PKP2 Gene Therapy Reduces Ventricular Arrhythmias, Reverses Ventricular Remodeling, Improves Heart Function, and Reduces Mortality in a Mouse Model of Arrhythmogenic Right Ventricular Cardiomyopathy

Iris Wu[1], Aliya Zeng[1], Amara Greer-Short[1], J. Alex Aycinena[1], Anley E. Tefera[1], Reva Shenwai[1], Farshad Farshidfar[1], Melissa Van Pell[1], Emma Xu[1], Chris Reid[1], Neshel Rodriguez[1], Beatriz Lim[1], Tae Won Chung[1], Joseph Woods[1], Aquilla Scott[1], Samantha Jones[1], Cristina Dee-Hoskins[1], Carolina G. Gutierrez[1], Jessie Madariaga[1], Kevin Robinson[1], Yolanda Hatter[1], Renee Butler[1], Stephanie Steltzer[1], Jaclyn Ho[1], James R. Priest[1], Xiaomei Song[1], Frank Jing[1], Kristina Green[1], Kathryn N. Ivey[1], Timothy Hoey[1], Jin Yang[1], Zhihong Jane Yang[1]

[1]Tenaya Therapeutics, Inc., 171 Oyster Point Blvd, 500, South San Francisco, CA, USA

Fundamental mechanisms beyond disruption of desmosome function are identified in a mouse model of ARVC. The observed PKP2 dose-function relationship indicates that cardiac-selective AAV9:PKP2 gene therapy may be a promising therapeutic approach to treat ARVC patients with *PKP2* mutations.

ABSTRACT

Introduction

Arrhythmogenic right ventricular cardiomyopathy (ARVC) is an inherited cardiomyopathy associated with ventricular arrhythmias and an increased risk of sudden cardiac death. Currently, there are no approved treatments that address the underlying genetic cause of this disease, representing a significant unmet need.

Purpose

Mutations in desmosome gene *Plakophilin-2 (PKP2)* account for approximately 40% of ARVC cases and result in reduced gene expression. Our goal is to examine the feasibility and the efficacy of restoration of PKP2 expression in a cardiac specific knock-out mouse model of *Pkp2*.

Methods

Adeno-associated virus 9 (AAV9) was used to deliver a wild-type copy of *PKP2* gene that is driven by a cardiac-selective promoter and expressed selectively in cardiomyocytes.

Results

We demonstrated that a single-dose AAV9-mediated *PKP2* gene therapy effectively prevented disease onset before overt cardiomyopathy and attenuated disease progression after overt cardiomyopathy. Restoration of PKP2 expression led to

- Restoring cellular structures of desmosomes and gap junctions
- Preventing or halting decline in left ventricular ejection fraction
- Preventing or reversing dilation of the right ventricle
- Attenuating ventricular arrhythmia event frequency and severity
- Preventing adverse fibrotic remodeling
- Significant extension of lifespan

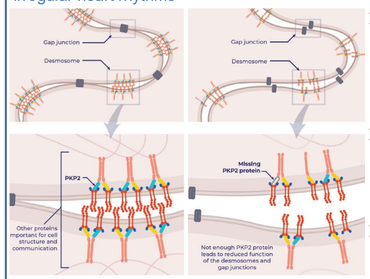
Durable restoration of *PKP2* expression led to highly coordinated and sustained correction of *PKP2*-associated transcriptional networks beyond desmosomes, revealing a broad spectrum of biological perturbances behind ARVC disease etiology.

Conclusion

These results indicate that a cardiac-selective AAV9-mediated *PKP2* gene therapy may be a promising one-time treatment for ARVC patients with *PKP2* mutations.

Background: *PKP2* mutation-mediated ARVC

- Severe and progressive genetic heart disease characterized by adverse remodeling and irregular heart rhythms

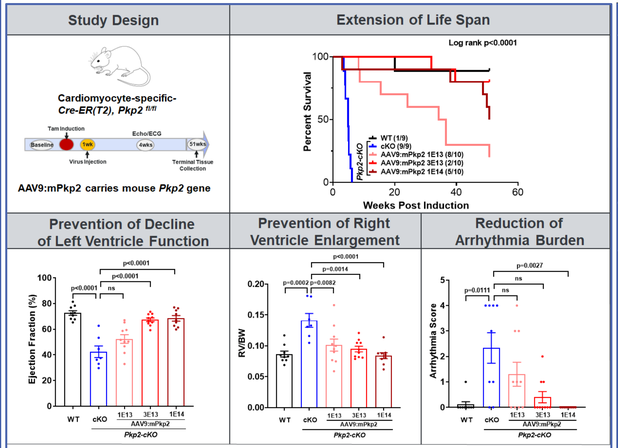


Asimaki et al. (2009) *NEJM*; Basso et al. (2011) *Nat. Rev. Cardiol.*; Corrado et al. (2017) *NEJM*; Oxford et al. (2017) *Cir. Res.*

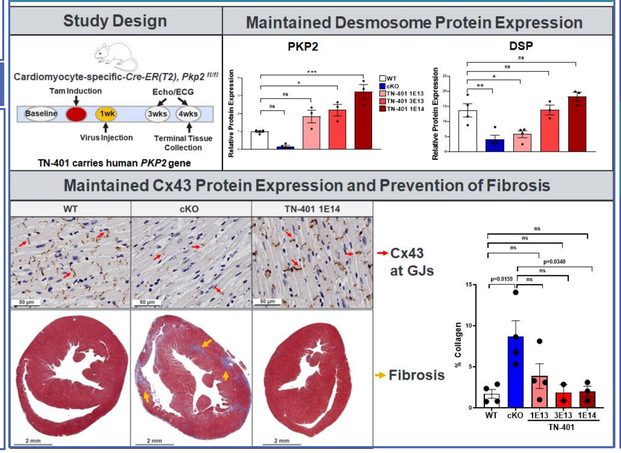
- *PKP2* mutations are the predominant genetic cause of ARVC
  - The mean age of presentation is <40 yo
  - Estimated to affect >70,000 patients in the U.S.
- *PKP2 m*utation is associated with reduction of protein levels and loss of desmosome structure and gap junctions (GJs) in human hearts
- Reduction of Connexin 43 (Cx43), a critical component of GJs, results in compromised electrical coupling and heterogeneous conduction between cardiomyocytes

RESULTS _ Preventive Mode of Treatment

AAV9:PKP2 Gene Therapy Demonstrated Dose-Dependent Disease Modification and Survival Benefit

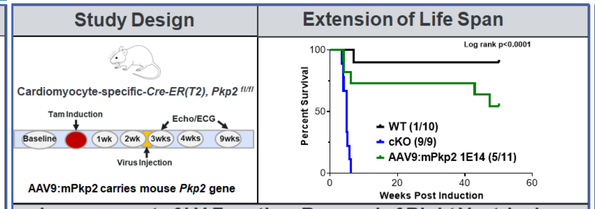


A Single Dose of AAV9:PKP2 Gene Therapy Restored Desmosomes and GJs and Prevented Fibrosis

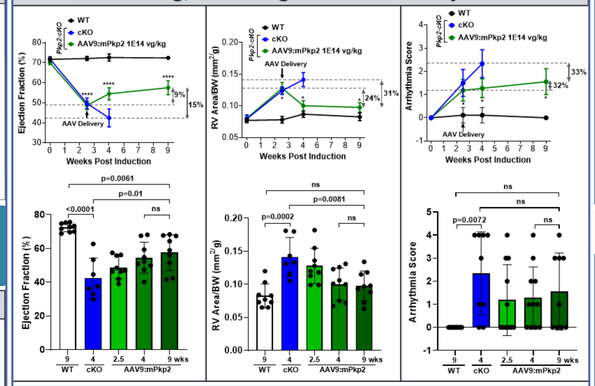


RESULTS _ Therapeutic Mode of Treatment

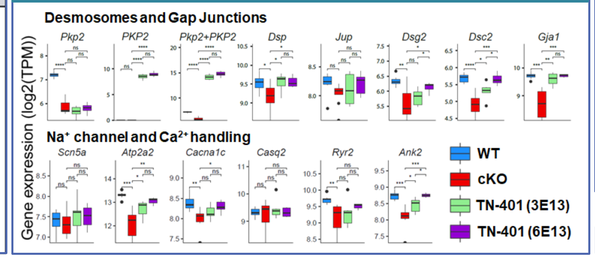
A Single Dose of AAV9:PKP2 Treatment Improved Heart Function and Survival after Onset of Disease



Improvement of LV Function, Reversal of Right Ventricular Remodeling, A Trending Reduction in Arrhythmias

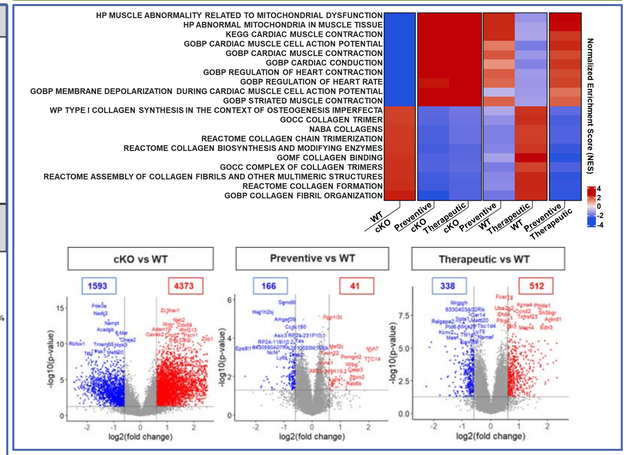


A Single Dose of AAV9:PKP2 Treatment Coordinated Restoration of Genes Encoding Desmosome and Ca^{2+} Handling System



RESULTS _ Therapeutic Mode of Treatment

Near Reversal of Enriched Genes in Mitochondrial Dysfunction, Cardiac Conduction and Fibrosis



CONCLUSIONS

Cardiac-selective AAV9:PKP2 gene therapy using a mouse model of ARVC:

- Largely prevented disease phenotypes before the onset of cardiomyopathy
- Reversed or halted disease progression after the onset of disease
- Led to a highly coordinated and durable correction of structural genes encoding desmosome, sarcomere, Ca^{2+}-handling system, and correction of multiple signaling pathways of metabolism, inflammation, and apoptosis.

We propose that cardiac-selective AAV9:PKP2 could be a beneficial gene therapy approach to reduce ventricular arrhythmias, slow down adverse right ventricular remodeling, improve heart function, and reduce mortality in ARVC patients with *PKP2* mutations. Correspondingly, the U.S. Food and Drug Administration (FDA) has provided clearance of Investigational New Drug (IND) application to initiate clinical testing of TN-401, Tenaya Therapeutics' AAV9:human PKP2 clinical drug candidate.

Detailed study information published on DOI : 10.1038/s43856-024-00450-w

ACKNOWLEDGEMENTS

We thank Dr. Mario Delmar and NYU for licensing *Pkp2-cKO* mouse.