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REVIEW ARTICLE

Nitrous oxide and perioperative outcomes

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Abstract There is emerging evidence related to the effects of nitrous oxide on important perioperative patient outcomes. Proposed mechanisms include metabolic effects linked to elevated homocysteine levels and endothelial dysfunction, inhibition of deoxyribonucleic acid and protein formation, and depression of chemotactic migration by monocytes. Newer large studies point to possible risks associated with the use of nitrous oxide, although data are often equivocal and inconclusive. Cardiovascular outcomes such as stroke or myocardial infarction were shown to be unchanged in previous studies, but the more recent Evaluation of Nitrous Oxide in the Gas Mixture for Anesthesia I trial shows possible associations between nitrous oxide and increased cardiovascular and pulmonary complications. There are also possible effects on postoperative wound infections and neuropsychological function, although the multifactorial nature of these complications should be considered. Teratogenicity linked to nitrous oxide use has not been firmly established. The use of nitrous oxide for routine anesthetic care may be associated with significant costs if complications such as nausea, vomiting, and wound infections are taken into consideration. Overall, definitive data regarding the effect of nitrous oxide on major perioperative outcomes are lacking. There are ongoing prospective studies that may further elucidate its role. The use of nitrous oxide in daily practice should be individualized to each patient's medical conditions and risk factors.

Keywords Nitrous oxide · Perioperative outcomes · Perioperative mortality · Perioperative morbidity

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Introduction

Nitrous oxide (N_2O) has remained a widely used inhaled anesthetic agent for the last 150 years due to its availability across the world. However, controversies exist with regard to its potential association with increased perioperative mortality, wound healing, cardiovascular risk, and pulmonary complications, among many others [1]. For example, multiple studies have demonstrated that the metabolic effects of N_2O are linked to elevated plasma homocysteine levels as well as endothelial dysfunction perioperatively [2–4]. However, its clinical significance has not been well defined or clearly demonstrated. To date, the Evaluation of Nitrous Oxide in the Gas Mixture for Anesthesia (ENIGMA-I and ENIGMA-II) trials are the largest randomized controlled trials evaluating perioperative patient outcomes [5, 6]. These studies have significant potential to provide valuable insights into the influence of N_2O on patient morbidity and mortality. This review focuses on N_2O and its role in systemic effects and perioperative patient outcomes as suggested by recent literature (Table 1).

Cardiovascular system

N_2O has been implicated in increasing perioperative vascular risk, including stroke and myocardial infarction. This

Table 1 Major perioperative outcomes related to nitrous oxide use

Trial or study	Total number of patients	Main result
GALA trial [12]	1,615	No clinically significant adverse effects of N_2O in patients undergoing carotid surgery when adjusted for baseline risk factors
Kozmary et al. [13]	70	No increased risk of myocardial ischemia or infarction in patients undergoing carotid surgery
POISE trial [14, 15]	5,104	No increased risk of adverse outcomes in patients who underwent non-cardiac surgery. Observational study
Mitchell et al. [16] and Slavik et al. [17]	70 in [16]; 7 in [17]	No detectable myocardial ischemia, even in patients with significant coronary artery disease undergoing cardiopulmonary bypass grafting
ENIGMA-I trial [5]	2,050	Duration of hospital stay did not change. However, the N_2O group showed increased duration of intensive care and increased occurrence of postoperative pneumonia, pneumothorax, pulmonary embolism, wound infection, myocardial infarction, venous thromboembolism, stroke, death within 2 days of surgery, less severe nausea and vomiting, as well as an increase in healthcare costs
ENIGMA-II trial [6]	Pending	Ongoing trial in patients at risk of coronary artery disease undergoing non-cardiac surgery to determine cardiovascular outcomes
Turan et al. [19]	49,016	N_2O was associated with decreased odds of 30-day mortality and in-hospital mortality and morbidity
Fleischmann et al. [36]	418	No significant difference in wound infection rates
IHAST trial [51–53]	1,000	No long-term effect on neurological or neuropsychological functions
Beilin et al. [66]	455	No significant difference in the clinical pregnancy or delivery rate

is based on clinical trials; however, no large randomized controlled trials existed until ENIGMA-I and ENIGMA-II [3, 4, 7]. With regard to the potential for postoperative adverse cardiovascular events, the role of N_2O in mediating or modulating adverse outcomes has led to a decades-old debate about its role [8, 9]. Further, the administration of N_2O typically results in a decrease in the concentration of a more cardiovascular depressant anesthetic at equivalent minimum alveolar concentration. This offers circulatory stability related, in part, to both the sparing of the potent anesthetic and to the direct effect of N_2O on peripheral vascular resistance [10, 11]. As a result, it is often difficult, if not impossible, to quantify and to characterize the perioperative cardiovascular risks related to the administration of N_2O.

A secondary analysis of the General Anesthesia Compared with Local Anesthesia for Carotid Surgery trial (GALA) demonstrated that the patients (total: 1,615) who received N_2O were not more likely to suffer stroke or myocardial infarction within 30 days of the operation [relative risk 1.12; 95 % confidence interval (CI) 0.73–1.73; $p = 0.63$] [12]. The study did find that patients who were exposed to N_2O were more likely to have preexisting coronary artery disease, peripheral vascular disease, and atrial fibrillation. When the authors adjusted the difference in baseline risk factors using logistic regression, the odds ratio was reduced to 1.09 (95 % CI 0.68–1.74; $p = 0.73$), still suggesting that there is no clinically significant adverse effect of N_2O in patients undergoing carotid surgery. This finding is similar to that obtained in a previous study by Kozmary et al. [13] (total: 70 patients), in which the authors did not find any trend suggesting a greater incidence of myocardial ischemia or infarction in patients undergoing carotid surgery. Similarly, in a post hoc subanalysis of the Perioperative Ischemic Evaluation (POISE) trial, N_2O was not associated with an increased risk of adverse outcomes in patients who underwent non-cardiac surgery (total: 5,104 patients) [14, 15]. It should be noted that the study was limited by the observational nature of the data. In cardiopulmonary bypass grafting, Mitchell et al. [16] and Slavik et al. [17] investigated the effects of N_2O on segmental left ventricular function using two-dimensional transesophageal echocardiography and changes in the ST segment of the electrocardiogram approximately two decades ago, but did not find any clinically detectable myocardial ischemia, even in patients with known significant coronary artery disease.

Based on the above studies, there does not seem to be any significant cardiovascular consequences of N_2O. However, the ENIGMA-I trial suggests otherwise.

The ENIGMA-I trial randomized 2,050 patients who underwent non-cardiac surgery lasting for more than 2 h to N_2O-based or N_2O-free anesthesia in 2007, with the primary endpoint being the duration of hospital stay [5]. The median duration of hospital stay did not differ between the two groups (7.0 vs. 7.1 days; $p = 0.06$). Many of the secondary endpoints, which included duration of intensive

care, the occurrence of postoperative pneumonia, pneumothorax, pulmonary embolism, wound infection, myocardial infarction, venous thromboembolism, stroke, and death within 2 days of surgery, did show that patients in the N_2O-free group had significantly lower rates of major complications (odds ratio 0.71; 95 % CI 0.56–0.89; $p = 0.003$) and less severe nausea and vomiting (odds ratio 0.4; 95 % CI 0.31–0.51; $p < 0.001$). Furthermore, they were more likely to be discharged from the intensive care unit on any given day than those in the N_2O-based group (hazard ratio 1.35; 95 % CI 1.05–1.75; $p = 0.02$). A long-term follow-up study (median 3.5 years) additionally showed that administration of N_2O was associated with increased long-term risk of myocardial infarction but not death or stroke (odds ratio 1.59; 95 % CI 1.01–2.51; $p = 0.04$) [18].

In 2013, a propensity-score-matched retrospective cohort analysis of 49,016 patients by Turan et al. [19] revealed that N_2O was associated with decreased odds of 30-day mortality and decreased odds of in-hospital mortality and morbidity. This contradicted the findings in ENIGMA-I. However, even though it included a large patient population, this study is difficult to interpret given its retrospective nature as well as the inherent limitations of propensity-score-matched comparisons in clinical studies [19–21]. Thus, the major difference between these two studies arises mainly from the nature of their design. Propensity score matching was applied in the study by Turan et al., which utilizes a non-experimental retrospective causal inference method to reduce bias in the clinical data. Specifically, each subject in the database is assigned a composite propensity score that is derived using multivariable logistic regression and takes these baseline characteristics into account. One subject in the group that received N_2O was matched to another subject in the group that did not receive N_2O according to their composite propensity score. Thus, this study is based on the assumption that subjects with similar propensity scores share similar baseline characteristics, and therefore such a process resembles a randomized controlled trial. On the other hand, in the ENIGMA-I study, a randomized controlled trial method that is considered the "gold standard" is employed. While there are many attractive features of propensity score matching, such as lower cost, shorter time to completion, and easy access to a larger group of subjects from a well-established database, the accuracy of this method requires prior knowledge of all pertinent covariates, so the propensity model remains valid. On the other hand, if there are unobserved and unbalanced variables then it is impossible to eliminate hidden bias using propensity score matching. As a result, a randomized controlled trial—despite its limitations and the difficulty involved in implementing it—is preferred for this exact reason, since it is capable of eliminating unknown bias. In summary, both studies provide additional insights into the safety profile of N_2O, but one needs to be cautious about drawing any definitive conclusions based on any single study, especially when the conclusions contradict one another. Finally, the decision to use N_2O should be made on an individual patient basis.

Given the conflicting data, a follow-up trial of 7,000 patients at risk for coronary artery disease (the ENIGMA-II trial) was designed, and to date has enrolled slightly over 1,000 subjects who are undergoing non-cardiac surgery [6]. As clarification emerges in the coming years with the conclusion of the ENIGMA-II trial regarding the role of N_2O in influencing cardiovascular outcomes, the agent should be tailored to each patient's needs as well as the individual cardiovascular response to N_2O.

Pulmonary system

It is well known that elimination of N_2O leads to diffusion hypoxia during emergence from anesthesia [22, 23]. As a result, one might postulate that those who receive N_2O during surgery might have a higher risk of developing complications. However, there was only scant evidence of such causality until the ENIGMA-I trial.

In the 1970s to 1980s, small randomized controlled trials with numbers of patients of <20 did not show any clinical significance in regards to pulmonary complications [24, 25]. The largest randomized controlled trials that study postoperative hypoxia and its relationship to N_2O were performed in the 1990s by Lampe et al. [26, 27]. Lampe randomized 270 patients to receive 60 % N_2O/40 % oxygen (O_2) or 100 % O_2 in various surgical settings that included total hip arthroplasty, carotid endarterectomy, and transsphenoidal hypophysectomy. While it was observed that patients in the N_2O group reached a lower intraoperative O_2 saturation that was statistically significant (but not clinically significant), there was no difference in postoperative O_2 saturation, the incidence of postoperative hypoxemia, cough, or sputum production. Furthermore, the group investigated long-term exposure to N_2O [26]. The second study was limited to patient populations who underwent surgical resection of acoustic neuroma lasting approximately 10 h [27]. Again, there was no increased incidence of postoperative hypoxemia, although only 26 patients were enrolled in the study.

In the ENIGMA-I trial, however, there was a significant statistical difference between the N_2O-free group and N_2O-based group in all areas of pulmonary complications. This included pneumonia (adjusted odds ratio 0.51; 95 % CI 0.27–0.97; $p = 0.04$) and atelectasis (adjusted odds ratio 0.55; 95 % CI 0.40–075; $p < 0.001$) [5]. There was no

difference in the duration of hospital stay between the study groups.

Given the conflicting data presented above, it is not unreasonable to remain conservative in one's daily practice and thus to avoid using N_2O, especially in patients prone to postoperative pulmonary complications. On the other hand, one should remember that many factors contribute to postoperative respiratory complications or hypoxemia. For example, age, as well as specific surgical sites, tend to play a more significant role than the type of inhalation agents employed intraoperatively [28, 29]. Meanwhile, although total intravenous anesthesia provides a different pharmacokinetic profile from inhalational techniques, there are conflicting results related to postoperative hypoxemia. Although at least two recent studies indicated an association between clinically significant postoperative hypoxemia and N_2O [30, 31], one showed no such correlation [32].

Surgical wound infection

Postoperative wound infection is a serious complication that can not only unnecessarily prolong hospital stay but also substantially increase healthcare costs. While multiple factors contribute to the frequency of surgical wound infection, including underlying immunocompromised states and timely administration of prophylactic antibiotics, the use of N_2O has been implicated due to in vitro evidence of its inhibition of deoxyribonucleic acid (DNA) formation via the inactivation of vitamin B_{12}, its inhibition of protein formation via the inhibition of methionine production, and its depression of chemotactic migration by monocytes [33–35]. As a result, Fleischmann et al. [36] conducted a randomized controlled study that involved 418 patients who were scheduled for colon resection and received either 65 % N_2O or 65 % nitrogen, with the primary outcome being the incidence of clinical postoperative wound infection. It was found that infection rates did not differ between patients who were given intraoperative N_2O and those who were given nitrogen (8 % in the N_2O group and 11 % in the nitrogen group, $p = 0.287$). While there are various definitions of surgical wound infection, Fleischmann's study demonstrated that, regardless of the definition, there was no statistical difference between the two groups. In fact, based on this study, the effect of N_2O on wound infection, if any, is trivial compared to other known perioperative factors such as body temperature and glucose control. The discrepancy between laboratory study and clinical experience is puzzling; although, one possible explanation is that the combination of N_2O and other inhalation agents leads to a lower requirement for other inhalation agents, since the minimum alveolar concentration is additive, which also impairs immune function, at least to some degree [37, 38]. It is, therefore, perceivable that the degree of immunosuppression is quite similar as long as inhalation agents are employed, regardless of whether it includes N_2O or not.

On the other hand, the ENIGMA-I trial demonstrated an adjusted odds ratio of 0.72 (95 % CI 0.52–0.98; $p = 0.036$), suggesting that more wound infections occurred in the N_2O-based group. However, it is possible that a higher inspired O_2 concentration in the N_2O-free group might have contributed to the lower postoperative surgical infection rate [39, 40].

Overall, given the complexity and multifactorial nature of postoperative surgical wound infection, it is not surprising that the question of the influence of N_2O administration on surgical wound infection remains largely unanswered.

Neuropsychological effects

The effect of N_2O on neuropsychological function has been an area of controversy, even in animal models. Some studies demonstrate preferable outcomes, while others report detrimental results [41–44]. Unfortunately, only a handful of randomized controlled trials have investigated functional outcomes. Most of those were limited to neurological outcome following cardiopulmonary bypass [45–47]. In theory, N_2O increases cerebral metabolic rate, cerebral blood flow, and therefore intracranial pressure [48]. N_2O also has the potential to increase intracranial air volume [49, 50]. In the setting of cerebral ischemia, glutamate excitotoxicity is known to exacerbate neurological injury, and blockade of *N*-methyl-D-aspartate (NMDA) receptors may attenuate the injury. In the latter case, since N_2O is a known NMDA antagonist, it is not surprising to see a reduction in infarct size after focal cerebral ischemia in animals [41]. The Intraoperative Hypothermia for Aneurysm Surgery Trial (IHAST) was a prospective randomized controlled trial which investigated the effect of intraoperative hypothermia in patients with aneurysmal subarachnoid hemorrhage who underwent surgical clipping [51]. Given the significant risk of developing cerebral ischemic events during aneurysmal clipping, the IHAST's database offers a unique opportunity to gain insights into both short- and long-term effects of N_2O on neurological outcomes via post hoc analysis [52, 53]. The initial post hoc analysis by McGregor et al. [52] included 1,000 patients and did not find any difference in delayed ischemic neurological deficit (20.1 % in the N_2O-free group vs. 24.5 % in the N_2O group; $p = 0.157$). The second post hoc analysis by Pasternak et al. [53] did show a greater risk of this short-term outcome (odds ratio 1.78; 95 % CI

1.08–2.95; $p = 0.025$) when the patient population was limited to those who received temporary proximal artery occlusion. In this second subgroup analysis, any intraoperative ischemic events would have occurred during exposure to N_2O, which argues against the use of N_2O when the risk of intraoperative ischemic events, whether incidental or intentional, is substantial. On the other hand, the long-term outcomes, such as any metric of neurological or neuropsychological function at 3 months, duration of hospital stay, or discharge destination, remained similar in both studies, which also suggests that the severity of delayed ischemic neurological deficit associated with N_2O was not sufficient to influence long-term outcomes. Admittedly, one has to acknowledge the limitations of both studies given the nature of post hoc analysis and potential biases. Yet, it would seem reasonable to assume that the use of N_2O does not appear to have a strong association with long-term adverse neurological outcome, even when patients are at substantial risk of developing neurological complications postoperatively.

Hepatobiliary system

Overall, in patients without pre-existing hepatic dysfunction, the incidence of serious hepatic insult from anesthetics is extremely low, with the exception of halothane [54]. While there have been in vitro studies that have demonstrated damage via changes in hepatic enzymes upon the administration of N_2O [55, 56], the clinical significance of such derangement remained unanswered until the randomized controlled trial performed by Lampe et al. [57] in 1990, where 100 patients were randomized to receive either 60 % N_2O/40 % O_2 or 100 % O_2 while undergoing total hip arthroplasty. The study did not find any evidence that suggested any linkage between N_2O and hepatic injury as estimated by liver function tests postoperatively.

Reproductive system

The reproductive consequences of the administration of N_2O have remained (and probably will remain) controversial, given the absence of prospective, randomized, blinded trials of such exposure due to ethical concerns. Numerous animal studies have demonstrated a teratogenic effect of N_2O [58–60]. However, it is difficult, if not impossible, to extrapolate the results from animal studies due to species differences. Unlike other inhalation agents, N_2O inactivates methionine synthase, which leads to inhibition of the conversion of homocysteine and methyltetrahydrofolate into methionine and tetrahydrofolate [61]. In particular, methionine is an essential amino acid, and tetrahydrofolate is important in the production of DNA. As a result, it is logical to cast doubts when it comes to using N_2O in pregnant patients. Retrospective epidemiologic studies using self-filled questionnaires have shown an association between N_2O exposure and adverse reproductive events, such as reduced fertility, lower birth weight, and higher risk of spontaneous abortion [62–65]. Yet, one needs to bear in mind the significant limitations of such studies, given their retrospective nature and potential biases and confounders, both known and unknown. More recently, Beilin et al. [66] conducted a retrospective multicenter anesthetic chart review in patients who underwent gamete intrafallopian transfer (GIFT) in order to examine whether anesthetic agents affect reproductive outcome. There was no statistically significant difference in the clinical pregnancy or delivery rate regardless of the anesthetic technique applied in this study. Again, while this result may be encouraging, the decision to include N_2O as part of the anesthetic plan for each patient needs to be individualized, given the limited scientific evidence to support one technique over another.

Hematologic system

Most of the hematologic effects of N_2O relate to possible side effects of bone marrow suppression, cobalamin deficiency, and folate deficiency, although studies have not been consistent in demonstrating such results, and many of them suffer from a limited sample of study subjects. Waldman measured red blood cell count, hemoglobin, mean red blood cell volume, reticulocyte count, platelet count, mean platelet volume, blood leukocyte level, and leukocyte differential in two groups of patients (a total of 49 patients), but did not show any clinical significance within the mean follow-up duration of < 2 weeks [67]. When the follow-up duration was extended to 3–5 weeks after brief N_2O exposure (40–80 min), as in the study of Deleu et al. [68], there was a reduction in serum folate level, especially in the elderly population, where symptoms of folate deficiency were observed in 3 patients (out of a total of 51 patients). These patients had preexisting low red blood cell folate levels and were randomized to an N_2O anesthetic. However, it is hard to explain why such symptoms were not apparent earlier. In addition, whether or not preoperative administration of folate or methionine loading would mitigate these side effects also remains controversial [69–72]. The paucity of evidence of adverse outcomes makes it challenging to draw a definitive conclusion, except to suggest that N_2O is relatively harmless for a limited duration in patients without any predisposing risk factors [73].

When it comes to bone marrow suppression, controversy has emerged, as some investigators strongly believe in the detrimental effect of N_2O while others have concluded that the agent has no significant side effects [74–76].

Cost analysis

One of the arguments for administering N_2O as part of routine anesthetic practice is its affordability. However, a retrospective cost analysis of the ENIGMA trial proves otherwise [77]. The rates of certain complications, including severe nausea or vomiting and wound infection, were higher in the N_2O group and resulted in a higher cost overall (total costs in the N_2O group were \$16,203 vs. \$13,837 in the N_2O-free group, $p = 0.002$). As a result, it is difficult to argue for the routine use of N_2O on the basis of its affordability.

Postoperative nausea and vomiting

N_2O has been linked to increased incidence of postoperative nausea and vomiting (PONV), and many have proposed to remove N_2O during anesthesia to reduce this risk in patients susceptible to PONV [78]. Given that PONV is a multifactorial phenomenon, it is challenging to quantify the extent of N_2O involvement. However, it is known that N_2O tends to cause bowel extension and produces changes in middle ear pressure [79–81]. It has also been shown that N_2O may act on dopamine and opioid receptors in the brain, which offers a possible mechanism for PONV associated with N_2O [82]. As a result, it is quite reasonable to avoid N_2O in certain patient populations when one wishes to minimize the risk of PONV.

Pediatric population

Nitrous oxide has long been used in the pediatric population for the purposes of sedation and general anesthesia [83]. Although systemic side effects are known to occur in both adult and pediatric populations, recent large outcomes studies [3, 5, 7, 12] did not specifically address this population, and therefore the short- and long-term effects of nitrous oxide use remain unclear. It is known, however, that nitrous oxide can have significant negative effects in patients with vitamin B_{12} deficiency and related inborn errors of metabolism. A recent case of the neurologic deterioration and death of a child who was anesthetized with nitrous oxide before the diagnosis of severe 5,10-methylenetetrahydrofolate reductase (MTHFR) deficiency was established has been reported [61]. MTHFR catalyzes the synthesis of 5-methyltetrahydrofolate. Therefore, any patient with a clinical presentation which fits that of severe MTHFR deficiency should be evaluated, and the diagnosis should be ruled out before nitrous oxide is administered.

Nitrous oxide has been found to increase homocysteine levels in children as well as adults [84]. The relative increase in the homocysteine level appears to be greater in children than in adults. Recent data also suggest that exposure for > 2 h is associated with a statistically significant increase in the postoperative homocysteine plasma concentration the morning after surgery in young children. The clinical significance of this increase is unknown [84–86].

Conclusion

Anesthesiologists are trained to minimize risk, to be prepared for the unexpected, and to be confident in their decisions. While large randomized controlled trials offer the best opportunity to shed light on the deep dark woods of ambiguity, we forget that such evidence is expressed on a continuum scale of reliability and applicability, whereas decision-making for each patient is a categorical process: yes or no. Uncertainty will therefore always remain, and it is paramount for physicians to recognize this intrinsic quality in the daily practice of medicine so that, through experience, one is capable of navigating through the sea of uncertainty with grace and wisdom.

While some may argue that N_2O should be removed from modern-day anesthesia practice given that there are many other alternatives that can be used to deliver anesthesia, we believe that such a conclusion may not be supported by the current literature.

In most instances, there is a lack of definitive data in most large-scale clinical trials to suggest the precise role of N_2O in the resultant morbidity or mortality. Ultimately, the decision of whether to utilize N_2O is left to each anesthesiologist's clinical judgment. Although many may argue that the choice of agent is not critical to the outcome based on the current literature, we believe that carefully weighing the risks and benefits for each patient and evolving clinical studies will allow practitioners to make the best decision regarding this interesting anesthetic agent in daily practice.

Conflict of interest None.

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