IJHSR
ijhighschoolresearch.org
REVIEW ARTICLE

Evaluating the Therapeutic Potential of Gene Therapy for RyR2 and CASQ2 Gene Mutations in Catecholaminergic Polymorphic Ventricular Tachycardia

Saathvik S. Koneru

Detroit Catholic Central High School, 27225 Wixom Rd, Novi, MI 48374, USA; koneru.saathvik@gmail.com

ABSTRACT: Catecholaminergic Polymorphic Ventricular Tachycardia (CPVT) is a rare inherited cardiac arrhythmia characterized by adrenergically induced ventricular tachycardia and sudden cardiac death in structurally normal hearts. The current medical therapies, like non-cardioselective beta blockers, flecainide, calcium channel blockers, dantrolene, implantable cardioverter-defibrillators (ICDs), and left cardiac sympathetic denervation (LCSD), reduce risk but do not reliably prevent life-threatening arrhythmic events. Breakthrough arrhythmias, device complications, and variable response to the medical therapies show the limitations of approaches that do not address the underlying molecular defect. This literature review combines preclinical and clinical literature on gene therapy strategies that directly target genetic defects that characterize CPVT. Experimental work in mouse and cell models demonstrates effective gene transfer, allele-specific silencing, CRISPR/Cas9-mediated editing, and CaMKII inhibition. These gene therapies normalize sarcoplasmic reticulum calcium handling and reduce ventricular arrhythmia burden. There have been some major breakthroughs, including SGT-501, a gene therapy designed to deliver a CASQ2 gene replacement to cardiomyocytes. These results support gene therapy as a mainline treatment rather than having a supporting role. These advancements represent a promising path towards a permanent gene therapy for CPVT, but careful assessment of the efficacy of treatments remains essential before widespread adoption.

KEYWORDS: Translational Medical Sciences, Disease Treatment and Therapies, CPVT, Gene Therapy.

Introduction

Catecholaminergic Polymorphic Ventricular Tachycardia (CPVT) is a genetic disease characterized by cardiac arrhythmias that can lead to dizziness, fainting, light-headedness, and even sudden cardiac death.[1] These symptoms first appear between the ages of 2 and 21. It has an estimated incidence of 1 in 10,000 and is triggered by both physical exertion and strong emotion.[2] If untreated, CPVT has a mortality rate of up to 50% by age 40.[3]

CPVT is classified as a channelopathy, indicating a gene mutation that causes a malfunction of the ion channels in the body.[4] CPVT's gene mutations have been classified into 5 subtypes, among which ryanodine receptor-2 (*RyR2*) and cardiac calsequestrin 2 (*CASQ2*) make up CPVT1 and CPVT2, respectively, and have the highest incidence of CPVT.[5] According to Figure 1, *RyR2* mutations are found in 70% of CPVT cases, and *CASQ2* mutations are found in 5% of CPVT cases.[6] The *RyR2* gene is autosomal dominant, and the *CASQ2* gene is autosomal recessive.[7] As shown in Figure 1, other CPVT gene variants like *CALM1, CALM2, CALM3, TRDN,* and *TECRL* appear, but their rarity makes them insignificant.[8]

The current treatments for CPVT are beta blockers, flecainide, calcium channel blockers, dantrolene, Implantable Cardioverter Defibrillator (ICDs), and Left Cardiac Sympathetic Denervation (LCSD).[9] There are some limitations in these treatments, so a permanent solution is required. Limitations are shown in the beta-blocker study by Mazzanti et al,[3] and the atropine study by Kannankeril *et al.*[10] Gene therapy is a medical approach that treats or prevents a disease by modifying a patient's genes to correct genetic defects.[11]

This review discusses the many different types of gene therapy, including gene transfer, allele silencing, gene editing, and modulation of signalling pathways using CRISPR-Cas9 technology.[12] The study of gene therapy is very new, so this paper will explore the challenges with gene therapy, innovative solutions to mitigate these challenges, and its possible uses in the future.[8] CPVT needs a permanent solution capable of addressing its genetic basis rather than solely mitigating symptoms.

Methodology

This paper is a thorough literature review of the most important studies in the field of CPVT. Since the disease is rare, the review analyses articles from 2002 to 2025. This timeframe allows for a comparison between previous medical therapy research and newer gene therapy approaches. Resources such as PubMed, SciSpace, and Google Scholar were necessary to identify the most relevant research for this review. Journals focused on cardiac research, such as AHA Journals and Frontiers in Cardiovascular Medicine, were used to obtain more information regarding CPVT. Search queues for the review included: CPVT, gene therapy, *RyR2, CASQ2*, CaMKII, ventricular tachycardia, and medical therapy. This paper will explore clinical trials that study the effects of medications for CPVT. Preclinical studies were also considered to evaluate previous research. This is a review of the different types of gene therapy and how they can treat CPVT. Any studies

DOI: 10.36838/IJHSR815.68

that included gene therapy targeting CaMKII proteins, *RyR2*, and *CASQ2* gene mutations were used. However, studies that targeted other genes or were not written in English were not included. This study is only a literature review; no original experimental research was conducted.

Pathogenesis:

A combination of gene mutations and adrenergic stimulation causes CPVT. There are gene mutations in *RyR2* and *CASQ2*, and adrenergic stimulation is caused by the activation of CaMKII. The mutations cause abnormal calcium handling in the cardiomyocytes, muscle cells of the heart. The cardiomyocytes leak excess calcium from the sarcoplasmic reticulum, accelerating heartbeat and precipitating ventricular arrhythmias. The adrenergic stimulation releases adrenaline and noradrenaline into the heart, making it beat faster and worsening the ventricular arrhythmias. As shown in Figure 1, these two factors lead to delayed afterdepolarizations (DADs) and other triggered activity, creating electrical instability that manifests as ventricular arrhythmias, which develop into CPVT.[13]

Detection Methods:

CPVT is a unique disease as patients don't have any structural abnormalities in the heart. Because they have no structural abnormalities, CPVT patients have a normal resting electrocardiogram (ECG).[14] As shown in Figure 1, resting ECGs and echocardiograms appear normal, making CPVT diagnosis extremely difficult as it relies heavily on exercise stress testing and genetic testing.[15] Genetic testing has become a critical tool, as it confirms mutations in the *RyR2* or *CASQ2* genes, allowing doctors to treat patients and screen other family members for any potential problems.[16] CPVT has been passed down through the family in about 30% of cases, showing the importance of genetic testing for early intervention if needed.[16]

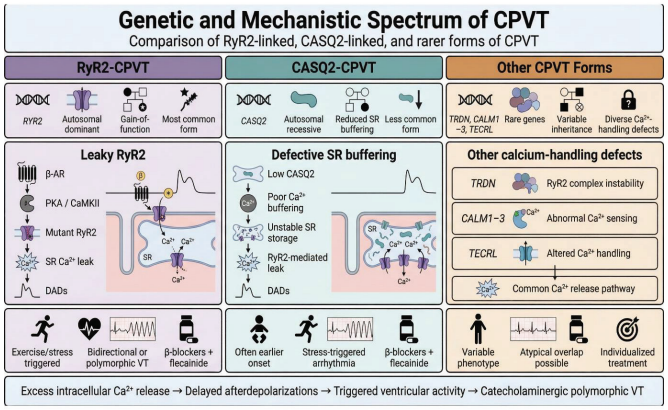

Figure 1: This figure compares the major genetic phenotypes of CPVT. RyR2-CPVT is expressed as an autosomal dominant condition caused by gain-of-function mutations that promote SR calcium leaks and DADs. CASQ2-CPVT is presented as an autosomal recessive form where impaired calcium buffering destabilizes SR calcium storage. Rarer forms involve genes such as TRDN, CALM1-3, and TECRL. Together, all pathways converge from excess intracellular calcium release to triggered ventricular activity and finally stress-induced polymorphic ventricular tachycardia. Made using BioRender.

Current Treatment Strategies :

Current treatments aim to reduce adrenergic stimulation and stabilize cardiac electrical activity. The treatments include beta blockers, flecainide, calcium channel blockers, dantrolene, Implantable Cardioverter Defibrillator (ICDs), and Left Cardiac Sympathetic Denervation (LCSD).[7]

Beta blockers work by inhibiting the effects of catecholamines like epinephrine and norepinephrine.[17] Cardioselective beta blockers target beta-1 receptors, and noncardioselective beta blockers target both beta-1 and beta-2 receptors. CPVT patients typically use noncardioselective beta blockers like nadolol and propranolol.[1] Beta blockers bind the beta receptors that bind with catecholamines and change the receptor. This stops the catecholamines from activating, inducing a slower heart rate, less stress, and a greater reduction in the incidence of abnormal heart rhythms.[2] However, beta blockers are not flawless and fail in 30% of patients.[8] As shown in Table 1, beta blockers can be used in combination with flecainide, CCBs, dantrolene, ICDs, and LCSD.

Flecainide, a sodium channel blocker, is used in conjunction with beta blockers as it reduces ventricular arrhythmias during exercise.[18] As shown in Table 1, flecainide works by directly blocking the *RyR2* receptor, binding to the *RyR2* channel's pores or outer surface to prevent the spontaneous calcium release common in CPVT patients. Flecainide also blocks the sodium channels, which are involved in the electrical signalling within heart cells.[19] Sodium channels let sodium ions in and out of the cell, triggering depolarization and repolarization, the mechanisms the heart uses to pump blood. Flecainide's blockage of the sodium and calcium channels stabilizes the heart's electrical activity and decreases the number of DADs that occur.[10] A side effect of flecainide is that it may trigger abnormal calcium release, precipitating arrhythmias.[20]

Calcium channel blockers work by inhibiting the entry of calcium ions into cardiomyocytes, regulating the heart's electrical activity, and lowering the likelihood of harmful arrhythmias.[21] Studies show that calcium channel blockers significantly reduce the frequency of fast heartbeats during exercise.[8] Calcium channel blockers specifically target L-type calcium channels, which are important for calcium entry into cardiomyocytes. CPVT patients most commonly use calcium channel blockers like verapamil and diltiazem.[22] However, as shown in Table 1, calcium channel blockers are used very limitedly as they offer less protection than sodium channel blockers and could worsen heart arrhythmias by allowing calcium leaks.[8]

Dantrolene, a skeletal muscle relaxant, reduces muscle spasms and tightness, causing relaxed muscle contraction.[20] As shown in Table 1, dantrolene works by inhibiting the release of calcium ions from the SR, helping prevent the abnormal calcium leaks associated with CPVT-1. It stabilises the *RyR2* calcium release channels to reduce the risk of triggering dangerous arrhythmias.[23] Dantrolene is also shown to reduce the frequency and duration of threatening arrhythmias in CPVT patients. The lack of long-term safety and efficacy data makes dantrolene a rare medication prescribed for CPVT patients.[13]

One of the two non-pharmacological treatments for CPVT involves the Implantable Cardioverter Defibrillator. As the

only machine that is commonly used to treat CPVT, an ICD monitors the heart's electrical activity and delivers electrical impulses to correct abnormal heart rhythms.[3] An ICD can be both a primary and secondary prevention based on the circumstances. ICDs are used when typical therapy with beta blockers and flecainide doesn't work and are recommended for patients who have survived cardiac arrest or have recurrent syncope.[1] However, there are serious side effects for ICDs. Even when ICDs emit appropriate shocks, there is a possibility that the shock can trigger catecholamines and worsen arrhythmias.[3] As shown in Table 1, another side effect is frequent electrical storms, which are inappropriate ICD shocks that can increase stress and catecholamine release. ICDs can be extremely useful in managing CPVT, but patients should be wary of side effects before using them.[1]

Left Cardiac Sympathetic Denervation is the only surgical treatment used for CPVT. It is only used for patients who are unresponsive or intolerant to beta-blocker/flecainide therapy.[24] As shown in Table 1, it is a minimally invasive surgical procedure that involves severing the sympathetic nerves on the left side of the heart. These nerves release stress hormones like epinephrine and norepinephrine, which can cause arrhythmias and abnormal heart rhythms. LCSD helps reduce the incidence of syncope, cardiac arrest, ventricular fibrillation, and sudden cardiac death.[9] LCSD is often used in combination with ICDs, as it provides an extra layer of protection from recurrent shocks in ICD patients. However, LCSD is not a complete cure for CPVT, and there is a possibility of sudden death. Even with its many positive effects and minimal side effects, LCSD is underutilized because of the challenges in predicting its effectiveness in patients.[8]

Table 1: Current treatment strategies for CPVT.

Treatment	Purpose	Results	Combination with other treatments
Beta Blockers	Bind to Beta 1 and 2 receptors that could activate catecholamines	Works as the primary treatment but fails in up to 30% of patients.	Flecainide, CCB, Dantrolene, ICD, LCSD
Flecainide	Blocks *RyR2* Channel Stops the flow of calcium and sodium into cardiomyocytes	Stabilizes electrical activity and lessens DADs, but increases the chance of abnormal calcium release	Beta Blockers
Calcium Channel Blockers (CCB)	Stops the flow of calcium from the SR into cardiomyocytes	Very limited results in reducing arrhythmias, but it may work in some patients	Beta Blockers
Dantrolene	Inhibits the release of calcium from the SR into cardiomyocytes	Very limited results in reducing arrhythmias, but it may work in some patients	Beta Blockers
ICD	Monitors heart rhythm and sends electrical shocks to correct abnormal rhythms.	A very effective primary and secondary treatment but it has a chance of producing harmful electrical storms.	LCSD
LCSD Surgery	Removes nerves that release epinephrine and norepinephrine	Very effective in stopping arrhythmias, but not used commonly because of variable patient response	ICD

Although common in use, these treatments are not 100% reliable and may not offer full protection in patients with CPVT. A better treatment that has been suggested is gene therapy. Gene therapy is a medical approach that treats or prevents a disease by modifying a patient's genes to correct genetic defects. For CPVT, we will discuss the *RyR2* and *CASQ2* genes and the CaMKII protein. These 2 genes and 1 protein are major components in causing and exacerbating CPVT.

Gene Background:

As shown in Figure 2, the *RyR2* gene encodes ryanodine receptor 2, a protein that forms calcium channels in the sarcoplasmic reticulum of cardiomyocytes. It uses calcium-induced calcium release to release calcium from the SR into the cytosol, but mutations in the gene can cause problems with the calcium release.[25] Mutations affect the production of the calstabin-2 protein, an important subunit that binds to the *RyR2* complex.[7] Calstabin-2 helps keep the calcium channel closed, preventing the opening of the channel when the calcium signal is absent. The mutation causes a disconnection between calstabin-2 and *RyR2* because of structural changes in the calstabin-2 protein that change its sensitivity to calcium. This structural change impacts the probability of the channel opening, leading to abnormal calcium release.[26] The abnormal calcium release can cause delayed afterdepolarizations (DADs), abnormal electrical activity following a heartbeat, leading to the polymorphic ventricular tachycardia that characterizes CPVT.[26]

As shown in Figure 2, the *CASQ2* gene makes calsequestrin, a calcium-binding protein. It acts as a calcium buffer in the SR, holding calcium in the cisterna of the SR after heart muscle contraction.[26] Calsequestrin stores calcium as a molecule of calsequestrin that can bind up to 50 Ca^{2+} ions.[25] *CASQ2* regulates calcium-induced calcium release and releases the bound calcium when the *RyR2* gene opens calcium channels in the SR.[7] However, the mutation in the *CASQ2* gene causes reduced expression or complete absence of *CASQ2* in cardiac tissue.[5] The mutation is the R33Q mutation, which disrupts the linear polymerization ability of *CASQ2*. This disruption directly inhibits *CASQ2*'s ability to bind Ca^{2+}. These mutations in the *CASQ2* gene cause abnormal calcium release, such as premature or spontaneous calcium release, which can cause DADs, leading to the polymorphic ventricular tachycardia that characterizes CPVT.[7]

However, CPVT needs both a pathogenic mutation and an environmental trigger in the form of adrenergic stimulation to be activated.[7] The pathogenic mutation in genes such as *RyR2* and *CASQ2* is inherited, while the adrenergic stimulation comes from catecholamines like epinephrine and norepinephrine, which are released upon stressful conditions.[26]

The adrenergic stimulation for CPVT is triggered by beta-adrenergic stimulation, the release of adrenaline during stress or exercise. The beta-adrenergic stimulation activates beta-adrenergic receptors that increase the heart rate and cause stronger muscle contractions by altering the activity of calcium ion channels.[7] To alter the activity of calcium ion channels, the beta-adrenergic receptors activate a protein kinase called CaMKII.[26]

CaMKII is a calmodulin-dependent protein kinase II, which is a serine/threonine-specific protein kinase that regulates the calcium complex. It is part of the fight-or-flight response, so it releases calcium to help the heartbeat faster and is activated by the binding of calcium and calmodulin. This binding triggers a conformational change, enabling it to phosphorylate target proteins involved in Ca^{2+} handling.[26] CaMKII phosphorylates *RyR2* in the SR, enabling calcium release from storage. However, CaMKII phosphorylation of a mutated *RyR2* gene can

DOI: 10.36838/IJHSR815.68

cause increased calcium leakage, which causes DADs and can ultimately lead to CPVT.[7] Stressful conditions can further activate CaMKII, which can cause even more calcium leakage and CPVT.[26]

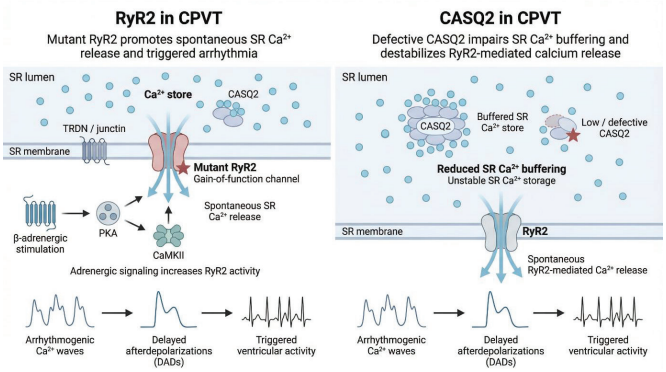

Figure 2: This figure illustrates the different mechanisms of RyR2 and CASQ2 mutations. Mutant RyR2 channels increase spontaneous SR calcium release during beta-adrenergic stimulation through PKA and CaMKII signaling. The excess signaling leads to arrhythmogenic calcium waves, DADs, and triggered activity. Defective CASQ2 weakens SR calcium buffering and destabilizes calcium storage, promoting abnormal RyR2-mediated calcium release. The figure emphasizes the shared downstream electrophysiologic consequence of disrupted calcium handling in CPVT. Made using Biorender.

Preclinical Studies:

A study by Bezzerides, Parker, and Pu tried to inhibit CPVT by blocking the CaMKII protein kinase in a CPVT tissue model. They tested this inhibition in tissue models made from blood samples of 2 CPVT-1 patients. These models reprogrammed the blood cells into induced pluripotent stem cells to make cardiomyocytes that carried the *RyR2* CPVT mutation. The scientists then stimulated the cells in an exercise stress test to initiate faster heartbeats to help reveal CPVT's underlying mechanisms. During the exercise stress test for the healthy heart tissue, the calcium waves moved through the tissue evenly. But for patients with CPVT, the calcium waves moved at varying speeds or didn't move at all. This abnormal calcium movement causes the dangerous arrhythmias characterized by CPVT. When the cells were stimulated at a faster speed, the healthy tissue handled it fine, but the CPVT tissue sustained recurrent arrhythmias. The scientists found that the CaMKII protein kinase modifies *RyR2* to trigger the cardiomyocyte to release more calcium, producing excessive calcium release and causing arrhythmias. When the scientists blocked CaMKII, the arrhythmias in the CPVT tissue model were eliminated.[11] The scientists also tested a gene therapy approach to inhibit CaMKII in a mouse model of CPVT. As shown in Table 2, the scientists engineered a special virus that selectively traveled into the heart and delivered AIP, a potent and selective inhibitor, into the mouse. Testing found AIP in 50% of heart cells, which was enough to block the effects of CaMKII. The scientist planned to refine the gene therapy strategies to test in larger animal models and eventually humans.[27]

As shown in Table 2, a study by Priori and Mazzanti *et al.* applied 4 gene therapy strategies to CPVT models: gene transfer, allele silencing, gene editing, and modulation of signalling pathways. Gene transfer is the process of introducing genetic material into a cell. As shown in Figure 3, gene transfer was tested in CPVT-2, where the delivery of a functional wild-type *CASQ2* gene replaced the missing or nonfunctioning gene to restore normal calcium-induced calcium release. A study injected mice with a deficient *CASQ2* with an AAV9-*CASQ2* treatment to develop a gene therapy strategy. The AAV9-*CASQ2* treatment is a functional *CASQ2* gene packaged into a recombinant adeno-associated viral (AAV) vector, which allows the treatment to go directly into the nucleus of the cell. Although only 30% of cardiomyocytes provided antiarrhythmic protection, within 1 year, the mice showed normal *CASQ2* levels, suppression of triggered activity arrhythmias, and absence of catecholamine-induced ventricular arrhythmia.[28]

As shown in Figure 3, allele silencing uses RNA interference to reduce the expression of a mutant allele by exerting a dominant negative effect. The scientists designed an allele silencing molecule (siRNA) to rescue the wild-type *RyR2* in *RyR2*-mutated CPVT. This approach identifies a siRNA that can selectively reduce the expression of the mutant allele without reducing the amount of wild-type protein. In its administration to both newborn and adult mice, the siRNA was able to reduce the mutant *RyR2* channels and increase wild-type *RyR2*, reducing triggered activity arrhythmias and suppressing catecholamine-induced ventricular tachycardia.[29] The study demonstrates that even a partial reduction of mutant *RyR2* protein reduces the chance of arrhythmias. The challenge with allele silencing is the lack of targeted therapy for each variant of CPVT-1, but the emergence of nonviral delivery systems could make this approach more feasible.

As shown in Figure 3, gene editing is a precise method of making targeted changes to an organism's DNA. CRISPR-Cas9 gene editing uses nonspecific DNA-cleavage proteins tethered to a guide RNA to target DNA double-strand breaks to correct the mutation. One pathway knocks out the mutant gene through the occurrence of insertions and deletions that lead to premature stop codons. Another one fixes the double-strand DNA breaks by using the cell's natural DNA-repairing pathways. Despite being a relatively new treatment, CRISPR-Cas9 genome editing disrupted the disease-causing allele in the cardiomyocytes of CPVT-1 mice without any detectable off-target mutagenesis, making this a viable opportunity for further development.[30]

As shown in Figure 3, modulation of cellular pathways is the process of altering or regulating the activity of intracellular signalling pathways. Modulation of cellular pathways for CPVT mediates the effects of catecholamines in cardiomyocytes or the proteins that interact with *RyR2*. This technique's breakthrough was the inhibition of CaMKII, which normalized calcium handling and suppressed arrhythmias in a CPVT-1 mouse model.[31] This technique shows the effectiveness of alternating paths to help a patient stop dangerous arrhythmias that can cause CPVT. All of these techniques are new and will need refinement before testing can happen in human patients.

Denegri *et al.* studied cardiac gene transfer to treat a mouse model with CPVT. The scientists used CPVT-2 mice, which are characterized by the deficiency or absence of the *CASQ2* protein. The CPVT-2 mice displayed evidence of DADs and arrhythmias in the presence of beta-adrenergic receptor stim-

ulation, showing the CPVT phenotype. Gene transfer with AAV9-CASQ2 at birth prevented the development of structural changes in the arrhythmogenic phenotype in the mice over a period of 12 months. As shown in Table 2, these effects were found even though only 40% of the cardiomyocytes were infected by AAV9 gene delivery, showing the bystander effects of gene transfer.[28] The scientists found structural improvements within the cell and reductions of DAD frequency in the presence of beta-adrenergic receptor stimulation after gene transfer of *CASQ2*. The scientists said that future studies should examine whether *CASQ2* gene transfer would stop *RyR2* calcium release and whether the amount of SR calcium can determine the probability of DADs. This study opens up the possibility of cardiac gene therapy as a therapeutic option for CPVT, but many challenges, such as efficacy, cost, and usability, remain before cardiac gene therapy can be effectively studied in humans.[28]

A study by Bezzerides *et al.* designed AAV9-GFP-AIP to introduce a CaMKII inhibitory peptide in CPVT mouse models. The AAV9 is the delivery vehicle, AIP is the CaMKII inhibitory peptide that would normalize calcium handling in cardiomyocytes, and GFP is a fluorescent protein that allows easy visualisation. The AAV9-GFP-AIP was injected into baby mice models, and the mice models underwent invasive electrophysiology testing between 2 and 4 months of age to view arrhythmic burden, defined as 3 or more premature ventricular contractions followed by tachycardia. As shown in Table 2, the AAV9-GFP-AIP was expressed in the heart with no significant expression in noncardiac tissue and effectively inhibited CaMKII, because both CaMKII indicators threonine 286 and threonine 17 were suppressed in the mouse models. The AIP inhibitory peptide greatly reduced ventricular arrhythmias induced by beta-adrenergic stimulation. In models without AAV9-GFP-AIP gene therapy, 9/18 mice models demonstrated a significant arrhythmic burden. However, in models with AAV9-GFP-AIP gene therapy, only 1/18 mice models showed a significant arrhythmic burden. This shows the ability of AAV9-GFP-AIP to reduce CPVT arrhythmia vulnerability. Though AAV gene therapy worked effectively in mouse models, additional research and refinement are necessary to use the same type of gene therapy in human patients.[30]

Table 2: Preclinical Studies.

Authors	Treatment	Model Type	Genes/ Proteins Targeted	Importance
Bezzerides[27] Parker[27] Pu[27]	AIP Inhibitor	Tissue Model Mouse Models	CaMKII	Proved that AIP is a potent inhibitor of CaMKII and that CaMKII modifies *RyR2* to cause CPVT.
Priori[3] Mazzanti[3]	Gene Transfer Allele Silencing Gene editing Modulation of signalling pathways	Mouse Models	*RyR2* *CASQ2*	Shows different gene therapy techniques and their effectiveness in mouse models, and how different vehicles are used to deliver the gene therapy
Denegri[28]	Cardiac Gene Transfer	Mouse Models	*CASQ2*	*CASQ2*-based gene therapy blocks the mutation, though only 40% of cardiomyocytes were affected, showcasing the bystander effect.
Bezzerides[27] Parker[27] Pu[27]	AAV9-GFP-AIP	Mouse Models	CaMKII	AIP inhibits CaMKII, reduces arrhythmic burden, and doesn't affect non-cardiac cells.

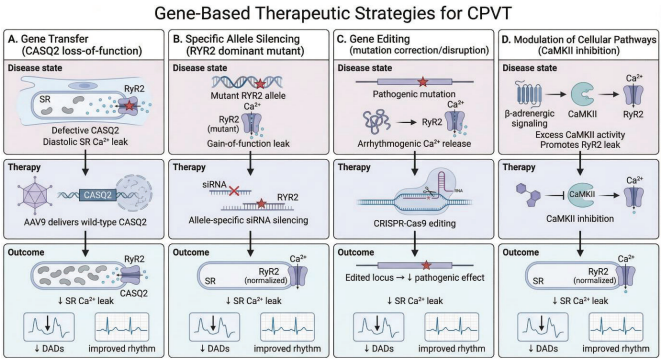

Figure 3: This figure summarizes four gene-based therapeutic approaches for CPVT. Gene transfer is presented for CASQ2 loss-of-function disease, using AAV9 to deliver wild-type CASQ2 and restore calcium buffering. Specific allele silencing targets dominant mutant RYR2 alleles with siRNA to reduce gain-of-function calcium leak. Gene editing uses CRISPR-Cas9 to correct pathogenic variants to reduce arrhythmogenic calcium release. Modulation of cellular pathways focuses on inhibiting CaMKII. Across all four strategies, the shared therapeutic goal is to reduce SR calcium leak, suppress DADs, and improve arrhythmic stability. Made using Biorender.

Clinical Studies:

Beta blockers block the effects of adrenaline and noradrenaline in the body. A study by Mazzanti *et al.* studied the difference of using only beta blockers versus using both beta blockers and an ICD. The study treated patients with only beta blockers. Patients who had other medications or surgery for CPVT were excluded from the study. Differences between selective and non-cardioselective beta blockers were also considered. The study explored a possible rationale for breakthrough events. A breakthrough event in this study was defined as a life-threatening arrhythmic event (LTAE), which included sudden cardiac death, aborted cardiac arrest, and hemodynamically nontolerated ventricular tachycardia. The study included 216 patients with CPVT-1. With only beta-blocker therapy, 28/216 (13%) of the patients experienced an LTAE. However, if a patient experienced an LTAE or syncope before diagnosis of CPVT, they were at an increased risk of an LTAE during beta-blocker therapy. The risk of an LTAE was increased sixfold when selective beta blockers were used compared to nonselective beta blockers. ICDs were implanted in 79/216 (37%) of patients and were shown to provide extra protection against LTAEs. As shown in Table 3, at the occurrence of LTAE, patients with ICDs survived with a 100% (18/18) survival rate versus patients without an ICD, who had a 60% (6/10) survival rate, demonstrating that non-cardioselective beta blockers are more effective than selective beta blockers and that ICDs offer additional protection for CPVT patients.

Flecainide is a sodium channel blocker that blocks electrical signals in the heart to stabilize and normalize a person's heart rhythm. A study by Berge *et al.* investigated whether flecainide combined with beta blockers reduced the incidence of arrhythmogenic events (AEs) in CPVT patients. An AE was defined as sudden cardiac death, sudden cardiac arrest, appropriate ICD shock, and arrhythmic syncope. The study included 247 patients, all of whom were on beta blockers; 28% had an ICD, and 9% had LCSD surgery. There were two study periods. The first, the pre-flecainide period, was the period after diagnosis

DOI: 10.36838/IJHSR815.68

during which beta blockers were prescribed until the start of flecainide. The second, the on-flecainide period, was the period after diagnosis in which both beta blockers and flecainide were used. During the pre-flecainide stage, 17% of patients experienced 58 AEs. During the on-flecainide stage, 9% of patients experienced 38 AEs. This demonstrates a significant decrease in AEs after initiating flecainide therapy. Among 167 patients who were symptomatic before CPVT diagnosis or during the pre-flecainide period, flecainide therapy was associated with significantly fewer AEs. In 41 patients who had more than one AE while on beta-blocker therapy alone, flecainide therapy resulted in significantly fewer AEs. As shown in Table 3, flecainide therapy in combination with beta blocker therapy significantly lowers the incidence of AEs and is particularly effective for symptomatic patients and those who experienced breakthrough events while on beta blocker therapy.

Atropine is an anticholinergic and antimuscarinic agent that blocks the signals of the parasympathetic nervous system. A study by Kannankeril *et al.* used atropine to prevent exercise-induced ventricular arrhythmias in CPVT patients. Preclinical evidence showed that elevating supraventricular rates with pacing or atropine protects against ventricular arrhythmias in CPVT mouse models. Kannankeril *et al.* tested whether this strategy could translate to human patients. 6 adults with CPVT underwent treadmill exercise testing with and without atropine. As shown in Table 3, atropine significantly increased resting sinus heart rate and reduced ventricular ectopy during exercise. Extra heartbeats were eliminated in four out of six patients, while exercise capacity and peak heart rates remained unchanged. There were also no adverse events reported, but the small sample size could skew these results. These findings suggest that raising sinus rates can become a novel therapeutic strategy in CPVT, especially for patients with recurrent arrhythmias despite standard therapy like beta blockers. However, the study's small sample size affects generalizability to the larger population. Larger trials are needed to confirm long-term safety and determine whether atropine can be integrated into standard CPVT treatment.

SGT-501 is an AAV-based gene therapy that treats CPVT. As shown in Table 3, SGT-501 will deliver a functional, full-length, codon-optimized copy of the *CASQ2* gene to cardiac muscle cells. This will increase *CASQ2* protein levels, which enhances buffering of free calcium in the SR. This buffering results in reduced calcium leak into the heart. Even though it increases *CASQ2* protein levels, SGT-501 also stabilizes *RyR2* mutated CPVT. SGT-501 should allow a normal cardiac rhythm with the potential to protect against ventricular arrhythmias. The FDA has approved this gene therapy as a first-in-class therapy to correct the underlying *RyR2* instability and calcium dysregulation causes of CPVT. This is the first gene therapy made for CPVT, making a groundbreaking impact in the treatment of CPVT. Patient testing will start in the 4th quarter of 2025 and will test the safety and efficiency of the gene therapy.

Table 3: Clinical Trials.

Clinical Trials ID	Genes being targeted	Gene targeting strategy	Other treatments	Results/Outcome
Mazzanti[3]	*RyR2*	Beta Blocker Therapy (BBT)	ICD Treatment	Non-selective beta blockers are more effective than selective beta blockers. When used with beta blockers, ICDs provide more protection against CPVT.
Berge[32]	*RyR2* *CASQ2*	Flecainide Therapy (FT)	Beta Blocker Therapy (BBT)	FT with BBT reduced ventricular ectopy but didn't reduce adverse events compared to BBT with placebo.
Kannankeril[10]	N/A	Atropine	N/A	Atropine elevated sinus rates to reduce or eliminate exercise-induced ventricular ectopy in patients with CPVT.
SGT-501[33]	*CASQ2*	SGT-501	N/A	Ongoing

Discussion

Current literature portrays CPVT as a genetic channelopathy with mutations in *RyR2* and *CASQ2* that disrupt calcium homeostasis, trigger DADs, and produce LTAEs.[3] A consistent theme is the difficulty of diagnosing CPVT as patients have structurally sound hearts, and normal resting electrocardiograms and echocardiograms.[14] Exercise stress testing and genetic testing remain the most reliable tools that detect arrhythmic responses to adrenergic stimulation, but reliance on specialized testing emphasizes simpler screening methods.[3]

Current therapies focus primarily on blocking the adrenergic stimulation and stabilizing cardiac electrical activity. Non-cardioselective beta blockers are the first-line treatment and reduce LTAEs, but adverse events in 30% of patients show the need for other therapies.[10] Flecainide affects both sodium channels and *RyR2*, making it an ideal complement to beta blockers. Clinical studies show that flecainide therapy with beta-blocker therapy reduces arrhythmic events, but long-term outcomes are still unknown.[17] It is very effective but requires careful patient selection because of proarrhythmic potential. Calcium channel blockers and dantrolene both target calcium channels, but their uncertain efficacy and lack of long-term safety data make their use very limited.[8] ICDs are used as primary and secondary prevention for CPVT, but inappropriate shocks, electrical storms, and psychological distress complicate their use.[8] LCSD is the only proven surgical method and diminishes adrenergic stimulation by cutting off the sympathetic nerves, effectively reducing arrhythmic events. Even though the surgery is very effective, LCSD is not used much due to variable patient responses and other side effects.[8] These findings illustrate that while current treatments function in some situations, a more definitive treatment is required. The possible treatment is gene therapy.

Gene therapy has been tested out in many different preclinical studies. Preclinical studies have focused on strategies like gene transfer, allele silencing, gene editing, and modulation of signalling pathways, which work in models but have significant challenges in translating them into effective therapy for humans. Obstacles such as efficient delivery, insufficient repair of the cardiomyocytes, and off-target modifications remain. Creating a mass reproducible gene therapy can also be cost-prohibitive. A company is not incentivized to fund gene therapy for CPVT because of its rarity and niche market. However, a company has finally created a gene therapy for

CPVT. Solid Biosciences has created an AAV-*CASQ2*–based gene therapy for CPVT patients. This is the first company to bankroll a major gene therapy treatment for CPVT, showing their willingness to invest money into a novel treatment. This gene therapy treatment and future ones will need to look at long-term safety and efficiency data like immune responses, off-target gene modification, and sustained transgene expression if they want to be successful. This breakthrough may help encourage other companies to invest in and advance research on gene therapy for CPVT. Gene therapy strategies will have to coexist with current treatments, as a combination may produce better outcomes than monotherapy alone. Patient care would be positively impacted by expanded research on the psychological impacts of CPVT.[8] As patients face lifestyle restrictions that lead to anxiety, social isolation, and decreased quality of life, gene therapy studies should also look at quality-of-life improvements and how patients deal with newfound freedom.

Discussion

Current literature portrays CPVT as a genetic channelopathy with mutations in *RyR2* and *CASQ2* that disrupt calcium homeostasis, trigger DADs, and produce LTAEs.[3] A consistent theme is the difficulty of diagnosing CPVT as patients have structurally sound hearts, and normal resting electrocardiograms and echocardiograms.[14] Exercise stress testing and genetic testing remain the most reliable tools that detect arrhythmic responses to adrenergic stimulation, but reliance on specialized testing emphasizes simpler screening methods.[3]

Current therapies focus primarily on blocking the adrenergic stimulation and stabilizing cardiac electrical activity. Non-cardioselective beta blockers are the first-line treatment and reduce LTAEs, but adverse events in 30% of patients show the need for other therapies.[10] Flecainide affects both sodium channels and *RyR2*, making it an ideal complement to beta blockers. Clinical studies show that flecainide therapy with beta-blocker therapy reduces arrhythmic events, but long-term outcomes are still unknown.[17] It is very effective but requires careful patient selection because of proarrhythmic potential. Calcium channel blockers and dantrolene both target calcium channels, but their uncertain efficacy and lack of long-term safety data make their use very limited.[8] ICDs are used as primary and secondary prevention for CPVT, but inappropriate shocks, electrical storms, and psychological distress complicate their use.[8] LCSD is the only proven surgical method and diminishes adrenergic stimulation by cutting off the sympathetic nerves, effectively reducing arrhythmic events. Even though the surgery is very effective, LCSD is not used much due to variable patient responses and other side effects.[8] These findings illustrate that while current treatments function in some situations, a more definitive treatment is required. The possible treatment is gene therapy.

Gene therapy has been tested out in many different preclinical studies. Preclinical studies have focused on strategies like gene transfer, allele silencing, gene editing, and modulation of signalling pathways, which work in models but have significant challenges in translating them into effective therapy for humans. Obstacles such as efficient delivery, insufficient repair of the cardiomyocytes, and off-target modifications remain. Creating a mass reproducible gene therapy can also be cost-prohibitive. A company is not incentivized to fund gene therapy for CPVT because of its rarity and niche market. However, a company has finally created a gene therapy for CPVT. Solid Biosciences has created an AAV-*CASQ2*–based gene therapy for CPVT patients. This is the first company to bankroll a major gene therapy treatment for CPVT, showing their willingness to invest money into a novel treatment. This gene therapy treatment and future ones will need to look at long-term safety and efficiency data like immune responses, off-target gene modification, and sustained transgene expression if they want to be successful. This breakthrough may help encourage other companies to invest in and advance research on gene therapy for CPVT. Gene therapy strategies will have to coexist with current treatments, as a combination may produce better outcomes than monotherapy alone. Patient care would be positively impacted by expanded research on the psychological impacts of CPVT.[8] As patients face lifestyle restrictions that lead to anxiety, social isolation, and decreased quality of life, gene therapy studies should also look at quality-of-life improvements and how patients deal with newfound freedom.

Conclusion

Gene therapy could revolutionize treatment, but current and future studies need to study not only physiological outcomes but also psychosocial and quality-of-life impacts. We need to be able to use preclinical findings to create gene therapy for humans efficiently. Investigations should explore the best combination of current treatments and new gene therapy. Overall, the evidence suggests that targeting RyR2 and CASQ2 gene mutations through gene therapy could offer a promising and more lasting treatment for CPVT by addressing the root genetic cause rather than only managing its symptoms.

Acknowledgments

I owe deep gratitude to my faculty mentors, who guided me through the entire process. Dr. Prahlad Parajuli ensured I understood the foundation of scientific principles used in research. Professor Kay Diaz helped me understand the nuances of how to write and present my facts. Professor Virgel Torremocha taught me how to organize my paper and navigate the difficult submission process. And finally, special thanks to my parents for all their support. They provided me with the energy and willpower to persevere, allowing me to capitalise on this great opportunity.

References

1. Hayashi, M.; Denjoy, I.; Extramiana, F.; Maltret, A.; *et al.* Incidence and Risk Factors of Arrhythmic Events in CPVT. *Circulation* 2009, *119*, 2426–2434. https://doi.org/10.1161/CIRCULATIONAHA.108.829267
2. Napolitano, C.; Priori, S. G.; Bloise, R.; Ronchetti, E. CPVT. *GeneReviews* 2007.

DOI: 10.36838/IJHSR815.68

3. Priori, S. G.; Blomström-Lundqvist, C.; Mazzanti, A.; Blom, N.; *et al.* ESC Guidelines for Ventricular Arrhythmias. *Eur. Heart J.* 2015, *36*, 2793–2867. https://doi.org/10.1093/eurheartj/ehv316.
4. Cerrone, M.; Priori, S. G. Genetics and Mechanisms of Arrhythmias. *Card. Electrophysiol. Clin*. 2019, *11*, 567–576. https://doi.org/10.1016/j.ccep.2019.08.007.
5. Medeiros-Domingo, A.; Bhuiyan, Z. A.; Tester, D. J.; Hofman, N.; *et al.* CASQ2-Associated CPVT. *J. Am. Coll. Cardiol.* 2009, *54*, 206–214. https://doi.org/10.1016/j.jacc.2009.02.070.
6. Priori, S. G.; Napolitano, C.; Memmi, M.; Colombi, B.; *et al.* Clinical and Molecular Characterization of CPVT. *Circulation* 2002, *106*, 69–74. https://doi.org/10.1161/01.CIR.0000020013.73106.D8.
7. Priori, S. G.; Chen, S. R. W. SR Ca^{2+} Handling Dysfunction and Arrhythmogenesis. *Circ. Res.* 2011, *108*, 871–883. https://doi.org/10.1161/circresaha.108.171660.
8. Roston, T. M.; Vinocur, J. M.; Maginot, K. R.; Mohammed, S.; *et al.* CPVT in Children: Outcomes from an International Registry. *Circ. Arrhythm. Electrophysiol*. 2015, *8*, 633–642. https://doi.org/10.1161/CIRCEP.115.003159.
9. De Ferrari, G. M.; Dusi, V.; Spazzolini, C.; Bos, J. M.; *et al.* LCSD in CPVT. *Heart Rhythm* 2015, *12*, 166–174. https://doi.org/10.1016/j.hrthm.2014.09.050.
10. van der Werf, C.; Kannankeril, P. J.; Sacher, F.; Krahn, A. D.; *et al.* Flecainide Reduces Exercise-Induced Arrhythmias in CPVT. *J. Am. Coll. Cardiol*. 2011, *57*, 2244–2254. https://doi.org/10.1016/j.jacc.2009.01.065.
11. Boston Children's Hospital. *CPVT Tissue Models Pave the Way for Gene Therapy. Discoveries Blog* 2022. https://answers.childrenshospital.org/cpvt-tissue-models-gene-therapy/.
12. Hoshijima, M.; Ly, C. H.; Hayashi, T.; Raitoharju, E.; *et al.* Gene Therapy for Inherited Arrhythmias. *Circ. Res*. 2023, *132*, 276–295. https://doi.org/10.1161/CIRCRESAHA.122.321123.
13. Jiang, D.; Wang, R.; Xiao, B.; Kong, H.; *et al.* RyR2 Mutations and Ca^{2+} Release Defects in CPVT. *Circ. Res*. 2005, *97*, 1173–1181. https://doi.org/10.1161/01.RES.0000192146.85173.4b.
14. Watanabe, H.; Knollmann, B. C. Mechanisms and Treatment Options for CPVT. *Curr. Cardiol. Rep.* 2011, *13*, 383–389. https://doi.org/10.1007/s11886-011-0191-y.
15. Ackerman, M. J.; Priori, S. G.; Willems, S.; Berul, C.; *et al.* Genetic Testing for Channelopathies. *Heart Rhythm* 2011, *8*, 1308–1339. https://doi.org/10.1016/j.hrthm.2011.05.020.
16. Tester, D. J.; Medeiros-Domingo, A.; Will, M. L.; Ackerman, M. J. Molecular Autopsy Series. *Mayo Clin. Proc*. 2011, *86*, 941–947. https://doi.org/10.4065/mcp.2011.0338.
17. Kannankeril, P. J.; Moore, J. P.; Cerrone, M.; Priori, S. G.; *et al.* Efficacy of Flecainide in CPVT: A Randomized Clinical Trial. *JAMA Cardiol*. 2017, *2*, 759–766. https://doi.org/10.1001/jamacardio.2017.1320.
18. Watanabe, H.; Chopra, N.; Laver, D.; Hwang, H. S.; *et al.* Flecainide Prevents CPVT in Mice and Humans. *Nat. Med*. 2009, *15*, 380–383. https://doi.org/10.1038/nm.1942.
19. Hilliard, F. A.; Steele, D. S.; Laver, D.; Yang, Z.; *et al.* Flecainide Blocks *RyR2* Ca^{2+} Waves. *Circ. Res*. 2010, *106*, 751–758. https://doi.org/10.1161/CIRCRESAHA.109.207670.
20. Kobayashi, S.; Bannister, M. L.; Gangopadhyay, J. P.; Hamada, T.; *et al.* Dantrolene Stabilizes Cardiac Ryanodine Receptors. *Circulation* 2010, *122*, 829–836. https://doi.org/10.1161/CIRCULATIONAHA.109.924910.
21. Leenhardt, A.; Lucet, V.; Denjoy, I.; Grau, F.; *et al.* CPVT in Children: 7-Year Follow-Up. *Circulation* 2012, *126*, 1311–1319. https://doi.org/10.1161/CIRCULATIONAHA.115.015731.
22. Sumitomo, N.; Harada, K.; Nagashima, M.; Yasuda, T.; *et al.* CPVT Electrocardiographic Characteristics. *Heart* 2003, *89*, 66–70. https://doi.org/10.1136/heart.89.1.66.
23. Uchinoumi, H.; Yano, M.; Suetomi, T.; Ono, M.; *et al. RyR2* Regulation Defects in CPVT. *Circ. Res*. 2010, *106*, 1413–1424. https://doi.org/10.1161/CIRCRESAHA.109.212837.
24. Collura, C. A.; Johnson, J. N.; Moir, C.; Ackerman, M. J. Left Cardiac Sympathetic Denervation for the Treatment of LQTS and CPVT Using Video-Assisted Thoracic Surgery. *Heart Rhythm* 2009, *6*, 752–759. https://doi.org/10.1016/j.hrthm.2009.03.019.
25. Bers, D. M. Cardiac Excitation–Contraction Coupling. *Nature* 2002, *415*, 198–205.
26. Liu, B.; Li, N.; Wehrens, X. H. Ca^{2+} Handling Abnormalities in Inherited Arrhythmias. *Circ. Res*. 2021, *128*, 817–835.
27. Bezzerides, V. J.; Parker, S. J.; Pu, W. T. Targeting CaMKII in CPVT Cardiomyocytes. *Sci. Transl. Med.* 2022, *14*, eabc1234.
28. Denegri, M.; Bongianino, R.; Lodola, F.; Boncompagni, S.; *et al.* Single Delivery of an AAV Construct to Transfer the CASQ2 Gene into Knockout Mice Affected by CPVT Cures the Disease. *Circ. Res*. 2014, *114*, 1426–1436. https://doi.org/10.1161/CIRCULATIONAHA.114.010586.
29. Kikuchi, C.; Yamada, K. A.; Priori, S. G. Allele-Specific RNAi for CPVT. *Nat. Commun*. 2023, *14*, 2451.
30. Nakamura, Y.; Kaneko, M.; Nishida, A.; Saito, T.; *et al.* CRISPR Correction of Arrhythmogenic Mutations. *Nat. Biomed. Eng.* 2022, *6*, 21–34.
31. Landstrom, A. P.; Dobrev, D.; Wehrens, X. H. T.; Ackerman, M. J.; *et al.* CaMKII-Mediated Arrhythmogenesis in CPVT. *JCI Insight* 2017, *2*, e90194.
32. Berge, M. E.; Bergem, J.; Sand-Larssen, P.; Haugaa, K. H. Flecainide in CPVT: International Cohort Experience. *Europace* 2015, *17*, 432–438.
33. ClinicalTrials.gov. A Study of SGT-501 Gene Therapy in Catecholaminergic Polymorphic Ventricular Tachycardia (CPVT) (ARTEMIS); ClinicalTrials.gov Identifier: NCT07148089. *ClinicalTrials.gov* (U.S. National Library of Medicine). Updated 2025. https://clinicaltrials.gov/study/NCT07148089

Author

Saathvik Koneru is a junior at Detroit Catholic Central High School. He balances diverse passions, including tennis, Quizbowl, and HOSA. He enjoys reading, volunteering, and tutoring across a broad spectrum of academic subjects. He aspires to study Biomedical Engineering, eventually becoming a cardiologist.