Behavioural Brain Research 253 (2013) 128–138

Contents lists available at SciVerse ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

Research report

Alterations in affective behavior during the time course of alcohol hangover

Analía G. Karadayian[a,b,*], María J. Busso[b], Carlos Feleder[c], Rodolfo A. Cutrera[b]

[a] Institute of Biochemistry and Molecular Medicine, School of Pharmacy and Biochemistry, University of Buenos Aires, Buenos Aires, Argentina
[b] Laboratory of Neurobiology and Rhythms, Department of Physiology, School of Medicine, Universidad de Buenos Aires, Buenos Aires, Argentina
[c] Albany College of Pharmacy and Health Sciences, Departments of Pharmaceutical Sciences and Arts and Sciences, Albany, USA

HIGHLIGHTS

- Anxiety-like behavior and fear-related signs are evidenced during alcohol hangover.
- Signs of depression were found 14 h after hangover onset.
- Pain perception disabilities were detected at the beginning of hangover.
- Changes in affective behavior are evidenced for 14–16 h during hangover.

ARTICLE INFO

Article history:
Received 15 May 2013
Received in revised form 2 July 2013
Accepted 7 July 2013
Available online 11 July 2013

Keywords:
Alcohol hangover
Anxiety-like behavior
Depression
Anhedonia
Nociception

ABSTRACT

Alcohol hangover is a temporary state described as the unpleasant next-day effects after binge-like drinking. Hangover begins when ethanol is absent in plasma and is characterized by physical and psychological symptoms. Affective behavior is impaired during the acute phase of alcohol intoxication; however, no reports indicate if similar effects are observed during withdrawal. The aim of this work was to study the time-extension and possible fluctuations in affective behavior during a hangover episode. Male Swiss mice were injected i.p. either with saline (control group) or with ethanol (3.8 g/kg BW) (hangover group). Anxiety, fear-related behavior and despair phenotype were evaluated at a basal point (ZT0) and every 2 h up to 20 h after blood alcohol levels were close to zero (hangover onset). Also, anhedonia signs and pain perception disabilities were studied. Mice exhibited an increase in anxiety-like behavior during 4 h and 14 h after hangover onset when evaluated by the elevated-plus maze and open field test respectively ($p < 0.05$). Fear-related behavior was detected in hangover animals by the increase of freezing and decrease of line crossings and rearing frequency during 16 h after hangover onset ($p < 0.001$). Depression signs were found in hangover mice during 14 h ($p < 0.05$). Hangover mice showed a significant decrease in pain perception when tested by tail immersion test at the beginning of hangover ($p < 0.05$). Our findings demonstrate a time-extension between 14 and 16 h for hangover affective impairments. This study shows the long lasting effects of hangover over the phase of ethanol intoxication.

1. Introduction

Alcohol hangover (AH) is a temporary state described as the unpleasant next-day effects after a binge-like drinking [1]. In humans, AH begins when ethanol (EtOH) is absent in plasma and is characterized by a cluster of physical symptoms which include drowsiness, nausea, diarrhea, fatigue and tremors along with psychological signs that involve anxiety and guilt [2,3]. In addition, it was demonstrated that humans suffering from acute alcohol withdrawal (hangover) self-reported depression [4] or a general "decreased mood" [5]. Moreover, it is known that pain-related effects of ethanol such as headache and hyperalgesia are evidence at least at the onset of AH [6,7].

In the case of experimental animals, hypo-activity [8], fluctuations in body temperature, anxiety-like behavior [9] and reduced wheel running activity are observed during the hangover state [10,11]. In addition, we have previously demonstrated a reduction in motor performance at the beginning of AH in mice [12] establishing also an association between this motor impairments and changes in brain cortex energetic metabolism [13].

Abbreviations: %FEO, proportion of entrance into open arms; %TSO, proportion of time spent in open arms; BAC, blood alcohol concentration; AH, alcohol hangover; EMP, elevated-plus maze; i.p., intraperitoneally; TE, total number of entries; ZT, zeitgeber time.

* Corresponding author at: Institute of Biochemistry and Molecular Medicine, School of Pharmacy and Biochemistry, University of Buenos Aires, Junín 956, C1113AAD, Buenos Aires, Argentina. Tel.: +5411 4964 8244; fax: +5411 4964 8244
E-mail address: analiakaradayian@conicet.gov.ar (A.G. Karadayian).

0166-4328/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.bbr.2013.07.011

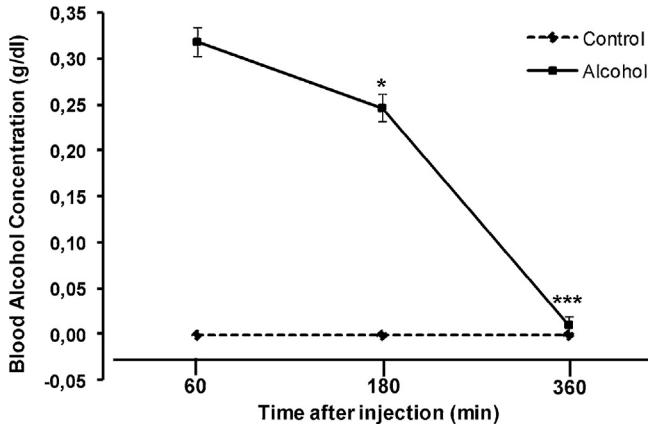

Fig. 1. Blood alcohol concentration after EtOH treatment.
Blood alcohol concentration (BAC) in controls (dotted line) and ethanol-treated (solid line) male Swiss mice was measured 60, 180 and 360 min after acute i.p. injections to determinate the onset of hangover. Values are expressed as mean ± SEM ($n = 15$ each group). $^{*}p < 0.05$ and $^{***}p < 0.001$, significantly different from BAC at 60 minutes. Independent samples *t*-test.

Particularly, it was shown that affective behavior is impaired during AH. In this sense, Gauvin et al. have shown that rats injected intraperitoneally (i.p.) with high doses of ethanol (3–4 g/kg) displayed a hangover-related anxiety behavior when tested 9 h after acute ethanol challenge [14,15]. Together with this, it has been recently demonstrated that, 18 h after acute EtOH administration (4 g/kg, i.p.), adult male rats present a reduced exploration into the elevated plus maze [16], and a significant social suppression [17] being both a response pattern that is consistent with an anxiogenic profile [18,19]. Similarly, Morse et al. reported a significant conditioned place aversion in the rat during hangover [20]. Likewise, Prediger et al. demonstrated a time-dependent development of anxiety-like behavior during AH in mice [21]. The evidence presented here together with the convergent findings from naturalistic methodology and the experimental investigations firmly suggest an increased anxiety-like behavior during alcohol hangover [22,23]. Nevertheless, the time-extension and possible fluctuations in affective behavior from the beginning to the end of a hangover episode were not explored.

In addition to hangover-related anxiety, it was demonstrated that rats exhibited brain reward deficits following acute exposure to ethanol [24]. This would indicate a negative affective component that, as mentioned above, is also observed in humans. Furthermore, Getachew et al. have verified that after chronic EtOH administration, rats exhibit an exaggerated immobility in the forced swim test, which reveal the "depressogenic" effects of alcohol [25]. Additionally, Walker et al. demonstrated an ethanol-induced depressive-like behavior mediated by alterations in the expression of brain neuropeptides [26]. Beyond anxiety and depression signs, it was established that hangover induced hyperalgesia and pain enhancement [11,27,28].

Although scientific evidences state that emotional behavior is affected during AH, it is unknown for how long these signs are presented and when individual physical and psychological symptoms are restored. Related to this, we have recently reported a time extension between 16 and 20 h for hangover motor and exploratory impairments in mice [29]; however, the different types and length of affective alterations together with the possible influence of light changes during AH throughout a day were not assessed. Taking all together into account, the aim of this work was to study the fluctuations in emotional behavior during AH in mice in order to achieve several goals: (1) to characterize the time course of anxiety and fear related-behavior; (2) to examine the possible manifestation of a despair phenotype; (3) to determinate the presence of anhedonia signs during a complete episode of alcohol induced hangover; (4) to evaluate possible pain perception disabilities at the beginning of the hangover and thus (5) to establish the time-extension of the possible affective behavior alterations.

2. Materials and methods

2.1. Animals

A total of 130 from five cohorts of male Swiss mice (*Mus musculus*) weighing 30–40 g were acquired from the School of Pharmacy and Biochemistry, Universidad de Buenos Aires, and housed in a soundproof room under conditions of controlled temperature (22 ± 2 °C) and humidity, with a 12-h light/dark cycle. Standard rat chow and tap water were provided ad libitum.

Animal handling, treatment and experimental procedures were reviewed in accordance with the guidelines of the National Institutes of Health (USA) and with Regulation 6344/96 of Argentina's National Drug, Food and Medical Technology Administration (ANMAT). Moreover, the present study had the legal ethical accreditation from Ethics Committee for Laboratory Animal Handling of the School of Medicine from Universidad de Buenos Aires where the protocol was performed. All efforts were made to minimize suffering and reduce the number of animals used.

2.2. Experimental procedure

Animals received intraperitoneal (i.p.) injections of 15% EtOH at a dose of 3.8 g/kg. Ethanol dose was previously applied in alcohol-induced hangover animal models [8,11]. Control mice received saline i.p. injections. In order to determine the animals' response to ethanol and the onset of hangover, fifteen mice from each group

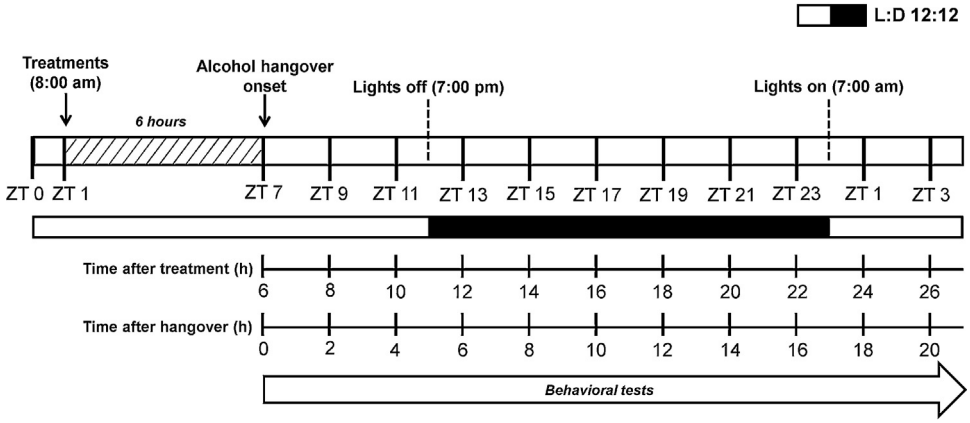

Fig. 2. Timeline and experiments.
Male mice received intraperitoneal treatment with saline or ethanol at a dose of 3.8 mg/kg or an equivalent of normal saline at 8:00 a.m. Behavioral tests were performed before and six hours after treatment when alcohol hangover began. ZT: Zeitgeber time; ZT12: 7:00 p.m.