be tested daily prior to performing tests on patient plasma.

**EXPECTED VALUES:**

A reference range study was conducted using triplicate specimens from 120 normal healthy adults. Approximately equal numbers of males and females were used. The PT results were as follows:

<table>
<thead>
<tr>
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<th>Between Run</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>13.7</td>
<td>13.7</td>
</tr>
<tr>
<td>SD</td>
<td>0.2</td>
<td>0.2</td>
</tr>
<tr>
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<td>31.1</td>
</tr>
<tr>
<td>SD</td>
<td>0.5</td>
<td>0.6</td>
</tr>
<tr>
<td>CV%</td>
<td>1.8</td>
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