Cardiac Ryanodine Receptor Phosphorylation as a Risk Factor for Sudden Cardiac Death in Congestive Heart Failure

Kenji A Fujita

A. Study Purpose and Rationale

Congestive heart failure is a major and increasing source of morbidity and mortality in industrialized societies, with an estimated prevalence of close to five million cases in the United States[1]. In patients with systolic dysfunction, causes of death are divided roughly equally between progressive pump failure and sudden cardiac death due to fatal arrhythmias[2] . Despite advances in treatment, five-year survival remains about 35%[3]. Death from arrhythmias has been particularly resistant to advances in treatment. In the CONSENSUS trial, sudden cardiac death was unaffected by optimal medical therapy[4].

One approach to the prevention of sudden cardiac death has been to identify subpopulations of heart failure patients who benefit from implantable cardiac defibrillators (ICDs). Multiple studies have shown a strong mortality benefit to ICDs in patients with low ejection fractions [5,6,7] . However, a common criticism of these studies is that they are overly broad in the definition of their patient populations, leading to the possible inclusion into their recommendations of patients who do not benefit from ICDs. Thus, an important area of investigation is to more finely segment populations shown to benefit from ICDs to identify sub-populations at particularly high risk.

Heart failure leads to a pathological hyperadrenergic state. A molecular consequence of chronic overstimulation of the P-adrenergic receptor is upregulation of cAMP-dependent protein kinase activity, with hyperphosphorylation of its targets. One target of interest is the cardiac ryanodine receptor (RyR2). Marx et al have shown that RyR2 is hyperphosphorylated in decompensated heart failure, with concomitant defects in calcium conductance properties[5] . The validity of this hypothesis has recently been supported by recent results showing promise for strategies to normalize RyR2 function [6] .

This proposed study will aim to demonstrate level of RyR2 phosphorylation as a risk factor for sudden cardiac death in advanced heart failure.

B. Study Design and Statistical Analysis

The study will enroll 320 patients with class IV congestive heart failure presenting to our electrophysiology lab for implantation of a device (usually either an ICD or a biventricular pacemaker). The size of the study is based on a chi-square analysis assuming an average event rate of 10% at one year, based on the results of the CONSENSUS trial[4]. The study is powered at the level of 80% to detect a difference of 10 percentage points between the two groups. This difference was chosen as the minimal difference of clinical interest for the purpose of risk stratification. Patients will be assigned to the two arms: low phosphorylation and high phosporylation; after analysis of an endocardial biopsy taken during the procedure. At the end of the study, the chi-square test on proportions will be used to look for a difference between the two groups of an event, defined as an episode of ventricular tachycardia or ventricular fibrillation sufficient to cause the device to fire.

C. Study Procedure

Patients will have an endocardial biopsy taken from the apex of the right ventricle at the time of device implantation. Samples will be frozen and stored until enrollment is completed. At this time, immunoblot analysis will be performed on all samples to determine reactivity to the anti-RyR2-2809P

antibody. Degree of reactivity will be gauged by optical density of the RyR2 band on autoradiogram, normalized to amount of protein loaded, as determined by reactivity to a standard anti-RyR2 antibody. Reactivity will then be expressed as a percentage of optical density, with the maximally reactive sample standardized to 100%.

Patients will be followed at one year, at which time their devices will be interrogated for episodes of ventricular tachycardia or ventricular fibrillation causing the device to fire.

At the end of the study, patients will be separated into "above average phosphorylation" and "below average phosphorylation" arms. The number of patients with events in each arm will be compared and analyzed for significant differences.

D. Study Drugs

None

E. Medical Device

None

F. Study Questionnaires

None

G. Study Subjects

Inclusion criteria:

- Over 18 years old
- Class IV CHF
- EF<30%
- Indication for implantation of an ICD or biventricular pacemaker

Exclusion criteria:

- Treatment with antiarrhythmic medications
- Known arrhythmia in past 60 days
- Life expectancy less than a year
- Creatinine > 2.0 or K > 6.0
- Presence of any metabolic disease expected to alter serum levels of calcium, potassium, or magnesium
- MI in past 60 days
- Plans to reside outside of the study area at one year from randomization

H. Recruitment of Subjects

Subjects under evaluation for device placement will be identified by their primary cardiologists and referred to the study coordinator.

I. Confidentiality of Study Data

All study data will be coded in a way that does not include identifying information which could be traced back to an individual patient. Data will be safeguarded as encrypted files by the study coordinator.

J. Potential Conflict of Interest

None

K. Location of Study

The study will be performed at New York Hospital, both at Columbia Presbyterian Medical Center and at Weill Medical Center. All biopsies will be taken by qualified electrophysiologists under standard conditions. Immunoblot analysis will be performed in a molecular biology laboratory at Columbia Presbyterian Medical Center.

L. Potential Risks

The only significant additional risk is that associated with the harvesting of the endocardial biopsy. This should pose minimal risk, as it is an established technique with which both medical centers have substantial experience. In addition, all patients will incur the risks inherent in the procedures they are undergoing. These risks will be explained in the standard consent forms for those procedures.

M. Potential Benefits

Given the fact that subjects will already be receiving devices, they are unlikely to benefit directly from the study. However, they will be helping to add to the scientific data base which may benefit future patients.

N. Alternative Therapies

N/A

O. Compensation to Subjects

No compensation is to be offered.

P. Costs to Subjects

This study will incur no additional costs to the subjects

Q. Minors as Research Subjects

No minors will be included in the study.

R. Radiation or Radioactive Substances

Patients will be exposed to low doses of ionizing radiation during fluoroscopy. The risks posed by this radiation exposure are explained in the standard consent forrns used before fluoroscopy.

S. References

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5. Moss AJ et al. Improved survival with an implanted defibrillator in patients with coronary disease at high risk for ventricular arrhythmia. *N Engl J Med* 1996; 335:1933-1940.
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8. Marx SO et al. PKA phosphorylation dissociates FKBP 12.6 from the calcium release channel (ryanodine receptor): defective regulation in failing hearts. *Cell* 2000 May 12; 101(4):365-76.
9. Wehrens XH et al. Protection from cardiac arrhythmia through ryanodine receptor-stabilizing protein calstabin2. *Science* 2004 Apr 9; 304(5668):292-6.