

A New Phase in Anesthetic-induced Neurotoxicity Research

SINCE their discovery, volatile anesthetics have been viewed as benign inhibitors of the central nervous system. Although the anesthetic state was always associated with potential risks, volatile anesthetics *per se* were thought to produce no central nervous system sequelae after discontinuation of the anesthetic. In fact, volatile anesthetics were viewed as beneficial to certain patients and capable of neuroprotection, presumably through central nervous system quiescence. However, recent experimental studies have seriously challenged this dogma, causing an astonishing paradigm shift in the field of anesthesiology, particularly regarding the care of infants given general anesthesia. In the current issue of ANESTHESIOLOGY, the work of Lemkuil *et al.* represents a shift in the focus of the studies of anesthetic-induced neurotoxicity.¹ This study presents an essential early step in understanding the molecular mechanisms underlying isoflurane-induced neurotoxicity. For an excellent detailed background, the reader is referred to the editorial review by Patel and Sun in ANESTHESIOLOGY.²

Jevtovic-Todorovic *et al.* first showed that the commonly used anesthetic combination of midazolam, nitrous oxide, and isoflurane was capable of inducing widespread apoptosis and neuronal degeneration throughout developing rat brains.³ They also demonstrated that these pathologic changes were accompanied by a learning defect that persisted into adulthood in the rat. They identified a critical window of exposure during the period of rapid synaptogenesis, from days 4 to 10 in postnatal rats.⁴ A previous report showed specifically that drugs that inhibit the *N*-methyl-D-aspartic acid receptor or enhance the γ -aminobutyric acid type A receptor were capable of inducing neuronal apoptosis during early development.⁵

It is now well established that, at least in rodents, volatile anesthetics in isolation are capable of inducing neurodegeneration in the developing nervous system. However, it remains unclear how great a risk perioperative anesthetic exposure poses to newborn humans. Retrospective studies have increased the possibility that significant learning defects may exist in children who have had multiple anesthetic exposures.^{6,7} In each patient, the studies are confounded by the fact that no child receives an anesthetic without having some comorbid condition requiring surgery or diagnostic study. It is extremely difficult to separate out the anesthetic effect from the condition necessitating the exposure. If, in fact, general anesthetics cause permanent developmental defects to humans during a critical window of vulnerability, the

implications to our current care of children obviously are enormous.

Most early work rightfully focused on the timing and the magnitude of anesthetic exposure necessary to cause neurotoxicity. However, little work has gone into determining the molecular mechanisms that trigger apoptosis, resulting in neuronal degeneration. The existence of such neurotoxicity is now on firm enough footing that it is clearly possible that our patients are at risk. Preventing the phenomenon requires an understanding of its cause(s). We simply cannot wait until we know the true magnitude of the risk to our patients before unraveling the molecular mechanisms behind this effect.

Until there is proof that humans are selectively not at risk for this neurotoxicity, it seems wise to minimize the risk. Of course, studies are being undertaken to determine whether altering the type of anesthetic delivered to infants will improve their outcomes. Because it is impossible to completely eliminate exposure of neonates to general anesthesia, it also seems prudent to identify mechanisms to prevent the untoward effects of anesthetics. We must move beyond studies that describe the magnitude and periods of vulnerability but leave us powerless to improve outcomes in exposed individuals.

Unfortunately, the mechanisms underlying the neurotoxicity are relatively unclear. Jevtovic-Todorovic *et al.* and Head *et al.* separately implicated brain-derived neurotrophic factor as a potentially important agent in the effect.^{8,9} Brain-derived neurotrophic factor is a crucial component of synaptogenesis, and its loss is known to induce neuronal apoptosis *via* induction of p75 neurotrophic receptors (p75^{NTR}). Head *et al.* found that inhibiting brain-derived neurotrophic factor function by a variety of treatments, including isoflurane exposure, led to neuronal apoptosis in cell culture and in hippocampal slices.⁹ These authors identified a role of p75^{NTR} and actin in the effects of isoflurane.

In this issue, Lemkuil *et al.* further tested the hypothesis that two proteins, p75^{NTR} and RhoA, are activated by isoflurane leading to depolymerization of actin, resulting in apoptosis.¹ If their hypothesis is correct, then there is hope that inhibition of this pathway could alleviate the anesthetic-induced neurotoxicity. They tested this hypothesis in both primary cell culture and in hippocampal slices. The authors found that RhoA was activated in neurons after isoflurane exposure, and that the actin-based cytoskeleton was depolymerized leading to an increase in markers for apoptosis. Fur-

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◆ This Editorial View accompanies the following article: Lemkuil BP, Head BP, Pearn ML, Patel HH, Drummond JC, Patel PM: Isoflurane neurotoxicity is mediated by p75^{NTR}-rhoA activation and actin depolymerization. ANESTHESIOLOGY 2011; 114:49-57.

thermore, inhibition of the p75^{NTR} and RhoA axis led to attenuation of the isoflurane-induced effects. Finally, stabilization of the actin cytoskeleton in hippocampal slices also inhibited the effects of isoflurane on neuronal structure. Lemkuil *et al.* conclude that the p75^{NTR}/RhoA pathway is involved in isoflurane-induced neurotoxicity.

There are some limits to the study that require follow-up. First, the hypothesis is not a blinded one. The authors have identified a logical candidate for triggering neuroapoptosis, but anesthetics are clearly very promiscuous drugs. It is quite possible that this pathway is only one of several that are involved. Second, the inhibitors used by the authors are thought to be specific to the pathway studied, but it is possible they have secondary effects on other targets affecting neurodegeneration. Third, the inhibitors used in the study are not complete inhibitors of the p75^{NTR}/RhoA pathway; thus, any remaining isoflurane effect could be from residual p75^{NTR}/RhoA activity or from other targets. In addition, the studies were primarily done in cell culture or with neuronal slice cultures. It will be essential to follow these with studies in whole animals measuring both neuronal and behavioral effects of the inhibitors. Finally, it is not known whether the role of brain-derived neurotrophic factors may actually be causative *in vivo* or the magnitude or uniqueness of its role in the anesthetic effect.

With the above caveats in mind, the study of Lemkuil *et al.* represents an important step forward in understanding anesthetic-induced neurotoxicity. These are the types of studies that may allow us more tools than simply avoidance to protect our vulnerable patients. We are truly entering the next phase in the study of anesthetics and neurodegeneration.

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