RNA Binding Protein RBM20 Regulates Gene Network Associated with Heart Muscle Contraction

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Introduction
Ribonucleic acid-binding motif protein 20 (RBM20) is highly expressed in skeletal and heart muscles, notably the heart. RBM20 deficiency can lead to heart muscle disease known as dilated cardiomyopathy (DCM), a leading cause of heart failure and a significant source of mortality and morbidity worldwide. RBM20 helps regulate gene splicing, a process in which one gene encodes for several proteins with similar functions. Given the role of RBM20 in the regulation of gene splicing, the identification of its downstream RNAs (ribonucleic acids) would shed light on human heart failure progression and provide strategies for molecular therapy. With these efforts, more than 30 downstream RNAs of RBM20 have been identified including a major titin RNA, a gene expressed in heart muscle for muscle contractile function; however, it is unknown whether RBM20 also regulates gene expression levels or changes in the heart.

Objectives
The objectives of this study are to investigate the expression level of how many and what genes are regulated by RBM20 in the heart muscle.

Materials and Methods
We used rats as animal models for our research. They were maintained on standard rodent feed using protocols approved by the University of Wyoming and University of Wisconsin–Madison animal use and care committees. Hearts were obtained from animals that were one, 20, and 49 days old after the animals were euthanized. The left chambers were dissected and used for this study because of their functional importance of pumping blood to peripheral tissues. RNAs and individual muscle cells were isolated from the heart tissues for gene expression and muscle cell contractile measurements (Fig. 1).

Results and Discussion
We compared gene expression in heart muscle from the rats with RBM20 expression (wild-type) and the rats without RBM20 (RBM20 knockout). We found that fewer genes are regulated by RBM20 at a younger age when compared to older ages. The expression level of about 90% of the regulated genes is increased. We also found that the upregulated genes are associated with heart failure. The proteins encoded by these upregulated genes can bind to a protein titin, a major muscle protein that can cause heart failure with changed expression level. Particularly, we revealed that RBM20 changes calcium levels in heart muscle and results in abnormal muscle contraction. These results suggest that RBM20 is important in regulating muscle gene expression, and its deficiency leads to increased gene expression in day 49 when compared to days one and 20, and, ultimately, heart failure.

Heart failure is a serious condition, and usually there is no cure. These patients normally have a weakened heart that cannot supply the body with enough blood, leading to fatigue and shortness of breath. In late stages of the disease, heart failure often leads to death. Our results provide new insights into the role of RBM20 in the progression of heart failure and novel therapeutic targets for molecular therapy; however, further study is needed to

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address the detailed mechanisms of how heart failure can be reversed by targeting RBM20.

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