Received: 23 February 2023 | Revised: 26 April 2023 | Accepted: 27 April 2023

DOI: 10.1111/adb.13288

Addiction Biology SSA WILEY

ORIGINAL ARTICLE

Mifepristone as a pharmacological intervention for stress-induced alcohol craving: A human laboratory study

Carolina L. Haass-Koffler[1,2,3,4] | Molly Magill[1,3] | Nazzareno Cannella[5] | Joshua C. Brown[6] | Elie G. Aoun[7] | Patricia A. Cioe[1,3] | Rajita Sinha[8] | Robert M. Swift[1,2,9] | Roberto Ciccocioppo[5] | Lorenzo Leggio[1,3,10,11,12,13]

[1]Center for Alcohol and Addiction Studies, Brown University, Providence, Rhode Island, USA

[2]Department of Psychiatry and Human Behavior, Warren Alpert Medical School, Brown University, Providence, Rhode Island, USA

[3]Department of Behavioral and Social Sciences, School of Public Health, Brown University, Providence, Rhode Island, USA

[4]Carney Institute for Brain Science, Brown University, Providence, Rhode Island, USA

[5]School of Pharmacy, Pharmacology Unit, University of Camerino, Camerino, Italy

[6]Department of Psychiatry, Harvard Medical School, McLean Hospital, Belmont, Massachusetts, USA

[7]Division of Law, Ethics and Psychiatry, Department of Psychiatry, College of Physicians and Surgeons, Columbia University, New York, New York, USA

[8]Yale Stress Center, Department of Psychiatry, Department of Neuroscience, Yale School of Medicine, Yale University, New Haven, Connecticut, USA

[9]Providence Veterans Affairs Medical Center, Providence, Rhode Island, USA

[10]Clinical Psychoneuroendocrinology and Neuropsychopharmacology Section, Translational Addiction Medicine Branch, NIDA IRP and NIAAA DICBR, Baltimore, Maryland, USA

[11]Medication Development Program, National Institute on Drug Abuse Intramural Research Program, National Institutes of Health, Baltimore, Maryland, USA

[12]Division of Addiction Medicine, Department of Medicine, Johns Hopkins University School of Medicine, Baltimore, Maryland, USA

[13]Department of Neuroscience, Georgetown University Medical Center, Washington, DC, USA

Correspondence
Carolina L. Haass-Koffler, Department of Psychiatry and Human Behavior, Brown University, 121 South Main Street, Providence, RI 02903, USA.
Email: carolina_haass-koffler@brown.edu

Funding information
This study was funded by the National Institute on Alcohol Abuse and Alcoholism (NIAAA; K01 AA023867 to CLH-K) and a Research Excellence Award from the Center of Alcohol and Addiction Studies, Brown University (CLH-K). Dr. Haass-Koffler is also supported by the NIAAA grants R01 AA026589, R01 AA027760 and R21 AA027614 and by the National Institute of General Medical Sciences (NIGMS), Center of Biomedical Research Excellence (COBRE, P20 GM130414). Drs. Magill and Cioe are supported by the NIAAA grants R01 AA027760 and R21 AA027614 (PI CLH-K). Dr. Leggio is supported by the National Institute on Drug Abuse Intramural Research Program and the NIAAA Division of Intramural

Abstract

Preclinical and clinical work suggests that mifepristone may be a viable treatment for alcohol use disorder (AUD). This was a Phase 1/2, outpatient, cross-over, randomized, double-blind, placebo-controlled trial with non-treatment-seeking individuals with AUD ($N = 32$). We assessed safety, alcohol craving and consumption, after 1-week mifepristone 600 mg/day administration, in a human laboratory study comprised of a single oral yohimbine administration (32.4 mg), a cue-reactivity procedure and alcohol self-administration. Safety was monitored by adverse events and hemodynamic parameters, alcohol craving by alcohol craving questionnaire and cue-induced saliva output. During the alcohol self-administration, we assessed alcohol pharmacokinetics, subjective effects and consumption. Outcomes were assessed using Generalized Estimating Equations and mediation analysis. Mild-moderate adverse events were reported in both conditions. There was no statistically significant difference between mifepristone and placebo in alcohol pharmacokinetics and subjective effects. Furthermore, blood pressure increased only in the placebo condition after the stress-induced laboratory procedures. Mifepristone, compared to placebo, significantly reduced

Clinical trial registration: Clinicaltrials.gov; NCT02243709
IND/FDA: 121984, mifepristone and yohimbine (Holder: Haass-Koffler).

Clinical and Biological Research (ZIA DA000635). The views expressed herein are those of the authors and do not reflect the official policy or position of the funding agencies.

alcohol craving and increased cortisol levels. Mifepristone-induced cortisol increase was not a mediator of alcohol craving. Mifepristone, compared to placebo, did not reduce alcohol consumption in the laboratory or in a naturalistic setting. This study successfully translated a developed preclinical procedure to a human laboratory study, confirming the safety of mifepristone in people with AUD and providing evidence to its role in reducing alcohol craving under stress procedures. The lack of effects on alcohol drinking may be related to the selection of non-treatment seekers and suggests future treatment-oriented trials should investigate mifepristone in people with AUD.

KEYWORDS
alcohol use disorder, glucocorticoids, noradrenergic, stress, yohimbine

1 | INTRODUCTION

Stress plays a key role in several neuropsychiatric disorders, including depression, anxiety[1] and alcohol use disorder (AUD).[2,3] Stress, combined with re-exposure to priming or to environmental cues previously associated with alcohol, exacerbates reoccurring drinking episodes both in rodents[4–6] and humans.[7,8] The mechanisms underlying these relationships are complex and include noradrenergic,[9] corticotropin releasing factor (CRF)[10] and glucocorticoid receptor (GR) pathways.[11,12]

Mifepristone, a GR/progesterone antagonist, is an FDA-approved medication for the termination of early pregnancy and for the treatment of hyperglycemia secondary to endogenous Cushing syndrome, in adults who have failed surgery or are not candidates for surgery. Mifepristone has been studied as a potential treatment for neuropsychiatric disorders including psychotic depression[13] and AUD.[14]

In our previous preclinical work, systemic administration of mifepristone, as well as its infusion in the central nucleus of the amygdala, reduced yohimbine-induced reinstatement of alcohol-seeking in alcohol-dependent Long Evans rats.[15] In male alcohol-dependent Wistar rats, mifepristone administration inhibited the development of alcohol escalation[16] and reduced alcohol-intake after extended abstinence.[14] In primates, cortisol mediated mifepristone effects on alcohol self-administration in a rhesus macaque AUD model.[17] However, in baboons under a chained schedule of reinforcement, mifepristone did not reduce alcohol-seeking or self-administration.[18] A more recent study that tested different novel GR compounds confirmed that, in addition to one GR modulator, mifepristone was the most effective drug in reducing alcohol consumption in alcohol-dependent animals.[19]

Taken together, the present literature supports a role of mifepristone in AUD but also suggests the need for additional studies to shed light on the mechanism(s) by which mifepristone may affect alcohol-related outcomes, particularly during stressful events.[14–16,20] With that in mind, in order to further understand the role of stress in mediating the potential beneficial effects of mifepristone in AUD, we aimed to provide a direct translation of our previous stress-induced preclinical work here.[15] As such, we utilized yohimbine (α2-receptor antagonist), rather than other stressors (psychological/physical), to specifically investigate the effects of mifepristone on noradrenergic activation. Glucocorticoids are secreted and bind to GR as part of the hypothalamic-pituitary-adrenal (HPA) axis stress response, which is further activated by the noradrenergic action of yohimbine. Of note, yohimbine is a well-validated pharmacological tool[21] that has been widely employed in preclinical alcohol research studies to evaluate the effect of noradrenergic activation.[15,22,23] As a pharmacological challenge, yohimbine was shown to activate the HPA axis in addition to increasing sympathetic nervous system activity[24] and increasing alcohol craving.[25] It is important to note that in a clinical laboratory setting, individuals may require also hydrocortisone to evoke a sustained cortisol response in addition to other physiological stress responses.[26,27]

We used a human laboratory paradigm designed to activate the noradrenergic system by a single oral dose of yohimbine (32.4 mg) paired with a cue-reactivity procedure, a priming alcohol dose and alcohol self-administration in an open bar laboratory. The primary outcome of this study was to test the safety of 1-week oral administration of mifepristone (600 mg/day) compared to placebo. Secondary outcomes included alcohol craving and consumption during the human experimental laboratory procedures. Other outcomes included monitoring the safety and efficacy of mifepristone (alcohol craving/consumption), compared to placebo, during the outpatient administration.

2 | MATERIALS AND METHODS

2.1 | Study design, setting and approval

This was a Phase 1/2, outpatient, cross-over, randomized, double-blind, placebo-controlled, human laboratory study (Figure 1). A cross-over design was chosen for this study because the within-subject variation was less than the between-subject variation and allowed for recruitment of less participants. The study was conducted at the Center for Alcohol and Addiction Studies, Brown University, Providence,

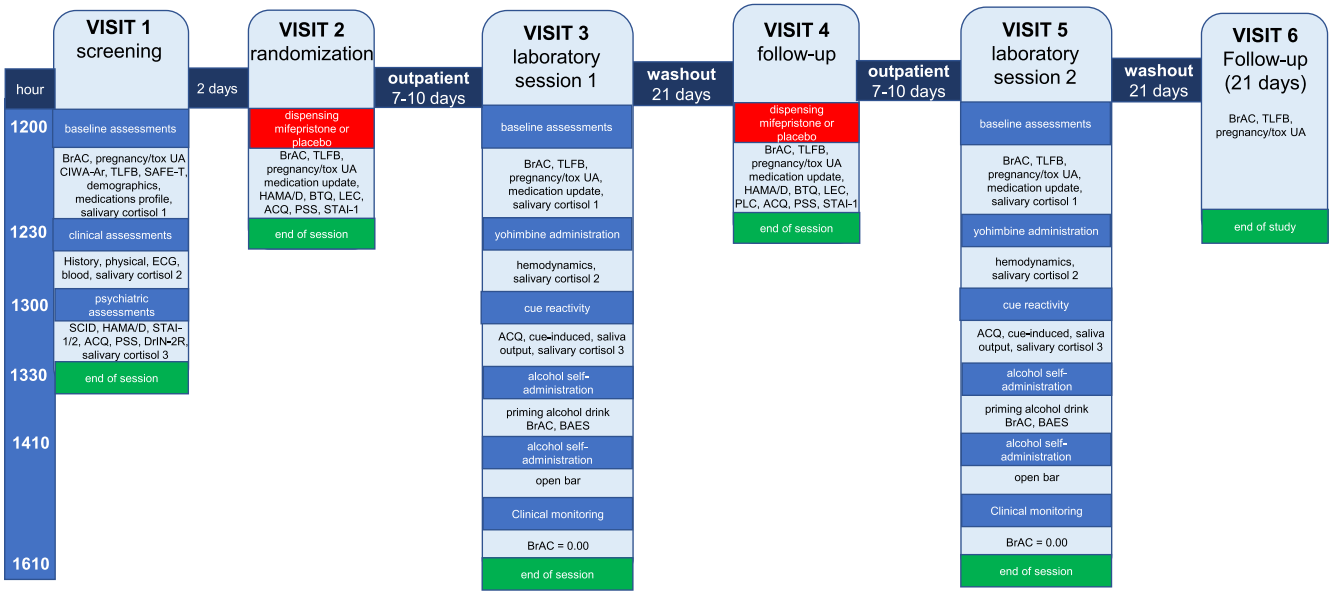

FIGURE 1 Study outline. Visit 1 (screening); Visit 2 (randomization, mifepristone or placebo); Visit 3: laboratory 1 (mifepristone or placebo), washout period (21 days); Visit 4 (follow-up and second condition: placebo or mifepristone); Visit 5: laboratory 2 (opposite condition, counter balanced); and Visit 6 (follow up). Legend: ADS, Alcohol Dependence Scale; ACQ, Alcohol Craving Questionnaire; BAES, Biphasic Alcohol Effects Scale; BrAC, breath alcohol concentration; BTQ, Brief Trauma Questionnaire; CIWA-Ar, Clinical Institute Withdrawal Assessment for Alcohol-revised; DrInc, Drinker inventory consequences; ECG, electrocardiogram; FHDA, Family History Density of Alcoholism; HAMA/HAMD, Hamilton Anxiety and Depression Rating Scale; LEC, Life Event Checklist; PSS, Perceived Stress Scale; SAFE-T, Suicide Assessment Five-Step Evaluation and Triage; SCID, Structured Clinical Interview for DSM-IV, STAI, Spielberger State Trait Anxiety; TLFB, Timeline Followback; UA, urine analysis.

RI, United States, from 2014 to 2021. The trial was approved by the Brown University Institutional Review Board, conducted under an FDA Investigational New Drug application (IND121984) and registered at clinicaltrials.gov (NCT02243709).

2.2 | Participants

After signing a written informed consent, a screening was performed to assess inclusion criteria: individuals who are non-abstinent, 21–65 years old, not seeking treatment for AUD (current Diagnostic and Statistical Manual of Mental Disorders Text Revision four edition diagnosis of alcohol dependence), meet criteria for moderate to heavy drinking (women: ≥2 drinks/day; men ≥3 drinks/day, during 90 days prior to screening) and good health as confirmed by medical history, physical examination, 12-lead electrocardiogram (ECG) and clinical laboratory tests. Females had to be postmenopausal for at least 1 year, surgically sterile or using a barrier, non-hormonal birth control method. All participants needed a breath alcohol content (BrAC) = 0.00 g/dl at each visit, be willing to take oral study medication and adhere to the study procedures.

Exclusion criteria included individuals seeking treatment for AUD; positive urine for opioids, benzodiazepines, cocaine, methamphetamine and tetrahydrocannabinol; diagnosed with a current substance use disorder other than alcohol or nicotine; met criteria for a lifetime diagnosis of bipolar disorder, schizophrenia or other psychotic disorders; active illness within the past 6 months of the screening visit that met the criteria for a diagnosis of Major Depressive Disorder (MDD) or Anxiety Disorder, or history of attempted suicide; clinically significant medical abnormalities: unstable hypertension, clinically significant abnormal ECG, bilirubin >150% of the upper normal limit (UNL), alanine aminotransferase/aspartate transaminase (ALT/AST) >5 times the UNL, creatinine clearance ≤60 dl/min; current use of psychotropic medications that may have an effect on alcohol consumption; current use of any medication involved in the metabolism of alcohol, such as aldehyde dehydrogenase, alcohol dehydrogenase and CYP2E1; current use of any medication (CYP3A4 inhibitor/substrate) that may interact with mifepristone; current use of any medication (CYP2D6 inhibitor/substrate) that may interact with yohimbine; history of seizure disorders; hypokalemia <3.5 mEq/L; participated in any behavioural and/or pharmacological study within the past 30 days; neuroendocrine disorders; taking corticosteroids; bleeding disorders; pre-existing QT prolongation on ECG (470 ms female; 450 ms male); history of porphyria; and not willing to engage in protected sex. Even though past clinical trials with mifepristone showed no increased depression,[28,29] MDD and anxiety disorder were ascertained with Structured Clinical Interview for DSM-IV (SCID-IV).[30]

Eligible participants were randomly assigned by computer allocation to 7-day treatment with either daily 600 mg mifepristone or placebo. After a 3-week washout period, to allow cortisol levels to return to baseline after mifepristone administration[31] and to avoid carryover effect, participants returned to the laboratory and received the crossover condition.

2.3 | Study drugs, dose justification and compliance

Mifepristone does not require a titration/taper schedule nor does it need to be adjusted by weight. The dose for this trail was based on previous work with individuals with AUD.[14] Furthermore, the effect of mifepristone treatment (short and long duration)[32] in clinical settings was shown to be safe and tolerable,[33,34] including in patients with diagnoses of depression, anxiety, post-traumatic stress disorder (PTSD)[35] and AUD.[14] Compliance was monitored by pill count and by saliva cortisol, as mifepristone increases the cortisol level by tenfold compared to placebo.[31]

The oral dose of yohimbine was based on prior studies in which yohimbine was administered to examine neuroendocrine activation in humans,[25,35,36] it does not need to be adjusted by weight and it was prepared/dispensed for each participant by a compounding pharmacy.

2.4 | Study procedures

2.4.1 | Visit 1 (screening)

Following a breath analyser (BrAC = 0.00 g/dl), participants signed a written-informed consent. The screening assessments included clinical assessments, medical history, physical examination, vital signs, ECG, blood/urine analysis and psychiatric assessments: Structured Clinical Interview for DSM-IV-TR Axis I Disorders (SCID-IV)[30]; Hamilton anxiety rating scale (HAM-A)[37] and depression scale (HAM-D)[38]; Spielberg State and Trait Anxiety Inventory (STAI Y-1 and Y-2)[39]; Perceived Stress Scale (PSS)[40]; and the Suicide Assessment Five-Step Evaluation and Triage (SAFE-T).[41] Alcohol consumption was measured using the timeline follow back (TLFB)[42] over 90 days prior screening. All visits were scheduled at the same time, in order to collect saliva samples to be measured both under basal 'Rest' state (Visit 1) and induced 'Stress' response (Visits 3 and 5) conditions. After a study physician approved the medical history and clinical laboratory tests, the participants were scheduled for Visit 2.

2.4.2 | Visit 2 (randomization)

After a BrAC = 0.00 g/dl, other assessments included TLFB,[42] Life Events (LEC)[43] and Brief Trauma Questionnaire (BTQ).[44] Study medication (mifepristone or placebo) was dispensed for 7–10 days (to facilitate participant schedule) outpatient administration, with the last dose of study drug administered in the laboratory.

2.4.3 | Visits 3 and 5 (alcohol laboratory session)

Participants were instructed not to consume alcohol for 24 h (BrAC = 0.00 g/dl), and Clinical Institute Withdrawal Assessment for Alcohol-revised (CIWA-Ar)[45] score of ≤10 was required. A sample of saliva was collected, and then a single oral dose of 32.4 mg yohimbine was administered to participants. The cue-reactivity began 30-min later, in order to allow yohimbine to take effect,[46] and a second sample of saliva was collected. The third saliva sample was collected after the cue-reactivity. The cue-reactivity was similar to previously published studies.[8,47] The water trial was included as a neutral control, and then, participants underwent two 3-min alcohol cue exposure trials. After every 3-min beverage exposure, participants rated their craving by completing the alcohol craving questionnaire (ACQ).[48] We also included cue-induced saliva output collected by using a cotton roll placed in the mouth of participants (weight, g) monitored at each trial of the cue-reactivity. Systolic/diastolic blood pressure (SBP/DBP, mmHg) and heart rate (HR, beats/min) were monitored continuously. Following the cue-reactivity, participants underwent the alcohol self-administration (ASA) procedure (priming alcohol drink and open bar). Participants received a priming dose of alcohol designed to raise blood alcohol levels to 0.05 g/dl, adjusted for sex and body weight. After the priming dose of alcohol, alcohol pharmacokinetics was measured by BrAC every 10 min, and stimulant/sedative effects of alcohol were assessed using the Biphasic Alcohol Effects Scale (BAES).[49–51]

The 'open bar' phase provided a total of eight standard drink unit (SDU), and all could be consumed within 120 min, with two trays of four drinks (0.015 g/dl/each) every 60 min.[49,52–54] As an alternative reinforcer for not drinking, we provided $3 per each drink not consumed. If the participant's BrAC reached 0.1 g/dl, the alcohol consumption ended. Participants waited until BrAC = 0.00 g/dl and hemodynamics normalized before being discharged.

2.4.4 | Visits 4 and 6 (washout and follow-up)

After a 3-week washout period, participants returned to receive the opposite medication (placebo or mifepristone) for a week. Participants then returned for a 21-day follow-up for final assessments.

2.5 | Statistical analysis

For all outcomes, we utilized an intention-to-treat (ITT) approach, where participants were examined based on their a priori randomized protocol and received at least one dose of the study medication (mifepristone or placebo).[55] The ITT analysis was also suitable for this crossover design,[56] as placebo was treated identically to the active drug condition (route, duration of administration and laboratory procedures).

Distributional characteristics of outcome measures were examined to evaluate similarity to the normal distribution, detailed descriptive analysis of demographics, substance use and clinical characteristics. The sedation scale for the BAES, cortisol and amylase had a skewness and kurtosis in excess of two; consequently, an outlier analysis was performed, and one outlier per group outside of ±3 interquartile range was treated as recommended.[57] Comparisons with these characteristics, in relation to enrolled versus completer status,

were performed using *t* tests to analyse continuous variables (age) and χ^2 for categorical variables (sex, race and smoking status). Attrition rates between the screening visit and follow-up visit were examined descriptively to assess for potential bias. In addition, a logistic regression was performed to test for possible bias due to period (placebo first, then mifepristone and mifepristone first and then placebo) or medication carryover (placebo and mifepristone), as done in our prior cross-over trial.[49] Effect size was reported as Cohen *d*.

2.5.1 | Primary outcomes

Safety and tolerability of oral administration of mifepristone was assessed after 7–10 days in outpatient setting and when it was administered with yohimbine and alcohol during the laboratory paradigms. We compared the number of adverse events (AEs) between the mifepristone and placebo condition via a χ^2. The results were presented using summary statistics: number of subjects (*n*); mean (*M*); standard deviation (*SD*) or frequency distributions (%). The safety and tolerability of mifepristone, compared to placebo, were also assessed by monitoring hemodynamic response, and alcohol pharmacokinetics and subjective effects, using Generalized Estimating Equations (GEE)[58] with robust standard errors and an unstructured correlation matrix. We conducted GEE with both laboratory paradigm procedures (time coded specifically for each analysis), medication (mifepristone/placebo) and visit (screening/laboratory) as within-subject factors. The model was specified to evaluate the effect of: drug by time (laboratory procedure) interaction, main effect of the drug (mifepristone/placebo condition) and main effect of time. Hemodynamic response included SBP, DBP and HR, with the laboratory paradigm procedures coded as time effect: t_0 = yohimbine administration, t_{30min} = pre and t_{60min} = post cue-reactivity.

2.5.2 | Secondary outcomes

Craving measures included alcohol craving questionnaire short form-revised (ACQ-SF-R)[59] and cue-induced saliva output (g). Values of the water trials for each dependent variable were inserted as covariate in the model (allowed for the dependent variable to be specific for alcohol), time coded: t_1 = alcohol trial 1 and t_2 = alcohol trial 2. Alcohol consumption of the mifepristone condition, compared to placebo, was assessed in the bar laboratory and in the outpatient setting during the 7-day medication administration and after 21-day post treatment. In the bar laboratory, alcohol consumption was measured by number of drinks consumed (*t* test). In the outpatient setting, alcohol consumption was measured by self-report using the TLFB method, reported as heavy drinking days (HDD) and drinks per week (DPW), with time coded as t_0: baseline and t_1: after 7-day mifepristone/placebo and t_2: 3-weeks after study medication administration, as conducted before,[14] to evaluate the long lasting effect of mifepristone.

Alcohol pharmacokinetics parameters included time to reach max concentration (T_{max}), max concentration (C_{max}) and area under the curve (AUC), calculated by $\int_{t=40}^{t=0}(BrAC)dx$ (t_0 = time pre and t_{40min} = 40-min post prime alcohol administration), and were analysed via data collected from the BrAC curve using confidence interval (*CI*) and interval estimate confidence, set at 95%. Subjective alcohol-related biobehavioural effects (stimulation/sedation) were measured by the Biphasic Alcohol Effects Scale (BAES)[51] on the alcohol biphasic curve. Data were collected from the breath alcohol content (BrAC) curve: t_{10min} = ascending and t_{20min} = descending limb.

2.5.3 | Mediation

Analyses for cortisol level, on ACQ and cue-induced saliva output, were conducted using a regression-based Macro Estimating Model[60] that estimated the indirect effect of a within-participant manipulation on outcomes. Mediation was tested using standard procedures (product of the *a* and *b* path coefficients), but difference scores were created for the mediator and outcome under mifepristone/placebo conditions. The dependent variables were ACQ and cue-elicited saliva output, and the mediator was the cortisol level after 7-day mifepristone (M_1) or placebo (M_2) administration. The indirect effect was tested with Monte Carlo *CI* (95%).

All statistical analyses were performed after participants had completed their follow-up visits, and the study database had been locked. All the statistical procedures were performed by IBM SPSS Statistics for Windows, version 27 with Macro MEMORE extension[9] (IBM Corp., Armonk, NY, USA), and GraphPad Prism (v.5) was used to generate figures (La Jolla, CA, USA). All statistical tests were two-tailed, and statistical significance was accepted if an alpha value $p < 0.05$ was obtained.

2.6 | Power analysis and missing data

This was a proof-of-concept trial to demonstrate the feasibility of the combined study design, the safety and tolerability of mifepristone and yohimbine while consuming alcohol and the potential value of testing mifepristone in an appropriately-powered larger RCT. In selecting a target sample size, we balanced power considerations and feasibility given the translational nature of this trial. Because of the within-subjects design, power to test the effects of the study drug was optimized for this modest sample size (originally $N = 20$ and then, after additional funding, increased to $N = 32$). For the safety and tolerability outcomes (primary) adverse events, difference was detected based on a judgement concerning the minimal effect, which has clinical relevance in the management of patients. In a noninferiority trial, the exact sample size could not be fixed in advance because it depends upon the chosen stopping guidelines.[61] Effect size reported as Cohen *d* was calculated for each analysis to describe how meaningful the difference was between mifepristone and placebo conditions.

For missing data approach, we first categorized missing data as missing completely at random (MCAR), missing at random (MAR) or

missing not at random (MNAR).[62] GEE analysis (using all available pairs of data to model missing values with maximum likelihood estimation) was deemed suitable for our analyses because no systematic differences existed between participants with missing data and those with complete data (MCAR).

3 | RESULTS

3.1 | Participants' characteristics and retention

The CONSORT diagram (extension for crossover trial)[63] is reported in Figure 2 and sociodemographic and baseline clinical characteristics of the participants in Table 1. One-hundred fifty-five participants were screened on the telephone, 46 were screened in person, 32 were randomized and 27 completed the study. Thirty-two received at least one dose of the study medication and were included in ITT analysis.

There was no difference in the attrition analysis conducted using period or medication (p's > 0.05) as predictors. Five individuals withdrew from the study. In the mifepristone condition, one individual did not attend the first laboratory visit due to a family emergency ($n = 1$, 3%). In the placebo condition, one individual was not compliant with the laboratory procedures, one individual experienced a non-serious adverse event, one participant was hospitalized for an event not related to the study procedures/medication and one individual ceased contact before attending the first laboratory procedures ($n = 4$, 13%). For the second laboratory session, one individual in the placebo condition was unable to complete the study in person due to COVID-19 in person restrictions ($n = 1$, 3%); however, the data in the naturalistic condition (no laboratory procedures) were completed with assessments collected remotely with an IRB-approved amendment.[64] There were no systematic differences between participants with missing data and those with complete data; therefore, data were considered missing completely at random (MCAR)[62] and used the GEE Standard Method.[65]

3.2 | Primary outcome

There were no serious adverse events (AEs) when the study medication was co-administered with yohimbine and alcohol in the laboratory. We observed three non-serious AEs (mifepristone: $n = 0$, 0%; placebo: $n = 3$, 10%; $p > 0.05$). Two individuals had an emesis episode after yohimbine and alcohol administration, and one individual experienced increased blood pressure (hypertensive urgency) after yohimbine administration, but before alcohol administration, however, blood pressure normalized after the alcohol administration. The safety and the tolerability of the laboratory procedures were also assessed by monitoring the hemodynamic function (SBP, DBP and HR) (Figure 3). For SBP, we found a drug by time interaction, where these increases were observed only in the placebo condition after the cue-reactivity (t_{60min}, $p = 0.020$), no significant main effects for drug and a significant time effect such that SBP increased from baseline after

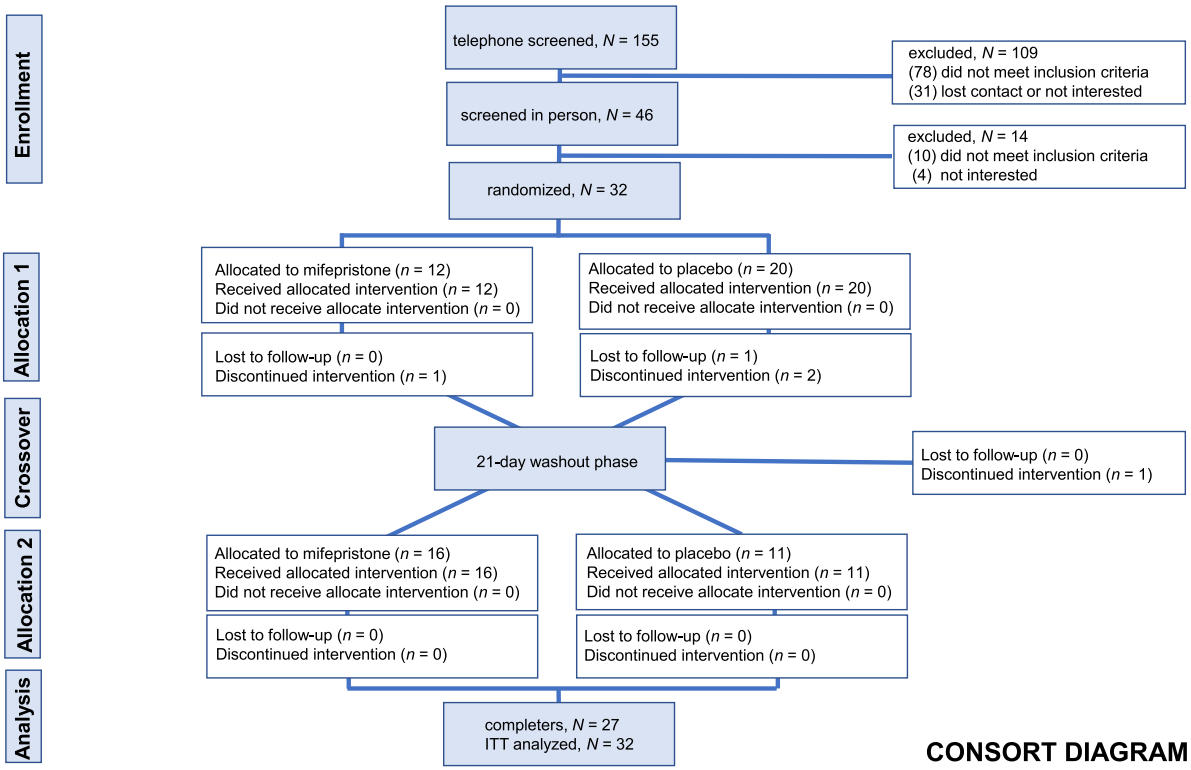

FIGURE 2 Consolidated Standards of Reporting Trials (CONSORT) extension for cross-over trials.

TABLE 1 Sociodemographic and baseline clinical characteristics at screening of the initial sample of participants who were enrolled and randomized in the study, expressed as *n* (%) or *M* ± (*SD*).

Number (*N*)	32
Male, *n* (%)	27 (84)
Hispanic, *n* (%)	4 (13)
White, *n* (%)	22 (69)
Age, *N* (*SD*)	43 (12)
Marital status: married/relationship, *n* (%)	11 (34)
Smoker *n* (%)	11 (34)
Cannabis *n* (%)	11 (34)
Age onset alcoholism (AOA) (*SD*)	23 (8)
Baseline drinking days (DD) (*SD*)[a]	74 (17)
Baseline heavy drinking days (HDD) (*SD*)[a]	46 (30)
Baseline drinks per week (DPW) (*SD*)[a]	39 (26)
Alcohol dependence scale (ADS) (*SD*)	8 (6)
Alcohol craving questionnaire (ACQ) (*SD*)	43 (15)
Drinker inventory consequences (DrInc) (*SD*)	32 (18)
Family history density alcoholism (FHDA) > 66% (*SD*)	17 (53)
Systolic blood pressure (SBP) (*SD*)	126 (17)
Diastolic blood pressure (DBP) (*SD*)	77 (9)
Heart rate (HR) (*SD*)	74 (14)
Body Mass Index (BMI) (*SD*)[b]	29 (11)
State-State Anxiety Inventory (STAI, state) (*SD*)	31 (10)
State-Trait Anxiety Inventory (*STAI, trait*) (*SD*)	34 (11)
Hamilton anxiety rating scale (HAMA) (*SD*)	4 (5)
Hamilton depression rating scale (HAMD) (*SD*)	3 (4)
Perceived stress scale (PSS) (*SD*)	11 (6)
PTSD criterion A (Brief Trauma questionnaire), *n* (%)	18 (46)
Life event checklist (PTSD criterion A) *n* (%)	26 (67)

[a]Alcohol consumption was measured by self-report using 90-day timeline follow back (TLFB) method.
[b]Mifepristone and yohimbine do not need to be adjusted by weight.

cue-reactivity ($p < 0.013$) (Figure 3A). For DBP, we found a significant drug by time interaction, where these increases were observed only in the placebo condition after the cue-reactivity (t_{60min}, $p < 0.001$), and a main effect of drug such that DBP was lower in the mifepristone condition compared to placebo ($p = 0.005$), and a main effect for time such that DBP increased from baseline to pre (t_{30min}, $p = 0.001$) and post (t_{60min}, $p = 0.002$) cue-reactivity (Figure 3B). Finally, for HR, there was no significant drug by time interaction, main effect for drug or time (*p's* > 0.05) (Figure 3C).

During the 7-day administration of mifepristone or placebo in an outpatient setting, we did not observe serious AEs related to the study drugs/procedure. Mild to moderate non-serious AEs were reported by both study conditions throughout the trial, with no difference (*p's* > 0.05) (Table S2). Additionally, no differences were observed in anxiety (HAM-A), depression (HAM-D) and stress (PSS) levels between the mifepristone and placebo conditions (*p's* > 0.05).

3.3 | Secondary outcomes

3.3.1 | Alcohol craving and cue-elicited saliva output

A time by drug interaction suggested decrease of craving for the mifepristone condition, compared to placebo condition ($p = 0.007$) at the alcohol trial 1, no main effect for drug ($p > 0.05$), but a significant main effect for time ($p < 0.001$), where increases of craving were observed in alcohol trial 2 (Figure 4A).

Analysis of cue-elicited saliva output revealed a drug by time interaction was observed both at the alcohol trial 1 ($p < 0.001$) and alcohol trial 2 ($p < 0.001$), with decrease of saliva output in the mifepristone condition, compared to placebo condition. Also, there was a significant main effect for drug ($p < 0.001$), where lower saliva output was observed in the mifepristone condition, compared to placebo condition, and a significant main effect for time showed saliva decreases at the alcohol trial 2 ($p < 0.001$) (Figure 4B).

3.3.2 | Cortisol as mediator of alcohol craving and cue-elicited saliva output

Analysis of salivary cortisol during the cue-reactivity revealed a drug by time interaction, indicating higher cortisol levels both pre (t_{30min} $p < 0.001$) and post (t_{60min} $p < 0.001$) cue-reactivity. Also, there was a significant main effect for drug ($p < 0.001$), such that higher cortisol was observed in the mifepristone condition, compared to placebo condition, with no main effect for time ($p > 0.05$) (Figure 4C). This result further supports that participants adhered to the mifepristone regimen, as cortisol increases with mifepristone administration.[31] Finally, to test if participants responded to the laboratory procedures, the increase of the HPA activation was confirmed when we compared the value of cortisol levels collected at the screening visit (basal) to the values collected during the laboratory visits (stress) only in the placebo condition (Figure S1).

For the mediation analysis, we defined the total effect (the *c path*) and the direct effect (the *c′ path*) of the mifepristone condition on improving craving outcomes (ACQ and cue-elicited saliva output). The defined indirect effect (*a* × *b path*) did not show a relationship between cortisol (mediator) and ACQ ($p > 0.05$), and cue-elicited saliva output ($p > 0.05$) at the alcohol trial 1 or by combining the alcohol 1 and 2 trials. As a result, the Monte Carlo *CI* around the product of the *a* and *b* path coefficients were non-significant (*p's* > 0.05) (Figure 4D).

3.3.3 | Alcohol consumption

In the open-bar laboratory session, participants consumed a small number of standard alcohol drinks both in the mifepristone (0.8 ± 0.3) and placebo (0.5 ± 0.2) conditions, with no significant difference between conditions ($p > 0.05$). During the naturalistic

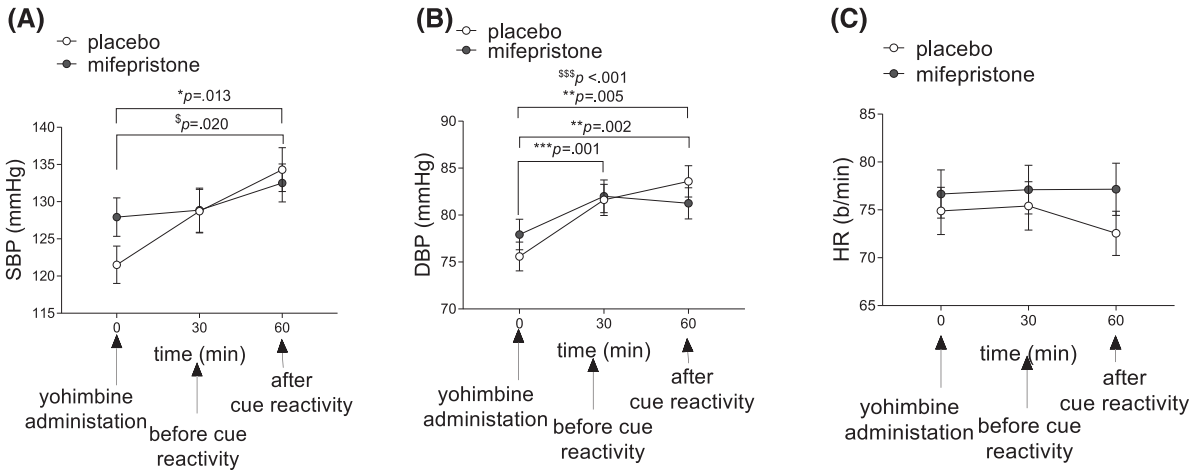

FIGURE 3 Hemodynamic function after administration of yohimbine paired to a cue reactivity. (A) SBP: a time by drug interaction, where these increases were observed only in the placebo condition post the cue-reactivity (t_{60min}, $b = 8.149$; $CI = 1.27, 15.03$; $p = 0.020$; $d = 0.321$); no significant main effect for drug, a significant time effect such that SBP increased from baseline to post cue-reactivity (t_{60min}, $b = 5.96$; $CI = 1.27, 10.65$; $p = 0.013$; $d = 0.602$). (B) DBP, a drug by time interaction, where these increases were observed only in the placebo condition after the cue-reactivity (t_{60min}, $b = 6.221$; $CI = 2.57, 9.88$; $p < 0.001$; $d = 0.018$), a significant main effect for drug, such that DBP was lower in the mifepristone condition compared to placebo condition ($b = -4.01$; $CI = -6.80, -1.215$; $p = 0.005$; $d = 0.118$); a main effect for time such that DBP increased from baseline to pre (t_{30min}, $b = 3.639$; $CI = 1.41, 10.21$; $p = 0.001$; $d = 0.615$) and post (t_{60min}, $b = 4.79$; $CI = 1.83, 7.75$; $p = 0.002$; $d = 0.602$) the cue-reactivity; and (C) HR: no significant effect (interaction and main effects, p's > 0.05). All data presented as mean ± *SEM*. $^{*}p < 0.05$ main effect; $^{\$}p < 0.05$ interaction. All Cohen *d* reported in Table S1.

outpatient setting, during the 7-day treatment and 21-day post treatment, participants reduced alcohol consumption; however, there was no difference between the mifepristone and placebo conditions (p's > 0.05).

3.3.4 | Drug-alcohol interaction

Drug-alcohol interaction was assessed by measuring alcohol pharmacokinetic parameters (C_{max}, T_{max} and AUC_{0-40}) and subjective response to alcohol (stimulation/sedation) (Figure 5). There were no differences in the mifepristone condition compared to placebo in the alcohol BrAC pharmacokinetic curve parameters (AUC, T_{max}, C_{max}) (p's > 0.05) (Figure 5A). Also, there were no significant differences in the mifepristone compared to placebo condition on the alcohol subjective effect in the Biphasic Alcohol Effects Scale (BAES) stimulation and sedation (p's > 0.05) scales (Figure 5B,C).

4 | DISCUSSION

The main finding of this study is that mifepristone, administered with yohimbine and alcohol, was safe in individuals with AUD. We also demonstrated that mifepristone, in a combined yohimbine/alcohol cue-reactivity paradigm, significantly reduced self-reported craving in the first alcohol challenge and reduced cue-elicited saliva output in both alcohol challenges. Mifepristone's effect in reducing yohimbine-induced alcohol craving was independent from the mifepristone-induced increase of cortisol level.

To our knowledge, this work represents the first translation to humans with AUD, of our previously reported pharmacological, stress-induced preclinical paradigm.[21] This bench-to-bed translation was successful both in terms of safety and methodological execution, as further indicated by the expected changes in physiological parameters, that is, increased blood pressure from pre to post cue-reactivity, a finding consistent with the known increase in blood pressure post cue-reactivity[66] and yohimbine[36] challenges.

Assessing the safety and tolerability of a study drug when co-administered with alcohol is important for both novel[67,68] and repurposed[69] medications under investigation for AUD. This approach is consistent with both FDA[70] and European Medicine Agency (EMA)[71] guidelines on the development of new AUD medications. We did not observe serious AEs, and non-serious AEs were encountered at similar frequencies in both mifepristone and placebo conditions. After the alcohol prime, mifepristone, compared to placebo, did not alter the alcohol pharmacokinetics nor did it affect the stimulation/sedation effects of alcohol. Together, the results of this study support the safety of mifepristone when co-administered with yohimbine and alcohol in people with AUD. Furthermore, the increases of blood pressure due to the stress-induced study paradigm (yohimbine administration and cue-reactivity) were higher in the placebo, compared to the mifepristone condition.

Mifepristone reduced the self-reported alcohol craving at the first, but not second, alcohol cue exposure; of note, the second alcohol cue exposure is known to further boost craving in cue-reactivity experiments.[72–74] On the other hand, the cue-elicited saliva output provided an objective biomarker for the effect of mifepristone in craving reduction in both challenges. This observation is in line with

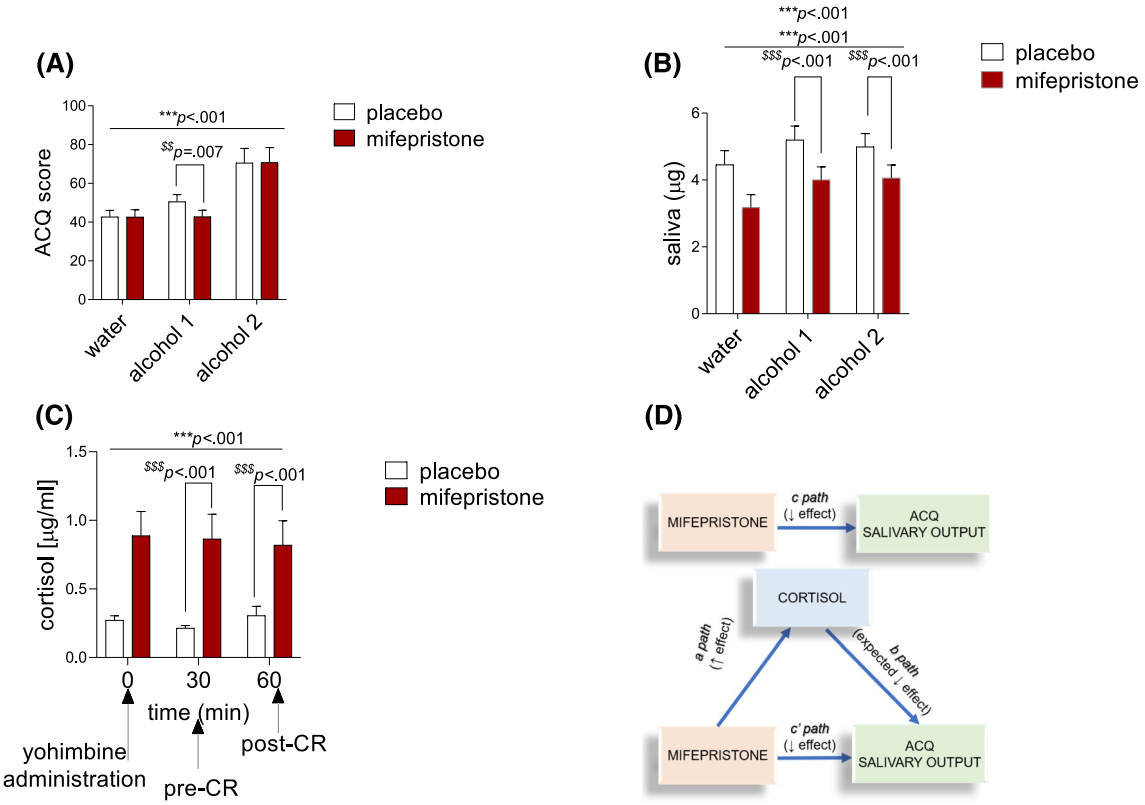

FIGURE 4 The effect of mifepristone compared to placebo on alcohol craving, cue-elicited saliva output and saliva cortisol. (A) ACQ: time by drug interaction ($b = -8.28$; $CI = -14.34, -2.22$; $p = 0.007$; $d = 0.481$) at the alcohol trial 1, no main effect for drug ($p > 0.05$), but a main effect for time ($b = 24.54$; $CI = 17.60, 31.48$; $p < 0.001$; $d = 0.246$) at the alcohol trial 1. (B) Cue-elicited saliva output: a time by drug interaction at the alcohol trial 1 ($b = -1.250$; $CI = -1.80, -0.70$; $p < 0.001$; $d = 0.727$) and alcohol trial 2 ($b = -1.253$; $CI = -1.76, -0.76$; $p < 0.001$; $d = 0.585$), a main effect for drug ($b = -1.373$; $CI = -1.94, -0.80$; $p < 0.001$; $d = 0.114$), a main effect for time ($b = -0.370$; $CI = -0.58, -0.16$; $p < 0.001$; $d = 0.623$) at the post cue-reactivity. (C) Cortisol: There was a drug by time interaction both pre- (t_{30min}: $b = 0.634$; $CI = 0.31, 0.99$; $p < 0.001$; $d = 0.741$) and post- (t_{60min}: $b = 0.49$; $CI = 0.29, -0.91$; $p < 0.001$; $d = 0.636$) cue-reactivity, a significant main effect for drug ($b = 0.54$; $CI = 0.22, 0.85$; $p < 0.001$; $d = 0.659$), but no main effect for time ($p > 0.05$). (D) Mediation model. Increase of cortisol level as mediator of alcohol craving, urge and saliva output after 7-day mifepristone administration before initiating any laboratory procedure. All Cohen *d* reported in Table S1.

cue-reactivity studies showing that salivation is associated less with conscious attention to alcohol but is more pronounced in individuals with serious AUD and is a strong predictor of alcohol consumption in the first period after detoxification.[75] The lack of mifepristone effect on the second self-reported craving could be due to the population of this study, which included 50% of individuals with history of trauma. A recent study showed that after a single prolonged stress exposure, only early mifepristone intervention (rather than later in life) improved fear extinction deficit and inhibited anxiety in rats.[76] Therefore, it is possible that a later intervention with mifepristone in a population with AUD and history of trauma is not sufficient to blunt a cue re-challenge in a yohimbine-induced alcohol craving paradigm. It also possible that this population would need a higher dose of mifepristone. In fact, a dose response of mifepristone was reported in a clinical study of patients with psychotic depression, where psychotic symptoms were reduced by mifepristone (1200 mg/day), and the effect was dependent on the blood level of mifepristone.[13]

While this trial was developed based on our original preclinical[15] and other translational[14,16] literature related to mifepristone in AUD, subsequent preclinical studies in an AUD model of rhesus monkeys showed that mifepristone decreased daily alcohol self-administration and that this effect was mediated by mifepristone-induced increase in cortisol.[17] When we tested this hypothesis in humans, we did not find that the effect of mifepristone on alcohol craving outcomes was mediated by increasing cortisol. Our results align with other clinical data showing that mifepristone's effects on reducing psychotic symptoms were independent of the increased plasma cortisol and adrenocorticotropin hormone.[13] This discrepancy from the preclinical study could be due to the fact that yohimbine was not administered in that monkey study.[17] Also, it is possible that salivary cortisol (as done in the present human study) is a more relevant measure for adrenocortical function than blood (as done in the previous monkey study). The saliva and serum total cortisol concentration have a non-linear relationship due to the rapid increase in saliva concentration once the serum cortisol-binding globulin is saturated.[77] Our clinical results align

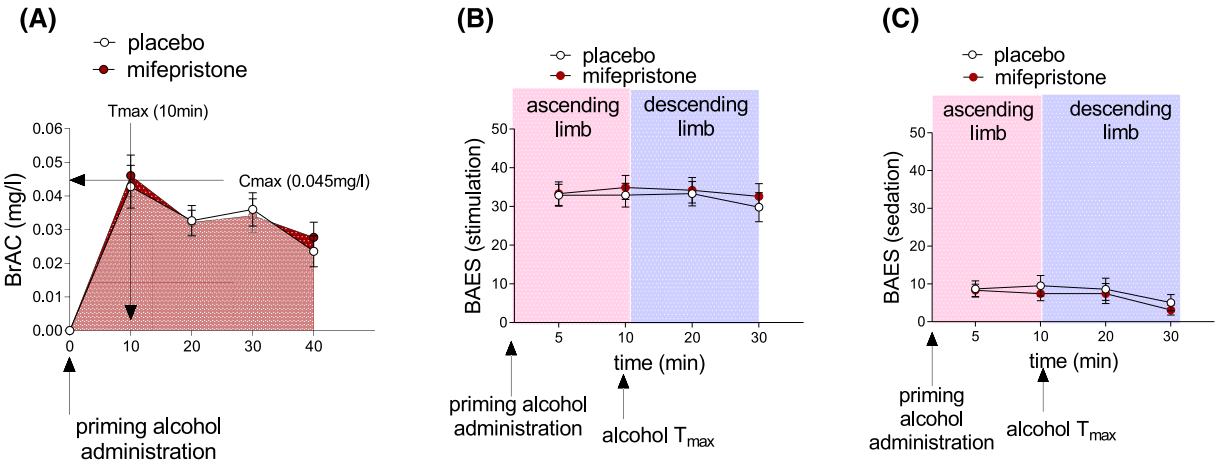

FIGURE 5 Alcohol pharmacokinetic and subjective response and after administration of yohimbine paired to a cue reactivity and alcohol self-administration paradigm: (A) alcohol pharmacokinetics: After the yohimbine administration, the cue-reactivity procedure and alcohol administration alcohol (BrAC to 0.03–0.05 mg/l), we found no difference in mifepristone, compared to placebo in the AUC, T_{max} and C_{max} (interaction and main effects, *p's* > 0.05). (B and C) Alcohol subjective effect: There was no significant difference in the mifepristone compared to placebo, in stimulation and sedation scales (interaction and main effects, *p's* > 0.05). All data presented as mean ± *SEM*. $^{*}p < 0.05$ main effect; $^{\$}p < 0.05$ interaction.

with our preclinical data which demonstrated that infusion of mifepristone directly into the amygdala suppressed yohimbine-induced reinstatement of alcohol-seeking, even though corticosterone levels were unaffected.[15] Our hypothesized involvement of the amygdala, rather than negative feedback on the hypothalamus, is further supported by other translational studies in AUD[14] and psychotic depression.[13]

In the open-bar laboratory session, participants, both in the mifepristone and placebo conditions, consumed only low amounts of alcohol, making it difficult to assess a medication effect in this paradigm (floor effect). In the 7-day treatment and 21-day post treatment, participants reduced alcohol consumption; however, contrary to a previous reported study,[14,16] there were no differences between mifepristone and placebo conditions. The lack of mifepristone effect during the naturalistic drinking (outpatient setting) could be due to our sample being individuals with high family history of AUD and history of early trauma. Case in point, Marchigian Sardinian alcohol preferring (msP) rats (a genetically selected rat model of AUD with phenotypic trait resembling anxiety and stress-related disorders such as PTSD) were less responsive to mifepristone's ability to reduce alcohol self-administration[78] or anxiety-like behaviour and startle responses.[79] Our results are also consistent with the previous baboon study where mifepristone did not reduce alcohol consumption,[18] supporting the hypothesis that in an individual with severe AUD who consumes large amounts of alcohol, mifepristone pharmacokinetics may be non-linear. In summary, given (1) the floor effect during the alcohol self-administration in the bar laboratory; (2) the unlikely ability that naturalistic drinking may be changed in an AUD population of non-treatment seekers; (3) the known differences between treatment-seekers versus non-treatment seekers for AUD[80]; (4) the fact that, unlike in our study, previous work[14] reported an effect of mifepristone in reducing alcohol drinking in people with AUD; and (5) the effects described here of mifepristone in reducing cue-induced craving and salivation, the latter being consistent with the previous findings[14]; taking all these factors together, this study provides support of the role of mifepristone as a novel treatment for AUD.

One of the major strengths of this study is the within-subject, cross-over design which provided the same set of participants acting as their own controls, increasing power and reducing variability.[49] The cortisol level also provided robust results for medication adherence, as it increases with mifepristone administration.[31] This is the first study in which yohimbine was paired to a cue-reactivity paradigm in an AUD population,[21] highlighting the translational efforts of this study bridging animal and human models.[81] The premise and the logistics of this work were paved by our preclinical study[15] using the same medications paired to alcohol laboratory paradigms. The robustness of the study paradigm was highlighted by participant retention, despite the long (3-week) washout period, which was necessary to allow the cortisol to return to basal levels after the mifepristone administration.

A major limitation of this study was not having a placebo-condition for yohimbine. As a result, we could not determine if the effect of stress induction was due to the interaction between yohimbine and cue-reactivity or cue reactivity alone. Regardless, both yohimbine and cue-reactivity are independently well-established and validated procedures for stress-induced alcohol craving and consumption.[21] Another limitation is that we did not collect mifepristone blood levels in an attempt to reduce a confounding variable of stress from blood draws. Therefore, we cannot ensure that clinical effects are dependent on mifepristone blood levels[13] nor can we determine dose–response relationships. From translational perspective, in our preclinical work,[15] we also utilize a yohimbine-induced alcohol reinstatement paradigm, which implies that the rodent model underwent a period of abstinence and return to alcohol. However, the participants included in this study were non-treatment AUD individuals

without abstinence period. To fully justify the translational 'reinstatement' paradigm, this human laboratory study should have included AUD participants with periods of abstinence. This procedure, in clinical setting, however, is not easy to obtain for safety and ethical considerations. Another limitation was the small sample size; nonetheless, this study may inform adequately powered future studies to ensure specific mechanism of action or precision medicine tailored for AUD endophenotypes.[82]

Finally, the low number of females enrolled in this study, due to the mifepristone FDA-indication, did not permit us to evaluate sex as a biological variable. From preclinical data, we can infer that mifepristone may have a different effect in males and females. For example, in nondependent Wistar rats, mifepristone reduced alcohol consumption only in females.[83] Further studies also should include additional alcohol endophenotypes as mifepristone did not reduced alcohol self-administration in alcohol-preferring msP rats of either sex.[78]

5 | CONCLUSION

In summary, this study provides important information on the safety of mifepristone as a medication to treat AUD. Our findings support the safety of mifepristone-alcohol combination in a human laboratory setting. The safety data of this trial supports the use of translational integration of yohimbine, combined with a cue-reactivity protocol and alcohol self-administration to evaluate the effects of stress-induced alcohol craving in humans.[84] In terms of efficacy, consistent with earlier clinical work,[14] we found an effect in reducing alcohol craving, an important behavioural marker and a diagnostic criterion in the DSM-5.[85,86] This translational trial fits with previous clinical studies that have utilized mifepristone in different psychiatry disorders, as mifepristone's effects on reducing craving were independent of the increased plasma cortisol.[13] Future studies, possibly testing higher doses, are warranted to assess mifepristone's alcohol consumption in patients with AUD and to best identify potential patients with AUD who are either responders or non-responders to mifepristone treatment.

AUTHOR CONTRIBUTIONS

Carolina L. Haass-Koffler is the lead for conceptualization and is responsible for funding acquisition, investigation, methodology, project administration, and analysis. Carolina L. Haass-Koffler, Patricia A. Cioe, Joshua C. Brown, Elie G. Aoun and Robert M. Swift carried out the experiments and collected the experimental data. Carolina L. Haass-Koffler and Molly Magill analysed the data. Carolina L. Haass-Koffler, Elie G. Aoun, Nazzareno Cannella, Roberto Ciccocioppo, Rajita Sinha, Joshua C. Brown, and Lorenzo Leggio provided substantial intellectual input on the interpretation of the results. All authors reviewed content and approved the final version of the manuscript.

ACKNOWLEDGEMENTS

The authors thank Dr. Leandro Vendruscolo for his input on the content of the manuscript and Brian Gully for proofreading the manuscript. We would also thank Michael Brickley, Zoe Brown, Danielle Giovenco, Victoria Long (Brown University) and Karen Hafez (Corcept Therapeutics) for providing logistics and technical support during the trial. The authors would also like to thank Prabhjot Singh and Talia Vasaturo-Kolondier for providing help with the database. The graphical abstract was created with BioRender.com.

CONFLICT OF INTEREST STATEMENT

The medication (mifepristone and matching placebo) was kindly provided by Corcept Therapeutics. Corcept Therapeutics did not have any role in the study design, execution or interpretation of the results, and this publication does not necessarily represent the official views of Corcept Therapeutics. CLH-K received travel support to CA to present the data to the Corcept Therapeutic Conference (September 2022). The other authors declare no competing interests.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID

Carolina L. Haass-Koffler https://orcid.org/0000-0002-5634-5679
Nazzareno Cannella https://orcid.org/0000-0002-2891-8679
Rajita Sinha https://orcid.org/0000-0003-3012-4349
Roberto Ciccocioppo https://orcid.org/0000-0003-3126-9240
Lorenzo Leggio https://orcid.org/0000-0001-7284-8754

REFERENCES

1. Vinkers CH, Kuzminskaite E, Lamers F, Giltay EJ, Penninx B. An integrated approach to understand biological stress system dysregulation across depressive and anxiety disorders. *J Affect Disord*. Mar 15 2021; 283:139-146. doi:10.1016/j.jad.2021.01.051
2. Haass-Koffler CL, Bartlett SE. Stress and addiction: contribution of the corticotropin releasing factor (CRF) system in neuroplasticity. *Front Mol Neurosci*. 2012;5:91. doi:10.3389/fnmol.2012.00091
3. Koob GF, Schulkin J. Addiction and stress: an allostatic view. *Neurosci Biobehav Rev*. 2019;106:245-262. doi:10.1016/j.neubiorev.2018.09.008
4. Liu X, Weiss F. Stimulus conditioned to foot-shock stress reinstates alcohol-seeking behavior in an animal model of relapse. *Psychopharmacology (Berl)*. 2003;168(1–2):184-191. doi:10.1007/s00213-002-1267-z
5. Zironi I, Burattini C, Aicardi G, Janak PH. Context is a trigger for relapse to alcohol. *Behav Brain Res*. 2006;167(1):150-155. doi:10.1016/j.bbr.2005.09.007
6. Janak PH, Chaudhri N. The potent effect of environmental context on relapse to alcohol-seeking after extinction. *Open Addict J*. Jan 1 2010; 3(2):76-87. doi:10.2174/1874941001003010076
7. Laberg JC. What is presented, and what prevented, in cue exposure and response prevention with alcohol dependent subjects? *Addict Behav*. 1990;15(4):367-386. doi:10.1016/0306-4603(90)90046-z
8. Rohsenow DJ, Niaura RS, Childress AR, Abrams DB, Monti PM. Cue reactivity in addictive behaviors: theoretical and treatment implications. *Int J Addict*. 1990;25(7A-8A):957-993. doi:10.3109/10826089109071030
9. Haass-Koffler CL, Swift RM, Leggio L. Noradrenergic targets for the treatment of alcohol use disorder. *Psychopharmacology (Berl)*. 2018; 235(6):1625-1634. doi:10.1007/s00213-018-4843-6

10. Dunn AJ, Swiergiel AH. The role of corticotropin-releasing factor and noradrenaline in stress-related responses, and the inter-relationships between the two systems. *Eur J Pharmacol*. 2008;583(2–3):186-193. doi:10.1016/j.ejphar.2007.11.069
11. Bryant RA, McGrath C, Felmingham KL. The roles of noradrenergic and glucocorticoid activation in the development of intrusive memories. *PLoS ONE*. 2013;8(4):e62675. doi:10.1371/journal.pone.0062675
12. Blaine SK, Sinha R. Alcohol, stress, and glucocorticoids: from risk to dependence and relapse in alcohol use disorders. *Neuropharmacology*. Aug 1 2017;122:136-147. doi:10.1016/j.neuropharm.2017.01.037
13. Block TS, Kushner H, Kalin N, Nelson C, Belanoff J, Schatzberg A. Combined analysis of mifepristone for psychotic depression: plasma levels associated with clinical response. *Biol Psychiatry*. 2018;84(1):46-54. doi:10.1016/j.biopsych.2018.01.008
14. Vendruscolo LF, Estey D, Goodell V, et al. Glucocorticoid receptor antagonism decreases alcohol seeking in alcohol-dependent individuals. *J Clin Invest*. 2015;125(8):3193-3197. doi:10.1172/JCI79828
15. Simms JA, Haass-Koffler CL, Bito-Onon J, Li R, Bartlett SE. Mifepristone in the central nucleus of the amygdala reduces yohimbine stress-induced reinstatement of ethanol-seeking. *Neuropsychopharmacology*. 2012;37(4):906-918. doi:10.1038/npp.2011.268
16. Vendruscolo LF, Barbier E, Schlosburg JE, et al. Corticosteroid-dependent plasticity mediates compulsive alcohol drinking in rats. *J Neurosci*. 2012;32(22):7563-7571. doi:10.1523/JNEUROSCI.0069-12.2012
17. Jimenez VA, Walter NAR, Shnitko TA, et al. Mifepristone decreases chronic voluntary ethanol consumption in rhesus macaques. *J Pharmacol Exp Ther*. 2020;375(2):258-267. doi:10.1124/jpet.120.000169
18. Holtyn AF, Weerts EM. Evaluation of mifepristone effects on alcohol-seeking and self-administration in baboons. *Exp Clin Psychopharmacol*. 2019;27(3):227-235. doi:10.1037/pha0000246
19. McGinn MA, Tunstall BJ, Schlosburg JE, et al. Glucocorticoid receptor modulators decrease alcohol self-administration in male rats. *Neuropharmacology*. 2021;188:108510. doi:10.1016/j.neuropharm.2021.108510
20. Fahlke C, Hard E, Eriksson CJ, Engel JA, Hansen S. Consequence of long-term exposure to corticosterone or dexamethasone on ethanol consumption in the adrenalectomized rat, and the effect of type I and type II corticosteroid receptor antagonists. *Psychopharmacology (Berl)*. 1995;117(2):216-224. doi:10.1007/BF02245190
21. Curley DE, Vasaturo-Kolodner TR, Cannella N, Ciccocioppo R, Haass-Koffler CL. Yohimbine as a pharmacological probe for alcohol research: a systematic review of rodent and human studies. *Neuropsychopharmacology*. 2022;47(12):2111-2122. doi:10.1038/s41386-022-01363-9
22. Bertholomey ML, Verplaetse TL, Czachowski CL. Alterations in ethanol seeking and self-administration following yohimbine in selectively bred alcohol-preferring (P) and high alcohol drinking (HAD-2) rats. *Behav Brain Res*. Feb 1 2013;238:252-258. doi:10.1016/j.bbr.2012.10.030
23. Richards JK, Simms JA, Bartlett SE. Conditioned cues and yohimbine induce reinstatement of beer and near-beer seeking in Long-Evans rats. *Addict Biol*. 2009;14(2):144-151. doi:10.1111/j.1369-1600.2008.00139.x
24. Stanford S. Central noradrenergic neurones and stress. *Pharmacol Ther*. 1995;68(2):297-342. doi:10.1016/0163-7258(95)02010-1
25. Umhau JC, Schwandt ML, Usala J, et al. Pharmacologically induced alcohol craving in treatment seeking alcoholics correlates with alcoholism severity, but is insensitive to acamprosate. *Neuropsychopharmacology*. 2011;36(6):1178-1186. doi:10.1038/npp.2010.253
26. Woodcock EA, Stanley JA, Diwadkar VA, Khatib D, Greenwald MK. A neurobiological correlate of stress-induced nicotine-seeking behavior among cigarette smokers. *Addict Biol*. 2020;25(4):e12819. doi:10.1111/adb.12819
27. Woodcock EA, Greenwald MK, Khatib D, Diwadkar VA, Stanley JA. Pharmacological stress impairs working memory performance and attenuates dorsolateral prefrontal cortex glutamate modulation. *Neuroimage*. Feb 1 2019;186:437-445. doi:10.1016/j.neuroimage.2018.11.017
28. Flores BH, Kenna H, Keller J, Solvason HB, Schatzberg AF. Clinical and biological effects of mifepristone treatment for psychotic depression. *Neuropsychopharmacology*. 2006;31(3):628-636. doi:10.1038/sj.npp.1300884
29. DeBattista C, Belanoff J, Glass S, et al. Mifepristone versus placebo in the treatment of psychosis in patients with psychotic major depression. *Biol Psychiatry*. 2006;60(12):1343-1349. doi:10.1016/j.biopsych.2006.05.034
30. First MB. *Structured Clinical Interview for DSM-IV Axis I Disorders*. Biometrics Research Department; 1997.
31. Gallagher P, Watson S, Elizabeth Dye C, Young AH, Nicol FI. Persistent effects of mifepristone (RU-486) on cortisol levels in bipolar disorder and schizophrenia. *J Psychiatr Res*. 2008;42(12):1037-1041. doi:10.1016/j.jpsychires.2007.12.005
32. Johanssen S, Allolio B. Mifepristone (RU 486) in Cushing's syndrome. *Eur J Endocrinol*. 2007;157(5):561-569. doi:10.1530/EJE-07-0458
33. Kathol RG, Anton R, Noyes R, Gehris T. Direct comparison of urinary free cortisol excretion in patients with depression and panic disorder. *Biol Psychiatry*. 1989;25(7):873-878. doi:10.1016/0006-3223(89)90267-9
34. Sachar EJ, Hellman L, Roffwarg HP, Halpern FS, Fukushima DK, Gallagher TF. Disrupted 24-hour patterns of cortisol secretion in psychotic depression. *Arch Gen Psychiatry*. 1973;28(1):19-24. doi:10.1001/archpsyc.1973.01750310011002
35. Wangelin BC, Powers MB, Smits JA, Tuerk PW. Enhancing exposure therapy for PTSD with yohimbine HCL: protocol for a double-blind, randomized controlled study implementing subjective and objective measures of treatment outcome. *Contemp Clin Trials*. 2013;36(2):319-326. doi:10.1016/j.cct.2013.08.003
36. Greenwald MK, Lundahl LH, Steinmiller CL. Yohimbine increases opioid-seeking behavior in heroin-dependent, buprenorphine-maintained individuals. *Psychopharmacology (Berl)*. 2013;225(4):811-824. doi:10.1007/s00213-012-2868-9
37. Hamilton M. Diagnosis and rating of anxiety. *Br J Psychiatry*. 1969;3-(special issue):76-79.
38. Hamilton M. A rating scale for depression. *J Neurol Neurosurg Psychiatry*. 1960;23(1):56-62. doi:10.1136/jnnp.23.1.56
39. Spielberg C. *Manual for the State-Trate Anxiety Inventory*. Consulting Psychologists Press; 1970.
40. Cohen S, Kamarck T, Mermelstein R. Perceived stress scale. Measuring stress: a guide for health and social scientists 1994;10(2):1–2.
41. Jacobs D. Suicide assessment five-step evaluation and triage (SAFE-T). 2009.
42. Sobell LC, Sobell MB. Timeline follow-back: a technique for assessing self-reported alcohol consumption. *Meas Alcohol Consumpt: Psychosoc Biochem Methods*. 1992;41-72. doi:10.1007/978-1-4612-0357-5_3
43. Gray MJ, Litz BT, Hsu JL, Lombardo TW. Psychometric properties of the life events checklist. *Assessment*. 2004;11(4):330-341. doi:10.1177/1073191104269954
44. Schnurr P, Vielhauer M, Weathers F, Findler M. Brief Trauma Questionnaire. 1999.
45. Sullivan JT, Sykora K, Schneiderman J, Naranjo CA, Sellers EM. Assessment of alcohol withdrawal: the revised clinical institute withdrawal assessment for alcohol scale (CIWA-Ar). *Br J Addict*. 1989;84(11):1353-1357. doi:10.1111/j.1360-0443.1989.tb00737.x
46. Owen JA, Nakatsu SL, Fenemore J, Condra M, Surridge DH, Morales A. The pharmacokinetics of yohimbine in man. *Eur J Clin Pharmacol*. 1987;32(6):577-582. doi:10.1007/BF02455991

47. Monti PM, Rohsenow DJ, Hutchison KE, et al. Naltrexone's effect on cue-elicited craving among alcoholics in treatment. *Alcohol Clin Exp Res*. 1999;23(8):1386-1394. doi:10.1111/j.1530-0277.1999.tb04361.x
48. Singleton E, Tiffany S, Henningfield J. Development and validation of a new questionnaire to assess craving for alcohol: problems of drug dependence 1994:289.
49. Haass-Koffler CL, Perciballi R, Magill M, et al. An inpatient human laboratory study assessing the safety and tolerability, pharmacokinetics, and biobehavioral effect of GET 73 when co-administered with alcohol in individuals with alcohol use disorder. *Psychopharmacology (Berl)*. 2021;239(1):35-46. doi:10.1007/s00213-021-06008-1
50. Haass-Koffler CL, Leggio L, Davidson D, Swift RM. Effects of idazoxan on alcohol pharmacokinetics and intoxication: a preliminary human laboratory study. *Alcohol Clin Exp Res*. 2015;39(4):594-602. doi:10.1111/acer.12658
51. Martin CS, Earleywine M, Musty RE, Perrine MW, Swift RM. Development and validation of the Biphasic Alcohol Effects Scale. *Alcohol Clin Exp Res*. 1993;17(1):140-146. doi:10.1111/j.1530-0277.1993.tb00739.x
52. Anton RF, Drobes DJ, Voronin K, Durazo-Avizu R, Moak D. Naltrexone effects on alcohol consumption in a clinical laboratory paradigm: temporal effects of drinking. *Psychopharmacology (Berl)*. 2004;173(1–2):32-40. doi:10.1007/s00213-003-1720-7
53. 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. *Psychopharmacology (Berl)*. 2002;160(1):19-29. doi:10.1007/s002130100919
54. Drobes DJ, Anton RF, Thomas SE, Voronin K. A clinical laboratory paradigm for evaluating medication effects on alcohol consumption: naltrexone and nalmefene. *Neuropsychopharmacology*. 2003;28(4):755-764. doi:10.1038/sj.npp.1300101
55. Schulz K, Altman D, Moher D. Consolidated standards of reporting trials group CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *BMJ*. 2010;340(mar23 1):c332. doi:10.1136/bmj.c332
56. Pearce K, Fisher L, Dixon D, Herson J, Frankowski R, Hearon M. In: Pearce KE, ed. *Intention to Treat in Clinical Trials, Statistical Issues in Drug Research and DEVELOPMENT*. Marcel Dekker; 1990.
57. Tukey JW. The future of data analysis. *Ann Math Stat*. 1962;33(1):1-67. doi:10.1214/aoms/1177704711
58. Zeger SL, Liang K-Y. Longitudinal data analysis for discrete and continuous outcomes. *Biometrics*. 1986;121-130(1):121. doi:10.2307/2531248
59. Singleton E. Alcohol craving questionnaire, short-form (revised; ACQ-SF-R): background, scoring, and administration. Baltimore, MD, USA. 1995.
60. Montoya AK, Hayes AF. Two-condition within-participant statistical mediation analysis: a path-analytic framework. *Psychol Methods*. 2017;22(1):6-27. doi:10.1037/met0000086
61. U.S. Department of Health and Human Services FDA. *Guidance for Industry, E9 Statistical Principles for Clinical Trials*. Vol. E9. Center for Drug Evaluation and Research (CDER) CfBEaRC; 1998:1998. https://www.fda.gov/downloads/drugs/guidancecomplianceregulatoryinformation/guidances/ucm073137.pdf
62. Little RJ, Rubin DB. *Statistical Analysis With Missing Data*. Vol. 793. John Wiley & Sons; 2019. doi:10.1002/9781119482260
63. Dwan K, Li T, Altman DG, Elbourne D. CONSORT 2010 statement: extension to randomised crossover trials. *BMJ*. 2019;366:l4378. doi:10.1136/bmj.l4378
64. Brown ZE, Portis SM, Fleig CL, Vasaturo-Kolodner T, Swift RM, Haass-Koffler CL. Randomized controlled trials for alcohol use disorder during the COVID-19 pandemic. *Alcohol*. 2021;92:21-24. doi:10.1016/j.alcohol.2021.01.001
65. Lin G, Rodriguez RN, SAS Institute Inc. Weighted methods for analyzing missing data with the GEE procedure. *Paper SAS166-2015*. 2015:1-8.
66. Fox HC, Hong KI, Siedlarz K, Sinha R. Enhanced sensitivity to stress and drug/alcohol craving in abstinent cocaine-dependent individuals compared to social drinkers. *Neuropsychopharmacology*. 2008;33(4):796-805. doi:10.1038/sj.npp.1301470
67. Haass-Koffler CL, Goodyear K, Long VM, et al. A phase I randomized clinical trial testing the safety, tolerability and preliminary pharmacokinetics of the mGluR5 negative allosteric modulator GET 73 following single and repeated doses in healthy volunteers. *Eur J Pharm Sci*. 2017;109:78-85. doi:10.1016/j.ejps.2017.07.031
68. Haass-Koffler CL, Goodyear K, Loche A, et al. Administration of the metabotropic glutamate receptor subtype 5 allosteric modulator GET 73 with alcohol: a translational study in rats and humans. *J Psychopharmacol*. 2018;32(2):163-173. doi:10.1177/0269881117746904
69. Haass-Koffler CL, Goodyear K, Zywiak WH, Leggio L, Kenna GA, Swift RM. Comparing and combining topiramate and aripiprazole on alcohol-related outcomes in a human laboratory study. *Alcohol Alcohol*. 2018;53(3):268-276. doi:10.1093/alcalc/agx108
70. GUIDANCE D. *Alcoholism: developing drugs for treatment*. Center for Drug Evaluation and Research (CDER); 2015.
71. Marquis P, Caron M, Emery M-P, Scott JA, Arnould B, Acquadro C. The role of health-related quality of life data in the drug approval processes in the US and Europe: a review of guidance documents and authorizations of medicinal products from 2006 to 2010. *Pharmaceut Med*. 2011;25(3):147-160. doi:10.1007/BF03256856
72. Haass-Koffler CL, Long VM, Farokhnia M, et al. Intravenous administration of ghrelin increases serum cortisol and aldosterone concentrations in heavy-drinking alcohol-dependent individuals: results from a double-blind, placebo-controlled human laboratory study. *Neuropharmacology*. 2019;158:107711. doi:10.1016/j.neuropharm.2019.107711
73. Leggio L, Zywiak WH, Fricchione SR, et al. Intravenous ghrelin administration increases alcohol craving in alcohol-dependent heavy drinkers: a preliminary investigation. *Biol Psychiatry*. 2014;76(9):734-741. doi:10.1016/j.biopsych.2014.03.019
74. Leggio L, Zywiak WH, McGeary JE, et al. A human laboratory pilot study with baclofen in alcoholic individuals. *Pharmacol Biochem Behav*. 2013;103(4):784-791. doi:10.1016/j.pbb.2012.11.013
75. Rohsenow DJ, Monti PM, Abrams DB, et al. Cue elicited urge to drink and salivation in alcoholics: relationship to individual differences. *Advances in Behaviour Research and Therapy*. 1992;14(3):195-210. doi:10.1016/0146-6402(92)90008-C
76. Lin CC, Cheng PY, Hsiao M, Liu YP. Effects of RU486 in treatment of traumatic stress-induced glucocorticoid dysregulation and fear-related abnormalities: early versus late intervention. *Int J Mol Sci*. 2022;23(10):5494. doi:10.3390/ijms23105494
77. Vining RF, McGinley RA, Maksvytis JJ, Ho KY. Salivary cortisol: a better measure of adrenal cortical function than serum cortisol. *Ann Clin Biochem*. 1983;20(Pt 6):329-335. doi:10.1177/000456328302000601
78. Benvenuti F, Cannella N, Stopponi S, et al. Effect of glucocorticoid receptor antagonism on alcohol self-administration in genetically-selected Marchigian Sardinian alcohol-preferring and non-preferring wistar rats. *Int J Mol Sci*. 2021;22(8):4184. doi:10.3390/ijms22084184
79. Vozella V, Cruz B, Natividad LA, et al. Glucocorticoid receptor antagonist mifepristone does not Alter innate anxiety-like behavior in genetically-selected Marchigian Sardinian (msP) rats. *Int J Mol Sci*. 2021;22(6):3095. doi:10.3390/ijms22063095
80. Haass-Koffler CL, Piacentino D, Li X, et al. Differences in sociodemographic and alcohol-related clinical characteristics between treatment seekers and nontreatment seekers and their role in predicting outcomes in the COMBINE study for alcohol use disorder. *Alcohol Clin Exp Res*. 2020;44(10):2097-2108. doi:10.1111/acer.14428

81. Nieto SJ, Grodin EN, Aguirre CG, Izquierdo A, Ray LA. Translational opportunities in animal and human models to study alcohol use disorder. *Transl Psychiatry*. 2021;11(1):496. doi:10.1038/s41398-021-01615-0
82. Haass-Koffler CL. Utilizing precision medicine to treat alcohol use disorder: a commentary on the alpha1 receptor antagonist. *Alcohol Clin Exp Res*. 2023;47(3):432-434. doi:10.1111/acer.15014
83. Logrip ML, Gainey SC. Sex differences in the long-term effects of past stress on alcohol self-administration, glucocorticoid sensitivity and phosphodiesterase 10A expression. *Neuropharmacology*. 2020;164:107857. doi:10.1016/j.neuropharm.2019.107857
84. Haass-Koffler CL, Sheffler DJ. Neuroscience targets and human studies: future translational efforts in the stress system. *Neuropsychopharmacology*. 2023;48(5):711-712. doi:10.1038/s41386-023-01539-x
85. Haass-Koffler CL, Kenna GA. Bacchus by Caravaggio as the visual diagnosis of alcohol use disorder from the fifth edition of the diagnostic and statistical manual of mental disorders (DSM-5). *Front Psych*. 2013;4:86. doi:10.3389/fpsyt.2013.00086
86. Haass-Koffler CL, Leggio L, Kenna GA. Pharmacological approaches to reducing craving in patients with alcohol use disorders. *CNS Drugs*. 2014;28(4):343-360. doi:10.1007/s40263-014-0149-3

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Haass-Koffler CL, Magill M, Cannella N, et al. Mifepristone as a pharmacological intervention for stress-induced alcohol craving: A human laboratory study. *Addiction Biology*. 2023;28(7):e13288. doi:10.1111/adb.13288