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Effects of 5-Hydroxytryptamine$_3$ Antagonist, Ondansetron, on Cigarette Smoking, Smoke Exposure, and Mood in Humans

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ZACNY, J. P., J. L. APFELBAUM, J. L. LICHTOR AND J. G. ZARAGOZA. *Effects of a 5-hydroxytryptamine$_3$ antagonist, ondansetron, on cigarette smoking, smoke exposure, and mood in humans.* PHARMACOL BIOCHEM BEHAV **44**(2) 387-391, 1993. – Recent studies in the animal laboratory indicate that 5-hydroxytryptamine$_3$ (5-HT$_3$) antagonists reduce the reinforcing effects of several psychoactive drugs, including nicotine. The purpose of our study was to determine if ondansetron, a selective 5-HT$_3$ antagonist, affected tobacco cigarette consumption in smokers. In the first experiment, a prospective, crossover, placebo-controlled trial was used in which subjects (N = 7) were exposed in an inpatient research unit to 0, 0.15, 0.3, or 0.45 mg/kg ondansetron in three equally divided doses given 4 h apart. In the second experiment, seven different subjects were exposed to the same trial except the dose range was reduced to about 10%: 0, 0.01, 0.02, or 0.04 mg/kg. In each experiment, order of dosing conditions was determined by a Latin square design. Dependent measures included number of cigarettes smoked during the 24-h session, biologic exposure levels (carbon monoxide and plasma nicotine levels), and mood. Number of cigarettes smoked and biologic exposure levels did not differ across drug conditions in either experiment. There were also no effects of ondansetron on mood. From our study results, we conclude that acute administration of ondansetron, at doses appropriate for antiemesis or at markedly lower doses, does not alter tobacco consumption or smoke exposure in humans.

Ondansetron Cigarette Tobacco Smoke exposure Nicotine Cotinine Carbon monoxide
Serotonin 5-HT$_3$ antagonist Mood Human

ONDANSETRON is a selective 5-hydroxytryptamine$_3$ (5-HT$_3$) receptor antagonist. Its clinical indication is for the prevention and treatment of chemotherapy-induced emesis and nausea (6,16,23). However, there are a number of preclinical studies that suggest other interesting applications of this 5-HT$_3$ antagonist in the area of drug abuse and pharmacotherapy [see (2,26) for reviews]. In animal models of anxiety and cognition/memory, 5-HT$_3$ antagonists have proven efficacious (3,10). In other studies, behavioral consequences of withdrawal (e.g., suppressed behavior) from chronic morphine, nicotine, diazepam, cocaine, and ethanol administration in rats and mice were attentuated by 5-HT$_3$ antagonists (8,9). Also, several studies have shown in rats that certain effects of ethanol, nicotine, and morphine are attenuated by 5-HT$_3$ antagonists. In one such study, MDL 72222, a selective 5-HT$_3$ antagonist, reduced alcohol consumption (14). In another study, the discriminative stimulus effects of ethanol were reduced by MDL 72222 (15). Carboni et al. (6) demonstrated that the reinforcing effects of nicotine, morphine, and ethanol, as assessed by place preference testing, were reduced by MDL 72222 and ICS-205,930, another selective 5-HT$_3$ antagonist. The way in which these 5-HT$_3$ antagonists reduced the reinforcing effects of the drugs was investigated by the same research group (5). In that study, dopamine (DA) release induced by nicotine, morphine, and ethanol, as measured in freely moving rats via transcerebral dialysis, was reduced in a dose-dependent fashion by ICS-205,930. It was concluded that 5-HT$_3$ antagonists reduce reinforcing effects of ethanol, nicotine, and morphine by blocking the ability of these drugs to stimulate the firing activity of DA neurons and subsequent release of DA into the synapse. It is possible that other behavioral effects of these drugs (discriminative stimulus effects) are also affected by the reduction in DA release.

If the reinforcing effects of nicotine are blunted by a 5-HT$_3$ antagonist, it is possible that 5-HT$_3$ antagonists could be used as pharmacotherapeutic agents for smokers interested in quit-

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ting smoking (28). On the other hand, smokers might increase their smoking rate to override or compensate for the effects of the 5-HT_3 antagonist, in much the same way that blockade of nicotinic receptors by mecamylamine, a nicotine antagonist, increases smoking rates (22,24). To investigate whether a 5-HT_3 antagonist alters cigarette consumption and, if so, in what direction, healthy cigarette smokers were given acute administration of ondansetron, a selective 5-HT_3 antagonist, in a clinical research ward while their cigarette consumption, smoke exposure, and mood were assessed.

METHOD

Subjects

This study was approved by the local Institutional Review Board. Informed consent from each subject was obtained prior to initiating the study. Subjects were cigarette smokers, aged 21–39 years, who had smoked regularly for at least 2 years prior to the study. Subjects reported at least 15 cigarettes/day on a regular basis and inhaling the tobacco smoke. Subjects were told that the purpose of the study was to examine the effects of drugs on mood and behavior, and, in the informed consent form, that the drug(s) to be used were a) commonly used in medical settings, b) may come from one of six classes (i.e., sedative/tranquilizer, stimulant, opiate, antiemetic, alcohol, or placebo), and c) would be clinically safe at the dose(s) used. Subjects were not told that the drug was to be studied in relation to their smoking behavior.

Prior to the first session, subjects attended a screening interview, at which point they completed the SCL-90 (a questionnaire designed to assess psychiatric symptomatology) (12) and a health questionnaire to determine their psychiatric and medical status. A structured psychiatric screening interview was conducted by one of the experimenters (J.G.Z.). Candidates with any history of significant psychiatric disorders or substance use disorder (1) were excluded. A physician performed a medical history and physical examination; volunteers with a history of neurological, cardiac, pulmonary, hepatic or renal disease, or any other medical contraindication were excluded from the study.

Subjects were instructed to refrain from using recreational drugs (including alcohol) for 24 h before and 12 h after the sessions. Payment for study participation was made after completion of the experiment.

Experimental Design

A prospective, crossover, placebo-controlled, double-blind trial was initiated in which subjects in each experiment were exposed across four sessions, spaced at least 1 week apart, to saline and three doses of ondansetron. Order of dosing conditions was established according to a Latin square. In Experiment 1, we chose our range of doses based upon the recommended therapeutic dose for treatment of nausea/emesis, 31.5 mg/daily in three equally divided doses. The doses we tested were 0, 0.15, 0.3, or 0.45 mg/kg ondansetron in three equally divided doses given 4 h apart. In Experiment 2, we tested a much lower range of doses—0, 0.01, 0.02, or 0.04 mg/kg in three equally divided doses—both because of the lack of effect in Experiment 1 and because of a recent study in which a relatively low dose of oral ondansetron, 0.25 mg b.i.d., reduced alcohol consumption in alcoholics (27).

Procedure

The experiment took place in a Clinical Research Center (CRC), an inpatient research unit at the University of Chicago Hospitals. Each subject had his own room, equipped with a bathroom, bed, chair, and TV. Subjects could engage in a variety of activities while they were at the CRC but could not leave the premises of the CRC. Most subjects spent their time studying, engaging in hobbies (e.g., drawing), and/or watching movies on the TV/VCR. Subjects arrived at the CRC between 2100 and 2200 h the evening before the first session and were discharged from the hospital 35 h later.

Each 24-h session was conducted from 0800–0759 h. At 0800 h, a nurse inserted an angiocatheter into a forearm vein and then attached the catheter to a 50-cc bag of saline containing the drug (or placebo). The contents of the bag were infused into the vein over a 5-min period. This drug administration regimen was repeated at 1200 and 1600 h. An ample supply of the subject's preferred brand of cigarettes was left in the subject's room at 0800 h. No restrictions were placed on subjects' smoking behavior.

Dependent Measures

24-h cigarette consumption. Twenty-four hour cigarette consumption, from 0800–0759 h, was monitored by having subjects record the time of day each cigarette was smoked and having them save butts from all cigarettes smoked. Typically, the self-motivated smoking count deviated from the butt count by no more than one or two. If there was a deviation between the two counts, the larger of the two numbers was considered the cigarette/day measure.

Smoke exposure levels. Smoke exposure was measured via carbon monoxide (CO) levels on subjects' breath and their venous levels of nicotine and cotinine. Expired-air CO levels, indicative of overall biologic smoke exposure, were collected using the procedure described in Zacny and Stitzer (30) at 0800, 0900, 1500, and 2100 h. Seven milliliters of blood were drawn from a forearm vein at 2000 h. Plasma nicotine and cotinine levels were determined by gas chromatography (18). Nicotine levels collected in the late afternoon or evening hours are considered representative of daily nicotine exposure (4). Cotinine, a metabolite of nicotine, is also considered a sensitive marker of tobacco smoke intake (4).

Subjective effects measures. Subjects several times during each session completed two mood forms: the Profile of Mood States (POMS) and a Symptoms Checklist. The POMS is a 72-item adjective checklist; subjects are instructed to rate each adjective using a five-point scale according to their feelings at that moment (21). Eight clusters of adjectives (scales) have been separated using factor analysis: anxiety, depression, anger, vigor, fatigue, confusion, friendliness, and elation. Two additional scales, arousal and positive mood, were derived from the other scales as follows: arousal = (anxiety + vigor) − (fatigue + confusion); positive mood = elation − depression. The POMS was completed at 0800, 0900, 1500, and 2100 h. On the Symptoms Checklist (17), subjects were asked to rate the degree to which they experienced each of 11 symptoms (cigarette craving, urge to smoke, irritability, difficulty in concentrating, headache, impatience, drowsiness, restless, anxious, depression, anger) during the previous 6-h time period using a 0–3 scale (0, not present; 1, mild; 2, moderate; and 3, severe). This checklist was completed at 1500 and 2100 h.

Data Analysis

Univariate analysis of variance (ANOVA) for repeated measures was used to analyze the dependent measures in this study. Factors used were dose (four levels) and, in most cases, time (four levels) (except for daily cigarette consumption and nicotine and cotinine levels). *F*-Values were considered significant for $p \leq 0.05$ with adjustments of within-factors degrees of freedom (Huynh–Feldt) to protect against violations of symmetry.

RESULTS

Four males and three females participated in Experiment 1 (mean age ± SD: 30.7 ± 4.9 years) and in Experiment 2 (mean age ± SD 25.4 + 5.1 years). Mean daily self-reported cigarette consumption of Experiment 1 subjects was 20.7 ± 4.5 cigarettes and for Experiment 2 subjects 27.1 ± 7.6 cigarettes. Average nicotine yield of subjects' cigarette brands in Experiments 1 and 2 were 0.8 ± 0.2 and 1.0 ± 0.3 mg, respectively.

Number of cigarettes smoked did not differ as a function of ondansetron dose in either experiment. Figure 1 shows that Experiment 2 subjects consumed more cigarettes, on average, than subjects in Experiment 1. Figure 1 also shows that ondansetron had no effect on expired-breath CO levels, plasma nicotine levels, or plasma cotinine levels. In both experiments, there was a significant time effect with CO levels, with levels increasing as the session progressed [Experiment 1, $F(3, 18) = 11.2$, $p < 0.002$; Experiment 2, $F(3, 18) = 14.5$, $p < 0.001$]. Ondansetron did not affect mood as assessed by the 10 scales of the POMS and the 11 adjectives/nouns from the Symptoms Checklist in either experiment. In Experiment 2, a significant time effect was obtained with the symptom headache, $F(1, 6) = 8.4$, $p < 0.02$. Average headache ratings at 1500 and 2100 h were 0.8 and 3.5, respectively and were nearly identical across the four dosing conditions.

DISCUSSION

A number of studies have shown in the animal laboratory that 5-HT_3 antagonists may have clinical potential beyond their antiemetic effects. Enhanced cognition and anxiolysis have been shown with these agents [cf. (3)]. Reinforcing and discriminative stimulus effects of drugs of abuse have been altered by 5-HT_3 antagonists (6,15). In particular, the reinforcing effects of nicotine have been reduced by 5-HT_3 antagonists (6), perhaps via their modulating role on nicotine-induced release of DA (5). Because the psychoactive component of tobacco thought to be responsible for its consumption is nicotine (25), and in rats 5-HT_3 antagonists have an effect on the reinforcing effects of nicotine, we reasoned that tobacco cigarette consumption in human smokers might also be altered. Using two different dose ranges of ondansetron, however, neither cigarette consumption nor smoke exposure levels were affected.

One criticism that might be directed at this study is the sample sizes in both experiments, seven subjects, which may have resulted in a lack of statistical power to detect a drug effect on the dependent measures in this study. This is a legitimate criticism, but it should be pointed out that in both studies there were no consistent increasing or decreasing trends in any of the smoking measures across ondansetron doses, which would suggest the drug was exerting an effect on smoking. Taking cotinine levels as an example, in Experiment 1 one subject showed an increase in cotinine levels across doses, two subjects showed a decrease, and the other four showed either no effect or no consistent trend across doses. In Experiment 2, two subjects showed an increase across doses, two subjects showed a decrease, and the other three showed no effect or consistent trend across doses. The same patterning of variability across subjects was evident with cigarette consumption and CO exposure. In summary, then, our results might be taken as preliminary due to the small sample sizes, but the data in any event does indicate lack of an effect of ondansetron on smoking consumption and smoke exposure.

At least three possibilities exist for the negative results obtained in this study besides the notion that 5-HT_3 antagonists have no effect on tobacco consumption in humans. First, it may be the case that had a longer time period than 24 h been used we would have eventually detected a drug effect. Indeed, ondansetron reduced alcohol consumption in alcoholics, but only after the drug had been given on a daily basis for 6 weeks (27). However, effects of 5-HT_3 antagonists have been obtained in several animal studies in which these drugs were given on an acute basis (5,6,8,9,13,29). Second, it could be argued that ondansetron does affect cigarette consumption but at a different dose range than the ones we tested. We limited our highest dose, 0.45 mg/kg, based upon the daily dose recommended for its clinical effect on nausea and vomiting. Considering both experiments, doses were used that differed almost 50-fold from each other, so we felt that an adequate range of doses were tested. It is still possible, though, that even a lower dose than the lowest dose tested in Experiment 2, 0.01 mg/kg (or 0.003 mg/kg three times daily), might have altered cigarette consumption. It has been noted in other studies that lower doses of 5-HT_3 antagonists are more likely to have behavioral effects than high doses of these agents (7,11). And, last, the setting in which the experiment took place, a hospital room, may have decreased the chances of detecting a drug effect on smoking or smoke exposure. Because the range of activities in which subjects could engage in this setting was somewhat limited, perhaps this would encourage the activities allowed to increase in frequency, including smoking. It should be acknowledged that setting is a potent variable that can influence drug consumption and subjective effects of drugs (19,20). We were limited to conducting the study in a hospital setting, though, because subjects had to be parenterally administered the drug several times over the course of the study day.

In the present study, we also found lack of an effect of ondansetron on mood, as measured by a standardized mood inventory and a symptoms checklist that included such items as cigarette craving, drowsiness, and anxious. We included mood as a dependent measure for two reasons: First, although it has been claimed in clinical studies that ondansetron (at doses similar to those used in Experiment 1) has no effects on mood, no study has systematically investigated this. Second, if ondansetron had decreased cigarette consumption in this study, it would have been important to document from a clinical standpoint that the decrease occurred in the absence of sedating or dysphoric effects of the drug, effects that would contraindicate its usage for smoking reduction therapy.

We conclude from our findings that, at least at the dose ranges tested, ondansetron neither increases nor decreases cigarette consumption or smoke exposure and does not alter ciga-

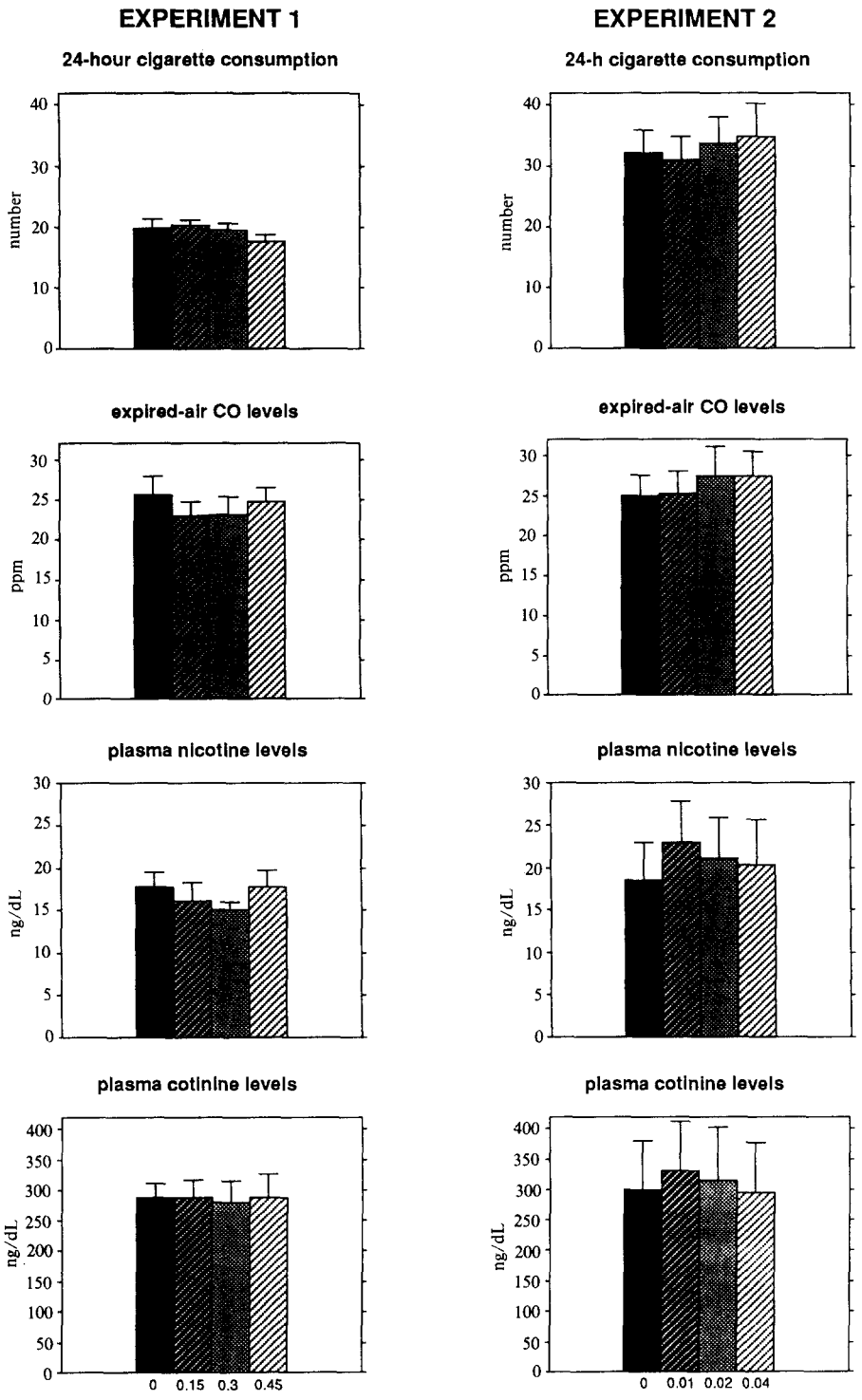

FIG. 1. Effects of ondansetron (Experiment 1: left; Experiment 2: right) on 24-h cigarette consumption, expired-air carbon monoxide levels [expressed in parts per million (ppm)], and plasma nicotine and cotinine levels (expressed as ng/dl). Ondansetron dose is expressed as the total amount of ondansetron given during a 24-h session—the dose was equally divided into three doses and administered at 0800, 1200, and 1600 h. Data is averaged across the seven subjects in each of the two experiments, brackets indicate SEM.

rette craving or the urge to smoke. Our study findings suggest that acute administration of ondansetron and perhaps other 5-HT_3 antagonists would not be beneficial in helping motivated smokers to cut down on their smoking. Whether chronic administration of these agents would affect cigarette smoking remains to be determined. Another related area to explore is the effects of 5-HT_3 antagonists on cigarette withdrawal symptomatology because behavioral suppression induced by nicotine withdrawal in mice and rats was attenuated by ondansetron (9).

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REFERENCES

1. American Psychiatric Association. Diagnostic and statistical manual of mental disorders (DSM-III-R). 4th ed. Washington, DC: American Psychiatric Association; 1987.
2. Barnes, J. M.; Barnes, N. M.; Cooper, S. J. Behavioural pharmacology of 5-HT_3 ligands. Neurosci. Biobehav. Rev. 16:107–113; 1992.
3. Barnes, J. M.; Costall, B.; Coughlan, J.; Domeney, A. M.; Gerrard, P. A.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S.; Tomkins, D. M.; Tyers, M. B. The effects of ondansetron, a 5-HT_3 receptor antagonist, on cognition in rodents and primates. Pharmacol. Biochem. Behav. 35:955–962; 1990.
4. Benowitz, N. L. The use of biologic fluid samples in assessing tobacco smoke consumption. In: Grabowski, J.; Bell, C. S., eds. Measurement in the analysis and treatment of smoking behavior. NIDA research monograph series 48. Rockville, MD: U.S. Government Printing Office; 1983:6–26.
5. Carboni, E.; Acquas, E.; Frau, R.; Di Chiara, G. Differential inhibitory effects of a 5-HT_3 antagonist on drug-induced stimulation of dopamine release. Eur. J. Pharmacol. 164:515–519; 1989.
6. Carboni, E.; Acquas, E.; Leone, P.; Di Chiara, G. 5-HT_3 receptor antagonists block morphine- and nicotine- but not amphetamine-induced reward. Psychopharmacology (Berl.) 97:175–178; 1989.
7. Costall, B.; Domeney, A. M.; Gerrard, P. E.; Jones, B. J.; Kelly, M. E.; Oakley, N. R.; Tyers, M. B. The anxiolytic activities of the 5-HT_3 antagonists GR38032F, ICS 205-930 and BRL 43694. In: Bevan, P.; Cools, A. R.; Archer, T., eds. Behavioural Pharmacology of 5-HT. Hillsdale, NJ: Lawrence Erlbaum; 1989:383–387.
8. Costall, B.; Jones, B. J.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S.; Tyers, M. B. The effects of ondansetron in rats and mice treated subchronically with diazepam. Pharmacol. Biochem. Behav. 34:769–778; 1989.
9. Costall, B.; Jones, B. J.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S.; Tyers, M. B. Ondansetron inhibits the behavioral consequences of withdrawing from drugs of abuse. Pharmacol. Biochem. Behav. 36:339–344; 1990.
10. Costall, B.; Naylor, R. J. Anxiolytic effects of 5-HT3 antagonists in animals. In: Rodgers, R. J.; Cooper, S. I., eds. 5-HT1A agonists, 5-HT3 antagonists and benzodiazepines: Their comparative behavioural pharmacology. Chichester, UK: John Wiley; 1991: 133–157.
11. Costall, B.; Naylor, R. J.; Tyers, M. B. The psychopharmacology of 5-HT_3 receptors. Pharmacol. Ther. 47:181–191; 1990.
12. Derogaitis, L. R.; Lipman, R. S.; Cozi, L. SCL-90: An outpatient psychiatric rating scale—preliminary report. Psychopharmacol. Bull. 9:13–17; 1973.
13. Dunn, R. W.; Carlezon, W. A.; Corbett, R. Preclinical anxiolytic versus antipsychotic profiles of the 5-HT_3 antagonists ondansetron, zacopride, 3-alpha-tropanyl-1*H*-indole-3-carboxylic acid ester, and 1-alpha-*H*-3-alpha,5-alpha-*H*-tropan-3-*yl*-3,5-dichlorobenzoate. Drug Dev. Res. 23:289–300; 1991.
14. Fadda, F.; Garau, B.; Marchei, F.; Colombo, G.; Gessa, G. L. MDL 72222, a selective 5-HT_3 receptor antagonist, suppresses voluntary ethanol consumption in alcohol-preferring rats. Alcohol Alcohol. 26:107–110; 1991.
15. Grant, K. A.; Barrett, J. E. Blockade of the discriminative stimulus effects of ethanol with 5-HT_3 receptor antagonists. Psychopharmacology (Berl.) 104:451–456; 1991.
16. Hesketh, P. J.; Gandara, D. R. Serotonin antagonists: A new class of antiemetic agents. J. Natl. Cancer Inst. 83:613–620; 1991.
17. Hughes, J. R.; Hatsukami, D. K.; Pickens, R. W.; Krahn, D.; Malin, S.; Luknic, A. Effects of nicotine on the tobacco withdrawal syndrome. Psychopharmacology (Berl.) 83:82–87; 1984.
18. Jacob, P. T.; Wilson, M.; Benowitz, N. L. Improved gas chromatographic method for the determination of nicotine and cotinine in biologic fluids. J. Chromatogr. 222:61–70; 1981.
19. Jones, R. T. Marijuana induced "high": Influence of expectation, social setting and previous drug experience. Pharmacol. Rev. 23: 359–369; 1971.
20. Lindman, R. Social and solitary drinking: Effects on consumption and mood in male social drinkers. Physiol. Behav. 28:1093–1095; 1982.
21. McNair, D.; Lorr, M.; Droppleman, L. Profile of mood states (manual). San Diego, CA: Educational and Industrial Testing Service; 1971.
22. Nemeth-Coslett, R.; Henningfield, J.; O'Keefe, M.; Griffiths, R. Effects of mecamylamine on human cigarette smoking and subjective ratings. Psychopharmacology (Berl.) 88:420–425; 1986.
23. Physician's desk reference Oradell, NJ: Medical Economics Company; 1992.
24. Pomerleau, C.; Pomerleau, O.; Majchrzak, M. Mecamylamine pretreatment increases subsequent nicotine self-administration as indicated by changes in plasma nicotine level. Psychopharmacology (Berl.) 91:391–393; 1987.
25. Rose, J. E. Jarvik, M. E.; Ananda, S. Nicotine preference increases after cigarette deprivation. Pharmacol. Biochem. Behav. 20:55–58; 1984.
26. Sellers, E. M.; Higgins, G. A.; Sobell, M. 5-HT and alcohol abuse. Trends Pharmacol. Sci. 13:69–75; 1992.
27. Sellers, E. M.; Higgins, G. A.; Tomkins, D. M.; Romach, M. K. Serotonin and alcohol drinking. NIDA Res. Monogr. 119:141–145; 1992.
28. Sellers, E. M.; Higgins, G. A. Tomkins, D. M.; Romach, M. K.; Toneatto, T. Opportunities for treatment of psychoactive substance use disorders with serotonergic medications. J. Clin. Psychiatry 52:S49–S54; 1991.
29. Yoshimoto, K.; McBride, W. J.; Lumeng, L.; Li, T. K. Alcohol stimulates the release of dopamine and serotonin in the nucleus accumbens. Alcohol 9:17–22; 1991.
30. Zacny, J. P.; Stitzer, M. L. Cigarette brand-switching. Effects on smoke exposure and smoking behavior. J. Pharmacol. Exp. Ther. 246:619–627; 1988.