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Title

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Permalink

Journal

ISSN

Authors

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Publication Date

2014

Copyright Information

Peer reviewed

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AMPA receptor modulation for enhancing plasticity and treating neuropathology
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AMPA receptor modulation for enhancing plasticity and treating neuropathology

by G. Lynch and C. M. Gall, *USA*

> Peripheral injection of an ampakine allowed monkeys to perform well beyond the level that could be achieved with weeks of training. Subsequent brain imaging studies showed that the drug produced a striking network effect: the monkeys engaged additional cortical areas while dealing with multiple, complex cues. These studies provide direct evidence that the network extension effect obtained in physiology experiments occurs in living animals and results in capacities beyond normal."

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*P*ositive allosteric modulators of α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate (AMPA)–type glutamate receptors ("ampakines" and functionally related compounds) constitute a relatively new class of psychoactive drugs that enhance fast, excitatory transmission in the brain. Because of this effect, ampakines reduce the threshold for inducing memory-related changes to synapses and improve learning in animals across species and paradigms. While most CNS drugs target neurons that project in a one-step, parallel fashion to a multitude of sites, ampakine-type agents act on the multiple connections found in serial brain networks. This results in a multiplier effect for the drug, likely to be most pronounced in the elaborate circuits found in the cortex, thereby intensifying cortical regulation of lower brain areas ("pharmacological encephalization"). Evidence that the compounds are effective in animal models of psychiatric disorders associated with abnormal brainstem activity is in agreement with this hypothesis. The possibility that expanding cortical networks will lead to cognitive, as opposed to memory, enhancement in normal brains is largely unexplored. Finally, positive modulators increase the production of brain growth factors that promote plasticity and neuronal viability; upregulation is associated with neuroprotection, growth, and improved functional outcomes in different disease models.

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*T*wenty years have passed since the introduction of peripherally administered compounds that positively modulate α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate (AMPA)–type glutamate receptors (ie, ampakines) and thereby rapidly enhance fast, excitatory transmissions in the brain.[1] The motivation for developing such drugs was relatively straightforward: increasing the size of postsynaptic responses should facilitate the opening of voltage sensitive N-methyl-D-aspartic acid (NMDA) receptors colocalized with AMPA receptors. Given that NMDA receptors trigger the learning-related long-term potentiation (LTP) effect, ampakines were expected to potently facilitate memory encoding.[2] These predictions have been confirmed by several groups. However, it quickly became evident that the drugs might have broader uses due to two effects, one obvious and the other not, that follow from augmented transmission at excitatory contacts. First, glutamatergic synapses mediate communication within the myriad networks used by the cortical telencephalon to regulate lower brain regions and to execute the dense computations underlying cognition. Positive modulators therefore have potential therapeutic applications in psychiatric disorders involving brainstem biogenic amine

systems[3] and, more speculatively, for enhancing cognition. Second, greater than normal excitatory postsynaptic currents (EPSCs) will increase the likelihood of cell discharges, an effect known to upregulate transcription of brain-derived neurotrophic factor (BDNF).[4] Such an effect raised the possibility of using ampakine-type drugs to treat brain damage.

These ideas received considerable preclinical support and, as might be expected, the positive modulator strategy evolved significantly over the subsequent two decades. This is an appropriate point to survey the status of the field and to consider possible future directions. This review begins with a brief description of the mode of action for ampakine-like drugs and then turns to specific applications. The issue of why, given their substantial success in animal studies, the compounds have not progressed further in clinical development will also be discussed.

Mode of action

The first, centrally active, positive AMPA receptor modulators were small benzamide structures ($\approx$300 Da)[5] that, with further development, resulted in two large subtypes. Later studies by other investigators, primarily at Servier, Eli Lilly, and Glaxo, generated pyridothiadiazine,[6] biarylpropylsulfonamide,[7] and 5'-alkyl-benzothiadiazide[8] families that similarly enhance AMPA receptor currents.[9] Compounds from each of these groups proved effective in various animal tests.

AMPA receptors are tetramers composed of several possible combinations of four homologous subunits (GluA1, GluA2, GluA3, and GluA4) each with RNA splicing variants ("flip" and "flop").[10] X-ray crystallography[11] and site-directed mutagenesis[12] work found that extracellular domains for individual GluA subunits create a V structure that contains the glutamate binding pocket. Notably, adjacent subunits form dimers (ie, two dimers per one tetrameric receptor). Ligand binding causes the extracellular V domains to converge and thereby trap the ligand; this generates tension on the transmembrane segment of the receptor and a shift to the channel open state. The V then reopens, releases the transmitter, and terminates ion flux, events referred to as "deactivation". However, binding can disrupt dimerization, leaving the V structures and the channel closed. This paradoxical (bound, but closed receptor) state is likely responsible for the pronounced desensitization of AMPA receptors following extended agonist treatment or repetitive

SELECTED ABBREVIATIONS AND ACRONYMS	
AMPA	α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate
BDNF	brain-derived neurotrophic factor
EPSC	excitatory postsynaptic current
EPSP	excitatory postsynaptic potential
LTP	long-term potentiation
NMDA	N-methyl-D-aspartic acid

high frequency transmitter release.[13] While it was commonly thought that desensitization terminates synaptic EPSCs, the decay phase of AMPA receptor–mediated synaptic currents appears to be governed by the rate of deactivation.[14] Notably, the rate constants for deactivation and desensitization are not greatly dissimilar, and routinely used experimental methods (eg, whole cell recording) can shift the balance between them in favor of the latter.[15] This is a point of some significance for the design of modulator experiments.

The binding pocket for ampakines was identified by x-ray crystallography of the receptor-drug complex. The pertinent site is found at the hinge of the extracellular V near the dimer interface,[16] a position that: (i) controls the kinetics for reopening of the V and transmitter release (deactivation); and (ii) maintains dimerization, thus preventing the receptor from becoming desensitized. These crystallography findings accord with physiological results showing that ampakines can slow in both deactivation and desensitization.[13] However, the relative effect on the two processes is variant-dependent; some modulators act predominantly on deactivation, others on desensitization, and still others on both. The first group increases the amplitude of synaptic responses while having minor effects on their duration (referred to as type A ampakines), while the second group (type B) causes a significant prolongation of the excitatory postsynaptic potential (EPSP) *(Figure 1, page 374)*.[17] As will be described, these two categories have substantially different functional effects.

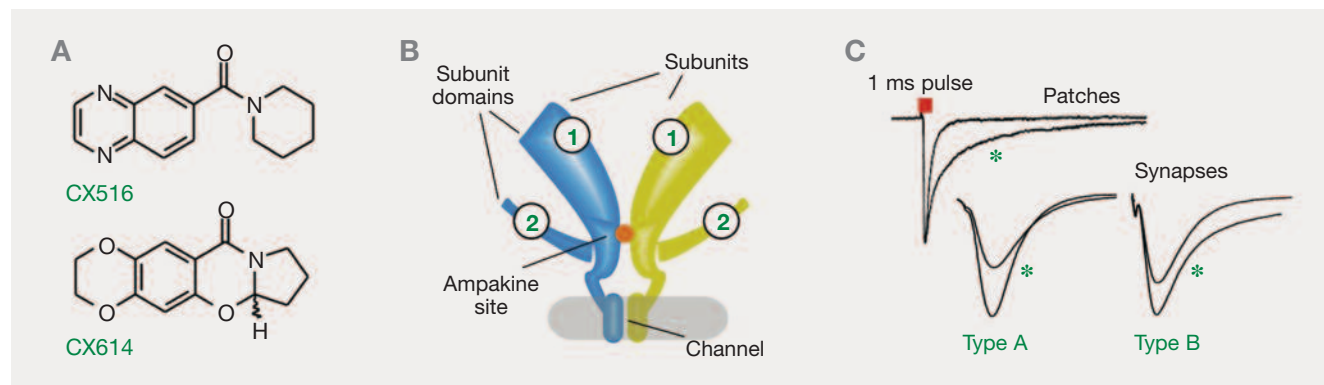## Enhancement of memory and cognition

◆ *Synaptic plasticity and memory*

Brain waves during learning in mammals, including humans, contain high levels of 4-8 Hz θ activity; recordings from individual neurons routinely detect multiple discharges (bursts) during the peaks of the rhythm. Remarkably, activation of inputs to the cells in a pattern that mimics θ bursts causes a marked and extremely persistent increase in the amplitude of EPSCs elicited by subsequent single stimulation pulses.[18,19] This long-term potentiation (LTP) effect is broadly accepted to be the substrate for many types of memory. If so, then there is a good possibility that enhancing LTP will produce corresponding effects on long-term memory. As mentioned, positive modulation of AMPA receptors potently facilitates the induction of LTP by reducing the voltage block on NMDA receptors, and there is now a large amount of literature showing that this is accompanied by the expected improvement in memory scores.

An overview of this material, the entirety of which cannot be adequately reviewed here, points to several general conclusions. A diverse array of positive modulators are effective across many different learning tests in animals and, where tested, without significant disturbances to behavior *(Figure 2, page 375)*.[20] Perhaps the most extensively tested single drug is Servier's S18986, which has positive effects on many hu-

Figure 1. *Mode of action for positive modulators of AMPA receptors.*
(A) Chemical structures for two functionally distinct, early stage ampakines (type A: CX516; type B: CX614). (B) The middle schematic illustrates the binding pocket for the drugs as deduced from site-directed mutagenesis and x-ray crystallography. Shown are two subunits of the tetrameric receptor. Note that each subunit has two extracellular domains ("1" and "2" in the schematic); the binding site for glutamate is found within the "V" formed by the two domains (not shown). The subunits form a dimer whose interface contains the ampakine pocket (orange circle). (C) Inward currents elicited by one-millisecond (ms) pulses of glutamate delivered to a patch excised from a hippocampal pyramidal cell. Note that application of an ampakine (green asterisk) greatly slows the recovery of the AMPA receptor–mediated current. This is the standard effect for positive modulators. Not shown here are effects in the presence of one-second glutamate pulses, during which the response quickly decays to 10% of its maximum value. Agents that block the decay are referred to as type B drugs; those that do not block the receptor desensitization, and yet produce the illustrated effect, are classified as type A. The bottom two traces show synaptic responses elicited by single stimulation pulses in a hippocampal slice. Type A ampakines increase the amplitude of field excitatory postsynaptic potential (fEPSP), while type B ampakines produce this effect, but also prolong the response.
Abbreviations: H, hydrogen; N, nitrogen; O, oxygen.
After reference 17: Lynch. Curr Opin Pharmacol. 2006;6:82-88. © 2006, Elsevier Ltd.

man-relevant memory types (including episodic, declarative, and relational[21]) and is effective after peripheral or central administration.[22] Work from other laboratories, using structurally different compounds, extends this list even further.[1,23,24] That positive modulators work in so many circumstances is perhaps surprising given that some forms of learning are not likely to be dependent on LTP.[25] This could indicate that drug effects on network processing, in these presumably non-LTP cases, facilitate attention and other processes required for acquisition, as opposed to acting directly on encoding mechanisms. Distinguishing between actions on processing vs encoding is thus a major issue with regard to interpreting memory enhancement effects.

Another noteworthy feature of memory enhancement with positive modulators is that it occurs in multiple species, including mice, rats, rabbits, and monkeys. This is encouraging with regard to human outcomes. The literature for humans is sparse, but at least one study, using an early, relatively weak ampakine, obtained evidence for enhancement in young, adult subjects.[26]

Most experiments searching for memory enhancement use a fixed amount of training followed by retention tests at some later time. These paradigms do not discriminate between accelerated learning vs strengthening of the memory trace. Acceleration has been established in a few cases, such as fear learning[27] and odor discrimination; in the latter, an ampakine reduced training requirements by almost 50%.[22,28] The strength of memory appears to have been explicitly tested only in a conditioning experiment in which ampakine-treated rats showed faster acquisition, but learned extinction proceeded at a normal rate, suggesting that the stability of the memory trace was not increased.[29] Additional work is needed on this topic, particularly with regard to type A vs type B drug variants. In all, an impressive body of work from many laboratories has confirmed the prediction that positive modulation will enhance memory in well-established learning paradigms. It will be of great interest to test for such effects in more cognitively demanding circumstances.[30]

◆ Network throughput and cognition

Positive AMPA receptor modulators are thus far the only agents known to enhance communication within cortical networks. Predictions about outcomes need to consider the high likelihood that the drugs act at many stages in serial circuits, something that is not the case for the great majority of psychoactive compounds. Serotoninergic neurons, for instance, do not string together to form long serial networks, but instead release more or less simultaneously at a very large number of scattered locations. The "serial sites of action" effect strongly suggests that the ultimate influence of an ampakine will depend upon the length and complexity of the network under study. Facilitation of transmission at one connection will lead to a greater number of cells that discharge at the next; repeated across many stages, each responding to the modulator, this would result in a multiplier effect for drug action. Confirmation was obtained in an experiment comparing the magnitude of EPSP increases at the first and third steps in a trisynaptic circuit that runs through the hippocampus: an ampakine concentration that produced a minor increase in synaptic responses at the initial step caused marked facilitation at the third.[31] Note that these arguments suggest that positive AMPA receptor modulation will have much greater effects in long networks, such as those found within the cortex, than in shorter ones.

The question then arises as to why the multiplier effect doesn't produce grossly enlarged EPSPs and seizures (notably, seizures are the primary unacceptable side effect of positive modulators). The likely reason that moderate concentrations do not cause epileptiform activity involves interneurons: enhancement of glutamatergic synapses on these inhibitory cells results in a slightly delayed suppression of further firing by projection neurons. In all, ampakines produce a potent, but brief, facilitation of rapid communication along a sequence of excitatory connections, thereby allowing a modest input to generate a sizable circuit output. Computational studies using integrated, multi-scale networks have provided a formal description of how enhanced excitatory transmission can lead to new and functionally useful outcomes from complex cortical systems.[32]

Could such effects enhance cognition in the sense of adding capabilities in normal humans? The only work addressing the issue used nonhuman primates and a very difficult problem. Peripheral injection of an ampakine allowed monkeys to perform well beyond the level that could be achieved with weeks of training. Subsequent brain imaging studies showed that the drug produced a striking network effect: the monkeys engaged additional cortical areas while dealing with multiple, complex
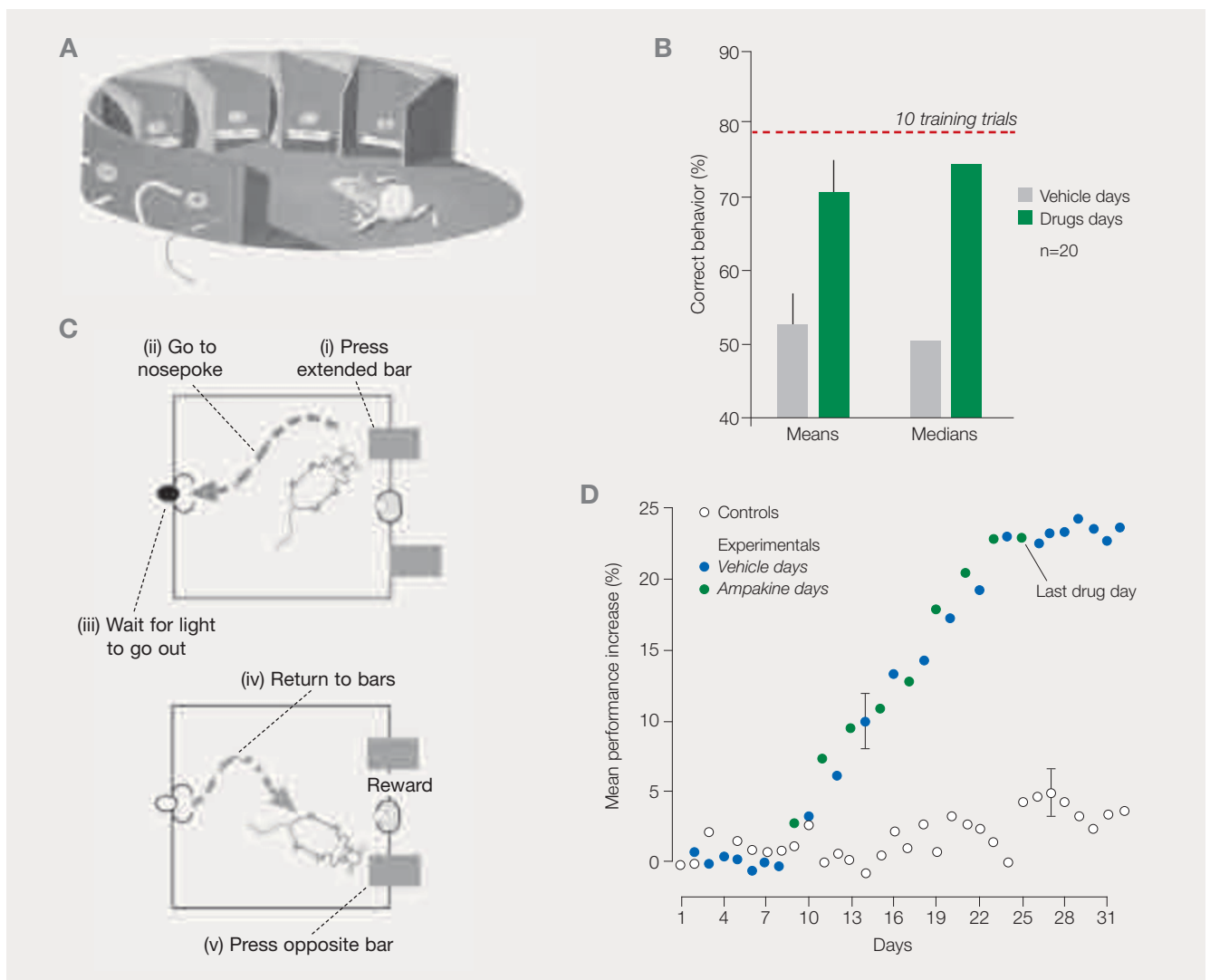
Figure 2. *Memory enhancement in two very different learning paradigms.*
(A) Well-trained rats were placed in an arena in which two novel odors were ejected simultaneously from any of six locations randomized from trial to trial. (B) Vehicle injected animals require 10 trials to form stable memory, as assessed on tests given 1 to 3 days later (red line). Five training trials produce chance scores on long-term retention (gray bars). However, the same animals showed robust memory when given a pretraining injection of an ampakine (green bars). (C) Enhancement in a complex, delayed nonmatch to sample task. Rats were trained for weeks to master a problem in which they were required to learn the following sequence: (i) press a bar to receive a small reward; (ii) move to the opposite side of test arena and perform a nose poke that activated a light; (iii) remain at that position until the light extinguished; (iv) return to the original location where two bars were now presented; and (v) press the bar that had not been originally selected. (D) As expected from the overtraining, control rats (open circles) did not improve over three additional weeks of testing. Rats given injections of an ampakine every other day (green circles) showed a steady increase in correct responses over days and this persisted after the end of drug treatment. These results[22] suggest that the ampakine enabled the animals to acquire novel information about the testing paradigm.
After reference 20: Hampson et al. J Neurosci. 1998;18:2748-2763. © 1998, Society for Neuroscience.

cues.[33] These studies provide direct evidence that the network extension effect obtained in physiology experiments occurs in living animals and results in capacities beyond normal.

◆ *Future directions*

An intriguing complexity to the ampakine/LTP interaction was introduced by the discovery that drug variants causing increases in EPSC amplitude only (type A) simply lower the threshold for potentiation, whereas those that increase both amplitude and duration (type B) produce this effect, but also elevate the LTP ceiling.[34] Both classes improve memory scores, but tests for behavioral differences are lacking. However, recent work describing new LTP timing rules suggests interesting possibilities. Those experiments showed that hippocampal LTP exhibits an analogue of the "spaced trials effect" (wherein retention is much better after multiple, temporally separated training sessions than after a single massed trial) that is a fundamental feature of learning. Specifically, a second episode of θ burst stimulation doubled the magnitude of LTP, but only when delivered with a one hour delay after a first train. This phenomenon appears to depend on the presence of a large population of synapses with high plasticity thresholds.[35,36] Pertinent to the present discussion, a type B ampakine caused a twofold increase in LTP after the first θ burst stimulation, but a subsequent train delayed by one hour produced no further enhancement. In essence, the drug removed the need for spaced trials to produce maximum potentiation. These results make clear predictions regarding behavior: (i) type A ampakines (which do not affect the LTP ceiling) will work within the distributed practice rules to reduce the amount of training needed to achieve stable memory; and (ii) type B compounds will eliminate the spaced trials effect by producing maximal encoding in a single training session.

AMPA receptor modulators and neuropsychiatric disorders

The interaction between the magnitude of modulator effects and network size is expected to produce a greater influence on the cortex than the lower brain. This would result in a kind of "pharmacological encephalization" in which the ampakine-influenced cortex exerts greater than normal regulation of: (i) brainstem and hypothalamic operations; and (ii) ascending projections to the forebrain. Such an outcome has considerable therapeutic potential for the many psychiatric conditions in which aberrant activity in subcortical systems is a contributing factor. Experimental testing of the idea produced encouraging results. Early work showed that acute ampakine treatment suppresses stereotypic activity caused by methamphetamine.[37] Follow-on experiments found that the ampakine increased neuronal activity in the cortex while depressing striatal fields,[38] thereby shifting cortico-subcortical balance in favor of the higher brain region. Excessive dopaminergic activity is widely thought to be a causal factor in schizophrenia, and antagonists of dopamine receptors are the most common treatment for the disease. It is of interest, then, that such

compounds act synergistically with ampakines in animal tests.[39] Unusual activity in ascending dopamine projections has also been implicated in attention deficit hyperactivity disorder (ADHD) and here again there is evidence that ampakines have positive effects. The compounds were found to produce dose-dependent suppression of hyperactivity in mice genetically modified to exhibit an ADHD-like syndrome.[40] Those studies were followed by successful phase 2 human trials using the type A ampakine CX717, developed in collaboration with Servier. Notably, a second group has recently reported positive results for attenuating locomotor activity in rats and reducing ADHD symptoms in a human trial.[41]

The pharmacological encephalization hypothesis suggests that enhanced cortical activity produced by ampakines will normalize activity in multiple ascending biogenic amine systems. Perhaps the most interesting test case involves serotonin, a transmitter intimately related to depression. Structurally diverse AMPA receptor modulators are multiply reported to be effective in different rodent models of the disorder.[17,42] Moreover, synergies with serotonin-based antidepressant medicines are evident: researchers at Eli Lilly found ampakine-like drugs cause a remarkable five- to tenfold leftward shift in dose-response curves for typical and atypical antidepressants in animal models.[43] Significant improvements have been described in at least one (modulator only) clinical study.[44]

◆ *Future directions*

The "pharmacological encephalization" hypothesis provides a perspective for considering potential applications of ampakines in a broad range of psychiatric disorders, but essential in vivo tests of the argument have not been reported. Such studies could involve conventional chronic recording techniques and are relatively straightforward. Beyond investigating the validity of a model with considerable explanatory power, the pertinent experiments could evaluate mechanisms (eg, serotonin and dopamine actions) that are likely to be homologous with those found in humans; the studies would thus be a valuable complement to behavioral screens in which rodent-human homologies remain uncertain. The synergies between ampakine-like drugs and two major classes of neuropsychiatric medicines (neuroleptics and antidepressants) call for further study. Approved treatments with the latter compounds produce negative psychological and/or neurological effects likely to be less prominent at lower dosages. Synergies might also serve to increase the safety margins of type B ampakines with regard to seizures.

Positive modulators and upregulation of brain growth factors

BDNF, a neurotrophin that is anterogradely transported and released from axon terminals,[45,46] promotes neuronal viability and facilitates learning-related synaptic modifications. Heightened levels of neuronal discharges cause a rapid increase in BDNF transcription,[47] leading to prolonged elevations in pro-

tein levels. These findings suggested that ampakine-mediated increases in excitatory drive would increase BDNF protein levels, an idea confirmed using peripheral injections.[4] Importantly, upregulation was induced by type B variants that have very short half-lives; this is in line with evidence that BDNF transcription responds quickly to increased firing and produces relatively stable products.[47,48] The following sections consider two classes of potential clinical applications for compounds that increase brain levels of BDNF.

◆ *Intellectual disability and memory loss*
Defects in LTP are found in animal models for each of nine, quite different, cases of intellectual disability or memory impairment so far tested.[30] Although these conditions have different etiologies (eg, stress, inflammation, aging, gene mutation, etc), the plasticity loss in each was traced to a shared end point failure in the cytoskeletal machinery responsible for LTP consolidation. There are reasons to suspect that increasing brain BDNF levels will circumvent this failure.

Induction of LTP by θ pattern stimulation activates multiple, small GTPase-initiated signaling pathways that trigger the assembly and later stabilization/elaboration of actin filament networks in the zone underlying excitatory synapses.[35] Manipulations that disrupt these events do not affect the initial expression of potentiation, but prevent its stabilization (consolidation) over the 10 to 15 minutes following induction. The actin signaling sequences involved in rapid LTP consolidation are regulated by multiple modulatory receptors found at synapses, prominent among which are tyrosine receptor kinase B (TrkB) receptors for BDNF.[35,49] As anticipated from this, brief infusions of the neurotrophin potently facilitate the production of LTP in normal animals.[50,51] They also rescue actin polymerization and LTP in models of late onset Huntington's disease (HD),[52] Fragile X syndrome,[53] and aging.[49] These encouraging findings set the stage for tests of whether chronic BDNF upregulation can be used as a general therapy for intellectual disability and memory impairment.

Several studies of this type have been conducted using 4 to 5 daily peripheral injections of an ampakine, which, despite a half-life of only several minutes, reliably increases BDNF levels in the hippocampus by 40% to 60%. Restoration of actin signaling and LTP was found in long-term ovarectomized rats[54] and in mouse models of both HD[55] and Angelman syndrome[56] *(Figure 3, page 378);* LTP recovery has also been described for middle-aged rats.[57] Where tested, learning defects were reduced or eliminated in conjunction with normalization of θ-driven actin filament assembly and LTP consolidation.[55,56] Notably, newly published work using small molecule BDNF receptor (TrkB) agonists[7,8] obtained positive results in rat aging.[58] Collectively, the above results suggest the possibility, using daily treatments of very short duration, of a therapeutic strategy with a broad spectrum of potential neuropsychiatric applications.

◆ *Degenerative diseases and brain damage*
While BDNF potently enhances activity-driven mechanisms underlying memory-related plasticity, it is more widely discussed with regard to positive actions on neuronal viability. The question that naturally arises in the latter context is whether the increased production elicited by positive modulation minimizes neuropathology. Several groups have reported positive tests of this idea. Experiments from Eli Lilly, using chronic peripheral administration, obtained a profound reduction in the degree of dopamine loss within the striatum in a rodent model of Parkinson's disease, a result that is most readily explained by extensive sprouting of remaining, intact axons.[59] Effects on the integrity of dopamine projections were accompanied by a pronounced improvement in locomotor scores. Of great clinical interest, these normalizing actions were also obtained when drug injections began two weeks after the destruction of dopaminergic neurons in brainstem.

A similar reduction in pathology was obtained in the R6/2 knock-in mouse model of early onset HD. These animals begin to exhibit motor symptoms at 3 to 4 weeks postnatal and then show progressively more intense signs of HD pathology in the striatum. Daily ampakine injections for seven weeks, starting at week three postnatal, substantially reduced multiple markers of the disease, including shrinkage of the striatum. This was accompanied by a striking improvement to near wild-type levels in motor functioning *(Figure 4 , page 379).*[60] Similar reductions in HD pathology in mouse models were recently described using peripheral administrations of a BDNF receptor agonist.[61]

Impressive evidence of neuroprotection associated with modulator-induced BDNF elevation was obtained by Servier researchers in a model of excitotoxic brain injury.[62] A parallel study using Servier compounds provided the first evidence that AMPA receptor modulators upregulate BDNF in the neonatal brain, and then showed that this effect is associated with a marked reduction of excitotoxic and inflammatory damage to the cortex.[63] Perinatal brain injury leads to profound clinical problems, and the results just described point to a novel, mechanism-based strategy for treating it.

Stroke is a leading cause of disability in adults, with treatment largely restricted to rehabilitative training. The positive effects of BDNF on learning-related plasticity suggested the possibility of using upregulation to enhance the beneficial effects of postinfarct practice. Positive results with a well-characterized animal model were recently described: daily peripheral injections with an ampakine, at dosages shown to substantially increase EPSPs, elevated cortical BDNF concentrations and markedly improved functional outcomes (motor performance) after six weeks of testing.[64] Critically, the ampakine-induced improvement was dependent upon BDNF: prolonged infusion into the peri-infarct zone of an extracellular BDNF scavenger eliminated the positive effects of drug treatment. This conclu-
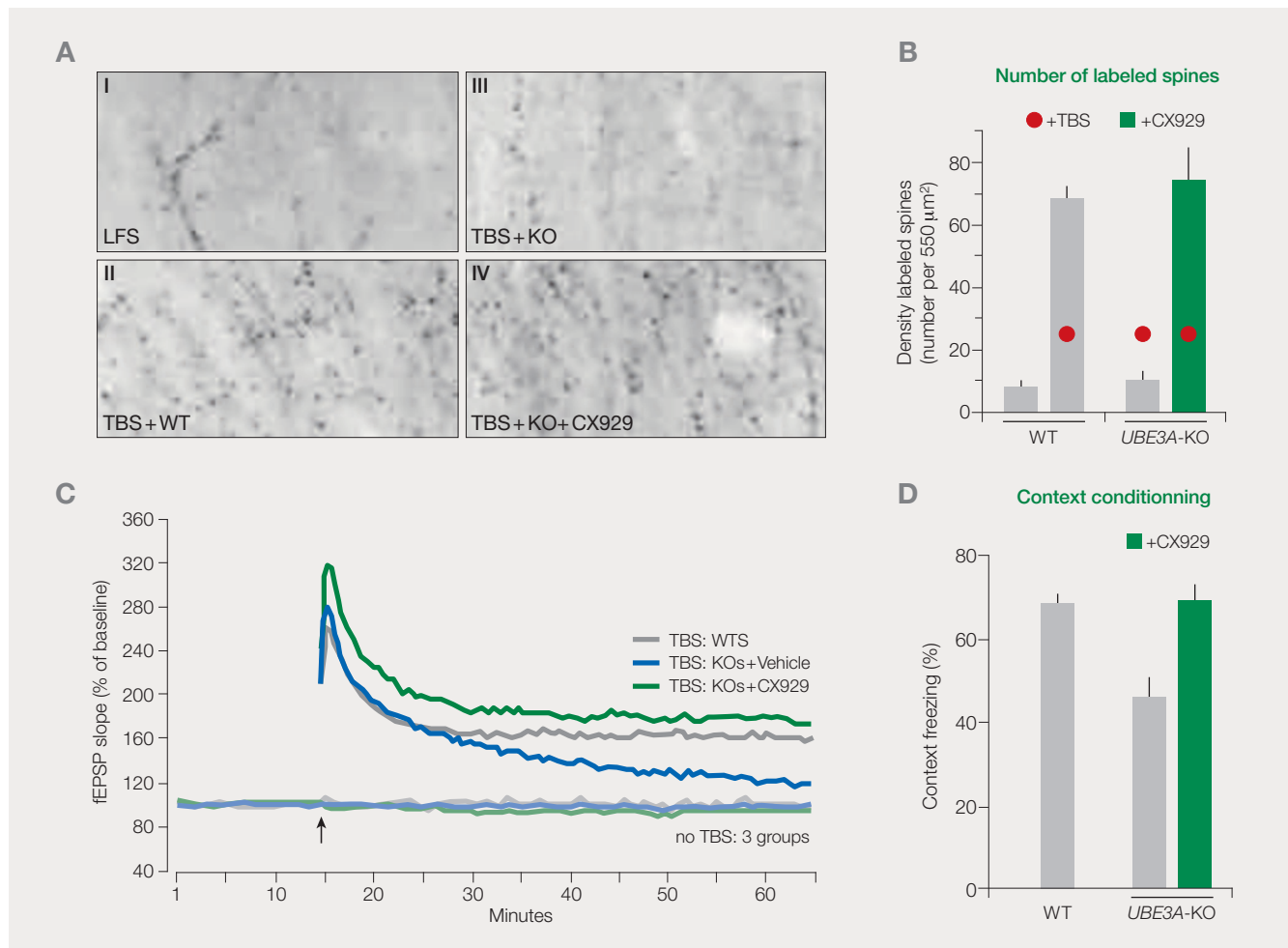
Figure 3. *Positive modulation of α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate (AMPA)–receptors rescues actin signaling, learning-related synaptic plasticity, and learning in a mouse model of Angelman syndrome.*[63]
(A) θ burst stimulation (TBS), which mimics neuronal activity occurring during learning, is known to cause actin polymerization in dendritic spines. (i) Low background labeling for spines with dense concentrations of polymerized action in hippocampus given low-frequency stimulation (LFS) of inputs. (ii) The number of labeled spines is markedly increased by TBS in wild type (WT) mice. (iii) However, TBS has little, if any, effect on spine actin polymerization in the UBE3A mutant mouse model of Angelman syndrome. (iv) Pretreating the mice with an ampakine (CX929) for 4 days completely restored TBS-driven actin polymerization in the mutants. (B) The positive modulator produced a full rescue of an essential step in producing learning-related synaptic plasticity. (C) As predicted from the actin studies, TBS does not cause stable increases in synaptic strength in vehicle-treated UBE3A–knock-out (KO) mice; four days of ampakine pretreatment restored long-term potentiation (LTP) in the mutants. (D) Angelman syndrome mice are defective in encoding long-term memory in a conventional behavioral task (context conditioning). This impairment is eliminated by the same four-day ampakine treatment that rescued actin signaling and LTP.
Abbreviation: fEPSP, field excitatory postsynaptic potential.
After reference 56: Baudry et al. Neurobiol Dis. 2012;47:210-215.© 2012, Elsevier Inc.

sion was reinforced by a demonstration that a type A ampakine did not elevate neurotrophin levels or improve stroke recovery. It seems clear from the selection of papers considered here that upregulating BDNF holds considerable promise for ameliorating the debilitating effects of brain injury.

◆ *Future directions*
Despite the relatively short time since the discovery that ampakines and allied compounds elevate brain BDNF levels, there is a now a surprisingly large amount of preclinical literature concerning the potential utility of the effect in treating intellectual disability. Missing from this is a description of how the treatments affect other, seemingly noncognitive components of syndromes associated with intellectual disability. How-

ever, a recent study in a mouse strain that exhibits many of the diagnostic features of autism, showed that daily injections with a type B compound reduced disturbances in social interaction.[65] More work of this type is needed to arrive at a reasonable appraisal of the broad utility of the ampakine/BDNF strategy for treating concomitants of intellectual disability.

This section of the review, dealing as it does with the most severe of conditions (degenerative diseases and brain injury), brings translational issues to the forefront. It does not appear from broadly available reports that any positive modulator has progressed to phase 3 trials, despite encouraging results in earlier stages of testing. There are likely to be several reasons for this. First, type B compounds (ie, those that increase both
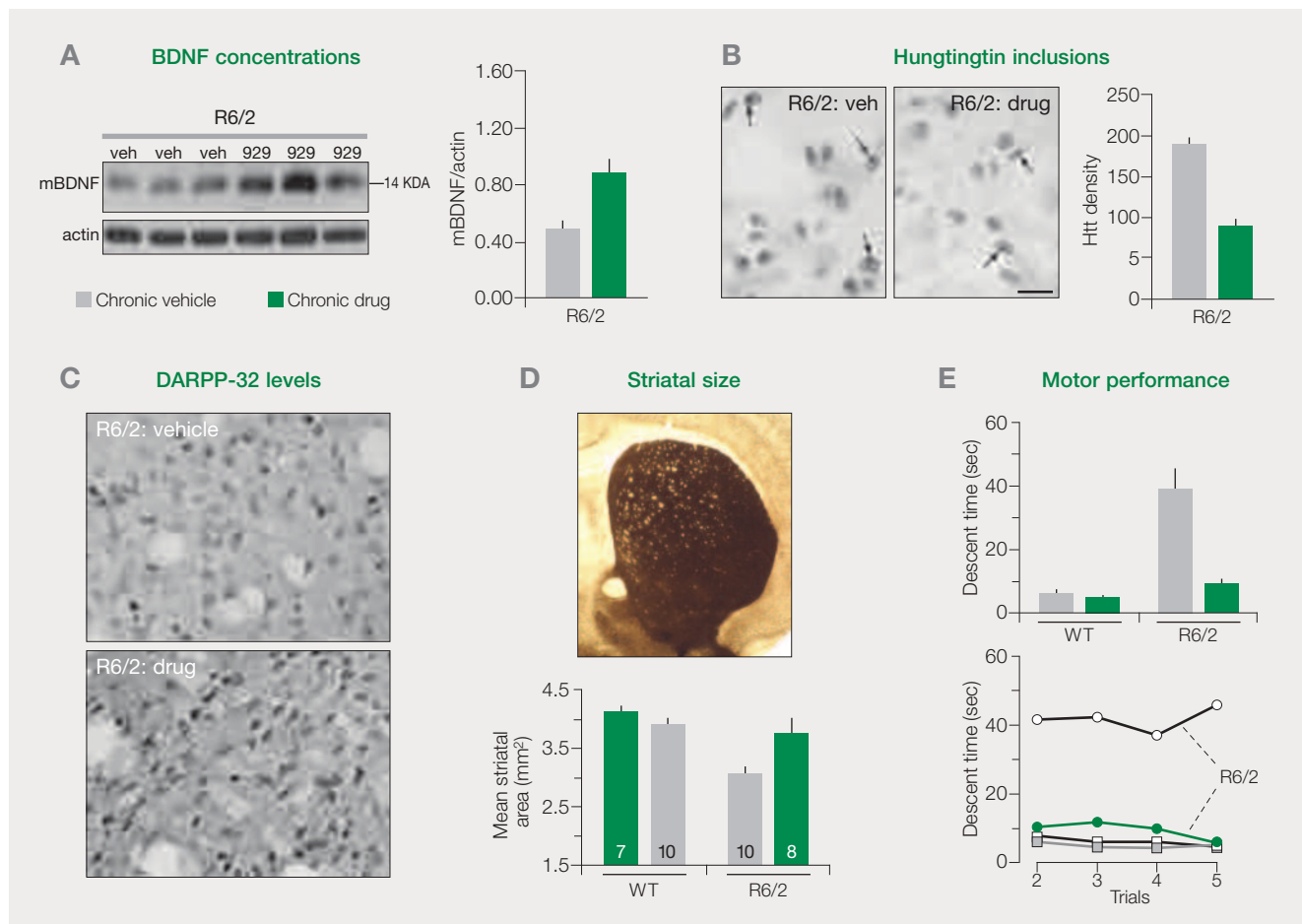
Figure 4. *Chronic treatment with an ampakine reduces pathology and restores motor functioning in a mouse model of early onset Huntington's disease (HD).*

R6/2 transgenic mice, overexpressing a fragment of mutant huntingtin protein, were given 7 weeks of daily vehicle (veh) or ampakine injections beginning at 3 weeks postnatal, when HD symptoms first appear. (A) The ampakine (CX929) treatments substantially elevated concentrations of mature brain-derived neurotrophic factor (mBDNF) in the striatum, the primary region showing HD-related pathology in the mutants. (B) A key feature of HD is the accumulation of huntingtin protein aggregates (Htt) in striatal neuronal nuclei (arrows). This effect is substantially reduced by chronic administration of the drug. (C) Another characteristic of HD is a sizable loss of the kinase, dopamine- and cAMP-regulated neuronal phosphoprotein (DARPP-32), from striatal neurons. As shown, the 7-week treatment with the positive modulator restored striatal DARPP-32 in R6/2 mice to normal levels. (D) Image of a coronal section through mouse forebrain shows the striatum densely labeled with acetylcholinesterase histochemistry. HD reduces striatal volume, an effect that is also evident in the R6/2 mouse by postnatal week 10. The ampakine (weeks 3 to 10) blocked shrinkage, yielding near wild type (WT) mean area measures. (E) R6/2 mice have a severe motor deficit in a "pole descent" test. The upper panel shows mean results for a set of descent attempts; note that chronic drug treatment caused a dramatic improvement in the mutants. Trial-by-trial analysis showed that vehicle injected R6/2 mice (open circles) do not improve with practice; mutants that received the drug (green filled circles) were indistinguishable from WTs (boxes) at the end of testing.

After reference 60: Simmons et al. Neurobiol Dis. *2011;41:436-444. © 2010, Elsevier Inc.*

the amplitude and duration of EPSPs) are generally more effective across indications and for elevating BDNF than type A variants (ie, those that affect amplitude only), but are more likely to cause seizures. Developing versions of these type B compounds with an acceptable safety margin may have been a major barrier to translation. Second, as is not uncommon, promising drugs can have side effects unrelated to their primary mode of action; this seems to be the case for an ampakine that proved effective in phase 2 ADHD work. Third, in view of the many potential indications being discussed, it is difficult to arrive at general conclusions regarding tolerable side effects, required efficacy levels, and safety margins; results for one situation may not apply to others. Additional points relevant to translation are noted below.

Final comments

The large body of work on positive AMPA receptor modulators establishes the perhaps surprising point that increasing the strength of fast excitatory transmission does not disturb a broad range of brain operations. It does, however, produce a number of beneficial effects either directly, via improved neuronal communication, or indirectly through increased growth factor production. Despite the many studies documenting these points, the field cannot be viewed as being well structured; put simply, it lacks broadly accepted guidelines and fundamental principles. A prominent example of this can be seen in the absence of data on how large an increase in EPSPs (amplitude and/or duration) is needed to produce a targeted functional outcome. Currently, efficacy is considered

only in terms of conventional dose-response curves with no discussion of neurobiological mechanisms, despite there being a clear primary mode of action. Discrepancies between effective doses for learning vs EPSP enhancement would have a major impact on further development of a candidate modulator, because it would point to the presence of unknown factors that regulate outcome potency.

There are also multiple reasons (eg, variations in AMPA receptor composition), and some evidence, to assume that ampakine variants are differentially effective across brain systems.[66] This constitutes a major barrier to the rational design of compounds intended for particular indications, since a drug could be exerting its greatest effects outside areas responsible for the desired effect. What seems called for is a type of regional "mapping" enterprise, perhaps using activity-dependent gene expression, to identify brain regions most affected by a candidate agent. Routine construction of such a map, during drug screening, was not a realistic possibility in the recent past, but advances in computer technology and the advent of automated microscopes suggest that it is now entirely fea-

sible. Note that maps would not only help explain differential behavioral effects, but could also point to unsuspected applications. Importantly, emerging methods not only allow for the analysis of regional actions, but also effects on interneuron-projection cell connections that constitute local circuits. Such information will likely prove critical for identifying agents that increase network throughput without greatly increasing the likelihood of seizures.

There is also considerable confusion about the duration of drug action needed to initiate BDNF production. Much of the work with animal models used an ampakine with a half-life of less than 15 minutes, and yet caused substantial increases in BDNF and positive effects in several animal models of intellectual disability. This raises the possibility that minimizing drug half-life might be a useful step towards reducing side effects of type B compounds. To conclude, while much has been accomplished, further progress will likely benefit from recognition of the unique complexities created by increasing excitatory transmission and the implementation of new analytical tools. ■

*G*ary Lynch *received his doctorate from Princeton University and then moved to the University of California, Irvine, where he is currently a Professor (Above-Scale). His early work produced foundational discoveries concerning two aspects of brain plasticity: growth after damage (axon sprouting) and learning-related synaptic modifications (long-term potentiation [LTP]). A search for drugs to enhance LTP and memory led to the invention of the first peripherally administered compounds (ampakines) to selectively enhance communication in cortical networks. Subsequent collaboration with Professor Gall revealed that ampakines increase transcription of a potent brain growth factor. Studies from their laboratories, and elsewhere, have shown that ampakines and closely related drugs have positive effects in animal models of psychiatric disorders and neuropathological conditions. Professor Lynch has recently described the synaptic chemistry responsible for LTP, and has shown that it is engaged by learning and is defective in many animal models of intellectual disability.*

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MODULATION DU RÉCEPTEUR AMPA POUR STIMULER LA PLASTICITÉ ET TRAITER LES PROCESSUS NEUROPATHOLOGIQUES

Les modulateurs positifs allostériques des récepteurs au glutamate AMPA (α-amino-3-hydroxyl-5-méthyl-4-isoxazole-propionate), les « ampakines » et composés fonctionnellement reliés, représentent une classe relativement nouvelle de médicaments psychoactifs qui favorisent une transmission cérébrale rapide des signaux synaptiques excitatoires. En raison de cet effet, les ampakines diminuent le seuil d'induction des modifications synaptiques liées à la mémoire et améliorent l'apprentissage parmi les espèces et les modèles animaux. Alors que la plupart des médicaments du système nerveux central agissent sur les neurones qui se projettent avec une seule connexion, de façon parallèle vers une multitude de sites, ceux du type ampakine agissent sur les nombreuses connexions des réseaux cérébraux en série. Ceci a pour conséquence de multiplier les effets du médicament, qui tendront de ce fait à plus prononcés dans les circuits corticaux complexes, intensifiant donc la régulation corticale induite par les aires cérébrales d'aval (« encéphalisation pharmacologique »). L'efficacité avérée de ces agents dans les modèles animaux de troubles psychiatriques associés à une activité anormale du tronc cérébral conforte cette hypothèse. Des réseaux corticaux étendus pourraient stimuler la fonction cognitive, et non mnésique, des cerveaux normaux mais cette éventualité est peu étudiée. Enfin, des modulateurs positifs augmentent la production cérébrale de facteurs de croissance favorisant la plasticité et la viabilité neuronales ; une régulation positive s'associe à la croissance, à la neuroprotection, et à des résultats fonctionnels améliorés dans différents modèles lésionnels.