Composite Biomarkers for Assessing Frailty Status in Stable Older Adults With Cardiovascular Disease

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Background: The relationship between frailty status and laboratory measurements in cardiovascular disease (CVD) remains unclear. We investigated which laboratory measurements indicated frailty in stable older CVD patients.

Methods and Results: One-hundred thirty-eight stable older CVD patients were evaluated by laboratory measurements, with frailty assessed using the Kihon Checklist (KCL). Laboratory measurements were compared between frail and non-frail groups. Across the entire cohort, mean age was 81.7 years, mean left ventricular ejection fraction was 57.8%, and mean plasma B-type natriuretic peptide was 182 pg/mL. KCL scores were used to divide patients into non-frail (n=43; KCL <8) and frail (n=95; KCL ≥8) groups. Serum iron was significantly lower in the frail than non-frail group (mean [±SD] 61.2±30.3 vs. 89.5±26.1 μg/dL, respectively; P<0.001). Blood urea nitrogen (BUN; 27.3±16.5 vs. 19.7±8.2 mg/dL; P=0.013) and C-reactive protein (CRP; 1.05±1.99 vs. 0.15±0.21 mg/dL; P=0.004) were significantly higher in the frail than non-frail group. Multivariate analysis revealed that serum iron, CRP, and BUN were significant independent predictors of frailty (β=-0.069, 0.917, and 0.086, respectively).

Conclusions: Frailty status was significantly associated with iron, CRP, and BUN in stable older CVD patients. Composite biomarkers (inflammation, iron deficiency, and renal perfusion) may be useful for assessing frailty in these patients.

Key Words: Biomarker; Cardiovascular disease; Frailty; Inflammation; Older adult

Frailty is an important concept in geriatric medicine, and understanding its etiology has become a fundamental aspiration of many researchers in the field of aging. Frailty is an aging-associated syndrome that produces subclinical dysfunction across multiple organ systems, leading to increased risk of mortality. Between 25% and 50% of patients with cardiovascular disease (CVD) are frail. Moreover, according to a systematic review, the prevalence of frailty in heart failure (HF) ranges from 18% to 54%. The development of frailty is linked to various conditions, such as chronic inflammation and changes in the immune and endocrine systems, and is associated with an increased risk of death. CVD, including HF, is the leading cause of morbidity in frail patients. The Kihon Checklist (KCL) was developed by the Japanese Ministry of Health, Labour and Welfare to identify older people with frailty in need of care; it is a reliable tool for predicting general frailty in the elderly. Biomarkers identified through the implementation of multivariate strategies may be used to support the detection of frailty. The progression of these biomarkers can be tracked over time or in response to interventions, and reveals the onset of complications, such as mobility disability, at a very early stage. Therefore, there is an increasing need to identify and validate robust biomarkers for frailty. Inflammation, as indicated, for example, by serum concentrations of C-reactive protein (CRP), interleukin (IL)-6, and tumor necrosis factor (TNF)-α, has been implicated in the pathogenesis of both frailty and HF, although the pathophysiology of both disorders is complex and includes multiple deranged pathways that require further elucidation. However, the importance of general laboratory measurements in assessing frailty in older adults with CVD remains unclear. Therefore, the aim of this study was to evaluate which laboratory measurements indicate frailty in stable older adults with CVD.
**Methods**

**Study Population**
We conducted a cross-sectional study of patients who were admitted to the Cardiology Department of the National Center of Geriatrics and Gerontology, Japan, between August 2016 and December 2019. The study population consisted of 138 patients with CVD who were at least 65 years old and were able to perform cardiopulmonary exercise testing, undergo laboratory measurements, echocardiography, and a physical function evaluation, and complete questionnaires. These assessments were performed after the patients had been medically stabilized.

The inclusion criteria were structural heart disease consisting of coronary artery disease (having experienced angina pectoris or myocardial infarction, with or without a history of revascularization procedures), symptomatic HF (including conditions such as non-ischemic cardiomyopathy, ischemia, tachycardia, bradycardia, valvular disease, and hypertension), and others (see below). Non-ischemic cardiomyopathies were defined as ventricular myocardial abnormalities in the absence of coronary artery disease or valvular, pericardial, or congenital heart disease. Tachycardia and bradycardia included atrial, supraventricular, and ventricular arrhythmias, sick sinus syndrome, and atrioventricular block in the absence of structural heart disease. Valvular heart disease was diagnosed on the basis of hemodynamic or echocardiographic findings or a history of valvular or congenital cardiac surgery. Hypertension was defined as a systolic blood pressure ≥140 mmHg, diastolic blood pressure ≥90 mmHg, or a history of treatment for hypertension. Included in the “others” category were aortic disease, peripheral artery disease, and other vascular diseases. HF was defined as pulmonary venous congestion or edema on chest X-ray plus any symptoms (e.g., dyspnea, ankle swelling, peripheral edema, or fatigue).

Exclusion criteria were severe respiratory dysfunction (those receiving long-term oxygen therapy for respiratory disease), liver dysfunction (Child-Pugh Class C), stroke, renal dysfunction (albuminuria and glomerular filtration rate category G5), malignant tumors carrying a prognosis of <1 year, difficulty walking 10 m even with a walking aid, a Mini-Mental State Examination score <18, and living in a nursing care facility before admission.

Only patients who were stable after admission were enrolled in the study (**Figure 1**). Of 228 patients with unscheduled hospital admittance due to progressing cardiovascular disease to the Cardiology Department of the National Center of Geriatrics and Gerontology, Japan, between August 2016 and December 2019, 138 were included in the present study. CPX, cardiopulmonary exercise.

**Figure 1.** Study flowchart for the present analysis. Of the 228 patients with an unscheduled admission due to progressing cardiovascular disease to the Cardiology Department of the National Center of Geriatrics and Gerontology, Japan, between August 2016 and December 2019, 138 were included in the present study. CPX, cardiopulmonary exercise.
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Variables were compared between CVD patients with and without frailty using Student’s t-test for unpaired data. The Chi-squared test was used to assess the significance of differences between dichotomous variables. Spearman’s rank and Pearson’s correlation coefficients were used to assess the relationships between KCL score and laboratory measurements. Multivariate linear regression analyses were used to identify factors that were independently associated with KCL score; the multivariate model included all baseline variables that had a significant correlation with KCL score in the Pearson’s correlation. All analyses were performed using SPSS 17.0 (SPSS Inc., Chicago, IL, USA). Two-sided P<0.05 was considered statistically significant.

Results

Patient Characteristics
Baseline clinical characteristics of the patients are presented in Table 1. In all, 138 consecutive older adult patients with CVD (78 (57%) men; mean age 81.7±6.6
The median plasma B-type natriuretic peptide (BNP) concentration was 182 pg/mL (interquartile range 42–272 pg/mL) and the mean left ventricular ejection fraction (LVEF) was 57.8±14.3%. On the basis of KCL scores, 68.4% of patients were frail (mean KCL score for all patients 10.7±5.7). The KCL score was significantly associated with hemoglobin level. 

**Table 1. Baseline Characteristics of the Study Population** *(n=138)*

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>n=138</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>81.7±6.6</td>
</tr>
<tr>
<td>Male sex</td>
<td>78 (57)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>22.1±4.1</td>
</tr>
<tr>
<td>Atrial fibrillation</td>
<td>44 (32)</td>
</tr>
<tr>
<td>KCL</td>
<td>10.7±5.7</td>
</tr>
<tr>
<td>No. robust/pre-frail/frail</td>
<td>12/31/95</td>
</tr>
<tr>
<td>Resting SBP (mmHg)</td>
<td>136±22</td>
</tr>
<tr>
<td>Resting HR (beats/min)</td>
<td>72±12</td>
</tr>
</tbody>
</table>

**Comparisons of Non-Frail and Frail Patients**

Subjects were allocated to 1 of 2 groups based on the absence (n=43) or presence (n=95) of frailty (Table 2). Age was significantly higher in the frail than non-frail group (P=0.019). Similarly, plasma BNP concentrations were significantly higher in the frail than non-frail group (P=0.038); however, the estimated glomerular filtration rate (eGFR) was comparable in both groups. Serum iron concentrations were significantly lower in the frail than non-frail group (61.2±30.3 vs. 89.5±26.1 μg/dL, respectively; P<0.001). Blood urea nitrogen (BUN) was significantly higher in the frail than non-frail group (27.3±16.5 vs. 19.7±8.2 mg/dL, respectively; P=0.013), as was serum CRP (1.05±1.99 vs. 0.15±0.21 mg/dL, respectively; P=0.004).

**Correlations Between Biomarkers and Frailty**

The KCL score was significantly associated with hemoglobin, albumin, BUN, iron, CRP, eGFR, and BNP in the Spearman’s rank and Pearson’s correlation analyses (Table 3). We then analyzed these significantly associated parameters for KCL score in multivariate analyses and found that serum iron and CRP concentrations and BUN were significant independent predictors of frailty (β=−0.069, 0.917, and 0.086, respectively; Table 3).

**Discussion**

The main aim of the present study was to elucidate the relationship between general biomarkers and frailty in older adults with CVD. Here, we report for the first time that serum iron and CRP and BUN concentrations are strongly associated with the presence of frailty in older adults with CVD. Frail patients scored significantly more poorly than non-frail patients on these items related to nutrition, inflammation, and protein catabolism. However, the frail group had people on a gradual scale from mild to severe frailty. In fact, the KCL scoring system runs from 0 to 25 points. Therefore, we thought it may be more important data-wise to correlate the baseline characteristics with the KCL score. Furthermore, we wanted to show which laboratory data contributed to the KCL scores. Among the blood biomarkers, iron, CRP, and BUN were regulatory factors independent of the deterioration of KCL in older adults with CVD.

**Iron**

Aging-related comorbidities are an emerging problem in patients with CVD. Among them, iron deficiency is an important therapeutic target regardless of the concomitant hemoglobin level. A recent study confirmed the relationship between reduced iron concentration and the occurrence of frailty syndrome. Iron deficiency affects up to 50% of CVD patients, and its association with poor quality of life, impaired exercise tolerance, and increased mortality rates has been widely established. Current European Society of Cardiology Guidelines for CVD recommend a diagnostic workup for iron deficiency in all CVD patients. Iron deficiency has detrimental effects in patients with coronary artery disease, HF, or pulmonary hypertension, and possibly in patients undergoing cardiac surgery. Perturbations of iron metabolism resulting in changes in iron status are observed in a variety of age-related medical conditions, including kidney disease, cancer, CVD, and neurodegenerative diseases.
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CRP
In older adults, there is a significant association between elevated levels of high-sensitivity CRP and the development of HF. Increased serum CRP concentrations are positively associated with increased severity of frailty in people aged >75 years, and increasing frailty is also associated with increasing TNF-α and IL-6 levels. Here, we chose to perform only those standard laboratory measurements that are used for health insurance purposes, so we did not check TNF-α and IL-6 levels. However, even in the absence of clinical signs, CRP may be useful in detecting frailty in older adult patients with CVD.

BNP
In patients with chronic HF, the BNP concentration provides powerful prognostic information regarding survival and deterioration of functional status. In the Valsartan Heart Failure Trial, patients with the greatest rise in BNP concentrations despite therapy had the highest rates of morbidity and mortality. Notably, in the present study

Table 2. Comparisons of Non-Frail and Frail Groups

<table>
<thead>
<tr>
<th></th>
<th>Non-frail group (KCL &lt;8; n=43)</th>
<th>Frail group (KCL ≥8; n=95)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>79.1±7.6</td>
<td>83.1±6.1</td>
<td>0.019</td>
</tr>
<tr>
<td>Sex (male/female)</td>
<td>25/18</td>
<td>53/42</td>
<td>0.724</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>24.1±3.6</td>
<td>21.0±3.3</td>
<td>0.001</td>
</tr>
<tr>
<td>Diuretics</td>
<td>17 (39)</td>
<td>61 (64)</td>
<td>0.056</td>
</tr>
<tr>
<td>Tolvaptan</td>
<td>9 (22)</td>
<td>22 (23)</td>
<td>0.878</td>
</tr>
<tr>
<td>ACE-I/ARBs</td>
<td>22 (52)</td>
<td>41 (43)</td>
<td>0.539</td>
</tr>
<tr>
<td>β-blockers</td>
<td>9 (22)</td>
<td>30 (32)</td>
<td>0.877</td>
</tr>
<tr>
<td>Spironolactone</td>
<td>9 (22)</td>
<td>22 (23)</td>
<td>0.878</td>
</tr>
<tr>
<td>Anticoagulants</td>
<td>17 (39)</td>
<td>36 (38)</td>
<td>0.948</td>
</tr>
<tr>
<td>Clinical data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LVEF (%)</td>
<td>62.1±9.4</td>
<td>55.7±15.9</td>
<td>0.082</td>
</tr>
<tr>
<td>E/e’</td>
<td>15.1±7.2</td>
<td>16.1±6.7</td>
<td>0.534</td>
</tr>
<tr>
<td>LAD (mm)</td>
<td>40.7±8.3</td>
<td>39.1±6.0</td>
<td>0.402</td>
</tr>
<tr>
<td>WBC (×10⁹/L)</td>
<td>57.3±16.3</td>
<td>59±22.4</td>
<td>0.741</td>
</tr>
<tr>
<td>Hb (g/dL)</td>
<td>13.3±1.9</td>
<td>11.5±1.9</td>
<td>0.001</td>
</tr>
<tr>
<td>Pt (g/dL)</td>
<td>20.4±5.1</td>
<td>20.1±7.3</td>
<td>0.87</td>
</tr>
<tr>
<td>TP (g/dL)</td>
<td>7.2±0.5</td>
<td>6.7±0.6</td>
<td>0.547</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>4.0±0.3</td>
<td>3.6±0.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>AST (IU/L)</td>
<td>22±4.3</td>
<td>23.5±18.2</td>
<td>0.705</td>
</tr>
<tr>
<td>ALT (IU/L)</td>
<td>21.7±11.2</td>
<td>20.7±40.3</td>
<td>0.906</td>
</tr>
<tr>
<td>LDH (IU/L)</td>
<td>205.1±35</td>
<td>204.5±58.5</td>
<td>0.963</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>19.7±8.2</td>
<td>27.3±16.5</td>
<td>0.013</td>
</tr>
<tr>
<td>Cr (mg/dL)</td>
<td>0.9±0.2</td>
<td>1.3±0.7</td>
<td>0.004</td>
</tr>
<tr>
<td>TC (mg/dL)</td>
<td>186±32</td>
<td>173±37</td>
<td>0.164</td>
</tr>
<tr>
<td>TG (mg/dL)</td>
<td>127.2±58.9</td>
<td>119.3±67.8</td>
<td>0.637</td>
</tr>
<tr>
<td>Fe (μg/dL)</td>
<td>89.5±26.1</td>
<td>61.2±30.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CRP (mg/dL)</td>
<td>0.15±0.21</td>
<td>1.05±1.99</td>
<td>0.004</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>6.1±0.4</td>
<td>6.2±0.8</td>
<td>0.602</td>
</tr>
<tr>
<td>BNP (pg/mL)</td>
<td>123.4±143.6</td>
<td>221.9±194.6</td>
<td>0.038</td>
</tr>
<tr>
<td>eGFR (mL/min/1.73m²)</td>
<td>56.4±14.1</td>
<td>47.7±23</td>
<td>0.101</td>
</tr>
</tbody>
</table>

Table 2. Comparisons of Non-Frail and Frail Groups

Unless indicated otherwise, data are given as the mean±SD or n (%). ALT, aspartate aminotransferase; AST, alanine aminotransferase; BUN, blood urea nitrogen; Cr, creatinine; CRP, C-reactive protein; E/e’, ratio of early transmitral flow velocity to early diastolic mitral annular velocity; Fe, iron; LAD, left atrial dimension; LDH, lactate dehydrogenase; Pt, platelets; TG, triglycerides; WBC, white blood cell count. Other abbreviations as in Table 1.

reflect poor global health status, rather than solely being an indicator of the severity of acute illness or unstable chronic disease. Silverberg et al first described the term “cardiorenal anemia syndrome”. This term has been widely used in recent years, now that we understand the importance of the associations among HF, renal failure, and anemia. High BUN has a negative effect on patient survival and reflects the extent of catabolism. In the acute phase of a critically ill patient, this catabolism may be beneficial, providing amino acids for hepatic gluconeogenesis and for the synthesis of proteins involved in immune functions, but persistent hypercatabolism in critically ill patients results in decreased immune function, which leads to increased mortality. In addition, Kameda et al reported that metabolite profiles efficiently distinguish frailty from non-frailty. Oxidative stress resulting from diminished antioxidant levels could be a key vulnerability for the pathogenesis of frailty, exacerbating illnesses related to human aging. Therefore, BUN is considered an integral marker of tissue necrosis, protein catabolism, and renal perfusion.
we found that serum iron and CRP concentrations and BUN were superior to BNP concentrations for the diagnosis of frailty in older adults with stable CVD. Ninety-one per cent of our 138 patients were admitted because of worsening HF. Even in patients in a stable condition after medical treatment, BNP is supposed to indicate disease severity or prognosis in HF. However, although BNP was significantly correlated with KCL, it was not a significant independent predictor of frailty according to the KCL in older adult patients with CVD. In assessing frailty, we need to pay attention to the evaluation of laboratory items other than powerful conventional prognostic markers, such as BNP, in the elderly CVD population.

**Frailty and CVD**

Frailty is a multidimensional syndrome based on an aggregate susceptibility to adverse health outcomes due to age- and disease-related deficits that accumulate across multiple domains. It is also associated with mortality. Several tools have been developed for assessing frailty, but there is no international standard measurement. The KCL, a self-administered questionnaire, is considered useful for frailty screening in older adult populations. As mentioned above, further research is needed to develop accurate composite markers that take these multimorbidities into account.

**Clinical Implications**

Laboratory measurements are commonly evaluated in daily practice because they are inexpensive, repeatable, and non-invasive tests. In the present study, we did not include specialized items relevant to frailty, such as IL-6 and TNF-α, in the laboratory measurements because we wanted to test only those biomarkers used in general assessments. To the best of our knowledge, the present study is the first to have investigated the ability of these standard laboratory measurements to detect frailty in older adults with stable CVD. The primary goals of CVD therapy are to improve quality of life and extend survival. The recognition of frailty within the medical community has created the need for diagnostic tests to determine when a patient’s physical ability has deteriorated.

**Study Limitations**

The present study was a single-center study with a small sample size. Moreover, we did not assess repeated measures over time or follow the incidence of cardiac events in the enrolled patients. We did not check ferritin levels, which are associated with iron levels. Nor did we check
IL-6 and TNF-α levels, which are also related to frailty. We also did not assess changes in the trajectory of exercise capacity or frailty due to medical intervention or cardiac rehabilitation.

Conclusions
Frailty status was significantly associated with serum iron and CRP concentrations and BUN in stable older adults with CVD. Composite biomarkers for inflammation, iron deficiency, and renal perfusion may be useful for assessing frailty in stable older adults with CVD.

Acknowledgments
The authors are indebted to the staff of the National Center for Geriatrics and Gerontology, particularly physiological technician Kimiko Hori.

Sources of Funding
This study was supported by 2016–2018 Ministry of Health, Labor and Welfare Geriatrics and Gerontology-sponsered research funds.

Disclosures
T.M. is a member of Circulation Reports’ Editorial Team. The remaining authors have no conflicts of interest to disclose.

Author Contributions
A.H., A.S., I.K., T.M., and H.A. supervised the research and prepared the text. K.H. and K.S. evaluated frailty. A.H., K.N., M.K., N.S., and A.S. evaluated patients. All authors reviewed the text and agree with the paper’s publication.

IRB Information
The present study was approved by the Ethics and Conflict of Interest Committee of the National Center for Geriatric and Gerontology (Reference no. 1272).

References


