

Figure 3

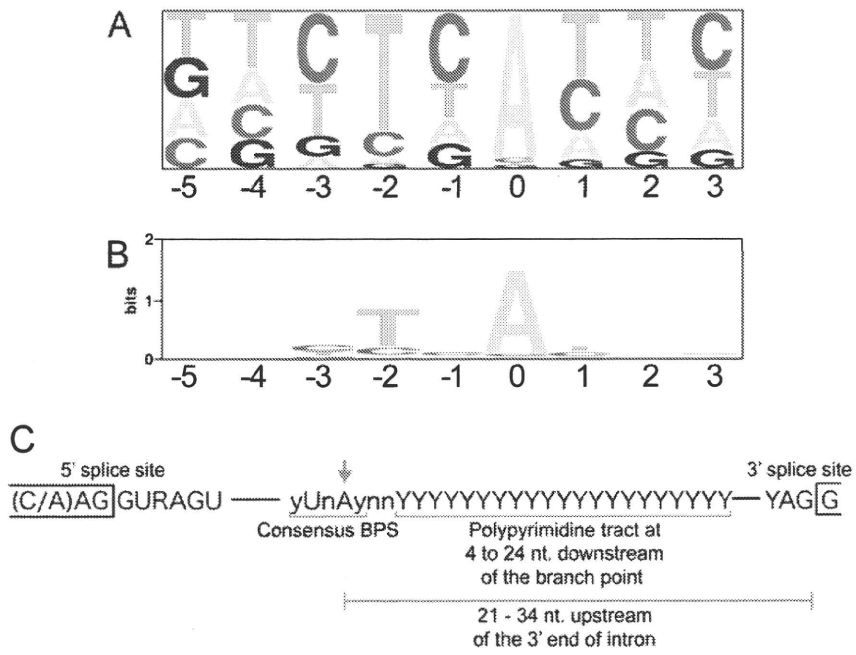


Figure 4

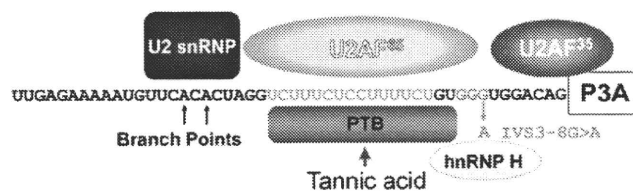


Figure 5

mRNA from normal allele

a 1 2 3 4 5 6 7 8 9 10 11 12

b 1 2 3 4 5 6 7 8 9 10 11 12 → Degraded
6 PTC

mRNA from mutant allele with 7-nt deletion

c 1 2 3 4 5 6 7 8 9 10 11 12 → Degraded
PTC

d 1 2 3 4 5 7 8 9 10 11 12
6

Figure 6

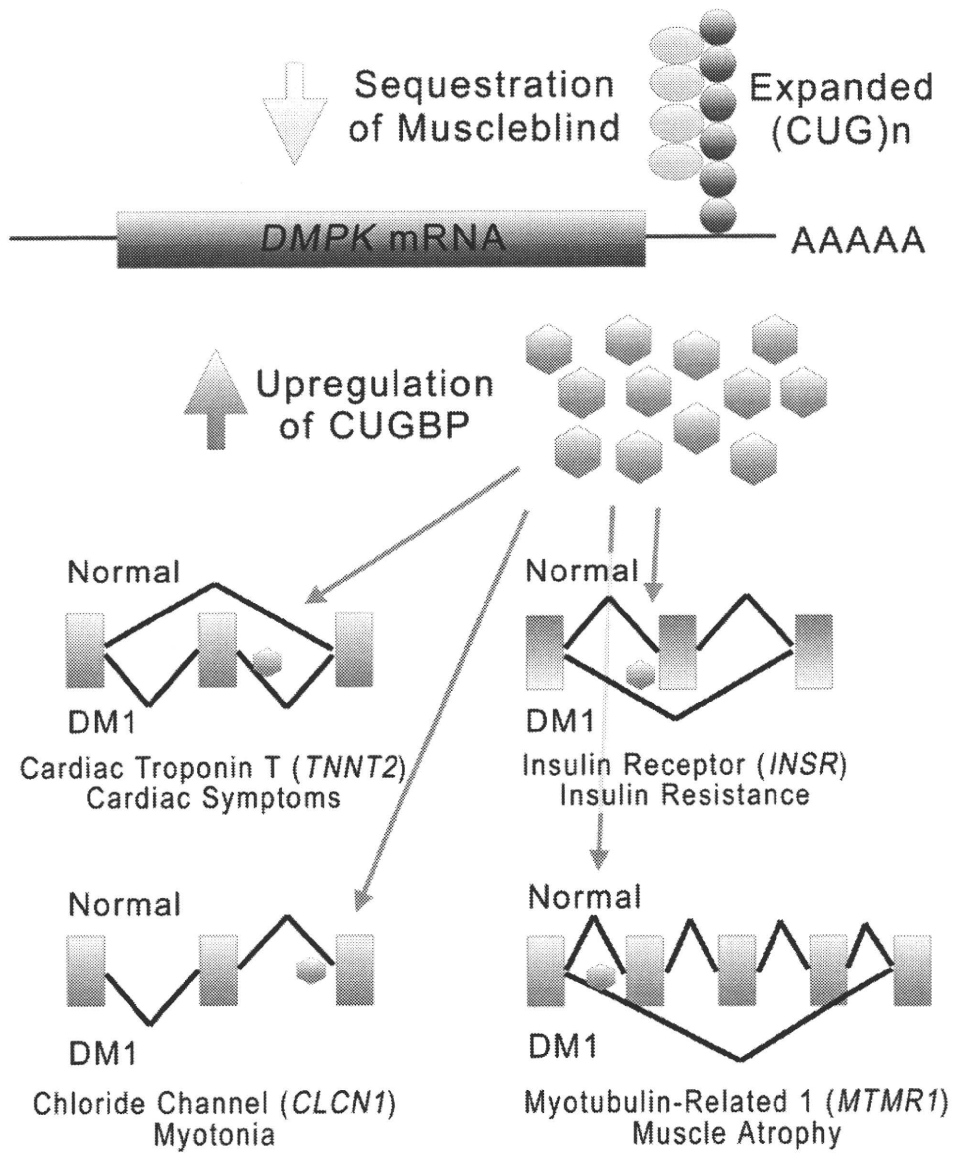


Figure 7

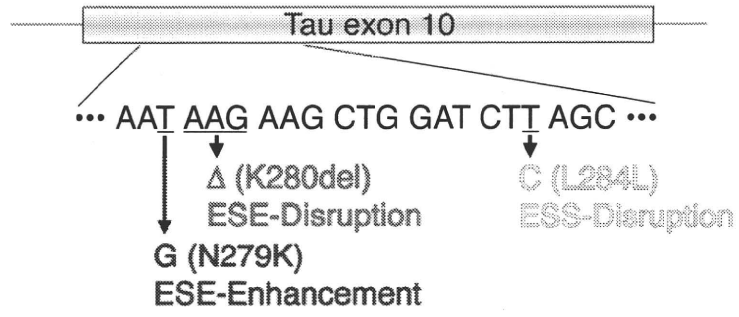
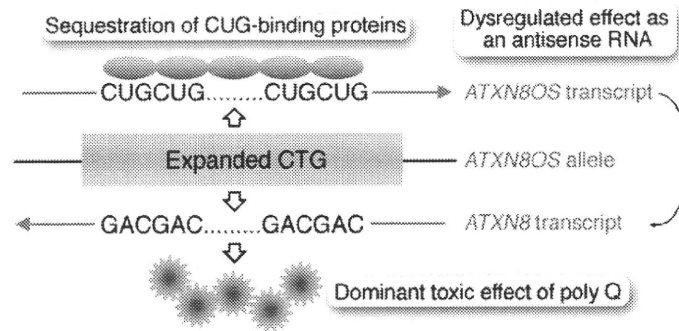
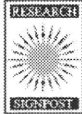


Figure 8





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Kerala, India

Pharmacology of Nicotinic Acetylcholine Receptors from the Basic and Therapeutic Perspectives, 2011
00-00 ISBN: 978-81-308-0454-5 Editor: Hugo R. Arias

8. Molecular defects of acetylcholine receptor subunits in congenital myasthenic syndromes

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Abstract. Congenital myasthenic syndromes (CMS) are caused by mutations in molecules expressed at the neuromuscular junction. Among eight defective molecules identified to date in CMS, mutations in the muscle nicotinic acetylcholine receptor (AChR) subunits are the first to be characterized and are prevalent. Mutations in the AChR subunit genes cause three phenotypes: (i) endplate AChR deficiency in which the number of AChR at the endplate is critically reduced; (ii) slow channel syndrome in which mutations either at the acetylcholine binding sites or at the ion channel pore increases synaptic response to acetylcholine and prolongs AChR channel opening events; and (iii) fast channel syndrome in which mutations either at the acetylcholine binding sites or at the long cytoplasmic loop between the third and fourth transmembrane domains compromise synaptic response to acetylcholine and shortens AChR channel openings. In addition, mutations in AChR subunit genes also cause fetal akinesia deformation sequence, and a single nucleotide polymorphism in the promoter region of the AChR $\alpha 1$ subunit is associated with early onset myasthenia gravis.

Introduction

Congenital myasthenic syndromes

Congenital myasthenic syndromes (CMS) are heterogeneous disorders caused by congenital defects of molecules expressed at the neuromuscular junction (Fig. 1). Each mutation affects the expression level of the mutant molecule and/or compromises the functional properties of the mutant molecule. The mutant molecules identified to date

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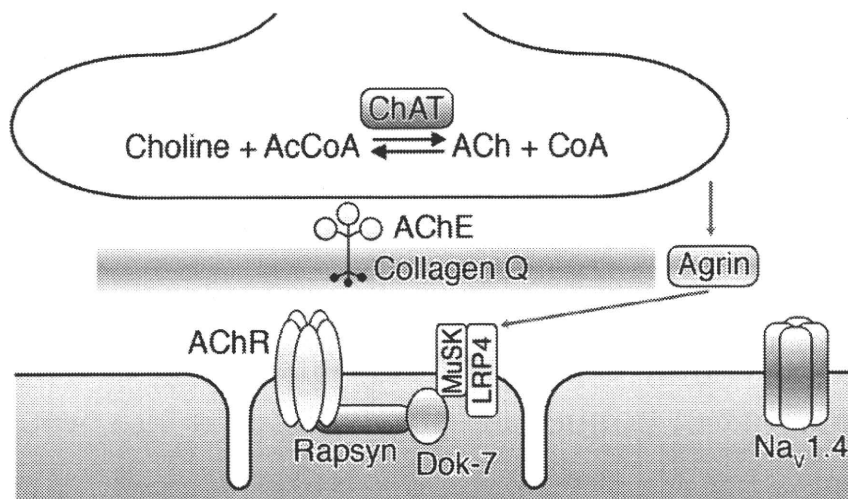


Figure 1. Schematic of molecules expressed at the neuromuscular junction. Mutated molecules in CMS include acetylcholine receptor (AChR), rapsyn, agrin, MuSK, Dok-7, skeletal muscle sodium channel type 1.4 (Na_v1.4), collagen Q, and choline acetyltransferase (ChAT).

include (i) acetylcholine receptor (AChR) subunits [1,2], (ii) rapsyn that anchors and clusters AChRs at the neuromuscular junction [3,4], (iii) agrin that is released from the nerve terminal and induces AChR clustering by stimulating the downstream LRP4/MuSK/Dok-7/rapsyn/AChR pathway [5], (iv) MuSK that transmits the AChR-clustering signal from agrin/LRP4 to Dok-7/rapsyn/AChR [6,7], (v) Dok-7 that transmits the AChR-clustering signal from agrin/LRP4/MuSK to rapsyn/AChR [8,9], (vi) skeletal muscle sodium channel type 1.4 (Na_v1.4) that spreads depolarization potential from endplate throughout muscle fibers [10], (vii) collagen Q that anchors acetylcholinesterase (AChE) to the synaptic basal lamina [11-13], (viii) choline acetyltransferase (ChAT) that resynthesizes acetylcholine from recycled choline at the nerve terminal [14]. AChR [15], MuSK [16,17], and LRP4 [18] are also targets of myasthenia gravis, in which autoantibody against these molecules compromises the neuromuscular transmission. This review focuses on molecular defects of AChR in CMS.

Muscle nicotinic acetylcholine receptor

Nicotinic AChRs are pentameric ligand-gated ion channels. The family of pentameric ligand-gated ion channels includes cationic AChRs, cationic serotonergic receptors (5HT₃), anionic glycine receptors, and anionic GABA_A and GABA_C receptors [19]. Heteromeric neuronal nicotinic AChRs are comprised of various combinations of α ($\alpha 2$ - $\alpha 7$) and β subunits ($\beta 2$ - $\beta 4$), whereas homomeric AChRs are formed by just one subunit type (e.g., $\alpha 7$ - $\alpha 9$) [20]. On the other hand, muscle nicotinic AChRs have only two forms: fetal AChR that carries the $\alpha 1$, $\beta 1$, δ , and γ subunits encoded by *CHRNA1*, *CHRN1*, *CHRND*, *CHRNG*, respectively, in the stoichiometry $\alpha 1_2\beta 1\delta\gamma$; and adult-type AChR that carries the ϵ subunit instead of the γ subunit in the stoichiometry $\alpha 1_2\beta 1\delta\epsilon$ [21]. The ϵ subunit is encoded by *CHRNE*. The muscle nicotinic AChR harbors

two binding sites for ACh at the interfaces between α - δ and α - γ/ϵ subunits [22,23]. Binding of a single ACh molecule opens the channel pore but for a short time. Binding of two ACh molecules stabilizes the open state of AChR, and AChR stays open for longer time. Cations but no anions pass through the channel pore of nicotinic AChRs. Unlike sodium, potassium, or calcium channels, AChRs, in general, have no selectivity for cations, but $\alpha 7$ AChRs have 10-20 times higher permeability for Ca^{2+} than for Na^+ .

Endplate AChR deficiency

Congenital deficiency of endplate AChRs is caused by mutations in genes encoding the AChR subunits. The mutated genes include *CHRNA1*, *CHRNB1*, *CHRND*, and *CHRNE*, but not *CHRNA1*. Endplate AChR deficiency is also caused by mutations in molecules that transmit signals for AChR clustering. These include *AGRN* encoding agrin [5], *MUSK* encoding MuSK [6,7], *DOK7* encoding Dok-7 [8,9], and *RAPSN* encoding rapsyn [3,4]. Mutations in the signaling molecules, however, are not within the scope of this review and are not addressed.

Two different groups of mutations of the AChR subunit genes cause endplate AChR deficiency. The first group includes null mutations in *CHRNE* encoding the AChR ϵ subunit. The null mutations are caused by frameshifting DNA rearrangements, *de novo* creation of a stop codon, and frameshifting splicing mutations. Large-scale in-frame DNA rearrangements also abolish expression of the AChR ϵ subunit. Mutations in the promoter region [24] and missense mutations [25] do not completely nullify the expression of ϵ , but the molecular pathological consequences are indistinguishable from those of null mutations. Lack of the ϵ subunit can be compensated for by the presence of the γ subunit that is normally expressed in embryos [26]. The patients can survive with γ -AChR even when ϵ -AChR is lacking. If a null mutation resides on the other AChR subunit genes, the affected individual should have no substituting subunit and cannot survive. Indeed, two such homozygous missense mutations are reported in *CHRNA1* and *CHRND* in lethal fetal akinesia disorders [27]. In general, mutations causing monogenic diseases should be very rare, because a single nucleotide substitution among the 3.0×10^9 nucleotides in a single allele should exhibit a certain phenotype that is recognized as a disease. Most single nucleotide substitutions are likely to be silent or to partially confer a variable phenotype observed in normal individuals. Most of the other nucleotide changes cause early embryonic lethality and we cannot observe such mutations in patients.

The second group of mutations affecting the AChR subunit genes includes missense mutations of *CHRNA1*, *CHRNB*, and *CHRND* encoding the AChR $\alpha 1$, $\beta 1$, and δ subunits, respectively. These mutations compromise the expression level of the mutant subunit and/or the assembly of AChRs, but do not completely abolish the expression of AChRs. Differences between mutations in *CHRNE* and those in *CHRNA1*, *CHRNB*, and *CHRND* are the tolerance to low expression of the affected subunit. The expression level of the ϵ subunit may go to zero, whereas a patient needs a certain amount of AChRs to be expressed at the endplate to survive when a mutation is in a gene for either the $\alpha 1$, $\beta 1$, or δ subunit. Patients with low-expressor mutations in *CHRNA1*, *CHRNB*, and *CHRND* tend to have a devastating course with high fatality. Some missense mutations in *CHRNA1*, *CHRNB*, *CHRND*, and *CHRNE* also affect the AChR channel kinetics. If a pathological effect due to aberration of the

channel kinetics is more than the degree of aberration of AChR expression, such a mutation is classified as slow channel or fast channel mutation.

In biopsied skeletal muscle, we observed several lines of evidence indicating a decreased number of AChRs at the endplate. Ultrastructural studies demonstrate simplified junctional folds at the endplate and reduced staining for AChRs. Miniature endplate currents (MEPC) are small in amplitude. As the number of ACh in a synaptic vesicle (quanta) is rather increased, the low MEPC amplitude directly indicates a reduced number of AChRs at the endplate. Endplate potentials (EPP) are also small in amplitude. Again, as the number of ACh released by a single nerve stimulus (quantal content) is rather elevated, the low EP amplitude indicates a reduced number of AChRs at the endplate. In patients with null mutations in *CHRNE*, single channel recordings of AChRs at the patient's endplates demonstrate low conductance and prolonged opening bursts, indicating expression of the fetal γ -AChR instead of the adult-type ϵ -AChR. The conductance of the adult-type ϵ -AChR is 80 pS, whereas that of the fetal γ -AChR is 60 pS. In patients with low-expressor mutations in either *CHRNA1*, *CHRN1*, or *CHRND*, single channel recordings demonstrate no or minor kinetic abnormalities.

As in autoimmune myasthenia gravis, endplate AChR deficiency is generally well controlled by regular dosages of anticholinesterases. Anticholinesterases inhibit the catalytic activity of AChE, which in turn increases the dwell time of ACh at the synaptic space and enables reassociation of ACh and AChRs. Anticholinesterases are effective in a wide range of diseases where the number of AChRs is reduced independent of its cause. Inadvertent or unexpected overdose of anticholinesterase, however, simulates endplate AChE deficiency [11,13,28]. In endplate AChE deficiency, the neuromuscular signal transduction is compromised by an excessive amount and prolonged dwell time of ACh in the synaptic space, which in turn induces three pathomechanisms: (i) staircase summation of endplate potentials, (ii) excessive desensitization of AChRs, and (iii) endplate myopathy caused by excessive influx of extracellular calcium. The three molecular mechanisms are identical to those observed in the slow channel congenital myasthenic syndrome, as described in the following section.

Slow channel congenital myasthenic syndrome

The second class of CMS due to mutations in the AChR subunit genes is the slow channel congenital myasthenic syndrome (SCCMS) (Table 1). SCCMS is an autosomal dominant disorder, in which a gain-of-function mutation on a single allele compromises the neuromuscular signal transduction [1]. The mutation causes prolonged AChR channel openings and increases the synaptic response to ACh (Fig. 2). There is a single reported case of autosomal recessive SCCMS, in which the ϵ L78P mutation minimally prolongs channel opening events and a mutant channel arising from a single allele is not sufficient to cause the disease phenotype [29]. In general, dominantly inherited disorders tend to develop after adolescence, because an individual carrying a mutant allele should get married and transmit the mutant allele to the next generation. In concordance with this notion, SCCMS tend to develop later in life and progresses slowly. Some patients with SCCMS, however, present early in life and become severely disabled even in the first decade.

Table 1. AChR mutations causing slow and fast channel syndromes.

Gene	Mutation	Domain	Reference
Slow channel syndrome			
<i>CHRNA1</i> (AChR α 1 subunit)	α 1G153S	ECD	[2,30]
	α 1V156M	ECD	[30]
	α 1N217K	M1	[31-33]
	α 1S226Y	M1	[34]
	α 1S226F	M1	[34,35]
	α 1V249F	M2	[36]
	α 1T254I	M2	[30]
	α 1S269I	M2-M3 linker	[30]
	α 1C418W	M4	[37]
<i>CHRN1</i> (AChR β 1 subunit)	β 1V229F	M1	[33,38]
	β 1L262M	M2	[39]
	β 1V266M	M2	[31,33]
<i>CHRN1</i> (AChR β 1 subunit)	β 1V266A	M2	[40]
	<i>CHRNA1</i> (AChR δ subunit)	δ S268F	M2
<i>CHRNE</i> (AChR ϵ subunit)	ϵ L78P	ECD	[29,42]
	ϵ L221F	M1	[29,43]
	ϵ I257F	M2	[44]
	ϵ V259F	M2	[45]
	ϵ V259L	M2	[46]
	ϵ T264P	M2	[1]
	ϵ V265A	M2	[47]
ϵ L269F	M2	[31,48]	
Fast channel syndrome			
<i>CHRNA1</i> (AChR α 1 subunit)	α 1V132L	ECD	[49]
	α 1F256L	M2	[50]
	α 1V285I	M3	[51]
<i>CHRNA1</i> (AChR α 1 subunit)	δ L42P	ECD	[52]
	δ E59K	ECD	[53]
<i>CHRNE</i> (AChR ϵ subunit)	ϵ P121L	ECD	[54]
	ϵ P121T	ECD	[55]
	ϵ D175N	ECD	[56]
	ϵ N182Y	ECD	[56]
	ϵ 1254ins18	LCP	[57]
	ϵ A411P	LCP	[58]
	ϵ N436del	LCP	[59,60]

In SCCMS, neuromuscular transmission defects are caused by three distinct mechanisms. First, staircase summation of endplate potentials causes depolarization of ECD, extracellular domain; M1-M4, transmembrane domains 1 to 4; LCP, long cytoplasmic loop. The membrane potential. Prolonged depolarization makes the voltage-gated skeletal sodium channel less responsive to endplate potential generated by opening of AChR ion channels. Second, mutant AChRs somehow tend to be desensitized [36], which reduces the number of AChRs that respond to the released ACh quanta. Third,

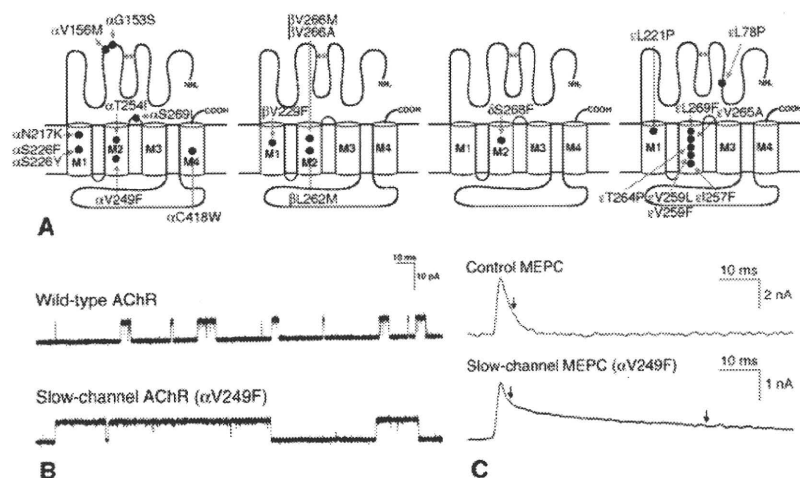


Figure 2. Slow channel syndrome. **(A)** Schematic diagram of AChR subunits with SCCMS mutations. **(B)** Single channel currents from wild-type and slow channel ($\alpha 1V249F$) AChRs expressed on HEK293 cells. **(C)** Miniature endplate current (MEPC) recorded from endplates of a control and a patient harboring $\alpha 1V249F$. The patient's MEPC decays biexponentially (arrows) due to expression of both wild-type and mutant AChRs.

prolonged opening of AChR causes excessive influx of extracellular calcium, which triggers the apoptosis pathway and gives rise to endplate myopathy [61]. In normal adult human ϵ -AChR, 7% of the synaptic current is carried by Ca^{2+} , which is higher than that in fetal human γ -AChR or muscle AChRs from other species [62]. This predisposes to endplate Ca^{2+} overloading when the channel opening events are prolonged. In addition, at least two SCCMS mutations, $\epsilon T264P$ [1] and $\epsilon V259F$ [45], increase the Ca^{2+} permeability 1.5- and 2-fold, respectively [63].

Slow channel mutations can be divided into two groups (Table 1). The first group includes mutations at the extracellular domain like $\alpha 1G153S$ [2], as well as at the N-terminal part of the first transmembrane domain like $\alpha 1N217K$ [32] and $\epsilon L221F$ [43]. These mutations increase the affinity for ACh binding, probably by retarding the dissociation of ACh from the binding site, which gives rise to repeated channel openings after a single event of ACh binding. The second group includes mutations at the second transmembrane domain (M2) that lines the ion channel pore. These mutations mostly introduce a bulky amino acid into the channel lining face, but $\epsilon T264P$ [1] introduces a kink into the channel pore, whereas $\beta 1V266A$ [40] and $\epsilon V265A$ [47] rather introduce a smaller amino acid into the pore. Mutations in M2 retard the channel closing rate α and variably enhance the channel opening rate β . Some mutations in M2 also increase affinity for ACh, which include $\alpha 1V249F$ [36], $\epsilon L269F$ [31], and $\epsilon T264P$ [1].

SCCMS can be effectively treated with conventional dosages of long-lived open channel blockers of AChR, such as the antiarrhythmic agent quinidine [64,65] and the antidepressant fluoxetine [66]. Quinidine reduces the prolonged burst duration of SCCMS to the normal level at $5 \mu M$ [64]. As the concentration of quinidine in the treatment of cardiac arrhythmia is 6-15 μM , 5 μM is readily attainable in clinical practice and indeed

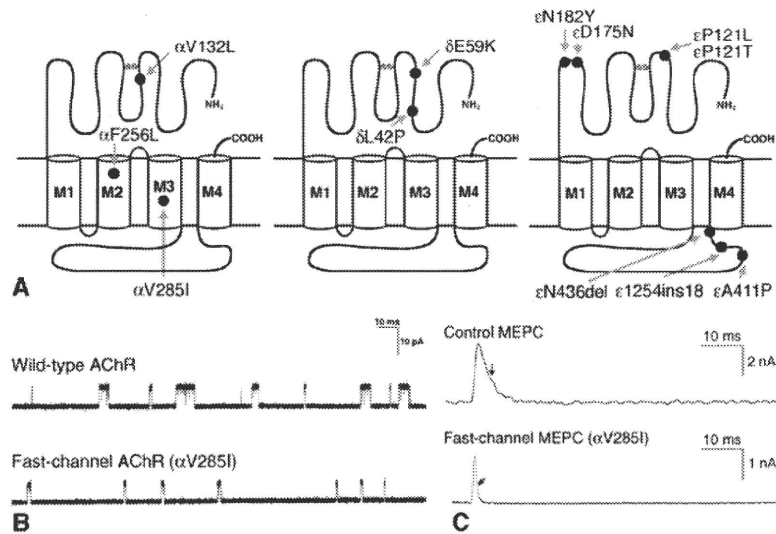


Figure 3. Fast channel syndrome. (A) Schematic diagram of AChR subunits with FCCMS mutations. (B) Single channel currents from wild-type and fast channel ($\alpha 1V285I$) AChRs expressed on HEK293 cells. (C) Miniature endplate current (MEPC) recorded from endplates of a control and a patient harboring $\alpha 1V285I$. The patient's MEPC decays faster than that of the normal control.

demonstrates significant effects [65]. Similarly, fluoxetine reduces the prolonged burst duration to the normal level at 10 μM , which is clinically attainable without adverse effects at 80 to 120 mg/day of fluoxetine [66].

Fast channel congenital myasthenic syndrome

The third class of CMS due to mutations in AChR subunit genes is the fast channel congenital myasthenic syndrome (FCCMS) (Table 1). FCCMS is kinetically opposite to SCCMS (Fig. 3). In FCCMS, the AChR becomes resistant to be transferred to an open state and prematurely comes back to a closed state, which results in insufficient depolarization of the endplate potential. The resulting pathophysiology is thus similar to endplate AChR deficiency, but the cause of inefficient endplate depolarization is due to qualitative defects of AChRs but not to quantitative defects as in AChR deficiency.

FCCMS is an autosomal recessive disorder. One allele carries a missense mutation that confers a fast closure of AChRs, and the other allele usually harbors a low-expressor or null mutation. As in heterozygous healthy parents of endplate AChR deficiency, we humans may completely lack 50% of each AChR subunit without any clinical symptoms. In FCCMS, a low-expressor or null mutation on one allele unmasks kinetic abnormalities of a FCCMS mutation on the second allele. Detailed kinetic analyses of FCCMS mutations have unmasked yet uncharacterized molecular architectures of the AChR subunits. Three such examples are presented here.

$\epsilon 1254ins18$ causes a duplication of STRDQE codons at positions 413 to 418 close to the C-terminal end of the long cytoplasmic loop (LCP) linking the third (M3) and fourth (M4) transmembrane domains. $\epsilon 1254ins18$ -AChR expressed on HEK293 cells opens in

three different modes. The opening probabilities of normal AChRs are clustered into a single large peak, whereas the ϵ 1254ins18-AChR shows three different peaks [57]. In all the three modes, the AChR is activated slowly and inactivated rapidly, which gives rise to an inefficient synaptic response to ACh. Another FCCMS mutation, ϵ A411P, in the LCP also destabilizes the channel opening kinetics. The channel opening probabilities of ϵ A411P-AChRs are widely distributed and do not form any discernible peaks [58]. Our analysis first disclosed that the function of LCP is to stabilize the open conformation of the AChR.

ϵ N436del is a deletion of Asn at the C-terminal end of the LCP. The deletion shortens the LCP and shifts a negatively charged Asp residue at codon 435 against M4. ϵ N436del-AChR decreases the duration of channel opening bursts 2.7-fold compared to the wild type due to a 2.3-fold decrease in gating efficiency and a 2.5-fold decrease in agonist affinity of the diliganded closed state. A series of artificial mutations established that the effects of ϵ N436del are not due to juxtaposition of a negative charge against M4 but to the shortening of the LCP. Deletion of the C-terminal residue of the LCP of the β 1 and δ subunits also results in fast-channel kinetics, but that in the α 1 subunit dictates slow-channel kinetics. Thus, the LCPs of four AChR subunits contribute in an asymmetric manner to optimize the activation of AChRs through allosteric links to the channel and to the agonist binding sites [59].

The mutation α 1V285I introduces a bulky amino acid into the M3 transmembrane domain and causes FCCMS (Fig. 3). Kinetic studies demonstrate that the mutation slows the channel opening rate β and speeds the channel closing rate α , which gives rise to a 15.1-fold reduction in the channel gating equilibrium constant θ ($= \beta/\alpha$). On the other hand, the mutation minimally affects affinity for ACh. The probability of channel openings decreased when we introduced Leu, a bulky amino acid, at position V285, but rather increased when we introduced smaller amino acids such as Thr and Ala. We observed similar effects when we introduced similar substitutions into the β 1, δ , and ϵ subunits. Thus, introduction of bulky amino acids narrows the channel pore, while introduction of smaller amino acids widens the channel pore. Our analysis first disclosed that the M3 domain backs up the channel-lining pore that is composed by the M2 transmembrane domains and has stereochemical effects on channel gating kinetics [51].

FCCMS can be effectively treated with anticholinesterases and 3,4-diaminopyridine. The mechanism of action of anticholinesterases is described in the section devoted to endplate AChR deficiency. The drug 3,4-diaminopyridine blocks the presynaptic potassium channel, which slows the repolarization of the action potential delivered to the nerve terminal [67]. The enhanced nerve action potential stimulates the presynaptic voltage-gated P/Q-type and N-type Ca^{2+} channels and increases Ca^{2+} influx to the nerve terminal, which then enhances synaptotagmin and the SNARE complex to facilitate the fusion of ACh vesicles to the presynaptic membrane. This increases the amount of ACh released by a single nerve stimulus and enhances AChR channel openings.

Other phenotypes associated with AChR mutations and a single nucleotide polymorphism

Mutations or a single nucleotide polymorphism (SNP) in muscle nicotinic AChR subunits also give rise to phenotypes other than CMS.

The first phenotype is fetal akinesia deformation sequence (FADS). Mutations in the AChR subunit genes cause neuromuscular transmission defects in embryos and restrict

intrauterine movements. As human embryos use the fetal γ -AChR by 33 weeks of gestation [68], mutations in *CHRNA1* and *CHRND* [27], as well as in *CHRNA1* and *CHRND* [69,70], as well as in *CHRNA1* and *CHRND* [27], cause FADS.

The second phenotype is early onset myasthenia gravis [71]. Promiscuous expression of a set of self-antigens occurs in medullary thymic epithelial cells to impose T-cell tolerance and to provide protection against autoimmune disorders. The AChR α subunit is one of those self-antigens. A SNP in the promoter region of *CHRNA1* compromises expression of the α 1 subunit in thymic epithelial cells, which increases the chance of developing myasthenia gravis 2.01- to 2.35-fold in individuals carrying the SNP.

Conclusions

We addressed three types of CMS that are caused by mutations in the AChR subunit genes.

Congenital deficiency of endplate AChRs is caused by mutations in *CHRNA1*, *CHRNBI*, *CHRND*, and *CHRNE* encoding the AChR α 1 β 1 δ and ϵ subunits, respectively. The mutations are classified into two groups. The first group includes mutations in *CHRNE* that nullify or significantly reduce the expression of the ϵ subunit. Patients survive with embryonic γ -AChR even when the adult-type ϵ -AChR is lacking. Null mutations in the other AChR subunit genes are likely to be fatal, which supports a general notion that we have no chance to identify mutations that result in lethal phenotypes. The second group of mutations includes missense mutations of *CHRNA1*, *CHRNBI*, and *CHRND*. These mutations compromise the expression level of the mutant subunit and/or the assembly of AChRs, but do not completely abolish the expression of AChRs. Differences between mutations in *CHRNE* and those in *CHRNA1*, *CHRNBI*, and *CHRND* are the tolerance to low expression of the affected subunit. As in autoimmune myasthenia gravis, endplate AChR deficiency is well controlled by anticholinesterases.

The slow channel congenital myasthenic syndrome (SCCMS) is an autosomal dominant disorder, in which a gain-of-function mutation causes prolonged AChR channel openings and increases the synaptic response to ACh. In SCCMS, neuromuscular transmission defects are caused by (i) staircase summation of endplate potentials, (ii) excessive desensitization of AChRs, and (iii) endplate myopathy caused by excessive influx of extracellular calcium. SCCMS mutations cause neuromuscular transmission defects either by increasing the affinity of AChR for ACh binding or by retarding the channel closing rate α and variably enhancing the channel opening rate β . SCCMS can be effectively treated with conventional dosages of long-lived open channel blockers of AChR, such as the antiarrhythmic agent quinidine and the antidepressant fluoxetine.

The fast channel congenital myasthenic syndrome (FCCMS) is caused by loss-of-function missense mutations in the AChR subunit genes. The mutations render the AChR resistant to be transferred to an open state and prematurely coming back to a closed state. Detailed kinetic analyses of FCCMS mutations have unmasked yet uncharacterized molecular architectures of the AChR subunits especially in the third transmembrane domain and in the long cytoplasmic loop. FCCMS can be effectively treated with anticholinesterases and 3,4-diaminopyridine.

Two more clinical phenotypes are associated with variations of the AChR subunit genes. Mutations in *CHRNA1* encoding the AChR α subunit cause another phenotype FADS by restricting intrauterine movement of an embryo. A SNP in the promoter region

of *CHRNA1* compromises expression of the $\alpha 1$ subunit in thymic epithelial cells, and increases the chance of developing myasthenia gravis.

Acknowledgements

Works in our laboratories were supported by Grants-in-Aid from the MEXT and the MHLW of Japan (to K.O.), and by the NIH grant (NS6277) and the Muscular Dystrophy Association grant (to A.G.E.).

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CHAPTER 8

CONGENITAL MYASTHENIC SYNDROMES

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CHAPTER OUTLINE

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MECHANISMS COMPROMISING THE SAFETY MARGIN OF NEUROMUSCULAR TRANSMISSION

THE CLASSIFICATION OF CONGENITAL MYASTHENIC SYNDROMES

THE INVESTIGATION OF CONGENITAL MYASTHENIC SYNDROMES

Clinical Observations

HISTORY AND EXAMINATION

THE INTRAVENOUS EDROPHONIUM TEST

EMG STUDIES

SEROLOGIC TESTS

Morphology

ROUTINE HISTOCHEMICAL STUDIES

CYTOCHEMICAL LOCALIZATION OF AChE

IMMUNOCYTOCHEMICAL LOCALIZATION OF AChR AND OTHER ENDPLATE ASSOCIATED PROTEINS

ELECTRON MICROSCOPY AND ELECTRON CYTOCHEMISTRY

¹²⁵I- α -bgt Binding Sites Per Endplate

In Vitro Electrophysiology Studies

CONVENTIONAL MICROELECTRODE STUDIES

Molecular Genetic Studies

EXPRESSION STUDIES

PRESYNAPTIC CMS

CMS Caused by Defects in Choline Acetyltransferase (ChAT)

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MORPHOLOGY

MOLECULAR STUDIES

TREATMENT

Paucity of Synaptic Vesicles and Reduced Quantal Release

Congenital Myasthenic Syndrome Resembling the Lambert-Eaton Syndrome

SYNAPTIC BASAL-LAMINA-ASSOCIATED CMS

Endplate Acetylcholinesterase Deficiency

CLINICAL ASPECTS

ELECTROPHYSIOLOGY FEATURES

MORPHOLOGY

PATHOPHYSIOLOGY

MOLECULAR PATHOGENESIS

DIAGNOSIS

THERAPY

Defects in β 2-Laminin

POSTSYNAPTIC CMS

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Slow-Channel Syndromes

CLINICAL ASPECTS

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