Date :

Title : INACTIVATION OF METHIONINE SYNTHETASE (MS) BY NITROUS OXIDE

Authors : D. D. Koblin, Ph.D., J. E. Watson, J. E. Deady, M.S., E. L. R. Stokstad, Ph.D., and E. I. Eger, II, M.D.

Affiliation: Department of Anesthesia, University of California, San Francisco, California 94143, and Department of Nutritional Sciences, University of California, Berkeley, California 94720

Introduction. Exposure of rats to 50% N_2O for 6 hr completely inactivates liver methionine synthetase (MS),[1] a vitamin B_{12}-dependent enzyme that synthesizes the amino acid methionine from homocysteine. The inactivation of this enzyme may play a role in the development of polyneuropathy and anemia seen in subjects chronically exposed to subanesthetic[2] and trace levels[3] of nitrous oxide. Because the effects of this process may be important to patients and operating room personnel, we studied the time course of inactivation, its dependence on anesthetic concentration, the time course of recovery of activity, and the possibility that other anesthetics might also produce inactivation.

Methods. Groups of eight adult male CD-1 mice were placed in a 20-l hyperbaric chamber and exposed to a range of N_2O concentrations (0 to 0.8 atm N_2O) for varying periods of time (0 to 4 hr). They were also exposed to xenon (0.8 atm), nitrogen (0.8 atm), halothane (0.6% atm), isoflurane (0.6% atm), and enflurane (1.0% atm), all in the presence of 1 atm O_2. Anesthetic concentrations were determined by gas chromatography. Following anesthetic exposure, livers were removed and homogenized in 0.01 M phosphate buffer (pH 7.3). The homogenate was centrifuged at 20,000 g for 80 min. Methionine synthetase activity was measured by incubating an aliquot of the supernatant with [$^{14}CH_3$]-tetrahydrofolate and homocysteine at 37 C for 1 hr, and then measuring the amount of radioactive methionine produced.[4] Methionine synthetase activity is expressed as the nanomoles of methionine produced per hour per gram of liver.

Results. The MS activity in livers of mice exposed to 0.8 atm N_2O decreased progressively as exposure times increased (table 1). Periods of exposure as short as 15 to 30 min produced significant inactivation. Exposure to xenon did not affect MS activity (table 1). Methionine synthetase activity was progressively inhibited as N_2O concentrations increased (table 2). For 4-hr exposure periods, 50% inactivation was achieved by approximately 0.1 atm N_2O (table 2). Methionine synthetase activity returned to 70% of its control value one day after exposure to 0.8 atm N_2O for 4 hr (table 3). Recovery of enzyme activity was nearly complete two days after exposure to N_2O (table 3). Exposure of mice to anesthetizing levels of halothane, enflurane, and isoflurane did not affect MS activity significantly. We conclude that brief exposures to nitrous oxide inactivate an essential metabolic enzyme, an effect that appears to be confined to anesthetic or subanesthetic, but not trace, levels. Thus, our results may have greater implications for patients than for operating room personnel. Studying chronic exposure to trace levels may alter this impression of innocuousness to operating room personnel.

References

1. Deacon R, Lumb M, Perry J, et al: Selective inactivation of vitamin B_{12} in rats by nitrous oxide. Lancet II:1023-1024, 1978

2. Amess JAL, Burman JF, Rees GM, et al: Megaloblastic hemopoiesis in patients receiving nitrous oxide. Lancet II:339-342, 1978

3. Cohen EN, Brown BW, Wu M, et al: Anesthetic health hazards in the dental operatory. Anesthesiology 51(3S):254, 1979 (abstract)

4. Sauer HJ, Jaenicke L: Einfacher Test zur Messung der Methionin-Synthetase-(MS-) Activität und seine Anwendungsmöglichkeiten in der Klinik. Klin Wochenschr 50:986-990, 1972

Table 1. Time Course of N_2O Inactivation

Group	MS Activity (mean ± SE) (nmol methionine/g/hr)
Shelf controls	296 ± 32.1
(15 min) 0.8 atm N_2O	197 ± 11.7
(30 min) 0.8 atm N_2O	54 ± 8.3
(60 min) 0.8 atm N_2O	56 ± 4.6
(120 min) 0.8 atm N_2O	41 ± 19.2
(240 min) 0.8 atm N_2O	16 ± 5.5
(240 min) 0.8 atm N_2	166 ± 11.5
(240 min) 0.8 atm Xe	302 ± 30.7

Table 2. Concentration Dependence of N_2O Inactivation

Group	MS Activity (mean ± SE) (nmol methionine/g/hr)
Shelf controls	297 ± 68.3
0.003 atm N_2O (4 hr)	304 ± 18.9
0.010 atm N_2O (4 hr)	277 ± 5.9
0.049 atm N_2O (4 hr)	242 ± 10.7
0.20 atm N_2O (4 hr)	101 ± 3.1
0.79 atm N_2O(4 hr)	58 ± 3.0

Table 3. Recovery of MS Activity after a 4-Hr Exposure to 0.8 atm N_2O

Group	MS Activity (mean ± SE) (nmol methionine/g/hr)
Shelf controls	413 ± 32.0
0 day recovery	49 ± 9.0
1 day recovery	279 ± 14.2
2 day recovery	333 ± 25.0
4 day recovery	367 ± 40.1
8 day recovery	323 ± 14.8