Pergamon

Pharmacology Biochemistry and Behavior, Vol. 51, Nos. 2/3, pp. 323-329, 1995

0091-3057/95 $9.50 + .00

0091-3057(94)00388-2

Effects of Subanesthetic Concentrations of Nitrous Oxide on Cold-Pressor Pain in Humans

VESNA PIREC,* TODD H. PATTERSON,* PANKAJ THAPAR,*
JEFFREY L. APFELBAUM* AND JAMES P. ZACNY†[1]

*Departments of *Anesthesia and Critical Care and †Psychiatry, The Pritzker School of Medicine, The University of Chicago, Chicago, IL*

Received 16 June 1994

PIREC, V., T. H. PATTERSON, P. THAPAR, J. L. APFELBAUM AND J. P. ZACNY. *Effects of subanesthetic concentrations of nitrous oxide on cold-pressor pain in humans.* PHARMACOL BIOCHEM BEHAV 51(2/3) 323-329, 1995.—Nitrous oxide (N_2O) has analgesic properties as determined in both animal and human research. In the present study, we sought to determine whether N_2O given in subanesthetic concentrations would reduce cold pressor (CP) -induced pain. A crossover, double-blind study was conducted in 10 healthy volunteers. Each subject participated in four separate sessions, and in each session the effects of one of four concentrations of N_2O in oxygen (0, 20, 30, and 40%) were assessed. The duration of inhalation was 40 min, and within each session, subjects immersed their nondominant arm in water (2-3°C) twice for 3 min (at 10 and 30 min intrainhalation). Pain intensity, the degree to which the pain was bothersome (measured on a verbal scale of 0-10, 0 = "not at all" and 10 = "extremely" painful/bothersome), and pain quality [measured by the short-form McGill Pain Questionnaire (SF-MPQ)] were assessed during the forearm immersion. Mood effects were measured with the use of visual analogue scales (VAS) in the presence and absence of pain. Self-reported pain intensity and bothersomeness, SF-MPQ ratings of "sharp pain" and "throbbing pain," and VAS rating of "unpleasant bodily sensations" were significantly reduced by N_2O ($p < 0.05$) in a concentration-dependent manner. Nitrous oxide had a number of effects on mood (e.g., increased VAS ratings of "stimulated," "high," "coasting," "carefree," and "having pleasant bodily sensations"). The cold-water immersion also influenced mood, but had little impact on modulating N_2O effects. Results from our study indicate that the CP test is a sensitive assay to measure the analgesic properties of subanesthetic concentrations of N_2O in humans.

Nitrous oxide Analgesia Pain Subjective Human Volunteer

NITROUS oxide (N_2O) at subanesthetic concentrations is used primarily for its sedative and anxiolytic effects in medical and dental practice. N_2O is also a known analgesic, which has been demonstrated in both animal and human research. Studies in animals have included various assays such as mouse writhing models (4,5,24,31), hot plate (31), tail-flick (4), hot water (21), and formalin tests (16). In most of these studies the effective analgesic dose of N_2O ranged from 55-80%. These concentrations are relatively high when compared to those used in humans to produce antinociception. In humans such pain assays as painful ischemia (8,40), von Frey hair technique (30), electrical pulp stimulation (2,7,32), tibial pressure pain, and thermal pain (28) have been used to demonstrate the analgesic efficacy of N_2O. Analgesic properties of N_2O have been found with concentrations as low as 20% (8). In a number of these studies, dose dependency for N_2O analgesia was found (2, 12,25,30).

In the present study we sought to determine whether N_2O would reduce cold-pressor pain (CP) in humans. The CP test, first used by Hines (18), has been used experimentally to induce pain, and it has proven to be a reliable method that is sensitive to pharmacologic and physiologic manipulations. It is a well-documented human tonic pain model (9) in which the immersion of the forearm in circulating ice water maintained at about 2°C produces a persistent aching or crushing pain. Because it induces tonic as opposed to phasic pain, the CP test produces sensations similar to clinical pain (e.g., postoperative pain, some forms of chronic pain) (36). This test has been used as a screening method to determine the analgesic efficacy of different drugs including opiates (26,39) and nonsteroidal anti-inflammatory agents (20,35). To our knowledge the CP test has not been used to assess the analgesic effects of N_2O at concentrations that are subanesthetic in nature. It has been used in a study involving anesthetic concentrations of N_2O

[1] Requests for reprints should be addressed to James P. Zacny, Department of Anesthesia and Critical Care, MC 4028, University of Chicago, 5841 S. Maryland Avenue, Chicago, IL 60637.

(29). Medical students inhaled concentrations that induced unconsciousness (60–80%), while at periodic intervals during the 3-h inhalation period their forearms were immersed in an ice bath. The response to the cold-induced pain was measured as a reflexive withdrawal of the immersed arm from the ice-cold water, in which a 3-min interval was taken as a cutoff time. Longer latencies to withdrawal were obtained during N_2O inhalation, relative to a no-inhalation time point. In the present study, we sought to determine whether concentrations lower than anesthetic ones would induce analgesia and, if so, whether the analgesia was concentration related. Such a study would be useful because results from this study could be compared with, and perhaps assimilated with, other studies that have tested N_2O's analgesic effects on other types of pain (e.g., thermal, mechanical, ischemic) in humans.

MATERIALS AND METHODS

Subjects

Ten healthy volunteers (eight men and two nonpregnant women), aged 21–35, participated in the study. Candidates who consumed at least one drink/week were scheduled for a screening interview with one of the research personnel. At the interview, prospective subjects completed the SCL-90 (a questionnaire designed to assess psychiatric symptomatology) (10) and a health questionnaire (designated to determine their psychiatric and medical status). Candidates with any significant psychiatric problems (such as any history of drug- or alcohol-related problems or Axis I psychiatric disorders) (1) were excluded. In addition, a physical examination and resting electrocardiogram were performed on prospective subjects. Those with significant medical problems (including adverse reaction to general anesthetics in the past) were excluded from the study. Female participants were screened for pregnancy once a week.

Before participating, each volunteer signed a written consent form that described the details in the study; in the consent form, subjects were told that the inhalation drugs to be used in the study were drugs commonly used in medical settings and that might come from one of six classes—sedative, stimulant, opiate, general anesthetic (at subanesthetic concentrations), alcohol, or placebo. Payment for the study was made during a debriefing session held after the study. The study was approved by the local institutional review board.

Experimental Design

Subjects participated in a randomized, placebo-controlled, crossover trial of four morning experimental sessions separated by at least 48 h (2 days), each lasting 2–2.5 h. During each session, subjects inhaled "placebo" (100% oxygen) or one of three different concentrations of N_2O in oxygen (20, 30, or 40%) for 40 min. The 40% N_2O concentration was the highest tested in our study, because at higher concentrations it has been documented that some people become uncooperative (6), and there is a risk of entering into the second stage of anesthesia (33). We chose 20% N_2O as the lowest concentration because a previous study done in our laboratory demonstrated that no psychoactive effects of the drug were observed with concentrations $< 20\%$ (11).

Experimental sessions took place in a laboratory located in the Department of Anesthesia and Critical Care. Drugs (both N_2O and oxygen) were administered by an anesthesiologist who was not blinded to the drug given but kept verbal communication with the subject to a minimum. The subject and the research technician administering tests were blinded to the agent being administered. The agents were administered through a clear, disposable, tight-fitting anesthesia mask (Vital Signs, Inc., Totowa, NJ) via a semiclosed circuit using an anesthesia machine (Narkomed; North American Drager, Telford, PA). Peppermint oil was added to the circuit in each session to mask the odor of N_2O. During the sessions subjects were comfortably seated, and between testing times they were allowed to engage in recreational activities that did not require much movement (e.g., reading, listening to the radio, watching television).

Cold-Pressor Test

The CP apparatus (36) consisted of a standard ice chest divided into two compartments by a wire screen. The tank was filled with water, and ice was added to one side of the screen. A cradle for the subject's forearm was positioned in the side of the chest with no ice and allowed the subject to rest the forearm while immersing it into the cold water. The water in the ice chest was constantly circulated by an aquarium pump. Each immersion of the nondominant arm lasted for 180 s. When the subject removed his or her forearm from the ice-cold water, he or she would immerse the arm in a container filled with lukewarm water for at least 5 min. Then, the experimenter would dry the arm with the towel and cover it with a blanket until the next immersion. Surface skin temperature was checked with a electronic digital thermometer (Fisher Scientific, Pittsburgh, PA) attached to a reusable temperature probe (YSI, Yellow Springs, OH) before each immersion, to avoid the effect of superficial cooling of the skin.

In the consent form, subjects were instructed that the ice-cold water would not result in any damage, but could induce extreme pain and some redness and numb feeling in the forearm. They were also told that those unpleasant effects would disappear soon after the withdrawal of the arm from the cold water.

Experimental Sessions

Each session consisted of three periods: baseline (BL), inhalation (INH), and recovery (REC). After coming into the laboratory, the subject was seated, the anesthesiologist placed the anesthesia mask placed over the subject's mouth and nose, and the BL period began. The BL period was 10 min, and the subject was informed that the air he or she was inhaling was drug free. The INH period lasted 40 min, during which time the subject inhaled either 100% oxygen or N_2O (20, 30, or 40%) in oxygen. At the beginning of the INH period subjects were told that the air they were going to inhale for the following 40 min might or might not contain a drug. Equilibration of the N_2O concentration (equalized N_2O concentration in the inhaled and exhaled air) takes about 10 min (33). The CP test was conducted 10 and 30 min into the INH period. During each CP trial, pain intensity, the extent to which it was bothersome, and mood were rated. Mood and psychomotor performance were also assessed at certain other time points. When the inhalation period was over, the anesthesiologist took off the mask and a REC period of 70 min commenced. During this time, the subject remained seated.

Dependent Measures

Table 1 shows the time line of events and when the various dependent measures (see subsequent description) were assessed.

TABLE 1
TIME LINE OF EVENTS AND WHEN MEASURES WERE COLLECTED

Min	−10:00*	0†	10:00‡	10:30‡	11:10‡	11:50‡	12:50‡	20:00	30:00‡	30:30‡	31:10‡	31:50‡	32:50‡	35:00	40:00§	100#
Pain measures																
Pain intensity				X	X	X	X			X	X	X	X			
Pain bothersome				X	X	X	X			X	X	X	X			
SF-MPQ					X						X					
Mood measures																
VAS						X		X				X		X		
ARCI	X							X						X		
Psychomotor measure																
DSST	X							X						X		X
Physiological measures																
SBP					X			X			X			X		
DBP					X			X			X			X		
HR					X			X			X			X		

*Baseline.
†Onset of inhalation of 0, 20, 30 or 40% N_2O.
‡Immersion of forearm in cold water.
§Offset of inhalation.
#Sixty minutes after offset of inhalation, i.e., recovery.

Pain assessments. Subjects were instructed to verbally rate the pain and its bothersomeness on a scale of 0–10 during the immersion of the arm in the ice-cold water (0 = not painful/bothersome at all and 10 = extreme pain/bothersomeness). The questions, "How painful is it?" and "How much does it bother you?" were asked at 30, 70, 110, and 170 s into the immersion. These assessments were done during both CP trials.

The short form of the McGill Pain Questionnaire (SF-MPQ) provides information on the sensory, affective, and evaluative dimensions of the pain experience (23). Fifteen descriptors (throbbing, shooting, stabbing, sharp, cramping, gnawing, hot-burning, aching, heavy, tender, splitting, tiring-exhausting, sickening, fearful, punishing-cruel) are listed to represent the sensory, affective, and evaluative dimensions of the pain experience. Each descriptor is ranked on an intensity scale from 0–3 (0 = none, 1 = mild, 2 = moderate, 3 = severe). Subjects were instructed to rate the pain descriptions during the cold-water immersion. The SF-MPQ was given to subjects during each of the two CP trials.

Subjective effects. The Addiction Research Center Inventory (ARCI) is a standardized true–false questionnaire designated to differentiate among classes of psychoactive drugs (17). Scales are derived from the questions corresponding to different drug effects. The 29 questions used in this study yielded scores for two subscales of the ARCI, the Morphine-Benzedrine Group Scale (designed to measure euphoria) and the Lysergic Acid Diethylamide Scale (LSD) (designed to measure dysphoria and somatic symptoms) (22).

The Visual Analogue Scale (VAS) measures mood states on a form consisting of 100-mm lines, each labeled with an adjective (i.e., stimulated, high, dizzy, nauseous, tingling, anxious, happy, sedated, down, confused, drunk, elated, coasting, carefree, having pleasant thoughts, having unpleasant thoughts, having pleasant bodily sensations, having unpleasant bodily sensations, in control of body, in control of thoughts, and hungry). Subjects were instructed to place a mark on each line indicating how they felt at the moment, ranging from "not at all" to "extremely."

Psychomotor performance. The Digit Symbol Substitution Test (DSST) is a simple paper-and-pencil test that provides a general measure of psychomotor performance (37). The test lasted 1 min, and subjects had to replace digits with an appropriate symbol. The score was the correct number of symbols drawn by the subject.

Physiologic measures. Pulse, systolic, and diastolic blood pressure were measured noninvasively (Hewlett Packard Model 54; Waltham, MA). The measures were assessed before the onset of inhalation and during the inhalation period, including when the forearm was immersed in the cold water.

Debriefing. After the last session, each subject was asked during a debriefing session several questions about the pain experienced during the forearm immersions in each session. These questions included describing how the pain felt in terms of qualitative terms, whether it decreased in magnitude in any of the sessions, and whether the drug effect decreased the pain sensation or merely distracted the subject from the pain.

Data Analysis

Repeated-measures analysis of variance (ANOVA) was used for statistical treatment of data. On "pain" and "bothersome" scores, ANOVAs were done on all 10 subjects with Concentration (four levels), Trial (two levels) and Time (four levels) as factors. Factors in the VAS, SF-MPQ ratings, and physiologic measures analyses were Concentration (four levels), Immersion (two levels), and Time (two levels). On both ARCI and DSST scores, ANOVAs were done on all 10 subjects with Concentration (four levels) and Time (three to four levels). F values were considered significant for $p < 0.05$ with adjustments of within-factor degrees of freedom (Huynh-Feldt) to protect against violation of symmetry assumptions. When appropriate, Tukey post-hoc tests were done, comparing oxygen responses vs. different drug concentrations at corresponding time points in the session.

RESULTS

Eight men and two women (age range 21–35, mean age 24) completed the study. One other volunteer dropped out after

the first session because of a self-reported panic reaction from the CP immersion. The mean number of alcoholic drinks consumed per week was four (mean 4; range 1-6 drinks/week). Five of our subjects smoked tobacco cigarettes occasionally (mean 3.6 cigarettes/day; range 1-6 cigarettes/day). Two subjects had previously received N_2O in a dental setting, but none of the subjects inhaled N_2O for recreational purposes in the past.

Pain Ratings

A significant Concentration × Trial interaction was obtained on pain intensity ratings ($p < 0.05$). Post-hoc testing revealed that during the first immersion trial, only 30 and 40% N_2O reduced pain intensity ratings (averaged across the four time points within the trial), but during the second immersion trial (averaged across the four time points within the trial), all three concentrations of N_2O significantly reduced these ratings (Fig. 1). Post-hoc testing also revealed that during the second immersion trial, the pain intensity ratings decreased in a concentration-dependent fashion, in that the ratings were significantly lower in the 40% condition than in the 20 and 30% N_2O conditions. A Concentration × Time interaction approached significance ($p = 0.08$). Pain intensity ratings tended to increase more across the four time points within the immersion period when placebo-oxygen was inhaled than when N_2O was inhaled. Percent reduction in pain intensity ratings, relative to oxygen-placebo ratings, in the 20, 30, and 40% N_2O conditions, collapsed across trials and time points within a trial, were 12, 24, and 36%, respectively.

A significant Concentration × Time interaction was obtained on pain bothersome ratings ($p < 0.05$). Post-hoc testing revealed that 30 and 40% N_2O significantly decreased bothersome ratings at all four time points within the immersion, as compared to oxygen (Fig. 2). Twenty percent N_2O had a significant attenuating effect on pain bothersomeness at the third and fourth time points during the immersion as compared to oxygen. As can be seen in the Fig. 2, pain bothersomeness ratings tended to increase across the four time points more in the placebo-oxygen condition than when N_2O was inhaled. Percent reduction in pain bothersomeness ratings, relative to oxygen-placebo ratings, in the 20, 30, and 40% N_2O conditions, collapsed across trials and time points within a trial, were 17, 34, and 38%, respectively.

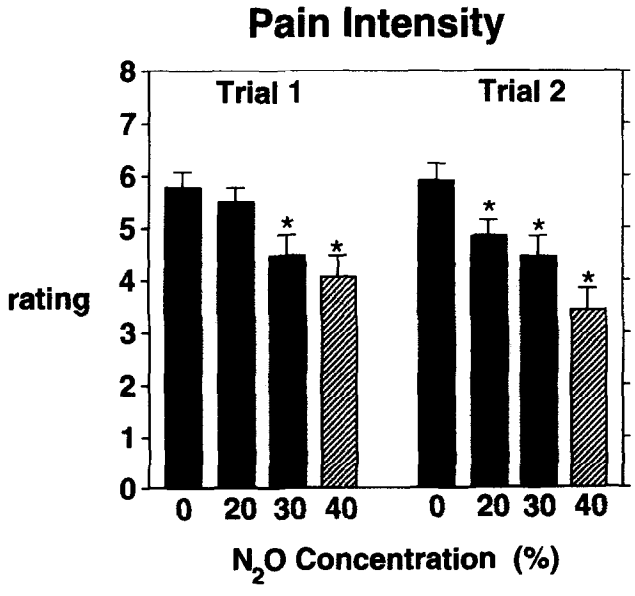

FIG. 1. Effects of 0, 20, 30, and 40% N_2O on pain-intensity ratings as a function of Trial. Each bar represents the average response of 10 subjects; brackets indicate SEMs. Asterisks above a bar indicate that the rating was significantly less in that condition than in the 0% N_2O condition.

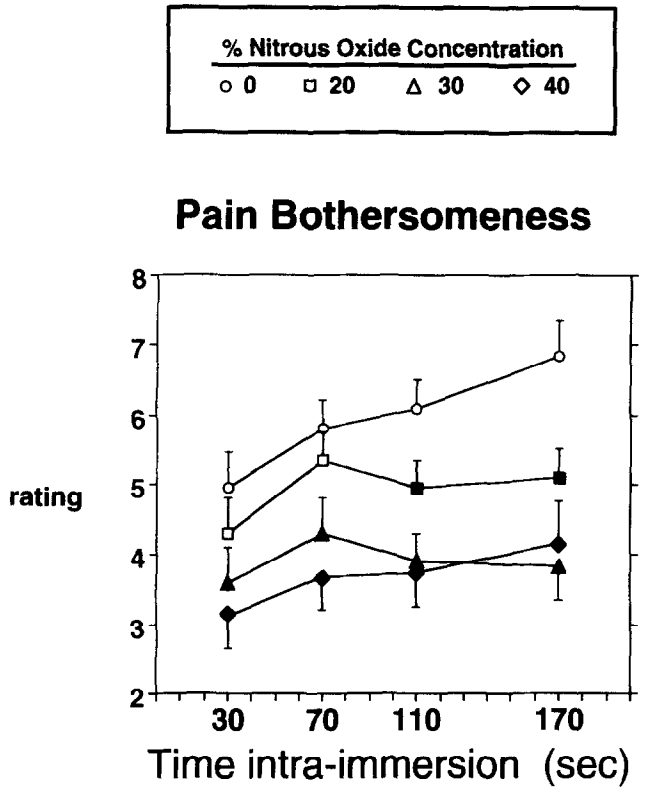

FIG. 2. Effects of 0, 20, 30, and 40% N_2O on pain bothersome ratings as a function of intra-immersion time. Each symbol represents the average response of 10 subjects; brackets indicate SEMs. Solid symbols indicate that the rating was significantly less in that condition than in the 0% N_2O condition.

SF-MPQ Ratings

Ratings > 1 were obtained on the following pain descriptors: sharp, throbbing, shooting, gnawing, and hot burning pain. However, a significant Concentration effect of N_2O was obtained only on throbbing pain ($p < 0.05$) and on sharp pain ($p < 0.01$) (Fig. 3). Both sharp pain and throbbing pain were significantly attenuated by 30 and 40% N_2O. Degree of attenuation was no greater in the 40% N_2O condition than in the 30% N_2O condition, indicative of a ceiling effect.

Subjective Effects

ARCI Nitrous oxide increased LSD scores in a concentration-dependent manner ($p < 0.01$). LSD scores were significantly higher in the 30 and 40% N_2O concentration conditions than in the oxygen-placebo condition. MBG scores were not significantly changed by N_2O inhalation.

VAS Nitrous oxide increased the ratings of stimulated, high, coasting, carefree, and having pleasant bodily sensations, in both the presence and absence of the immersion (all

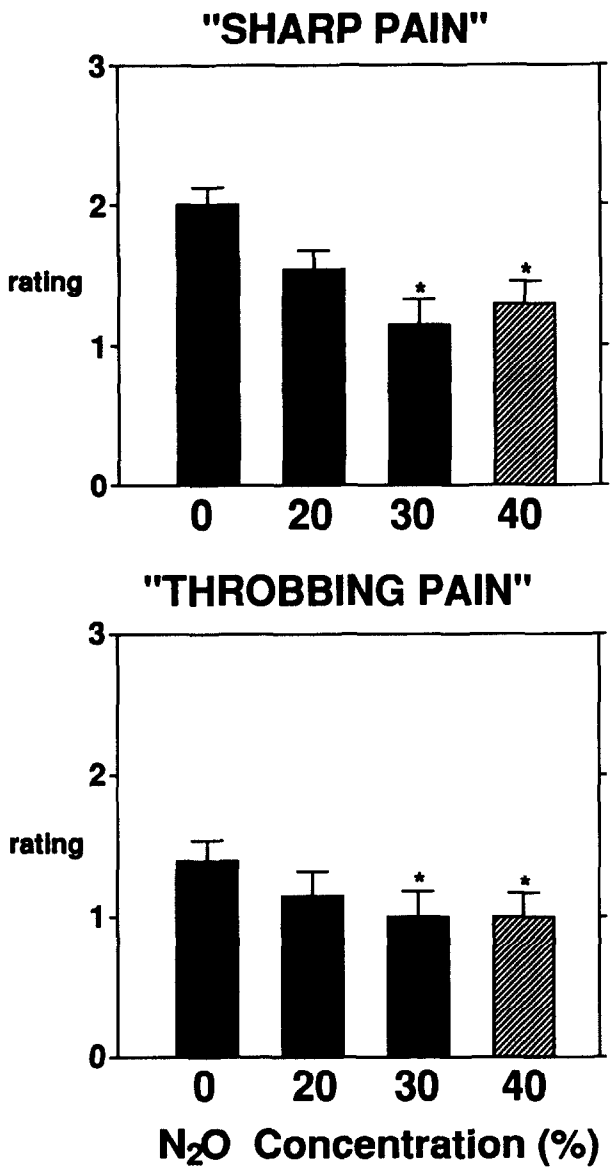

FIG. 3. Effects of 0, 20, 30, and 40% N_2O on sharp pain and throbbing pain ratings from the SF-MPQ. Each bar represents the average response of 10 subjects; brackets indicate SEMs. Asterisks above a bar indicate that the rating was significantly less in that condition than in the 0% N_2O condition.

$p < 0.05$). Marginally significant Concentration effects were obtained on elated ($p = 0.06$) ratings, which increased during N_2O inhalation, and in-control-of-body ($p = 0.06$) ratings, which decreased during N_2O inhalation. Significant Immersion effects were observed on the ratings of anxious, carefree, having pleasant thoughts, having pleasant bodily sensations, having unpleasant bodily sensations, and in control of thoughts. The cold-water immersion increased ratings of anxious and having unpleasant bodily sensation, whereas it decreased ratings of carefree, having pleasant thoughts, having pleasant bodily sensation, and in control of thoughts. Significant Concentration × Immersion interactions were obtained on having unpleasant bodily sensations and tingling. In the presence of pain, N_2O (30 and 40%) decreased ratings of unpleasant bodily sensations, but in the absence of pain, these ratings were low and the drug had no effect (Fig. 4). Tingling ratings in the oxygen-placebo condition were significantly higher during immersion (mean ± SEM: 31.3 ± 6.5) than in the absence of immersion (mean rating 7.8 ± 2.1), which accounted for the Concentration × Immersion interaction. Tingling ratings increased during N_2O inhalation in the absence of immersion, but the increase was not significant.

Psychomotor Effects

Nitrous oxide significantly decreased performance on the DSST ($p < 0.001$). All three N_2O concentrations produced significant impairment in DSST scores as compared to the inhalation of 100% oxygen. Decreases in number of symbols correctly drawn in the 0, 20, 30, and 40% N_2O conditions (20-min intra-inhalation minus baseline) were 0, 5.2, 7.7, and 9.2, respectively.

Physiologic Parameters

A significant Concentration effect was obtained with systolic blood pressure ($p < 0.05$). Post-hoc testing revealed that systolic blood pressure was significantly lower in the 20% N_2O condition than in the oxygen-placebo condition, but the absolute difference between conditions, 7 mm Hg, was relatively small and of little clinical significance. Nitrous oxide had no effect on diastolic blood pressure or heart rate. The cold-water immersion produced a significant increase in systolic ($p < 0.01$) and diastolic ($p < 0.01$) blood pressure and heart rate ($p < 0.05$). Average systolic blood pressure was 131 ± 1.6 mm Hg in the immersion condition, and 118 ± 1.5 mm Hg when the arm was not immersed. Average diastolic blood pressure was 86 ± 1.0 mm Hg in the immersion condition, and 71 ± 0.9 mm Hg when the arm was not immersed. Finally, average heart rate was 65 ± 1.3 beats per minute (bpm) in the immersion condition, and 62 ± 1.2 bpm when the arm was not immersed.

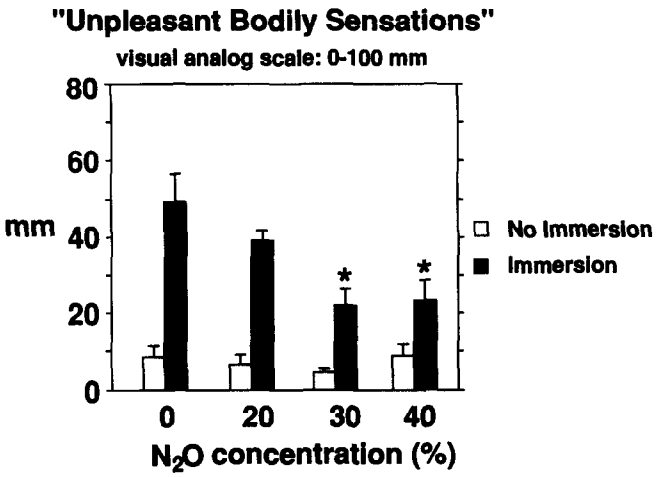

FIG. 4. Effects of 0, 20, 30, and 40% N_2O on the VAS rating of unpleasant bodily sensations as a function of whether the forearm was immersed in icy cold water. Each bar represents the average response of 10 subjects; brackets indicate SEMs. Asterisks above a solid bar (immersion) indicate that the rating was significantly less in that condition than in the 0% N_2O condition when the arm was immersed.

Debriefing

All 10 subjects reported decreased pain sensation during N_2O inhalation. Five of 10 subjects were able to rate the degree of pain sensation from session to session, in accordance with the different drug concentration. In other words, they reported the least pain during the inhalation of 40% N_2O, whereas the pain increased with decreasing concentrations of drug inhaled. Three of 10 subjects reported feeling the same pain intensity when they were inhaling 30 and 40% N_2O. There was considerable intersubject variability in terms of rating the qualitative nature of the pain. Seven subjects reported that the pain was actually reduced by N_2O, as opposed to the drug merely producing a distraction from the painful stimulus.

DISCUSSION

Several measures collected in this study demonstrated that N_2O reduced pain induced by cold. First we asked subjects four times during the 180-s cold-water immersion to verbally rate pain intensity and bothersomeness. Nitrous oxide at subanesthetic concentrations reduced in a dose-dependent fashion ratings of pain intensity and bothersomeness (i.e., aversiveness) (Figs. 1 and 2). An interesting finding emerged in that bothersome ratings tended to increase across the 3-min time period more so when oxygen-placebo was inhaled, as opposed to N_2O (Fig. 2). Second, two ratings on the SF-MPQ were reduced by N_2O. Third, VAS ratings of having unpleasant bodily sensations were reduced during the immersion in a dose-dependent manner by N_2O. Finally, we asked subjects during the debriefing, before they were informed about the drug that they inhaled, about their recollections regarding the cold-water immersion across the four sessions. The majority of subjects were able to remember that the immersion generated less pain during those sessions in which they inhaled N_2O. Taken as a whole, these results provide convincing evidence that the type of pain engendered by cold is reduced by subanesthetic concentrations of N_2O in humans. Thus, at both subanesthetic and anesthetic [see (29)] concentrations, the CP test is a sensitive assay for detecting the pain-reducing qualities of N_2O.

For the most part our findings are consistent with other studies examining the effects of N_2O on different types of pain in humans. In the present study, concentrations as low as 20% reduced self-reported intensity and aversiveness of cold-induced pain. We established that the pain reduction was concentration related in that 40% N_2O produced a greater reduction in intensity and bothersome ratings than did 20% N_2O. Other studies used fairly low concentrations of N_2O (e.g., 20–30%) in demonstrating its analgesic properties on qualitatively different nociceptive stimuli, including heat (28), electrical stimulation of tooth pulp (2,7,32), ischemia (8,40), and tibial pressure (28,38). In those studies that examined a range of concentrations of N_2O, analgesia was related to concentration in a fairly orderly fashion (2,38).

There was no evidence of acute tolerance occurring to the analgesic effects of N_2O in this study. On the contrary, 20% N_2O, which was not analgesic in the first trial (10 min intra-inhalation), was analgesic in the second trial (30 min intra-inhalation) (Fig. 1). Acute tolerance of the analgesic effects of N_2O was obtained in both animals (3) and humans (29,38), but the inhalation interval studied tended to be longer than the 40-min trial used in the present study.

How might N_2O be exerting its analgesic effects? There is a large body of research evidence involving both infrahumans and humans to suggest that the endogenous opiate system (EOS) is involved in mediating the analgesic effects of N_2O [cf. (15)]. Most of the studies involved challenge with an opiate antagonist before or during N_2O inhalation, during which a nociceptive stimulus was applied. In the majority of these studies, an opiate antagonist challenge reversed, in part or totally, the analgesia that was produced by N_2O [e.g., (27)]. Naloxone also antagonizes N_2O-induced locomotor activity in mice in the same way that this antagonist blocks morphine-induced locomotor activity (19). Further, cross-tolerance exists between morphine, a classic μ-opiate agonist, and N_2O, suggesting shared mechanisms of action (5).

One caveat of the present study is that a negative control drug was not employed (i.e., a psychoactive drug that has been shown previously to have no effect on pain responses). One could argue that demand characteristics of the experiment would bias subjects toward reporting a diminution in pain with any drug that produced psychoactive drug effects. In fact, in an interesting series of studies, Dworkin et al. (13,14) demonstrated that the N_2O could become either more analgesic or actually algesic, depending on the specific set of instructions given to subjects at the beginning of the study regarding N_2O's effects. We would argue against this interpretation for our study, though, for two reasons. First, the studies of Dworkin et al. manipulated instructions, and under their control conditions in which subjects were given minimal instructions (as our subjects received), they found a normal analgesic response. Only under the special instructions conditions did Dworkin et al. find either potentiated analgesia or algesia. Second, studies have shown that drugs with mood-altering effects do not necessarily produce reductions in pain reports when a painful stimulus is applied. A study conducted several years ago (34) and one recently completed in our laboratory (41) used both an opiate (analgesic) and a benzodiazepine, both of which have mood-altering effects, and only the opiate reduced CP-induced pain, whereas the benzodiazepine had no effect. Thus, although it is possible that demand characteristics may have been operating in the present study, we believe it to be unlikely, based on the responses by subjects to the different measures used in this study to assay pain.

An interesting component of the present study is that we asked subjects to rate the subjective effects of N_2O while they had their arm immersed in the cold water. There have been numerous studies examining the mood-altering effects of N_2O, but all of these studies were done while subjects were in a pain-free state. We found little difference in subjective effects of N_2O between presence vs. absence of cold-water immersion. Subjects reported increased ratings of stimulated, high, coasting, and carefree, both in the presence and absence of pain. Whether mood-altering effects of other psychoactive drugs without analgesic properties would be unaltered by pain is still open to question, and remains another interesting research avenue to explore.

In conclusion, in terms of its intensity and bothersomeness (aversiveness), cold-induced pain is reduced by subanesthetic concentrations of N_2O. Taking this study into account with a number of other studies, it is clear that N_2O reduces a number of different types of pain. The neurochemical mechanisms mediating the analgesic effects of N_2O appear to involve the endogenous opiate system, although other mechanisms may be involved. Future studies using this method of inducing a tonic sort of pain while collecting measures of pain in real time might employ drug antagonist challenges (naloxone, flumazenil) to determine to what extent a given receptor is involved in N_2O analgesia.

ACKNOWLEDGEMENTS

This research was supported in part by a grant from the National Institute on Drug Abuse (DA08391). The authors express their appreciation to Alicia Toledano, Sc.D., for her advice regarding the statistical analysis of the data.

REFERENCES

1. American Psychiatric Association. Diagnostic and statistical manual of mental disorders. 3rd edition, revised. Washington, DC: American Psychiatric Association; 1987.
2. Benedetti, C.; Chapman, C. R.; Colpitts, Y. H.; Chen, A. C. Effect of nitrous oxide concentration on event related potentials during painful tooth stimulation. Anesthesiology 56:360–364; 1982.
3. Berkowitz, B. A.; Finch, A. D.; Hynes, M. D.; Ngai, S. H. Tolerance to nitrous oxide in rats and mice. Anesthesiology 51: 309–312; 1979.
4. Berkowitz, B. A.; Finch, A. D.; Ngai, S. H. Nitrous oxide analgesia: Reversal by naloxone and development of tolerance. J. Pharmacol. Exp. Ther. 203:539–547; 1977.
5. Berkowitz, B. A.; Ngai, S. H.; Finch, A. D. Nitrous oxide "analgesia" resemblance to opiate action. Science 194:967–968; 1976.
6. Block, R. I.; Ghoneim, M. M.; Hinrichs, J. V.; Kumar, V.; Pathak, D. Effects of subanesthetic concentration of nitrous oxide on memory and subjective experience: Influence of assessment procedures and types of stimuli. Hum. Psychopharmacol. 3:257–265; 1988.
7. Chapman, C. R.; Benedetti, C.; Butler, S. H. Nitrous oxide effects on cerebral evoked potential to pain; partial reversal with narcotic antagonist. Anesthesiology 51:135–138; 1979.
8. Chapman, W. P.; Arrowood, J. G.; Beecher, H. K. The analgesic effects of low concentrations of nitrous oxide compared in man with morphine sulfate. J. Clin. Invest. 22:871–875; 1943.
9. Chen, A. C. N.; Dworkin, S. F.; Haug, J. Human pain responsivity in a tonic pain model: Physiological determinants. Pain 37: 143–160; 1989.
10. Derogatis, L. R.; Lipman, R. S.; Cozi, L. HSCL-90: An outpatient psychiatric rating scale: Preliminary report. Psychopharmacol. Bull. 9:13–17; 1973.
11. Dohrn, C.; Lichtor, J. L.; Finn, R. S.; Uitvlugt, A.; Coalson, D. W.; Rupani, G.; de Wit, H.; Zacny, J. P. Subjective and psychomotor effects of nitrous oxide in healthy volunteers. Behav. Pharmacol. 3:19–30; 1992.
12. Dundee, J. W.; Moore, J. Alterations in response to somatic pain associated with anesthesia. IV. The effect of subanesthetic concentrations of inhalation agents. Br. J. Anesth. 32:453–459; 1960.
13. Dworkin, S. F.; Chen, A. C.; LeResche, L.; Clark, D. W. Cognitive reversal of expected nitrous oxide analgesia for acute pain. Anesth. Analg. 62:1073–1077; 1983.
14. Dworkin, S. F.; Chen, A. C.; Schubert, M. M; Clark, D. W. Cognitive modification of pain: Information in combination with nitrous oxide. Pain 19:339–351; 1984.
15. Gillman, M. A. Analgesic nitrous oxide should join the opioid family after almost two centuries. S. Afr. J. Sci. 89:25–27; 1993.
16. Goto, T.; Marota, J. J. A.; Crosby, G. Nitrous oxide induces preemptive analgesia in the rat that is antagonized by halothane. Anesthesiology 80:409–416; 1994.
17. Haertzen, C. A. Development of scales based on patterns of drug effects, using the Addiction Research Center Inventory (ARCI). Psychol. Rep. 18:163–194; 1966.
18. Hines, E. A.; Brow, G. E. A standard stimulus for measuring vasomotor reactions: Its application in the study of hypertension. Staff Mtng. Mayo Clin. 8:332–335; 1932.
19. Hynes, M. D.; Berkowitz, B. A. Nitrous oxide stimulation of locomotor activity: Evidence for an opiate-like behavioral effect. J. Pharmacol. Exp. Ther. 209:304–308; 1979.
20. Jones, S. F.; McQuay, H. J.; Moore, R. A.; Hand, C. W. Morphine and ibuprofen compared using the cold pressor test. Pain 34:117–122; 1988.
21. Lawrence, D.; Livingston, A. Opiate-like analgesics in general anesthetics. Br. J. Pharmacol. 73:435–442; 1981.
22. Martin, W. R.; Sloan, J. W.; Sapira, J. D.; Jasinski, D. R. Psychologic, subjective and behavioral effects of amphetamine, methamphetamine, ephedrine, phenmetrazine and methylphenidate in man. Clin. Pharmacol. Ther. 12:245–258; 1971.
23. Melzack, R. The short-form McGill Pain Questionnaire. Pain 30: 191–197; 1987.
24. Mueller, J. L.; Quock, R. M. Contrasting influences of 5-hydroxytryptamine receptors in nitrous oxide antinociception in mice. Pharmacol. Biochem. Behav. 41:429–432; 1992.
25. Parkhouse, J.; Henrie, J. R.; Dunkan, G. M.; Rome, H. P. Nitrous oxide analgesia in relation to mental performance. Anesthesiology 128:44–54; 1960.
26. Posner, J.; Telekes, A.; Crowley, D.; Philipson, R.; Peck, A. W. Effects of an opiate on cold-induced pain and the CNS in healthy volunteers. Pain 23:73–82; 1985.
27. Quock, R. M.; Curtis, B. A.; Reynolds, B. J.; Mueller, J. L. Dose-dependent antagonism and potentiation of nitrous oxide antinociception by naloxone in mice. J. Pharmacol. Exp. Ther. 267:117–122; 1993.
28. Robson, J. G.; Harold, M. B.; Davenport, M. B.; Sugiyama, R. Differentiation of two types of pain by anesthetics. Anesthesiology 26:31–34; 1965.
29. Rupreht, J.; Dworacek, B.; Bonke, B.; Dzoljic, M. R.; van Eijhdhoven, J. H. M.; de Vlieger, M. Tolerance to nitrous oxide in volunteers. Acta Anesthesiol. Scand. 29:635–638; 1985.
30. Seevers, M. H.; Bennett, J. H.; Pohle, H. W.; Reinardy, E. W. The analgesia produced by nitrous oxide, ethylene and cyclopropane in the normal human subjects. J. Pharmacol. Exp. Ther. 59: 294–300; 1937.
31. Smith, E. H.; Rees, J. M. H. The effects of naloxone on the analgesic activities of general anesthetics. Experientia 37:289–290; 1981.
32. Sonnenschein, R. R.; Jamison, R.; Loveseth, L. J.; Cassels, W. H.; Ivy, A. C. A study of the mechanism of nitrous oxide analgesia. J. Appl. Physiol. 1:254–258; 1948.
33. Stanski, D. R. Monitoring depth of anesthesia. In: Miller, R. D., ed. Anesthesia. 3rd edition. New York: Churchill Livingstone; 1990:1001–1029.
34. Stern, J. A.; Brown, M.; Ulett, G. A.; Sletten, I. A comparison of hypnosis, acupuncture, morphine, valium, aspirin, and placebo in the management of experimentally induced pain. In: Edmonston, W. E., Jr., ed. Conceptual and investigative approaches to hypnosis and hypnotic phenomena. New York: Annuals of the New York Academy of Sciences; 1977:175–193.
35. Telekes, A.; Holland, R. L.; Peck, A. W. Indomethacin: Effects on cold-induced pain and nervous system in healthy volunteers. Pain 30:321–328; 1987.
36. Turk, D. C.; Meichenbaum, D.; Genest, M. Pain and behavioral medicine—A cognitive-behavioral perspective. New York: Guilford Press; 1983:84–89.
37. Wechsler, D. The measurement and appraisal of adult intelligence. Baltimore, MD: Williams and Wilkins; 1958.
38. Whitham, J. G.; Morgan, M.; Hall, G. M.; Petrie, A. Pain during continuous nitrous oxide administration. Br. J. Anaesthesiol. 48:425–429; 1976.
39. Wotherspoon, H. A.; Kenny, G. N. C.; McArdle, C. S. Analgesic efficacy of controlled-release dihydrocodeine. Anaesthesia 46: 915–917; 1991.
40. Yang, J. C.; Crawford, W. C.; Ngai, S. H. Antagonism of nitrous oxide analgesia by naloxone in man. Anesthesiology 52: 414–417; 1980.
41. Zacny, J. P.; Coalson, D.; Young, C.; Klafta, J.; Rupani, G.; Thapar, P.; Choi, M.; Apfelbaum, J. L. A dose-response study of the effects of intravenous midazolam on cold pressor-induced pain. Anesth. Analg. 80:521–525; 1995.