Journal Club

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Impaired Cardiorespiratory Function during Focal Limbic Seizures: A Role for Serotonergic Brainstem Nuclei

Jabir Mohamed[1] **and Brian W. Scott**[2]

[1]Institute of Medical Science, University of Toronto, Toronto, Ontario M5S 1A8, Canada, and [2]Neurosciences & Mental Health, Hospital for Sick Children, Toronto, Ontario M5G 1X8, Canada

Review of Zhan et al.

Seizures that impair consciousness and cardiorespiratory function have a significant, negative impact on patient safety and quality of life. If uncontrolled, such seizures can limit a patient's driving ability, productivity at school and work, or worse, lead to death (Blumenfeld, 2012). Sudden unexpected death in epilepsy (SUDEP) is the most devastating consequence of refractory epilepsy, but the precise mechanism remains unknown. Nonetheless, evidence suggests a combination of cardiorespiratory compromise and decreased arousal (Massey et al., 2014). Therefore, understanding how seizures cause loss of consciousness and cardiac or respiratory arrest is essential. Progress on this front has been aided by two hypotheses (Richerson and Buchanan, 2011; Blumenfeld, 2012), which attribute these deficits to different neuromodulators, specifically acetylcholine and serotonin [5-hydroxytryptamine (5-HT)].

One hypothesis, the network inhibition hypothesis, proposes that impaired consciousness in temporal lobe seizures results from the loss of cholinergic input to the cortex (Blumenfeld, 2012). Data from intracranial EEG, neuroimaging, and animal models support this view. Across studies, decreased cerebral blood flow and widespread slow-wave activity are observed in the frontoparietal association cortex during temporal lobe complex partial seizures (Blumenfeld, 2012). These sleep-like changes are correlated with suppressed firing of cholinergic neurons in the basal forebrain and pedunculopontine tegmental nucleus (Motelow et al., 2015). This is presumed to result from aberrant activation of subcortical "inhibitory" structures with rich GABAergic projections to these arousal centers, such as the lateral septum and anterior hypothalamus (Motelow et al., 2015).

The network inhibition hypothesis does not explain the mechanisms leading to cardiorespiratory dysfunction, and ultimately, SUDEP, however. Therefore, Richerson and Buchanan (2011) proposed a role for the 5-HT raphe nuclei in the midbrain and medulla, in both arousal and cardiorespiratory impairment induced by seizures. In general, 5-HT exhibits anticonvulsant properties and agents that elevate 5-HT levels, such as serotonin reuptake inhibitors, block many seizure types (Bagdy et al., 2007). Work in transgenic mice provided initial evidence linking 5-HT dysfunction and SUDEP. Specifically, mice lacking 5-HT neurons experience postictal respiratory arrest and increased mortality after audiogenic or electrically induced seizures (Richerson and Buchanan, 2011).

To further investigate the role of 5-HT in seizure-induced loss of consciousness and cardiorespiratory dysfunction, Zhan et al. (2016) measured changes in the firing of medullary and midbrain raphe neurons in rats during and after seizure induction. Given that 5-HT neurons stimulate breathing and arousal (Richerson and Buchanan, 2011), Zhan et al. (2016) hypothesized that a reduction in the activity of these neurons during and after a seizure would correlate with impaired cardiorespiratory function and consciousness.

Seizures were triggered from the dorsal hippocampus in lightly anesthetized animals, and multiunit and single-unit activity was recorded along with changes in breathing and heart rate. Respiratory rate, tidal volume, and minute ventilation were all decreased during and after seizures. Heart rate showed a slight, but significant, decrease during and following seizures (Zhan et al., 2016, their Fig. 1*A*). Analyses of multiunit activity in the medullary and midbrain raphe indicated that overall neuronal activity was significantly reduced during and after seizures (Zhan et al., 2016, their Fig. 1*B*). Similarly, single-unit activity of serotonergic medullary raphe neurons was markedly reduced both during and after seizures (Zhan et al., 2016, their Fig. 1*B*). In contrast, changes in the firing rate of individual midbrain serotonergic neurons were inconsistent, with some cells showing decreased firing, while others showed increased firing (Zhan et al., 2016, their Fig. 8*C*). Consequently, changes occurring during the ictal and postictal periods were not

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Correspondence should be addressed to Jabir Mohamed, University of Toronto, Room 4352, Medical Sciences Building, 1 King's College Circle, Toronto, ON M5S 1A8, Canada. E-mail: jabir.mohamed@mail.utoronto.ca.

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statistically significant (Zhan et al., 2016, their Fig. 1*B*).

This study revealed important effects of seizures on arousal and cardiorespiratory modulating centers of the 5-HT system. Zhan et al. (2016) provide strong evidence that reduction in medullary raphe serotonergic firing is associated with cardiorespiratory impairment during and after seizures. Ultimately, these findings provide the basis to further explore the cardiorespiratory centers that may be disrupted and a model in which to test treatments that may benefit persons with epilepsy who are at risk for SUDEP.

How might seizures affect cardiorespiratory centers downstream from the medullary raphe? Under normal conditions, 5-HT neurons in the medulla promote respiration: they increase phrenic motoneuron activity, facilitate respiratory rhythmogenesis, and contribute to the CO_2-driven central respiratory chemoreflex (Hodges and Richerson, 2010). Based on these previous findings, one might speculate that seizure-induced depression of medullary 5-HT neuronal firing leads to decreased excitation of respiratory centers, which in turn leads to hypoventilation. However, the mechanisms underlying seizure-induced respiratory dysfunction may differ with an increasing number and severity of seizures. On one hand, repeated generalized seizures could heighten the activation of inhibitory pathways (Dlouhy et al., 2015), leading to further reductions in 5-HT neuronal firing and more pronounced hypoventilation. It is also possible that hyperactivity resulting from repeated generalized seizures could disrupt normal network activity in respiratory centers, particularly when the epileptic discharge spreads into the brainstem (McIntyre and Gilby, 2008). Thus, abnormally decreased or increased activity in the same networks may underlie respiratory dysfunction induced by seizures, which is similar to what has been proposed to explain how different seizure types can cause loss of consciousness (Blumenfeld, 2012).

While respiration might become progressively slowed with seizure repetition and generalization, heart rate might increase progressively. According to several authors, increases in heart rate are correlated with the volume of cortical areas recruited during the seizure, particularly areas of central autonomic control such as the insular, anterior cingulate and prefrontal cortices (Eggleston et al., 2014). Consequently, as seizures worsen and more cortical areas become involved (McIntyre and Gilby, 2008), heart rate would progressively increase. Consistent with this, Pansani et al. (2011) observed a correlation between the number and severity of seizures induced by electrical amygdala kindling and ictal tachycardia. Similarly, the intense muscular and neuronal activity associated with tonic-clonic seizures presents an increased demand for oxygen; as such, the heart must pump harder to supply oxygen to the muscles and to sustain the profound increases in cerebral blood flow (Blumenfeld, 2012).

What pathways lead to reduced activity of 5-HT neurons in the medulla raphe nuclei? Zhan et al. (2016) suggest amygdala involvement, based on work in humans and animals (Pansani et al., 2011; Dlouhy et al., 2015). Either of the major efferent pathways of the amygdala, the ventral amygdalofugal pathway and the stria terminalis, could conceivably affect the activity of medullary neurons. The amygdalofugal pathway carries output from the central nucleus of the amygdala to medullary regions implicated in apnea (Verner et al., 2008). The stria terminalis is the main output pathway from the amygdala to subcortical structures, such as the lateral septum and the hypothalamus (De Olmos and Ingram, 1972; Ono et al., 1985). Recently, the orexin neurons of the lateral hypothalamus have been implicated in respiratory control. These cells respond to changes in CO_2 levels, project to respiratory centers in the brainstem, and stimulate breathing (Nattie and Li, 2012). Distinguishing between these direct and indirect pathways will be critical in determining a target for therapies aimed at preserving cardiorespiratory function in human temporal lobe seizures.

One of the more interesting findings from this study was that the midbrain population of serotonergic neurons did not show significant decreases in firing during and after seizures, likely because some neurons had increased responses and others had decreased responses. Given that a subset of midbrain 5-HT neurons also expresses CO_2 chemoreceptors, and their activation promotes arousal (Richerson and Buchanan, 2009), it is possible that seizures could depress the chemosensitive 5-HT neurons in the midbrain raphe. Assessing the chemoresponsiveness of midbrain 5-HT neurons following extracellular recordings is needed to test this possibility. Alternatively, it is likely that nonserotonergic neurons of midbrain raphe contribute to impaired consciousness during seizures. Midbrain GABAergic neurons innervate and inhibit neighboring 5-HT neurons (Bagdy et al., 2000), so it would be worth exploring how seizures affect the firing of these neurons. In fact, an unexplained observation from neuroimaging studies in patients during temporal lobe complex partial seizures is that the medial/intralaminar and brainstem reticular formation are hyperperfused (Blumenfeld, 2012). One possibility is that neuronal activity is dominated by inhibitory activity—that is, seizure discharge activates GABA-releasing interneurons within the arousal centers—which is metabolically demanding yet leads to reduced cortical arousal. Another possibility is that glutamatergic projection neurons in the midbrain raphe, which promote arousal by exciting neurons in the ventral tegmental area and lateral hypothalamus that activate the cortex (Hioki et al., 2010), are suppressed during seizures. This would support the Blumenfeld hypothesis, and warrants further investigation. Recording single-unit activity from midbrain raphe GABAergic and glutamatergic neurons would help resolve these mysteries.

Additional work will be required to establish a causal relationship between 5-HT deficits and cardiorespiratory dysfunction in this seizure model. However, it is important to note that the present model of single seizures may have limited relevance to SUDEP, given that people with epilepsy experience many seizures. Thus, a model involving repeated spontaneous seizures (e.g., the kainic acid model) or repeated triggered seizures (e.g., the kindling model) is needed to corroborate these findings. That being said, it is necessary to examine whether pretreatment with serotonin reuptake inhibitors or agonists can suppress or prevent the seizure-induced depression of medullary raphe neuronal firing and cardiorespiratory dysfunction. More sophisticated interventions include optogenetic or pharmacogenetic activation of the medullary serotonergic nuclei during and after seizures to determine whether selective activation of these neurons restores normal cardiorespiratory function. In accordance with the hypothesis of Richerson and Buchanan (2011), these interventions should reduce the incidence of respiratory arrest induced by seizures. If successful, treatment with selective serotonin reuptake inhibitors, serotonin agonists, or deep brain stimulation of medullary raphe targets should be evaluated as strategies to improve cardiorespiratory function in patients with epilepsy.

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