Pharmacology Biochemistry & Behavior, Vol. 26, pp. 749–755. © Pergamon Journals Ltd., 1987. Printed in the U.S.A. 0091-3057/87 $3.00 + .00

Harman Induces Preference for Ethanol in Rats: Is the Effect Specific for Ethanol?

HANS ROMMELSPACHER, CORNELIA BÜCHAU AND JÜRGEN WEISS

Department of Neuropsychopharmacology, Free University Ulmenallee 30, D-1000 Berlin 19, F.R.G.

Received 21 July 1986

ROMMELSPACHER, H., C. BÜCHAU AND J. WEISS. *Harman induces preference for ethanol in rats: Is the effect specific for ethanol?* PHARMACOL BIOCHEM BEHAV **26**(4) 749–755, 1987.—Increasing concentrations of either ethanol, etonitazene, clomethiazole or midazolam were offered to male Wistar rats for 21 days. Between day 8 and day 21, the animals were treated with several doses of harman, harmalan, and tetrahydronorharman (tetrahydro-β-carboline) by means of continuous intraventricular infusion. Harman and THN induced a significant preference for ethanol in a dose-dependent manner. Harman was approximately three times more potent than THN. The amount of ethanol consumed during the second and third weeks of the experimental period correlated with the harman concentration in the brain after the cessation of the treatment ($p<0.01$). Harman infusion attenuated the clomethiazole intake, whereas that of etonitazene and midazolam was not affected as compared with CSF-treated rats. By counting licking movements, it was found that the rats drank ethanol and water at distinct time periods with the pattern dependent on the concentration of the ethanol solution offered. The intervals between the maxima were 6 to 8 hours at low ethanol concentrations. Relatively high concentrations caused a disruption of the regular rhythms in favour of shorter ones with increasing intervals between the maxima (3 hr, 4 hr, 5 hr intervals). Harman treatment (27 nmol/hr) disturbed the regular rhythms at lower ethanol concentrations but mimicked the ultradian rhythm which was observed at high ethanol concentrations in CSF-treated animals. The observed coincidence of water and ethanol intake was uncoupled if the highest ethanol concentration in both treatments was offered. Thus, treatment with harman changed the rhythm of fluid intake in a direction which was detected in CSF-treated rats only at relatively high ethanol concentrations.

Beta-carbolines Harman Tetrahydronorharman Harmalan Preference for ethanol Etonitazene Midazolam Clomethiazole Ultradian rhythm

INTRACEREBROVENTRICULAR infusion of approximately 40 nmol/hr tetrahydronorharman (THN, tetrahydro-β-carboline) over a period of 12 days virtually increased the ethanol intake of male Sprague Dawley rats by seven fold per day; in one rat it was up to 16.6 g/kg/day, despite increasing concentrations of ethanol [15]. Thus, even though the concentration of ethanol became more gustatorily aversive towards the end of the drinking period, the rats nevertheless selected increasing quantities of ethanol. When they were once again given the choice of alcohol 1 month after the cessation of the intraventricular application, the rats resumed ingestion of ethanol although there had been no access to the drug in the interim [15]. These studies were continued with Kuopio-Wistar rats which spontaneously consumed an average of 0.9 g/kg/day to 6.5 g/kg/day depending on the ethanol concentration offered [3,27]. The authors demonstrated that infusion of 47 nmoles/hr THN or 1-Me-THN caused a 100% increase in voluntary ethanol consumption with a lack period of 6 days. The same dose of 6-MeO-THN, a serotonergic β-carboline, was ineffective.

These findings are remarkable, in that they might help to generate hypotheses for why some rats/humans drink more ethanol than others. Many attempts have been undertaken to explain the preference for ethanol. For example, drugs which suppress REM-sleep, like noradrenaline-, dopamine-, and 5-hydroxytryptamine-uptake inhibitors administered to rats in the postnatal period, increased the voluntary ethanol consumption of the adult animals [8] as did REM-sleep deprivation [1]. Unspecific stimulants like amphetamine and nicotine also increased ethanol intake. This is probably due to its sedative activity [20,21]. Placing rats or monkeys in a new environment induced increased ethanol consumption as well [12,26]. Infantile handling resulted in increased preference for 10% ethanol compared with water [11]. Pups whose mothers were exposed to ethanol consumed significantly more ethanol in a preference test compared to control offspring [4]. These are only a few examples which demonstrate that a common denominator for explaining ethanol preference is far from being established. Therefore, continuing the investigations about the effect of β-carbolines on ethanol preference seemed worthwhile. A further reason for pursuing this work is the observation that some of the BC's occur in vivo and increase in the brain and urine of rats after a load with ethanol [22] and are excreted in the urine of alcoholics in higher concentrations than in that of non-alcoholics [2,23].

METHOD

Male Wistar rats (breeder: Hagemann Boesingfeld, F.R.G.) weighing 290–390 g, were maintained individually in

macrolon cages (43×26×15 cm). The animal room was air conditioned (21–24°C: air humidity 50±5%) with a 12/12 hr light/dark cycle installed. Each rat's selection was tested by a standard three-bottle two-choice technique [14]. Three drinking tubes were offered with one tube containing a solution of ethanol which was increased every other day as follows: 3, 5, 7, 9, 11, 13, 15, 17, 20, 25, 30% (v/v). Each solution was prepared in tap water with 95% ethanol. The second tube was filled with water, whereas the third tube served as dummy and was empty. The position of the three tubes was interchanged each day in random order to prevent the rat from developing a position habit.

Chronic Infusion Procedure

Unselected rats were anesthetized with chloralhydrate (300 mg/kg IP) and mounted in a stereotaxic apparatus (David Kopf, small animal stereotaxic instrument). An infusion cannula (inner diameter 0.8 mm) was implanted so that the tip rested in the lateral ventricle and fixed on the surface of the skull with two screws and dental cement. The day of the operation was the first day on which the lowest concentration of ethanol was offered (day one of the experimental period). Consumption of ethanol and water was measured daily by weighing each bottle.

After 7 days, osmotic minipumps (either ALZET 2001 or ALZET 2 ML 1) filled with artificial cerebrospinal fluid (CSF; pyrogen-free) or the respective β-carboline (BC) dissolved in CSF were implanted under the skin of the neck and connected with the cannula by silicone tubing. Seven days later, a second minipump was implanted. The remaining solution from the reservoirs of several pumps was analyzed by HPLC with respect to a possible decomposition of the BC's. At the end of the treatment, the position of the cannula was verified by histology in some animals. The harman concentration was measured in the brain of the vast majority of the rats.

Measurement of the Number of Licks

Animals treated either with CSF or 27 nmol/hr harman were placed in individual cages from 16 hr until 8 hr. The dark period lasted from 20 hr until 8 hr. The licking apparatus consisted of two bottles in which a metallic ball was inserted in the outlet. The ball served as a valve and could easily be moved back by the tongue of the rats. This action elicited an electrical impulse which was counted, added up separately for each bottle for a period of one hour, and registered. The time course for the water and ethanol consumption was calculated based on the number of licking movements, their distribution over the registration period and the fluid consumption.

Analytical Methods

Determination of blood ethanol. The concentration of ethanol was measured in blood samples collected after decapitation utilizing an enzymatic method based on the formation of NADH by the alcoholdehydrogenase (Boehringer, Mannheim, F.R.G.). The extinction at 365 nm was determined by a photometer (Zeiss, PMQ II). The standard curve was found linear up to 6 g/l of ethanol.

Determination of harman in rat brain. The method for measuring the concentration of harman was described previously [22]. In summary, rats were decapitated by a guillotine at the end of the twenty first day following the implantation of the cannula. The brains were rapidly removed and homogenized in 50 vol. (w/v) of 0.45 mol/l perchloric acid. Two aliquots of the homogenate containing approximately 300 mg tissue were utilized further. Two ng harman standard (free base) were added to one of the samples. Both aliquots were centrifuged at 26000×g for 10 min (+5°C) in a Spinco L-50 centrifuge (Beckman Ltd., Fife, UK). The supernatant was decanted. The pellet was suspended in 2.5 ml of 0.45 mol/l perchloric acid, homogenized and centrifuged as above. Both supernatants were combined and made basic (pH 10.2) by potassium hydroxide. Following a further centrifugation step (26000×g, 10 min), harman was extracted twice into 20 ml diethylether which was evaporated to dryness. The residue was dissolved in 150 μl methanol. A 100 μl aliquot was spotted on silica gel thin layer plates (HPTLC quality, Merck, Darmstadt, F.R.G.) using a Linomat III apparatus (Camag, Muttenz, Switzerland). The plates were developed in chloroform/methanol (92:8). The authentic fluorescence was measured by a densitometer (Camag, 302 nm excitation, emission: cut off filter at 400 nm). The amount of harman extracted from the tissue was calculated by the internal standard method (for details see [22]).

Assessment of decomposition of BC's in the minipumps during the infusion period. After the end of the treatment period, the remainder of the BC solution was collected from the minipumps and pooled. Furthermore, 3 minipumps were filled with either harman, THN or harmalan solutions and wrapped in aluminum foil. They were implanted under the skin of the neck without connecting tubes. Seven days later, the concentration of BC's in the solutions was measured by HPLC (Hewlett Packard, B 3680). The BC's were eluted from a RP 18 column by a 17 mmol/l sodiumphosphate buffer, pH 8, and 80% methanol (Uvasol®-grade) using an UV-spectrometer adjusted to 254 nm for detection.

Statistics

Analysis of least squares. The time-course of the ethanol consumption was analyzed with respect to linear trends. For this purpose, calculations of the least squares were conducted separately for the period before and during infusion. The calculation utilized the means of each regimen and day. The calculated regression coefficients were tested under two aspects: Whether they were significantly different from zero (level: 5%) and whether the coefficients of the BC-treated animals differed from those of CSF-treated rats. In both cases the calculated parameters followed a Student-*t*-distribution [28].

Auto- and crosscorrelation. Some of the time courses were analyzed with respect to ultradian rhythms as well as the temporal relationship between ethanol and total fluid consumption. For that purpose, a parametric auto- and crosscorrelation procedure was applied [13,24]. The calculation of the crosscorrelation was performed by measuring the product-moment-correlation between the values of two time series based on given temporal changes. One of them was shifted in one hour steps in the course of the calculation. High positive and negative correlations at certain moments pointed to a temporal correlation of both parameters. The calculated correlation values could be used to assess significant agreements [28]. In principle, the autocorrelation utilizes the same procedure as the crosscorrelation however, only one time series is compared with itself. This method is suited to revealing rhythmic changes of a given parameter [13, 19, 29].

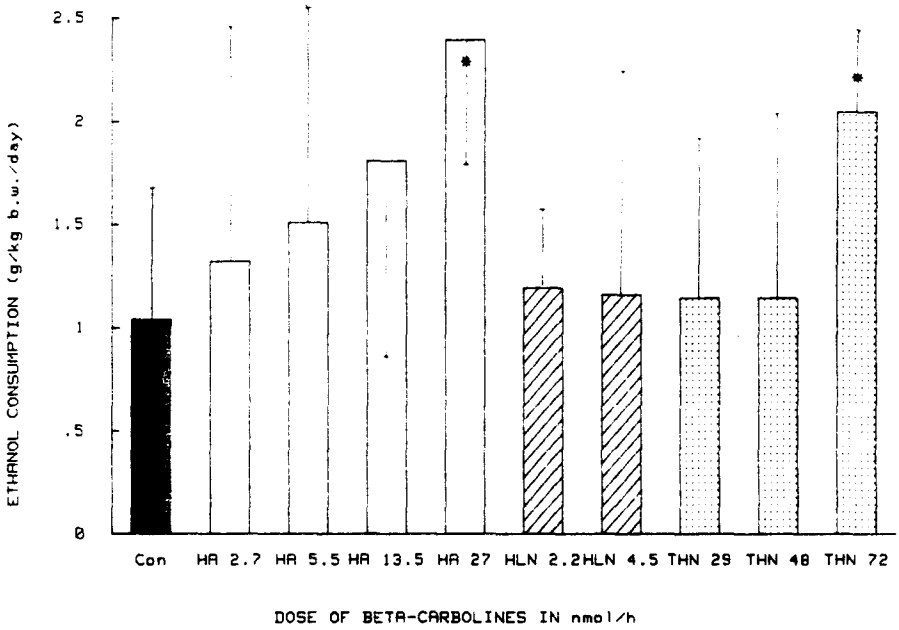

FIG. 1. Values are the means±SD of 4 to 13 rats. Con: vehicle-treated controls, THN: tetrahydronorharman. HA: harman; HLN: harmalan. *$p<0.001$.

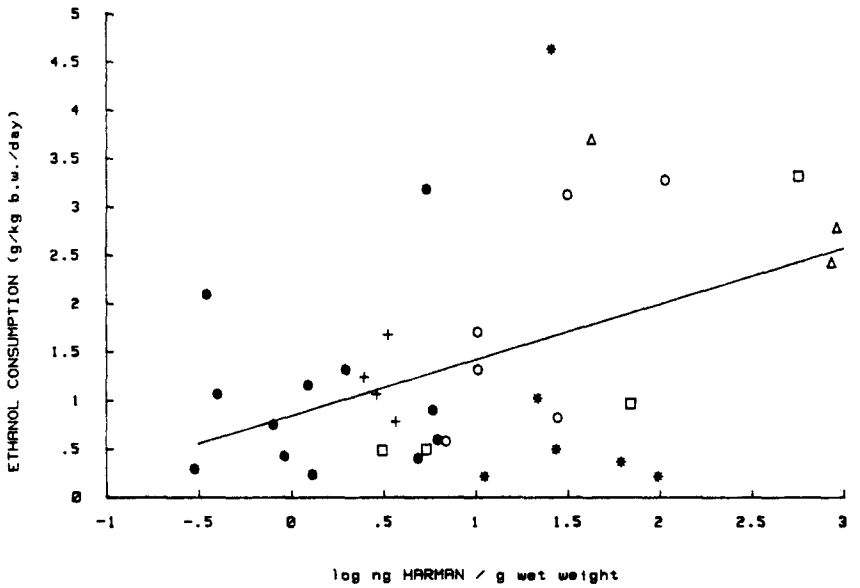

FIG. 2. Animals were treated IVT with either CSF, harmalan or harman for 14 days. The mean of the ethanol consumption per day is depicted against the concentration of harman in the brain measured at the end of the observation period. The values represent results from single rats. The slope of the calculated regression line is different from zero ($p<0.01$). ●: CSF-treated controls; ○: harman (5.5 nmol/hr); +: harmalan (2.2 nmol/hr); □: harman (2.7 nmol/hr); △: harman (27 nmol/hr); *: harmalin (4.5 nmol/hr).

RESULTS

Effect of Various β-Carbolines on the Consumption of Ethanol in a Free Choice Trial

The effect of various BC's on ethanol consumption is depicted in Fig. 1. During the observation period, the second and third weeks of the experiment, the ethanol concentration increased from 9 to 30%. Continuous infusion of harman into the lateral ventricle of the brain induced a dose-dependent increase of the consumption of ethanol. The highest dose caused significant differences compared with CSF-treated animals. Both doses of harmalan, namely 2.2 nmol/hr and 4.5 nmol/hr, elicited no statistically significant rise in the consumption of ethanol whereby the variance clearly increased. Higher concentrations of harmalan could not be dissolved in CSF. Only the highest dose of THN (72.1 nmol/hr) provoked an increased ethanol consumption ($p<0.001$).

The water and the total fluid intake were not affected by any treatment. The highest doses of harman and THN tended to increase the fluid intake at approximately the middle of the infusion period. However, the tendency was reversed and reached amounts of controls toward the end of the experiment. From approximately day 7 until the end of the experimental period the volume of the ethanol solution ingested did not change much and consisted of 1/6 to 1/8 of total fluid consumed. The body weight in all groups decreased between 5 and 11%.

At the end of the experimental period, the concentration of harman was determined in the brain of the vast majority of rats. As expected, infusion of harman induced a dose-dependent increase. Harmalan infusion led to an increase of harman levels in the brain as well. The rise was dose-dependent as 2.2 nmol/hr harmalan was ineffective whereas 4.5 nmol/hr caused a significant change ($p<0.001$; Fig. 2). 29.48 and 72.1 nmol/hr THN did not alter harman levels suggesting that THN is neither converted into harman nor inhibits the metabolism. The correlation of the concentration of harman in the brain and the ethanol consumption is depicted in Fig. 2. The symbols of the calculated values are depicted differently for the respective treatments. The calculated regression line has a positive slope which is different from zero [correlation coefficient 0.445; $t(33)=2.856$, $p<0.01$].

The finding that the concentration of harman in the brain was not altered by THN suggests a different mode of action of THN from that of harman. However, both β-carbolines might independently activate a common mechanism leading to an increased consumption of ethanol with harman clearly more effective than THN.

Effect of Harman on the Consumption of Etonitazene, Clomethiazole, and Midazolam

The findings described above did not allow a conclusion about the specificity of the facilitating activity of harman and THN with respect to the consumption of ethanol. Therefore, other drugs with known dependence potential differing in their chemical structure and medical indication were included in the study. Increasing concentrations of drugs were offered to the rats in a free choice paradigm whereby the concentration was changed every other day. The concentrations of the water-soluble opioid etonitazene were as follows: 0.1, 0.2, 0.4, 0.6, 0.8, 1, 1.5, 2, 2.5, 3 μg/ml fluid. The opioid was consumed in increasing amounts which were not altered by the infusion of harman (Fig. 3A). The clomethiazole consumption increased during the observation period

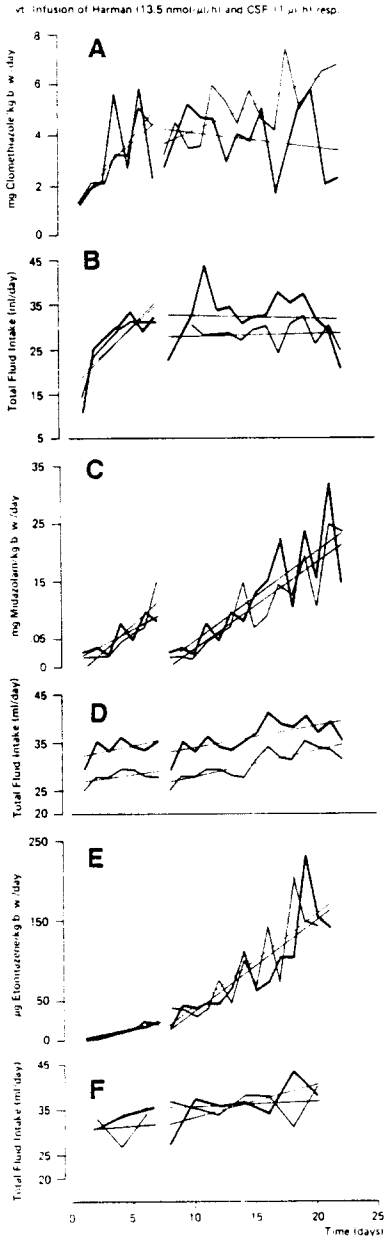

FIG. 3. Time course of the consumption of etonitazene (C), clomethiazole (A), and midazolam (B) as well as of total fluid (drugs + tap water; B, D, F) during the IVT infusion of harman in a free choice trial. At day 1, cannulae were implanted with the tip in the lateral ventricle. At days 7 and 14 the rats received minipumps filled with harman or vehicle (CSF). Values are the means from 8 rats. The time course of the means from rats treated with harman is represented by lines drawn through, whereas that from CSF-treated rats is plotted with dotted lines. The straight lines represent the respective regression lines.

(Fig. 3C). The slope of the regression line was somewhat attenuated after the beginning of the infusion period at day 8, indicating that harman suppressed the increase ($p<0.01$, comparison of the slopes of both regression lines). The total fluid intake, which was lowered after the implantation of the cannula (day 1; Fig. 3D) reached a plateau after about 5 days. The consumption of midazolam was not altered by harman if compared with CSF-treated rats. The same was true for the fluid intake (Fig. 3E and F).

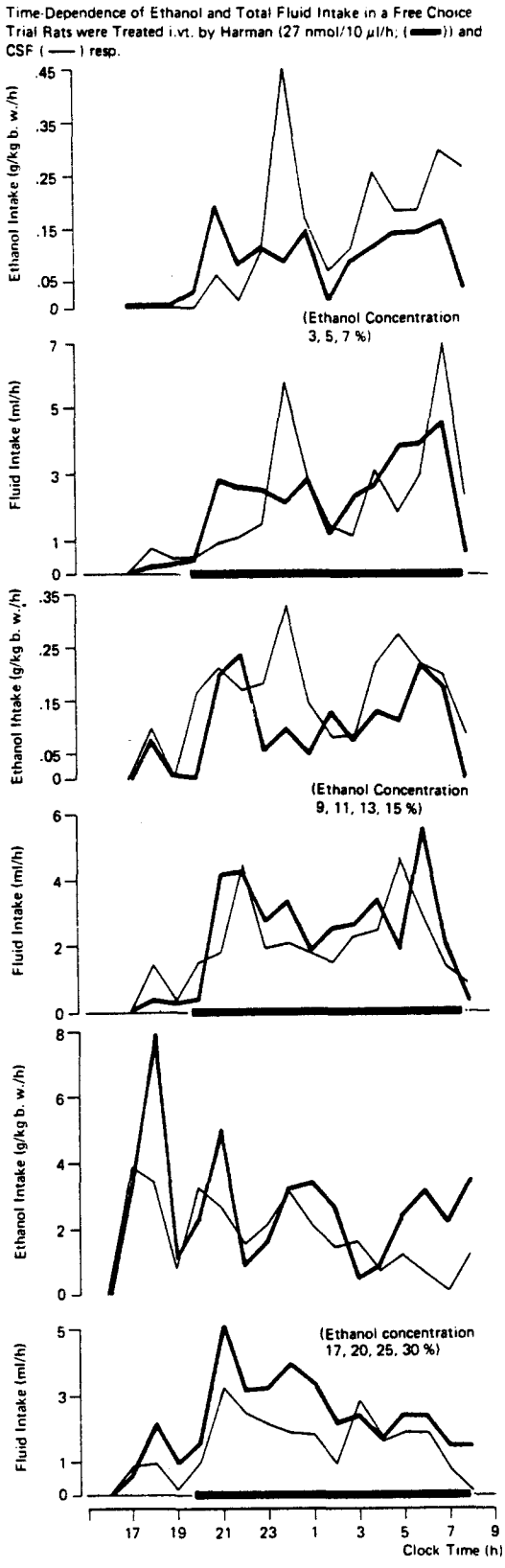

FIG. 4. Rats were kept on a 12/12 hr light/dark cycle (dark period indicated by the dark bar on the abscissa). The concentrations of ethanol offered in a free choice trial are indicated in the graphs. Six naive animals were treated with each concentration. The values are the means from those rats treated with the concentrations of ethanol indicated in the respective graph.

Effects of Harman on the Ultradian Rhythm of the Fluid and Ethanol Consumption Respectively

Two groups of rats (6 animals each) treated with CSF (10 µl/hr) and harman (27 nmol/10 µl/hr) respectively were placed in cages which were connected with an apparatus to register licking movements. The observation period lasted from 16 hr until 8 hr; the dark period was from 20 hr until 8 hr. Various concentrations of ethanol were offered. The findings with ethanol concentrations from 3, 5 and 7% 9, 11, 13 and 15% as well as 17, 20, 25 and 30% were compiled (same experimental set up as described for ethanol treatment in the Method section). As shown in Fig. 4, the rats drank ethanol solutions as well as water at distinct time periods. The pattern depended on the concentration of ethanol in the solution. CSF-treated animals showed two maxima (3–7% and 9–15% ethanol) at approximately 0 hr/22 hr and 7 hr/5 hr, respectively, for the ethanol consumption. Harman-treated animals consumed ethanol over a longer period of time beginning earlier (21 hr) with less clear cut maxima. Independent of the treatment, the rats drank ethanol and water during the same period of time.

The rhythm changed at the highest concentration of ethanol, namely 17–30%. As observed with harman-treated rats with lower ethanol concentrations, the first maximum was shifted to an earlier timepoint in harman as well as CSF-treated rats. It is striking that the 3 maxima for the ethanol intake were found between 17 hr and 1 hr for both CSF- as well as harman-treated rats (Fig. 4, lower section).

The rhythm was evaluated in more detail by auto- as well as crosscorrelation analysis. The autocorrelation of CSF-treated rats, with respect to fluid consumption, revealed a clear periodic behavior with a recurrence of 6 to 8 hours. However, this regular behavior disappeared when the highest concentration of ethanol was offered. Under these conditions, the intervals between the maxima increased progressively from 3 hours to 4 hours to 5 hours (data not shown).

Harman-treated rats displayed a broader maximum for the fluid consumption suggesting an interindividual variation of the periodicity. In no group was a regular rhythm ascertained by statistical means.

The autocorrelation, with respect to ethanol consumption of animals treated with CSF, revealed a clear periodicity with the lower doses of ethanol ($\alpha \leq 5\%$). The pattern corresponded to that of fluid intake (6–8 hours). At the medium doses, the distance of the recurrence was not statistically significant ($\alpha > 5\%$). At the highest doses of ethanol, a shorter rhythm appeared of 3–4 hr ($\alpha \leq 5\%$).

The crosscorrelation analysis which allowed comparison of the pattern of the water and ethanol consumption during the observation period yielded a good temporal correlation ($\alpha < 0.1\%$) for the lowest ethanol concentration for CSF as well as harman-treated rats. Under these conditions the animals drank predominantly ethanol. A good to moderate temporal correlation still existed for both groups with respect to the medium concentrations (9%, 11%, 13%, 15%) ($\alpha < 5\%$) whereby the proportion of ethanol solution consumption was less. At high ethanol concentrations (17%, 20%, 25%, 30%) no temporal correlation existed between the intake of water and the ethanol solution (uncoupling). This was true for both treatments.

Determination of Ethanol in Blood

The blood was collected at 9 a.m. As elaborated above,

the animals drank only little in the morning hours. The ethanol concentration was below the limit of detection except in those rats which had drunk large amounts of ethanol, e.g., those treated with 72.1 nmol/hr THN (0.09–0.18 g/l).

Stability of β-Carbolines During the Infusion Period

Minipumps containing the respective β-carboline were implanted under the skin of rats for 7 days. Thereafter, the concentration was measured by HPLC with UV-detection (254 nm) and compared with the concentration before the operation. The decomposition was 5.5% for THN, 7.4% for harman and 11.3% for harmalan. The differing stability can be explained by the ease of oxidation of the BC's. Neither the formation of harman from harmalan nor norharman from THN could be detected. The oxidation products were not identified.

DISCUSSION

In the early 70's Virginia Davis suggested that alkaloids may be formed in mammals and men which are directly or indirectly responsible for the pathogenesis of alcoholism. According to this hypothesis, acetaldehyde reacts with catecholamines yielding tetrahydroisoquinolines (TIQ's) and it reacts with indoleamines yielding β-carbolines (BC's). Furthermore, acetaldehyde might inhibit aldehydedehydrogenase competitively inducing an increase of the concentration of aldehydic metabolites (products of the monoamine-oxidase reaction) of catecholamines. The aldehydes subsequently form more complex alkaloids with the non-metabolized catecholamines namely tetrahydropapaverolines (THP's) [5,7].

Infusion of such alkaloids like salsolinol and THP into the brain ventricle led to an increased voluntary intake of ethanol in rats and monkeys [16,18]. Comparison of the effectiveness of salsolinol with that of THN revealed an approximately 100 fold higher potency of the BC. Since the experiments were performed with synthetic substances, it is important to note that THN occurs in rat brain and other tissues under physiological conditions [9]. Several other BC's were detected in human and rat tissue as well [10]. A load with ethanol induced an increase of the BC harman. However, the dose required was high (5 g/kg PO) [22].

In animals treated with 72.1 nmol/hr THN, which drank at most an average of 3.5 g ethanol/day, no increased levels of harman were found in the brain. This was to be expected from previous studies [22] assuming THN does not affect the synthesis or the metabolism of harman. The findings further demonstrate that at least two β-carbolines, namely THN and harman, are able to increase ethanol consumption in a free choice trial. Harman was about three times more potent than THN. Thus, harman is the most potent compound known so far to induce a preference for ethanol in rats. Furthermore, at least two mechanisms induce ethanol drinking without apparent interdependence. It is noteworthy that THN, a BC probably not derived from acetaldehyde, can induce ethanol preference in rats.

Several studies deal with the possible relationship of TIQ's with opioid mechanisms. Repeated intraventricular infusions of THP induced increased voluntary drinking of ethanol. This effect was attenuated by naloxone and naltrexone, as well as morphine [6,17]. The effect depends on the regimen of morphine administration in that only repeated injections attenuate the THP-induced increase of ethanol consumption. In another experimental set up it was observed that rats did not generalize the effect of the opioid fentanyl to either ethanol, THP, salsolinol or 3-carboxysalsolinol [25]. These data did not support a biochemical link between ethanol and opiates. Thus, a possible link of opioid mechanisms and TIQ's is far from being established. The findings of this study would support the view of a specific action of harman on voluntary ethanol intake since neither the intake of the opioid etonitazone nor that of clomethiazole and midazolam was increased by the BC. However, it should be noted that the effect of harman on tolerance mechanisms cannot be assessed by the experimental conditions utilized. The findings presented in Fig. 3 would indicate the increased consumption of the opioid was produced by harman (provided harman induces tolerance to etonitazene).

The specificity of the effect of harman for ethanol has also been supported by behavioral studies. Harman suppresses the physiological rhythm of water and ethanol intake in favour of shorter rhythms with increasing intervals. Similar effects were elicited by ethanol, whereby the effects became evident with increasing doses. However, further studies are required to substantiate the hypothesis that harman and ethanol have a similar impact on fluid intake.

ACKNOWLEDGEMENTS

The authors thank Dr. J. Wolffgramm for advice and for performing statistical calculations, and Mrs. Regina Hill for excellent technical assistance. The study was supported by the 'Deutsche Forschungsgemeinschaft.'

REFERENCES

1. Aalto, J. and K. Kiianmaa. Role of brain monoaminergic systems in the increased ethanol drinking caused by REM-sleep deprivation. *Alcohol Alcohol* **21:** A30, 1986.
2. Allen, J. R. F., O. Beck, S. Borg and R. Skroeder. Analysis of 1-methyl-1,2,3,4-tetrahydro-β-carboline in human urine and cerebrospinal fluid by gas-chromatography-mass spectrometry. *Eur J Mass Spectrom Biochem Med Environ Res* **1:** 171–177, 1980.
3. Airaksinen, M. M., M. Mähönen, L. Tuomisto, P. Peura and C. J. P. Eriksson. Tetrahydro-β-carbolines: Effect on alcohol intake in rats. *Pharmacol Biochem Behav* **18:** Suppl 1, 525–529, 1983.
4. Bond, N. W. and E. L. DiGiusto. Effects of prenatal alcohol consumption on open-field behaviour and alcohol preference in rats. *Psychopharmacologia* **46:** 163–165, 1976.
5. Cohen, G. and M. Collins. Alkaloids from catecholamines in adrenal tissue: possible role in alcoholism. *Science* **167:** 1749–1751, 1970.
6. Critcher, E. C., C. I. Lin, J. Patel and R. D. Myers. Attenuation of alcohol drinking in tetrahydroisoquinoline-treated rats by morphine and naltrexone. *Pharmacol Biochem Behav* **18:** 225–229, 1983.
7. Davis, V. E. and M. J. Walsh. Alcohol, amines, and alkaloids: a possible biochemical basis for alcohol addiction. *Science* **167:** 1005–1007, 1970.
8. Hilakivi, L. A. and J. D. Sinclair. Effect of neonatal clomipramine treatment on adult alcohol drinking in the AA and ANA rat lines. *Pharmacol Biochem Behav* **24:** 1451–1455, 1986.

9. Honecker, H. and H. Rommelspacher. Tetrahydronorharman (tetrahydro-β-carboline), a physiologically occurring compound of indole metabolism. *Naunyn Schmiedebergs Arch Pharmacol* **305:** 135–141, 1978.
10. Johnson, J. V., R. A. Yost, O. Beck and K. F. Faull. The use of tandem mass spectrometry for the identification and quantitation of tryptolines (tetrahydro-β-carbolines) in tissue extracts. *Prog Clin Biol Res* **183:** 161–178, 1985.
11. Jones, B., R. Goldstine, M. Gurley and E. Reyes. Appetite for alcohol: Influence of genetics and early experience. *Neurobehav Toxicol Teratol* **7:** 125–127, 1985.
12. Kraemer, G. W. and W. T. McKinney. Social separation increases alcohol consumption in rhesus monkey. *Psychopharmacology (Berlin)* **86:** 182–189, 1985.
13. Lange, F. H. *Korrelationselektronik*. 2nd edition. Berlin: VEB Verlag Technik, 1962.
14. Myers, R. D. and R. B. Holman. A procedure for eliminating position habit in preference-aversion tests for ethanol and other fluids. *Psychol Sci* **6:** 235–236, 1966.
15. Myers, R. D. and C. L. Melchior. Differential actions on voluntary alcohol intake of tetrahydroisoquinolines or a β-carboline infused chronically in the ventricle of the rat. *Pharmacol Biochem Behav* **7:** 381–392, 1977.
16. Myers, R. D. and C. L. Melchior. Alcohol drinking: abnormal intake caused by tetrahydropapaveroline in brain. *Science* **196:** 554–556, 1977.
17. Myers, R. D. and E. C. Critcher. Naloxone alters alcohol drinking induced in the rat by tetrahydropapaveroline (THP) infused ICV. *Pharmacol Biochem Behav* **16:** 827–836, 1982.
18. Myers, R. D., M. L. McCaleb and W. D. Ruwe. Alcohol drinking induced in monkey by tetrahydropapaveroline (THP) infused into the cerebral ventricle. *Pharmacol Biochem Behav* **16:** 995–1000, 1982.
19. Peschel, M. and H. Otto. *Statistische Methoden in der Regeltechnik-Korrelations- und Spektralanalyse. Reihe Automatisierungstechnik*. Band 106. Berlin: VEB Verlag Technik, 1970.
20. Potthoff, A. D. and G. Ellison. Low level continuous amphetamine administration selectively increases alcohol consumption. *Psychopharmacology (Berlin)* **77:** 242–245, 1982.
21. Potthoff, A. D., G. Ellison and L. Nelson. Ethanol intake increases during continuous administration of amphetamine and nicotine, but not several other drugs. *Pharmacol Biochem Behav* **18:** 489–493, 1983.
22. Rommelspacher, H., H. Damm, S. Strauß and G. Schmidt. Ethanol induces an increase of harman in the brain and urine of rats. *Naunyn Schmiedebergs Arch Pharmacol* **327:** 107–113, 1984.
23. Rommelspacher, H., H. Damm, L. Schmidt and G. Schmidt. Increased excretion of harman by alcoholics depends on events of their life history and the state of the liver. *Psychopharmacology (Berlin)* **87:** 64–68, 1985.
24. Schlitt, H. *Stochastische Vorgänge in linearen und nichtlinearen Regelkreisen*. Berlin: VEB Verlag Technik, 1968.
25. Shearman, G. T. and A. Herz. Ethanol and tetrahydroisoquinoline alkaloids do not produce narcotic discriminative stimulus effects. *Psychopharmacology (Berlin)* **81:** 224–227, 1983.
26. Sinclair, J. D. and R. J. Senter. Increased preference for ethanol in rats following alcohol deprivation. *Psychosom Sci* **8:** 11–12, 1967.
27. Tuomisto, L., M. M. Airaksinen, P. Peura and C. J. P. Eriksson. Alcohol drinking in the rat: Increases following intracerebroventricular treatment with tetrahydro-β-carbolines. *Pharmacol Biochem Behav* **17:** 831–836, 1982.
28. Weber, E. *Grundriß der biologischen Statistik*. Stuttgart: Gustav Fischer Verlag, 1967.
29. Zwiener, U. *Pathophysiologie neurovegetativer Regelungen und Rhythmen*. Jena: Gustav Fischer Verlag, 1976.