Treatment of HFpEF with nitrate supplement: A double-blind, placebo controlled trial including patients with atrial fibrillation

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INTRODUCTION

Heart failure (HF) is the most common principal diagnosis for hospital admission in patients over 65 years old. [10] According to the 2015 American Heart Association Statistical Update, one in nine deaths has HF mentioned on the death certificate.[1] It is estimated that 6.5 million Americans ≥20 years of age have heart failure. Projections show the prevalence of HF will increase 46% from 2012 to 2030. [11] There are two types of HF, those with reduced ejection fraction (HFrEF) and those with preserved ejection fraction (HFpEF). Approximately half of patients with the clinical syndrome of HF have preserved systolic function. HEpEF is becoming more prevalent with aging of the population and obesity. [1]

While there are nine American Heart Association class I recommendations for the treatment of HFrEF, there are only two class I recommendations for the treatment of HFpEF, which are controlling blood pressure and the use of diuretics to relieve symptoms.[3] Spironolactone may also be particularly effective. [4]

Exercise training is another approach to improving symptoms, however it may be poorly tolerated. Nitrate supplement in the form of concentrated beetroot juice was recently shown to improve exercise tolerance in patients with HFpEF.[5] Beetroot juice contains high concentration of NO_3 which is absorbed in the gut, concentrated in the salivary glands and secreted in saliva. This is metabolized to NO_2 by symbiotic bacteria on the tongue. NO_2 is swallowed, absorbed and enters the blood stream, where, in the presence of deoxyhemoglobin and acidosis, it is further reduced to NO resulting in intense vasodlitation and mitochondrial activation.

Patients with diastolic dysfunction are often asymptomatic at rest but complain of dyspnea with exertion. Increase in heart rate with exercise causes reduced diastolic filling time and increases left sided filling pressure. Borloug, et al demonstrated this with right heart catheterization and supine exercise in patients with diastolic dysfunction. Infusion of NO_2 resulted in decreased filling pressures and increased cardiac output. [9]

Neo40 is a new product made from concentrated beetroot juice in the form of a lozenge designed to dissolve on the tongue. This bypasses the enterosalivary circulation resulting in immediate production of NO instead of a delay of several hours seen when drinking beetroot juice. It also caontains nitrite reductase which promotes the final step in NO production.

Nitroglycerine in the form of isosorbide mononitrate is oxidized by endothelial nitric oxide synthetase causing widespread vasodilatation and hypotension. It was recently shown to be ineffective in treating

HFpEF. [6] NO_3 supplement causes vasodilatation only in the setting of hypoxia and acidocis resulting in targeted vasodilatation.

NO may have direct effect on diastolic function via titin. Titin is a giant multi-functional sarcomeric filament that is important in maintaining the structural integrity of the contracting sarcomere[7] and together with the extracellular matrix for defining diastolic stiffness.[8] Titins elasticity is dependent on phosphorilation which in turn is dependent on NO.

We propose to study the effect of Neo40 on patients with HFpEF.

STUDY OBJECTIVES

The objective of this project is to determine if NEO40, when consumed twice daily by subjects with HFpEF, will increase exercise tolerance, decrease symptoms and improve quality of life for patients.

Primary endpoint:

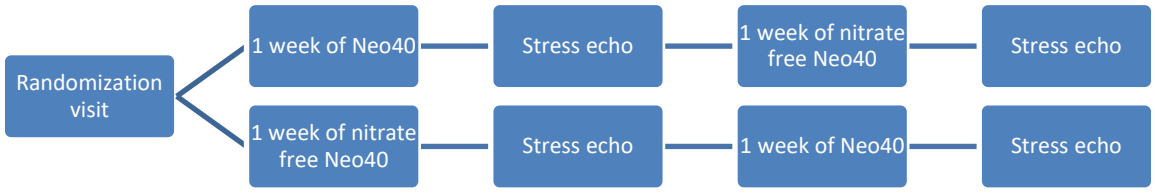
- Exercise Echocardiogram
  - Total time on the treadmill
  - Metabolic equivalents (METS)[6]
  - Average E/E'
  - Estimation of right ventricular systolic pressure using standard echo measurements
  - Quality of life questionnaire changes

Secondary endpoints:

- Change in blood pressure measurements
- Change in heart rate measurements
- Change in 6 minute walk test results

STUDY DESIGN

This is a double blind cross over study. Each subject will consume the NEO40 for two weeks; one week with NEO40 and one week with nitrate free NEO40 (placebo). The project will randomly assign which the subject consumes first.



Approximately 25 subjects will be enrolled at Pen Bay Medical. Interim analysis will be done when 12 subjects have completed the study or at approximately six months, whichever comes first. The protocol will be reevaluated to determine if any amendment should be made as well as to assess safety and efficacy of the NEO40.

Inclusion Criteria

1. Males and females aged 18 years or greater
2. Diagnosis of HFpEF, defined as:
    a. Symptomatic with one or more of the following: orthopnea, paroxysmal nocturnal dyspnea, lower-extremity edema, dyspnea on exertion; and
    b. Ejection fraction > 50%; and
    c. Ratio of early mitral inflow velocity to septal tissue Doppler velocity >8; and
    d. One or more of the following:
        i. Left atrium measurement > 34 mL/m^2
        ii. Elevated N-terminal pro-brain natriuetic peptide level within the past 12 months
        iii. Long-term loop diuretic use for control of symptoms
        iv. Elevated filling pressures on prior cardiac catheterization
3. Stable medical therapy, defined as: no change in cardiac medications within 30 days
4. Willing to comply with the protocol and provide written informed consent

Exclusion Criteria:

1. Non-cardiac condition causing limitation of exercise tolerance
2. Acute coronary syndrome, myocardial infarction or cardiac revascularization within 60 days
3. Clinically significant valvular disease, defined as moderate-severe or severe stenosis or insufficiency
4. Significant ischemia seen on stress testing within the past 12 months which was not revascularized
5. Subjects who have taken an investigational medication within the past 30 days
6. History of allergic reaction to beets
7. Unwilling to comply with the protocol requirements
8. Systolic blood pressure of <100 at screening
9. The subject has a significant medical condition and/or conditions that would interfere with treatment, safety or compliance with the protocol

STUDY PROCEDURES

Screening Visit: After the subject has provided informed consent, the Study Coordinator will collect basic demographic information; the patient's cardiovascular history and measure subject's heart rate and blood pressure. The subject will complete the World Health Organization Quality of Life (WHOQOL) -

BREF questionnaire. The subject will then be randomized by an unblinded pharmacy staff member to either receive the NEO40 or the nitrate-free NEO40 for the first week and 16 doses will be dispensed to the subject, along with a dosing diary and written instructions regarding the dosing and diary.

Visit 2: Seven days (+/- 2 days) later, the subject will return for the second study visit. Blood pressure and heart rate will be measured. The quality of life questionnaire will be re-administered and the diary will be collected at this visit. The exercise echocardiogram will be performed by one of the investigators. The exercise echocardiogram will be done 2-3 hours after the subject ingests their last dose of NEO40 for this study period. Another 16 doses of either NEO40 or nitrate-free NEO40 will be dispensed to the subject along with another diary.

During the exercise testing, the Borg scale of perceived exertion will be utilized and recorded with each subject approximately one minute prior to each incremental stage increase. The goal will be to have each subject exercise to a perceived "ten" on the one-to-ten scale.

Visit 3: Seven days (+/- 2 days) later, the subject will return for the end-of-study visit. Blood pressure and heart rate will be measured. The quality of life questionnaire will be re-administered and the second diary will be collected. The exercise echocardiogram will be performed by one of the investigators. The exercise echocardiogram will be done within 30 minutes of the subject ingests their last dose of NEO40 for this study period.

RISKS

Exercise Echocardiogram

The risks of the exercise echocardiogram include fainting, disorders of heartbeats, abnormal blood pressure response, chest pain and, very rarely, a heart attack. A cardiologist will be in attendance during the exercise echocardiogram.

Neo40

A subject could be allergic to beets.

CONFIDENTIALITY

Subject identity will remain confidential. Records will be kept in the locked Clinical Research. No reports which come from this study will identify the subjects in any way. Only basic demographics, such as age and gender will be reported.

STORAGE OF STUDY MATERIALS

NEO40 will be stored at room temperature in the pharmacy of Penobscot Bay Medical Center.

SUBJECT STIPENDS OR PAYMENTS AND COSTS

Subjects will not be compensated for participation in this study. Neo40 will be provided free of charge to the subjects. There will be no charge to the subjects, nor their insurance plan, for any procedures related to this study.

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