

# Predictive Value of the Rapid Whole Blood Agglutination D-Dimer Assay (AGEN SimpliRED) in Community Outpatients with Suspected Deep Venous Thrombosis

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## Abstract

**Context:** D-dimer assay has been used to screen patients with deep venous thrombosis (DVT). Because both the predictive value and sensitivity/specificity of the test vary according to the type of assay, prevalence, and pretest probability of DVT, clinicians must know the local performance of the d-dimer assay.

**Objective:** To evaluate the predictive value of the rapid whole blood agglutination d-dimer Assay (AGEN SimpliRED) in community outpatients with suspected DVT in the Kaiser Permanente (KP) Mid-Atlantic Region.

**Design:** Retrospective, randomized, cross-sectional review of electronic medical records of patients with suspected DVT who underwent d-dimer testing for venous thromboembolism.

**Methodology:** A total of 5104 patients with suspected venous thromboembolism underwent d-dimer testing using AGEN SimpliRED from April 2001 to December 2002. A total of 551 electronic medical records were reviewed, and results of d-dimer assay and compression ultrasonography were tabulated. Records were analyzed to determine later diagnosis of DVT or unexplained death occurring as late as six months after initial testing.

**Results:** Electronic records showed a 5.3% disease prevalence. Ten patients were excluded from data analysis. A total of 129 (23.8%) patients had positive d-dimer; the positive predictive value was 20.2% (CI, 13.2% to 27%). A total of 412 (76.1%) patients had negative test results; three of these patients had DVT shown by compression ultrasonography; negative predictive value was 99.3% (CI, 98.4% to 100%). Calculated sensitivity was 89.7%; specificity was 79.9%.

**Conclusion:** In the outpatient setting, the rapid whole blood agglutination d-dimer assay (AGEN SimpliRED) used in combination with both clinical judgment and compression ultrasonography exhibited a high negative predictive value comparable with previously reported values.

\*Approved by KP Mid-Atlantic Institutional Review Board on April 2004.

## Introduction

In the evaluation and management of suspected deep venous thrombosis (DVT), clinical evaluation alone is not sufficient to confirm or exclude presence of the disease.<sup>1,2</sup> The reference standard for diagnosing DVT is by venography, which is invasive. Because of its noninvasive nature, compression ultrasonography (CUS) has replaced venography; however, although CUS is highly sensitive and specific for symptomatic *proximal* DVT, this technique is not as sensitive as venography for detecting thrombus in the *distal* vein of the calf.<sup>3,4</sup> Because approximately 13% to 30% of affected patients have distal DVT that may propagate proximally,<sup>5,6</sup> a second examination is recommended five to seven days later to detect unvisualized calf thrombi that may have propagated proximally.<sup>7-12</sup> This additional examination can be costly, inconvenient, and could still miss acute DVT. In past years, d-dimer assays have been studied as a tool to aid diagnosis of thromboembolic disease. D-dimers are products of fibrin degradation when fibrin in the thrombus is lysed by plasmin. D-dimer assays cannot differentiate between clots associated with spontaneous venous thromboembolism and other causes of thrombus (eg, sepsis, trauma, surgery, malignancy, postoperative states, posttraumatic states, infection, autoimmune disease, inflammatory disease).<sup>13</sup> The combination of clinical decision rules or guidelines, d-dimer assay, and CUS also has

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been studied for diagnosis of DVT. These reports show that when used and interpreted in the proper clinical setting, the d-dimer assay provides a safe, cost-effective, clinician/patient-friendly means of ruling out DVT.<sup>2,7,14-16</sup>

Several assays are used to measure d-dimer: ELISA, which is the most accurate but which is lengthy and costly;<sup>8,17,18</sup> the latex agglutination test, which has high false-negative rates;<sup>19</sup> and the red blood cell agglutination test, which has wide ranges of specificity and sensitivity.<sup>20</sup> These tests have different characteristics; and because each test uses different reagents, the reported sensitivity and specificity for each assay cannot be applied interchangeably.<sup>7,21</sup> Because both predictive value and sensitivity/specificity of the d-dimer test vary according to the type of assay and pretest probability of DVT,<sup>2,13</sup> clinicians must understand the indications for and limitations of d-dimer measurement in the diagnosis of DVT and must inquire whether the assay's performance has been investigated locally.

The AGEN SimpliRED d-dimer test kit currently used in our laboratory is an autologous red cell agglutination assay which uses a chemical conjugate of a monoclonal antibody specific to d-dimer. D-dimer levels in excess of 0.12 mg/L result in visible agglutination of whole blood.

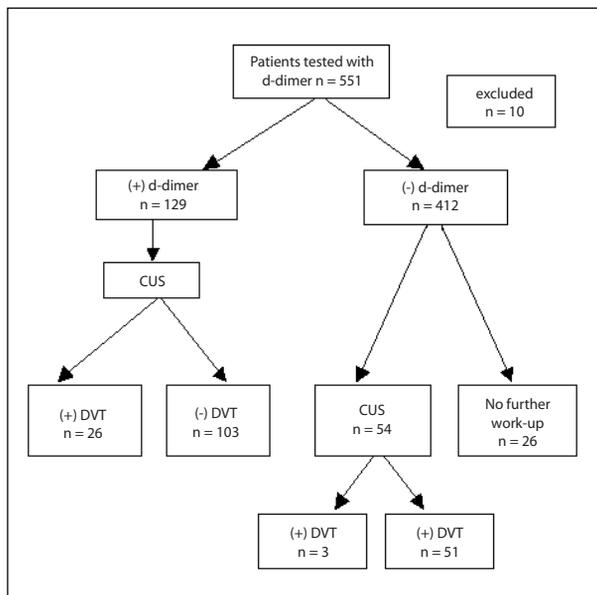


Figure 1. Summary of results of SimpliRED and Compression Ultrasonography (CUS) in patients with suspected deep venous thrombosis (DVT).

Table 1. Referral guideline for suspected deep venous thrombosis (DVT) <sup>a</sup>	
Clinical indications for referral	<p>Patients with suspected DVT who are also at high risk for DVT should be referred to the Radiology Department for evaluation by CUS.</p> <p>Patients who are not at high risk of DVT may be referred to the laboratory for a d-dimer study.</p> <p>High risk for DVT may be defined as:</p> <ul style="list-style-type: none"> <li>• three or more MAJOR criteria and no alternate diagnosis</li> <li>OR</li> <li>• two or more MAJOR criteria and two or more MINOR criteria and an alternate diagnosis</li> </ul> <p>Major criteria for DVT:</p> <ul style="list-style-type: none"> <li>• Active cancer</li> <li>• Paralysis, paresis, immobilization</li> <li>• Surgery &lt;4 weeks ago, bedridden &gt;3 days</li> <li>• Positive family history of DVT (&gt;2 relatives)</li> <li>• Thigh and calf swollen (measure and compare the circumference of each leg at midcalf and above the knee)</li> <li>• Calf &gt;3 cm of normal calf</li> </ul> <p>Minor criteria for DVT:</p> <ul style="list-style-type: none"> <li>• Trauma &lt;60 days</li> <li>• Pitting edema of the leg</li> <li>• Erythema</li> <li>• Dilated veins</li> <li>• Hospitalization &lt;6 months</li> </ul>

<sup>a</sup> Since 2003, guideline recommends CUS for pregnant patients.

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**Table 2. D-dimer assay diagnostic performance in 541 patients with clinical signs of deep venous thrombosis (DVT)**

Results of d-dimer assay	DVT present	DVT absent	Total
Positive (n = 129)	26	103	129
Negative (n = 412)	3	409	412
Total	29	512	541

Reported sensitivity ranges from 77% to 100%, and reported specificity ranges from 64% to 75%.<sup>22,23</sup>

Use of the AGEN SimpliRED d-dimer assay began in the KP Mid-Atlantic States Region in April 2001. We analyzed the predictive value of the rapid whole blood agglutination d-dimer assay in community outpatients seen for suspected DVT.

**Methods**

Over the 20-month period extending from April 2001 to December 2002, 5104 d-dimer tests using AGEN SimpliRED were conducted on outpatients in whom a clinician suspected venous thromboembolism. The tests were performed at ten different laboratory sites.

We conducted a retrospective, randomized review of the electronic medical records of 551 patients who had d-dimer testing for suspected DVT (Figure 1). Results of d-dimer testing were categorized according to test result. Current guidelines recommend CUS for all patients with positive d-dimer test results. Results of CUS were tabulated. Each medical record of patients with negative d-dimer results was reviewed to identify patients who subsequently had recorded results of CUS. The decision to refer patients with negative d-dimer for CUS was made independently by each clinician on the basis of physical findings as well as clinical judgment. Although we currently have a *Referral Guideline for Evaluation of Suspected DVT*<sup>24</sup> (Table 1) that categorizes patients into risk levels by using several major/minor criteria, this guideline is meant to be informational and does not replace reasonable, independent clinical judgment.

All records showing a negative d-dimer and no CUS as well as records that showed positive d-dimer but negative CUS results were further reviewed to determine later diagnosis of DVT or unexplained death occurring as long as six months after initial testing.

Data analysis was performed using the standard Bayesian statistical formula and calculations.

**Results**

Of the 551 patients whose electronic medical records were reviewed, ten patients were excluded for the fol-

lowing reasons: no electronic records documenting examination (seven patients); negative d-dimer with previous diagnosis of DVT several weeks before the test and preexisting receipt of anticoagulation therapy (one patient); negative d-dimer but patient unavailable for follow-up (one patient); positive d-dimer but additional testing refused (one patient).

Of the 541 records (Table 2), 29 showed DVT diagnosed by CUS, indicating a 5.3% prevalence (95% CI, 3.5% to 7.3%). For 129 (23.8%) patients, records showed positive d-dimer; and 412 (76.1%) patients tested negative. Of the 129 patients who tested positive, 103 patients had negative results of CUS, and CUS was used to diagnose 26 patients with DVT; these results indicated 89.7% sensitivity (95% CI, 78.6% to 100%) and 20.2% positive predictive value (95% CI, 13.2% to 27%). Of the 412 patients who tested negative, 54 patients (13.1%) were referred for CUS on the basis of a clinician’s evaluation and judgment. CUS was used to diagnose DVT in 3 of the 54 patients, indicating 79.9% specificity (95% CI, 76.4% to 83.4%); the negative predictive value was 99.3% (95% CI, 98.4% to 100%), and the false-negative rate was 0.7%.

Of the 541 patients, 183 (34%) had CUS. This group represented all patients with positive d-dimer and those with negative d-dimer referred for CUS by the clinician. None of the patients with positive d-dimer who had negative CUS or negative d-dimer results who did not have CUS had later diagnosis of DVT or unexplained death within six months after initial d-dimer testing.

We also reviewed the electronic medical records of the three patients who had false negative results of d-dimer testing. One of these patients was a 44-year-old man with a history of alcoholism who was seen for right lower leg pain and swelling, had negative results of d-dimer testing, and right peroneal clot shown subsequently by CUS. The second patient with false negative test results was a 25-year-old man with a history of DVT (in 1992) following fibular stress fracture and both grandparents with history of blood clots. He was initially seen for left lower leg pain and erythema and was treated with cephalixin; two days later, he returned with swelling of the left leg and received d-dimer testing that yielded negative results. Because of the patient’s medical history, CUS was done; this technique showed extensive superficial venous thrombotic disease as well as a partial narrowing in the popliteal vein. The third patient with false-negative test results was a 35-year-old woman with a history of advanced cervical cancer treated with radiation who was initially seen for a two-week history of bilateral swelling of the lower extremi-

ties. D-dimer test results for this patient were negative. CUS showed left-sided DVT.

## Discussion

Our study showed sensitivity of 89.7%, specificity of 79.9%, negative predictive value of 99.3%, and a positive predictive value of 20.2%. These results are comparable with several published studies, which showed high negative predictive value in patients who are at low to moderate risk for DVT.<sup>7,15</sup> Several reports show that sensitivity decreased with higher disease prevalence.<sup>14</sup> Our three patients with false negative d-dimer test results were at high risk for DVT. The question arises as to whether d-dimer testing should have been omitted in these patients and the patients instead referred directly for CUS. Review of electronic medical records for each of the 54 patients with negative d-dimer results who had CUS showed that the risk for DVT among these patients ranged from low to moderate to high on the basis of our current guideline (categorizing patients into risk levels by using major and minor criteria).

The decision to refer patients with negative d-dimer test results for CUS was made by each clinician independently on the basis of physical signs and clinical judgment. Several factors may influence a clinician's choice to refer a patient for CUS. Some clinical instances (for example, pregnancy) might not have been captured in the guideline: Our guideline, which was revised in April 2003, recommends CUS for pregnant patients because this population has a high rate of false-positive d-dimer test results.<sup>25</sup> The guideline for managing suspected DVT is mainly informational and is not meant to substitute or replace clinical judgment. Several reports show that clinical assessment of pretest probability—whether performed empirically or by prediction rule—did not alter the overall prevalence of DVT among patients who had been assigned a low pretest probability of having the condition.<sup>2,26</sup>

We also noted other benefits of using d-dimer assay. The approximate turnaround time for the test is 30 minutes, a timespan that indicates timesaving. In addition, before we started using the d-dimer assay, all 551 patients would have been referred for CUS. With introduction of the d-dimer assay, CUS was administered to 34% of the patients—those with positive as well as those with negative d-dimer test results—who were adjudged to be at risk for DVT. The other 66% of patients (ie, those who did not have CUS) did not have later diagnosis of DVT or unexplained death within six months after initial testing. This finding supports other

studies that show incorporating the d-dimer assay with clinical assessment has reduced the need for CUS without apparent compromise of safety.<sup>2,14,15,27,28</sup>

We conclude that, used in combination with clinical judgment and CUS, the rapid whole blood agglutination d-dimer assay (AGEN SimpliRED) had a 99.3% negative predictive value and a 20.2% positive predictive value in the outpatient setting—results comparable with previous reports.

## Study Limitations

Among the study population, clinical probability for DVT ranged from low to moderate to high—a finding that may lower the sensitivity/specificity of the test because this probability may vary according to the prevalence and pretest probability of DVT. That is, the lower the prevalence of the disease, the higher the negative predictive value.<sup>2,13</sup> Although venography is the reference standard used to rule out DVT, in our true-negative and false-positive population we based true negativity on endpoints such as a patient having neither a later diagnosis of DVT nor unexplained death within six months after initial testing. ❖

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## Prediction

Prediction is extremely difficult. Especially about the future.

—Niels Bohr, 1885-1962, Danish physicist