Hypertrophic cardiomyopathy (HCM) is a common genetic heart condition affecting approximately 1 in 500 individuals, where the heart muscle becomes thick and blood flow is restricted. The condition is characterized by a thickening of the ventricular wall as a result of enlarged cardiac myocytes, changes in blood pressure due to restricted blood flow, and arrhythmias. The most prevalent form of familial HCM arises from a missense mutation in the gene encoding the beta-myosin heavy chain protein, resulting in a change of amino acid 403, from Arg-Gln to His (MYH7-R403Q). The study of diseases affecting cardiomyocytes has been advanced by the advent of stem cell technology which has enabled the production of stem cell-derived cardiomyocytes in sufficient quantities to facilitate large scale in vitro research. Further advances in stem cell technology have enabled the production of induced pluripotent stem (iPS) cells from any individual, regardless of health status, to address the needs of diseases affecting cardiomyocytes. Research continues to focus on the role of cellular hypertrophy, including the up-regulation of fetal genes, cytoskeletal rearrangements, and an increase in cardiomyocyte size. We show that induced and cardiac hypertrophy in iPS cell-derived CM and show autonomous contractile activity similar to the control iPS cell-derived CM. MYH7-R403Q CM and cardiomyocytes (CM) can be produced from any iPS cell in a collection and used to gain a better understanding of mechanisms involved in complex heart diseases. Here we describe the iPS cell-derived CM from normal and MYH7-R403Q carriers.

Hypertrophy can be induced in normal human donor iPS cell-derived CM with exposure to Endothelin-1 (ET-1). HCM-induced CM exhibit classic hallmarks of cardiac hypertrophy including up-regulation of fetal genes, cytoskeletal rearrangements, and an increase in cardiomyocyte size. We show that induced and inherited HCM in iPS cell-derived CM have common features. Exposure to ET-1 from MYH7-R403Q iPS cells exhibit cardiac morphology, and showed autonomous contractile activity similar to the control iPS cell-derived CM. MYH7-R403Q CM and ET-1 induced HCM in normal CM have similar basal gene expression. ET-1 induction increases BNP expression in both control and MYH7-R403Q cardiomyocytes, but basal BNP levels are higher in MYH7-R403Q cardiomyocytes. These data show the progression of HCM characteristics in MYH7-R403Q cardiomyocytes and underscore the advantages of modeling cardiovascular disease with iPS cell technology.

Somatic cells from human adult tissue are obtained via non-invasive methods and reprogrammed to generate iPS Cells using non-integrating vectors expressing a variety of factors. The resulting iPS cells are cultured and expanded in defined media in feeder-free conditions. These cells can be grown indefinitely or cryopreserved. Truly pluripotent iPS cells can be differentiated into terminal cell types representing those in the human body derived from all three germ layers, mesoderm, endoderm, and ectoderm. Our reprogramming technology ensures the iPS cells we produce are truly pluripotent, enabling us to produce differentiated cells for diverse donor samples.

Large scale manufacturing capabilities, allows us to produce reproducible terminal cell lines from any iPS cell line in quantities sufficient for screening experiments. Consistent production makes these terminally differentiated cells an ideal model system for studying biology in vitro. In addition, we have devised cryopreservation methods that enable researchers to store terminally differentiated materials, and enhanced workflows taking cells from the freeze to assay in less than five days. Taken together, these technologies are revolutionizing our ability to study mechanisms of induced, infectious, and inherited human diseases in a dish and to develop treatments for these diseases.

One powerful feature of iPS cell technology is the ability to generate patient-derived stem cell lines from individuals affected by inherited disorders. This poster describes the production and characterization of cardiomyocytes from a patient with hypertrophic cardiomyopathy.

### Abstract

Hypertrophy can be induced in normal human donor iPS cell-derived CM with exposure to Endothelin-1 (ET-1). HCM-induced CM exhibit classic hallmarks of cardiac hypertrophy including up-regulation of fetal genes, cytoskeletal rearrangements, and an increase in cardiomyocyte size. We show that induced and inherited HCM in iPS cell-derived CM have common features. Exposure to ET-1 from MYH7-R403Q iPS cells exhibit cardiac morphology, and showed autonomous contractile activity similar to the control iPS cell-derived CM. MYH7-R403Q CM and ET-1 induced HCM in normal CM have similar basal gene expression. ET-1 induction increases BNP expression in both control and MYH7-R403Q cardiomyocytes, but basal BNP levels are higher in MYH7-R403Q cardiomyocytes. These data show the progression of HCM characteristics in MYH7-R403Q cardiomyocytes and underscore the advantages of modeling cardiovascular disease with iPS cell technology.

### Power of iPS Technology

Somatic cells from human adult tissue are obtained via non-invasive methods and reprogrammed to generate iPS Cells using non-integrating vectors expressing a variety of factors. The resulting iPS cells are cultured and expanded in defined media in feeder-free conditions. These cells can be grown indefinitely or cryopreserved. Truly pluripotent iPS cells can be differentiated into terminal cell types representing those in the human body derived from all three germ layers, mesoderm, endoderm, and ectoderm. Our reprogramming technology ensures the iPS cells we produce are truly pluripotent, enabling us to produce differentiated cells for diverse donor samples.

Large scale manufacturing capabilities, allows us to produce reproducible terminal cell lines from any iPS cell line in quantities sufficient for screening experiments. Consistent production makes these terminally differentiated cells an ideal model system for studying biology in vitro. In addition, we have devised cryopreservation methods that enable researchers to store terminally differentiated materials, and enhanced workflows taking cells from the freeze to assay in less than five days. Taken together, these technologies are revolutionizing our ability to study mechanisms of induced, infectious, and inherited human diseases in a dish and to develop treatments for these diseases.

One powerful feature of iPS cell technology is the ability to generate patient-derived stem cell lines from individuals affected by inherited disorders. This poster describes the production and characterization of cardiomyocytes from a patient with hypertrophic cardiomyopathy.

### Human iPS Cell-derived Cardiomyocytes Carrying MYH7-R403Q Exhibit Aspects of Hypertrophic Cardiomyopathy In Vitro

Eugenia Jones, Natsuyo Aoyama, Jun Wang, Michael McLachlan, Randy Learich, Tom Burke, Coby Carlson, and Blake Anson

### Functional Data

- **Baseline differences:** Contrast CM (left) and MYH7-R403Q (right) in the amount of BNP expressed in the unstimulated state (red = BNP, blue = DAPI), suggesting innate signs of a cardiac hypertrophy phenotype in the mutant CM. Additionally, the number of BNP-positive cells can be rescued by small molecules such as verapamil.

### Future Directions

Induced pluripotent stem cell technology enables disease research with the human cell type of interest. Using genetic engineering, isogenic backgrounds facilitate more robust studies of phenotypic relationships. This poster demonstrates the use of genetically engineered iPSC-derived cardiomyocytes for the study of hypertrophy.

Our next step is to differentiate genetically engineered iPSC cells, harboring arrhythmogenic alleles in ion channels to determine if the phenotype is recapitulated in cardiomyocytes in vitro. In addition, we are producing cardiomyocytes from iPSC cells derived from patients with polygenic cardiac hypertrophy, who are part of the Hypergen study cohort. These cells will be available to researchers as cardiomyocytes, later this year as part of the MyCell Disease and Diversity panel. The iPSC cell lines will be available in an NHLBI bank.