



A deeper insight into how GABA-B receptor agonism via baclofen may affect alcohol seeking and consumption: lessons learned from a human laboratory investigation

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Abstract

Previous studies suggest that GABA-B receptor agonism may represent an effective pharmacological approach to treat addictive disorders. Baclofen is a selective GABA-B receptor agonist which has been investigated as a potential treatment for alcohol use disorder. However, research is needed to understand the biobehavioral mechanisms underlying baclofen's effect on alcohol use. In the present randomized, double-blind, placebo-controlled study, thirty-four alcohol-dependent individuals were randomized to receive baclofen (30 mg/d) or placebo for a week, and then participated in a laboratory experiment consisting of three procedures: alcohol cue-reactivity, priming, and self-administration. During the experiment, craving and other subjective responses to alcohol were assessed, and blood samples were collected for pharmacokinetic measurements. The effects of baclofen on the relationships between different alcohol-related laboratory parameters were investigated. Baclofen pharmacokinetic parameters and their correlations with behavioral measures were also examined. Results showed that baclofen disrupted the link between alcohol priming and self-administration, as indicated by significant interaction effects between drug condition (baclofen vs. placebo) and some of the priming variables (alcohol craving: $F_{3,9} = 6.03$, $p = 0.01$; alcohol sedation: $F_{3,6} = 7.16$, $p = 0.01$) on the total amount of alcohol self-administered. Considerable interindividual variability in baclofen pharmacokinetic parameters was observed. Maximum plasma concentrations of baclofen negatively correlated with cue-induced alcohol craving ($r = -0.57$, $p = 0.03$) and priming-induced ratings of 'like more' ($r = -0.59$, $p = 0.02$). In conclusion, baclofen may work by dissociating the link between an initial drink (priming) and subsequent alcohol consumption (self-administration). Considerable pharmacokinetic variability is an important factor to take into account when employing baclofen as a treatment for alcohol use disorder.

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Introduction

Baclofen, a lipophilic derivative of γ -aminobutyric acid (GABA) with high blood-brain barrier penetrance, acts as a selective agonist of the metabotropic GABA-B receptor (GABA_BR). GABA_BRs are widely expressed throughout the central nervous system (CNS) at both pre- and post-synaptic terminals. Activation of GABA_BRs potentiates second messengers that, through desensitizing calcium channels and sensitizing potassium channels, hyperpolarizes the neuron to reduce neuronal excitability and inhibit neurotransmitter release [1]. Therefore, GABA_BR agonism has been considered as a potential therapeutic approach for disorders involving aberrant neuronal signaling. Baclofen is approved by the Food and Drug Administration (FDA) as a treatment for muscle spasticity. In addition to this approved indication, previous studies

suggest that baclofen may represent a treatment for alcohol use disorder (AUD) [2].

In rodent models, baclofen administration reduces alcohol intake and suppresses acquisition, maintenance, and reinstatement of alcohol seeking behavior [3–11]. Administration of high doses of baclofen before alcohol presentation significantly increased alcohol consumption [12, 13], while operant conditioning studies show a decrease [14–18], or no change [19], in alcohol administration in rodents given moderate doses of baclofen. Preclinical data collectively suggest that alcohol seeking and consumption can be altered by baclofen administration, although the results vary across different studies, possibly due to differences in rodent strain, alcohol exposure, experimental paradigm, baclofen dosage, and other factors (for review, see ref. [20]). In several human studies, treatment with baclofen (30–80 mg/d) reduced alcohol craving and drinking, and prolonged abstinence in alcohol-dependent individuals [21–25], while other studies employing similar dosing found no effect of baclofen on alcohol-related outcomes [26–29]. Administering higher doses of baclofen has also produced mixed results: treatment with baclofen (up to 270 mg/d) improved abstinence rates in one study [30], but other studies found no difference between baclofen- (up to 180 mg/d) and placebo-treated individuals [31, 32]. The results of three recent meta-analyses underscore the heterogeneity in effect sizes, enrolled populations, adjunct behavioral therapies, and baclofen doses in the previous studies [33–35], while the overall conclusion is that baclofen seems superior to placebo as a treatment for AUD.

Additional research is needed to understand the biobehavioral mechanisms underlying baclofen's function. Human laboratory paradigms pose an informative approach to examine the neurobiological and behavioral effects of medications [36], and a few studies have utilized this methodology to investigate how baclofen works in relation to alcohol use. In one experiment, baclofen, compared with cyproheptadine (as an active control), increased subjective ratings of stimulation and sedation, with no significant effect on cue- or alcohol-induced craving [37]. Another study evaluating the effect of baclofen on alcohol cue-reactivity also found no significant effect on craving or attention to alcohol during the procedure [25]. Evans and Bisaga (2009) reported that a single dose of baclofen, administered prior to a fixed-dose alcoholic beverage, increased alcohol sedation, with no effect on other subjective responses (e.g., stimulation, drug liking) or alcohol-induced craving [38]. Finally, in a recent human laboratory study, we found that baclofen significantly increased subjective effects of alcohol (i.e., feeling high and intoxicated), and attenuated the positive association between post-priming breath alcohol concentration (BrAC) and the amount of alcohol self-administered [39]. These findings

suggest that baclofen's function, in relation to alcohol use, may be mediated through disrupting the link between an initial drink and subsequent alcohol consumption—a hypothesis requiring further investigation.

To investigate baclofen's biobehavioral mechanisms of action, the present study examined whether and how baclofen may alter the relationship between alcohol-related parameters assessed in a well-controlled and ecologically valid laboratory setting, using data from our study cited above [39]. In addition, while baclofen has been tested as a treatment for AUD for some years, most of the pharmacokinetic data come from studies in healthy individuals or those with neurological disorders. Considerable inter-individual variability was found in the few studies that have assessed baclofen pharmacokinetics in alcohol-dependent individuals [40, 41], highlighting the need for more research in this regard. Therefore, we also characterized pharmacokinetic parameters of baclofen and examined whether these parameters correlate with alcohol-related behavioral measures in the laboratory.

Materials and methods

Participants and setting

Potential candidates were first screened through a phone interview, followed by an in-person screening visit at the National Institute on Alcohol Abuse and Alcoholism (NIAAA). The study inclusion/exclusion criteria (Appendix S1) were assessed, and eligible individuals were enrolled after providing written informed consent. Non-treatment-seeking male and female individuals (21–65 years old) with a current diagnosis of alcohol dependence [Structured Clinical Interview for Diagnostic and Statistical Manual of Mental Disorders—Fourth Edition—Text Revision (DSM-IV-TR) Axis I Disorders (SCID)] and high trait anxiety [Spielberger State-Trait Anxiety Inventory (STAI)—trait version score ≥ 40] were enrolled in this study. The protocol was approved by the National Institutes of Health (NIH) Addictions Institutional Review Board (IRB), and was registered at ClinicalTrials.gov (NCT01751386). Study visits were conducted at the NIH Clinical Center in Bethesda, MD. Alcohol administration procedures were performed consistent with the NIAAA Council Guidelines on Alcohol Administration (<https://www.niaaa.nih.gov/Resources/ResearchResources/job22.htm>). For a detailed description of the parent study, see ref. [39].

Design and procedures

This was a between-subjects, randomized, double-blind, placebo-controlled human laboratory study. All procedures

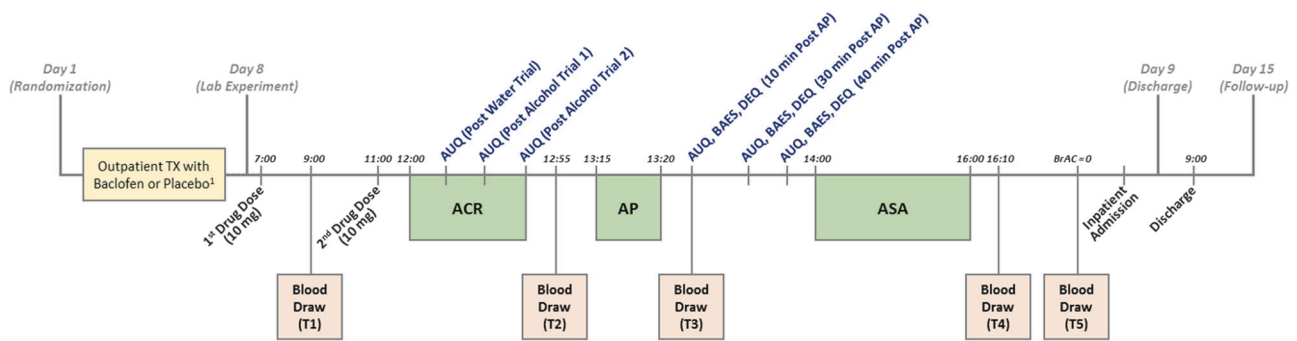


Fig. 1 Schematic outline of the study procedures and assessments. ¹Baclofen dosage was 15 mg/day (5 mg t.i.d.; titration phase) for 3 days, followed by 30 mg/day (10 mg t.i.d.; target dose) until the laboratory experiment. For additional details, see Table S1. *ACR*

alcohol cue-reactivity, *AP* alcohol priming, *ASA* alcohol self-administration, *AUQ* alcohol urge questionnaire, *BAES* biphasic alcohol effects scale, *BrAC* breath alcohol concentration, *DEQ* drug effects questionnaire, *TX* treatment

followed a predetermined timetable, as shown in Fig. 1. With an allocation ratio of 1:1, participants were randomized to receive oral racemic baclofen or placebo for one week in an outpatient setting. The initial dose of baclofen was 15 mg/day (5 mg t.i.d.; titration phase) for 3 days, followed by 30 mg/day (10 mg t.i.d.; target dose). After being on the target dose for at least 4 days, participants were brought back for an alcohol laboratory experiment (Table S1). The laboratory experiment was conducted during a full-day visit. Participants were asked to abstain from consuming alcohol 24 h prior to this visit and had to have a BrAC of 0 g/dL in the morning to proceed with the study procedures. They were also asked to take the first daily medication dose (10 mg) at home, before coming to the clinic. Standardized meals were served (400 kcal each, details in Appendix S2), and participants took the second daily medication dose (10 mg) at 11:00 am. This schedule was employed to allow participants receive the second dose of baclofen (or placebo) 1 h prior to the laboratory experiment, under direct observation of the research team. The laboratory experiment was conducted in a private bar-like room, included each participant's preferred alcoholic beverage, and consisted of three consecutive procedures: alcohol cue-reactivity (ACR), alcohol priming (AP), and alcohol self-administration (ASA), as outlined below and previously described [39, 42].

Alcohol cue-reactivity (ACR)

After an initial relaxation period, participants were exposed to a neutral beverage (water trial), followed by two consecutive alcohol trials. During each 3-minute trial, participants were asked to sniff the beverage upon hearing high tones and to stop sniffing upon hearing low tones. At the end of each trial, craving for alcohol was assessed via the alcohol urge questionnaire (AUQ). Cue-induced alcohol craving was calculated as follows: $\Delta AUQ = [\frac{1}{2} (AUQ$

score post alcohol trial 1 + AUQ score post alcohol trial 2) - AUQ score post alcohol water trial].

Alcohol priming (AP)

The alcohol content of this priming drink was calculated based on each participant's total body water, to raise the blood alcohol concentration to 0.03 g/dL. The priming drink had to be consumed within 5 min. Participants rated their craving for alcohol, using the AUQ, and other subjective responses to alcohol, using the biphasic alcohol effects scale (BAES) and a modified drug effects questionnaire (DEQ). Assessments were performed at three time-points: 10, 30, and 40 min after AP. The highest rating among the three time-points for each variable was considered as the peak score. For more details about the instruments used, see Appendix S3.

Alcohol self-administration (ASA)

During a 2-hour session, participants were given the opportunity to self-administer up to 8 mini-drink ad libitum. As an alternative reinforcer, \$3.00 per mini-drink was provided for the ones not consumed. Each mini-drink was designed to raise the blood alcohol concentration by 0.015 g/dL. A safety limit of BrAC \geq 0.12 g/dL was set, which would result in halting the session (this scenario did not happen during the present study). Total amount of alcohol self-administered was recorded as the primary outcome of this experiment, as reported in ref. [39].

At the end of the laboratory experiment, participants were monitored until their BrAC reached 0 g/dL; they stayed in an inpatient unit and were discharged the following morning. Baclofen (or matched placebo) was continued for three additional days for safety reasons (5 mg t.i.d., starting the evening after the laboratory session) (Table S1). The study was concluded with a brief follow-up

visit, approximately one week after the laboratory experiment.

Blood collection and processing

At the beginning of the laboratory experiment visit, a saline lock intravenous catheter was inserted into the antecubital fossa of the non-dominant arm for multiple blood draws. Blood samples were collected at the following time-points (Fig. 1 and Table S2): (T1) upon arrival, i.e., approximately two hours after the first baclofen dose; (T2) 10 min after the ACR procedure, i.e., approximately two hours after the second baclofen dose; (T3) 10 min after the priming drink was consumed; (T4) 10 min after the ASA session; and (T5) when participants' BrAC reached 0 g/dL. Prior to each visit, participants were asked to start fasting at midnight; therefore, the first blood draw (T1) was performed under fasting condition. A baseline (pre-drug) blood sample was also collected before dispensing baclofen or placebo (T0), which was not used for the present pharmacokinetic analyses. At each time-point, blood was collected into a lavender cap spray-coated K2EDTA tube (BD Vacutainer®) and centrifuged within 30 min post-collection (relative centrifugal force: $1700 \times g$, temperature: 4°C , centrifugation time: 15 min). The extracted plasma was pipetted into microtubes and stored in a -80°C freezer until analysis. After completing the study and breaking the blind, blood samples collected from the baclofen-treated participants were used for pharmacokinetic measurements.

Baclofen assay and quantification

A liquid chromatography-tandem mass spectrometry (LC-MS/MS) assay was developed at the Clinical Pharmacokinetics Research Laboratory of the University of Rhode Island to quantify baclofen in the plasma samples obtained during this study. Briefly, the LC-MS/MS system consisted of an ACQUITY UPLC system, coupled to an API 3200 triple quadrupole mass spectrometer, equipped with an electrospray ionization source to detect positive ions in multiple reaction monitoring (MRM) mode. The following MRM transitions (precursor \rightarrow product ion) were selected: m/z 214.2 \rightarrow 151.3 for baclofen and m/z 218.9 \rightarrow 155.3 for internal standard (IS). The method was validated according to the latest version of FDA guidance for industry on bioanalytical method validation [43]. In order to extract baclofen from plasma for quantification, one part of the plasma samples was mixed with three parts of IS solution in methanol (50 ng/mL). The mixtures were then centrifuged ($10,000 \times g$ at 4°C) for 10 min. Finally, 5 μL of the supernatant was withdrawn and injected into the LC-MS/MS system for quantification. For more details, see Appendix S4.

Statistical methods

The parent study showed that baclofen, compared to placebo, had no significant main effect on the total amount of alcohol self-administered [39]. In the present analyses, we examined whether baclofen modulated the relationship between ACR/AP (alcohol craving and other subjective responses to alcohol) and ASA (amount of alcohol consumed). Mixed-effects models were used, with drug condition (baclofen vs. placebo) and each ACR/AP variable as fixed effects, participants as the random effect, and grams of alcohol consumed during ASA as the dependent variable. A total number of nine variables were tested in individual models: Δ AUQ during ACR, peak AUQ during AP, peak BAES (two subscales) during AP, and peak DEQ (five questions) during AP. As recommended by Bender and Lange [44], multiplicity adjustment was not performed for this exploratory/hypothesis-generating analysis. A list of potential covariates (age, gender, race, number of SCID criteria, average drinks per drinking days based on alcohol Timeline Followback 90 days prior to the screening visit, and peak BrAC during AP) were tested in the initial run of each model; significant covariates were retained in the final model analysis. Within the baclofen group, non-compartmental analysis (Phoenix WinNonlin, Pharsight Corp., Mountain View, CA—Version 5.2.1) was used to characterize pharmacokinetic parameters of baclofen. As a marker of total drug exposure, the area under the plasma concentration-time curve (AUC) was determined and extrapolated to 16 h (the time of the first baclofen dose in the morning was considered as 0; see Table S2). Partial AUCs were also calculated, using the log-linear trapezoidal rule. Other estimated pharmacokinetic parameters were maximum plasma concentration (C_{\max}), the time of C_{\max} (T_{\max}), half-life ($t_{1/2}$), and apparent plasma clearance at steady state (CL_{ss}/F). Finally, Pearson's correlation coefficients evaluated bivariate associations between baclofen's pharmacokinetic parameters (AUC and C_{\max}) and the laboratory behavioral variables (alcohol drinking, craving, and subjective responses). All data were examined for normal distribution and statistical outliers were removed prior to analysis. IBM SPSS Statistics for Windows (IBM Corp., Armonk, NY—Version 20.0) was used for data analysis, and the significance level was set at $p < 0.05$ (two-tailed).

Results

Study sample

A sample size of $N = 34$ was planned based on previous human laboratory studies (Cohen's d effect size: ≥ 0.5 ,

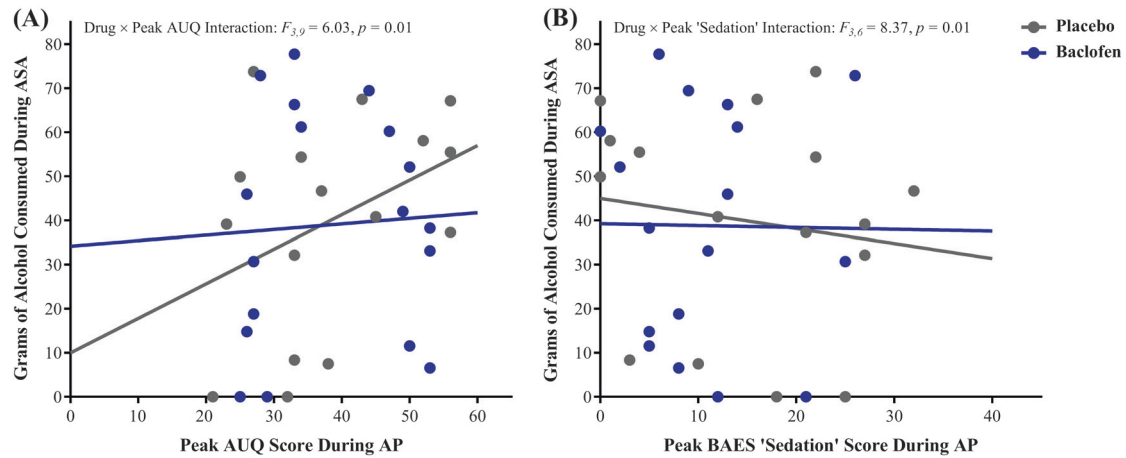


Fig. 2 The effect of baclofen vs. placebo on the relationship of (A) peak AUQ score and (B) peak BAES 'sedation' score during AP with the amount alcohol consumed during ASA. For other AP variables, see

Figure S2. AP alcohol priming, ASA alcohol self-administration, AUQ alcohol urge questionnaire, BAES biphasic alcohol effects scale

power: 80%, two-tailed significance level: 0.05) [37, 45]. To reach the target sample size, we randomized 39 individuals, and after 5 drop-outs, the final sample included 18 participants in the baclofen group and 16 participants in the placebo group (Figure S1). Baseline and demographic characteristics of the study sample are summarized in Table S3.

Exposure-response analyses

ACR-ASA

Baclofen did not affect the relationship between cue-induced alcohol craving during ACR and the amount of alcohol self-administered during ASA, as indicated by no significant drug \times Δ AUQ interaction effect on the grams of alcohol consumed ($F_{1,8} = 1.85, p = 0.21$).

AP-ASA

There was a significant drug \times peak AUQ interaction effect on the grams of alcohol consumed ($F_{3,9} = 6.03, p = 0.01$), where baclofen blunted the positive association between alcohol-induced craving during AP and the amount of alcohol self-administered during ASA (Fig. 2a). A significant drug \times peak BAES 'sedation' effect was also detected ($F_{3,6} = 8.37, p = 0.01$) (Fig. 2b). Other alcohol-induced subjective measures showed a similar pattern, but the interaction terms did not reach statistical significance (p 's ≥ 0.05) (Figure S2).

Pharmacokinetic parameters

Plasma concentrations of baclofen were measured in 17 individuals (14 males, 3 females); blood samples were not

Table 1 Pharmacokinetic parameters of baclofen estimated in individuals with alcohol dependence^a

Parameter	N	Mean	SD
AUC _{0→16} (h*ng/mL) ^b	17	1033.50	403.14
AUC _{0→4} (h*ng/mL)	17	253.17	165.29
AUC _{4→8} (h*ng/mL)	17	446.50	169.53
AUC _{8→12} (h*ng/mL)	15	357.62	148.86
AUC _{12→16} (h*ng/mL)	15	221.30	145.16
C _{max} (ng/mL) ^c	14	84.90	50.84
t _{1/2} (h)	11	4.42	0.98
CL _{ss} /F (mL/h)	17	60866.52	52123.17

AUC area under the plasma concentration-time curve, CL_{ss}/F apparent plasma clearance at steady state, C_{max} maximum plasma concentration, N number, SD standard deviation, T_{max} time of C_{max}, t_{1/2} half-life

^aBaclofen was administered in an outpatient setting (15 mg/day for 3 days, followed by 30 mg/day for at least 4 days). Blood samples for pharmacokinetic measurements were obtained on the day of the alcohol laboratory experiment, during which the first and the second daily medication doses (10 mg each) were respectively taken in the morning and one hour before the alcohol laboratory experiment started

^bFor AUC calculations, the time of the first baclofen dose in the morning was considered as 0 and extrapolated to 16 h (see Table S2)

^cT_{max} data for each participant are presented in Table S4 (range: 2–4 h, median: 2 h)

available for one participant in the baclofen group due to poor venous access. Pharmacokinetic parameters estimated for each participant are presented in Table S4, and the summary statistics are presented in Table 1.

Pharmacokinetic-behavior correlations

There was a significant negative correlation between baclofen C_{max} and cue-induced alcohol craving during ACR ($r = -0.57, p = 0.03$) (Fig. 3a). In addition, baclofen C_{max}

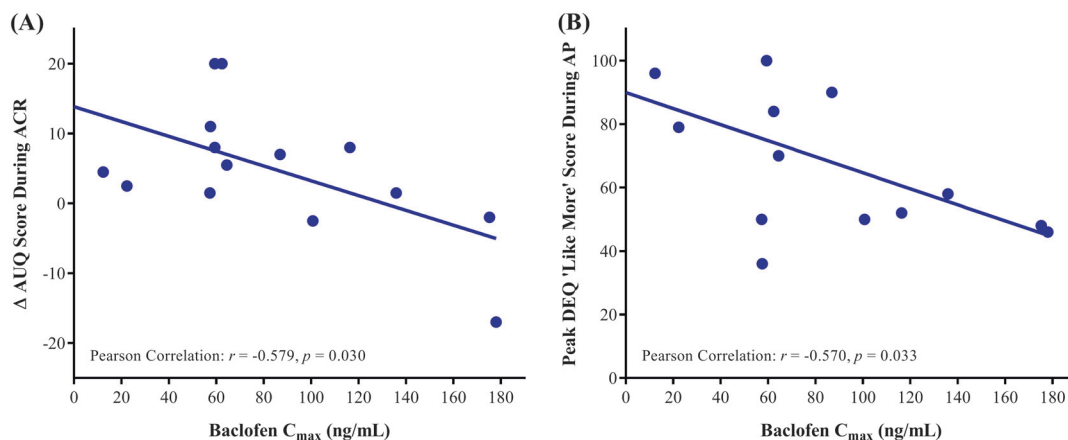


Fig. 3 Bivariate correlations between baclofen C_{max} and (A) Δ AUQ score during ACR and (B) peak DEQ 'like more' score during AP. ACR alcohol cue-reactivity, AP alcohol priming, AUQ alcohol urge

questionnaire, C_{max} maximum plasma concentration, DEQ drug effects questionnaire

negatively correlated with peak DEQ 'like more' during AP ($r = -0.57$, $p = 0.03$) (Fig. 3b). No other significant correlations were found between baclofen pharmacokinetic parameters and the laboratory behavioral assessments (p 's ≥ 0.05) (Table 2).

Discussion

We previously reported that baclofen, compared to placebo, did not significantly reduce alcohol craving or self-administration in this study. Baclofen, however, flattened the relationship between the peak BrAC during priming and subsequent alcohol self-administration, and amplified the effects of alcohol, e.g., feeling high and intoxicated [39]. The latter observation, i.e., increased subjective response to alcohol after baclofen administration, is consistent with findings of the two human laboratory studies conducted before [37, 38]. In the present set of analyses, we found that baclofen blunted the link between an initial alcohol priming and the amount of alcohol self-administration in a human laboratory setting. This pattern was observed for most of the AP variables, and statistically significant results were detected for alcohol-induced craving (assessed via AUQ) and sedation (assessed via BAES). To our knowledge, previous human laboratory studies did not report the effect of baclofen on the relationship between alcohol-related parameters, hence we cannot directly compare our findings with these studies. For this reason and given the exploratory/hypothesis-generating nature of this work, the results need to be confirmed in future studies. Nevertheless, the present data propose a biobehavioral mechanism through which baclofen acts, i.e., by attenuating the effects of an initial drink on subsequent alcohol consumption. This interpretation was further corroborated by the finding that

higher blood concentrations of baclofen (as indicated by baclofen C_{max}) were associated with lower ratings of 'like more' on DEQ after consuming the priming drink (verbatim of the DEQ item: *would you like more of what you received, right now?*).

According to several lines of basic and clinical research, administering a priming dose of alcohol stimulates operant responding for alcohol and provokes alcohol seeking and consummatory behaviors—a phenomenon known as the "alcohol priming effect". In rodent models, for example, priming injections of alcohol after a period of extinction leads to reinstatement of alcohol self-administration [46, 47]; a similar priming effect has also been observed for other drugs of abuse [48]. Interestingly, baclofen has been shown to block priming-induced reinstatement and escalation of drug use in numerous animal experiments [49–53]. These preclinical observations are consistent with the present human findings where baclofen was found to disrupt the link between priming-induced effects and alcohol self-administration. Beyond baclofen, the present work is an example of the importance of human laboratory experiments in studying how medications affect alcohol-related outcomes—an informative approach that may illuminate novel biobehavioral mechanisms of action compared to those classically seen with FDA-approved medications for AUD.

Evidence from human research further supports the notion that an initial low-to-moderate dose of alcohol increases subsequent alcohol consumption. A number of factors have been found to facilitate the aforementioned "alcohol priming effect", including (but not limited to) alcohol-induced craving and other subjective responses, interoceptive stimuli, attentional bias, alcohol expectancies, behavioral economic demand, and impaired control [54–59]. A human laboratory study conducted by

Table 2 Bivariate correlations between baclofen pharmacokinetic parameters and human laboratory behavioral assessments

Baclofen PK parameter	Laboratory assessments ^a									
	Δ AUC	Peak AUC	Peak BAES 'sedation'	Peak BAES 'stimulation'	Peak DEQ 'drug effects'	Peak DEQ 'feel the effects'	Peak DEQ 'like the effects'	Peak DEQ 'like more'	Peak DEQ 'feel high'	Peak DEQ 'feel intoxicated'
AUC ₀₋₁₆ (h*ng/mL) ^b	<i>r</i> -0.09	0.08	-0.10	-0.04	-0.13	0.15	0.17	-0.27	-0.40	0.23
	<i>p</i> 0.73	0.74	0.69	0.86	0.60	0.56	0.49	0.28	0.10	0.36
C _{max} (ng/mL)	<i>r</i> -0.57	-0.36	0.20	-0.25	-0.02	0.01	-0.57	-0.09	-0.16	0.05
	<i>p</i> 0.03	0.20	0.50	0.37	0.94	0.95	0.03	0.75	0.58	0.85

ACR alcohol cue-reactivity, AP alcohol priming, ASA alcohol self-administration, AUC area under the plasma concentration-time curve, AUC₀₋₁₆ alcohol self-administration, BAES alcohol effects scale, C_{max} maximum plasma concentration, DEQ drug effects questionnaire, PK pharmacokinetic

^aA AUC was assessed during ACR. Grams of alcohol consumed was assessed during ASA. Other variables were assessed during AP

^bThe time of the first baclofen dose in the morning was considered as 0 and extrapolated to 16 h (see Table S2)

Christiansen and colleagues (2017) showed that priming with a placebo drink (that is 'believed' to contain alcohol, but does not), compared to a control drink (that is known to contain no alcohol), significantly increased craving, subjective intoxication, and ad libitum beer consumption [60]. These findings suggest that anticipated effects of alcohol may play a more prominent role than its pharmacological effects per se in mediating the influence of a priming on alcohol-related outcomes. Consistent with this concept, the present study found that baclofen modulated the relationship between alcohol priming and self-administration without affecting alcohol pharmacokinetics, as indicated by no significant changes in BrAC (Figure S3).

Alcohol-induced craving and sedation are significant predictors of alcohol self-administration in human laboratory studies. Bujarski and colleagues (2018), for example, administered an initial dose of intravenous alcohol (alcohol challenge), followed by a progressive ratio self-administration, and found that craving and sedation during the alcohol challenge, respectively, predicted higher and lower alcohol self-administration [61]. In our study, baclofen, as compared to placebo, blunted the positive association between alcohol-induced craving (during AP) and self-administration. In addition, while baclofen did not affect the relationship between cue-induced craving (during ACR) and self-administration, higher blood concentrations of baclofen (as indicated by baclofen C_{max}) were associated with lower ratings of cue-induced craving. Craving is a prominent hallmark of addiction, and self-report measures of craving considerably correlate with objective measures of substance use, in both laboratory and naturalistic settings [54]. From a neuroscientific perspective, craving is a multidimensional symptom with diverse biobehavioral substrates, and targeting craving may represent an effective approach to reduce alcohol drinking and/or to prevent relapse [62-64].

In addition to craving, we also found that baclofen, as compared to placebo, flattened the negative association between alcohol-induced sedation (during AP) and self-administration. Experiencing sedative effects typically leads to lower alcohol consumption, and Wardell and colleagues (2015) showed that alcohol-induced craving mediates this link [65]. It has been well established that the level of sensitivity to alcohol effects plays a key role in shaping alcohol-related behaviors [66, 67]. Accordingly, subjective response to alcohol may represent a targetable biobehavioral domain in the development of medications for AUD [36, 68]. In the present study, we found that GABA_BR agonism via baclofen altered the relationship between alcohol-induced subjective effects during AP and alcohol consumption during ASA (Figure S2). Our results also suggest that baclofen may be more efficacious in reducing alcohol drinking in those patients who experience higher

craving and lower sedation after one drink (i.e., priming). Contrary to Evans and Bisaga (2009) findings [38], baclofen did not increase sedation in this study. As previously discussed in ref. [39], methodological differences between the two studies (e.g., differences in the enrolled population, alcohol dosage, baclofen dosage, and length of baclofen administration) may explain, at least in part, these dissimilar results. It has been shown that GABA_BRs regulate alcohol sensitivity at the cellular/molecular level [69–72]. Chronic alcohol exposure also leads to neuroadaptive changes in GABA_BRs [73, 74], which may further modulate the effects of baclofen in heavy-drinking alcohol-dependent individuals.

A causal role of GABA_BRs in alcohol reinforcement has been previously explained [75–77]. It is also known that alcohol enhances GABAergic neurotransmission and, among other sites of action, activates GABA_ARs, which leads to hyperpolarization of the neurons through facilitating chloride influx into the cell [78, 79]. Potentiation of GABA_BRs also hyperpolarizes neurons by increasing potassium and decreasing calcium permeability [1]. It is therefore plausible to hypothesize that GABA_BR agonism, in combination with alcohol, may pose an additive or synergistic effect on GABAergic neurons and decrease the amount of alcohol needed to have the same effect on GABAergic neurotransmission. While electrophysiological studies are required to directly test this hypothesis, our behavioral findings show that GABA_BR agonism via baclofen modulates the response to an initial drink, such that subsequent drinking is less reinforcing. Accordingly, it has been suggested that baclofen may act as a partial substitution therapy by increasing subjective response to alcohol and mimicking alcohol's effects [37–39, 80]. It appears that a certain level of alcohol drinking is required for baclofen to exert its optimal function – a notion supported by previous findings that baclofen is more effective in heavy than light drinkers [34, 81]. From a clinical perspective, it is crucial to identify subpopulations of patients who better respond to baclofen, e.g., heavy-drinking alcohol-dependent individuals. Interestingly, a recent study in baboons showed that baclofen suppressed alcohol self-administration when the treatment was initiated during ongoing alcohol access, but not during alcohol abstinence [82]. Baclofen may, therefore, serve as a harm reduction pharmacotherapy by helping heavy-drinking individuals reduce their alcohol consumption levels [83]. Along these lines, the possibility of interaction between baclofen and alcohol should be further investigated in future studies.

Consistent with two previous reports in alcohol-dependent individuals [40, 41], we found considerable interindividual variability in baclofen pharmacokinetic parameters. In addition to other factors, this high variability may be partially responsible for the heterogeneity of

response to baclofen and the inconsistency of previous reports. Our pharmacokinetic-behavior analyses provided a deeper insight into baclofen's effects on alcohol-related outcomes. For instance, baclofen was not superior to placebo in reducing cue-induced alcohol craving in our aggregate analyses [39], but here we found that higher blood concentrations of baclofen (C_{max}) were associated with lower cue-induced alcohol craving during the ACR. These results suggest that, while all participants received the same dose of baclofen, interindividual variability in exposure to baclofen (resulting from pharmacokinetics variability) may have washed out the signal in the aggregate sample. Future studies should investigate whether individually tailored dosing vs. fixed-dose treatment may be a better approach in the use of baclofen for AUD. In addition, the significant negative correlation between baclofen C_{max} and cue-induced craving suggests that doses higher than 30 mg/day may be needed to see an effect, particularly in AUD patients with no clinically significant liver impairment, as the sample enrolled in this study. 30 mg/day seems a sufficient dose of baclofen to reduce drinking in AUD patients with alcoholic liver disease (ALD) [22, 24], but higher doses may be needed in those without ALD. Nonetheless, additional pharmacokinetic studies with baclofen are required in the alcohol field, and such studies should be conducted in patients with and without ALD.

The results of this study should be viewed in the context of its limitations. The sample size was relatively small. A limited number of females were enrolled and, therefore, possible sex differences in behavioral outcomes and/or baclofen pharmacokinetics could not be studied. Participants could not self-administer more than eight mini-drinks, as the number was limited due to safety reasons. Only one dose of baclofen was used and, consequently, the effect of higher doses of baclofen on the reported outcomes remains unknown. Previous data suggest that the strength of alcohol priming effect varies by the dose of alcohol, as well as the time between priming and self-administration [84]. Here, both the dosage and the timing were fixed, thus, the findings may not be generalizable to other experimental scenarios. Also, an alcoholic beverage was always used as the priming, and we did not run comparison sessions with placebo and/or control drinks; therefore, we cannot fully disentangle the pharmacological vs. anticipated aspects of the priming effect. Finally, the finding of an effect of baclofen on the relationship between an initial drink and subsequent alcohol consumption was not an a priori hypothesis, and the present results should be further examined in prospective confirmatory studies.

In conclusion, our results suggest that GABA_BR agonism via baclofen administration uncouples the link between an initial drink and subsequent alcohol consumption—a potential biobehavioral mechanism of baclofen's effects on

alcohol-related outcomes. In addition, considerable variability in pharmacokinetic measures is an important factor to take into account when employing baclofen as a treatment for AUD.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

- Bettler B, Tiao JY. Molecular diversity, trafficking and subcellular localization of GABAB receptors. *Pharmacol Ther.* 2006;110:533–43.
- Agabio R, Sinclair JM, Addolorato G, Aubin HJ, Beraha EM, Caputo F et al. Baclofen for the treatment of alcohol use disorder: the Cagliari statement. *Lancet Psychiatry.* [In Press].
- Daoust M, Saligaut C, Lhuintre JP, Moore N, Flipo JL, Boismare F. GABA transmission, but not benzodiazepine receptor stimulation, modulates ethanol intake by rats. *Alcohol.* 1987;4:469–72.
- Colombo G, Vacca G, Serra S, Brunetti G, Carai MA, Gessa GL. Baclofen suppresses motivation to consume alcohol in rats. *Psychopharmacol (Berl).* 2003;167:221–4.
- Colombo G, Addolorato G, Agabio R, Carai MA, Pibiri F, Serra S, et al. Role of GABA(B) receptor in alcohol dependence: reducing effect of baclofen on alcohol intake and alcohol motivational properties in rats and amelioration of alcohol withdrawal syndrome and alcohol craving in human alcoholics. *Neurotox Res.* 2004;6:403–14.
- Williams KL, Nickel MM, Bielak JT. Baclofen blocks yohimbine-induced increases in ethanol-reinforced responding in rats. *Pharmacol Biochem Behav.* 2016;144:20–25.
- Maccioni P, Bienkowski P, Carai MA, Gessa GL, Colombo G. Baclofen attenuates cue-induced reinstatement of alcohol-seeking behavior in Sardinian alcohol-preferring (sP) rats. *Drug Alcohol Depend.* 2008;95:284–7.
- Bianchi PC, Carneiro de Oliveira PE, Palombo P, Leao RM, Cogo-Moreira H, Planeta CDS, et al. Functional inactivation of the orbitofrontal cortex disrupts context-induced reinstatement of alcohol seeking in rats. *Drug Alcohol Depend.* 2018;186:102–12.
- Colombo G, Serra S, Brunetti G, Atzori G, Pani M, Vacca G, et al. The GABA(B) receptor agonists baclofen and CGP 44532 prevent acquisition of alcohol drinking behaviour in alcohol-preferring rats. *Alcohol Alcohol.* 2002;37:499–503.
- Maccioni P, Lorrai I, Contini A, Leite-Morris K, Colombo G. Microinjection of baclofen and CGP7930 into the ventral tegmental area suppresses alcohol self-administration in alcohol-preferring rats. *Neuropharmacology.* 2018;136(Pt A):146–58.
- Quintanilla ME, Perez E, Tampier L. Baclofen reduces ethanol intake in high-alcohol-drinking University of Chile bibulous rats. *Addict Biol.* 2008;13:326–36.
- Smith BR, Boyle AE, Amit Z. The effects of the GABA(B) agonist baclofen on the temporal and structural characteristics of ethanol intake. *Alcohol.* 1999;17:231–40.
- Smith BR, Robidoux J, Amit Z. GABAergic involvement in the acquisition of voluntary ethanol intake in laboratory rats. *Alcohol Alcohol.* 1992;27:227–31.
- Janak PH, Michael Gill T. Comparison of the effects of allo-pregnanolone with direct GABAergic agonists on ethanol self-administration with and without concurrently available sucrose. *Alcohol.* 2003;30:1–7.
- Anstrom KK, Cromwell HC, Markowski T, Woodward DJ. Effect of baclofen on alcohol and sucrose self-administration in rats. *Alcohol Clin Exp Res.* 2003;27:900–8.
- Stromberg MF. The effect of baclofen alone and in combination with naltrexone on ethanol consumption in the rat. *Pharmacol Biochem Behav.* 2004;78:743–50.
- Walker BM, Koob GF. The gamma-aminobutyric acid-B receptor agonist baclofen attenuates responding for ethanol in ethanol-dependent rats. *Alcohol Clin Exp Res.* 2007;31:11–18.
- Lorrai I, Maccioni P, Gessa GL, Colombo G. R (+)-Baclofen, but Not S(-)-Baclofen, Alters Alcohol Self-Administration in Alcohol-Preferring Rats. *Front Psychiatry.* 2016;7:68.
- Czachowski CL, Legg BH, Stansfield KH. Ethanol and sucrose seeking and consumption following repeated administration of the GABA(B) agonist baclofen in rats. *Alcohol Clin Exp Res.* 2006;30:812–8.
- Agabio R, Leite-Morris KA, Addolorato G, Colombo G Targeting the GABAB Receptor for the Treatment of Alcohol Use Disorder. In: Colombo G (ed). *GABAB Receptor.* Springer International Publishing: Cham, 2016, pp 287-307.
- Addolorato G, Caputo F, Capristo E, Domenicali M, Bernardi M, Janiri L, et al. Baclofen efficacy in reducing alcohol craving and intake: a preliminary double-blind randomized controlled study. *Alcohol Alcohol.* 2002;37:504–8.
- Addolorato G, Leggio L, Ferrulli A, Cardone S, Vonghia L, Mirijello A, et al. Effectiveness and safety of baclofen for maintenance of alcohol abstinence in alcohol-dependent patients with liver cirrhosis: randomised, double-blind controlled study. *Lancet.* 2007;370:1915–22.
- Addolorato G, Leggio L, Ferrulli A, Cardone S, Bedogni G, Caputo F, et al. Dose-response effect of baclofen in reducing daily alcohol intake in alcohol dependence: secondary analysis of a randomized, double-blind, placebo-controlled trial. *Alcohol Alcohol.* 2011;46:312–7.
- Morley KC, Baillie A, Fraser I, Furneaux-Bate A, Dore G, Roberts M et al. Baclofen in the treatment of alcohol dependence with or without liver disease: multisite, randomised, double-blind, placebo-controlled trial. *Br J Psychiatry* 2018;212:362–9.
- Leggio L, Zywiak WH, Edwards SM, Tidey JW, Swift RM, Kenna GA. A preliminary double-blind, placebo-controlled randomized study of baclofen effects in alcoholic smokers. *Psychopharmacol (Berl).* 2015;232:233–43.
- Garbutt JC, Kampov-Polevoy AB, Gallop R, Kalka-Juhl L, Flannery BA. Efficacy and safety of baclofen for alcohol dependence: a randomized, double-blind, placebo-controlled trial. *Alcohol Clin Exp Res.* 2010;34:1849–57.
- Hauser P, Fuller B, Ho SB, Thuras P, Kern S, Dieperink E. The safety and efficacy of baclofen to reduce alcohol use in veterans

- with chronic hepatitis C: a randomized controlled trial. *Addiction*. 2017;112:1173–83.
28. Morley KC, Baillie A, Leung S, Addolorato G, Leggio L, Haber PS. Baclofen for the treatment of alcohol dependence and possible role of comorbid anxiety. *Alcohol Alcohol*. 2014;49:654–60.
 29. Ponizovsky AM, Rosca P, Aronovich E, Weizman A, Grinshpoon A. Baclofen as add-on to standard psychosocial treatment for alcohol dependence: a randomized, double-blind, placebo-controlled trial with 1 year follow-up. *J Subst Abus Treat*. 2015;52:24–30.
 30. Muller CA, Geisel O, Pelz P, Higl V, Kruger J, Stickel A, et al. High-dose baclofen for the treatment of alcohol dependence (BACLAD study): a randomized, placebo-controlled trial. *Eur Neuropsychopharmacol*. 2015;25:1167–77.
 31. Beraha EM, Saleminck E, Goudriaan AE, Bakker A, de Jong D, Smits N, et al. Efficacy and safety of high-dose baclofen for the treatment of alcohol dependence: A multicentre, randomised, double-blind controlled trial. *Eur Neuropsychopharmacol*. 2016;26:1950–9.
 32. Reynaud M, Aubin HJ, Trinquet F, Zakine B, Dano C, Dematteis M, et al. A Randomized, Placebo-Controlled Study of High-Dose Baclofen in Alcohol-Dependent Patients-The ALPADIR Study. *Alcohol Alcohol*. 2017;52:439–46.
 33. Bschor T, Henssler J, Muller M, Baethge C. Baclofen for alcohol use disorder—a systematic meta-analysis. *Acta Psychiatr Scand*. 2018;138:232–42.
 34. Pierce M, Sutherland A, Beraha EM, Morley K, van den Brink W. Efficacy, tolerability, and safety of low-dose and high-dose baclofen in the treatment of alcohol dependence: A systematic review and meta-analysis. *Eur Neuropsychopharmacol*. 2018;28:795–806.
 35. Rose AK, Jones A. Baclofen: its effectiveness in reducing harmful drinking, craving, and negative mood. A meta-analysis. *Addiction*. 2018;113:1396–406.
 36. Yardley MM, Ray LA. Medications development for the treatment of alcohol use disorder: insights into the predictive value of animal and human laboratory models. *Addict Biol*. 2017;22:581–615.
 37. Leggio L, Zywiak WH, McGeary JE, Edwards S, Fricchione SR, Shoaff JR, et al. A human laboratory pilot study with baclofen in alcoholic individuals. *Pharmacol Biochem Behav*. 2013;103:784–91.
 38. Evans SM, Bisaga A. Acute interaction of baclofen in combination with alcohol in heavy social drinkers. *Alcohol Clin Exp Res*. 2009;33:19–30.
 39. Farokhnia M, Schwandt ML, Lee MR, Bollinger JW, Farinelli LA, Amodio JP, et al. Biobehavioral effects of baclofen in anxious alcohol-dependent individuals: a randomized, double-blind, placebo-controlled, laboratory study. *Transl Psychiatry*. 2017;7:e1108.
 40. Marsot A, Imbert B, Alvarez JC, Grassin-Delye S, Jaquet I, Lancon C, et al. High variability in the exposure of baclofen in alcohol-dependent patients. *Alcohol Clin Exp Res*. 2014;38:316–21.
 41. Chevillard L, Sabo N, Tod M, Labat L, Chasport C, Chevalyre C, et al. Population pharmacokinetics of oral baclofen at steady-state in alcoholic-dependent adult patients. *Fundam Clin Pharmacol*. 2018;32:239–48.
 42. Farokhnia M, Sheskie MB, Lee MR, Le AN, Singley E, Bouhlal S, et al. Neuroendocrine response to GABA-B receptor agonism in alcohol-dependent individuals: Results from a combined outpatient and human laboratory experiment. *Neuropharmacology*. 2018;137:230–9.
 43. Guidance for Industry: Bioanalytical Method Validation *US Department of Health and Human Services (HHS), Food and Drug Administration (FDA), Center for Drug Evaluation and Research (CDER), Center for Veterinary Medicine (CVM)* 2013.
 44. Bender R, Lange S. Adjusting for multiple testing—when and how? *J Clin Epidemiol*. 2001;54:343–9.
 45. O'Malley SS, Krishnan-Sarin S, Farren C, Sinha R, Kreek MJ. Naltrexone decreases craving and alcohol self-administration in alcohol-dependent subjects and activates the hypothalamo-pituitary-adrenocortical axis. *Psychopharmacol (Berl)*. 2002;160:19–29.
 46. Le AD, Quan B, Juzytch W, Fletcher PJ, Joharchi N, Shaham Y. Reinstatement of alcohol-seeking by priming injections of alcohol and exposure to stress in rats. *Psychopharmacol (Berl)*. 1998;135:169–74.
 47. Hauser SR, Deehan GA Jr., Knight CP, Toalston JE, McBride WJ, et al. Parameters of context-induced ethanol (EtOH)-seeking in alcohol-preferring (P) rats: temporal analysis, effects of repeated deprivation, and EtOH priming injections. *Alcohol Clin Exp Res*. 2016;40:2229–39.
 48. Shaham Y, Shalev U, Lu L, de Wit H, Stewart J. The reinstatement model of drug relapse: history, methodology and major findings. *Psychopharmacol (Berl)*. 2003;168:3–20.
 49. Spano MS, Fattore L, Fratta W, Fadda P. The GABAB receptor agonist baclofen prevents heroin-induced reinstatement of heroin-seeking behavior in rats. *Neuropharmacology*. 2007;52:1555–62.
 50. Fattore L, Spano MS, Cossu G, Scherma M, Fratta W, Fadda P. Baclofen prevents drug-induced reinstatement of extinguished nicotine-seeking behaviour and nicotine place preference in rodents. *Eur Neuropsychopharmacol*. 2009;19:487–98.
 51. Campbell UC, Lac ST, Carroll ME. Effects of baclofen on maintenance and reinstatement of intravenous cocaine self-administration in rats. *Psychopharmacol (Berl)*. 1999;143:209–14.
 52. Gabriele A, See RE. Lesions and reversible inactivation of the dorsolateral caudate-putamen impair cocaine-primed reinstatement to cocaine-seeking in rats. *Brain Res*. 2011;1417:27–35.
 53. Rocha A, Kalivas PW. Role of the prefrontal cortex and nucleus accumbens in reinstating methamphetamine seeking. *Eur J Neurosci*. 2010;31:903–9.
 54. de Wit H. Laboratory-based assessment of alcohol craving in social drinkers. *Addiction*. 2000;95(Suppl 2):S165–169.
 55. Paulus MP, Stewart JL. Interoception and drug addiction. *Neuropharmacology*. 2014;76(Pt B):342–50.
 56. Duka T, Townshend JM. The priming effect of alcohol pre-load on attentional bias to alcohol-related stimuli. *Psychopharmacol (Berl)*. 2004;176:353–61.
 57. Amlung M, McCarty KN, Morris DH, Tsai CL, McCarthy DM. Increased behavioral economic demand and craving for alcohol following a laboratory alcohol challenge. *Addiction*. 2015;110:1421–8.
 58. Hicks JA, Schlegel RJ, Friedman RS, McCarthy DM. Alcohol primes, expectancies, and the working self-concept. *Psychol Addict Behav*. 2009;23:534–8.
 59. Wardell JD, Le Foll B, Hendershot CS. Preliminary evaluation of a human laboratory model of impaired control over alcohol using intravenous alcohol self-administration. *J Psychopharmacol*. 2018;32:105–15.
 60. Christiansen P, Townsend G, Knibb G, Field M. Bibi ergo sum: the effects of a placebo and contextual alcohol cues on motivation to drink alcohol. *Psychopharmacol (Berl)*. 2017;234:827–35.
 61. Bujarski S, Jentsch JD, Roche DJO, Ramchandani VA, Miotto K, Ray LA. Differences in the subjective and motivational properties of alcohol across alcohol use severity: application of a novel translational human laboratory paradigm. *Neuropsychopharmacology*. 2018;43:1891–9.
 62. Sinha R. The clinical neurobiology of drug craving. *Curr Opin Neurobiol*. 2013;23:649–54.
 63. Farokhnia M, Momenan R, Leggio L. (2017): Advancing medication development for addiction-behavioral and neuroimaging

- outcomes as indirect biomarkers of target engagement. *Addiction*. 2017;112:1869–70. Commentary on Schmitz et al.
64. Spanagel R, Zieglansberger W. Anti-craving compounds for ethanol: new pharmacological tools to study addictive processes. *Trends Pharmacol Sci*. 1997;18:54–59.
65. Wardell JD, Ramchandani VA, Hendershot CS. A multilevel structural equation model of within- and between-person associations among subjective responses to alcohol, craving, and laboratory alcohol self-administration. *J Abnorm Psychol*. 2015;124:1050–63.
66. Morean ME, Corbin WR. Subjective response to alcohol: a critical review of the literature. *Alcohol Clin Exp Res*. 2010;34:385–95.
67. Schuckit MA. Low level of response to alcohol as a predictor of future alcoholism. *Am J Psychiatry*. 1994;151:184–9.
68. Ray LA, Bujarski S, Roche DJ. Subjective response to alcohol as a research domain criterion. *Alcohol Clin Exp Res*. 2016;40:6–17.
69. Ariwodola OJ, Weiner JL. Ethanol potentiation of GABAergic synaptic transmission may be self-limiting: role of presynaptic GABA(B) receptors. *J Neurosci*. 2004;24:10679–86.
70. Misgeld U, Bijak M, Jarolimek W. A physiological role for GABAB receptors and the effects of baclofen in the mammalian central nervous system. *Prog Neurobiol*. 1995;46:423–62.
71. Wan FJ, Berton F, Madamba SG, Francesconi W, Siggins GR. Low ethanol concentrations enhance GABAergic inhibitory postsynaptic potentials in hippocampal pyramidal neurons only after block of GABAB receptors. *Proc Natl Acad Sci USA*. 1996;93:5049–54.
72. Weiner JL, Valenzuela CF. Ethanol modulation of GABAergic transmission: the view from the slice. *Pharmacol Ther*. 2006;111:533–54.
73. Roberto M, Gilpin NW, O'Dell LE, Cruz MT, Morse AC, Siggins GR, et al. Cellular and behavioral interactions of gabapentin with alcohol dependence. *J Neurosci*. 2008;28:5762–71.
74. Peris J, Eppler B, Hu M, Walker DW, Hunter BE, Mason K, et al. Effects of chronic ethanol exposure on GABA receptors and GABAB receptor modulation of 3H-GABA release in the hippocampus. *Alcohol Clin Exp Res*. 1997;21:1047–52.
75. Vlachou S, Markou A. GABAB receptors in reward processes. *Adv Pharmacol*. 2010;58:315–71.
76. Maccioni P, Zaru A, Loi B, Lobina C, Carai MA, Gessa GL, et al. Comparison of the effect of the GABABeta receptor agonist, baclofen, and the positive allosteric modulator of the GABAB receptor, GS39783, on alcohol self-administration in 3 different lines of alcohol-preferring rats. *Alcohol Clin Exp Res*. 2012;36:1748–66.
77. Augier E, Dulman RS, Damadzic R, Pilling A, Hamilton JP, Heilig M. The GABAB positive allosteric modulator ADX71441 attenuates alcohol self-administration and relapse to alcohol seeking in rats. *Neuropsychopharmacology*. 2017;42:1789–99.
78. Abrahao KP, Salinas AG, Lovinger DM. Alcohol and the brain: neuronal molecular targets, synapses, and circuits. *Neuron*. 2017;96:1223–38.
79. Siggins GR, Roberto M, Nie Z. The tipsy terminal: presynaptic effects of ethanol. *Pharmacol Ther*. 2005;107:80–98.
80. Chick J, Nutt DJ. Substitution therapy for alcoholism: time for a reappraisal? *J Psychopharmacol*. 2012;26:205–12.
81. Leggio L, Garbutt JC, Addolorato G. Effectiveness and safety of baclofen in the treatment of alcohol dependent patients. *CNS Neurol Disord Drug Targets*. 2010;9:33–44.
82. Holtyn AF, Kaminski BJ, Weerts EM. Baclofen and naltrexone effects on alcohol self-administration: Comparison of treatment initiated during abstinence or ongoing alcohol access in baboons. *Drug Alcohol Depend*. 2017;179:47–54.
83. Mann K, Aubin HJ, Charlet K, Witkiewitz K. Can reduced drinking be a viable goal for alcohol dependent patients? *World Psychiatry*. 2017;16:325–6.
84. Rose AK, Duka T. Effects of dose and time on the ability of alcohol to prime social drinkers. *Behav Pharmacol*. 2006;17:61–70.