Nitrous oxide (N_2O) toxicity and cobalamin (B_{12})-dependent metabolism

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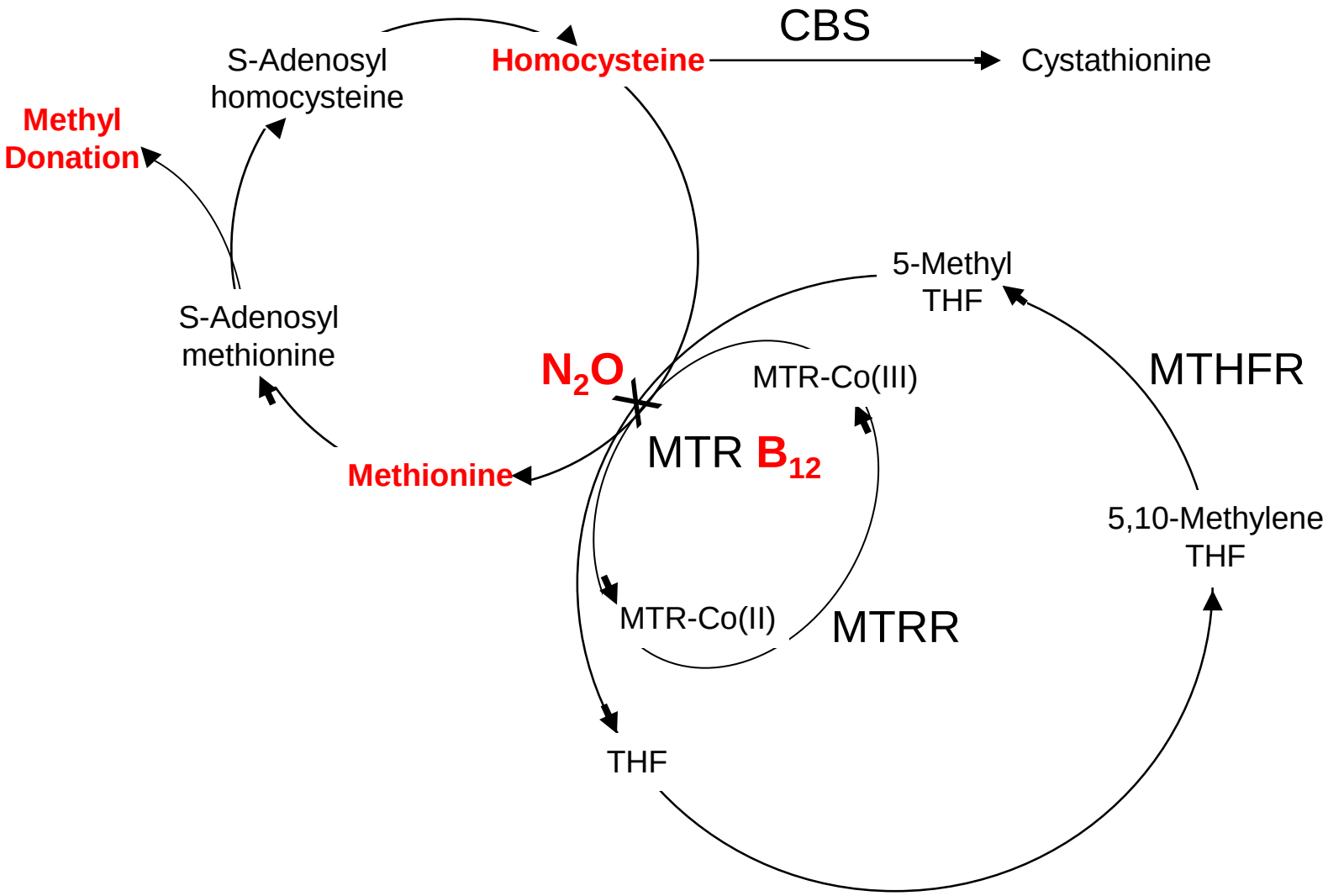
No conflicts

History of N_2O use

- 1810s - First recognition of N_2O analgesic properties
- 1840s - First demonstration of N_2O anesthetic properties
- 20th century - N_2O "balanced" with inhaled volatile agents to reduce toxicity and side effects
- 1938 - Food, Drug and Cosmetic Act
- § 201.161 Medical gases (April 1, 2017).
- "(a) Oxygen, nitrogen, carbon dioxide, helium, and **nitrous oxide** gases intended for drug use, and medically appropriate combinations of any of these gases intended for drug use, **are exempted from the requirements** of § 201.100(b)(2) and (3), and **(c)(1)"**
- "**(c)(1)**: (1) Labeling on or within the package from which the drug is to be dispensed bears adequate information for its use, including indications, effects, dosages, routes, methods, and frequency and duration of administration, and any **relevant hazards, contraindications, side effects, and precautions under which practitioners licensed by law to administer the drug can use the drug safely and for the purposes for which it is intended**, including all purposes for which it is advertised or represented;"
- **The FDA has not investigated or established the safety and efficacy of N_2O**
- Ask your colleagues and patients if they know this

History of N_2O toxicity

- Late 1940s - N_2O uniformly lethal in treatment of tetanus pain in post-war Europe
- 1970s - **Subacute** combined degeneration of the spinal cord after **recreational use**
- 1990s – **Acute** combined degeneration of the spinal cord after **clinical use** in standard concentrations and durations (e-mail for reference list)
- 1990s – N_2O NIOSH and OSHA regulated at **8 ppm** vs. FDA (and DEA) **un**regulated at **700,000** ppm +
- 2000 - **Acquired** predisposition to N_2O neurotoxicity (Felmet *et al*.)
- 2003 - **Genetic** predisposition to N_2O neurotoxicty (Selzer *et al*.)
- N_2O is the only inhaled anesthetic that will kill everyone who breathes it at clinical concentrations for 5-7 days
- Issues are individual susceptibility, duration and concentration of use, cellular reserves of methionine, and how much damage is acceptable


CBS
S-Adenosyl
homocysteine
Homocysteine
Cystathionine
Methyl
Donation
5-Methyl
THF
S-Adenosyl
methionine
N_2O
MTR-Co(III)
MTHFR
MTR B_{12}
Methionine
5,10-Methylene
THF
MTR-Co(II)
MTRR
THF

N_2O Inactivation of Methionine Synthase

- N_2O inactivates methionine synthase **in everyone**
- Mean half time of human hepatic methionine synthase inactivation - 45 minutes – **CNS unknown**
- Recovery > 4 days to resynthesize enzyme *i.e.*, **not** when nitrous oxide is DCed
- Serial exposure particularly harmful
- Human **fetal/neonatal** and **geriatric** kinetics of inactivation and reactivation are **unknown** in any tissue
- N_2O inhibition of methionine synthase is **rapid, potent, and irreversible in all patients,** and **harmful in a substantial proportion of patients** that are not identified before exposure
- Deleterious consequences are **long-lasting**, not “short acting”
- N_2O’s **toxicity is distinct from** N_2O’s **site(s) of anesthetic action**

Prevalent *MTHFR* Mutations

- MTHFR activity is reduced > 70% with common SNPs
- ***MTHFR* (C677T)** (Hogan *et al*. *Clin Med Res* 2009;7:69)
  - Homozygote – 13%
- ***MTHFR* (A1298C)**
  - Homozygote – 9%
- No compound homozygotes – lethal?
- % varies by geographic origin of ancestors
- Additional polymorphisms in intrinsic and extrinsic single carbon pathways

B-vitamin deficiency

- "The prevalence of vitamin **B12 deficiency**, whether defined as low vitamin B12 or metabolically significant vitamin B12 deficiency increased with age in all three studies, from about **1 in 20** among people aged 65-74 years **to 1 in 10 or even greater** among people aged 75 years or greater. The prevalence of **folate deficiency** also increased with age, and was **similar** to that for vitamin B12 deficiencies."
- Clarke *et al*. Vitamin B12 and folate deficiency in later life. *Age Ageing* 2004;33:34-41.
- **23% incidence of B12 deficiency in third trimester and childhood**
- Sukumar *et al*. Prevalence of vitamin B12 deficiency and its effect on offspring birth weigh: A systematic review and meta-analysis. *Am J Clin Nutr* 2016;103:1232-1251.
- Added to deficiencies in folate and B6
- **Combined genetic and acquired susceptibilities in at least 5% of patients**

Components of prospective study before and after N_2O exposure

- B **vitamin levels** in plasma, red blood cells and other cells
- Single carbon pathway **substrate and product levels** – homocysteine, methionine, etc.
- Single carbon pathway **genotypes and epi-genotypes (DNA methylation)**
- **Enzyme activities** in target tissues
- **Cellular indices** of bone marrow deficiency in circulating cells
- **Cellular indices of genotoxicity** *e.g.,* in cord blood, etc. (SCE, comet assay)
- **Clinical outcomes** – *e.g.,* neurotoxicity, marrow toxicity, other tissues, immediate **and** long term outcomes
- **Xenon** as a positive control

Informed consent: The precautionary principle

- N_2O has **no established benefit in anesthetic maintenance** compared to contemporary anesthetics
- N_2O use is exploding outside the O.R. – *e.g.,* labor and delivery, emergency rooms and ambulances, pain and psychiatry, pediatric procedural "sedation", dental and oral surgery
- Patients are **never tested** for susceptibilities
- Patients and families are **never informed** of specific risks (ask for anesthesia informed consent law review in *Journal of Law and Medicine* (in press))
- N_2O is a **bone marrow gas**
- N_2O is a **nerve gas**
- The effects of N_2O on **regeneration of tissues** after trauma and surgery have not been reported to date (ask to share submission-ready data)

???

- If newly developed and introduced, **could N_2O achieve FDA approval now**?
- Would a pharma executive wish to apply in view of minimal benefits and assured risks?
- Would academic investigators wish to conduct appropriate prospective trials?
- Would patients and parents of children wish to provide **fully informed consent (*i.e.*, "not FDA approved")** to participate in randomized trials antecedent to FDA approval?

Thanks!

Action Steps

- Must-read literature
- Clinical and research informed consent guidelines
- Research priorities
  - Labor & delivery
- Publication guidelines