Anticonvulsant Drugs Are Neuronal Network-Modifying Agents (NMAs)

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Introduction

Despite decades of research, it is still not really understood precisely how anticonvulsant drugs act to control the epilepsies, which are chronic neurological disorders. The acute actions of anticonvulsants have been evaluated on isolated neurons in vitro, which is the standard approach to defining mechanisms of action. These acutely determined mechanisms may help explain the ability to stop ongoing seizures, which likely contribute to the ability of chronically-administered anticonvulsants to prevent seizures. However, translating these findings from the dish (in vitro) to the intact brain is problematic. A major reason for this quandary is the complexity of the human brain, which contains billions of neurons and trillions of synapses. A true understanding of the relevant actions of these drugs requires studying neurons in the intact brain that are located in brain regions that are important to the generation of the seizures using therapeutic doses of the anticonvulsants. Neuronal networks play important roles in all seizures, including those once considered to be strictly localized (focal) in nature. It is becoming clear that neuronal networks in the intact brain have the capability to exhibit “additional” properties that arise from neuronal interactions between nuclei within the network that may not be present in isolated neurons even from the same brain structure (Faingold, 2004; Faingold and Blumenfeld, 2015). These “new” properties that arise from the intact network are called “emergent properties,” and recent findings indicate that anticonvulsant drugs can act selectively on these properties to exert their therapeutic effects (Faingold, 2014b; Faingold and Blumenfeld, 2015). Therefore, agents that are currently classified as anticonvulsants are more accurately termed neuronal network-modifying agents (NMAs) (Faingold, 2009). A major reason for using this drug classification is that many anticonvulsant NMAs are also effective in chronic pain disorders as well as a number of neuropsychiatric disorders, such as anxiety, obesity, alcoholism, migraine, and mania. Each of these disorders is mediated by a neuronal network in the brain (Faingold and Blumenfeld, 2014a).

Background

Approaches to Understanding CNS Drug Mechanisms

Early in the development of CNS drugs, investigators proposed that most agents exert therapeutic effects by selective actions on neurons in specific brain sites rather than by acting globally on all neurons. An example of this earlier neuropharmacology paradigm is the theory that sedative-hypnotic drugs produce sleep by selective actions on the brainstem reticular formation, a wakefulness center, which was a key element in the “centrencephalon,” an early example of what we now call a neuronal network (Penfield and Jasper, 1954). This theory was never disproven and has actually been refined in recent studies (Sukhotinsky and Devor, 2014). However, for a number of years this approach was superseded by another paradigm, based on the fact that most CNS drugs affect neurotransmitter receptors or ion channels, mechanisms that had been identified by studying isolated neurons in vitro. Subsequent work has shown that the selective effects seen in in vitro studies are not always seen in the same neurons in the intact animal in vivo, as discussed below. Such inconsistencies have emphasized that the neuronal network approach may yield a more accurate and complete understanding of CNS drug action, especially since it has become well-established that neurological and neuropsychiatric diseases are mediated by neuronal networks rather than single CNS neurons.
in one locus within the brain (Faingold and Blumenfeld, 2014a). The inconsistencies between actions of drugs in vivo versus in vitro can be explained by the fact that neurons within networks may express additional emergent properties at which drugs can potentially act. These emergent properties may be absent from the same neurons in vitro, because important influences on these neurons are lost when the neurons are isolated (see Fig. 5).

**Network Perspective on Brain Function**

The many complex functions that the brain performs involve many different distributed neuronal networks. The term neuronal network has been used in a wide variety of ways and in different contexts (Stam, 2014; Faingold and Blumenfeld, 2014a). This review will concentrate on relatively large-scale networks, each of which consists of several distinct brain regions. These networks were defined using direct recordings of neuronal action potentials within specific brain sites (network hubs) that were initially identified, using neuroanatomically-based techniques, such as neuroimaging (Faingold and Blumenfeld, 2015). For example, a comprehensive network has been identified that controls normal locomotion, and another distinct widespread network mediates hearing (Jordan and Slawińska, 2014; Brozoski and Bauer, 2014). Neuronal network connections can be strengthened and, in unusual cases, new ones can even be formed, which modify brain function, both positively, as seen in learning (Schafe, 2014) and negatively, as seen in epilepsy (N’Gouemo et al., 2014). Many other brain disorders involve disrupted neuronal network functions, as seen in degenerative diseases, such as Parkinson’s disease (Lakraj et al., 2014). Other brain disorders are due to the formation of abnormal connectivity within neuronal networks due to neuroplasticity, for example fiber sprouting and other network expansion mechanisms that occur in several chronic forms of epilepsy (N’Gouemo et al., 2014).

Although neuroanatomical studies have identified essential details about the connections between brain neurons, a functional neuronal network can greatly differ from the neuroanatomy of that pathway. Functional studies indicate that a significant proportion of anatomical connections, when activated physiologically, only result in subthreshold responses, limiting the activation of many neurons in the network. Such subthreshold responsiveness is seen to an extensive degree in non-primary sensory and motor brain regions in areas of the brain termed "conditional multi-responsive" (CMR) regions (Faingold, 2008; Faingold et al., 2014b). CMR brain regions are particularly important in the ability of the brain networks to undergo functional changes. These subthreshold responses, which are so predominant in CMR brain regions, can reach threshold under pathophysiological conditions, resulting in the activation of a specific disease network. Such findings have led to the idea that CNS disorders are largely governed by what could be termed the “Law of Conservation of Networks” (Faingold, 2004). That is, the pathophysiology of CNS disorders is mediated largely by normal neuronal networks that are interacting abnormally with other normal networks. For example, an epilepsy model called audiogenic seizures (AGS), described in detail below, involves an aberrant interaction between the normal auditory network and the normal network for locomotion to produce the bilaterally symmetrical motor (convulsive) seizure in response to intense acoustic stimuli. Thus, the convulsion occurs due to excessive stimulation of the auditory pathway, which extensively activates the locomotor network that is only transiently activated by acoustic stimuli under normal conditions, as seen in the acoustic startle response (Faingold and Tupal, 2014). Multiple network interactions can also occur, such as the interaction of the auditory and locomotor network seen in AGS, which subsequently interacts importantly with the respiratory network in certain AGS models. This additional network interaction mediates seizure-induced respiratory arrest, leading to death that is seen in DBA/1 and DBA/2 mouse models of sudden unexpected death in epilepsy (SUDEP), which is a major problem in human epilepsy (Faingold and Tupal, 2014; Faingold et al., 2015; Thurman et al., 2014). Many mechanisms are known to control network function (Table 1), including volume transmission of neuroactive agents, such as CNS drugs, mediated by diffusion from blood vessels and via the cerebrospinal and extracellular fluid (Agnati et al., 2014). Volume transmission is especially important for all systemically-administered CNS drugs, such as anticonvulsant NMAs.

**Networks in Epilepsy**

As noted above, all forms of epilepsy involve a distributed epileptic network rather than just abnormal neurons in one brain location (focus). This view is based on studies of small networks in vitro, animal models of temporal lobe epilepsy, human intracranial EEG, and human and animal neuroimaging. Neuroimaging is one of the currently most useful approaches to networks. However, the minimum time scale that is achievable with neuroimaging is too great to observe neuronal firing changes that occur during a seizure, which is usually relatively brief. Neuroimaging often does not indicate the nature (inhibition or excitation) of neuronal firing changes, which are the critical events underlying elaboration of the seizure (Faingold and Blumenfeld, 2014c). Although neuroimaging can be very useful for correlating anatomical and physiological changes associated with neurological or psychiatric disorders, neuroimaging data cannot determine if the brain regions that exhibit changes are requisite, ancillary, or compensatory to the disease process (Faingold and Blumenfeld, 2015). Direct observation of neuronal firing changes recorded with extracellular micro-electrodes in behaving organisms that occur before and during the seizure or other behavioral manifestation of the disorder allows observation at a millisecond time scale of events. This provides details about the occurrence of excitation or inhibition during the manifestation as well as after the event, such as during the post-ictal depression period seen after seizures. The neuronal recording approach allows observation of the dynamic functional changes that occur in each network nucleus. The effects of anticonvulsant NMAs on neuronal firing within the requisite network nuclei have further illuminated the mechanisms of network operation in epilepsy and provided important insights on the actions of these agents, as discussed in detail below.
Current Classification of Anticonvulsant NMA Mechanisms

The current mechanisms of anticonvulsant NMA action, which were largely determined in vitro, include, for the most part, effects on neurotransmitter actions or directly on ion channels (Bialer et al., 2015). Enhancement of the inhibitory action of gamma-aminobutyric acid (GABA) at GABAA receptors is thought to contribute importantly to the anticonvulsant effects of barbiturates, benzodiazepines, vigabatrin, and tiagabine, and this action may contribute to the anticonvulsant effects of valproic acid and gabapentin. Use-dependent block of sodium channels is another important anticonvulsant drug mechanism for phenytoin, carbamazepine, oxcarbazepine, rufinamide, lacosamide, and lamotrigine. Opening of potassium channels by ezogabine is a newer anticonvulsant drug mechanism. Antagonism of specific types of calcium channels is another mechanism of anticonvulsant NMA action, including ethosuximide, gabapentin, and pregabalin. Blockade of glutamate receptors has also been observed with perampanel, barbiturates, felbamate, and lamotrigine. Many anticonvulsant NMAs exert multiple actions when examined at the single cell level in vitro. However, the actions of these anticonvulsant NMAs have received minimal evaluation in seizure networks, and the limited seizure network research that has been done has shown differential effects on neurons within specific nuclei of the networks, as discussed below. Such differential effects may be particularly relevant to therapeutic mechanisms when the doses given to the intact animal do not greatly exceed those required to produce anticonvulsant effects in the same animal. By limiting the doses the non-therapeutic (adverse) effects that are exerted by all anticonvulsants in excessive doses can be reduced or eliminated, as discussed below. Many anticonvulsant NMAs exert multiple mechanisms of action in vitro, but one of these mechanisms may actually predominate in vivo at therapeutic doses. Interestingly, even within the same brain structure anticonvulsant drugs that block sodium channels only inhibit neurons in the excitatory subnetwork and do not reduce the activity of neurons in the inhibitory subnetwork of the same structure at the same doses (Pothmann et al., 2014). These findings suggest that these excitatory neurons possess an emergent property that is not present on the inhibitory neurons (Faingold and Blumenfeld, 2015), despite the importance of sodium channels in generation of action potentials in both types of neurons.

Audiogenic Seizure Models

Seizure models in animals have played a key role in the discovery of all antiepileptic drugs. In vitro testing cannot replace animal models, because it cannot model the entire spectrum of pharmacodynamic actions required for seizure blockade, does not provide information about the pharmacokinetics that occur in intact organisms, and obscures potentially important toxicological effects that may occur in the intact animal.

Only a few epilepsy models have well-established neuronal networks (Faingold and Blumenfeld, 2014c), and this review will emphasize a well-defined network in an inherited epilepsy model, since genetics is well-known to play an important role in a significant percentage of human epilepsies. Genetically epilepsy-prone rats (GEPRs), which model generalized convulsive seizures, are
abnormally sensitive to many seizure induction methods, including intense acoustic stimuli, which induce audiogenic seizures (AGS) (Jobe et al., 1992; Faingold and Naritoku, 1992). There are numerous naturally-occurring rodent forms of AGS, and a number of gene knockout mouse strains also exhibit AGS susceptibility. AGS can also be readily induced in rodents by several different treatments (Faingold et al., 2014a). In all forms of AGS, high intensity acoustic stimulation results in ictal behaviors that begin with wild running and progress to generalized (all-limb) clonus or tonus. Under conditions of seizure repetition the tonus can be followed by post- tonic generalized clonus, closely mimicking human tonic-clonic seizures, as discussed below. The terminal convulsive behavior is followed by post-ictal depression of behavior, including loss of the righting reflex, like that seen in human generalized tonic-clonic seizures in humans. In human epileptic patients, generalized clonus and tonus occur, but these convulsions are rarely evoked by acoustic stimuli, although they may be evoked by visual stimuli. This sensory difference between humans and rodents may be due to the fact that rodents, unlike humans, are nocturnal and rely less than humans do on the visual network and rely more on other senses, especially the auditory network. Many anticonvulsant NMAs have been developed using AGS models, and the anticonvulsant effect of levetiracetam was initially observed in an AGS model but not in the models that were standard for drug screening at the time.

**Neuronal Network for Audiogenic Seizures**

Inherited AGS models provide specific experimental advantages for network determination and functional studies. No invasive techniques are needed to induce the seizure, and there is the ability to precisely control the seizure-inducing stimulus, which allows external control of seizure induction and the ability to examine responses to stimuli at intensities that are below as well as above the seizure threshold. Research on the neuroanatomy and neurophysiology of the auditory system is also well-developed (Bauer, 2014). Once it was realized that the locomotion network was also involved in the AGS network, this well-developed neuroscience knowledge base provided further important information (Jordan and Sławińska, 2014). This established neuroscience knowledge base is readily applicable to AGS, including potential candidate sites within the auditory and locomotor pathways to evaluate for involvement in AGS.

**Approaches to Seizure Networks**

The use of chronic stereotaxically implanted cannulae and neuronal recording electrodes to experimentally probe the AGS network in unanesthetized and behaving rats allows determination of the role of specific brain structures in the epilepsy network (Faingold and Blumenfeld, 2014b). Based on the ability to block AGS by inhibition of specific putative network sites, the requisite structures in the seizure network were determined. In subsequent experiments extracellular neuronal responses to acoustic stimuli were recorded initially at intensities below that which will initiate AGS and then at a seizure-inducing intensity. Neuronal firing and behavior were recorded on video (split-screen) simultaneously during AGS. The neuronal firing data were analyzed using post stimulus-time histograms, and quantified neuronal firing changes were statistically compared (Faingold, 2012).

**AGS Network Findings**

Using these approaches the inferior colliculus (IC) was established as the consensus seizure initiation site. The major defect that leads to AGS susceptibility in GEPRs is the reduced effectiveness of GABA-mediated acoustically-evoked inhibition in IC neurons that occurs at higher stimulus intensities in normal animals (Faingold, 2002; Faingold et al., 2014a). This reduced inhibition allows high intensity acoustic stimuli to induce excessive firing of IC neurons, which in turn results in abnormally intense activation of neurons in midbrain locomotor network sites to which the IC projects [deep layers of superior colliculus (DLSC), periaqueductal gray (PAG), pontine reticular formation (PRF)], and substantia nigra reticulata (SNr) (Fig. 1). Thus, excessive activity in the normal auditory network abnormally activates the locomotor network, which projects to the spinal cord and produces the bilaterally symmetrical convulsive behaviors.

The neuronal responsiveness to acoustic stimuli in each of the AGS network sites in GEPRs is significantly greater than in normal animals, even in response to stimuli below the seizure threshold (Faingold et al., 2014a). The firing patterns of neurons in the AGS network during the seizure change dramatically in each site. However, the patterns are consistently different in each site, as the seizure progresses, depending on the temporal sequence of convulsive behaviors that is occurring. Neuronal recording studies indicate that the seizure network operates hierarchically during AGS; that is, intense activation of each network nucleus occurs in a consistent and specific order, which precedes and likely initiates each convulsive behavior, as they appear sequentially (Fig. 2). Thus, IC neurons fire most intensely just prior to and during AGS initiation, while neurons in DLSC fire most intensely as the wild running begins. PAG and PRF neurons fire most intensely just prior to and during tonic convulsions, PRF and SNr neurons fire most intensely just prior to and during tonic hind limb extension. During post-ictal depression only neurons in the PRF and SNr remain active until recovery of the righting reflex, when neurons in all sites become active again. These findings suggest that the network hierarchy begins with IC dominance (greatest change from pre-seizural firing), but the DLSC becomes dominant during wild running. Then the PAG, PRF, and SNr become dominant during tonic behavior, while the PRF and SNr dominate during tonic hind limb extension.
Anticonvulsant NMA Action on the AGS Network

As noted above, abnormal acoustically-evoked neuronal firing prior to seizure occurs in all AGS network nuclei of the GEPR, and the effects of several anticonvulsant drugs on these elevated neuronal responses were evaluated in these seizure nuclei network in awake, behaving GEPRs. Several of the anticonvulsant NMAs examined (at doses that effectively reduce AGS) exerted differential effects on firing of neurons in each network nucleus (see Fig. 3). Since this network operates hierarchically, this suggests that the site in the network at or closest to the seizure initiation site in IC is likely to be a critical site for drug action when it is administered at the lowest effective dose and acts as the critical therapeutic target for each specific anticonvulsant NMA.

For example, competitive N-methyl-D-aspartate (NMDA) receptor antagonists are effective anticonvulsants against many forms of experimental seizures, including AGS in GEPRs. Systemic administration of these agents at anticonvulsant doses, prior to seizure induction, results in significant reductions of IC neuronal responses to acoustic stimuli at all stimulus intensities, suggesting that competitive NMDA receptor antagonists act in the AGS network at or afferent to the IC. However, an uncompetitive NMDA receptor antagonist, MK-801 (dizocilpine), does not alter the responses of IC neurons, despite the fact that this agent blocks AGS in very low doses (Faingold, 2014b).

Tiagabine blocks the GABA transporter, prolonging its action. Systemically administered tiagabine blocked AGS in GEPRs, but unlike the competitive NMDA receptor antagonists, significant IC neuronal firing reduction prior to seizure was seen only at high acoustic intensities. The time course of the reduced IC neuronal firing paralleled that of AGS suppression with tiagabine, suggesting that tiagabine enhanced that form of GABA_A receptor-mediated inhibition in IC neurons that is prominent at high acoustic intensities (Faingold, 2002). However, other anticonvulsant drugs that block AGS, including phenytoin, do not affect IC neuronal responses to acoustic stimuli, indicating that their actions are exerted at network sites efferent to the IC.

The PAG and PRF are implicated in generation of the tonic convulsive behaviors. AGS in GEPRs that culminate in tonic hind limb extension, and elevated acoustically-evoked rapid tonic and/or burst neuronal firing, respectively, immediately preceding...
tonic convulsions, have been observed in these brain sites. Phenytoin can block AGS completely, but lower doses of this agent can block the tonic phase of AGS selectively. Anticonvulsant doses of phenytoin induce consistent changes in PAG and PRF neuronal firing and behavior in GEPRs (Faingold, 2014b). Phenytoin in doses that selectively suppressed tonic convulsions did not consistently alter PAG neuronal responses to acoustic stimuli prior to seizure, but the same doses of phenytoin resulted in significant suppression of PRF pre-seizural acoustically-evoked neuronal firing and suppressed firing during seizure as well. Higher doses of phenytoin which completely blocked AGS, significantly reduced PAG acoustically-evoked neuronal firing and more greatly decreased PRF firing. These results suggest a critical role for PRF, but not PAG neurons, in generation of tonic convulsive behaviors of AGS. The suppression of PAG and PRF neuronal firing induced by phenytoin, associated with complete seizure blockade, is consistent with vital roles for both structures in the seizure network. The differential suppressive effect of phenytoin on both pre-seizural and seizural neuronal firing in PRF as compared to the lack of effect on PAG firing indicate that this experimental approach identified the most sensitive therapeutic target for the action of this anticonvulsant NMA. For an anticonvulsant NMA to completely block seizures, however, actions on multiple network sites, including the PRF and PAG in the GEPR, may be needed to significantly affect the emergent properties of the seizure network. Interestingly, as discussed in detail below, the PAG is also implicated in the pain network, and during the post-ictal period, it has been observed that analgesia lasting for hours occurs after seizures, including AGS in GEPRs. The PAG also plays a major role in this seizure-induced reduction in pain sensitivity, since focal blockade of this structure will reverse the post-ictal analgesia (Samineni et al., 2011).

Another agent with anticonvulsant actions, the uncompetitive NMDA antagonist, MK-801, did not consistently affect pre-seizural acoustically-evoked neuronal firing in IC, DLSC, PRF, or PAG, despite blocking AGS in GEPRs. However, MK-801 induced...
a significant increase of neuronal firing in SNr neurons concomitant with suppression of AGS (Fig. 4), indicating that SNr neurons are an important therapeutic target for this agent. In contrast, MK-801 induced minimal effect on neuronal firing when perfused directly onto SNr neurons in brain slices in response to a broad range of electrical stimuli and drug concentrations. These findings suggest that the significantly increased firing induced by systemically administered MK-801 is an effect on the emergent properties of the AGS network, critically involving SNr neurons (Faingold, 2004, 2014b). How can a drug produce a major effect on specific neurons in vivo, but not produce this effect on the same neurons in vitro? As noted above and detailed in the example in Fig. 5, there are a number of influences that control neuronal function. In the intact unanesthetized brain in vivo, all of these mechanisms are present. However, many of these mechanisms are absent and others may be greatly altered in the same neurons isolated in vitro (Faingold, 2014a).

If we examine the influences exerted on neurons in a specific brain site, such as the IC, we can see that there are numerous influences, some of which are shown in Fig. 5. When IC neurons are studied in in vitro brain slices, many of these influences are eliminated (eg, ascending and descending projections) or greatly modified (eg, ionic and O2 levels) (Fig. 5). Also, achieving an appropriate concentration of the drug in vitro is highly problematic. This is exemplified by the “amplification” phenomenon. If we examine the influences exerted on neurons in a specific brain site, such as the IC, we can see that there are numerous influences, some of which are shown in Fig. 5. When IC neurons are studied in in vitro brain slices, many of these influences are eliminated (eg, ascending and descending projections) or greatly modified (eg, ionic and O2 levels) (Fig. 5). Also, achieving an appropriate concentration of the drug in vitro is highly problematic. This is exemplified by the “amplification” phenomenon.

**Figure 3** Diagram of the neuronal network for audiogenic seizures (AGS) with emergent properties (indicated by cylinders) in each requisite site at which systemically-administered drugs that block these seizures may act at therapeutic doses. The network is organized as a hierarchy beginning with the acoustic stimulus (1) input into the auditory pathway (2–4), including neurons in these nuclei (up to the level of the inferior colliculus (IC) (5), which is the consensus seizure-initiating site. The IC projects to the brainstem locomotion network nuclei, including the deep layers of superior colliculus (DLSC) (6), projecting to the periaqueductal gray (PAG) (7) and substantia nigra reticulata (SNr) (8) and brainstem reticular formation (BRF) (9) which project to the spinal cord (10). The hierarchical activation of each requisite network nucleus produces the sequential behaviors of AGSz (wild running followed by tonic flexion and tonic extension) (11). (The critical structure that initiates each behavior is shown below the behavior.) Neurons in the BRF and SNr are the only regions that are active during the post-ictal behavioral depression that follows the tonic extension behavior. Therapeutic doses of several drugs with anticonvulsant properties act to inhibit neurons in the IC, including competitive (c-) NMDA antagonists, such as 2-amino-7-phosphonoheptanoate and GABA uptake inhibitor, tiagabine, as well as ethanol. Therapeutic doses of other drugs that are effectively anticonvulsant exert no effect on IC neurons, including an uncompetitive (uc-) NMDA antagonist (MK-801), gabapentin, lamotrigine, and phenytoin. Other effective anticonvulsant NMAs, such as gabapentin, act to reduce PAG neuronal firing. Therapeutic doses of phenytoin selectively act to inhibit neurons in the BRF of the pons. The SNr is the target of MK-801, but the effect is to increase neuronal firing. The emergent property of each nucleus is seen as a confluence of influences onto the neurons in each nucleus, including neuroactive substances (red squares) released onto the neurons via synaptic transmission (ST) and from the blood vessels, cerebrospinal and extracellular fluids via volume transmission (VT), as shown in the expanded diagram of an emergent property on the left of the network. Systemically administered drugs reach each site via VT. Direct stimulation of any site within the network, either chemically or electrically, can affect seizure susceptibility and may modify the emergent properties in the affected site. (CN, cochlear nucleus; SOC, superior olivary complex, which are auditory structures important for input to the IC). Reproduced from Faingold, C.L., Blumenfeld, H., July 6, 2015. Targeting neuronal networks with combined drug and stimulation paradigms guided by neuroimaging to treat brain disorders. Neuronsci. 7, 454-469 (Epub ahead of print) with permission.
that occurs in vivo, so that much smaller concentrations of drugs affecting seizures are needed in vivo as compared to in vitro to
obtain the same effect (Narahashi et al., 2007).

The realization that functioning brain networks often exhibit emergent properties (Faingold, 2014a) has led to the hypothesis
that these properties themselves are potentially critical targets for the action of CNS drugs (Faingold, 2004). When neurons are iso-
lated (in vitro) from their network, regional selectivity of drug action can be lost. In this situation, a neuron in vivo may be affected
by a given drug, but the same drug may not affect the same neuron in vitro, as seen with MK-801. Conversely, the emergent property
theory also suggests that a neuron may not be affected by a given drug in vivo, even if the same drug has clear effects on the same
neuron in vitro, which has been observed with ethanol. Thus, the network can actually alter the sensitivity of neurons to a drug, as
seen in data comparing the regional effects of ethanol in vivo versus in vitro (see Faingold, 2004).

Networks and CNS Pathophysiology

As noted above, CNS neuronal networks often express emergent properties. Specific elements within these networks may be a critical
therapeutic target on which CNS drugs, including anticonvulsant NMAs, exert their pharmacological effect. These emergent
properties of network neurons can result from the intensification of a specific receptor or channel or result from a critical interaction of certain of the many influences affecting these neurons in that specific site (Faingold, 2014a), causing these cells in this site to be a selective target for the therapeutic action of the specific anticonvulsant NMA. However, this emergent property may be absent in the same neurons when they are isolated in a brain slice or culture in vitro (Faingold, 2004, 2014a). The net drug effect in an intact organism is the sum of all the actions exerted on various levels of these networks by the drug, and may vary by dose, particularly with doses in the toxic range. It is well-established that neurological and psychiatric disorders are also mediated by networks of brain nuclei (Faingold and Blumenfeld, 2014a). A drug’s effect may be exerted on multiple networks or common elements of these networks, as discussed in detail below for the anticonvulsant NMA, gabapentin.

Anticonvulsant NMAs successfully treat seizure disorders, but many of these same drugs are also effective in treating chronic pain disorders and psychiatric diseases, such as anxiety disorders and bipolar disorder, as mentioned above. Each of these disorders is thought to be subserved by specific brain networks (Faingold and Blumenfeld, 2014a). The gate control theory of pain is an example of a prototypical neuronal network in the spinal cord (Melzack and Wall, 1965; Faingold and Tupal, 2014), and several techniques, including neuroimaging, have identified a neuromatrix that is activated in chronic pain syndromes (Roy et al., 2014; Sukhotinsky and Devor, 2014). Neuroimaging differences are seen between patients in pain as compared to pain-free individuals, and many of these differences are reversible with effective drug therapy. As noted above, many anticonvulsant NMAs effectively treat chronic pain syndromes. These agents may act by affecting emergent properties in specific sites within the neuronal network involved in the pathophysiology of pain.

**Anticonvulsants in Epilepsy and Pain Networks**

Anticonvulsant NMAs, including gabapentin and pregabalin, are also effective in the treatment of neuropathic pain syndromes (Bialer, 2012; Mehta et al., 2014). Epilepsy and pain are mediated by separate neuronal networks in the brain, but these networks also contain common brain regions (Linnman et al., 2012). Thus, human and animal neuroimaging studies have shown that a number of brain areas, including the PAG, are strongly implicated in pain networks, and functional changes in these pathways are induced by several drugs that possess analgesic properties (Wager et al., 2013; Becerra et al., 2013). The brainstem and the
PAG, in particular, are among the areas that show significant neuroimaging changes when gabapentin and other drugs that exert analgesic properties are administered in humans and animals (Iannetti et al., 2005; Governo et al., 2008; Takemura et al., 2011).

In chronic CNS disorders, such as chronic pain syndromes and progressive epilepsies, additional emergent properties of neurons may develop that become the critical targets for the action of anticonvulsant NMAs, such as gabapentin. Recent evidence suggests that the PAG may be a major nexus for both epileptic and chronic pain neuronal networks that is the target of anticonvulsant NMAs, such as gabapentin, and explain why these drugs are effective in these different classes of CNS disorders. Gabapentin exerts effects on PAG neurons in AGS in a somewhat different manner from that of phenytoin described above. Repeated, periodic induction of AGS results in AGS kindling, which increases seizure duration and, in GEPRs (substrain 9), induces an additional generalized clonus phase that follows the tonic extension seizure [post- tonic clonus (PTC)] (Naritoku et al., 1992; N’Gouemo et al., 2014). Systemic administration of gabapentin in AGS-kindled GEPRs blocks this PTC behavior, and the seizures temporarily revert to the unkindled pattern of behaviors that end in tonic hind limb extension (Tupal and Faingold, 2012). In AGS kindling the brainstem network expands to include the amygdala (N’Gouemo et al., 2014), and focal blockade of the amygdala will also cause the AGS-kindled seizure to revert temporarily to the unkindled pattern (Feng et al., 2001). The pathway between the amygdala and PAG is implicated in production of PTC, and AGS kindling-induced changes in this pathway were evaluated by recording PAG neuronal responses evoked by electrical stimulation of the amygdala (Tupal and Faingold, 2012). Electrical stimuli in the amygdala evoked intensity-dependent PAG neuronal firing that was enhanced significantly above that seen prior to AGS kindling (see N’Gouemo et al., 2014). Gabapentin blocked PTC in AGS-kindled GEPRs, but the other convulsive behaviors (wild running and tonic hind limb extension) seen during the seizures before AGS kindling were not affected by this same drug dose. Simultaneous with the block of PTC, gabapentin significantly reduced PAG neuronal responses to amygdala stimulation (Fig. 6). These neuronal response patterns returned to levels similar to those seen prior to AGS kindling (Tupal and Faingold, 2012). These data suggest that the amygdala to PAG pathway may be critical in mediating the emergence of PTC during AGS kindling. The ability of gabapentin to suppress this pathway to the PAG may be important for its anticonvulsant effects in AGS-kindled GEPRs. In addition, an effect on PAG neurons may also contribute to gabapentin’s effectiveness in anxiety disorders and chronic pain, since the networks that mediate these CNS disorders also involve the pathway between the amygdala and PAG, which is supported by recent data (Samineni et al., 2012), as discussed below.

Chronic pain syndromes often occur as an adverse effect of cancer chemotherapeutic drugs, such as paclitaxel (Yared and Tkacuk, 2012). Neuroimaging data in pain suggest that gabapentin may act on the PAG, as noted above, and the effect of this agent on PAG neuronal firing in response to noxious stimuli of PAG neurons was evaluated in awake, behaving rats. Gabapentin was administered to rats that had previously been treated with a paclitaxel administration paradigm that induces a chronic pain syndrome (Flatters and Bennett, 2004), and the changes in pain threshold and PAG neuronal firing were examined (Fig. 6). The nociceptive response in this chronic pain model was significantly reduced by gabapentin in the same dose that blocked PTC in kindled GEPRs. However, gabapentin in the same dose did not affect the nociceptive response or PAG neuronal firing in normal

Figure 6  An anticonvulsant network-modifying drug (gabapentin, 50 mg kg\(^{-1}\) i.p.) significantly reduced periaqueductal gray (PAG) neuronal firing and seizure severity in a repetitive epilepsy model [kindled audiogenic seizures (AGS)] in GEPR-9s 60 min after administration. The same dose of this drug significantly reduced PAG neuronal firing and thermal pain in a chronic pain model (paclitaxel), but this dose had no effect on PAG neuronal firing in acute (thermal) pain. In the left pair of bars a significantly reduced PAG (ventrolateral) neuronal responsiveness to amygdala (central) electrical stimulation was induced by gabapentin at all stimulus intensities tested [repeated measures ANOVA and post-hoc paired t-test (\(p < 0.01\))]. The middle set of bars shows thermal-evoked PAG neuronal responses in rats treated with a chronic pain-inducing protocol (paclitaxel), and a significant reduction in PAG neuronal responses was also observed 60 min after gabapentin. However, in an acute pain protocol (radiant heat presented to the paw at 53 °C) the same dose of gabapentin induced no significant change in PAG neuronal firing. Data in the left pair of bars are from awake, behaving GEPR-9s subjected to AGS kindling and in the middle and right pairs of bars are from normal rats. *Significance at \(p < 0.01\) (Repeated measure ANOVA). Reproduced from Faingold, C.L., 2014b. Neuronal network effects of drug therapies for CNS disorders. In: Faingold, C.L., Blumenfeld, H. (Eds.), Neuronal Networks in Brain Function, CNS Disorders, and Therapeutics, Academic Press/Elsevier, San Diego, pp. 443–465 with permission.
rats prior to paclitaxel treatment. Thus, the same dose of gabapentin significantly reduced the firing of PAG neurons to nociceptive stimuli in the chronic pain model. However, this dose of gabapentin did not affect PAG neuronal firing or responses to the nociceptive stimuli in the rats that were not subject to chronic pain (Samineni et al., 2012). These data, along with the effects of gabapentin in the AGS kindling paradigm discussed above, suggest that a similar change in responsiveness of PAG neurons appears to emerge in both of these chronic conditions. This change causes PAG neurons to become a critical target for therapeutic doses of gabapentin in both pain and epilepsy, which is consistent with the important role of the PAG in the neuronal networks for both disorders. In both cases the therapeutic doses are the same, and in each case this effect is seen only after chronic expansion of the network is induced. Based on the concept of emergent properties (Faingold, 2014a), an additional (emergent) property appears to develop in the PAG after either chronic protocol that causes PAG neurons to become highly sensitive to gabapentin, but this sensitivity is absent prior to these network expansion-inducing experiences (N’Gouemo et al., 2014). Gabapentin is not the only anticonvulsant NMA that is effective in epilepsy and pain. Phenytoin has long been used in chronic pain syndromes (Cheshire, 2007), and, as noted above, this anticonvulsant NMA also inhibits PAG neuronal firing in GEPRs, further supporting the concept that PAG neurons may be important in both pain and epilepsy networks.

Gabapentin, which is proposed to act primarily by binding to alpha2 delta Ca\(^{2+}\) channels (N-type) in vitro (Rogawski and Bazil, 2008; Geisler et al., 2015), may act on the PAG, because the network influences occurring during chronic pain or chronic epilepsy induce an increased number of channels or channel affinity for this drug. Alternatively, gabapentin has also been reported to increase the concentration of GABA in the brains of experimental animals (Richerson and Wu, 2004), and human imaging studies also indicate significant increases in GABA after gabapentin administration (Cai et al., 2012). This GABAergic mechanism may become more sensitive to gabapentin in the intact network due to the chronic conditions. The emergent property may also develop because both of these mechanisms are activated. It is also possible that the mechanisms responsible for the effectiveness of this anticonvulsant NMA in the chronic conditions may involve secondary events triggered by the disease process. Thus, the receptor subtypes or subunits may undergo switching due to the chronicity of the conditions, and these altered subunits may become a drug target because of increased sensitivity of the particular subunit (Ueda, 2006; Matta et al., 2011). Another possible effect of chronic CNS disorders is that intracellular mechanisms, such as NMDA receptor-mediated increases in Ca\(^{2+}\) entry into neurons, cause altered network function and expansion due to chronic network activations caused by seizure repetition or chronic pain (Grabenstein, 2012; N’Gouemo et al., 2014).

Several other mechanisms of action have been proposed for gabapentin (Zhang et al., 2013; Kumar et al., 2012). The changes induced by chronic activation of a network may cause these “minor” mechanisms to become expressed in specific network nuclei to a greater extent. Resolving these possibilities will require extensive further investigation. Thus, these emergent properties may be selective for the specific network sites in which they are observed, or the same emergent property may also occur in networks in related disorders, such as different forms of epilepsy, as well as different classes of CNS disorders, as seen with gabapentin in chronic pain and epilepsy discussed above (Faingold, 2014b).

Neuroimaging techniques have also identified pathophysiological networks in psychiatric disorders, such as anxiety, that express major differences between anxious patients and non-anxious individuals (see Schaie, 2014), and many of these differences are also reversible with successful therapy. Evidence suggests that certain anticonvulsant NMAs (e.g., valproate, lamotrigine, pregabalin, and gabapentin) may be effective in treating anxiety disorders. It may be worthwhile noting that the PAG is also a key site in the neuronal network that mediates anxiety (Linnman et al., 2012), suggesting a commonality of network structures (PAG) in anxiety, pain and epilepsy, and all these neurological disorders are treatable with the anticonvulsant NMA, gabapentin.

**Future Directions**

This neuronal network approach adds another dimension (emergent property alteration) to the understanding of how anticonvulsants block seizures. This approach also sheds light on the other therapeutic effects exerted by these agents, including usefulness in chronic pain syndromes and anxiety disorders, leading to the idea that anticonvulsants are more properly classified as neuronal network modifying agents (NMAs). Additional experimentation is required to further develop the value of this approach. Systematic explorations are needed of the variety of emergent properties that exist in other epileptic neuronal networks, as well as in the other neurological disorders, such as chronic pain, in which these drugs are effective. There is a hierarchy of levels within the nervous system at which emergent properties may occur. The concept that an emergent property is a critical target for CNS drug action was developed in the GEPR, and additional anticonvulsant NMAs need to be evaluated in this model. More comparisons between in vitro versus in vivo effects of the same agent are needed. The generality of this approach also needs to be tested in closely related seizure models (Faingold et al., 2014a). The mechanisms need to be examined in other seizure models to identify whether differences and similarities occur. This methodology can generate a compendium of emergent targets for drug action. This approach is potentially important for the development of improved drug treatment for epilepsy, and a similar approach may be applicable to other CNS disorders, such as anxiety and pain, to determine if there are common actions, as suggested by the findings with gabapentin discussed above.

Seizure networks are subject to plasticity, which may result in the expression of additional emergent properties. For example, partial seizures initially involve a relatively circumscribed neuronal network, but if the seizure secondarily generalizes, the seizure network expands and additional properties emerge. These properties may be additive with the original emergent properties. Blockade of these separable emergent properties may require drugs with different actions. Alternatively, effective anticonvulsants...
may act at multiple levels. As noted above, gabapentin, for example, acts on isolated alpha(2) delta Ca2+ channels, which can be considered the “ground level” in the hierarchy of emergent properties affected by this agent. However, gabapentin also acts on emergent properties at higher organizational levels in vivo, including specific network nuclei, as seen in the studies cited above where it selectively targets PAG neurons within the AGS network and in a chronic pain network in non-epileptic animals. Identification of how the anticonvulsant NMAF affect the emergent properties subserving the pathophysiology of epilepsy and of chronic pain is vital in order to identify drugs that more selectively target these properties. The studies described above apply this approach by investigating network mechanisms for seizure and chronic pain control and evaluating the anticonvulsant NMAF on neurons in the nuclei of these networks. These studies are yielding a detailed understanding of the way these network functions and how anticonvulsant NMAF modify these mechanisms. The results of initial studies indicate that specific sites within the intact network are most sensitive to therapeutic doses of a given drug; this site sensitivity may be different with each specific anticonvulsant examined, since even drugs with similar mechanisms of action can act at different brain sites, as discussed for the NMDA receptor antagonists. However, the findings with the anticonvulsant NMAF, gabapentin, indicate that a specific brain nucleus, the PAG, may play a common role in treatment of both seizure and chronic pain, suggesting the possibility that this may also occur with other anticonvulsant NMAF that are effective in multiple CNS disorders.

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References


Bialer, M., 2012. Why are antiepileptic drugs used for nonepileptic conditions? Epilepsia 53 (Suppl. 7), 26–51.


