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Acute Alcoholic Intoxication and Naloxone. Effects on Visual Evoked Potential

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Experimental assays analyzing visual evoked potential (VEP) changes during an acute alcoholic intoxication were carried out in two groups of cats: One with continuous ethanol (0.06 g/kg · min) i.v. perfusion. Another one with a naloxone (400 μg/kg) i.v. injection 10 min before ethylic perfusion. Naloxone potentiates alcohol effects on VEP parameters, and on the appearance of isoelectric postpotential and flat VEP.

Key words: Ethylic intoxication, Naloxone, Visual evoked potential, Cat.

Within the last decade various studies (2, 4, 5, 8) have suggested that the central action of endogenous opiates and ethanol may have common points in their mechanisms.

The selective depletion of brain calcium induced by the administration of morphine, ethanol or the isoquinoline salsolinol has been shown to be inhibited by the administration of the opiate antagonist naloxone (9, 11, 12).

BLUM *et al.* (1) observed that naloxone prevented dependence and withdrawal syndromes in mice exposed to ethanol and suggested that tetrahydroisoquinolines (TIQs), compounds formed from the condensation of catecholamines and the ethanol metabolite, acetaldehyde, link ethyl alcohol and opiates and may explain their common effects in the brain.

These findings along with the identification of the endogenous opiates (15) have led to the speculation that some of the central effects of ethanol could be mediated through opiates, either by the formation of isoquinolines or by the release of endogenous opiates, and that naloxone, therefore, could antagonise these effects.

From the time when the first case-re-

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port of alcoholic coma reversal after the administration of naloxone appeared (14), a succession of communications with disparate data has been published (8). The results obtained in human volunteers and experimental animals are also contradictory, and lead to the conclusion that the possible role of endogenous opiates in acute alcoholic intoxication is still an object of investigation.

In order to evaluate the effect of ethanol and naloxone on the central nervous system the visual evoked potential (VEP) was studied systematically, by analyzing its quantitative and qualitative changes, during the acute alcoholic intoxication, in the hypotesis that naloxone administered before ethanol would block opiate receptor sites, neutralising thus the ethanol effects that depend on opiate or isoquinoline levels.

Materials and Methods

The study was carried out on 10 male adult cats, weighing between 3 and 4 kg (3.55 ± 0.33 SD), with chronic electrodes Ag/AgCl implanted over the dura on the right primary visual area 17, by stereotaxis method at coordinates 21 mm from the bregma and 3 mm from the midline (P4). As electrical reference a frontal electrode was used. Animals were fastened in a specially made muzzle apparatus and introduced into a sound-proof, dark Faraday chamber. Visual responses were evoked by a stroboscope (Estrobolume 1540) which generates, in xenon lamp, a flash lasting 12 μs, frequency of 0.5 Hz and an intensity of 4×10^6 candles. The stroboscope was placed outside the Faraday chamber separated, with a double glazed window, 30 cm from the head of the animal. The EEG signal was filtered (0.1-100 Hz) and amplified (× 100) by means of a Keithley 103A differential amplifier. VEPs lasting 500 ms were obtained throughout the experiment from the average of 200 responses using an Intertechnique Didac 800 averaging computer and the analysis of the VEP obtained was carried out by means of an Intertechnique SA-40 wave analyzer. If this way of registering the VEP, is taken into account, the occasional influence of interfering factors could be considered negligible.

Alcohol was administered by sustained i.v. perfusion, with a physiological 5 % glucose solution, at a dose of 0.06 g/kg · min. Experimental assays were carried out in two groups of cats with a systematically analyzed VEP: One with a continuous ethanol i.v. perfusion; and other with a previous i.v. injection of a single dose of naloxone (400 μg/kg b. w.) before 10 min i.v. ethanol perfusion, as in the first group.

The VEP parameters analyzed were the following: latency, defined as the time, in ms, to the end of the isoelectric line, or until a negative (N_1) or a positive (P_1) peak occurred after the visual stimulus. Amplitude index N_1-P_1, given by the difference in height from the baseline between N_1 and P_1, related to the difference N_1-P_1 of the control VEP. The time of the disappearance of VEP main complexes and the time of isoelectric postpotential and flat VEP appearance were also calculated.

For analysis venous blood samples were taken from 4.5 to 49.5, every 4.5 min and 58.5 minutes after initiating alcohol perfusion. Alcohol concentration was measured by spectrophotometry using the Trinder method (Boheringer).

For statistical procedures the means and standard deviations of each VEP parameter obtained in both groups were calculated. Once the statistical normality (D'Agostino's test) and the homoscedasticity (Bartlett's test) of the data were verified, comparisons were made using the Student's t test and the covariance test. The initial values of the different VEP parameters before they suffered any modification attributed to alcohol or naloxone, were considered as control.

Results

Alcohol perfusion produced a progressive increase in VEP latency (fig. 1), which was significant in relation to control values 36 minutes, approximately, after initiating alcohol perfusion ($p < 0.02$); N_1 latency (45 min, $p < 0.05$) and P_1 latency (27 min, $p < 0.05$). It also produced a progressive decrease of the amplitude index N_1-P_1, which was significant 18 minutes after initiating the perfusion ($p < 0.05$).

Alcohol perfusion, after 10 minutes of the naloxone administration, produced an earlier increase (18 min, $p < 0.01$) in VEP latency, N_1 latency and P_1 latency; and an earlier decrease in the amplitude index N_1-P_1 (9 min, $p < 0.01$).

Table I. *Time (min) from the beginning of ethanol perfusion to the disappearance of VEP main components (P_n, N_n), appearance of isoelectric post-potential (I. P.) and flat VEP.*
Alcohol: Continuous i.v. perfusion of ethanol (0.06 g/kg·min). Naloxone-Alcohol: Injection of a single dose of naloxone (400 μg/kg). After 10 minutes, continuous i.v. ethanol perfusion.

	Alcohol	Naloxone-Alcohol	p
P_3	33.41 ± 9.04	21.01 ± 10.77	< 0.02
N_3	46.39 ± 14.02	33.18 ± 11.05	< 0.02
P_2	59.01 ± 16.10	47.17 ± 12.01	< 0.01
N_2	77.40 ± 20.06	49.50 ± 12.30	< 0.01
P_1	97.15 ± 33.50	68.12 ± 16.31	< 0.02
N_1	99.24 ± 33.07	70.37 ± 19.35	< 0.02
I. P.	85.23 ± 28.18	58.50 ± 15.34	< 0.01
Flat VEP	103.00 ± 31.39	74.20 ± 21.44	< 0.02

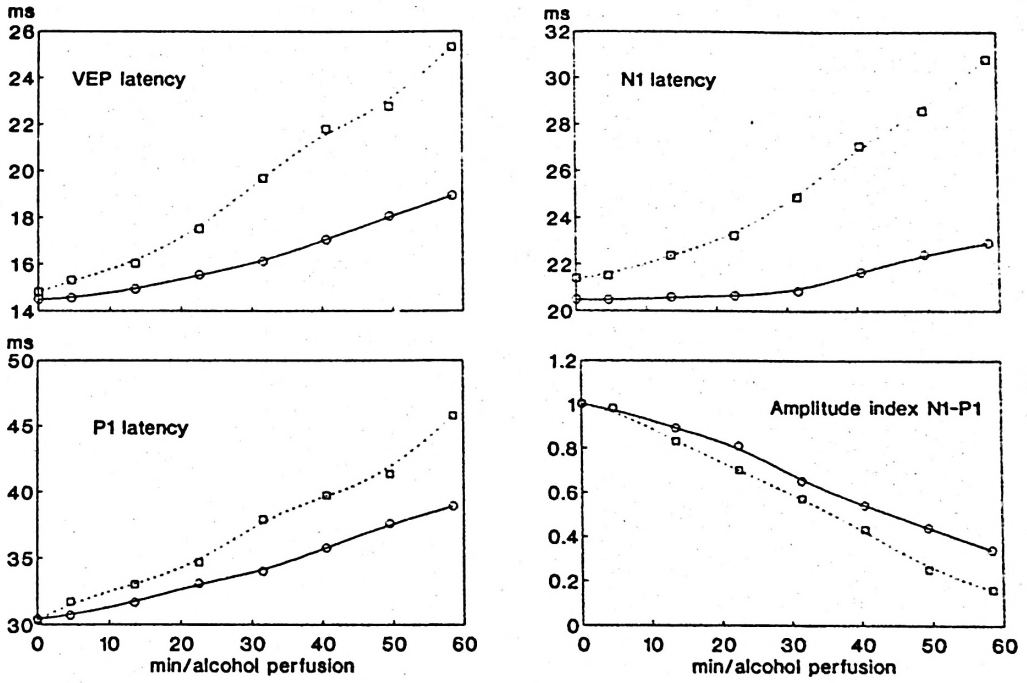

Fig. 1. *Naloxone effects on the ethylic intoxication. Changes of VEP latency, latency of N_1, latency of P_1 and amplitude index N_1-P_1.*
Continuous ethanol i.v. perfusion (0.06 g/kg · min) (—). Injection of a single dose of naloxone (400 μg/kg) 10 minutes before continuous ethanol i.v. perfusion (- - -).

Significant differences ($p < 0.001$) were found between both groups in latency of VEP, latency of N_1 and latency of P_1. No significant differences were found in the amplitude index N_1-P_1, due perhaps to data dispersion.

Moreover, when naloxone was injected before ethanol perfusion the disappearance of main VEP components occurred earlier. It also produced a significantly earlier appearance of isoelectric postpotential and flat VEP (table I).

No significant differences between both groups were obtained in alcohol levels 18, 36 and 54 min after initiating perfusion.

Discussion

The VEP modifications observed during the acute alcoholic intoxication are similar to those described by other authors (3, 5, 6).

Naloxone administered before ethanol perfusion, in our experimental conditions, potentiates alcohol effects in a process in which endogenous opiates are probably involved.

The administration of naloxone, is known to induce a transient up-regulation of μ-opiate receptor sites (10) which appears 5 minutes after its administration and persists for approximately 2 h during which naloxone is present *in vivo*. This change affects the number of receptor sites but not their affinity.

Active μ-opiate receptors may exist in equilibrium with an equal concentration of inactive silent receptors, althoug its mechanism is not known. Naloxone blocking of active receptors leads to the activation or unmasking of silent receptors. Thus when naloxone blocking decreases, since its function is limited in time, the result is an up-regulation in density.

For this reason, by the time alcohol perfusion commences the number of opiate receptor sites can be supposed to have increased in the group that receives naloxone before alcohol.

Furthermore, acute alcohol administration increases β-endorphin levels in certain brain areas (13), and its metabolite, acetaldehyde, is condensed with dopamine by the Pictet-Spengler TIQ generating reaction, that interacts at opiate receptor sites preferentially μ (7). Its concentration can be considered to increase in both groups similarly to the effect of ethanol perfusion. In the group that receives naloxone before alcohol, therefore, and owing to the increase in the number of opiate receptor sites, the toxic effect of alcohol would be equivalent to a greater dose than that really perfused.

Resumen

Se estudia el efecto de la naloxona (400 μg/kg) sobre el potencial evocado visual (PEV) durante la intoxicación alcohólica aguda, en dos grupos de gatos: Uno sólo con perfusión continua i.v. de alcohol (0,06 g/kg · min); otro con una inyección i.v. de naloxona (400 μg/kg) 10 min antes de iniciarse la perfusión i.v. de alcohol. La naloxona administrada antes del etanol aumenta el efecto tóxico del alcohol, lo cual influye sobre los componentes del PEV, y sobre la aparición de postpotencial isoeléctrico y del PEV plano.

Palabras clave: Intoxicación etílica, Naloxona, Potencial evocado visual, Gato.

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