

Aujla *et al.*² note that patients with cystic fibrosis and chronic lung colonization by bacteria have elevated pulmonary lymph node production of IL-22, but it is unclear whether the cytokine is beneficial or harmful in the pathophysiology of the disease. Thus the utility of IL-22 as a drug remains to be determined.

It seems possible that administration of antimicrobial peptides might be therapeutically effective. Aujla *et al.*² show that IL-22 induces production of lipocalin 2, which is critical for the microbicidal effects of this

cytokine. Furthermore, Zheng *et al.*¹ were able to rescue *Il22*-deficient animals with *C. rodentium* infections by supplementing a key antibacterial protein—RegIII γ —a somewhat surprising finding, given that Gram-negative bacteria have been reported to be resistant to the effects of RegIII γ . Nevertheless, manipulation of IL-22 and its signaling pathways may have the potential to provide treatments for diverse conditions ranging from inflammatory bowel disease to HIV.

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Reducing glutamate signaling pays off in fragile X

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A recent theory about the basis for fragile X syndrome is now validated in a mouse model. The findings point the way to treatment options targeting group 1 metabotropic glutamate receptors.

Basic science on disease mechanisms is driven by hypotheses that persist long enough to be frequently disproved or complicated by tests of principle in animal models. Rarely, but all the more compellingly, a scientific model derived from basic research turns out to be appropriate for actual medical therapy in people. An exciting example is the report by Dolen *et al.*¹ in a recent issue of *Neuron*.

This study is a major step toward the ultimate goal of treating fragile X syndrome (FXS), an inherited form of mental retardation and the predominant monogenetic cause of autism. The study is also a genetic test of the 'mGluR theory' of FXS, which postulates that an important consequence of FXS is excessive signaling through group 1 metabotropic glutamate receptors (mGluRs), a specific subfamily of seven-transmembrane receptors involved in different forms of synaptic plasticity. The theory holds that the disease might be treated by mGluR antagonists. Dolen *et al.*¹ now show that genetic reduction of mGluR signaling can reverse several typical FXS symptoms in mice and provide the first comprehensive evidence that the mGluR theory of FXS holds true.

FXS is characterized by a trinucleotide expansion in the 5' UTR of the *FMR1* gene. The expansion prevents expression of the fragile X mental retardation protein (FMRP), an mRNA-binding protein and negative regulator of translation². FMRP seems to put the breaks on local protein synthesis at synapses in response to activation of group 1 mGluRs³. Synaptic stimulation of group 1 mGluRs by 3,5-dihydroxyphenylglycine (DHPG, a potent agonist of group 1 mGluRs) triggers phosphorylation of translation initiation factors and leads to increased translation of specific mRNAs at synapses^{2,3}.

In a landmark publication for FXS research, Huber and colleagues reported that FMRP knockout mice have an exaggerated form of mGluR-induced long-term depression of synaptic transmission in the hippocampus (mGluR-LTD) (ref. 4 and references cited therein). In contrast to wild-type, mGluR-LTD expression in the FMRP knockout mice did not require new protein synthesis (ref. 4 and references cited therein). Interestingly, a subsequent study showed that DHPG-evoked translational responses of certain FMRP target mRNAs are dysregulated at FMRP-deficient synapses⁵.

These and other reports culminated in the hypothesis that FXS is a disease of excess basal translation^{1,3} and simultaneous loss of mGluR-induced translation⁵, as a result of uncontrolled signaling via group 1 mGluR receptors³ (Fig. 1). Pharmacologic antagonism of group 1 mGluR5 (with, for instance, 2-methyl-6-(phenylethyl)pyridine (MPEP)) has therefore been an attrac-

tive strategy to reverse FXS symptoms^{6–8}. By feeding MPEP to flies, McBride *et al.*⁶ could reverse defects in neuronal morphology and synaptic plasticity in a *Drosophila melanogaster* FXS model. Several other studies used FMRP knockout mice to show that MPEP ameliorates the hyperexcitability of FXS neurons, *in vitro* in brain slices as well as *in vivo* in living mice^{7,8}.

These experiments, although a good indication of the *in vivo* relevance of the mGluR theory for humans, have important drawbacks: they only address a very limited spectrum of FXS phenotypes, largely disregarding the cognitive impairments, and, furthermore, they do not provide a final proof of the role of mGluR signaling in FXS because the applied antagonist MPEP has substantial off-target effects, such as inhibition of *N*-methyl-D-aspartic acid receptors⁹. Thus, there has been little evidence so far that the mGluR theory indeed proves true *in vivo*, especially in mammals.

The work of Dolen *et al.*¹ is the first study using a genetic, instead of a pharmacologic, approach to reduce mGluR5 activity in FXS. The authors crossed FMRP knockout mice with mice heterozygous for the gene encoding mGluR5, generating double mutants with a 50% decrease in mGluR5 expression¹. This reduction in mGluR5 signaling was sufficient to rescue six out of seven FXS phenotypes (Fig. 1). Notably, the only phenotype that could not be rescued was a non-neurological defect, increased size of testes.

Importantly, and as opposed to several earlier studies, Dolen *et al.*¹ focused on phenotypes that correlate with the human disease pattern.

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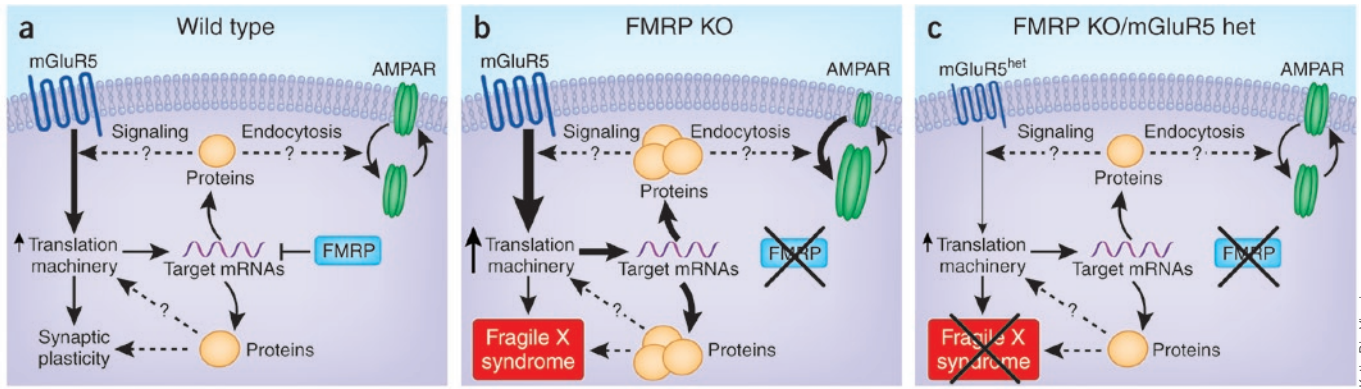


Figure 1 Genetic reduction of metabotropic glutamate receptor 5 (mGluR5) signaling rescues fragile X syndrome (FXS) in mice. (a) mGluR5 signaling in wild-type mice activates the translation machinery and induces specific protein synthesis-dependent forms of synaptic plasticity. Some of the mGluR5-regulated mRNAs are translationally suppressed by fragile X mental retardation protein (FMRP). (b) In FMRP knockout mice (FMRP KO), FMRP target mRNAs are translated excessively and mGluR5 signaling is exaggerated. (c) Dolen *et al.*¹ now show that genetic reduction of mGluR5 signaling in mGluR5 heterozygous mice (mGluR5 het) restores translation rates and rescues FXS phenotypes in FMRP KO. Putative functions of the proteins encoded by FMRP target mRNAs might include control of α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPA) endocytosis, cell signaling or translation.

Defects rescued by lowering mGluR5 expression included a specific form of memory failure that might reflect the cognitive impairments in humans with FXS, as well as two phenotypes that have perfect correlates in people with FXS: higher susceptibility to seizures and altered morphology of dendritic spines. It is noteworthy that mice heterozygous for the gene encoding mGluR5 were indistinguishable from wild-type mice in regard to these phenotypes, indicating that the observed rescue is not a compound effect of opposing defects, but a true reversal of FXS symptoms.

The absence of any major abnormalities in the heterozygous mice is especially important because a total knockout of mGluR5 expression in mice leads to severe impairment in synaptic plasticity¹⁰, suggesting that complete silencing of mGluR5 function in humans would probably have devastating effects and therefore no therapeutic benefit.

This work substantially contributes to the hope that reducing excessive mGluR5 signaling could be successfully applied to treat people with FXS. However, as gene therapy in humans is still far from being a realistic application, attempts to treat FXS in the near future will still have to focus on pharmacologic reduction of mGluR signaling. Recently, several pharmaceutical companies have begun to develop and test new, highly specific and potent drugs that antagonize mGluR5 signaling for the treatment of epilepsy and FXS, and these compounds will probably be tested in people with FXS soon.

Although comprehensive and compelling, the work of Dolen *et al.*¹ still leaves unanswered questions: why are not all phenotypes rescued by reduction of mGluR5 signaling? What about those FXS phenotypes that represent more subtle deficits in higher cognition and are therefore not reflected by the mouse model, such as autistic behavior? There is certainly an aspect of FXS that cannot be rescued by mGluR5 inhibition.

In addition to the DHPG-induced LTD, two other forms of hippocampal LTD that are dependent on protein synthesis, but independent of mGluR1 and mGluR5 activation, are also exaggerated in FMRP knockout mice¹¹. These three LTD-stimulation paradigms converge on a common signaling mechanism through the small GTPase Gq, indicating that detailed dissection of the affected downstream signaling pathways will be crucial for the development of alternative or conjunctive therapies to treat FXS. In this case, it will be of special interest to analyze whether any components of these LTD-related signaling pathways are direct FMRP targets and thus translationally dysregulated in FXS, providing the rationale for a feedback inhibition mechanism (Fig. 1).

In the future, a detailed analysis of how mGluRs and other signals directly affect FMRP, and thereby influence the expression of specific proteins important for learning and memory, will be essential to develop a full therapeutic regimen. One way to fine-tune FMRP-mediated translational regulation at synapses might involve rapid, transient dephosphorylation of FMRP, as suggested by Narayanan *et al.*¹².

Notably, a recent study showed that mGluR5-dependent AMPA receptor internalization, a common feature of protein synthesis-dependent LTD, is elevated in FMRP-deficient neurons¹³ (Fig. 1). Intriguingly, at least four reported targets of FMRP—PSD95, MAP1B, APP and Arc/Arg3.1—have been implicated in the regulation of AMPA receptor trafficking and endocytosis^{14,15}, suggesting that reduction of mGluR activity may not be the only way to effectively treat FXS.

The study by Dolen *et al.*¹ not only provides an important test of the mGluR theory, but also shapes our thinking and guides us toward an effective therapy for fragile X syndrome.

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