The Citrate aPTT is affected by poor technique including blood collection and the transfer of blood to the sample well. The accuracy of the test is largely dependent upon the quality of the blood specimen, which may be affected by:

- Foaming of the sample
- Hemolysis of the sample
- Clotted or partially clotted blood

Results greater than 300 seconds should be considered abnormally high and the test should be repeated or reported as $>300$ seconds.

Performance Characteristics

**Reference range**

Citrated whole blood samples were obtained from normal healthy animals and tested with the Citrate aPTT assay. Reference ranges were developed as follows:

<table>
<thead>
<tr>
<th>Species</th>
<th>Canine</th>
<th>Feline</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reference Range (sec)</td>
<td>72–102</td>
<td>65–119</td>
</tr>
</tbody>
</table>

Limitations

Test results under 12 seconds and over 300 seconds are not reported. Instead, either an "Out of range - Lo" or "Out of range - Hi" message is displayed.

References


Suggested Readings


Intended Use

The Citrate aPTT is a unitized coagulation test intended for in vitro use in performing a quantitative, one-stage Activated Partial Thromboplastin Time (aPTT). The Citrate aPTT test is performed using citrated canine or feline whole blood. This test is to be used in the Coag Dx™ Analyzer and is also compatible with the SCA2000™ Coagulation Analyzer.

**This test is for veterinary use only and not for human diagnostic use.**

Summary and Explanation

The Citrate aPTT is a measure of the intrinsic and common coagulation pathways, which involves all coagulation factors except Factors VII and III (tissue factor). The aPTT is a modification of the Partial Thromboplastin Time (PTT). The PTT uses a phospholipid derived from either brain or lung tissue to mimic the role of platelets in the coagulation process. The aPTT contains a contact activating substance to standardize the activation of Factor XII, thereby providing a more precise and sensitive assay. The addition of a contact activator such as glass, kaolin or diatomaceous earth distinguishes the aPTT from the PTT.

The Citrate aPTT assay resolution is achieved through the use of a platelet factor 3 substitute and a kaolin activator, and does not require an incubation step. Since little specimen processing is required, Citrate aPTT results are obtained in less than four minutes. In addition, because the test is performed using a citrated whole blood sample, testing can be performed for up to 2 hours after sample collection without affecting test results.

Principle of Operation

The coagulation analyzer utilizes a mechanical end point clotting mechanism in which clot formation occurs within the disposable Citrate aPTT cartridge. Following citrated whole
blood sample introduction, the analyzer precisely measures 15 microliters of blood and automatically moves it into the test channel within the Citrate aPTT cartridge. The remainder of the blood sample, not needed for testing, is automatically drawn into the waste channel of the cartridge. Sample/reagent mixing and test initiation are also performed automatically, requiring no operator interaction. After mixing with the reagent, the sample is moved back and forth within the test channel and observed for clot formation.

The clot detection mechanism consists of a series of LED optical detectors aligned with the test channel of the cartridge. The speed at which the blood sample moves between the detectors is measured. As clot formation begins, blood flow is impeded and the movement slows. The analyzer recognizes that the clot end point has been achieved when the movement decreases below a predetermined rate. The Citrate aPTT whole blood clotting time is reported in whole seconds.

Reagents
Each box of Citrate aPTT test cartridges contains:

- 10 pouches, each containing one Citrate aPTT test cartridge and one dessicant

The Citrate aPTT test cartridge is a self-contained disposable test chamber preloaded with a dried preparation of kaolin, phospholipid, calcium salts, stabilizers and buffers. Each cartridge is individually packaged in a pouch. Cartridge pouches are stamped with a lot-specific expiration date.

Caution: All used test cartridges should be considered as potentially infectious, handled with care and disposed of properly.

Storage and Stability
When refrigerated (2°–8°C), the foil-pouched Citrate aPTT cartridges are stable until the marked expiration date. Room temperature storage (15°–30°C) is optional for unopened, pouchged cartridges. Citrate aPTT cartridges should not be exposed to temperatures in excess of 37°C.

Note: Room temperature recathing is to a maximum of 4 weeks, but must never exceed the marked expiration date. Redating is necessary if stored at room temperature. Mark the outer box with the new expiration date when cartridges are stored at room temperature.

Sample Collection
Blood samples to be used for coagulation testing must be collected in the following manner to prevent contamination with tissue thromboplastin, or indwelling intravenous (I.V.) solutions that interfere with the coagulation assays. Poorly collected blood samples with visible clotting or debris accumulation must be discarded and a fresh sample collected.

Patient excitement should be minimized as this can increase platelet count, platelet aggregation, and the levels of von Willebrand Factor (vWF), fibrinogen, and Factors V and VIII. Prolonged venous stasis and excessive probing for the vessel should be avoided. Use of the cephalic or saphenous veins are advised as bleeding is easier to control from these sites (Green, et al, 1995). If a syringe is used, it should have a 23-gauge needle or larger. Use of excessive force when expelling the blood specimen through the needle may cause hemolysis.

Note: It is recommended that coagulation testing be performed within two hours of sample collection if stored at room temperature.

Note: Blood samples for testing should be collected at least 5 minutes prior to testing to allow for adequate mixing of the sodium citrate with the sample.

Material provided
- Citrate aPTT test cartridges
- 11 single-use disposable pipettes

Material required (not provided)
- Coagulation analyzer
- Evacuated tube containing sodium citrate (3.2%)
- Blood collection needle

Operating Instructions
Before performing any assay, refer to the IDEXX Coag Dx™ Analyzer Operator’s Manual for detailed operating instructions.

Operating Precautions
Do NOT use cartridges that are past their marked expiration date, or that have been improperly stored.

Do NOT force a cartridge into the analyzer. If resistance to insertion is encountered, gently remove the cartridge and examine the cartridge slot. Remove any obstruction before attempting further use of the analyzer.

Specimen collection and handling for all coagulation testing requires careful adherence to guidelines. As with all diagnostic tests, test results should be scrutinized in light of a specific patient’s condition and anticoagulant therapy. Any results exhibiting inconsistency with the patient’s clinical status should be repeated or supplemented with additional test data.

Syringe sample, from indwelling line or venipuncture
Note: The amount of blood required to adequately flush the line until it is free of contaminants is dependent on the amount of solution contained within the line. Greater volumes will be required to clear longer lines.

1. Collect a sufficient sample to fill the blood collection tube.
2. Attach a needle to the syringe. Carefully puncture the stopper of an evacuated test tube containing sodium citrate (3.2%) and allow blood to be drawn into the tube until the flow stops.
3. Mix gently. Discard the syringe with the needle.

Test procedure
Refer to the IDEXX Coag Dx™ Analyzer Operator’s Manual if any fault message should appear during this procedure.

1. Insert a test cartridge into the cartridge opening of the analyzer. The cartridge must be inserted with the blood reservoir facing up. The analyzer will automatically identify the test cartridge and display the test type.
2. During the warming stage, observe the display for fault messages.