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Title : EFFECT OF CHRONIC HALOTHANE EXPOSURE ON NEUROTRANSMITTER SYSTEMS IN DISCRETE BRAIN REGIONS

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Introduction. Recent studies indicate that chronic exposure to only trace levels of halothane (2-bromo-2-chloro-1,1,1-trifluoroethane), as may occur in the operating room environment, may have adverse effects on personnel. Halothane associated pathological changes in several organs, including the brain, have been reported.[1] In addition, halothane has been shown to produce changes in human behavior.[2, 3]

Since behavioral changes may be associated with changes in neurotransmitter systems, the present study was designed, using the rat as an experimental model, to assess the effect of chronic exposure to subanesthetic concentrations of halothane on adrenergic, dopaminergic, cholinergic and GABAergic systems in discrete brain nuclei.

Methods. Thirty six male Sprague Dawley rats weighing approximately 185 g were used. All rats had free access to normal laboratory diet and water.

Rats were randomly divided into two groups (18 rats/group), placed in covered cages (22x26x46 cm; 6 rats/cage) and designated for halothane or air (control) exposure. Exposures were for 8 hrs/day, 5 days/week during an eight week period. Each rat was weighed weekly.

Halothane was regulated with a Fluotec MK2 fluothane vaporizer, using compressed air, resulting in a flow of 0.25 percent halothane (2500 ppm) at two liters per minute in air.

Twenty four hours after the last exposure regime, the rats were sacrificed by decapitation; the brains were quickly dissected and frozen in dry ice. Samples of liver, muscle and serum were taken from each rat, quickly frozen and stored until further analysis for halothane content. Testes and adrenal glands were removed and weighed, as a measure of possible halothane associated stress.

The frozen brains were sliced into 300 µm sections, using a cryostat, and discrete regions were removed using cannulae of various diameter size (Table 1). The resulting tissue pellets were dispersed by ultrasound in various media, and aliquots subsequently taken for determination of protein[4], norepinephrine (NE)[5] and dopamine (DA)[5] content. In addition, activities of glutamic acid decarboxylase (GAD)[6] and choline acetyltransferase (ChAT)[7], marker enzymes for GABAergic and cholinergic systems respectively, were determined.

Results. No significant difference in mean body weight was observed between halothane exposed and control rats during the first three weeks of exposure period. On the fourth week of exposure, halothane exposed rats exhibited a significant ($P<0.01$) decrease in body weight compared to air-exposed (control) rats. This relative difference in mean body weight was apparent throughout the subsequent 4 week exposure period.

Gonad and adrenal weights of halothane-exposed animals were not significantly different from control rats.

Liver, muscle and serum from each halothane-exposed rat were assayed for halothane content. Halothane was not detected.

The concentration of NE and DA and the activity of GAD and ChAT were determined in twenty different brain regions of halothane exposed and control rats. Halothane exposed rats exhibited a significant ($P< 0.01$) 41% reduction in dopamine concentration in the ventral tegmental area compared to control rats. Halothane exposure had no significant effect on DA concentration of the other brain regions. In contrast, halothane exposure resulted in no significant changes in regional NE concentrations.

GAD activity of halothane exposed rats was significantly decreased in the nucleus accumbens ($P< 0.01$; 60%)and in the medial preoptic nucleus ($P< 0.01$; 35%) and significantly increased ($P<0.01$; 39%) in the lateral preoptic nucleus when compared to control animals. No significant difference in GAD activity in the other brain regions was observed.

ChAT activity was significantly increased in globus pallidus ($P<0.01$; 125%), caudate nucleus ($P<0.01$; 43%) and N. accumbens ($P<0.01$; 41%) and significantly decreased ($P<0.01$; 43%) in central gray of the halothane-exposed compared to control rats. The activity of ChAT was unaffected in the other brain regions studied.

Discussion. The present study clearly demonstrates that chronic exposure to subanesthetic concentrations of halothane results in significant changes in certain neurotransmitter systems in different discrete nuclei. Since the observed changes are not unidirectional or ubiquitous, it may be concluded that the changes would not have been apparent if neurotransmitter changes were assessed at the level of whole brain, or in gross brain structures.

It is interesting to note that the observed changes occurred in nuclei associated with motor activity (basal ganglia), emotional behavior (limbic structures, e.g., N. accumbens, ventral tegmental area and preoptic nuclei) and pain (central gray). The significance of these data in relation to the reported changes in behavior, manifested after chronic halothane exposure, cannot be assessed at present.

References.

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