Assessment of Coagulation and Hemostasis Biomarkers in a Subset of Patients With Chronic Cardiovascular Disease

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Abstract
Measurement of a single marker of coagulation may not provide a complete picture of hemostasis activation and fibrinolysis in patients with chronic cardiovascular diseases. We assessed retrospective orders of a panel which included prothrombin fragment 1.2 (PF1.2), thrombin: antithrombin complexes, fibrin monomers, and D-dimers in patients with heart assist devices, cardiomyopathies, atrial fibrillation and intracardiac thrombosis (based on ordering ICD-10 codes). During 1 year there were 117 panels from 81 patients. Fifty-six (69%) patients had heart assist devices, cardiomyopathy was present in 17 patients (21%) and 29 patients (36%) had more than 1 condition. PF1.2 was most frequently elevated in patients with cardiomyopathy (61.1%) compared to those with cardiac assist devices (15.7%; \( P = 0.0002 \)). D-dimer elevation was more frequent in patients with cardiac assist devices (98.8%) compared to those patients with cardiomyopathy (83.3%; \( P = 0.014 \)). Patients with cardiomyopathy show increases of PF1.2 suggesting thrombin generation. In contrast, elevations of D-dimers without increase in other coagulation markers in patients with cardiac assist devices likely reflect the presence of the intravascular device and not necessarily evidence of hemostatic activation.

Keywords
heart disease, coagulation, prothrombin factor 1.2, d-dimer, fibrin monomers

Introduction
The National Patient Safety Goals from the Joint Commission encourage American hospital administrations to conduct internal reviews of anticoagulation safety practices.¹ A variety of tests can be used to monitor the coagulation system. Most hospital laboratories offer measurement of D-dimer as a marker of recent or ongoing fibrinolysis and clinicians may use the results to assess risk of thrombosis.² Importantly, D-dimer values above the reference range can be seen in a variety of situations not necessarily related to activation of the coagulation system, including advanced age³,⁴ and in the presence of intravenous catheters.⁵ This stresses the importance of the test’s negative predictive value, as D-dimer results within the reference range allow clinicians to rule-out certain conditions such as disseminated intravascular coagulation (DIC), pulmonary embolism, or deep vein thrombosis. Fibrin monomers are another analyte frequently offered by clinical laboratories. Increased levels of fibrin monomers indicate active cleavage of fibrinogen to fibrin and reflect concentration of thrombin activity.⁶ Levels of fibrin-monomers may predict left atrial appendage thrombosis in elderly patients with acute ischemic stroke.⁷ Concentrations of fibrin monomers vary in some hypercoagulable states (e.g., pregnancy, hormone replacement, chemotherapeutic agents, anti-angiogenesis medications) but are reliably increased in patients with DIC and malignancies.⁸,⁹ Some use fibrin monomers to monitor anticoagulant treatment.¹⁰,¹¹

Activation of the coagulation system upstream of fibrin monomer generation can be assessed by measuring prothrombin fragment 1.2 (PF1.2) and thrombin-antithrombin complexes (TATs). PF1.2 assesses ongoing activation and

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Results were segregated into their diagnostic ICD-10 groups; demographic information and anticoagulation treatment.

Collection of data from the clinical chart included classification of diseases managed by the World Health Organization. The ICD-10 is the 10th revision of the International Classification of Disease (ICD) codes corresponding to cardiac assist devices, cardiomyopathies, atrial fibrillation, and intracardiac thrombosis. Table 1 presents the specific codes to cardiac assist devices, cardiomyopathies, atrial fibrillation, or intracardiac thrombosis (Table 1 presents the specific codes and their descriptor). The ICD-10 is the 10th revision of the classification of diseases managed by the World Health Organization. Collection of data from the clinical chart included demographic information and anticoagulation treatment. Results were segregated into their diagnostic ICD-10 groups; however, some patients had more than 1 code associated with their panel result. When this occurred, we only counted the patient once in the category of cardiac assist devices as all these patients had the ICD-10 code for the device plus another code (cardiomyopathy or thrombosis).

Plasma samples for testing MOCHA parameters were obtained in coagulation tubes containing 3.2% sodium citrate. D-dimer levels were measured using high sensitivity latex dimer assay (Instrumentation Laboratories, Bedford, MA). PF1.2 and TATs were measured using Enzygnost ELISA kit (Siemens Healthcare, Tarrytown, New York, NY). Soluble fibrin monomer assays were performed using latex immunoassay (Stago, Parsippany, NJ).

We calculated the following for each parameter: the mean, median, range, and number of results that were above the reference range. A 2 tailed Fisher exact test using the online GraphPad calculator (https://www.graphpad.com/quickcalcs/contingency1.cfm) was used to calculate \( P \) values when comparing number of abnormal results for each of the parameters. Based on the evaluation of the MOCHA markers taken together, the sample results were segregated into 3 groups and correlated with the anticoagulation received at the time the MOCHA panel was performed. These groups included:

A. Patients with increased PF1.2 independent of elevation of the other parameters were considered to have prothrombin conversion to thrombin.
B. Patients with increased D-dimer together with elevated TATs and/or fibrin monomer suggested clot formation and fibrinolysis.
C. Patients with only D-dimer, or TAT, or fibrin monomer elevation were considered to have a non-specific panel.

The study was performed in accordance with Emory’s institutional review board (IRB). The IRB defined that a consent form was not necessary as physicians ordered the tests as part of the patient’s treatment and independent of this retrospective study. In addition, the study was deemed an expedited IRB review as results present patient data in aggregate.

Results

A total of 117 MOCHA panels from 81 patients were reviewed. This included 47 males and 34 women with an average age of 51 years (range 19-85). The most frequent ICD-10 code was for cardiac assist device (56 patients) followed by cardiomyopathy (17 patients). Upon chart review, diagnoses of the patients with the ICD-10 related to cardiomyopathy included coronary artery disease (7 patients), ischemic heart disease (3), cardiogenic shock (2), valvular insufficiency (1), sickle cell vaso-occlusive disease (1), sinus tachycardia (1), hypertrophic cardiomyopathy (1), and viral myocarditis (1). There were 29 patients with more than 1 ICD-10 code assigned; 24 had an ICD-10 code for cardiac assist device and cardiomyopathy while 5 had cardiac assist device and intracardiac thrombosis. Table

### Table 1. ICD 10 Used With Descriptors.

| ICD-10 code | Descriptor                           |
|-------------|-------------------------------------|
| Z95.811     | Presence of heart assist device      |
| I42.9       | Cardiomyopathy, unspecified, including primary and secondary causes and can be accompanied by heart failure or tachycardia |
| I51.3 or I23.6 | Intracardiac thrombosis, not elsewhere specified (independent of location)-OR- Thrombosis of atrium, auricular appendage, mural and ventricle or following complications of an acute myocardial infarction |
| I48         | Persistent atrial fibrillation or flutter, independent of chronicity (persistent), paroxysmal, atypical or other |

Abbreviation: ICD, International Classification of Disease.
2 presents demographic data, total number of patients, and number of measurements for each ICD-10 code. The table also shows the number of repeat MOCHA panels that these patients had, including repeats occurring within a 4-week period. Patients with cardiac assist devices had repeat testing more frequently compared to the other groups.

Table 3 shows the results for the different parameters for the MOCHA panels for each ICD-10 code. Of note, PF1.2 was most frequently elevated in patients with cardiomyopathy compared to the other ICD-10 codes. This difference was statistically significant compared to the patients with cardiac assist devices ($P \leq .001$). Also of note, was the high frequency of D-dimer elevation in patients with cardiac assist devices ($P = .014$, compared with cardiomyopathy patients). Analysis of the 24 patients with both cardiomyopathy and cardiac assist device ICD-10 codes showed similar biomarker behavior to that observed in patients with only cardiac assist device for the majority the parameters. Specifically, all showed elevated D-dimer and only 4 showed an increase in PF1.2.

Table 4 presents the correlation of the 3 MOCHA interpretation groups with the anticoagulation treatment, changes that occurred in those patients after repeat testing, and location (inpatient versus outpatients). Most of the patients with cardiomyopathy were considered to have prothrombin conversion, while those patients with cardiac assist devices were considered to have clot formation with fibrinolysis ($P \leq .001$). Twenty-four patients with cardiac assist devices had repeat testing. In these 24 patients, their panels changed from either prothrombin conversion or clot formation with fibrinolysis to either a non-specific panel or clot formation with fibrinolysis. Review of anticoagulation treatment showed that all patients with cardiac assist devices (56) were anticoagulated. Fifty (89%) patients were on warfarin while the remaining 6 (11%) patients were on heparin due to recent device implantation. Of the 50 patients on warfarin, 12 (24%) were considered to have prothrombin conversion while 22 (39%) had a second code of cardiac assist device.
molecular heparin, or warfarin; 4 (23%) were receiving platelet inhibitors (aspirin or P2Y12 inhibitors); and the remaining 4 were not receiving anticoagulation or antiplatelet therapy. Of the 9 patients receiving anticoagulants, 6 (54%) were considered to have prothrombin conversion, which was the same as 3 of the 4 patients receiving platelet inhibitors and 2 of the 4 patients not receiving platelet inhibitors or anticoagulation treatment. Lastly, all of the 8 patients with thrombosis or atrial fibrillation were receiving anticoagulant treatment and 4 (50%) were considered to have prothrombin conversion.

Discussion

Our results demonstrate that patients with cardiac assist devices commonly have D-dimer elevations, while increases in other MOCHA parameters are infrequent. Over 50% of MOCHA panels were interpreted as non-specific in this group, as there was only elevation of D-dimer. Increases of D-dimer are expected in this group of patients as even the insertion of an antecubital line can result in elevations of this marker.5 Also notable was the fact that patients with dual ICD-10 codes behaved mostly as patients with cardiac assist devices which may be due to the fact that they were adequately anticoagulated. In a previous analysis of cardiac assist devices in our institution, D-dimers were elevated when measured at baseline, during routine visits, after the device had been in place, and when patients had a thrombotic event.15 Of relevance, others have found that patients with recent myocardial infarcts that had reduction of D-dimer, either by pharmacological means or spontaneously, had a decreased risk of new ischemic events.16 This literature suggests that normalization or decrease of D-dimer may have significant value in patients with cardiac assist devices. Although not all patients with cardiac assist devices had repeat tests in our study, these were the patients with more frequent repeat testing suggesting that clinicians monitor patients using this biomarker panel.

In our series the frequency of elevation of fibrin monomer was similar for each ICD-10 code. Elevated values occurred in approximately half of our cases independent of the ICD-10 code. Some researchers have found that fibrin monomer had higher sensitivity than the other MOCHA parameters to predict deep venous thrombosis after surgery, others have found a strong association with ischemic heart disease, while others have found increased levels of fibrin monomer in patients with thrombi in the left atrial appendage.7,17,18 Our study could not corroborate results from the previous studies including heart thrombosis as we did not have enough patients with isolated intracardiac thrombi. In our study the percent of cases with increased levels of TATs was higher compared to fibrin monomer, but the frequency of increase (around 70%) was similar in all the ICD-10 codes. Many laboratories do not offer measurement of TATs.

In our series, the PF1.2 marker showed significant differences between heart assist devices and cardiomyopathy. This analyte was elevated in less than 15% of patients with cardiac assist devices while it was elevated in over 60% of patients with

| Table 4. Correlation of Results for Each ICD-10 Code and Anticoagulation Being Received. |
|---------------------------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Anticoagulation                         | Prothrombin conversion (A) % | Formed clot with fibrinolysis (B) % | Non-specific changes (C) % | Repeated panel that changed |
|---------------------------------------------|-----------------|-----------------|-----------------|-----------------|
| Device (56 patients)                      |                  |                  |                  |                  |
| Warfarin (mostly outpatients)             | 12              | 21              | 17              | 30              | 22             | 39              | 5 went from A to B; 18 went from B to C |
| Heparin (mostly inpatients)               | 2               | 4               | 1               | 2               | 2              | 2               | 1 went from B to C |
| Cardio-myopathy (17 patients)             |                  |                  |                  |                  |
| Warfarin (outpatients)                    | 0               | 0               | 0               | 0               | 2              | 12              | 1 repeated with no change |
| Heparin (mostly inpatients)               | 6               | 35              | 0               | 0               | 1              | 6               | 0 |
| Platelet inhibitors (outpatients)         | 3               | 18              | 0               | 0               | 1              | 6               | 0 |
| No anticoagulation (outpatients)          | 2               | 12              | 0               | 0               | 2              | 12              | 0 |
| Intra-cardiac thrombosis (4 patients)     |                  |                  |                  |                  |
| Warfarin (outpatient)                     | 1               | 25              | 0               | 0               | 0              | 0               | 1 repeated went from A to C |
| Heparin (mostly inpatients)               | 2               | 50              | 1               | 25              | 0              | 0               | 0 |
| Atrial fibrillation (4 patients)          |                  |                  |                  |                  |
| Heparin (inpatients)                      | 0               | 0               | 1               | 25              | 1              | 25              | 0 |
| Tranexemic acid (inpatient)               | 1               | 25              | 0               | 0               | 0              | 0               | 0 |
| Apixaban (outpatient)                     | 1               | 25              | 0               | 0               | 0              | 0               | 1 repeat went from A to C |

Abbreviation: ICD, International Classification of Disease.

(A) Increase in PF1.2 independent of elevation of the other parameters. (B) Increased D-dimer together with increased TAT and/or fibrin monomers. (C) Only D-dimer, TAT, or fibrin monomers elevated were considered as non-specific.
thrombosis in patients with cancer. Ota et al found it to be not routinely offered by laboratories. Increased amounts of myopathy had very frequent elevation of PF1.2 suggesting prothrombin conversion independent of the use of warfarin or heparin. Similarly, our cases with ICD-10 codes of thrombosis and atrial fibrillation were all anticoagulated but still 1 half showed prothrombin conversion. The only ICD-10 group with patients without anticoagulant or antiplatelet treatment was the cardiomyopathy group. 

In conclusion, patients with cardiomyopathy tend to have increased levels of PF1.2 indicating prothrombin conversion. In comparison, patients with cardiac assist devices had increased D-dimer while their PF1.2 was mostly within reference range, suggesting adequate anticoagulation in this group of patients. A prospective, real-time study of a cohort of patients with cardiac conditions would likely be useful to further identify how clinicians respond to MOCHA parameter results.

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