Nitrous oxide induces subacute combined degeneration by affecting vitamin B12 metabolism

Historical documents give inconsistent statements about the timing of the separation of nitrous oxide (N_2O). The first synthesis of N_2O is generally believed to have been made by Joseph Priestley in 1776.[1-3] By 1793, Dr. Thomas Cochrane had moved from Oxford to Brisbane, where he established pneumatic mechanisms including N_2O.[1,4] Subsequently, Humphrey Davy discovered that N_2O can reduce pain during the surgery and gave an extensive N_2O lecture. In 1844, Horace Wells realized that the gas could be used as an anesthetic[5] and extracted one of his molars under N_2O anesthesia. N_2O played an important role in dentistry after the 1870s, and subsequently, in the 1930s, N_2O became the mainstay of intrapartum pain relief. N_2O was invented by European scientists in the 18th and 19th centuries and has been used as an anesthetic for over 150 years.[6]

N_2O is used clinically as a safe anesthetic, especially in dentistry and pregnancy, while in other respects, according to the literature, N_2O shows early promise as a rapid antidepressant in patients with the treatment-resistant major depressive disorder.[7,8] The neuroprotective effect of N_2O on cerebral ischemia/reperfusion,[9] N_2O did not improve the neuroprotective effect after cerebral ischemia/reperfusion. Ischemic stroke is the most common cerebrovascular accident with high morbidity and mortality,[10] but the incidence of subacute comorbid myelopathy is also increasing, and N_2O reuptake has increased in the adolescent population in recent years.[11] Global Drug Survey 2012, an international online survey of young people, with more than 22,000 respondents reported that nearly half of UK respondents had used N_2O repeatedly at some point, 10% in the past 12 months.[12] In the 2016 Global Toxicology Survey, N_2O accounted for 8.7%[13] and was the second most common drug in this age group. N_2O is a very commonly used drug, especially in the United Kingdom and the United States (lifetime prevalence rates of 38.6% and 29.4%).[14] The number of published papers and related patients has increased rapidly every year since the first case was reported in China in 2016.[15] In conclusion, the use of N_2O outside of hospitals is gradually increasing, especially among adolescents.

Clinical manifestations and mechanisms: The clinical manifestations of subacute combined degeneration (SCD) caused by N_2O abuse are nonspecific and are mainly divided into chronic and subacute onset. Low doses of N_2O can produce pleasurable sensations and even hallucinations, but these disappear quickly. Of course, all patients with N_2O abuse experience symptoms of neurotoxicity, mainly including numbness and weakness of the limbs and decreased sensation, autonomic dysfunction, and cognitive impairment. The mechanism by which N_2O abuse causes SCD remains unclear but appears to be due to vitamin B12 deficiency.[16] Long-term inhalation of N_2O can interfere with the metabolism of vitamin B12 and may cause pathological changes due to its deficiency.[17] With prolonged inhalation time and increased inhalation volume, N_2O will oxidize cobalt ions, resulting in vitamin B12 as a coenzyme inactivation of metabolic processes (**Figure 1**),[18] preventing homocysteine from being converted to methionine, and methylmalonyl-coenzyme A cannot be isomerized to succinyl-coenzyme A,[19] resulting in degeneration of the central nervous system and formation of SCD in the myelin sheath and spine.

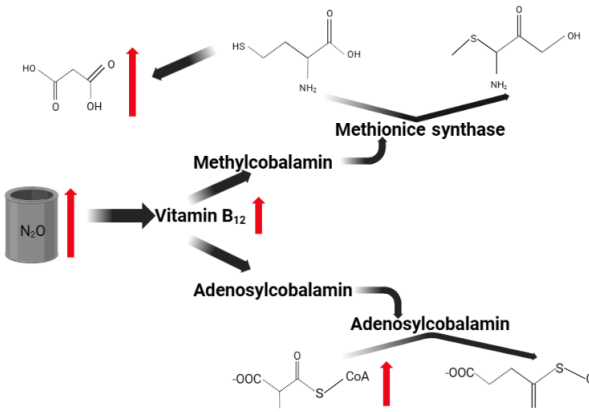

Figure 1: Simplified diagram of increased homocysteine and methyl-malonic acid levels due to inactivation of vitamin B12.
Note: Created with BioRender.com. CoA: Coenzyme A; MMA: methylmalonic acid.

Diagnosis, treatment, and prognosis: Diagnosis and differential diagnosis are usually based on the patient's clinical manifestations, biochemical tests, and imaging studies. Biochemical tests show mainly a decrease in vitamin B12, an increase in methylmalonic acid and homocysteine,[20-22] and magnetic resonance imaging (MRI) showed an abnormal T2-weighted signal in the spine.[23] Cessation of N_2O and oral or intramuscular vitamin B12 supplementation are the mainstays of treatment,[24] and with reasonable treatment, neurological symptoms may take months to resolve, and many patients report residual symptoms.[14,25,26] The speed and extent of recovery are inversely correlated with N_2O intake and the extent of the spinal cord and peripheral nerve damage.

Methods: Using "nitrous oxide (N_2O)" and/or "subacute combined degeneration (SCD)" as the keywords, a literature search was conducted through the PubMed database with the following inclusion criteria: (1) literature case reports from 2015 to 2020; (2) history of N_2O abuse; (3) age ≥ 18 years old; (4) have neurological signs and symptoms; (5) exclude other diseases that lead to decreased vitamin B12, such as autoimmune gastritis, congenital vitamin B12 deficiency, and folic acid deficiency; (6) only cases reported in English. The clinical manifestations, physical examination, biochemical examination, imaging examination, treatment, and prognosis were analyzed for the eligible cases. The case report was approved by Ethics Committee of Nantong First People's Hospital (No. 2023KT102) and obtained written informed consent from each patient.

Case report: We included 16 N_2O abusers from 2015 to 2020 (**Additional Table 1**), all of whom were adolescents and had prominent neurological symptoms at the onset of symptoms, the most common being numbness, weakness, and motor paresthesia. Neurological examinations mainly indicated Romberg's sign

positive, Babinski's sign positive, and Brudzinski's sign positive. Biochemical tests indicated decreased vitamin B12, with or without increased methylmalonic acid and (or) homocysteine levels. A small number of patients developed megaloblastic anemia. On imaging, MRI T2 signals showed varying degrees of hyperdensity in the spine, with an inverted V or "rabbit ear" sign on cervical MRI in some patients. Treatment mainly included discontinuation of N_2O and oral or intramuscular injection of vitamin B12. The serum vitamin B12 level of the patient improved rapidly, and neurological symptoms also recovered to varying degrees.

Discussion: With the increase in N_2O use in adolescents, the incidence of SCD gradually increases. Why does excessive N2O cause SCD? The pathogenic mechanism of N_2O remains unclear, and numerous studies have shown that N_2O oxidizes cobalt ions in vitamin B12, rendering it inactive.[27] Cobalamin is mainly used as an enzyme cofactor in the human body. Inactive cobalamin prevents the body from synthesizing succinyl-coenzyme A and methionine, which are essential for the methylation of myelin sheaths, thus, leading to demyelination of the nervous system.[28] Furthermore, serum homocysteine in patients with N_2O abuse is closely associated with nail thrombosis.[29]

Additional Table 1 shows 17 patients, all of whom ingested N_2O for at least several weeks to as long as 3 years. These adolescents obtained N_2O because small N_2O steel tanks were sold in foreign supermarkets as a food additive, resulting in the foaming structure of fresh milk powder,[30] while N_2O is readily available, cheap, and legal in adolescents and thus cannot be detected in routine positive drug screening. When adolescents inhale N_2O, the partial pressure of N_2O first increases in the lungs and then increases in the blood. N_2O acts quickly after inhalation for a few seconds, and its high elimination rate makes the patient return to normal quickly. The gas, N_2O, has minimal short-term effects on heart function, making it considered safe for adolescents. This is due to its quick and complete elimination from the body after inhalation. However, a potential concern is that adolescents may not recognize the side effects of N_2O and may consume it for extended periods of time. And yet another recent lockdown due to the coronavirus disease 2019 pandemic was associated with higher rates of psychiatric symptoms and substance abuse,[31] including N_2O use.

In the majority of patients with a history of N_2O abuse and neurological symptoms consistent with it, the diagnosis is relatively straightforward, but clinically it is also crucial to emphasize the consideration of N_2O abuse in the differential diagnosis because some patients have atypical neurological signs and symptoms. A normal serum vitamin B12 level does not indicate accurate or timely cellular availability of vitamin B12,[32] as N_2O causes vitamin B12 inactivation rather than a true deficiency. In addition to patients with reduced vitamin B12, there are other factors that can cause a decrease in vitamin B12 levels, including metabolic diseases, inflammatory infections, and tumors. It is essential to identify and diagnose these underlying diseases. Differential diagnosis of diseases that lead to vitamin B12 reduction, such as advanced deficiency, requires all clinicians to perform additional differential diagnostic tests, such as checking the patient's homocysteine and methylmalonic acid levels. It is used to check the differential diagnosis of spinal cord MRI and other semen lesions, such as neurosarcoidosis, central and peripheral neuropathy caused by acquired immune deficiency syndrome and syphilis, peripheral nerve vasculitis, etc. Classical SCD lesions are hyperintense on posterolateral T2-weighted spinal cord and can appear as an inverted V shape on axial MRI,[33,34] but only half of the patients with N_2O-related spinal cord subacute lesions have MRI abnormalities. Therefore, to diagnose a patient, we should perform a physical examination. According to the main symptoms of the patient, the etiology of subacute spinal cord lesions was excluded, and comprehensive judgment was made based on biochemical examination and imaging examination.

Conclusion: Taken together, N_2O abuse mostly leads to subacute lesions of the spinal cord, mainly manifested as limb weakness, dyskinesia, and paresthesia, accompanied by decreased serum vitamin B12 and abnormal T2 signal high density in the posterior and lateral columns of the spinal cord. Among these people who abuse N_2O, the main group is adolescents, which is related to their lack of awareness of the harm of N_2O and national policies.

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Additional file

***Additional Table 1:** Clinical characteristics, treatment and prognosis of 16 patients with nitrous oxide abuse.*

References

1. Buslov A, Carroll M, Desai MS. Frozen in time: a history of the synthesis of nitrous oxide and how the process remained unchanged for over 2 centuries. *Anesth Analg.* 2018;127:65-70.
2. Smith WD. A history of nitrous oxide and oxygen anaesthesia. IA. The discovery of nitrous oxide and of oxygen. *Br J Anaesth.* 1972;44:297-304.
3. Lew V, McKay E, Maze M. Past, present, and future of nitrous oxide. *Br Med Bull.* 2018;125:103-119.
4. Levere TH. Dr. Thomas Beddoes and the establishment of his pneumatic institution: a tale of three presidents. *Notes Rec R Soc Lond.* 1977;32:41-49.
5. Gillman MA. Mini-Review: A brief history of nitrous oxide (N_2O) use in neuropsychiatry. *Curr Drug Res Rev.* 2019;11:12-20.

6. Schwilden H, Schüttler J. 200 years of nitrous oxide (laughing gas)--and the end of an era? *Anasthesiol Intensivmed Notfallmed Schmerzther*. 2001;36:640.
7. Nagele P, Zorumski CF, Conway C. Exploring nitrous oxide as treatment of mood disorders: basic concepts. *J Clin Psychopharmacol.* 2018;38:144-148.
8. Nagele P, Duma A, Kopec M, et al. Nitrous oxide for treatment-resistant major depression: a proof-of-concept trial. *Biol Psychiatry.* 2015;78:10-18.
9. Hoffmann U, Sheng H, Ayata C, Warner DS. Anesthesia in experimental stroke research. *Transl Stroke Res.* 2016;7:358-367.
10. Zhao T, Zhu T, Xie L, et al. Neural stem cells therapy for ischemic stroke: progress and challenges. *Transl Stroke Res*. 2022;13:665-675.
11. Domont P, Debaize S. Extra hospital use of nitrous oxide for recreational purposes. *Rev Prat*. 2020;70:1143-1149.
12. Winstock AR, Griffiths P, Stewart D. Drugs and the dance music scene: a survey of current drug use patterns among a sample of dance music enthusiasts in the UK. *Drug Alcohol Depend.* 2001;64:9-17.
13. GOV.UK. Drug misuse: findings from the 2018 to 2019 CSEW. https://www.gov.uk/government/statistics/drug-misuse-findings-from-the-2018-to-2019-csew. Accessed March 11, 2020.
14. Kaar SJ, Ferris J, Waldron J, Devaney M, Ramsey J, Winstock AR. Up: The rise of nitrous oxide abuse. An international survey of contemporary nitrous oxide use. *J Psychopharmacol.* 2016;30:395-401.
15. Zheng D, Ba F, Bi G, Guo Y, Gao Y, Li W. The sharp rise of neurological disorders associated with recreational nitrous oxide use in China: a single-center experience and a brief review of Chinese literature. *J Neurol.* 2020;267:422-429.
16. Agarwal P, Khor SY, Do S, Charles L, Tikaria R. Recreational nitrous oxide-induced subacute combined degeneration of the spinal cord. *Cureus.* 2021;13:e19377.
17. Hathout L, El-Saden S. Nitrous oxide-induced B12 deficiency myelopathy: perspectives on the clinical biochemistry of vitamin B12. *J Neurol Sci.* 2011;301:1-8.
18. Vallès M, Soler D, Albu S, Kumru H. Neurological complications of nitrous oxide consumption. A case report. *Rev Neurol*. 2021;72:261-262.
19. Al-Sadawi M, Claris H, Archie C, Jayarangaiah A, Oluya M, McFarlane SI. Inhaled nitrous oxide 'whip-its!' causing subacute combined degeneration of spinal cord. *Am J Med Case Rep*. 2018;6:237-240.
20. Saji AM, De Jesus O. *Spinal cord subacute combined degeneration*. Treasure Island (FL): StatPearls Publishing; 2023.
21. Temple C, Horowitz BZ. Nitrous oxide abuse induced subacute combined degeneration despite patient initiated B12 supplementation. *Clin Toxicol (Phila).* 2022;60:872-875.
22. Bao L, Li Q, Li Q, et al. Clinical, Electrophysiological and radiological features of nitrous oxide-induced neurological disorders. *Neuropsychiatr Dis Treat.* 2020;16:977-984.
23. Check L, Abdelsayed N, Figueroa G, Ragunathan A, Faris M. Subacute combined degeneration of the cervical spine secondary to inhaled nitrous-oxide-induced cobalamin deficiency. *Cureus.* 2022;14:e21214.
24. Shah K, Murphy C. Nitrous oxide toxicity: case files of the carolinas medical center medical toxicology fellowship. *J Med Toxicol.* 2019;15:299-303.
25. Winstock AR, Ferris JA. Nitrous oxide causes peripheral neuropathy in a dose dependent manner among recreational users. *J Psychopharmacol.* 2020;34:229-236.
26. Thompson AG, Leite MI, Lunn MP, Bennett DL. Whippits, nitrous oxide and the dangers of legal highs. *Pract Neurol.* 2015;15:207-209.
27. Edirisinghe SP. Homocysteine-induced thrombosis. *Br J Biomed Sci.* 2004;61:40-47.
28. Cheng HM, Park JH, Hernstadt D. Subacute combined degeneration of the spinal cord following recreational nitrous oxide use. *BMJ Case Rep.* 2013;2013.
29. Norris F, Mallia P. Lesson of the month 2: A case of nitrous oxide-induced pancytopenia. *Clin Med (Lond).* 2019;19:129-130.
30. Einsiedler M, Voulleminot P, Demuth S, et al. A rise in cases of nitrous oxide abuse: neurological complications and biological findings. *J Neurol.* 2022;269:577-582.
31. Jiang J, Shang X. Clinical-radiological dissociation in a patient with nitrous oxide-induced subacute combined degeneration: a case report. *BMC Neurol*. 2020;20:99.
32. Xiao CP, Ren CP, Cheng JL, et al. Conventional MRI for diagnosis of subacute combined degeneration (SCD) of the spinal cord due to vitamin B-12 deficiency. *Asia Pac J Clin Nutr.* 2016;25:34-38.
33. Lim CC. Neuroimaging in postinfectious demyelination and nutritional disorders of the central nervous system. *Neuroimaging Clin N Am.* 2011;21:843-858, viii.
34. Lan SY, Kuo CY, Chou CC, et al. Recreational nitrous oxide abuse related subacute combined degeneration of the spinal cord in adolescents - A case series and literature review. *Brain Dev*. 2019;41:428-435.

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Additional Table 1 Clinical characteristics, treatment and prognosis of 16 patients with nitrous oxide (N_2O) abuse

Patient No.	Age/sex	Date	Symptoms	N_2O abuse	Neurological examination	Vit. B_{12}	HCY	Spinal MRI changes	Treatment	Prognosis
1[1]	27/F	2015	LN+LW	3 years	R (+)	N	N	C3-T12	1000 mg of cyanocobalamin was injected daily for 5 days, and then 1000 mg per week for 4 weeks	Stable discharge after 3 days
2[2]	20/F	2017	LN+LW+H	1 year	R (+); BA (+); MMSE (23)	N	N	C1-T2	Vitamin B12 injections (1 mg per day) and the cessation of N_2O exposure	After 3 months, the sensory and gait symptoms were significantly relieved and the cognitive function was completely recovered
3[3]	20/M	2018	LN+H	DU	Patellar emission (+); Tendon hyperreflexia	↓	DU	C2-C4	Active vitamin B12 supplementation (dose unknown)	The symptoms were relieved after 4 months
4[4]	29/F	2018	LW	DU	BA (+);	↓	DU	T4	Stop using N_2O and vitamin B12 replacement therapy	Unknown
5[5]	24/M	2018	LN+LW	5 months	R (+); Deep hypoesthesia of foot	N	↑	C1-C7	Vitamin B12 1mg was injected every day for more than 1 week	Paresthesia improved and ataxia partially improved
6[5]	22/F	2018	LN+LW	3 months	Inferior muscle strength level II; R(DU);	↓	↑	C1-C7	1 mg of vitamin B12 was injected intramuscularly for 2 weeks and at the outpatient clinic every 2 weeks for 4 weeks	Three months later, the proprioception of the upper limbs was normal and the proprioception of the lower limbs was improved. Sensory abnormalities of lower limbs were further improved, and dysuria was also partially improved
7[6]	19/F	2019	LW	A few weeks	Left upper extremity hyperreflexia	↓	DU	C3-C6	Intramuscular injection of vitamin B12	Transferred to a hospital in China for continuous rehabilitation
8[6]	19/F	2019	DU	6 months	Gait and trunk ataxia	↓	DU	C1-C7	Vitamin B12 replacement therapy	Transferred to a spinal rehabilitation institution in China
9[6]	18/F	2019	LN+LW	9 months	Hyperthyroidism of upper and lower limbs	↓	DU	C1-T12	Vitamin B12 replacement therapy	Unknown
10[7]	22/F	2019	LN+LW	6 months	R (+);	↓	DU	C1-C7	They were treated with daily intramuscular cobalamin (1 mg) for 2 weeks, followed by oral medication	Neurological function improved gradually
11[7]	33/M	2019	LW+LW	4 months	R (+)	↓	DU	C1-C7	They were treated with daily intramuscular cobalamin (1 mg) for 2 weeks, followed by oral medication	Neurological function improved gradually
12[8]	22/M	2019	LW	4 months	hypertonicity; hyperreflexia	↓	DU	C1-C6	Take high-dose vitamin B12 and inject 1 mg intramuscularly every day	After the 7th day, the nervous system improved
13[9]	24/M	2020	LW+LW	3 months	R (+); BA (+);	↓	↑	C2-C6	High dose supplementation with intramuscular vitamin B12 (1.5 mg per day),	After 1 month, the symptoms of weakness and paresthesia were

									oral folic acid (15 mg per day), and N_2O withdrawal	relieved
14[10]	18/F	2020	LW+LW	1 month	Limb muscle strength (↓)	N	↑	T3-T6	Stop inhaling N_2O and smoking, supplement Mecobalamin capsule (500 μ g three times a day), compound vitamin B (one tablet three times a day), and rehabilitation treatment for 1 mon	The symptoms completely disappeared 9 mon after discharge
15[10]	21/M	2020	LN+LW	1 month	R (+); BA (+);	↑	↑	C2-C6	Stop inhaling N2O, Mecobalamin capsule (500 mg three times a day), vitamin B complex (one tablet three times a day), and rehabilitation treatment, and start the treatment for 23 d	Numbness symptoms have improved, but the patient still difficult to walk independently. He can walk independently 4 mon after discharge
16[11]	22/M	2020	LN+LW	3 years	HO (+); R (+);	N	N	C1-C7	Intramuscular injection of hydroxocobalamin lasted for 3 mon	After 5 d, the divine function was partially restored

Note: "↑": Up; "↓": down; BA: Babinski sign; BR: Brudzinski sign; DU: details unknown; F: female; H: hypoesthesia; HCY: homocysteine; HO: Hoffmann sign; LN: limb numbness; LW: limb weakness; M: male; MMSE: Mini-Mental State Examination; MRI: magnetic resonance imaging; N: normal; R: Romberg sign; Vit.: vitamin.

References

1. Pugliese RS, Slagle EJ, Oettinger GR, Neuburger KJ, Ambrose TM. Subacute combined degeneration of the spinal cord in a patient abusing nitrous oxide and self-medicating with cyanocobalamin. *Am J Health Syst Pharm.* 2015;72:952-957.
2. Yuan JL, Wang SK, Jiang T, Hu WL. Nitrous oxide induced subacute combined degeneration with longitudinally extensive myelopathy with inverted V-sign on spinal MRI: a case report and literature review. *BMC Neurol.* 2017;17:222.
3. Antonucci MU. Subacute combined degeneration from recreational nitrous oxide inhalation. *J Emerg Med.* 2018;54:e105-e107.
4. Anderson D, Beecher G, van Dijk R, Hussain M, Siddiqi Z, Ba F. Subacute combined degeneration from nitrous oxide abuse in a patient with pernicious anemia. *Can J Neurol Sci.* 2018;45:334-335.
5. Choi C, Kim T, Park KD, Lim OK, Lee JK. Subacute combined degeneration caused by nitrous oxide intoxication: a report of two cases. *Ann Rehabil Med.* 2019;43:530-534.
6. McArdle DJT, Gaillard F. Pernicious azotaemia? A case series of subacute combined degeneration of the cord secondary to nitrous oxide abuse. *J Clin Neurosci.* 2020;72:277-280.
7. Kwon YJ, Rho JH, Hwang J, Baek SH. Unhappy end of 'happy balloons': subacute combined degeneration caused by nitrous oxide gas. *J Clin Neurol.* 2019;15:118-119.
8. Chaplin K, Bower P, Man MS, et al. Understanding usual care for patients with multimorbidity: baseline data from a cluster-randomised trial of the 3D intervention in primary care. *BMJ Open.* 2018;8:e019845.
9. Jiang J, Shang X. Clinical-radiological dissociation in a patient with nitrous oxide-induced subacute combined degeneration: a case report. *BMC Neurol.* 2020;20:99.
10. Zhao B, Zhao L, Li Z, Zhao R. Subacute combined degeneration induced by nitrous oxide inhalation: Two case reports. *Medicine (Baltimore).* 2020;99:e19926.
11. Seed A, Jogia M. Lessons of the month: Nitrous oxide-induced functional vitamin B(12) deficiency causing subacute combined degeneration of the spinal cord. *Clin Med (Lond).* 2020;20:e7-e9.