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GPR56: Its Regulation in GABAergic Neurons and Possible Involvement in Neuro Developmental Disorders

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Abstract

One-third of patients with epilepsy have intractable seizures that cannot be controlled with any currently available antiepileptic drugs. It has been well known that there are patients afflicted with epilepsy with mutations in some G protein-coupled receptors (GPCRs). Patients with the 15-bp deletion within the upstream region of non-cording exon 1m of human G Protein-Coupled Receptor 56 (*GPR56)* gene exhibit cortical malformation and develop young-onset epilepsy. In our recent report, to understanding the mechanism underlying the etiological role of this 15-bp deletion, we examined the function of a cis-regulatory element containing this 15-bp in the control of expression pattern of *GPR56* using marmoset as a nonhuman primate animal model. We showed that the cis-regulatory element drives expression of *GPR56* preferentially in GABAergic neurons in developing brain and suggest that pathogenic mechanisms of epilepsy in patients with a 15-bp deletion may be explained in part by the developmental abnormality or dysfunction of GABAergic neurons.

Keywords: Epilepsy; G Protein-Coupled Receptor (GPCR); G Protein-Coupled Receptor 56 (*GPR56*); GABAergic neuron

Introduction

Epilepsy is a chronic brain disease with recurrent seizures resulting from excessive firing of cerebral neurons. Epilepsy affects approximately 1% of the population, and they are estimated at more than 50 million cases worldwide [1]. In recent years, the focus of epilepsy treatment has shifted significantly from surgical treatments to non-invasive drug therapy. There are more than 20 types of antiepileptic drugs (AEDs), including channel blockers and γ-aminobutyric acid (GABA) transaminases. However, the mechanism of action of many AEDs are still not well known. Moreover, at least one-thirds of patients have intractable seizures that cannot be controlled with any of the AEDs currently available [2]. Therefore, it is desirable to find a molecular target to suppress the abnormal firing of neurons by an unprecedented mechanism.

Literature Review

GPR56 and patients with a 15-bp deletion within its cis-regulatory element

It has been well known that mutations in some G protein-coupled receptors (GPCRs) cause epilepsy [3]. G-protein-coupled receptor 56 (*GPR56*) is one such GPCRs, and patients with mutations in *GPR56* gene exhibit cortical malformation (a cobblestone-like lissencephaly) called bilateral frontoparietal polymicrogyria (BFPP) and develop young-onset epilepsy, as well as mental retardation and gait difficulty [4,5].

Previous studies have clearly demonstrated the crucial roles of *GPR56* in cortical development [6]. In the developing brain, neural stem cells proliferate in the ventricular zone that lies adjacent to the lateral ventricle and their daughter cells migrate toward the pial surface, developing into excitatory glutamatergic neurons. Loss of *Gpr56* in mice causes many cellular abnormalities in the cerebral cortex, including reduced proliferation of neuronal progenitor cells [7] and overmigration of developing neurons [6].Consequently, *Gpr56*-deficient mice exhibit disorganized cortical lamination and BFPP [6]. Although the function of *GPR56* in glutamatergic progenitors and oligodendrocyte precursor cells has been well studied [8], the role of *GPR56* in GABAergic neuronal lineage has not been addressed, which is important for understanding the mechanism underlying the pathogenesis of epilepsy.

In our recent report, we, for the first time, reported the predictable role of *GPR56* in GABAergic neuronal development [9]. The human *GPR56* gene consists of 14 exons, and has multiple presumptive promoters encompassing distinct non-cording first exon. The gene has 17 or more alternative transcription start sites that may promote transcription of mRNAs encoding identical *GPR56* protein with distinct expression profiles in the brain.

A 15-bp deletion within a cis-regulatory element upstream of one of the noncoding first exon, exon 1m (e1m), of *GPR56* has recently been identified in individuals with polymicrogyria restricted to the regions around the Sylvian fissure. Patients with this 15-bp deletion suffer from medically refractory seizures from a young age, as well as with intellectual disabilities but without evident motor disabilities [7].

Epilepsy caused by mutations in non-cording region

Until now, only a few genes have been found, in which mutations in non-cording regions cause epilepsy. As a well-known example, *SCN1A* gene produces voltage-gated sodium channel Nav1.1 that expresses in a GABAergic neuron [10-12]. Mutations in its promoter region cause *SCN1A* haploinsufficiency and reduce the amount of Nav1.1. As a consequence, the reduced sodium currents in GABAergic inhibitory neurons lead to the hypoexcitability of inhibitory neurons, initiating epilepsy [13]. An attractive hypothesis would be that, in the case of *GPR56* , the 15-bp deletion in the e1m cis-regulatory element produces intact GPR56 protein [7], but leads to inaccurate temporal and/or spatial expression of GPR56.

To clarify how the 15-bp deletion within the *hGPR56* e1m upstream region leads to the symptoms including epilepsy, we focused on the 0.3 kbp minimal promoter region containing this 15-bp (0.3 k *hGPR56* e1m) and examined the etiological role of this region by using common marmoset (*Callithrix jacchus*), a nonhuman primate animal model [9][14-16]. We generated transgenic marmoset in which EGFP is expressed under the control of *0.3 k hGPR56* e1m *(hGPR56 e1m-EGFP).* Using the technique of immunohistochemistry, we showed that *hGPR56* e1m-EGFP expression was mainly detected in developing neurons, while endogenous *GPR56* is expressed in the ventricular zone of the cerebral cortex as well as in developing neurons. Furthermore, while the total *GPR56* protein is evenly expressed in both GABAergic and glutamatergic neurons, *hGPR56* e1m-EGFP is preferentially expressed in GABAergic neurons in layer V in the cerebral cortex. The expression of e1m-driven EGFP is also detected in the developing ganglionic eminence (GE), a region where most inhibitory interneurons originate. Thus, the e1m cis-element may drive EGFP expression not only in mature GABAergic neurons but also in the progenitor cells of interneurons in the GE during development. These results support the idea that 15 bp deletion within e1m cis-element may have a significant effect on the expression pattern of *GPR56* in GABAergic neurons in human developing brain.

Epilepsy caused by the developmental abnormality of GABAergic neurons

Like *GPR56,* homeobox transcription factor DLX families and their target gene ARX mainly contribute to the migration of neuronal cells in developing neocortex. These genes are also known to be involved in differentiation of GABAergic inhibitory neurons at embryonic stages [17,18]. Dysfunction of GABAergic neuron development caused by mutations in *DLX* and *ARX* is frequently associated with epilepsy [17,18]. In humans, exon 1m of *GPR56* gene shows an order of higher expression in the fetal brain compared to the adult brain [7]. Moreover, our data suggest that e1m cis-element drives *GPR56* expression in GABAergic neurons irrespective of subtype. Therefore, *GPR56* driven by e1m cis-element may play a role in the earlier development of GABAergic neuron before its subtype determination [19]. Similar to the case for *DLX* and *ARX* mutations, our data may imply that pathogenic mechanisms of epilepsy in patients with a 15-bp deletion within e1m region may be explained in part by the developmental abnormality or dysfunction of GABAergic neurons.

Antiepileptic drug

Vigabatrin (γ-vinyl-GABA) is a structural analogue of GABA and thus acts as a GABA-transaminase inhibitor by reducing metabolism and degradation of GABA and increasing GABA concentrations. Interestingly, Vigabatrin is used as an antiepileptic drug that has been reported to relieve symptoms in some patients with mutations in *GPR56* gene [20]. This fact strongly supports our conclusion that a 15-bp deletion within a cis-regulatory element of *GPR56* may lead to developmental abnormality or dysfunction of GABAergic neurons, leading to the onset of epilepsy. Because the mechanism of action of Vigabatrin is known to be different from that of $GABA_AR$ activators such as benzodiazepines and barbiturates, more detailed studies are needed to develop GPCRs as antiepileptic and/or antiepileptogenic targets.

Discussion and Conclusion

The present study with transgenic marmoset showed that the e1m cis-element of human *GPR56* drives expression of *GPR56* preferentially in GABAergic neurons in developing brain. This fact strongly supports our conclusion that a 15-bp deletion within a cis-regulatory element of *GPR56* may leads to developmental abnormality or dysfunction of GABAergic neurons, initiating epilepsy. Recently, there are many GPCR-targeted drugs which are approved by the US Food and Drug Administration, but no AEDs are known to suppress epileptic seizures by acting directly on GPCRs so far. It is promising that our study may lead toward a breakthrough in developing a new AED targeting GPCR and achieving complete remission in refractory patients by an unprecedented.

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References

1. World Health Organization (2019) Epilepsy.
2. Sirven JI. (2021) Evaluation and management of drug-resistant epilepsy. In: UpToDate, Post TW (Ed), UpToDate. Waltham MA.
3. Yu Y, Nguyen DT, Jiang J (2019) G protein-coupled receptors in acquired epilepsy: Druggability and translatability. Progress in Neurobiology 183: 101682.
4. Piao X, Hill RS, Bodell A, Chang BS, Basel-Vanagaite L, et al. (2004) G protein-coupled receptor-dependent development of human frontal cortex. Science 303: 2033-2036.
5. Ganesha RA, Venkataraman K, Sirdeshmukh R (2019) GPR56: An adhesion GPCR involved in brain development, neurological disorders and cancer. Brain Research. 1747: 147055.
6. Li S, Jin Z, Koirala S, Bu L, Xu L, et al. (2008) GPR56 regulates pial basement membrane integrity and cortical lamination. J Neurosci 28:5817-5826.
7. Bae BI, Tietjen I, Atabay KD, Evrony GD, Johnson MB, et al. (2014) Evolutionarily dynamic alternative splicing of GPR56 regulates regional cerebral cortical patterning. Science 343: 764-768.
8. Langenhan T, PiaoX, Monk KR (2016) Adhesion G protein-coupled receptors in nervous system development and disease. Nat Neurosci 17:550-561.
9. Murayama AY, Kuwako K, Okahara J, Bae B, Okuno M, et al. (2020) The polymicrogyria-associated GPR56 promoter preferentially drives

gene expression in developing GABAergic neurons in common marmosets. Sci Rep 10: 21516.

10. Nakayama T, Ogiwara I, Ito K, Kaneda M, Mazaki E, et al. (2010) Deletions of SCN1A 5' genomic region with promoter activity in Dravet syndrome. Human mutation 31, 820-829.
11. Higurashi N, Uchida T, Lossin C, Misumi Y, Okada Y, et al. (2013) A human Dravet syndrome model from patient induced pluripotent stem cells. Mol Brain 6:19.
12. Gao QW, Hua LD, Wang J, Fan CX, Deng WY, et al. (2017) A Point Mutation in SCN1A 5' Genomic Region Decreases the Promoter Activity and Is Associated with Mild Epilepsy and Seizure Aggravation Induced by Antiepileptic Drug. Molecular neurobiology 54:2428-2434.
13. Holmes GL, Noebels JL. (2016) The Epilepsy Spectrum: Targeting Future Research Challenges. Cold Spring Harbor perspectives in medicine 6.
14. Miller CT, Freiwald WA, Leopold DA, Mitchell JF, Silva AC, et al. (2016) Marmosets: A Neuroscientific Model of Human Social Behavior. Neuron 90:219-233.
15. Okano H, Sasaki E, Yamamori T, Iriki A, Shimogori T, et al. (2016) Brain/MINDS: A Japanese National Brain Project for Marmoset Neuroscience. Neuron 92: 582-590.
16. Okano H (2021) Current Status of and Perspectives on the Application of Marmosets in Neurobiology. Annu Rev Neurosci 44:27-48.
17. Cobos I, Calcagnotto ME, Vilaythong AJ, Thwin MT, Noebels JL, et al. (2005) Mice lacking Dlx1 show subtype-specific loss of interneurons, reduced inhibition and epilepsy. Nat Neurosci 8: 1059-1068.
18. Marsh ED, Golden JA (2012) Developing Models of Aristaless-related homeobox mutations in Jasper's Basic Mechanisms of the Epilepsies. 4th edition.
19. Lim L, Mi D, Llorca A, Marin O (2018) Development and functional diversification of cortical interneurons. Neuron 100: 294-313.
20. Bellusci M, Trivisano M, de Palma L, Pietrafusa N, Vigevano F, et al. (2016) Vigabatrin efficacy in GPR56-associated polymicrogyria: The role of GABAA receptor pathway. Epilepsia 57: 1337-1338.