Christine J. Chung
Doris Duke Clinical Research Fellow
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Hepcidin as a Novel Biomarker of Congestive Hepatopathy in Advanced Heart Failure Patients

I. Study Purpose and Rationale

Background:

Heart Failure

Heart failure (HF) is a prevalent disease affecting approximately five million Americans and 10% or more of those aged 70 years or older [1]. Despite significant advances in therapy, HF remains a significant cause of hospital admissions and patients who are symptomatic have a one-year mortality rate approaching 45% [2]. The high mortality rate in HF patients may be attributable at least in part to frequently present co-morbidities [3].

Congestive Hepatopathy in the Setting of Advanced HF

Congestive hepatopathy refers to the spectrum of chronic liver injury caused by passive congestion in the setting of right-sided heart failure or any other cardiopulmonary condition leading to elevated central venous pressure. Liver function abnormalities are common in HF patients and are associated with a worse prognosis [3]. It is difficult, however, to estimate the true incidence of congestive hepatopathy because the condition is often subclinical and fibrosis can develop before significant abnormalities in liver function tests (LFTs) become evident [4].

A study of 59 patients with advanced HF undergoing evaluation for either cardiac transplantation or LVAD implantation revealed the vast majority had histopathological evidence of hepatic fibrosis [5]. When compared to advanced HF patients without hepatic fibrosis, those with fibrosis were more likely to have more severely impaired LV systolic function with ejection fraction <35%, indicating a correlation between worsening hemodynamic compromise and increasing prevalence of liver dysfunction. Subset analysis of patients who underwent liver biopsy at the time of evaluation for LVAD or transplant showed patients with no or mild fibrosis were more likely to proceed to surgery and to be alive at the time of the study. Thus, knowledge of the presence and severity of liver damage in HF patients has important implications for prognosis and clinical decision-making.

Management of congestive hepatopathy is centered on treating the underlying cardiac disease and restoring forward cardiac output, which improves LFTs and reduces ascites. Both LVAD implantation and cardiac transplantation have been shown to improve and even reverse the congestive liver injury seen in HF. It seems reasonable, therefore, to suspect that patients

with evidence of congestive hepatopathy who are refractory to medical therapy and suitable operative candidates could benefit from earlier consideration for surgical management.

Need for Novel Biomarkers of Congestive Hepatopathy in Patients with Advanced HF

Since existing laboratory studies inadequately reveal the presence of congestive hepatopathy, a sequela of HF associated with worse clinical outcomes that can preclude patients from receiving a cardiac transplant or LVAD, there is a need for more sensitive biomarkers that alert clinicians to the presence of hepatic injury in the setting of HF. If such a biomarker could be shown to detect the presence of congestive hepatopathy at earlier stages of fibrosis, it could join the existing panel of laboratory tests used to monitor hepatic function in HF patients and aid in determining candidacy for LVAD implantation or cardiac transplantation.

Hepcidin as a Potential Biomarker

Hepcidin is a peptide synthesized in the liver that plays a key role in iron homeostasis. It acts to regulate intestinal uptake of dietary iron and efflux of intracellular iron to the plasma from macrophages of the reticuloendothelial system by binding to the only known cellular iron transporter, ferroportin, and causing its internalization. Levels of hepcidin are increased by iron loading and decreased in anemia, hypoxia, and erythropoiesis. During infection and inflammation, hepcidin is thought to be induced by cytokines such as IL-6, TNFα, and IL-1β and subsequently to contribute to anemia of inflammation [6].

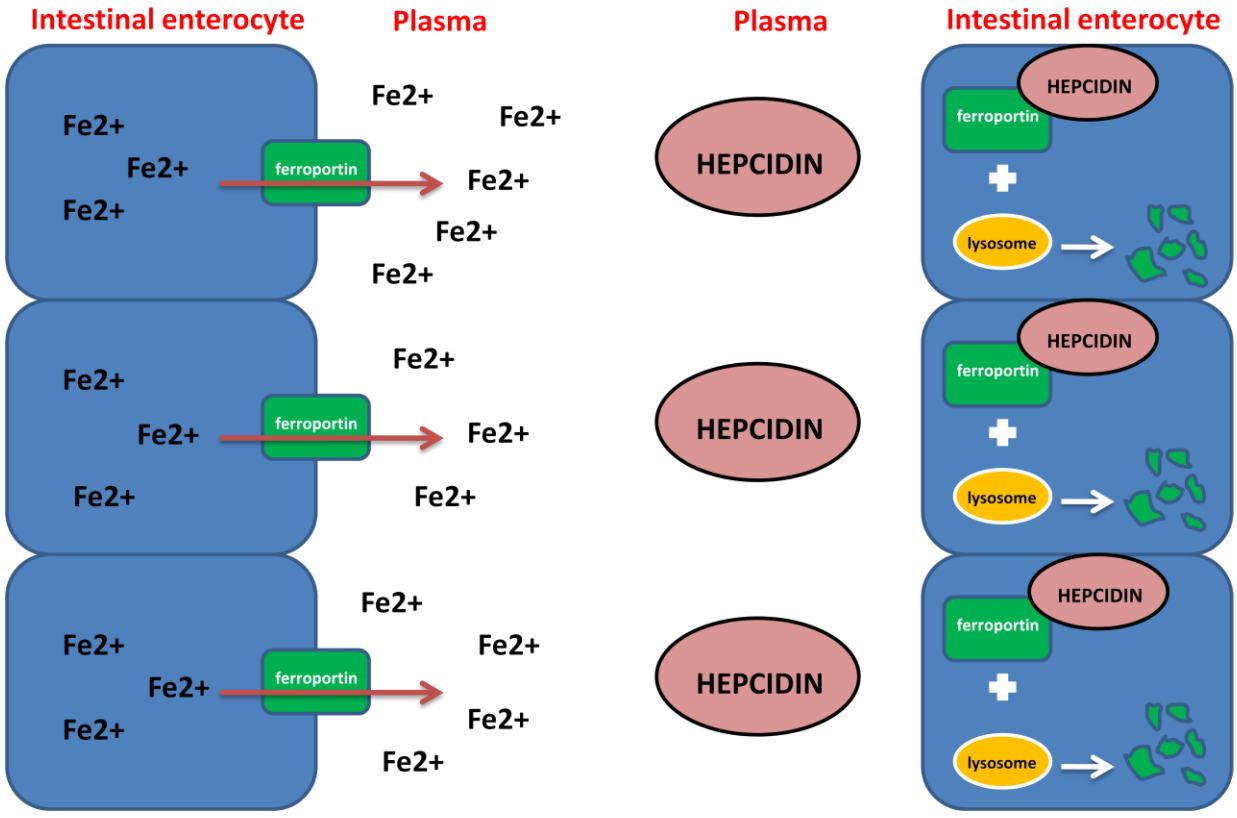

Figure 1. Mechanism of action of hepcidin and its role in iron regulation

The purpose of our study is to investigate the potential role of hepcidin as a novel biomarker to aid in the prognostic assessment of patients with heart failure. Morrow and de Lemos have proposed three criteria by which to gauge the clinical potential of a novel cardiovascular biomarker. First, it should be easily measurable. Second, it should provide new information not available to the clinician through other channels. And finally, it should affect clinical management [7].

As recently as 2008, the methodologies for measuring serum levels of the physiologically relevant form of hepcidin were cumbersome and not readily available [8]. However, since then, several commercially available enzyme-linked immunosorbent assay (ELISA) kits for the detection of human hepcidin have entered the market, and as research on the potential link between hepcidin and myriad diseases is a very active area, there is reason to presume that such kits will continue to become more widely available and at increasingly lower cost.

We first propose to investigate whether hepcidin levels do indeed correlate with liver function tests and the presence of congestive hepatopathy. Then, we will compare its sensitivity, specificity, and positive predictive value with those of the currently available markers of hepatic damage in order to determine whether it is an equally good or better biomarker. If hepcidin correlates with the presence of congestive hepatopathy at earlier stages of hepatic damage and fibrosis, it most certainly will provide new information that will impact management of HF patients.

Hypothesis #1: Congestive hepatopathy and subsequent inflammation in the liver due to HF upregulate hepcidin levels.

Hypothesis #2: Elevated hepcidin levels will normalize after hemodynamic correction by either LVAD placement or cardiac transplantation.

Hypothesis #3: Hepcidin levels will predict reversibility of liver damage in advanced HF patients as well as clinical outcomes.

II. Study Design

This will be a retrospective analysis of patients seen in the CUMC/NYPH Heart Failure Clinic. We have a database of serum and plasma samples collected from 2008 until the present under IRB Protocol AAAF1150, "Blood analysis in patients with heart failure before and after LVAD placement, after cardiac transplantation and controls." We currently have approximately 200 patients in our database. Of those, a sub-group of an estimated 50-60 patients have serial samples taken before and after LVAD implantation. The patients will be sorted into one of five categories: controls, mild to moderate HF, severe HF, HF patients on LVAD support, and HF patients post-cardiac transplantation.

III. Statistical Analysis

Statistical analyses will be performed using a commercially available software program, such as SPSS. A prior study of hepcidin levels in HF patients found values of serum hepcidin to not follow a normal distribution [9]. If we find the same result in our data, we will subject it to logarithmic transformation. The unpaired Student's t-test will be used to analyze the difference in the change in hepcidin levels between the two cohorts. Univariate linear regression analysis will be used to determine whether hepcidin correlates with each of the individual markers of liver function. P-values of <0.05 will be considered statistically significant.

As this is a retrospective analysis, the number of subjects is pre-determined. We estimate a total sample of 50-60 patients in our existing database with serum taken before and after LVAD implantation. With a sample size of 50 patients, the smallest difference between groups that would be statistically significant is 35ng/mL of hepcidin. With a sample size of 60 patients, the smallest effect that would be statistically significant is 32ng/mL of hepcidin. A validation study of one hepcidin ELISA found that the average median value of hepcidin in several different chronic inflammatory conditions such as multiple myeloma and chronic kidney disease was approximately 335ng/mL, whereas the median in healthy volunteers was 88.5ng/mL [8]. To the best of our knowledge, there have not been any studies conducted in which the change in hepcidin caused by a therapeutic intervention was measured. Thus, it is difficult to predict the magnitude of the change in hepcidin upon LVAD implantation in our patients. However, as there is a much larger difference between median values in diseased versus normal patients than the effect size we could detect at 80% power, a sample of 50-60 patients should be sufficient to detect any significant difference between the two cohorts.

IV. Study Procedures

General characteristics such as age, gender, etiology of heart failure, New York Heart Association (NYHA) functional class, and past medical history will be collected through chart review.

Values for markers of liver function such as ALT, AST, alkaline phosphatase, γ-glutamyl transpeptidase (GGT), direct, indirect, and total bilirubin, albumin, and prothrombin time will also be collected from the medical record.

ELISA will be employed as the method of measuring serum levels of the 25-amino acid, bioactive form of hepcidin.

V. Study Drugs

N/A.

VI. Medical Devices

No medical devices will be implanted for the purposes of this study.

VII. Study Questionnaires

N/A.

VIII. Study Subjects

The study will be conducted on serum samples obtained from patients seen in the CUMC/NYPH Heart Failure Clinic.

We will not exclude patients with co-morbidities known to be common in HF patients and associated with a chronic inflammatory state such as chronic kidney disease and anemia. In our statistical analysis, we will control for the presence of co-existing inflammatory conditions in the cohort of patients with HF and congestive hepatopathy.

IX. Recruitment of Subjects

This is a retrospective study and does not entail any additional patient recruitment.

X. Confidentiality of Study Data

All patient samples collected in our database are linked to a study identification number, which allows patient data to remain confidential. Any information obtained for the purposes of this study will be kept confidential in compliance with the standards set by HIPAA.

XI. Potential Conflict of Interest

None.

XII. Location of the Study

The study will be conducted in the laboratory of Dr. P. Christian Schulze in the CUMC/NYPH Presbyterian Hospital Building (PH 8-405).

XIII. Potential Risks

This is a retrospective study and does not pose any additional risks to the patients.

XIV. Potential Benefits

This study is unlikely to bring any direct benefit to the patients involved. However, we hope the information obtained will be beneficial to other HF patients in the future.

XV. Alternative Therapies

This is a retrospective study and does not involve any therapeutic interventions.

XVI. Compensation to Subjects

None.

XVII. Minors as Research Subjects

No minors will be involved in this study.

XVIII. Radiation or Radioactive Substances

N/A.

XIX. References

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