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Chloral Hydrate Is Not Acceptable for Anesthesia or Euthanasia of Small Animals

To the Editor:—A recent paper[1] reported experiments, using laboratory rats, on the effect of intraamygdala infusion of a γ-aminobutyric acid type A antagonist on propofol-induced amnesia for inhibitory avoidance training, as well as on expression of activity-regulated cytoskeleton-associated protein in the hippocampus. This work may elucidate the neural mechanisms of the amnestic effects of propofol, as well as the neurobiological mechanisms of general anesthesia and memory more generally.

The experiments reported in this paper required stereotaxic neurosurgery to implant cannulae aimed at the basolateral amygdala, and rats were subsequently euthanized for determination of activity-related cytoskeleton-associated protein levels or histologic verification of cannula placement. The authors used chloral hydrate in both procedures; for surgical anesthesia in the first and euthanasia in the second. Chloral hydrate is not a suitable drug in either case. Chloral hydrate is regarded by many to produce hypnosis and not anesthesia.[2] It does not provide analgesia and causes marked respiratory depression at doses required for surgical anesthesia.[3] Apart from its inadequate anesthetic properties, 20% chloral hydrate is extremely irritating and therefore unsuitable for intraperitoneal use. It is associated with ileus in rats,[4] as well as peritonitis and gastric ulcers.[5] Its use by intraperitoneal injection for survival surgery is not recommended.[5] Thus, it is not the most refined choice of agent for the surgical procedure in which cannulae are chronically implanted to make drug infusions into the amygdala. The authors also used a higher dose of chloral hydrate for euthanasia. However, chloral hydrate is not an acceptable agent for euthanasia according to the guidelines of the American Veterinary Medical Association†; its use for this purpose has been proscribed for some time.[6]

There are no scientific justifications for using chloral hydrate for these experiments, as many other agents would be more suitable for both surgical anesthesia and euthanasia without interfering with the experimental endpoints. Indeed the chloral hydrate-induced hypoxemia which must occur during euthanasia as respiration becomes depressed†, may compromise the experimental aims in terms of measuring protein and messenger ribonucleic acid levels of an activity-related protein. The noxious stimulus of an intraperitoneal irritant is not only inhumane, but if it leads to peritonitis the rats will be abnormal at the time of testing.

It seems that chloral hydrate has traditionally been used to provide anesthesia where the avoidance of agents with known receptor interactions is desirable. But it is likely that chloral hydrate has unknown receptor interactions. Therefore choosing a different agent whose receptor interactions are better characterized could be beneficial, not only in terms of animal welfare but also in terms of data interpretation.

The publication of this paper in ANESTHESIOLOGY concerns us, because the standard of laboratory animal anesthesia used in this research is not acceptable.

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† AVMA Guidelines on Euthanasia, 2007. http://www.avma.org/issues/animal_welfare/euthanasia.pdf. Last accessed 11-10-08.

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In Reply:—We sincerely thank Baxter *et al.* for their interests in our article and their valuable information about the use of chloral hydrate for rats' anesthesia and euthanasia in our experiment.

First, we would like to emphasize that we do not think the reliability of our experimental results was influenced by chloral hydrate. Chloral hydrate was used in all the experimental groups, thus its interpretations were comparable among these groups. Our significant findings could not be simply induced by it. In addition, the mechanisms of most anesthetics, including their effects on Arc expression, are still obscure. Furthermore, sevoflurane has even been proved to inhibit Arc transcription.[1] Under this condition, choosing any other anesthetic for rat euthanasia may produce the similar unpredicted interpretations. Therefore, we believe that to a great extent, our results and conclusions are reliable.

Second, we designed our experiment on the basis of a great deal of published articles on authority journals. The method as intraperitoneal injection of chloral hydrate was wildly used to rats for some kinds of surgeries, particularly with the word as "anesthesia." For example, Rodríguez Manzanares *et al.*, Bredeloux *et al.*, and Sammut *et al.* all use chloral hydrate to anesthetize rats for stereotaxic neurosurgery to implant cannulae.[2–4] Actually, in recent years, chloral hydrate is still widely used to anesthetize rats. However, we agree with the view of Baxter *et al.* that some other anesthetics (like Phenobarbital sodium) may be more suitable in this type of surgery because of the side effects of chloral hydrate illustrated by them. Fortunately, the overwhelming

As noted by the letter from Dr. Baxter, chloral hydrate is unacceptable for anesthesia or for euthanasia, although it has been used for these purposes in the past. We regret that the peer review system, both in this journal and in other prominent journals as noted by Dr. Yu, or the institutional animal care and use committees do not always catch this animal care issue, and we will strive to do so and not publish work using chloral hydrate for these purposes in the future. —James C. Eisenach, M.D., Editor-in-Chief.

majority of rats in our experiment recovered well from the neurosurgery, with normal appetite and defecation.

Third, we admit that we neglect the potential problem of using chloral hydrate for euthanasia of rats. Chloral hydrate is a traditional anesthetic in animal experiments, and before we performed our study we also found that it is used for killing rats by either decapitation or cardiac perfusion in respectable published articles.[2,5,6] Moreover, our research was approved by the Institutional Animal Care and Use Committee with no questions. Therefore, we never doubted the use of chloral hydrate for rat euthanasia. Now we feel deeply sorry for the possibility of inflicting pain on the animals because of using chloral hydrate.

Finally, we would like to extend our sincere gratitude to Baxter *et al.* for letting us understand animal euthanasia more deeply. We will pay much more attention to the euthanasia issue, adopt proper and scientific animal welfare methods, and try our best to decrease harm to the experimental animals as much as possible in future research.

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Low-dose Spinal Anesthesia with Low-dose Phenylephrine Infusions for Cesarean Delivery: Better but Not Necessarily Best

To the Editor:—We read with interest the recently published report by Langesæter *et al.*[1] together with the accompanying editorial,[2] describing the use of minimally invasive continuous cardiac output monitoring during spinal anesthesia for cesarean delivery. Based on derived values for cardiac output, the authors advocated allowing a 10 to 20% decrease in blood pressure and questioned as unnecessary the use of higher doses of phenylephrine given to maintain blood pressure near baseline. The findings from this paper are important and are a substantial contribution to our knowledge of the hemodynamic changes during regional anesthesia in parturients. However, we believe that some caution is required when extrapolating these findings to recommendations for everyday clinical practice.

Although placement of peripheral arterial catheters was regarded as "minimally invasive," we believe that most clinicians would find them difficult to justify in routine elective cases and will continue to rely on the use of noninvasive blood pressure monitoring, despite its intermittent nature of measurement. From our previous work, we consider that, when using noninvasive blood pressure monitoring, the clinically optimal approach is to titrate a phenylephrine infusion with the target of maintaining systolic blood pressure near to the baseline value. Using this approach, with both manual[3–5] and automated[6] control, the incidence of nausea and vomiting is significantly reduced, as compared with less strict blood pressure control[4]; this is a clearly defined and important endpoint, the clinical relevance of which is more immediately apparent as compared with derived values for cardiac output.

When using phenylephrine to maintain maternal blood pressure, it is important to appreciate that although this potent vasopressor has a faster onset and can be more accurately titrated than ephedrine,[7] control is not perfect and some variation (overshoot and undershoot) between target and actual blood pressure is to be anticipated. For example, in a previous study,[4] when we targeted a systolic blood pressure of 80% of baseline (equivalent to allowing a 20% decrease as advocated by Langesæter *et al.*[1]), in actuality in the first 15 min after intrathecal injection, 96% of patients had one or more measurements $< 80\%$ of baseline with 25% of all measurements $< 80\%$ (fig. 1), and 52% of patients had one or more measurements $< 70\%$ of baseline with

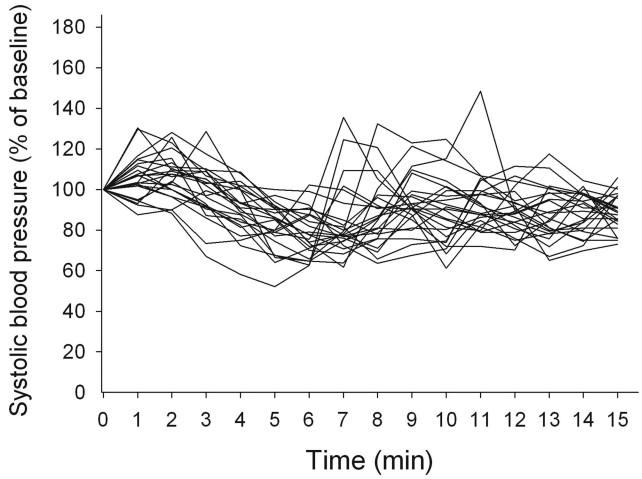

Fig. 1. Changes in systolic blood pressure in the first 15 min after spinal anesthesia in patients having cesarean delivery with a phenylephrine infusion titrated with the target of maintaining values at the target of 80% of baseline. Twenty-five percent of all measurement were < 80% of baseline and 7% of all measurements were < 70% of baseline.[4]

7% of all measurements $< 70\%$. As a consequence, 40% of patients experienced nausea or vomiting. In comparison, when we targeted a systolic blood pressure of 100% of baseline, 29% of patients had one or more measurements $< 80\%$ of baseline with 4.7% of all measurements $< 80\%$; 8% of patients had one or more measurements $< 70\%$ of baseline with 2% of all measurements $< 70\%$; and only 4% of patients experienced nausea or vomiting. Phrased in another way, our results suggest that to maintain maternal comfort, phenylephrine should be titrated whenever blood pressure begins to decrease. Delaying or withholding administration while permitting a decrease in blood pressure to some arbitrary percentage threshold below baseline as suggested by Langesæter *et al.*[1] risks the likelihood that blood pressure