

**Study Title:** Pharmacogenetics of Adverse Outcomes after Nitrous Oxide Anesthesia

**PI:** Peter Nagele, MD

**I. Abstract**

**Background and significance:** Recent studies have shown that nitrous oxide (N<sub>2</sub>O) anesthesia may be associated with an increased risk of adverse cardiovascular outcomes. It is well-known that N<sub>2</sub>O inhibits vitamin B<sub>12</sub>-dependent enzymes and as a result increases plasma homocysteine concentrations. Homocysteine has been identified as risk factor for cardiovascular disease. Therefore elevations in homocysteine after N<sub>2</sub>O may be a causative factor in N<sub>2</sub>O toxicity. In a previous investigation, we found that patients who carry a homozygous mutation in the *MTHFR* gene develop higher homocysteine levels after N<sub>2</sub>O anesthesia than non-carriers. These patients might be at higher risk for adverse cardiac outcomes from N<sub>2</sub>O. Thus, there may be a pharmacogenetic mechanism to account for the adverse cardiac outcomes from N<sub>2</sub>O. Moreover, prevention of N<sub>2</sub>O-increased homocysteine concentrations in these high risk patients by perioperative vitamin B<sub>12</sub> and folate supplementation might decrease the incidence of adverse cardiac outcomes.

**Hypothesis:** Patients carrying a homozygous *MTHFR* 677C>T or 1298 A>C variant allele will have a higher incidence rate of postoperative myocardial ischemia after N<sub>2</sub>O anesthesia [detected by serial TnI measurements] due to elevated homocysteine levels than normal "wild-type" non-carriers, and that the incidence rate will be reduced if they receive perioperative vitamin B<sub>6</sub>/B<sub>12</sub>/folate supplementation.

**Primary outcome:** Myocardial ischemia in the first 72 hours after surgery

**Outcome definition:** Peak serum troponin I concentration during the first 3 postoperative days

**Secondary outcome:** Composite endpoint of 30-day mortality and major cardiac morbidity (non-fatal MI)

**Design:** Randomized controlled trial. Patients will be randomized to receive vitamin supplementation or placebo before surgery. All patients will receive N<sub>2</sub>O during surgery. Mendelian randomization of *MTHFR* genotype.

**Intervention:** Vitamin B<sub>6</sub>/B<sub>12</sub>/folate supplementation preoperatively

**Study setting:** Barnes-Jewish-Hospital

**Patients:** Patients scheduled for major vascular surgery with previously diagnosed coronary artery disease

**Statistical Approach:** Comparison of two groups: *MTHFR* homozygous vs. heterozygous/wild-type patients. General linear model will be fit to the data after normalizing transformation (e.g. log troponin).

**Anticipated result:** Patients carrying a homozygous *MTHFR* 677C>T or 1298 A>C variant allele will have a 50% increased peak TnI due to elevated homocysteine compared to non-carriers. Secondly, treatment with vitamin B<sub>6</sub>/B<sub>12</sub>/folate will prevent the homocysteine increase as well as the increase in peak troponin and myocardial ischemia.