CYFIP2 Network Research Roadmap

This roadmap collects the knowledge, thoughts, ideas, and opinions of key stakeholders in the effort to develop treatments for Developmental Epileptic Encephalopathy 65 (DEE65), a condition caused most often by dominant, de novo pathogenic variants in the Cyfip2 gene. The roadmap is meant to serve as a guide to the Cyfip2 Network and allies worldwide in setting priorities for funding and other support to further research toward solutions for DEE65 patients.

Table of Contents

Honzik, Czech Republic

CYFIP2 NETWORK

The Cyfip2 Network was created in 2023 to raise awareness, support children and families affected by DEE65, and to fund research into developing treatments. The Cyfip2 Network has:

- Partnered with RARE-X to create an easy to access online patient Data Collection Plan
- Initiated development of an antisense oligonucleotide (ASO) therapy targeting the Cyfip2 Arg87Cys variant
- Secured funding to generate a conditional mouse model of the Cyfip2 Arg87Cys variant
- Brought together researchers from around the world with diverse scientific backgrounds to discuss Cyfip2 research in a series of virtual meetings

www.cyfip2network.org

This Research Strategy Map presents the detailed plan for supporting the development of treatments for DEE65 aimed at the root cause of the disease. In addition to the above stated research activities, interviews with the researchers and families of patients have suggested further activities aimed toward therapy development. These will be presented as potential programs the Cyfip2 Network could support, because achieving the goal of curing this disease cannot be confined to a single project or a single therapeutic option.

The strategic priorities also include non-research functions of the Cyfip2 Network, including supporting families with resources and community, and growing global awareness of DEE65. Some of those activities overlap with the research effort and will be briefly touched on in this document.

Eli, United Kingdom

About DEE65

DEE65, is a rare genetic condition that leads to seizures and prevents children from reaching developmental milestones like walking and speaking. It is one type of developmental epileptic encephalopathy, a severe form of epilepsy that is accompanied by developmental delay and encephalopathy. A recent review found that over 900 different genes were associated with DEE(Poke et al., 2023). DEE65 is caused by pathogenic variants in the Cyfip2 gene. There is no treatment or cure for this disease.

In the Unknown

GeneDx is on a mission to shorten and prevent the diagnostic odyssey. By providing clear, accurate, and meaningful genetic information, our comprehensive genetic tests help guide healthcare decisions, fuel the discovery of new genetic causes of disease, and accelerate the development of new therapies.

Since 2015 in the US, GeneDx has identified 27 patients with a pathogenic or likely pathogenic variant in the CYFIP2 gene.

In addition to helping individuals by enabling their genetic diagnosis, each patient tested at GeneDx enables us to more precisely interpret the genetic information of future patients we test—helping even more families find answers.

There is power in numbers. When patients come together, we can do great things.

This number is current as of February 2025 and is subject to change. For example, patients may opt out of data sharing or be part of institutional agreements which prohibits data sharing, new patients may be identified, and variant classifications may evolve.

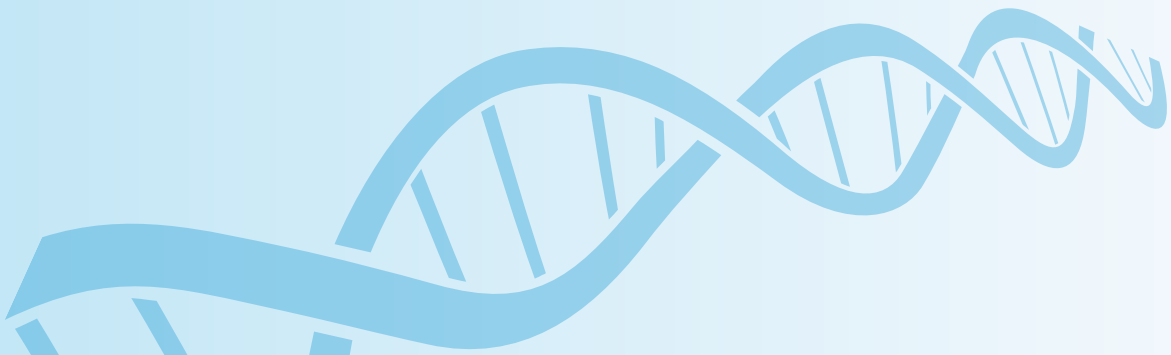
GeneDx

Courtesy of GeneDx, Current as of February 2025

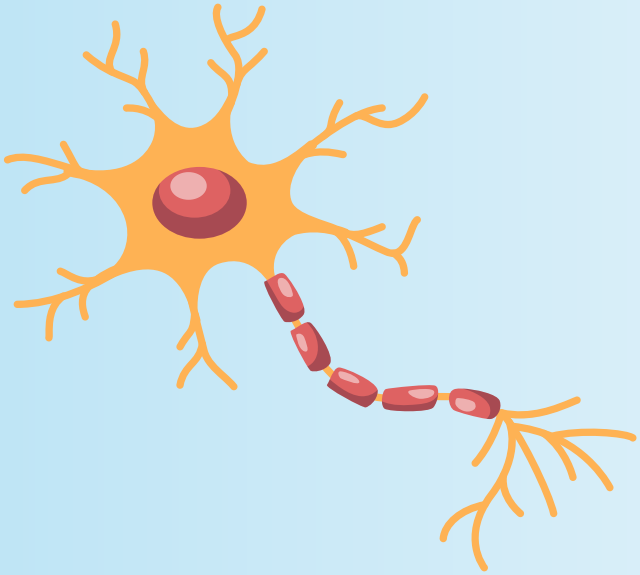
About Cyfip2

Cyfip2 is an abbreviation of Cytoplasmic Fragile X Messenger Ribonucleoprotein 1 Interacting Protein 2. The gene was first discovered in 2001 by researchers studying Fragile X syndrome(Schenck et al., 2001). The genetic cause of Fragile X syndrome is a gene called FMRP, and the Cyfip2 protein acts as a partner to FMRP. However, it was not until 9 years later that biochemists studying a protein called actin that another role of Cyfip2 became clear(Chen et al., 2010). Cyfip2, which is mainly found in neurons in the brain, keeps control over the protein complex that remodels the cytoskeleton, a network of protein cables inside the cell. When Cyfip2 is not functioning correctly the protein cables are laid down in places where they cause problems for the neurons, and brain development and function are affected.

To date, clinicians and scientists have confirmed that 26 different genetic variants in Cyfip2 are linked to DEE65, with two other variants suspected to be linked to the disease(Begemann et al., 2021; Zweier et al., 2019; Begemann, unpublished data). Almost half of the variants occur on Arginine 87, a critical amino acid for controlling the remodeling of the cytoskeleton. Most of the other variants also appear involved in this cytoskeleton control. Most DEE65 patients experience epilepsy, decreased muscle tone, and severe intellectual disability/developmental delay. A comprehensive natural history study of 46 patients is in progress (Begemann, unpublished).

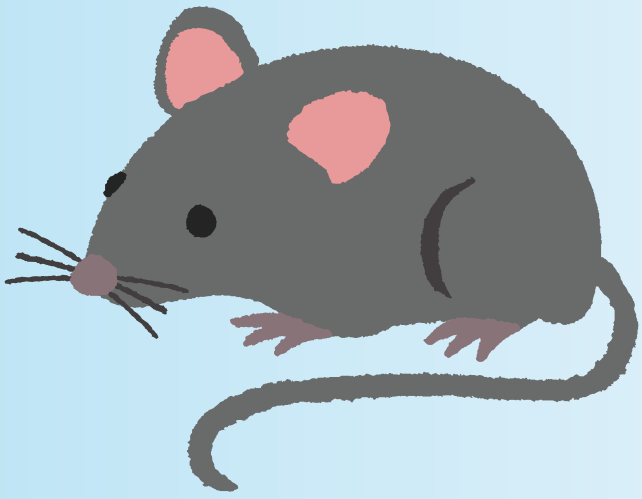


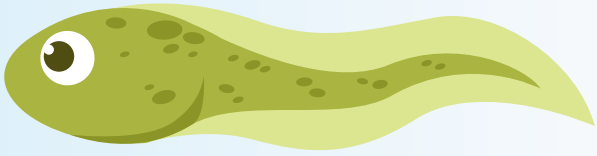
Background



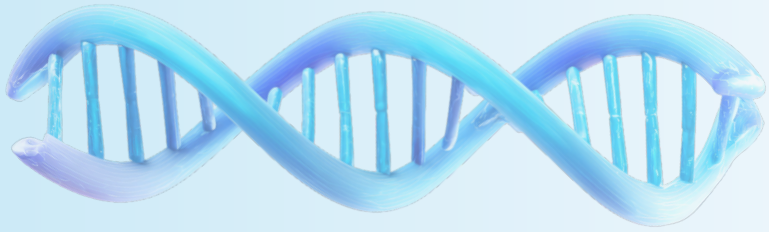
Disease models

Models of DEE65 that could be used to discover and test treatments for the disease include mouse and zebrafish Cyfip2 knockouts, a tadpole in which Arg87Cys Cyfip2 mRNA has been added temporarily, and a mouse with a genomic edit to change Cyfip2 to the Arg87Cys variant on one of the two copies. In addition, samples from patients have been used to create cell lines that carry several of the disease causing variants.





Treatments in Development



Treatments in development for DEE65 are in their early stages. Therapeutic options include a selective removal of the pathogenic messenger RNA by antisense oligonucleotide (ASO), repurposed drugs selected from those already approved by the FDA and other regulators, and compounds that regulate other aspects of the biological pathway impaired by Cyfip2 variants.

> “Recent studies at the molecular, cellular, and animal model levels are gradually uncovering the pathological mechanisms underlying CYFIP2-associated neurodevelopmental disorders. However, there are still significant gaps in the development of therapeutic strategies, indicating a need for further progress and collaboration between clinical and basic research.” Ma et al, 2024

Description of the Disease

Patients are currently diagnosed with DEE65 based on a clinical presentation, including intractable seizures that begin within the first six months of life, delayed psychomotor development, microcephaly, facial dysmorphisms, and hypotonia. This cluster of symptoms is sometimes termed Ohtahara or West syndrome (Scheffer et al., 2025). Since this cluster of symptoms indicates a genetic cause, patients are often evaluated with whole genome sequencing (WGS) or whole exome sequencing (WES). With over 900 different genes potentially leading to DEE, finding the identity of the genetic cause for each patient is a painstaking process.

Description of the Disease

The term DEE was introduced in 2017 to differentiate epilepsy syndromes where seizures are the cause of developmental delay from those where the developmental delay is an independent disease symptom. At that time it was clear that most of the DEE syndromes were caused by genetic variants. The most common form of DEE is Dravet syndrome. DEEs are complex neurological disorders and are therefore difficult to diagnose. Diagnosis and identification of the genetic cause often fail to lead to an effective treatment plan, but because some DEE syndromes are treatable with drugs, surgery, or dietary changes, it is important to establish the genetic cause as early as possible(Samanta et al., 2025).

DEE65 is a very rare subset of DEE. In an epidemiology study in Scotland in 2021 the incidence of DEE was approximately 1 in 1200 children born(Symonds et al., 2021). The most common DEE, Dravet syndrome, makes up about 7.5% of DEE cases in that study. It is very difficult to estimate the incidence of a disease as rare as DEE65. The latest natural history report from Anais Begemann in Switzerland identifies 46 DEE65 patients that have been reported in the literature or directly identified in Dr. Begemann's clinical practice. With the incomplete adoption of genetic testing worldwide it is probable that more cases exist but are undiagnosed.

West Syndrome

Ohtahara Syndrome

or

DEE65

Many rare diseases are given multiple names as different physicians describe related, or often the same set of symptoms independently. These syndromes are differentiated from one another based on the type of seizure, the age of onset, and the progression of symptoms. In the age of genetic testing, a new system of naming and classification has arisen, where each genetic cause of the disease is given a unique identifier, such as DEE65 for patients with Cyfip2 variants. Giving the syndrome a name like West or Ohtahara can give doctors an overall picture of the symptoms the patient has and suggest treatment regimens. Knowing the precise genetic cause may not lead to greater understanding of symptoms or lead to different treatment courses in the short term, but it could suggest a long term plan for drug development. Therefore we can still use the name of a doctor who described a set of symptoms long before the era of modern genetic testing (Dr. West described the syndrome that bears his name in 1840) as a shorthand for a complex and variable syndrome.

Signs and Symptoms

Patients with DEE65 all experience intellectual disability and developmental delay. Most also have epilepsy, consisting of different types of seizures (including tonic, myoclonic, and tonic-clonic seizures, absences, and epileptic spasms) and abnormalities in their MRI and EEG tests. The majority of patients exhibit hypotonia (muscle weakness) and about half show smaller head size than is normal for their age and weight (microcephaly). Many patients also exhibit visual problems (either optic atrophy, cortical vision impairment, or crossed eyes), feeding difficulties, and behavioral problems that are similar to autism spectrum disorder(Begemann et al., 2021). Some patients have been found to lack the normal complement of neutrophils in their blood. Although the reported incidence of this neutropenia is low, it is not a test that would normally be ordered in a work up for an epilepsy patient. Therefore it is possible that Cyfip2 variants also affect either the production or survival of neutrophils and could therefore compromise the patient's response to infection.

Isabella and her family, Brazil

"We place our hope in science to significantly improve Isabella's quality of life, especially when it comes to her communication."
-Isabella's Mom

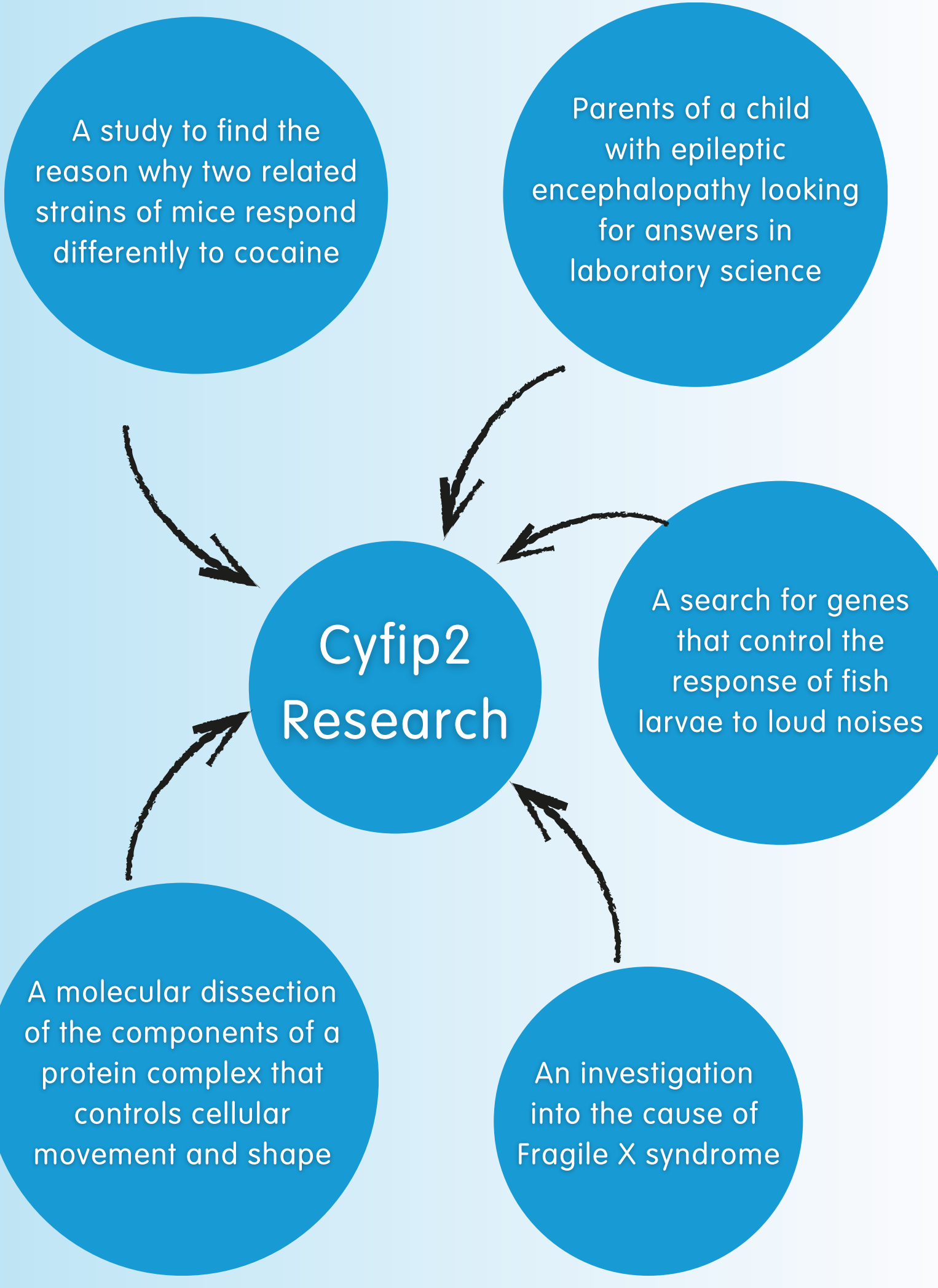
Current Treatment Options

Some DEE65 patients have been treated successfully with anti-seizure medications as reviewed in Squire, 2025. The authors mention the effectiveness of valproic acid and lacosamide, but the antiseizure medications that have been reported in case studies varies considerably. Sometimes caregivers can take on the responsibility to choose an effective treatment regimen from within a set of medications and doses give by their doctors. Encouraging clinicians to share their experience treating DEE65 patients with one another may shorten the time needed to optimize treatment for future patients. Research in mice and anecdotal evidence from patients raises the possibility that seizure activity may subside a few years after birth but reappear later in life.

The non-seizure symptoms of DEE65, like other DEEs, are difficult to characterize and have few treatment options. Caregivers and parents of DEE patients report communication and gross motor function as their top priorities for treatments(Hecker et al., 2024). DEE65 parents expressed that while seizures are the major focus of medical visits and day to day interventions, the non-seizure symptoms, including sleep problems, are equally if not more concerning.

Research Landscape

Research into the function of Cyfip2 began in 1999, when the gene was first called PIR121(Saller et al., 1999). It was not until 2018 that clinicians in Japan linked four cases of DEE with variants in Arginine 87 of Cyfip2(Nakashima et al., 2018). While the number of laboratories studying Cyfip2 and DEE65 remains small, significant progress in our understanding of the causes of the disease has resulted from scientific interest in several aspects of Cyfip2 function.


A study to find the reason why two related strains of mice respond differently to cocaine
Parents of a child with epileptic encephalopathy looking for answers in laboratory science
Cyfip2 Research
A search for genes that control the response of fish larvae to loud noises
A molecular dissection of the components of a protein complex that controls cellular movement and shape
An investigation into the cause of Fragile X syndrome

How do scientists come to be interested in studying Cyfip2 and DEE65?

Scientists take up an area of study for many reasons. Some are guided to a particular research project by their career mentors. Others become intrigued by a scientific report and initiate a new line of investigation. Sometimes patients or caregivers of patients request that a researcher investigate their disease, sparking a lifelong interest in finding treatments. The leading Cyfip2 researchers were asked what led to their interest in this area of biology. Their responses varied, but all led to significant contributions to the field.

Honoring the memory of Marcellus, United States

Case Histories

For an ultra rare disease, every patient that is described to the scientific community adds significant data to our understanding of that disease. Clinicians who discover a genetic diagnosis of a DEE usually report that result in a scientific paper called a case history. To date there have been 8 case history reports in the literature, including two larger reports that compile the previous case histories, called retrospective natural histories(Amato et al., 2025; Arisaka et al., 2021; Begemann et al., 2021; Da Silva Cardoso et al., 2023; Nakashima et al., 2018; Peng et al., 2018; Salokivi et al., 2024; Zhong et al., 2019; Zweier et al., 2019). These reports compare the clinical characteristics of each patient as well as the genetic cause of the disease (the specific pathogenic variant in Cyfip2). The resulting data allows for a genotype/phenotype correlation, a grouping of patients based on their disease course and their specific mutation. With a very small data sample, this genotype/phenotype correlation is imperfect. It is impossible at this point to predict the clinical course a patient might experience based on their pathogenic variant. However, it does appear so far that variants at position 87 lead to more severe symptoms than variants at other parts of the gene.

Gain or Loss of Function?

To develop an effective treatment for DEE65 it is important to determine if the disease is caused by a "gain of function" or a "loss of function" of Cyfip2. A gene that is "lost" or mutated in such a way that no functional protein is synthesized could be treated by gene replacement. However, when a gene gains function, or takes on a different, damaging role in the cell because of a change in the amino acid sequence, reducing the amount of the damaging protein would be an effective treatment.

One important piece of information used to determine if Cyfip2 pathogenic variants represent a gain or loss of function is the genotype/phenotype correlation. If patients exist that harbor a complete loss of the gene, a null variant, then a loss of function mechanism is possible. However, in the most recent overview of the case histories Dr. Anais Begemann has found that all of the described case histories from patients where the genetic cause is confirmed feature small changes in the gene that would not result in a null variant. Therefore it is most likely given the information available to date that gain of function in Cyfip2 is the mechanism for DEE65. There are more ways to determine whether gain or loss of function is the mechanism for DEE65 that will be discussed below.

Cyfip2 Data Collection Program

The Cyfip2 Network has partnered with RARE-X to collect information about DEE65 patients and facilitate research and treatment development. The Data Collection Program allows families to participate in research at no cost and with no required clinic visits.

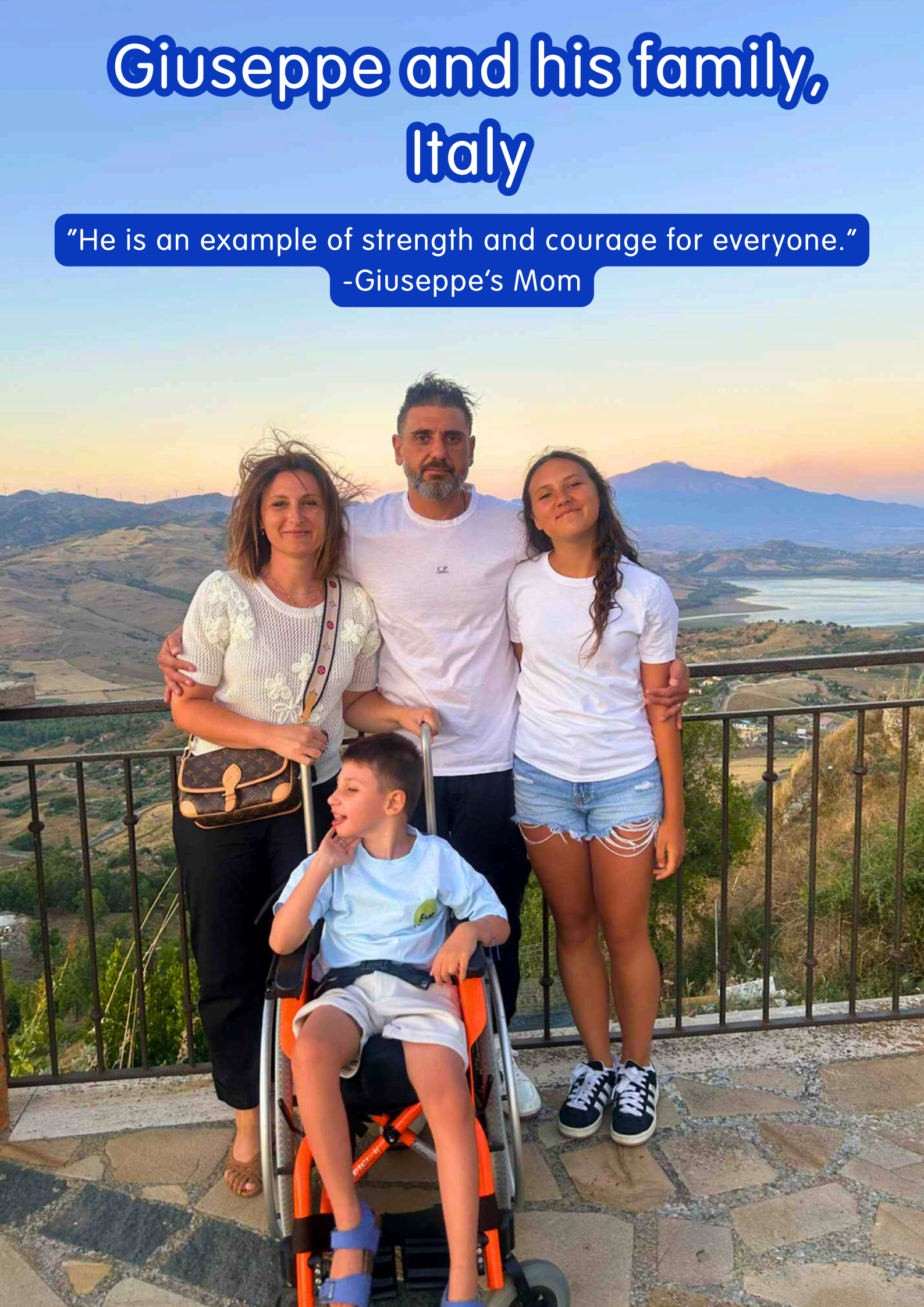
When caregivers log into the platform they are given a “head to toe” survey of the patient. Based on the answers to that survey, additional surveys are generated. In addition to seizure and spasm details, the surveys may include questions about sleep disturbances, psychomotor impairments, communication ability, aberrant behavior, gastrointestinal issues, visual problems, and immune health. The platform will also collect information about medical interventions, diet, physical therapy and patient and caregiver quality of life. This information will help researchers study the mechanism of the disease and potentially create treatments for patients.

Sign Up Here

RARE-X
A Research Program of Global Genes

Giuseppe and his family, Italy

"He is an example of strength and courage for everyone."
-Giuseppe's Mom



Cyfip2 Biochemistry

Cyfip2 was first identified as a gene that was turned on by a mutant form of the protein p53(Saller et al., 1999), which is often mutated in human cancers. The connection between Cyfip2 and p53 has led to dozens of research articles on the role of Cyfip2 in cancer. While these cancer focused studies do not directly connect with the role of Cyfip2 variants in DEE65, the continued interest in the protein from the very active and well funded cancer research community could serve to advance DEE65 therapeutic development.

Landon, United States

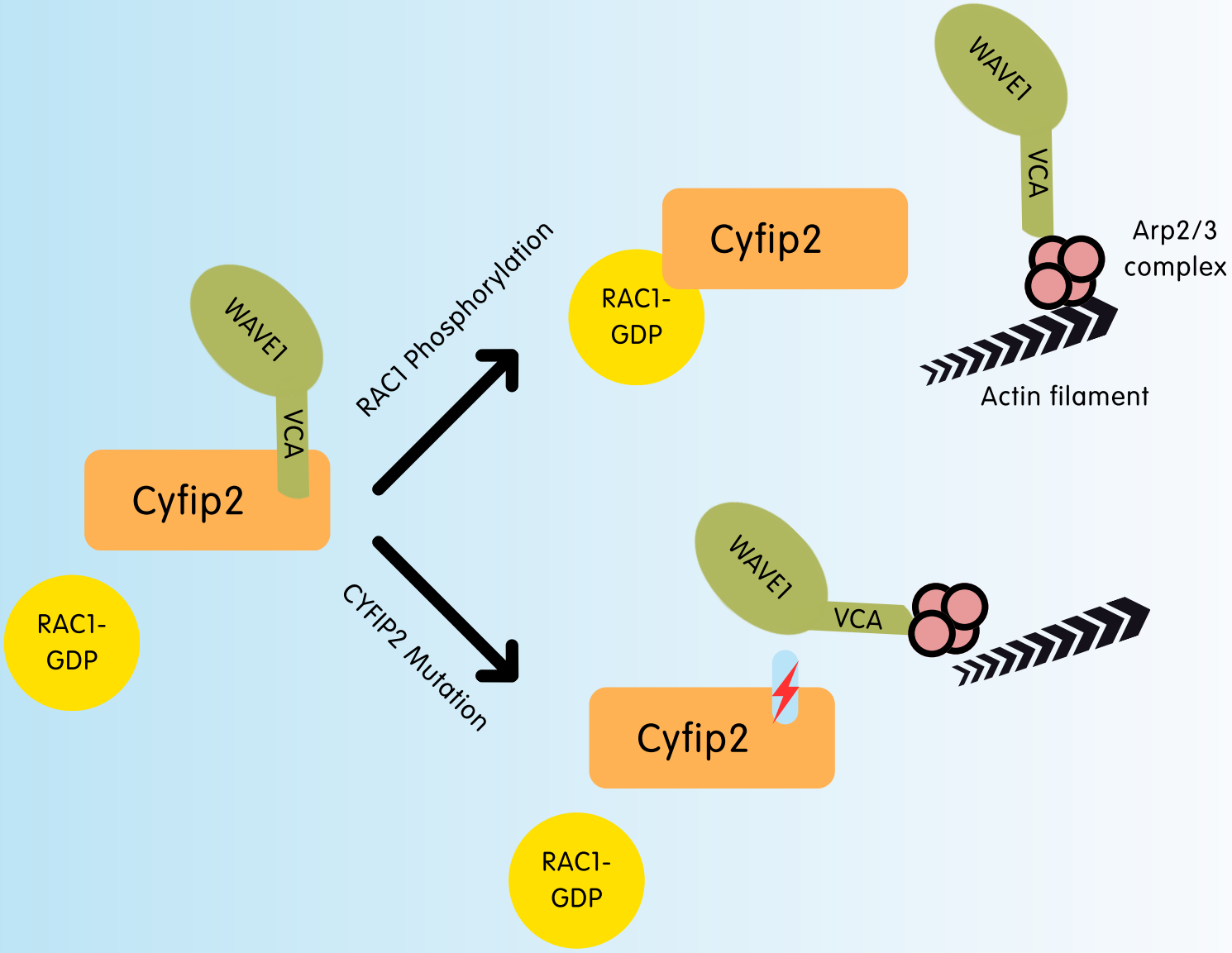
Cyfip2 Biochemistry

Cyfip2 interacts with a protein called FMRP (Fragile X Mental Retardation Protein, or Fragile X Messenger Ribonucleoprotein). Using a method to "fish" new protein binding partners out of a yeast cell using FMRP as the "bait" two new genes were discovered, Cyfip1 and Cyfip2 (Schenck et al., 2001). Cyfip1 and 2 are among the most studied of the FMRP associated proteins, and yet their role in FMRP's functions are still not clear. FMRP binds to RNA, controlling the process by which RNA is read to produce proteins (translation). Cyfip1 is apparently important to coordinate RNA translation with cytoskeleton remodeling(DeRubeis et al., 2013). Cyfiip1 and Cyfip2 are very similar to one another (88% of the amino acids in the protein are identical) but they have different jobs in the cell.

The WAVE Regulatory Complex

In 2010 a new role for the Cyfip proteins was uncovered. Cells are held together by lipid membranes that form the walls and protein filaments that maintain internal rigidity like the cables that hold up a suspension bridge. One of the proteins that make up these filaments is called actin. The Wave Regulatory Complex, or WRC, is an important switch that determines where and when actin filaments are formed. Scientists at UT Southwestern were examining the components of the WRC and found that Cyfip1 plays an important role in organizing the WRC and holding a component called the VCA in an inactive state(Chen et al., 2010). Cyfip2, which is nearly identical to Cyfip1, can take this role in the WRC as well. When the Cyfip proteins are not functioning properly the VCA is active at all times and actin filaments form in places where the cell doesn't need them.

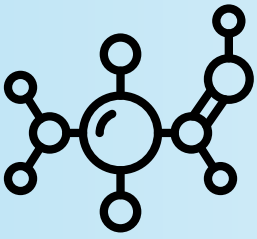
In neurons the precise formation of actin filaments is important to guide neuron extensions called dendrites and axons to their proper targets, and to form the connections at those targets called synapses(DeRubeis et al., 2013; Han et al., 2014; Y. Kim et al., 2024). While there has been no direct demonstration that loss of actin filament control, axon guidance, and proper synapse formation leads to the symptoms of DEE65, that is the prevailing hypothesis for how Cyfip2 pathogenic variants cause disease.

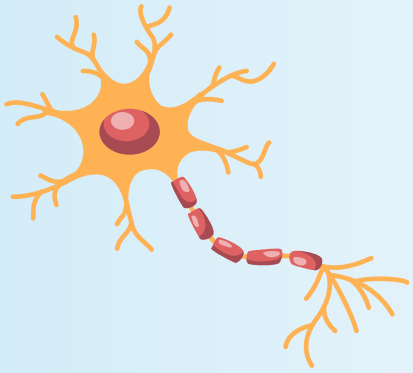


Schematic drawing of the Wave Regulatory Complex and the proteins that interact with it. Cyfip2 is a large protein that acts as a scaffold for assembly of the complex. It also keeps the part of WAVE1 called VCA in a pocket, preventing it from triggering actin filament assembly until the protein Rac1 activates the complex. When Cyfip2 can't hold VCA in that pocket (the red lightning bolt) actin filaments are formed even when Rac1 is not activating the process. These extra actin filaments form in parts of the cell where they are not needed and impair neuron function (Zweier et al., 2019)

Cyfip2 Research Tools

To study any disease, whether to gain a greater understanding of the cause, refine the diagnosis, or develop new treatments, scientists have to recreate the disease in a controlled lab setting. The simplest form of disease models may require a few proteins packed into a crystal lattice that can be illuminated by X-rays. More complex are mixtures of proteins, nucleic acids, and chemical compounds that reproduce the interactions between cell components. Some disorders can be modeled in cell culture while others may require an entire living organism to capture all of the mechanisms that lead to disease progression. The most complex lab models of disease involve mammals with the pathological variant introduced into their DNA.

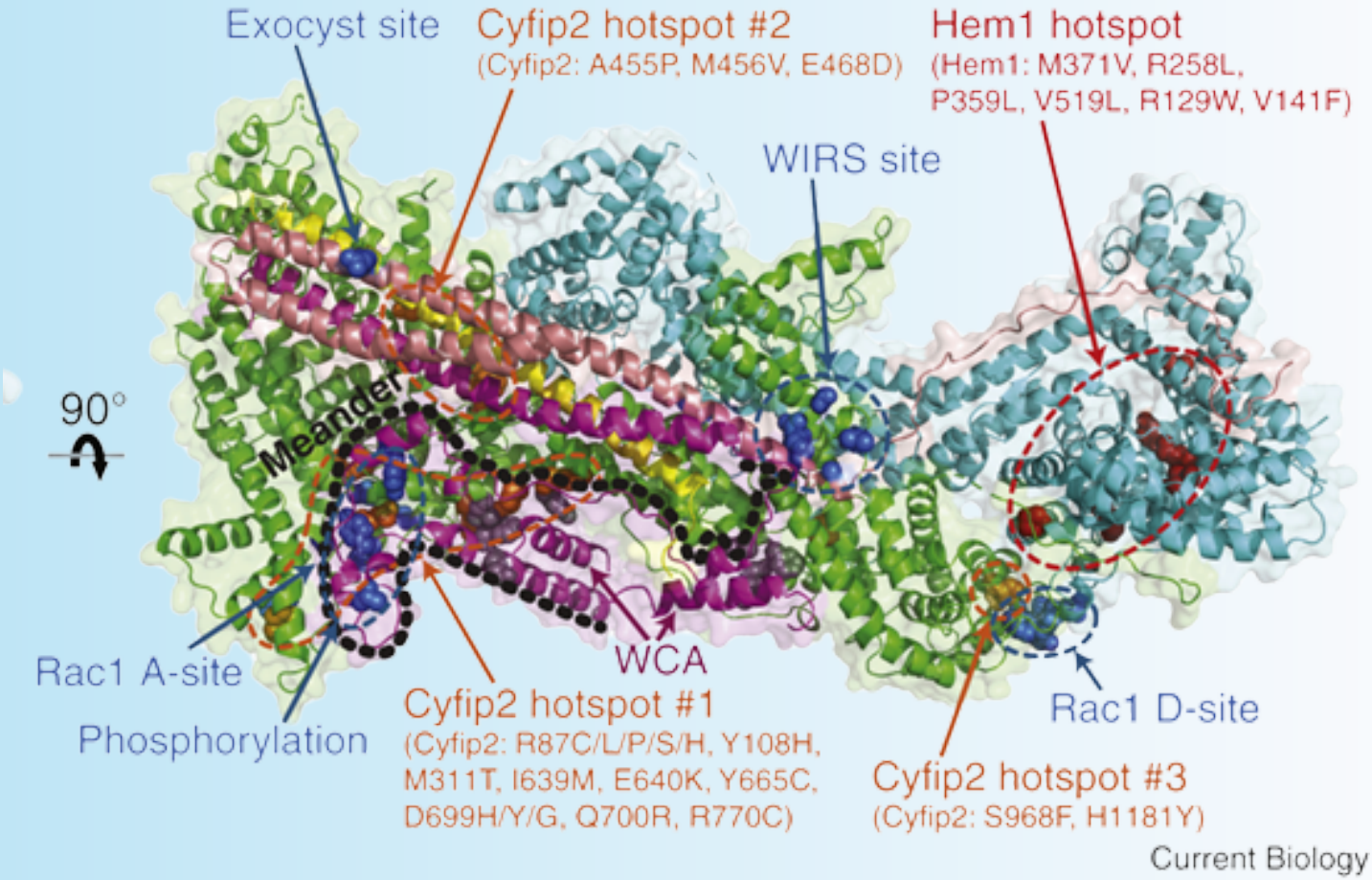




Increasing complexity

WRC Crystal Structure

Cyfip2 is a member of at least two protein complexes. While the X-ray crystal structure of the Wave Receptor Complex containing Cyfip1 has been reported(Chen et al., 2010), the structure that contains Cyfip2 has not. The two proteins share a great deal of similarity and this has allowed a recreation of the WRC structure with Cyfip2 in a computer model(Biembengut et al., 2022). This "in silico" model can be further animated by a computational process called molecular dynamics to reveal the mechanism of WRC dysregulation by Arginine 87 variants. It can also be used to screen for compounds that might reverse the effect of those variants(Venturi Biembengut et al., 2024).



Cell Lines

The research tools needed to introduce any mutation into a cell's DNA allow scientists to create all of the known pathogenic variants in the cell type of their choice. However, it is preferable in many cases to use cells collected from patients and close family members since the rest of the patient's genome may influence the effect of the Cyfip2 variant. Cells can be collected from blood, cheek swabs, biopsies or, in the case of a 2023 paper(Da Silva Cardoso et al., 2023) urine specimens. Fibroblasts from DEE65 patients in culture show a deficit in the formation of a structure called a dorsal ruffle (Begemann et al., 2021). This is both an indicator of actin filament dysfunction as well as a potential starting point to create a cell-based screening assay for a drug repurposing effort.

> When I am working on a cell line in the lab, I remind myself that this is a cell from a patient who has a family. It is important to stop and think about that sometimes.
> – Dr. Isabelle Zaboroski Silva

Cell Lines

Animal models (to be discussed below) have shown that Cyfip2 activity loss impairs the ability of neurons in the developing brain to find their intended target(Cioni et al., 2018). Patient derived cell lines can be induced to become neurons in culture. If the DEE65 causing variants disrupt axon guidance the same way that Cyfip2 gene knockout does, it should be evident in the way the neurons grow in culture. This study is in progress at Washington University St. Louis.

Flies and Fish

Fruit flies are extraordinarily useful model organisms for genetic studies, because they are quick to reproduce and relatively easy to genetically manipulate. The fruit fly has only one form of Cyfip. By removing part of the protein the researchers found that fruit fly Cyfip plays a role in axon guidance, synapse formation and actin polymerization(Schenck et al., 2003). They did not analyze behavior in the flies. There have been no reports of DEE65 causing variants in fruit flies.

Zebrafish share many of the attractive features of fruit flies for genetic studies but have the added benefit that they are vertebrates. A mutant zebrafish strain called nevermind, in which axons meant for the visual cortex fail to find their targets, is a null variant for Cyfip2(Pittman et al., 2010). A separate study looking for genes that regulate the fish larva's response to stimuli also showed that a Cyfip2 null variant (this one called triggerhappy) had a reduced startle threshold(Marsden et al., 2018). This model allowed scientists to test the effect of removing and adding back the gene at different times, and they found that the change in startle threshold was reversible. While this finding may not be directly applicable to DEE65 causing variants in human brains, it is one indication that the role of Cyfip2 involves maintenance of neuron function and not simply development of functional neural networks. Development of a zebrafish expressing DEE65-causing variants in Cyfip2 is underway at North Carolina State University in the laboratory of Dr. Kurt Marsden.

Mouse Models

Compared to fish and invertebrates, mammals are more difficult to keep in the lab, take up more space and cost more to keep healthy. Despite these drawbacks, most scientists consider mouse models essential to understanding how a diseases develop and whether treatments will be effective in patients. Like fruit flies and zebrafish, the tools needed to manipulate the genes of mice are well developed. Unlike fruit flies and zebrafish mice can be dosed using routes that are similar to the way human patients would be dosed: orally for some pharmaceutical drugs, intravenous, or into the center of the brain or spinal cord for oligonucleotides and viral gene therapies.

The first mouse models exploring Cyfip2 came about serendipitously. Scientists were studying the response of mice to cocaine and found two closely related strains of laboratory mice responded differently to the drug. The strain C57Bl/6J reacted more strongly than the strain C57BL/6N(Kumar et al., 2013). These two mouse strains differ only slightly in their genetic code. One difference in Cyfip2 that makes the protein less stable proved to be the cause of the lowered cocaine response in the 6N mice. This result led to an exploration of the role of Cyfip2 in impulsive behavior and reward circuits that control not only cocaine responses but also alcohol intake and binge eating(Hartmann et al., 2023).

Mouse Models

Using a strategy called knock-out, where a gene is altered so no protein product is produced, Kihoon Han and colleagues showed that Cyfip2 is essential for mouse survival(Han et al., 2014). Since most genes exist in two copies or alleles (one from each parent) these researchers studied mice that only kept one allele of the gene (heterozygous knockouts). These mice displayed hyperactivity and appeared quite similar to mice lacking FMRP. While the knockout strategy is an important way to explore how a gene affects biological pathways, a more laborious procedure is necessary to study a pathogenic variant that results in a gain of function. It was not until 2023 that such a mouse model was generated, and this accomplishment represents a major advance in the development of therapies.

Mice expressing the Arg87Cys variant in Cyfip2 in one of their two copies of the gene have reduced body weight and strength(Kang et al., 2023). They display spontaneous epilepsy-like spasms which disappeared as the mice aged but recurred when they reached adulthood(Ma et al, 2025). They had a lower response to social interaction in tests that are used to study autism spectrum in mice. The mice have an increased sensitivity to the seizure inducing drug PTZ. All of these results suggest that the Arg87Cys Cyfip2 mouse is a valid model of the human disease. The difference in the heterozygous knockout mice and those expressing the Arg87Cys variant also are strongly suggestive that the disease mechanism involves a gain of function. These mice should be useful in predicting the disease course in patients and would also be helpful in testing any new therapeutic strategies, including pharmaceutical drugs and genetically targeted therapies.

New Mouse Model

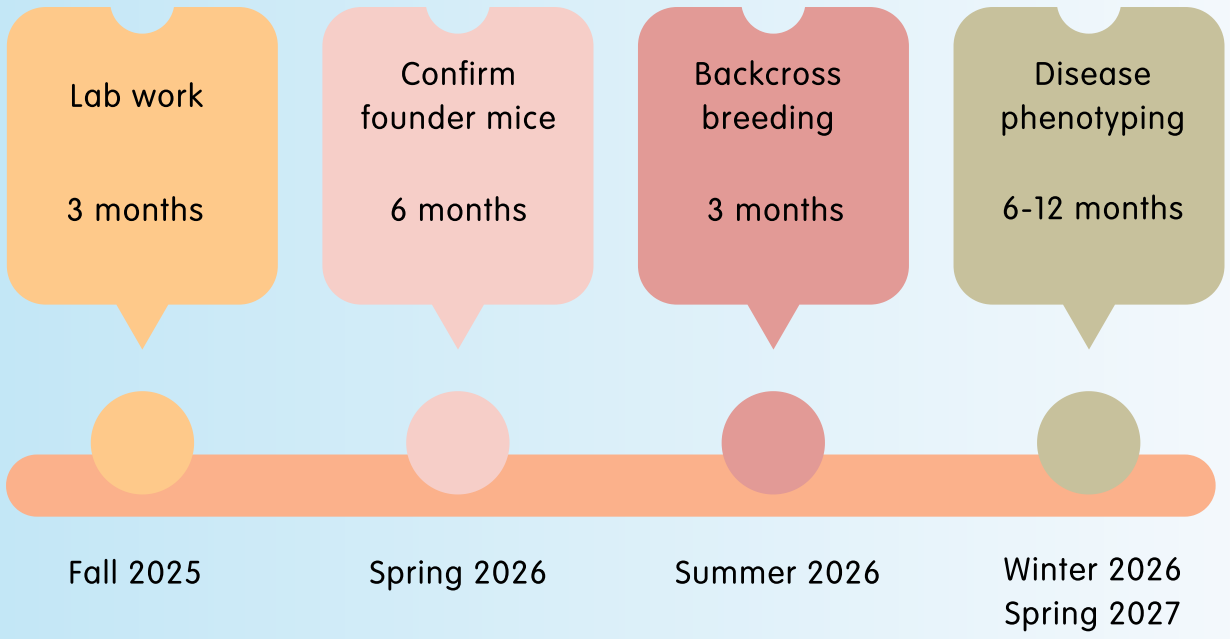
The mouse model developed by Dr. Han and colleagues at Korea University represents a major step in the understanding of DEE65 and the development of treatments. However, one of the major gaps in our understanding of DEE65 that expert scientists identified is the time and place where Cyfip2 dysregulation affects neurons. This information is key to developing treatments, because those treatments will have to create their effect at a specific time (the time of treatment) and in a specific location (depending on how the treatment is delivered to the patient). We need to ensure that the time and place of drug effect is consistent with the time and place where the disease mechanism occurs.

For this reason, the Cyfip2 Network has partnered with the Czech Center for Phenogenomics (CCP) to produce a conditional mouse model of DEE65. In this model, the pathogenic variant Arg87Cys is introduced in such a way that it will only be expressed in cells in the mouse that also express the protein Cre. This will allow scientist to breed mice that only express the pathogenic variant in one particular type of neuron, for example, or only express the Arg87Cys form at some defined point after birth. This increased flexibility demonstrates the remarkable power of mouse genomics.

After a successful grant application, the CCP has generously offered to create the conditional mouse model at no cost to the Cyfip2 Network. Once the mice are born and the conditional expression of the pathogenic gene has been confirmed, the mice will have to be characterized in a series of experiments. The budget for performing these experiments is still in development.

Developing therapeutics for these kids involves an intense amount of work, but we still need to work harder. – Dr. Vivek Kumar

Mouse Model Generation at the Czech Center for Phenogenomics



Our immediate goal is to delineate the detailed neurobiological changes occurring in the brain of Cyfip2 Arg87Cys mice. Based on those insights we aim to identify and validate therapeutic targets. – Dr Kihoon Han

Tadpoles

Finally, the African Clawed Frog (Xenopus laevis) has become an important research tool for the major reason that the eggs are so large they can be injected with proteins and nucleic acids with far less difficulty than other vertebrate cells. Injecting Xenopus eggs with Cyfip2 wild type RNA resulted in slightly hyperactive tadpoles, whereas injecting Cyfip2 Arg87Cys RNA caused the tadpoles to develop spontaneous seizure behavior and abnormal brain activity(Panthi et al., 2022). This model represents another strong indicator of a gain of function mechanism, since producing the seizure phenotype did not require removing the wild type Cyfip2. These tadpoles could also be used to screen potential treatments, in higher numbers than would be possible for mice.

Fin and his family, Switzerland

Disease Models Available

Model	Location
Patient-derived fibroblasts (6 different lines)	University of Zurich
Patient-derived iPSCs (2 patients, both Arg87Cys)	Institute Carlos Chagas/Fiocruz
Patient-derived fibroblasts	Coriel Institute (not cataloged)
Xenopus Laevis tadpoles expressing 2 different pathogenic variants of Cyfip2	Univeristy of Otago, New Zealand
Cyfip2 knockout zebrafish	NCSU
Cyfip2 knockout mice	International Mouse Phenotyping Consortium
Cyfip2 Arg87Cys edited mice	Korea University

Honzik, Czech Republic

"I hope Honzík will learn to walk and talk better. I would also like to help others."

-Honzik's Mom

Gaps in the Research Landscape

While significant progress has been made since Cyfip2 was first identified as the gene associated with DEE65, much remains to be done:

Basic scientific understanding

- Where and when is Cyfip2 expression important in brain development?
- What proteins make up the WRC in partnership with Cyfip2?
- How is Cyfip2 activity regulated?
- What is the role (if any) of the Cyfip2/FMRP interaction in DEE65?
- How do the variants other than Arg87x affect activity?

Research tools

- Conditional mouse model (allows researchers to restrict the pathogenic variant to certain tissues or to certain times during development)
- Complex cell based models, such as 3D culture and brain organoids (to explore what cell types are most vulnerable to Cyfip2 pathogenic variants and to reproduce seizure-like responses in a more controllable model system)

What can Cyfip2 Network and their allies do to help?

Purpose	Project	Research Location/ Investigator	Cost
Disease Model Generation	Collect biological samples from patients to create cell lines (fibroblasts or iPSCs)	University of Brescia, Italy/ Alessandro Barbon	
Disease Model Generation	Characterize Arg87Cys Cyfip2 mice being generated at Czech Institute	UC Davis, CCP	

Therapeutic Development

Photo: Eli during a DMI Intensive Course!

Therapeutic Development

One of the most important goals of the Cyfip2 Network is to accelerate the development of disease modifying therapies for DEE65. Current treatments focus on seizure control, which is important, but does not address the other aspects of the disease.

> "Morbidities include developmental delay and regression resulting in intellectual disability; psychiatric features including autism spectrum disorder, mood disorders, anxiety, and psychosis; gastrointestinal, musculoskeletal, respiratory, and cardiac manifestations, together with a considerably increased mortality rate."
> Scheffer et al., 2025

On a positive note, we are entering into a new era of medicine where genetically targeted drugs that can reduce, replace or even repair defective genes are currently in development for rare and even ultra-rare diseases.

Drug Repurposing

Creating an entirely new pharmaceutical drug is an expensive and time consuming process and there is little economic incentive to pursue new treatments for ultra-rare diseases. However, finding a new use for an old drug is relatively inexpensive and fast. A little over 300 chemical compounds that act on targets within the brain have been approved for use in humans, and while none of those drugs were approved to treat DEE65, it is possible that one of them has a biological effect that would be beneficial to patients.

Lucas, United States

Drug Repurposing

Discovering which of the approved drugs might benefit DEE65 patients can be done in a few different ways. Researchers at the Brazilian institute Fiocruz used a computer model of Cyfip2 to "dock" chemical compounds into the Arg87Cys location(Venturi Biembengut et al., 2024). They found 11 compounds that they predicted would bind tightly. To test these results against the actual protein they added the drugs to cells to test whether they would change the temperature at which the protein became denatured. They found that 8 compounds increased the thermal stability of Cyfip2, indicating that these compounds bind to the protein. The next step to determine if these compounds would be beneficial to DEE65 patients is to test them in a cell-based assay that would indicate either a restoration of normal Cyfip2 function or removal of the unwanted function of the Arg87Cys variant. At least three labs are actively creating such cell-based assays: Patricia Shigunov's lab at Fiocruz, Joseph Dougherty's lab at Washington University St. Louis, and Eileen Kennedy's lab at the University of North Carolina. It is vitally important to use an assay that accurately reflects the disease, so it would be helpful to use a few different cell-based assays for this purpose. The compounds should also be tested in the context of other pathogenic variants, because those variants also cause changes in the same region of the protein as Arg87Cys. This would require the creation of a cell line from patients with the other pathogenic variants.

Drug Repurposing

A second way to discover potential drugs to repurpose is to perform a high throughput screen in a relevant assay. This could be a cell-based assay (for example the dorsal ruffle test in fibroblasts described above(Begemann et al., 2021)), or a test in animals small enough to use in an experiment with over 300 conditions (tadpoles (Panthi et al., 2022) or zebrafish, for example). Development of an assay suitable for high throughput screening of approved drugs is an activity that is financially within reach of patient advocacy groups and has potential to significantly impact the search for effective treatments.

Isabella, Brazil

Therapeutic Development

Drug Repurposing

University of North Carolina, Chapel Hill

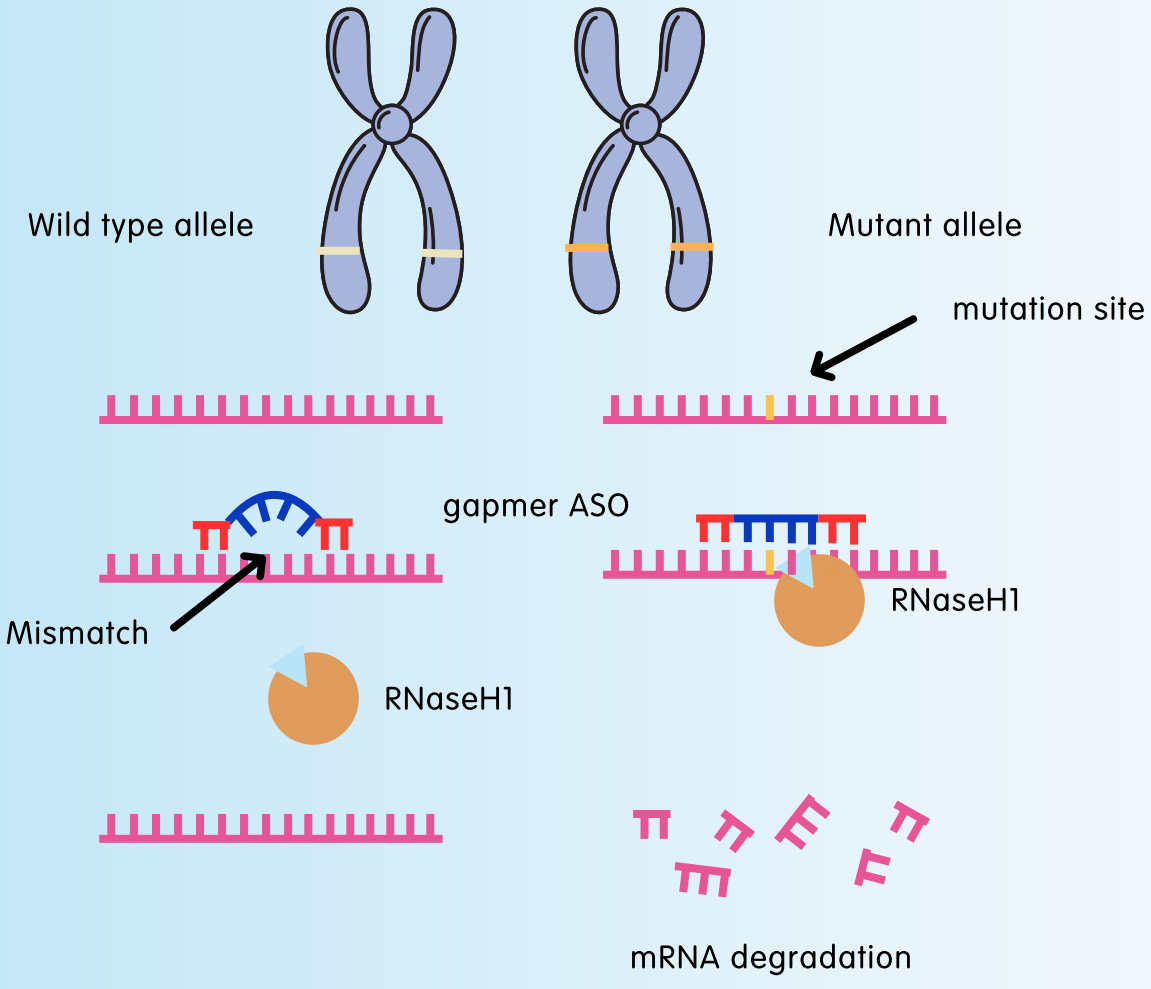
Another approach to repurposing drugs is to take advantage of the fact that in addition to DEE65, Cyfip proteins are also involved the process of cancer growth and metastasis. Researchers at the University of Georgia and University of North Carolina Chapel Hill have developed compounds that alter the function of the Wave Regulatory Complex(Limaye et al., 2022). Those compounds act on cells that express the wild type Cyfip proteins, but it will be an important next step to determine if they remove the harmful effects of pathogenic variants of Cyfip2. While these treatments are still a long way from human studies, they are being developed for a disease that attracts a great deal of resources which will move them forward more quickly than any treatment specific for DEE65.

Allele selective ASOs

Cyfip2 is present in two alleles in every cell (one on each chromosome 5). In DEE65 patients one allele is normal and the other copy carries a pathogenic variant. The variant is often only one nucleotide different from the normal allele (called a single nucleotide polymorphism, or snp, pronounced "snip"), but this one nucleotide is enough to produce a protein that has a harmful effect on the cell and ultimately the patient. In mice, removal of both alleles of Cyfip2 is not tolerated. The mice with no Cyfip2 at all don't live past birth. Mice with only one allele of the gene live past birth, although they do have some differences from those with both alleles(Han et al., 2014)(G. H. Kim et al., 2020; Zhang et al., 2019, 2020). These differences are not as severe as those seen in the mice expressing Arg87Cys in one allele(Kang et al., 2023). Considering this mouse data along with the fact that all DEE65 patients studied to date express some form of Cyfip2 (as opposed to a null variant) suggest that the disease mechanism is a gain of function. Therefore it may be beneficial to patients to remove the pathogenic allele, to reverse the gain of function, if it possible to do so and leave the normal allele in place.

Allele selective ASOs

A therapeutic approach called RNA interference, gene silencing or knockdown could achieve this allele selective removal of Cyfip2. An oligonucleotide that is complementary to the Cyfip2 messenger RNA, an antisense oligonucleotide or ASO, could be constructed that exactly matches, for example, the Arg87Cys variant. Because the ASO doesn't exactly match the normal allele, it would not interfere with the production of the normal protein. Such allele-specific gene silencing oligonucleotides have been generated for Huntington's disease(Conroy et al., 2022). The design of allele-specific ASOs is described in the paper and the authors showed that the method is applicable to other genes.



Allele selective ASOs

Design of an allele-specific ASO for the Arg87Cys variant would benefit the most DEE65 patients, but it would be even more beneficial to design an ASO that could silence any pathogenic allele. In the laboratory of Joseph Dougherty at the Washington University of St. Louis, researchers identified a “silent” snp called rs1823035 in the Cyfip2 gene that causes no change in the protein sequence and is found in approximately 1/3 of alleles. By targeting one or the other version of this silent variant, it may be possible to silence the pathogenic allele without affecting the normal allele.

What can Cyfip2 Network and their allies do to help?

Purpose	Project	Research Location/ Investigator	Cost
Drug repurposing	Test compounds from in silico screen in cell-based model	Fiocruz/Shigurov	
Drug repurposing	Screen compounds in zebrafish	NCSU/Marsden	
New drug development	Test Cyfip2 targeting stapled peptides in cell-based model	UNC/Kennedy	
Allele-specific gene silencing	Test pathogenic variant specific ASOs in cell-based model	WUSTL/Dougherty	


Ada and her family,
United States

Treatments with potential for all DEEs

Because DEE65 shares symptoms and prognosis with other DEEs it is worth exploring treatments that are effective in treating other DEEs and examining whether they might have utility in treating DEE65.

Bexicaserin

With the acquisition of Longboard Pharmaceuticals by Lundbeck, the development of the first treatment aimed at all DEEs has moved to a large company with significant resources and expertise at moving neurological drugs towards market approval. Bexicaserin acts on a specific serotonin receptor 5-HT2C. Drugs like fenfluramine that target serotonin receptors non-selectively reduce seizures but cause unwelcome side effects (sedation and valvular heart disease, among others). Compounds that specifically activate 5-HT2C were considered promising because mice lacking 5-HT2C display spontaneous seizures(Tecott et al., 1995). Bexicaserin reduces seizures in DEE patients and has so far raised no serious safety concerns(Chan et al., 2025). The drug is currently in a Phase III trial, the final stage before commercial approval.

Treatments with potential for all DEEs

BMB-101

Similar to Bexicaserin, BMB-101 acts on the 5-HT2C receptor. It is currently in a Phase 2 trial for DEE. It is important to note that seizure frequency is the primary endpoint in both studies of 5-HT2C agonists (BMB-101 and Bexicaserin). While there has been no convincing data that reduction in seizures impacts cognitive development in DEE, the only hint that a drug can improve cognition in epilepsy is fenfluramine(Soto-Insuga et al., 2025), which as a serotonin releasing agent will act on all serotonin receptors. Therefore, it is possible that these better tolerated serotonin agonists (selective for the 5-HT2C receptor) could have a similar benefit, but it may be difficult for the researchers to tell if such an effect exists without an appropriate outcome measure (see next section).

Relutrigine

Praxis Precision Medicine has developed a sodium channel blocker that has shown efficacy in animal models of non-sodium channel epilepsies. The company demonstrated efficacy in SCN2A/SCN8A patients and is expanding the studies to include all DEE patients. While the company states that "all genetically driven DEEs result in hyperactivation of sodium channels" it would be important to test the compound in a model of DEE65 to ensure that seizures in those models are also reduced.

Therapeutic Development

Ravicti

The Amgen drug Ravicti, or glycerol phenylbutyrate, is used to control ammonia levels in urea cycle disorders. The drug showed anti-seizure activity in models of SLC6A1 and STXBP1 epilepsy. Researchers at Weill Cornell Medicine are conducting a study of the drug in all DEEs following an initial arm with only SLC6A1 and STXBP1patients(Stone et al., 2025).

Epidiolex/Cannabidiol/CBD oil

The FDA approved Epidiolex to treat two forms of epilepsy, Dravet Syndrome and Lennox-Gastaut syndrome, in 2018. Both are forms of DEE. "The currently available data suggest that response to treatment with a highly purified, plant-derived CBD oil-based solution can be seen in patients across a broad range of epilepsy disorders and etiologies." (Lattanzi et al., 2021) A report of the successful treatment of a single DEE65 patient with cannabidiol (de Góes et al., 2022) does not contain any information about the dose, schedule or source of the drug, so it is difficult to assess the effectiveness of this treatment.

Ketogenic or modified Atkins diet

Some forms of DEE, including Dravet Syndrome, respond well to a ketogenic diet (Sharma & Tripathi, 2013) . However, some subtypes of DEE are not amenable to this treatment, and the diet can actually make patients worse (Ko et al., 2018). Since there is no reported efficacy of this treatment in the literature it is best (as with any intervention) to proceed with caution.

Clinical Trials for DEEs

As reported above, parents of DEE65 patients expressed that while seizure control is important, developmental delay is as important if not more so when considering a therapy's effectiveness. Developing a therapeutic intervention that treats these non-seizure symptoms will require a way to measure improvement in the patient. These outcome measures, whether used in natural history or interventional clinical trials, must be:

1) Relevant to the patient's quality of life and well being

2) Applied consistently by different clinical evaluators and at different locations

3) Responsive to change, either improvement or worsening

Other than Stoke Therapeutics/Biogen's Phase 3 treatment for Dravet syndrome, zorevunersen, clinical trials studying disease modifying treatments for DEE are still in their early stages. There is still time to prepare for clinical trials for disease modifying treatments in a way that will ensure effective drugs reach the patients who need them.

Clinical Trials for DEEs

Even though many drugs have been investigated and approved that reduce seizure burden in patients with different forms of epilepsy, measuring whether a drug reduces seizure burden is still not as simple or reliable as investigators would like to rigorously prove that a drug is effective. Seizures often happen at home, can occur at any time of day or night, and even the most attentive and observant caregivers cannot always determine what kind of seizure the patient is experiencing, when it started, or what triggered it. Despite this limitation, most treatments that are investigated for DEE will use a seizure burden count as a primary outcome measure.

Giuseppe and his family, Italy

The Inchstone Project

Because precision therapies are expected to not only reduce seizure frequency and intensity, but also improve brain development and intellectual disability, outcome measures in the clinical trials must be sensitive to the expected change. The Developmental Epileptic Encephalopathy Project (DEE-P) has initiated a program to develop those outcome measures, called the Inchstone Project (because it is focused on smaller improvements than typical milestone measurements). The project includes adapting methods of measuring skills to visually impaired children, determining what a meaningful change in motor function represents to parents of DEE children, and developing new outcome measures appropriate to patients who might perform on the "floor" of the standard Bayley or Vineland development scales(Hecker et al., 2024). Measuring any improvement that a treatment brings about will be difficult and will require new tools to accurately capture subtle gains in patients who start with profound impairment.

The Inchstone Project aims to accelerate outcome measures development to the point of FDA approval for DEE patients. Such outcome measures must be more sensitive to incremental improvements than the tools that have been used for less severe developmental delays. The project assembles behavioral domains and the smallest improvement that caregivers would consider important and identifies patterns in the data(Downs et al., 2025). This is a first step in the development and selection of clinical outcomes assessments that could be used in clinical trials for DEEs as new treatments are tested.

What can Cyfip2 Network and their allies do to help?

Purpose	Project	Location/ Investigator	Cost
Optimizing the effectiveness of existing seizure treatments	Collect treatment results from patients and form consensus opinion	All DEE65 cllinicians	
Compare EEGs from DEE65 patients with other disorders	EEG Bank	Combined Brain, all DEE65 clinicians	
Clinical trial readiness	Inchstone Project www.inchstone project.org	All DEE65 caregivers and clinicans	

Conclusions

Compared to the more common forms of DEE (Dravet syndrome/SCN1A, SCN2A CDKL5) the development of treatments for DEE65 is at an early stage. However, lessons learned from the treatment of those other DEEs will make DEE65 treatments move from scientific discovery to clinical studies to patient care more quickly. The study of Cyfip2 function in contexts other than DEE65 will also drive research forward, although the applicability of the basic science results may not be immediately apparent. Patient caregivers can accelerate this process directly by donating biosamples (for creating cell-based disease models), participating in natural history studies (to better predict the clinical course, producing data that can act as a virtual placebo group for small studies) and by supporting the earliest, risky stages of research. Organizations like the Cyfip2 Network also play an important role in scientific research by bringing together investigators from very different worlds who can learn from one another. In March of 2025 the Cyfip2 Network hosted a virtual meeting where scientists studying mouse cocaine seeking behavior, zebrafish noise responses, actin filament organization and epileptic encephalopathy exchanged ideas and initiated collaborations. This role as network hub is an invaluable component of the process of scientific discovery.

Glossary

Actin – a protein that forms filaments or cables inside cells

Agonist – a substance that activates a receptor to transmit a signal into a cell

Allele – one copy of a gene (usually used in the context where a patient has two different alleles)

Antisense oligonucleotide (ASO) – a string of nucleotides that complements a sequence of RNA or DNA and therefore attaches itself to the RNA or DNA at a specific site

Arginine – one of the 20 amino acids and a frequent site of pathogenic variants

Axon – a long projection that leads from the body of a neuron to another cell

Cell line – a modified cell from a patient or an animal that lives in an incubator and can be multiplied several times to provide research material for several experiments. Cell lines can also be frozen and transferred from one lab to another

Conditional mouse model – a genetically modified mouse that allows researchers to control where and when a pathogenic variant is expressed

Cytoskeleton – the network of filaments, including actin filaments, that give a cell rigidity and allow it to move

De novo – a pathogenic variant that is not inherited from either parent

Denature – a process in which the 3 dimensional shape of a protein is altered so that it no longer performs its normal function. The protein usually becomes insoluble. The albumin in egg white is denatured when the egg is cooked

Dendrite – one of many short projections that lead from the body of a neuron (usually one neuron's axon connects with another neuron's dendrite)

Dominant – a genetic condition in which one allele of a gene controls the phenotype of the organism no matter what the other allele is (such as brown eyes)

Dorsal ruffle – a ring of actin filaments that is found on top of many cultured fibroblasts

Fibroblast – a type of cell that is found in many parts of the body and is easy to use to create a cell line

Gain of function – a change that gives a protein a new activity (in the context of genetic disease the new activity is usually deleterious to health)

Gene replacement therapy – a treatment that delivers a new, healthy copy of a gene to certain tissues in a patient

Genomic edit – a method of changing the DNA sequence in a living organism, either to create a disease model or to cure a disease in a patient

Genotype/phenotype correlation – a comparison of the pathogenic variants (gene sequence) with physical characteristics such as disease signs and symptoms

Hypotonia – loss of muscle strength and control

In silico – an experiment that takes place in a computer using data supplied by a researcher

Interventional clinical trial – a clinical study in which a treatment is given to the patient to determine if the treatment is effective

Knockout – removing a gene from DNA to create a cell-based or animal model

Loss of function – a change that removes the normal function of a protein (in the context of genetic disease the loss is deleterious to health)

Messenger RNA or mRNA – an oligonucleotide produced by a cell that holds the instructions for making a protein

Molecular dynamics – the rapid movement of parts of a molecule in relation to other parts

Natural history study – a clinical study in which patient data is collected over time with no treatment (or with a consistent standard of care treatment) to collect information about how a disease usually progresses

Neutrophil – a blood cell involved in fighting off certain kinds of infection

Non-selective – a substance that affects many different biological targets

Nucleotide – a unit of DNA or RNA that makes up the genetic code (A, T, G or C in DNA)

Null variant – a genetic variant that results in no functional protein (similar to loss of function)

Outcome measure – a measurement used in a clinical trial to determine disease severity, progression, or response to a treatment

Pathogenic variant – an alternative sequence of a gene that results in a genetic disease

Phase 1 clinical trial – a clinical study that determines how much drug is safe to give to patients (sometimes performed in healthy subjects)

Phase 2 clinical trial – a clinical study that determines what dose of drug is effective in patients

Phase 3 clinical trial – a clinical study that determines whether a drug is eligible for commercial distribution

Repurposed drug – a drug that has been approved for commercial distribution for one disease that is effective at treating another disease

Serotonin receptor – a family of proteins that detect the neurotransmitter serotonin and transmit a signal into a cell. These receptors are responsible for the activity of many psychoactive substances including treatments for mental disorders.

Sodium channel blocker – a class of substances that reduce the flow of sodium through a cell membrane. This class includes natural toxin, pain medications, heart arrythmia drugs as well as anti-convulsants.

Thermal stability- the ability of a protein to withstand denaturation in response to heat. This property is usually changed when a substance binds to the protein, so thermal stability is a relatively simple way of screening compounds for a specific biological activity.

Ultra-rare disease – a disease that affects fewer than 1 in 50,000 people

Whole exome sequencing – a method of evaluating all of the expressed genes in a patient to find potential pathogenic variants. This method only sequences about 2% of the genome and cannot evaluate variants that are outside of the expressed genes, which may have effects on health.

Whole genome sequencing – a method of evaluating all of a patient's DNA to find pathogenic variants. This method is more expensive than whole exome sequencing and the results are more difficult to interpret.

Wild type – a gene sequence that is free of deleterious variants, or an animal that has no pathogenic variants in any gene. This definition is never absolute, since any change in gene sequence may have an affect on animal health under certain conditions. For example, both the C57Bl/6J and C57Bl/6N mouse strains are considered wild type, but the C57Bl/6N has a variant in Cyfip2 that affects reward seeking behavior.

Timeline of Cyfip2 Research

The simplest way to view these publications is to visit Pubmed and enter the PMID in the search bar.

1999

Saller E, Tom E, Brunori M, Otter M, Estreicher A, Mack DH, Iggo R.
Increased apoptosis induction by 121F mutant p53.
EMBO J. 1999 Aug 16;18(16):4424-37.
PMID: 10449408

Cyfip2 was first identified in a screen of genes that are affected by a mutant transcription factor called p53. The authors called the gene 121F-specific p53 inducible RNA or PIR121. They did not know the function of the gene

2000

Spranger S, Rommel B, Jauch A, Bodammer R, Mehl B, Bullerdiek J.
Interstitial deletion of 5q33.3q35.1 in a girl with mild mental retardation.
Am J Med Genet. 2000 Jul 17;93(2):107-9.
PMID: 10869111.

This is a case report that identifies a possible cause of mental retardation in a 4 year old girl, with psychomotor delay and seizures. The region of chromosome 5 that is deleted contains Cyfip2, but there is no proof that Cyfip2 alteration is the cause of the girl's symptoms.

2001

Schenck A, Bardoni B, Moro A, Bagni C, Mandel JL.
A highly conserved protein family interacting with the fragile X mental retardation protein (FMRP) and displaying selective interactions with FMRP-related proteins FXR1P and FXR2P.
Proc Natl Acad Sci U S A. 2001 Jul 17;98(15):8844-9.
PMID: 11438699

The first scientific report that used the name Cyfip2, this research study started out as a search for proteins associated with the Fragile X Mental Retardation Protein (FMRP). They did not assign a function for Cyfip2 in this study, but they did find it in neurons, and specifically near synapses, that connect nerves together. FMRP controls messenger RNA that provides the instructions for making proteins in neurons.

2003

Schenck A, Bardoni B, Langmann C, Harden N, Mandel JL, Giangrande A.
CYFIP/Sra-1 controls neuronal connectivity in Drosophila and links the Rac1 GTPase pathway to the fragile X protein.
Neuron. 2003 Jun 19;38(6):887-98.
PMID: 12818175.

This follow up to the 2001 paper from the same group now identifies a function for the Cyfip proteins. Fruit flies (Drosophila) have only one form of Cyfip. Removing this gene from the flies results in defects in neuron structures called axons and connections between neurons called synapses. They did not examine the behavior of the flies.

2010

Chen Z, Borek D, Padrick SB, Gomez TS, Metlagel Z, Ismail AM, Umetani J, Billadeau DD, Otwinowski Z, Rosen MK. Structure and control of the actin regulatory WAVE complex. Nature. 2010 Nov 25;468(7323):533-8.
PMID: 21107423

The actin cytoskeleton is a complex of protein cables that define the cell's shape. The cables are constantly changing with the cell's needs, and this process must be carefully controlled. A complex called the WAVE regulatory complex (WRC) is one controller of the actin cytoskeleton. This paper showed the way that Cyfip1 (in this paper called Sra1) fits into the WRC. Cyfip2, which is very similar to Cyfip1, also fits into the WRC in the same way, and plays the same role in communicating from signal inputs to cytoskeletal outputs.

Pittman AJ, Gaynes JA, Chien CB.
nev (cyfip2) is required for retinal lamination and axon guidance in the zebrafish retinotectal system.
Dev Biol. 2010 Aug 15;344(2):784-94.
PMID: 20537992

Scientists studying development of the nervous system like to use zebrafish because like humans they are vertebrates, but unlike humans they are transparent during development. This study used a mutant zebrafish called nevermind in which neurons went astray on the way to their target organs. They discovered that the mutant fish lacked Cyfip2, and hypothesized that either the actin cytoskeleton controlling actions or the RNA binding actions of Cyfip2 were necessary for neurons to find their way.

2013

Kumar V, Kim K, Joseph C, Kourrich S, Yoo SH, Huang HC, Vitaterna MH, de Villena FP, Churchill G, Bonci A, Takahashi JS.
C57BL/6N mutation in cytoplasmic FMRP interacting protein 2 regulates cocaine response.
Science. 2013 Dec 20;342(6165):1508-12.
PMID: 24357318

Studying two closely related strains of laboratory mice, these scientists found that one strain responded to repeated cocaine administration differently than the other. They searched for the difference between these two strains and found that the one that the strain that was less sensitive to cocaine had a variant form of Cyfip2 that decreased the stability of the protein. They hypothesized that changes in the connections between nerves in the brain were responsible for these differences.

2014

Abekhoukh S, Bardoni B.
CYFIP family proteins between autism and intellectual disability: links with Fragile X syndrome.
Front Cell Neurosci. 2014 Mar 27;8:81.
PMID: 24733999

This review article speculates that because Cyfip1 and Cyfip2 can interact with FMRP, they may also prove to be involved in intellectual disability or autism spectrum disorders. While there is no new data, the hypothesis is remarkably prescient.

2015

Han K, Chen H, Gennarino VA, Richman R, Lu HC, Zoghbi HY.
Fragile X-like behaviors and abnormal cortical dendritic spines in cytoplasmic FMR1-interacting protein 2-mutant mice.
Hum Mol Genet. 2015 Apr 1;24(7):1813-23.
PMID: 25432536

Interested in studying the partners of FMRP, these authors generated Cyfip2 knockout mice. The mice lacking both copies of Cyfip2 did not live long past birth, but the mice lacking one copy (and retaining one copy) showed hyperactivity, similar to mice lacking FMRP. This data is strongly suggestive that Cyfip2 activity is important for FMRP function. [It is important to note that a knockout (complete loss of the protein) may be very different from expression of a protein with a variant that disrupts the activity.]

2018

Marsden KC, Jain RA, Wolman MA, Echeverry FA, Nelson JC, Hayer KE, Miltenberg B, Pereda AE, Granato M.
A Cyfip2-Dependent Excitatory Interneuron Pathway Establishes the Innate Startle Threshold.
Cell Rep. 2018 Apr 17;23(3):878-887.
PMID: 29669291

Searching for genes that affect the responses of zebrafish larvae to loud noises, these authors introduced random mutations over the entire fish genome. The mutations that affected the "startle response" but did not affect the fish's ability to hear the noise or move in response included a change in Cyfip2 that is expected to remove the entire function of the protein. Restoring the full gene later in the fish's developmental program restored the startle response to normal levels. The role of Cyfip2 in this biological pathway is likely due to the actin cytoskeleton remodeling function.

2018

Nakashima M, Kato M, Aoto K, Shiina M, Belal H, Mukaida S, Kumada S, Sato A, Zerem A, Lerman-Sagie T, Lev D, Leong HY, Tsurusaki Y, Mizuguchi T, Miyatake S, Miyake N, Ogata K, Saitsu H, Matsumoto N.
De novo hotspot variants in CYFIP2 cause early-onset epileptic encephalopathy.
Ann Neurol. 2018 Apr;83(4):794-806.
PMID: 29534297.

Major breakthrough:

An investigation into the genetic causes of epileptic encephalopathy in 699 patients revealed Arginine 87 variants in Cyfip2 in 4 individuals. Arginine is particularly susceptible to spontaneous mutation because the DNA code that calls for arginine is vulnerable to chemical alteration. Arginine 87 is predicted to stabilize the interaction of Cyfip2 with WAVE, resulting in a WAVE activity that is always in the "on" position regardless of cell requirements for actin fiber growth. Indeed, when the variant Cyfip2 was added to cells, runaway actin fibers resulted. This key paper not only describes the syndrome later named DEE65 for the first time, it suggests the mechanism by which variant Cyfip2 leads to consequences in neurons.

2018

Cioni JM, Wong HH, Bressan D, Kodama L, Harris WA, Holt CE.
Axon-Axon Interactions Regulate Topographic Optic Tract Sorting via CYFIP2-Dependent WAVE Complex Function.
Neuron. 2018 Mar 7;97(5):1078-1093.e6.
PMID: 29518358

In this technological tour de force paper, the authors use gene knockout and replacement methods in fish and frogs to show that Cyfip2 plays an important role in sorting axons as the nervous system develops. They go on to demonstrate that of the two distinct roles Cyfip2 has been assigned, its interaction with the WRC and subsequent effects on the actin cytoskeleton are crucial to the function of axon sorting, while RNA binding is not required.

Peng J, Wang Y, He F, Chen C, Wu LW, Yang LF, Ma YP, Zhang W, Shi ZQ, Chen C, Xia K, Guo H, Yin F, Pang N.
Novel West syndrome candidate genes in a Chinese cohort.
CNS Neurosci Ther. 2018 Dec;24(12):1196-1206.
PMID: 29667327

An analysis of 72 Chinese patient with West syndrome (infantile epilepsy with delayed development) revealed 17 candidate genes that likely cause the disease, including an arginine to cysteine variant at position 87 of Cyfip2.

2019

Zhang Y, Kang HR, Han K.
Differential cell-type-expression of CYFIP1 and CYFIP2 in the adult mouse hippocampus.
Anim Cells Syst (Seoul). 2019 Nov 24;23(6):380-383.
PMID: 31853374

Despite their similarity, it is clear that Cyfip1 and Cyfip2 play different roles in biology. Here the authors show that the two proteins are located in different cell types in the brain, with Cyfip1 found in neurons and astrocytes in discrete regions of the brain, while Cyfip2 is expressed all over the brain but only in neurons.

Zhong M, Liao S, Li T, Wu P, Wang Y, Wu F, Li X, Hong S, Yan L, Jiang L.
Early diagnosis improving the outcome of an infant with epileptic encephalopathy with cytoplasmic FMRP interacting protein 2 mutation: Case report and literature review.
Medicine (Baltimore). 2019 Nov;98(44):e17749.
PMID: 31689829

An infant girl in China showing symptoms of West syndrome was diagnosed with DEE65 following whole exome sequencing. She had a R87L mutation in Cyfip2.

2019

Zhang Y, Kang H, Lee Y, Kim Y, Lee B, Kim JY, Jin C, Kim S, Kim H, Han K.
Smaller Body Size, Early Postnatal Lethality, and Cortical Extracellular Matrix-Related Gene Expression Changes of Cyfip2-Null Embryonic Mice.
Front Mol Neurosci. 2019 Jan 4;11:482.
PMID: 30687000

We know from previous reports that mice completely lacking Cyfip2 expression do not survive. This study attempts to determine why the mice die shortly after birth. However, the brains of the Cyfip2 lacking mice appear normal using the methods these authors employ, leaving the question unanswered for now.

2019

Zweier M, Begemann A, ..., Rauch A.
Spatially clustering de novo variants in CYFIP2, encoding the cytoplasmic FMRP interacting protein 2, cause intellectual disability and seizures.
Eur J Hum Genet. 2019 May;27(5):747-759.
PMID: 30664714

Important milestone:

Rare disease research depends on collecting information from as many patients as possible in natural history studies. This first natural history study in DEE65 describes 12 patients with 8 different genetic variations. All of these variants except for one are located in parts of the protein that interact with WAVE and the authors speculate that all of the variants result in an "always on" state of WAVE actin polymerization. One of these variants results in a shortened protein, where the missing piece is not thought to contact WAVE but rather another member of the WRC, called NCKAP1. Most of these patients displayed seizures and all were reported to show a developmental delay, in most cases severe. It is important to note that up to this publication all DEE65-causing variants in Cyfip2 produce a protein, meaning the disease is caused by an altered function of Cyfip2, not a reduced amount of the protein. This is in contrast to the mouse models that were described above, in which the mouse cells produce half the normal amount of protein. It is not unusual that two different diseases can result from altered function and complete loss of a protein, and this may be the case for Cyfip2 based on the evidence so far.

2020

Arisaka A, Nakashima M, Kumada S, Inoue K, Nishida H, Mashimo H, Kashii H, Kato M, Maruyama K, Okumura A, Saitsu H, Matsumoto N, Fukuda M.
Association of early-onset epileptic encephalopathy with involuntary movements - Case series and literature review.
Epilepsy Behav Rep. 2020 Dec 17;15:100417.
PMID: 33490948

Four patients with similar symptoms, one of which had a pathogenic variant in Cyfip2, are compared. A distinctive type of movement disorder, characterized by small, rapid, repetitive movements (choreiform movements) is described as a distinctive feature of DEE65.

Kim GH, Zhang Y, Kang HR, Lee SH, Shin J, Lee CH, Kang H, Ma R, Jin C, Kim Y, Kim SY, Kwon SK, Choi SY, Lee KJ, Han K.
Altered presynaptic function and number of mitochondria in the medial prefrontal cortex of adult Cyfip2 heterozygous mice.
Mol Brain. 2020 Sep 11;13(1):123.
PMID: 32917241

The proposed roles of Cyfip2 so far include the association with the RNA controlling protein FMRP and participation in the actin cytoskeleton remodeling WRC. This paper proposes another role, regulating the location of mitochondria in the neuron. Mitochondria are essential for many neuron functions and they need to be present at specific locations in the cell to carry out these tasks. Mice lacking one copy of Cyfip2 have fewer mitochondria in the extreme ends of the neurons, and Cyfip2 was found as a component of the mitochondria themselves.

2020

Lee Y, Zhang Y, Kang H, Bang G, Kim Y, Kang HR, Ma R, Jin C, Kim JY, Han K.
Epilepsy- and intellectual disability-associated CYFIP2 interacts with both actin regulators and RNA-binding proteins in the neonatal mouse forebrain.
Biochem Biophys Res Commun. 2020 Aug 13;529(1):1-6.
PMID: 32560809.

Up until this report the explanation of the cause of DEE65 had focused on the role of Cyfip2 in the WRC. However, Cyfip2 was first identified as a partner of the RNA controlling protein FMRP. In this paper the authors examined what proteins Cyfip2 sticks to inside cells. While they did find other members of the WRC, they also found other RNA binding proteins. Next they tested the effect of Cyfip2 expression on the formation of RNA/protein structures called stress granules. While they found wild type Cyfip2 as a component of stress granules, the pathogenic variants R87C, R87P and R87L not only do not associate with stress granules, their presence prevents the stress granules from forming. This may explain how the R87 variants lead to a more severe disease than the variants found on other parts of Cyfip2.

2020

Ghosh A, Mizuno K, Tiwari SS, Proitsi P, Gomez Perez-Nievas B, Glennon E, Martinez-Nunez RT, Giese KP.
Alzheimer's disease-related dysregulation of mRNA translation causes key pathological features with ageing.
Transl Psychiatry. 2020 Jun 16;10(1):192.
PMID: 32546772

This article proposes a potential deleterious effect of eliminating one copy of Cyfip2, since the mice with one copy remaining developed pathology similar to Alzheimer's Disease. However, it's important to note the detail that the mouse strain they used to create the mice was the C57BL/6N, which has a variant in Cyfip2 causing the protein to be less stable. These results shouldn't be directly compared to the previous characterization of Cyfip2 heterozygous knockouts, which used the C57BL/6J strain.

Schaks M, Reinke M, Witke W, Rottner K.
Molecular Dissection of Neurodevelopmental Disorder-Causing Mutations in CYFIP2.
Cells. 2020 May 29;9(6):1355
PMID: 32486060

Although researchers had already guessed that the DEE65 causing variants in Cyfip2 were turning WAVE on inappropriately, it had never been directly tested. In this paper the authors used a cell assay called lamellipodia formation to show that while wild type Cyfip2 could not bypass the "on switch" for actin polymerization, the DEE65 causing variants could. The truncating mutant describe in the natural history study did not have the same activity, suggesting that it works to cause intellectual disability in a different way. Another important accomplishment in this paper is the development of a cell-based assay for identifying potential therapeutics. While the measurement of lamellipodia in cells is not easy, it may form the basis for a high throughput screen with some modifications.

2021

Begemann A, Sticht H, Begtrup A, Vitobello A, Faivre L, Banka S, Alhaddad B, Asadollahi R, Becker J, Bierhals T, Brown KE, Bruel AL, Brunet T, Carneiro M, Cremer K, Day R, Denommé-Pichon AS, Dyment DA, Engels H, Fisher R, Goh ES, Hajianpour MJ, Haertel LRM, Hauer N, Hempel M, Herget T, Johannsen J, Kraus C, Le Guyader G, Lesca G, Mau-Them FT, McDermott JH, McWalter K, Meyer P, Õunap K, Popp B, Reimand T, Riedhammer KM, Russo M, Sadleir LG, Saenz M, Schiff M, Schuler E, Syrbe S, Van der Ven AT, Verloes A, Willems M, Zweier C, Steindl K, Zweier M, Rauch A.
New insights into the clinical and molecular spectrum of the novel CYFIP2-related neurodevelopmental disorder and impairment of the WRC-mediated actin dynamics.
Genet Med. 2021 Mar;23(3):543-554.
PMID: 33149277

Demonstrating the power of patient/caregiver engagement in the scientific process, this comprehensive natural history study represents a significant follow up to the 2018 report, with an additional 16 patients expressing missense variants and 3 with suspected loss of function. 11 new variants were described as well as three more Arg87Cys patients. All patients showed developmental delay, half with seizures. The three patients with loss of function variants had a less severe presentation and it is possible that the Cyfip2 variants did not cause their symptoms. The patient fibroblasts were cultured and the actin cytoskeleton of the cells was examined. The patient cells displayed a reduced presence of a structure called a dorsal ruffle.

2022

Biembengut ÍV, Shigunov P, Frota NF, Lourenzoni MR, de Souza TACB.
Molecular Dynamics of CYFIP2 Protein and Its R87C Variant Related to Early Infantile Epileptic Encephalopathy.
Int J Mol Sci. 2022 Aug 5;23(15):8708.
PMID: 35955843

An x-ray crystallography study established the structure of the WRC that included Cyfip1. The similarity between Cyfip1 and Cyfip2 is sufficient for these authors to create a simulated computer model of the WRC containing wild type and DEE65 causing variants of Cyfip2. According to these simulations, replacing Arginine 87 with cysteine results in a destabilization of an important substructure of Cyfip2.

Chaya T, Ishikane H, Varner LR, Sugita Y, Maeda Y, Tsutsumi R, Motooka D, Okuzaki D, Furukawa T.
Deficiency of the neurodevelopmental disorder-associated gene Cyfip2 alters the retinal ganglion cell properties and visual acuity.
Hum Mol Genet. 2022 Feb 21;31(4):535-547.
PMID: 34508581

Previous natural history studies have shown that over half of DEE65 patients exhibit visual problems. Using a mouse strain that cannot express Cyfip2 in the retina (conditional knock out mice) these authors show that while the retinas look normal and respond the same to light whether they express Cyfip2 or not, the mice without Cyfip2 in their retinas do have some changes in gene expression in the retina and have some differences in their ability to track movement with their eyes.

2022

Panthi S, szyszka P, beck CW
expression of mRNA encoding two gain-of-function cyfip2 variants associated with Dee65 results in spontaneous seizures in xenopus laevis tadpoles
biorxiv preprint, 2022
https://www.biorxiv.org/content/10.1101/2022.12.07.519540v1.full.pdf

This report describes a potentially valuable vertebrate model of DEE65. African clawed frogs (Xenopus laevis) are an important research tool. These researchers injected frog embryos with messenger RNA encoding either wild type or two different pathogenic variants of Cyfip2. The tadpoles hatched from eggs given the pathogenic variant displayed spontaneous seizure activity.

Limaye AJ, Bendzunas GN, Whittaker MK, LeClair TJ, Helton LG, Kennedy EJ.
In Silico Optimized Stapled Peptides Targeting WASF3 in Breast Cancer.
ACS Med Chem Lett. 2022 Mar 8;13(4):570-576.
PMID: 35450347

Cyfip2 was first identified as a protein associated with cancer progression. These researchers developed a drug that binds to Cyfip2 and disrupts its function in the WRC. This approach could have therapeutic benefit in DEE65 patients.

2023

Salokivi T, Parkkola R, Rajendran Y, Bharadwaj T, Acharya A, Leal SM, Järvelä I, Arvio M, Schrauwen I.
A novel variant in CYFIP2 in a girl with severe disabilities and bilateral perisylvian polymicrogyria.
Am J Med Genet A. 2024 Apr;194(4):e63478.
PMID: 37975178

This report describes a girl with a novel pathogenic variant in Cyfip2, Valine 551 Leucine that causes a distinct but overlapping set of symptoms as the DEE65 variants describe to date.

Silva ILZ, Gomes-Júnior R, da Silva EB, Vaz IM, Jamur VR, de Freitas Souza BS, Shigunov P.
Generation of an induced pluripotent stem cell line from a patient with epileptic encephalopathy caused by the CYFIP2 R87C variant.
Hum Cell. 2023 Nov;36(6):2237-2246
PMID: 37646972.

Cell based models are an important tool in the study of any disease, but for rare diseases they are particularly important. In this report cells were collected from a urine sample of a patient, reprogrammed into induced pluripotent stem cells. These cells can then be used to examine the effect of this particular pathogenic variant (R87C) on the function of relevant cell types such as neurons.

2023

Ma R, Zhang Y, Li H, Kang HR, Kim Y, Han K.
Cell-autonomous reduction of CYFIP2 is insufficient to induce Alzheimer's disease-like pathologies in the hippocampal CA1 pyramidal neurons of aged mice.
Anim Cells Syst (Seoul). 2023 Mar 24;27(1):93-101.
PMID: 36999135

Mice lacking one copy of Cyfip2 from the point of conception go on to develop Alzheimer's disease like features as they age. This report shows that if Cyfip2 is completely removed after the mice are born, no such Alzheimer's disease pathologies result. This study suggests that Cyfip2 plays an important role during prenatal development but possibly a different role after birth.

Da Silva Cardoso J, Gomes R, Abreu M, Parente Freixo J, Falcão Reis C, Garrido C.
Clinical Role of Codon 87 of the CYFIP2 Gene in Early Infantile Epileptic Encephalopathy: A Clinical Case Description.
Cureus. 2023 Feb 22;15(2):e35323
PMID: 36968925

This case study reveals a new molecular cause of DEE65, a deletion of three amino acids, including R87. The presentation of the disease is similar to that of patients harboring a missense variant such as Arg87Cys.

2023

Kang M, Zhang Y, Kang HR, Kim S, Ma R, Yi Y, Lee S, Kim Y, Li H, Jin C, Lee D, Kim E, Han K.
CYFIP2 p.Arg87Cys Causes Neurological Defects and Degradation of CYFIP2.
Ann Neurol. 2023 Jan;93(1):155-163
PMID: 36251395.

Important Research Tool:

Development of treatments for disease depends on research models, ideally live animal models, and because our understanding of mouse genetics is far advanced compared to other mammals, scientists tend to generate mouse models first.

These authors use a genetic editing tool to change one copy of a mouse (they used the C57BL/6N strain) Cyfip2 gene to the Arg87Cys variant. The mice grew to adulthood but with reduced body weight and strength. They showed signs of neurological problems including autism-like behaviors, spontaneous spasms and greater sensitivity to the seizure indcing drug PTZ.

Decreased organization of the brain in the Arg87Cys mice and increased inflammation progressed with age.

Overall this animal model displays excellent correlation with human disease and would be a valuable research tool for the creation of new therapeutics. The mouse strain used has a less stable form of Cyfip2 (Ser968Phe), so it would be important to repeat this study using the C57BL/6J strain with the more stable form of the protein.

2023

Poke G, Stanley J, Scheffer IE, Sadleir LG.
Epidemiology of Developmental and Epileptic Encephalopathy and of Intellectual Disability and Epilepsy in Children.
Neurology. 2023 Mar 28;100(13):e1363-e1375
PMID: 36581463

While it is difficult to establish the prevalence of a rare disease, these authors attempt to calculate the frequency of DEE in a Scottish population.

2024

Deslauriers JC, Ghotkar RP, Russ LA, Jarman JA, Martin RM, Tippett RG, Sumathipala SH, Burton DF, Cole DC, Marsden KC.
Cyfip2 controls the acoustic startle threshold through FMRP, actin polymerization, and GABAB receptor function.
bioRxiv [Preprint]. 2024 Feb 5:2023.12.22.573054.
PMID: 38187577

Cyfip2 has two major functions in biology, participation in the WRC to control actin polymerization and binding to FMRP to suppress RNA translation. Up to this point the WRC activity of Cyfip2 has explained all of the known effects of Cyfip2 mutation or loss. In this report, which expands on an earlier study on startle response in zebrafish larvae, both biological activities come together for the first time. Cyfip2 mutants that are unable to bind the WRC regulator Rac1 and mutants that cannot bind FMPR are both unable to restore normal function to fish lacking Cyfip2. One function of Cyfip2 in this context is the activation of a neurotransmitter receptor called GABAB and when GABAB is activated by a drug called Baclofen the normal startle response is restored.

2024

Venturi Biembengut Í, de Castro Andreassa E, de Souza TACB.
Identification of CYFIP2 Arg87Cys Ligands via In Silico and In Vitro Approaches.
Biomedicines. 2024 Feb 21;12(3):479.
PMID: 38540093

Therapeutic development:

Up to this point all of the research on DEE65 has been aimed at understanding the biology behind the disease. The authors of this study looked for approved drugs that might stick to the Arg87Cys form of Cyfip2 and therefore alter its function.

Using a technique called molecular docking, the authors found compounds that they predicted would bind preferentially to Cyfip2 Arg87Cys over the wild type protein. Using a cell culture the 8 most promising compounds were found to bind Cyfip2 Arg87Cys
.
The next steps for testing these compounds as potential treatments involve showing that they prevent the pathogenic function of the protein, and have a positive effect on symptoms in a disease model such as the mouse.

As the first demonstration of a potential treatment for DEE65 this paper represents a transition of the research on this disease into a new phase.

2024

Xie S, Zuo K, De Rubeis S, Ruggerone P, Carloni P.
Molecular basis of the CYFIP2 and NCKAP1 autism-linked variants in the WAVE regulatory complex.
Protein Sci. 2025 Jan;34(1):e5238.
PMID: 39660913

Using a rigorous computational method the authors examine the effect of each DEE65-causing variant on Cyfip2 structure and interaction with other members of the WRC. They confirm that all of the variants that cause disease also disrupt the function of Cyfip2 in controlling WAVE function.

Hecker J, Conecker G, Chapman C, Hommer R, Ludwig NN, Sevinc G, Te S, Wojnaroski M, Downs J, Berg AT.
Patient-advocate-led global coalition adapting fit-for-purpose outcomes measures to assure meaningful inclusion of DEEs in clinical trials.
Ther Adv Rare Dis. 2024 Jun 22;18:26330040241249762
PMID: 38911512

Describes the Inchstone Project and the results of the work on developing new outcome measures for DEEs.

2025

Kim HG, Berdasco C, Nairn AC, Kim Y.
The WAVE complex in developmental and adulthood brain disorders.
Exp Mol Med. 2025 Feb;57(1):13-29.
PMID: 39774290

This review article outlines the parallels between disease causing variants in WAVE1, NCKAP1, Cyfip2 and other members of the WRC and related pathways. It also gives a background on the "upstream" pathways that regulate the WRC, some of which may represent promising therapeutic targets for DEE65.

Ma R, Kim US, Chung Y, Kang HR, Zhang Y, Han K.
Recent advances in CYFIP2-associated neurodevelopmental disorders: From human genetics to molecular mechanisms and mouse models.
Brain Dev. 2025 Feb;47(1):104302.
PMID: 39603202.

An excellent review of the recent literature that shows the differences between Cyfip2 complete knockout, partial knockout, and Arg87Cys replacement, and hypothesizes that different pathogenic variants could impact distinct Cyfip2 functions and disease symptoms.

References

Amato, M. E., Frías, M., Cerisola, A., Roldán, M., & Ortigoza-Escobar, J. D. (2025). Novel CYFIP2 Frameshift Variant Linked to Dyskinetic Crises: Functional Studies Show Impaired Cell Motility. Clinical Genetics, n/a(n/a). https://doi.org/https://doi.org/10.1111/cge.14774

Arisaka, A., Nakashima, M., Kumada, S., Inoue, K., Nishida, H., Mashimo, H., Kashii, H., Kato, M., Maruyama, K., Okumura, A., Saitsu, H., Matsumoto, N., & Fukuda, M. (2021). Association of early-onset epileptic encephalopathy with involuntary movements – Case series and literature review. Epilepsy and Behavior Reports, 15. https://doi.org/10.1016/j.ebr.2020.100417

Begemann, A., Sticht, H., Begtrup, A., Vitobello, A., Faivre, L., Banka, S., Alhaddad, B., Asadollahi, R., Becker, J., Bierhals, T., Brown, K. E., Bruel, A.-L., Brunet, T., Carneiro, M., Cremer, K., Day, R., Denommé-Pichon, A.-S., Dyment, D. A., Engels, H., … Rauch, A. (2021). New insights into the clinical and molecular spectrum of the novel CYFIP2-related neurodevelopmental disorder and impairment of the WRC-mediated actin dynamics. Genetics in Medicine, 3(23), 543–554. https://doi.org/10.1038/s41436

Biembengut, Í. V., Shigunov, P., Frota, N. F., Lourenzoni, M. R., & de Souza, T. A. C. B. (2022). Molecular Dynamics of CYFIP2 Protein and Its R87C Variant Related to Early Infantile Epileptic Encephalopathy. International Journal of Molecular Sciences, 23(15). https://doi.org/10.3390/ijms23158708

Chan, R., Orevillo, C., O'Connell, G., McLin, D., Polega, S., Srinivas, N. R., & Kaye, R. (2025). Safety, Tolerability, Pharmacokinetics, Pharmacodynamics, and Food Effect of Bexicaserin in Healthy Participants: A First-in-Human Randomized, Double-Blind, Placebo-Controlled Single Ascending Dose Escalation Phase 1 Study. Clinical Pharmacology in Drug Development. https://doi.org/10.1002/cpdd.1600

Chen, Z., Borek, D., Padrick, S. B., Gomez, T. S., Metlagel, Z., Ismail, A. M., Umetani, J., Billadeau, D. D., Otwinowski, Z., & Rosen, M. K. (2010). Structure and control of the actin regulatory WAVE complex. Nature, 468(7323), 533–538. https://doi.org/10.1038/nature09623

Cioni, J. M., Wong, H. H. W., Bressan, D., Kodama, L., Harris, W. A., & Holt, C. E. (2018). Axon-Axon Interactions Regulate Topographic Optic Tract Sorting via CYFIP2-Dependent WAVE Complex Function. Neuron, 97(5), 1078-1093.e6. https://doi.org/10.1016/j.neuron.2018.01.027

Conroy, F., Miller, R., Alterman, J. F., Hassler, M. R., Echeverria, D., Godinho, B. M. D. C., Knox, E. G., Sapp, E., Sousa, J., Yamada, K., Mahmood, F., Boudi, A., Kegel-Gleason, K., DiFiglia, M., Aronin, N., Khvorova, A., & Pfister, E. L. (2022). Chemical engineering of therapeutic siRNAs for allele-specific gene silencing in Huntington's disease models. Nature Communications, 13(1). https://doi.org/10.1038/s41467-022-33061-x

Da Silva Cardoso, J., Gomes, R., Abreu, M., Parente Freixo, J., Falcão Reis, C., & Garrido, C. (2023). Clinical Role of Codon 87 of the CYFIP2 Gene in Early Infantile Epileptic Encephalopathy: A Clinical Case Description. Cureus. https://doi.org/10.7759/cureus.35323

de Góes, F. V., de Andrade Ramos, J. T. M., da Silva Fontana, R., de Carvalho Serão, C. L., Kok, F., & Gandelman Horovitz, D. D. (2022). Cannabidiol Successful Therapy for Developmental and Epileptic Encephalopathy Related to CYFIP2. The Open Neurology Journal, 16(1). https://doi.org/10.2174/1874205x-v16-e2203290

DeRubeis, S., Pasciuto, E., Li, K. W., Fernández, E., DiMarino, D., Buzzi, A., Ostroff, L. E., Klann, E., Zwartkruis, F. J. T., Komiyama, N. H., Grant, S. G. N., Poujol, C., Choquet, D., Achsel, T., Posthuma, D., Smit, A. B., & Bagni, C. (2013). CYFIP1 coordinates mRNA translation and cytoskeleton remodeling to ensure proper dendritic Spine formation. Neuron, 79(6), 1169–1182. https://doi.org/10.1016/j.neuron.2013.06.039

Downs, J., Haywood, S., Ludwig, N. N., Wojnaroski, M., Hommer, R., Muzyczka, K., Hecker, J. E., Conecker, G., Keeley, J., & Berg, A. T. (2025). Caregiver-reported meaningful change in functional domains for individuals with developmental and epileptic encephalopathy: A convergent mixed-methods design. Developmental Medicine and Child Neurology. https://doi.org/10.1111/dmcn.16363

Han, K., Chen, H., Gennarino, V. A., Richman, R., Lu, H. C., & Zoghbi, H. Y. (2014). Fragile X-like behaviors and abnormal cortical dendritic spines in Cytoplasmic FMR1-interacting protein 2-mutant mice. Human Molecular Genetics, 24(7), 1813–1823. https://doi.org/10.1093/hmg/ddu595

Hartmann, M. C., McCulley, W. D., Holbrook, S. E., Haney, M. M., Smith, C. G., Kumar, V., & Rosenwasser, A. M. (2023). Cyfip2 allelic variation in C57BL/6J and C57BL/6NJ mice alters free-choice ethanol drinking but not binge-like drinking or wheel-running activity. Alcohol: Clinical and Experimental Research, 47(8), 1518–1529. https://doi.org/10.1111/acer.15137

Hecker, J. E., Conecker, G., Chapman, C., Hommer, R., Ludwig, N. N., Sevinc, G., Te, S., Wojnaroski, M., Downs, J., & Berg, A. T. (2024). Patient-advocate-led global coalition adapting fit-for-purpose outcomes measures to assure meaningful inclusion of DEEs in clinical trials. In Therapeutic Advances in Rare Disease (Vol. 18). SAGE Publications Ltd. https://doi.org/10.1177/26330040241249762

Kang, M., Zhang, Y., Kang, H. R., Kim, S., Ma, R., Yi, Y., Lee, S., Kim, Y., Li, H., Jin, C., Lee, D., Kim, E., & Han, K. (2023). CYFIP2 p.Arg87Cys Causes Neurological Defects and Degradation of CYFIP2. Annals of Neurology, 93(1), 155–163. https://doi.org/https://doi.org/10.1002/ana.26535

Kim, G. H., Zhang, Y., Kang, H. R., Lee, S. H., Shin, J., Lee, C. H., Kang, H., Ma, R., Jin, C., Kim, Y., Kim, S. Y., Kwon, S. K., Choi, S. Y., Lee, K. J., & Han, K. (2020). Altered presynaptic function and number of mitochondria in the medial prefrontal cortex of adult Cyfip2 heterozygous mice. Molecular Brain, 13(1). https://doi.org/10.1186/s13041-020-00668-4

Kim, Y., Ma, R., Zhang, Y., Kang, H. R., Kim, U. S., & Han, K. (2024). Cell-autonomous reduction of CYFIP2 changes dendrite length, dendritic protrusion morphology, and inhibitory synapse density in the hippocampal CA1 pyramidal neurons of 17-month-old mice. Animal Cells and Systems, 28(1), 294–302. https://doi.org/10.1080/19768354.2024.2360740

Ko, A., Jung, D. E., Kim, S. H., Kang, H. C., Lee, J. S., Lee, S. T., Choi, J. R., & Kim, H. D. (2018). The efficacy of ketogenic diet for specific genetic mutation in developmental and epileptic encephalopathy. Frontiers in Neurology, 9(JUL). https://doi.org/10.3389/fneur.2018.00530

Kumar, V., Kim, K., Joseph, C., Kourrich, S., Yoo, S. H., Huang, H. C., Vitaterna, M. H., De Villena, F. P. M., Churchill, G., Bonci, A., & Takahashi, J. S. (2013). C57BL/6N mutation in cytoplasmic FMRP interacting protein 2 regulates cocaine response. Science, 342(6165), 1508–1512. https://doi.org/10.1126/science.1245503

Lattanzi, S., Trinka, E., Striano, P., Rocchi, C., Salvemini, S., Silvestrini, M., & Brigo, F. (2021). Highly Purified Cannabidiol for Epilepsy Treatment: A Systematic Review of Epileptic Conditions Beyond Dravet Syndrome and Lennox–Gastaut Syndrome. In CNS Drugs (Vol. 35, Issue 3, pp. 265–281). Adis. https://doi.org/10.1007/s40263-021-00807-y

Limaye, A. J., Bendzunas, G. N., Whittaker, M. K., Leclair, T. J., Helton, L. G., & Kennedy, E. J. (2022). In Silico Optimized Stapled Peptides Targeting WASF3 in Breast Cancer. ACS Medicinal Chemistry Letters, 13(4), 570–576. https://doi.org/10.1021/acsmedchemlett.1c00627

Marsden, K. C., Jain, R. A., Wolman, M. A., Echeverry, F. A., Nelson, J. C., Hayer, K. E., Miltenberg, B., Pereda, A. E., & Granato, M. (2018). A Cyfip2-Dependent Excitatory Interneuron Pathway Establishes the Innate Startle Threshold. Cell Reports, 23(3), 878–887. https://doi.org/10.1016/j.celrep.2018.03.095

Nakashima, M., Kato, M., Aoto, K., Shiina, M., Belal, H., Mukaida, S., Kumada, S., Sato, A., Zerem, A., Lerman-Sagie, T., Lev, D., Leong, H. Y., Tsurusaki, Y., Mizuguchi, T., Miyatake, S., Miyake, N., Ogata, K., Saitsu, H., & Matsumoto, N. (2018). De novo hotspot variants in CYFIP2 cause early-onset epileptic encephalopathy. Annals of Neurology, 83(4), 794–806. https://doi.org/10.1002/ana.25208

Panthi, S., Szyszka, P., & Beck, C. W. (2022). Expression of mRNA encoding two gain-of-function cyfip2 variants associated with DEE65 results in spontaneous seizures in Xenopus laevis tadpoles. https://doi.org/10.1101/2022.12.07.519540

Peng, J., Wang, Y., He, F., Chen, C., Wu, L. W., Yang, L. F., Ma, Y. P., Zhang, W., Shi, Z. Q., Chen, C., Xia, K., Guo, H., Yin, F., & Pang, N. (2018). Novel West syndrome candidate genes in a Chinese cohort. CNS Neuroscience and Therapeutics, 24(12), 1196–1206. https://doi.org/10.1111/cns.12860

Pittman, A. J., Gaynes, J. A., & Chien, C. Bin. (2010). Nev (cyfip2) is required for retinal lamination and axon guidance in the zebrafish retinotectal system. Developmental Biology, 344(2), 784–794. https://doi.org/10.1016/j.ydbio.2010.05.512

Poke, G., Stanley, J., Scheffer, I. E., & Sadleir, L. G. (2023). Epidemiology of Developmental and Epileptic Encephalopathy and of Intellectual Disability and Epilepsy in Children. Neurology, 100(13), E1363–E1375. https://doi.org/10.1212/WNL.0000000000206758

Saller, E., Tom, E., Brunori, M., Le Otter, M., Estreicher, A., Mack, D. H., & Iggo, R. (1999). Increased apoptosis induction by 121F mutant p53. In The EMBO Journal (Vol. 18, Issue 16).

Salokivi, T., Parkkola, R., Rajendran, Y., Bharadwaj, T., Acharya, A., Leal, S. M., Järvelä, I., Arvio, M., & Schrauwen, I. (2024). A novel variant in CYFIP2 in a girl with severe disabilities and bilateral perisylvian polymicrogyria. American Journal of Medical Genetics, Part A, 194(4). https://doi.org/10.1002/ajmg.a.63478

Samanta, D., Bhatia, S., Hunter, S. E., Rao, C. K., Xiong, K., Karakas, C., Reeders, P. C., Erdemir, G., Sattar, S., Axeen, E., Sandoval Karamian, A. G., Fine, A. L., Keator, C. G., Nolan, D., & Schreiber, J. M. (2025). Current and Emerging Precision Therapies for Developmental and Epileptic Encephalopathies. In Pediatric Neurology (Vol. 168, pp. 67–81). Elsevier Inc. https://doi.org/10.1016/j.pediatrneurol.2025.04.010

Scheffer, I. E., French, J., Valente, K. D., Auvin, S., Cross, J. H., & Specchio, N. (2025). Operational definition of developmental and epileptic encephalopathies to underpin the design of therapeutic trials. Epilepsia, 66(4), 1014–1023. https://doi.org/10.1111/epi.18265

Schenck, A., Bardoni, B., Langmann, C., Harden, N., Mandel, J.-L., & Giangrande, A. (2003). CYFIP/Sra-1 Controls Neuronal Connectivity in Drosophila and Links the Rac1 GTPase Pathway to the Fragile X Protein the postsynaptic site of most excitatory synapses in mammalian brains (for review see Luo, 2002). Fragile X syndrome, the most frequent cause of hered-itary mental retardation, is caused by the absence of the RNA binding protein FMRP (for review see Bardoni. In Neuron (Vol. 38).

Schenck, A., Bardoni, B., Moro, A., Bagni, C., & Mandel, J.-L. (2001). A highly conserved protein family interacting with the fragile X mental retardation protein (FMRP) and displaying selective interactions with FMRP-related proteins FXR1P and FXR2P. Procedings of the National Academy of Sciences USA, 15(98), 8844–8849. www.pnas.orgcgidoi10.1073pnas.151231598

Sharma, S., & Tripathi, M. (2013). Ketogenic Diet in Epileptic Encephalopathies. Epilepsy Research and Treatment, 2013, 1–5. https://doi.org/10.1155/2013/652052

Soto-Insuga, V., Conejo Moreno, D., González-Alguacil, E., Aledo Serrano, A., Navarro Abia, V., Gretel Pinzón-Acevedo, A., Lamagrande Casanova, N., Duat Rodríguez, A., Cantarín Extremera, V., & García Peñas, J. J. (2025). Fenfluramine: an effective treatment for developmental epileptic encephalopathies beyond Dravet and Lennox–Gastaut Syndromes. Journal of Neurology, 272(6), 397. https://doi.org/10.1007/s00415-025-13135-8

Stone, A., Burré, J., Wayland, N., & Grinspan, Z. M. (2025). Phenylbutyrate for monogenetic epilepsy: Literature review. Epilepsy Research, 217, 107621. https://doi.org/https://doi.org/10.1016/j.eplepsyres.2025.107621

Symonds, J. D., Elliott, K. S., Shetty, J., Armstrong, M., Brunklaus, A., Cutcutache, I., Diver, L. A., Dorris, L., Gardiner, S., Jollands, A., Joss, S., Kirkpatrick, M., McLellan, A., MacLeod, S., O'Regan, M., Page, M., Pilley, E., Pilz, D. T., Stephen, E., … Zuberi, S. M. (2021). Early childhood epilepsies: Epidemiology, classification, aetiology, and socio-economic determinants. Brain, 144(9), 2879–2891. https://doi.org/10.1093/brain/awab162

Tecott, L. H., Sun, L. M., Akana, S. F., Strack, A. M., Lowenstein, D. H., Dallman, M. F., & Julius, D. (1995). Eating disorder and epilepsy in mice lacking 5-HT2C serotonin receptors. Nature, 374(6522), 542–546. https://doi.org/10.1038/374542a0

Venturi Biembengut, Í., de Castro Andreassa, E., & de Souza, T. A. C. B. (2024). Identification of CYFIP2 Arg87Cys Ligands via In Silico and In Vitro Approaches. Biomedicines, 12(3). https://doi.org/10.3390/biomedicines12030479

Zhang, Y., Kang, H., Lee, Y., Kim, Y., Lee, B., Kim, J. Y., Jin, C., Kim, S., Kim, H., & Han, K. (2019). Smaller body size, early postnatal lethality, and cortical extracellular matrix-related gene expression changes of Cyfip2-null embryonic mice. Frontiers in Molecular Neuroscience, 11. https://doi.org/10.3389/fnmol.2018.00482

Zhang, Y., Kang Hyae, R., Lee, S. H., Kim, Y., Ma, R., Jin, C., Lim, J. E., Kim, S., Kang, Y., Kang, H., Kim, S. Y., Kwon, S. K., Choi, S. Y., & Han, K. (2020). Enhanced Prefrontal Neuronal Activity and Social Dominance Behavior in Postnatal Forebrain Excitatory Neuron-Specific Cyfip2 Knock-Out Mice. Frontiers in Molecular Neuroscience, 13. https://doi.org/10.3389/fnmol.2020.574947

Zhong, M., Liao, S., Li, T., Wu, P., Wang, Y., Wu, F., Li, X., Hong, S., Yan, L., & Jiang, L. (2019). Early diagnosis improving the outcome of an infant with epileptic encephalopathy with cytoplasmic FMRP interacting protein 2 mutation: Case report and literature review. Medicine, 98(44), e17749. https://doi.org/10.1097/MD.0000000000017749

Zweier, M., Begemann, A., McWalter, K., Cho, M. T., Abela, L., Banka, S., Behring, B., Berger, A., Brown, C. W., Carneiro, M., Chen, J., Cooper, G. M., Finnila, C. R., Guillen Sacoto, M. J., Henderson, A., Hüffmeier, U., Joset, P., Kerr, B., Lesca, G., … Rauch, A. (2019). Spatially clustering de novo variants in CYFIP2, encoding the cytoplasmic FMRP interacting protein 2, cause intellectual disability and seizures. European Journal of Human Genetics, 27(5), 747–759. https://doi.org/10.1038/s41431-018-0331-z