Local spinal cord glucose utilization in conscious and halothane-anaesthetized rats

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The authors used the 2-[¹⁴C]deoxyglucose method to study local spinal and cerebral glucose utilization simultaneously during 1.2 per cent halothane anaesthesia in adult Sprague-Dawley rats. In conscious animals (n = 5) the rate of glucose utilization in lumbar spinal gray matter was about 50 per cent lower than that of cerebral cortex. Halothane anaesthesia (n = 6) reduced spinal cord and cerebral metabolic rate. Spinal glucose utilization was reduced 12–35 per cent, but this was less than the 45–70 per cent decrease halothane produced in 8 of 16 cerebral structures examined and was independent of the hypotension produced. These results indicate that halothane is a spinal metabolic depressant but that its effects on this tissue are substantially less than those it has on many cerebral structures.

Key words

SPINAL CORD: metabolism, regional, glucose utilization, regional; BRAIN: metabolism, regional; GLUCOSE UTILIZATION: local; ANAESTHESIA: general, halothane; MEASUREMENT TECHNIQUES: autoradiography.

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Since most studies of central nervous system metabolism during anaesthesia have not included spinal cord in the analysis, it is unclear whether the spinal cord is subject to the profound metabolic changes usually associated with anaesthesia. The fact that most anaesthetics reduce brain metabolism^{1–3} does not necessarily imply equal effects on the spinal cord because these tissues are fundamentally different in some important respects. For example, the spinal cord has a lower neuronal density,^{4,5} lower rates of blood flow and metabolism,^{6–9} and is possibly less responsive to certain physiological stimuli than the brain.¹⁰ Such differences suggest that the spinal cord may be less responsive to anaesthetic effects as well. Indeed, in a previous study⁶ we found that pentobarbitone anaesthesia reduced spinal metabolic rate considerably less than it did the metabolism of many cerebral structures but that 70 per cent nitrous oxide stimulated metabolism in both tissues. The purpose of the present experiments, therefore, was to determine the nature of halothane's effect on spinal metabolism and to compare its spinal action with its effect on the brain.

Methods

Local spinal and cerebral glucose utilization were measured simultaneously with the 2-[¹⁴C]deoxyglucose (2-DG) technique¹¹ in 16 adult, male Sprague-Dawley rats. All animals were anaesthetized with one per cent halothane and 70 per cent nitrous oxide for femoral artery and vein catheterization. The five control animals were then partially restrained with a pelvic plaster cast and allowed to awaken. The remaining 11 animals had a tracheostomy performed, were paralyzed with gallamine (2 mg intravenously with periodic supplementation), and were mechanically ventilated with a small-animal ventilator (Harvard Apparatus Co., Boston, MA). Once surgery was complete, nitrous oxide was discontinued and the inspired halothane concentration adjusted to 1.2 per cent (Beckman LB-2 infra-red analyzer, Fullerton, CA), a dose equivalent to 1 MAC in the rat.¹² Arterial blood gas tensions and pH and mean arterial blood pressure (MAP)

were measured in all animals. Rectal temperature was also measured and maintained with a heat lamp. Since halothane anaesthesia reduces blood pressure, we evaluated the effect of hypotension on spinal glucose utilization by dividing experimental animals into two groups: the blood pressure of Group I animals ($n = 6$) was not adjusted (they received only saline intravenously) while Group II rats ($n = 5$) received an intravenous infusion of phenylephrine to maintain blood pressure near control levels.

Glucose utilization was measured following at least 30 min of equilibration at 1.2 per cent halothane or, in control animals, at least 2 h after anaesthesia was discontinued. Animals received an intravenous bolus injection of 2-DG ($125 \mu\text{g} \cdot \text{kg}^{-1}$) and arterial blood was sampled at frequent timed intervals during the 45-min experiment. The plasma concentrations of 2-DG and glucose were determined by scintillation counting and enzymatic analysis, respectively. Animals were sacrificed with an overdose of pentobarbitone and the lumbar spinal cord was rapidly removed and frozen in isopentane (-50°C). To allow comparison of spinal cord and brain effects of halothane, the brains of control and Group I animals were also removed and frozen. Tissue sections 20 microns thick were later cut in a cryostat at -22°C and exposed along with a set of previously calibrated methylmethacrylate standards (Amersham, Arlington Heights, IL) on OM-1 x-ray film (Eastman Kodak, Rochester, NY).

Autoradiographs of lumbar spinal cord were analyzed with a computerized image-processing system¹³ and brain autoradiographs from control and Group I rats were analyzed by manual densitometry (Densichron Model PPD, Sargent Welch, Skokie, IL). All optical density measurements were made bilaterally in a minimum of five sections. Glucose utilization was calculated from tissue ^{14}C concentrations, the rate and lumped constants of normal rat brain, and the time course of plasma glucose and 2-DG concentrations according to the operational equation of the method.¹¹ The three sets of physiologic and spinal cord glucose utilization data were analyzed with Dunnett's *t* test for multiple comparisons¹⁴ and a *t* test for unpaired data was used to analyze the two sets of cerebral metabolic data.

Results

Differences in physiologic variables between the groups were anticipated (Table I). Use of high inspired oxygen concentrations produced a higher arterial PO_2 in experimental animals but we know of no evidence that such hyperoxia affects the metabolic rate of neural tissue. Group I animals were hypotensive and had a slightly lower arterial pH than controls, while animals receiving

TABLE I Physiologic variables (mean $\pm$ SEM)

	Control ($n = 5$)	Group I ($n = 6$)	Group II ($n = 5$)
Mean arterial blood pressure (kPa)	17.18 $\pm$ 0.39	8.97 $\pm$ 0.25*	17.23 $\pm$ 0.72
Rectal temperature ($^\circ\text{C}$)	36.6 $\pm$ 0.3	37.3 $\pm$ 0.3	37.3 $\pm$ 0.5
Arterial pH	7.47 $\pm$ 0.01	7.43 $\pm$ 0.01†	7.46 $\pm$ 0.02
PaCO_2 (kPa)	4.58 $\pm$ 0.08	5.14 $\pm$ 0.24	4.85 $\pm$ 0.18
PaO_2 (kPa)	11.43 $\pm$ 0.46	49.46 $\pm$ 2.92*	38.29 $\pm$ 4.02*

* $p < 0.01$ as determined by Dunnett's *t* statistics.

† $p < 0.05$.

TABLE II Spinal glucose utilization ($\mu\text{mol} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$) (mean $\pm$ SEM)

	Control ($n = 5$)	Group I ($n = 6$)	Group II ($n = 5$)
Gray matter			
Lamina(e) I–III	40 $\pm$ 2	35 $\pm$ 2	30 $\pm$ 1†
IV–VI	53 $\pm$ 3	37 $\pm$ 2*	35 $\pm$ 1*
VII	55 $\pm$ 2	37 $\pm$ 3*	35 $\pm$ 1*
VIII	57 $\pm$ 2	37 $\pm$ 3*	36 $\pm$ 1*
IX	54 $\pm$ 2	35 $\pm$ 3*	33 $\pm$ 1*
X	56 $\pm$ 3	40 $\pm$ 3*	36 $\pm$ 1*
White matter			
Dorsal	21 $\pm$ 2	15 $\pm$ 1	17 $\pm$ 1
Lateral	32 $\pm$ 2	23 $\pm$ 1†	23 $\pm$ 1†
Ventral	29 $\pm$ 1	20 $\pm$ 1*	22 $\pm$ 1*

* $p < 0.01$ as determined by Dunnett's *t* statistics.

† $p < 0.05$.

phenylephrine (Group II) had a normal blood pressure and pH.

Glucose utilization was measured in five gray and three white matter regions of lumbar spinal cord (Table II) and in 16 regions of brain (Table III). In the conscious animals, spinal white matter glucose utilization was approximately one-half the rate of spinal gray matter, which, in turn, was only one-half as metabolically active as cerebral cortex. Within spinal gray and white matter, however, there was little metabolic heterogeneity (Table II). Laminae I–III and dorsal white matter were the least metabolically active areas of these tissues. In contrast, the brain of conscious rats showed considerable metabolic heterogeneity and most structures had a higher metabolic rate than the spinal cord (Table III).

Halothane proved to be a spinal metabolic depressant regardless of whether blood pressure was normal or low (Table II). Spinal glucose utilization was reduced 12–35

TABLE III Cerebral glucose utilization ($\mu\text{mol} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$) (mean SEM)

	Control (n = 5)	Group I (n = 6)
Frontal cortex	110 $\pm$ 2 ^a	58 $\pm$ 5*
Sensorimotor cortex	97 $\pm$ 3 ^a	56 $\pm$ 3*
Visual cortex	86 $\pm$ 3	54 $\pm$ 3*
Auditory cortex	130 $\pm$ 6 ^a	68 $\pm$ 6*
Medial geniculate body	111 $\pm$ 7	48 $\pm$ 3*
Inferior colliculus	171 $\pm$ 10	50 $\pm$ 3 ^{ab}
Thalamus: lateral nucleus	98 $\pm$ 4	53 $\pm$ 3*
Caudate nucleus	94 $\pm$ 1	80 $\pm$ 5†
Globus pallidus	57 $\pm$ 2	38 $\pm$ 4*
Substantia nigra	62 $\pm$ 5 ^a	59 $\pm$ 9
Hippocampus	78 $\pm$ 1	63 $\pm$ 4*
Dentate gyrus	58 $\pm$ 1	45 $\pm$ 3 ^{ab}
Hypothalamus	51 $\pm$ 1	38 $\pm$ 3*
Amygdala	47 $\pm$ 1	34 $\pm$ 2*
Cingulate gyrus	100 $\pm$ 3	55 $\pm$ 4*
Cerebellar cortex	50 $\pm$ 1 ^a	37 $\pm$ 1 ^{ab}

* $p < 0.01$ by grouped t test.† $p < 0.05$.^an = 4 animals, ^bn = 5 animals.

per cent in hypotensive animals (Group I), but the changes in laminae I–III and dorsal white matter were not statistically significant. Hypotension evidently contributed little to this effect, since no more than an additional 13 per cent decrease in spinal metabolic rate occurred when MAP was maintained normal with phenylephrine. In contrast to its spinal metabolic effects, halothane reduced cerebral glucose utilization to a greater extent in many structures. Thus, halothane reduced glucose utilization 45–70 per cent in frontal, auditory, and sensorimotor cortices, thalamus, and cingulate gyrus but decreased metabolic rate in caudate nucleus, globus pallidus, hippocampus, and amygdala 15–33 per cent. Therefore, in eight of 16 structures, halothane reduced cerebral glucose utilization 10–40 per cent more than spinal glucose utilization.

Discussion

The spinal cord is one of the least metabolically active structures in the central nervous system.^{6,8,9} In the monkey^{8,9} and rat,⁶ for example, lumbar spinal cord gray matter utilizes glucose at a rate which is 50–60 per cent lower than the cerebral cortex. Moreover, the thoracic area has a lower rate of metabolism than lumbar or cervical spinal cord^{7,9} and spinal white and dorsal horn gray matter are less metabolically active than ventral gray matter.^{6,7,9} These regional variations in metabolic rate parallel and presumably account partially for regional differences in spinal cord blood flow. Thus, the low rates of perfusion in spinal white and dorsal horn gray matter and thoracic

spinal cord⁷ probably reflect the low metabolic demand of these areas.

Perhaps because of this low rate of metabolism (and, presumably, blood flow), the spinal cord may respond differently than the brain to anaesthesia. Halothane, for example, markedly increases, and pentobarbitone profoundly reduces cerebral blood flow,¹ yet both drugs evidently have similar effects on the spinal circulation of the dog.¹⁵ The specific nature of this spinal blood flow effect (i.e., whether it increases or decreases) is not established, however, because comparisons with the conscious state have not been reported and the studies have not been repeated. Insofar as metabolic changes in the spinal cord during anaesthesia are concerned, we recently reported⁶ that nitrous oxide increases both spinal and cerebral metabolic rate 15–25 per cent in paralyzed and mechanically ventilated rats. Pentobarbitone, on the other hand, decreases utilization of glucose by the spinal cord only 10–20 per cent while simultaneously reducing cerebral glucose utilization as much as 50 per cent.⁶ Halothane substantially decreases spinal glucose utilization (Table II) but resembles pentobarbitone in that it reduces spinal metabolism less than brain metabolism. Thus, halothane reduced spinal glucose utilization no more than 35 per cent but decreased glucose utilization as much as 45–70 per cent in eight of 16 brain regions examined (e.g., cortical, thalamic, and auditory structures (medial geniculate, inferior colliculus)). Since the anaesthetized animals were paralyzed and mechanically ventilated in order to prevent hypercarbia, it is not entirely accurate to attribute the effects reported here to halothane alone. For example, partial restraint may stress animals somewhat and some believe this leads to an artificially high control metabolic state.¹⁶ Nevertheless, a control group of partially restrained conscious animals seems preferable to paralyzed rats mechanically ventilated with N₂O because spinal glucose utilization is 15–25 per cent higher with the latter regimen.⁶ In addition, it is unlikely that muscle paralysis of anaesthetized animals materially affected the results because cerebral glucose utilization in our animals was comparable to that reported previously in spontaneously breathing halothane-anaesthetized rats³ and because barbiturates have similar regional cerebral metabolic effects in unparalyzed¹¹ and paralyzed¹⁷ animals.

Additional studies are required to determine whether less profound spinal than cerebral metabolic depression is characteristic of the anaesthetic state or, like patterns of regional brain metabolism during anaesthesia,^{2,3,6} is a function of the anaesthetic agent. It seems, however, that halothane and pentobarbitone are qualitatively similar in that both produce substantially greater metabolic depression in cortical and metabolically active subcortical structures than the spinal cord. The reasons for a lesser

metabolic response of the spinal cord than brain to anaesthesia are not known. Based on the established relationship between metabolic rate and neuronal activity in the central nervous system,¹⁸ we speculate that the spinal cord's low spontaneous metabolic rate reflects, in part, relatively low neuronal activity which, if eliminated, leads to only a small decrement in metabolism. It is interesting in this regard that spinal shock,⁹ which is characterized by paralysis and areflexia, and spinal anaesthesia¹⁹ are associated with only 10–20 per cent decreases in spinal glucose utilization.

This work suggests that one must extrapolate cautiously to the spinal cord from information on the cerebral metabolic effects of anaesthetics (particularly if the results are weighted toward cortical structures) because the brain and spinal cord evidently respond somewhat differently to anaesthesia. However, on the basis of evidence that some general anaesthetics decrease spinal metabolic rate and, presumably, alter spinal blood flow, concern that anaesthesia might bias the outcome of certain experimental studies (particularly those concerned with the pathophysiology or therapy of trauma or ischaemia) is warranted. On the other hand, the observation that spinal metabolism does not decrease as much as brain metabolism during administration of halothane or pentobarbitone suggests that metabolic depression may not figure prominently in the protective action of some anaesthetics in spinal low-flow states.^{20–22}

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Résumé

Les auteurs ont utilisé la méthode du 2-[14 C] déoxyglucose afin d'étudier l'utilisation du glucose cérébral et médullaire simultanément lors d'une anesthésie à 1.2 pour cent d'halothane chez des rats Sprague-Dawley adulte. Chez les animaux conscients ($n = 5$) le taux d'utilisation du glucose dans la matière grise médullaire lombaire était de 50 pour cent plus bas que celui du cortex cérébral. L'anesthésie à l'halothane ($n = 6$) a réduit le taux métabolique dans la moëlle épinière et dans le cerveau. L'utilisation du glucose par la moëlle a été réduite de 12-35 pour cent mais ceci était moindre que la diminution de 45-70 pour cent produite par l'halothane dans 8 des 16 structures cérébrales étudiées et ceci était indépendant de l'hypotension produite. Ces résultats indiquent que l'halothane est un dépresseur du métabolisme de la moëlle mais ses effets sur ce tissu sont substantiellement moindres que ceux qu'il possède sur plusieurs structures cérébrales.